UNIVERSITÀ DI MILANO
"CENTRO DINO FERRARI"
PER LA DIAGNOSI E LA TERAPIA DELLE MALATTIE
NEUROMUSCOLARI E NEURODEGENERATIVE

FONDAZIONE I.R.C.C.S. CA' GRANDA
OSPEDALE MAGGIORE POLICLINICO
ISTITUTO DI RICOVERO E CURA A CARATTERE
SCIENTIFICO DI NATURA PUBBLICA

COLLABORAZIONI INTERNAZIONALI

E

FRONTESPIZI

LAVORI SCIENTIFICI

2019

" CENTRO DINO FERRARI"

Sezione di Neuroscienze
Dipartimento di Fisiopatologia Medico-Chirurgica e dei Trapianti
Università degli Studi di Milano
Fondazione I.R.C.C.S. Ca' Granda - Ospedale Maggiore Policlinico

COORDINATORE SCIENTIFICO PROF. NEREO BRESOLIN
Via F. Sforza, 35 - 20122 Milano – Tel. 02.5503.3809 - Fax 02.503.20448
E-mail: nereo.bresolin@unimi.it - centro.dinoferrari@policlinico.mi.it - www.centrodinoferrari.com

UNIVERSITÀ DI MILANO
"CENTRO DINO FERRARI"
PER LA DIAGNOSI E LA TERAPIA DELLE MALATTIE
NEUROMUSCOLARI E NEURODEGENERATIVE

FONDAZIONE I.R.C.C.S. CA' GRANDA
OSPEDALE MAGGIORE POLICLINICO
ISTITUTO DI RICOVERO E CURA A CARATTERE
SCIENTIFICO DI NATURA PUBBLICA

Istituti di ricerca del "Centro Dino Ferrari":

- **IRCCS FONDAZIONE CA' GRANDA OSPEDALE MAGGIORE POLICLINICO UNIVERSITA' DEGLI STUDI DI MILANO**
  - **Laboratorio di biochimica, Genetica e Colture Cellulari**
  - **Laboratorio di Cellule Staminali Neurali**
  - **U.O.S.D. Malattie Neuromuscolari e Rare**
  - **U.O.S.D. Malattie Neurodegenerative – Unità Valutativa Alzheimer (U.V.A.)**
  - **Centro Sclerosi Multipla**
  - **Laboratorio Parkinson e altri Disturbi del Movimento**
  - **Laboratorio di Cellule Staminali**
- **IRCCS ISTITUTO AUXOLOGICO ITALIANO – UNIVERSITA' DEGLI STUDI DI MILANO**
  - **U.O. Neurologia - Stoke Unit**
  - **Laboratorio di Neuroscienze**

COORDINATORE SCIENTIFICO PROF. NEREO BRESOLIN
Via F. Sforza, 35 - 20122 Milano – Tel. 02.5503.3809 - Fax 02.503.20448
E-mail: nereo.bresolin@unimi.it - centro.dinoferrari@policlinico.mi.it - www.centrodinoferrari.com

Centri Internazionali di Ricerca che collaborano con il "Centro Dino Ferrari"

- Prof. S. Przedborski **Motor Neuron Center, Columbia University, Columbia University of New York – USA**
- Prof. B. Kaspar - **Avexis, Lake Forest, IL, USA A**.
- Prof. Kathrin Meyer- **Nation Wide Children's Hospital**
- Prof. E. Hedlund - **Karolinska Institutet, Stockholm, SWEDEN.**
- Prof. S. Di Mauro - **Columbia University of New York – USA.**
- Prof. L. Stefanis Biomedical Foundation – **Academy of Athens, Grecia.**
- Dr. Marc Ruepp and Prof. Müehlemann O, **University of Bern.**
- Prof. H. Moulton **Oregon University, USA**
- Prof. Federico Mingozzi, **Genethon, Inserm, Evry, Università Parigi-Saclay; Università Pierre and Marie Curie Parigi 6 e INSERM U974, France**
- Dr. Enrico Bertini, **IRCCS Ospedale Bambino Gesu', Roma 13**
- Prof. Luca Imeri, **Dipartimento di Fisiologia Umana, Università degli Studi di Milano, Milano**
- Dr. Uberto Pozzoli, **IRCCS E. Medea Bosisio, Parini, Italy**
- Prof. ML Gelmi, **Dipartimento di Scienze Farmaceutiche, Università degli Studi di Milano.**
- Prof. Cristina Guardia - Laguarta**, USA Columbia University New York, N.Y., USA**
- Prof. D. Re**, Columbia University New York, N.Y., USA**
- Prof. Catarina Quinzii, PhD, **Columbia University, New York, N.Y., USA**
- Prof. Jeroen Pasterkamp **University Medical Center Utrecht, Utrecht University, Olanda**
- Prof. Philip Van Damme **Department of Neurology Universitair Ziekenhuis Leuven, Belgio**
- Dr. Simona Lodato **Humanitas Research Center, Milano**
- Dr. Maria Grazia Biferi**, Insitut de Myologie Parigi, Francia**

- NYU Movement Disorders, Fresco Institute for Parkinson's Disease, New York University – USA
- Dr. Daniel Claassen il **Vanderbilt University Medical Center, Nashville US.**
- Prof. Kyproula Christodoulou, **The Cyprus Institute of Neurology and Genetics, Cyprus School of Molecular Medicine**
- Prof. Bearne Udd - **Tampere University, FINLANDIA**
- Prof. Martin Rossor - **UCL School of Life and Medical Sciences, London, UNITED KINGDOM.**
- Prof. Bengt Winblad, **Karolinska Institutet, Stockholm, SWEDEN**.
- Dr. Luis Garcia – **Institut de Myologie, Université Pierre et Marie Curie, Paris, FRANCE.**
- Prof. Camillo Ricordi, **University of Miami (UM), Miami, Florida**
- Prof. Giulio Cossu, **Institute of Infalmmation and repair, University of Manchester, Manchester, UK**
- Prof. Pura Muñoz Cánoves**, Department of Experimental and Life Sciences, Pompeu Fabra University, Barcelona, Spain**
- Prof. Jacques Tremblay**, Centre de recherche, Centre hospitalier de l'Université de Montréal, (CRCHUM), Montréal, Québec, Canada**
- Dr. Joao Carlos da Silva Bizario – **Muscular Dystrophy Research Center** AADM/UNAERP, **Ribeirao Preto, SP, BRAZIL.**
- Prof. Humberto Cerrel Bazo, **Dipartimento Medicina riabilitativa AUSL Piacenza**
- Prof. Adolfo Lopez de Munain Arregui**, Grupo Nerogenética, Hospital Donostia-Unidad Experimental San Sebastian, Espana**
- Prof. Kay Davies**, Department of Physiology, Anatomy and Genetics, University of Oxford, Oxford, UK**
- Maurilio Sampaolesi**,** Stem Cell Research Institute, University Hospital Gasthuisberg, Leuven, Belgium, Human Anatomy Section, **University of Pavia, Pavia, Italy, Interuniversity Institute of Myology (IIM), Italy**
- Gillian Butler-Brown and Vincent Mouly, Institut de Myologie, Institut national de la sante´ et de la recherche me´ dicale, and **L'Universite´ Pierre et Marie Curie Paris, Paris, France**
- Prof. Giuseppe Perale**, I.B.I. S/A, Svizzera, Dipartimento di Chimica, Materiali e Ingegneria Chimica "Giulio Natta" Sezione Chimica Fisica Applicata, Politecnico di Milano, Milano**
- Prof. Roberto Maggi, **Facoltà di Farmacia Università degli Studi di Milano**
- Prof. Mario Pellegrino, **Dipartimento di Ricerca Traslazionale e delle Nuove Tecnologie in Medicina e Chirurgia, Università di Pisa**
- Prof. Daniele Cusi, **Università degli Studi di Milano**

- Anna Spada, U.O. di Endocrinologia e Diabetologia, **Dipartimento di Scienze Mediche, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico**
- Lorenza Lazzari e Rosaria Giordano**,** Cell Factory Center for Transfusion Medicine, Cell Therapy and Criobiology, Department of Regenerative Medicine, **Fondazione IRCCS Cà Granda Ospedale Maggiore Policlinico di Milano**
- Giuseppe D'Antona**,** Department of Molecular Medicine, **University of Pavia, Pavia, Italy** LUSAMMR, Laboratory for Motor Activities in Rare Diseases, Sport Medicine, Centre Voghera, Voghera, Italy
- Mauro Pluderi e Nadia Grimoldi**,** UO Neurochirurgia, **Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano**
- Giorgio Pajardi, Dipartimento di Scienze Cliniche e di Comunità, **Università degli Studi di Milano, Direttore U.O.C. di Chirurgia e Riabilitazione della Mano Ospedale S. Giuseppe Milano, Gruppo MultiMedica IRCCS**
- Dr Yuri D'Alessandra – Unità di Immunologia e genomica funzionale, **Centro Cardiologico Monzino IRCCS, Milan, Italy**
- Prof. Stefano Biressi – **Centro di Biologia Integrata – CIBIO, Università degli Studi di Trento**
- Prof. Lorenzo Bello - **Neurochirurgia Oncologica, Humanitas, Milano**
- Prof. Alberto Priori - **U.O.C. Neurologia, Ospedale San Paolo, Milano**
- Prof. Pietro Mauri – **Istituto di Tecnologie Biomediche, Consiglio Nazionale delle Ricerche (CNR-ITB), Milano**
- D.ssa Barbara Cassani – **Instituto di Genetica e Biomedicina (IRGB), National Research Council (CNR) Milano**
- Prof. Sabrina Sacconi – **Nice University Hospital, Nice, France**
- Dr. E. M. Mercuri - **Pediatric Neurology and Nemo Clinical Centre, Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario "A. Gemelli", Rome, Italy**
- Dr. F. Alessandrino - **Neuroradiology Department, IRCCS C. Mondino Foundation, Pavia, Italy**
- Dr. P. Ripellino - **Department of Neurology, Neurocenter of Southern Switzerland, Lugano, Switzerland**
- Dr. M. Filosto **- Center for Neuromuscular Diseases, Unit of Neurology, ASST Spedali Civili and University of Brescia, Brescia, Italy**
- Dr. S. Ravaglia - **Emergency Neurology, IRCCS Mondino Foundation, Pavia, Italy**

- Dr. O. Musumeci - **Department of Clinical and Experimental Medicine, UOC di Neurologia e Malattie Neuromuscolari, University of Messina, Messina, Italy**

- Dr. A. Toscano - **Department of Clinical and Experimental Medicine, UOC di Neurologia e Malattie Neuromuscolari, University of Messina, Messina, Italy**

- Dr. C. Lamperti - **Unit of Medical Genetics and Neurogenetics, Fondazione IRCCS Istituto Neurologico 'Carlo Besta', Milan, Italy**

- Dr. G. Siciliano - **Neurological Clinic, University of Pisa, Pisa, Italy**

- Dr. J. Laporte - **Department of translational medicine and Neurogenetics, Institut de Génétique et de Biologie Moléculaire et Cellulaire (IGBMC), Illkirch, France, Strasbourg University, Illkirch, France**

- Dr. J. Böhm - **Department of translational medicine and Neurogenetics, Institut de Génétique et de Biologie Moléculaire et Cellulaire (IGBMC), Illkirch, France, Strasbourg University, Illkirch, France**

- Dr. J. A. Ross - **Centre for Human and Applied Physiological Sciences, School of Basic and Medical Biosciences, Faculty of Life Sciences and Medicine, Guy's Campus, King's College London, London SE1 1UL, UK**

- Dr. C. Fiorillo - **IRCCS Gaslini and Department of Neuroscience, Rehabilitation, Ophthalmology, Genetics, Maternal and Child Health, Genoa University, Genoa, Italy**

- Dr. E. Bertini - **IRCCS Ospedale Bambino Gesu', Roma 13**

- Dr. F. Triulzi **- UOC Neuroradiologia, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico**

- Dr. F. M. Santorelli - **Fondazione Stella Maris, Pisa**

- Dr. Bernard Brais – **McGill University/Montreal Neurological Hospital and Institute, Montreal, Quebec, CANADA**

- Dr. Gorka Orive – **BTI Biotechnology Institute, Vitoria-Gasteiz, Alava, SPAIN**

- Dr. Diana Nordling – **Cincinnati Children's Hospital, Cincinnati, OH, USA**

- Prof. Fulvio Mavilio – **Institut Genethon, Evry, FRANCE.**

- Prof. Guglielmo Foffani, **Hospital Nacional Parapléjicos, Toledo, SPAIN.**

- Prof. John Rothwell, Sobell Department, **UCL Institute of Neurology**, **London, UNITED KINGDOM.**

- Prof. Elena Moro, **Department of Psychiatry and Neurology , University Hospital Center of Grenoble, FRANCE.**

- Dr. Andre Brunoni, **Department of Neurosciences and Behavior, Institute of Psychology**, **University of Sao Paulo, Sao Paulo, BRAZIL.**

- Prof. Dale J. Lange, M.D. Chair, **Department of Neurology Neurologist-in-Chief, Hospital For Special Surgery - New York, USA.**
- Prof. Hiroshi Mitsumoto, **MD, DSc Head, Eleanor and Lou Gehrig MDA/ALS Research Center, Department of Neurology, Columbia University Medical Center, New York, USA.**
- Prof. John E. Landers, - **University of Massachusetts Medical School, Worcester, MA, USA.**
- Prof. Merit E. Cudkowicz, MD, MSc - **Neuromuscular Division Neurology - Massachusetts General Hospital Wang Ambulatory Care Center - Boston, MA, USA**
- Prof. Stanley H. Appel MD, **Edwards Distinguished Endowed Chair for ALS Director, Methodist Neurological Institute Chair, Department of Neurology - Houston, Texas, USA.**
- Prof. Rosa Rademakers - **Mayo Clinic Florida, Department of Neuroscience Jacksonville, Florida, USA.**
- Prof. Kendall Jensen – **Tgen -The Translational Genomics Research Institute, Phoenix, AZ, USA.**
- Prof. Matthew Alexander - **Children's Hospital Boston - Boston, MA, USA.**
- Prof. Christopher Klein- Mayo Clinic - **Department of Neurology- Rochester**.
- Prof. Joshua Selsby, **Iowa State University – Ames - USA.**
- Prof. Alexey Belkin, **University of Maryland – Baltimore - USA.**
- Prof. Toshifumi Yokota- **University of Alberta – Edmonton - CANADA.**
- Prof. Xiping Cheng- **University of Michigan - Ann Arbor - USA.**
- Prof. Alexis Brice, **Sorbonne Universités, Université Pierre et Marie Curie Paris 06, Unité Mixte de Recherche (UMR) S 1127, Institut du Cerveau et de la Moelle Épinière (ICM), Paris France**
- Prof. Isabelle Le Ber, **Sorbonne Universités, Université Pierre et Marie Curie Paris 06, Unité Mixte de Recherche (UMR) S 1127, Institut du Cerveau et de la Moelle Épinière (ICM), Paris France**
- Prof. Bruno Dubois, **Institute de la Mémoire et de la Maladie d'Alzheimer (IM2A), Département de Neurologie, Hôpital de la Pitié-Salpêtrière, AP-HP, Paris, France; INSERM, CNRS, UMR-S975, Institut du Cerveau et de la Moelle Epinière (ICM), Paris, France; Sorbonne, Universités, Université Pierre**
- Prof. Adriana Maggi, Centro di Biotecnologie Farmacologiche, **Dipartimento di Scienze Farmacologiche, Università di Milano**
- Dott. Gianluigi Forloni, **Istituto di Ricerche Farmacologiche Mario Negri, Milano**
- Prof. Stefano Cappa, **Università di Pavia**
- Prof. Alessandro Padovani, **Università di Brescia**
- Prof. Innocenzo Rainero, **Università di Torino**

- ❖ Prof. Claudio Franceschi, **Università di Torino**
- ❖ Dott. Claudia Verderio, **CNR, Milano**
- ❖ Prof. Robert P. Lisak, **Dip. di Neurologia, Detroit, USA**
- ❖ Prof. Philip Scheltens, Dr. Yolande Pijnenburg, **Dept. of Neurology, VU University Medical Center, Amsterdam, The Nertherlands**
- ❖ Prof. Janine Diehl-Schmid, **Technical University of Munich, Munich, Germany**
- ❖ Prof. Simon Ducharme McGill **University, Montreal, Canada**
- ❖ Prof. Glenda Halliday, **University of Sydney, Australia**
- ❖ Dr. Anne Cross, **University of Saint Louis, USA**
- ❖ Prof. Jean Charles Lambert, **Lille, France**
- ❖ Dr. Jonathan Rohrer, UCL, **London, UK**
- ❖ Prof. An Goris, **University of Leuven, Belgium**
- ❖ Dr. Manuel Comabella, **Universitat Autònoma de Barcelona, Barcelona, Spain**
- ❖ Prof. Andreas Reif, **University of Frankfurt, Germany**
- ❖ Prof. Albert Ludolph, **Dept. Neurology – University of Ulm, Germany.**
- ❖ Prof. Amman Al-Chalabi, **King's College – London, United Kingdom.**
- ❖ Prof. Christopher Shaw, **King's College – London , United Kingdom**
- ❖ Prof. John Powel, **King's College – London , United Kingdom**
- ❖ P.D. Dr. Marcus Weber**, St. Gallen, CH**
- ❖ Prof. E. I Rugarli **– Institute for Genetics CECAD Research Center University of Cologne Joseph-Stelzmann, Köln Germany**
- ❖ Prof. W Griffith – **Institute of Mass Spectrometry College of Medicine Grove Building Swansea University Wales, UK**
- ❖ Dr. Edward J Hollox – **Department of Genetics, University of Leicester, Leicester, UK**
- ❖ Dr. Nasser M. Al-Daghri, **Biochemistry Department College of Science, King Saud University, Kingdom of Saudi Arabia Riyadh 11451(KSA)**
- ❖ Prof. Prince Mutaib, **Biochemistry Department, College of science, King Saud University, Riyadh, KSA**
- ❖ Prof. Lu Qilong **- McColl-Lockwood Laboratory for Muscular Dystrophy Research, Neuromuscular/ALS Center, Carolinas Medical Center, Charlotte, North Carolina, USA.**

- Dr. Angelo Monguzzi **, Dept. Scienze dei Materiali, Università degli Studi di Milano-Bicocca**
- Dr. Maurilio Bruno – **Istituto Ortopedico Galeazzi, IRCCS**
- Dr. Francesco Nicassio **– Department of Experimental Oncology, European Institute of Oncology, IFOM-IEO Campus**
- Prof. Graziella Messina – **Dept. Bioscienze, Università degli Studi di Milano**
- Prof. Angelo Poletti – **Dept. Di Science Farmacologiche e Biomolecolari, Università degli Studi di Milano**
- Prof. Silvio Bicciato**, bioinformatics unit, Faculty of Biosciences and Biotechnologies, University of Modena and Reggio Emilia**
- Prof. Enrico Tagliafico **, clinical Biochemistry, University of Modena and Reggio Emilia**
- Prof. Sergio Abrignani, **National Institute of Molecular Genetics (INGM), Milan, Italy**
- Prof. Silvano Bosari, **Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano,**
- Prof. Carlo Agostoni**, Università degli Studi, IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano**
- Prof. Lorenza Lazzari, **Department of Regenerative Medicine, Fondazione IRCCS Cà Granda Ospedale Maggiore Policlinico di Milano**
- Prof. Rosaria Giordano**, Department of Regenerative Medicine, Fondazione IRCCS Cà Granda Ospedale Maggiore Policlinico di Milano**
- Dr. Massimo Aureli, **Università Degli Studi di Milano, Ospedale san Raffaele, Milano**
- Prof. Michela Deleidi, **Department of Neurodegenerative Diseases, University of Tübingen**
- Prof. Cristina Barlassina – **Dept. Health Sciences, Università degli Studi di Milano**
- Prof. Irene Cettin, **Centro di Ricerche Fetali Giorgio Pardi, Università degli Studi di Milano - Polo Universitario Ospedale L. Sacco di Milano**
- Prof. Paola Rossi**, Dipartimento di Scienze Fisiologiche e Farmacologiche cellulari e molecolari- Sezione di Fisiologia dell'Università di Pavia**
- Prof. Umberto Galderisi – **Dipartimento di Medicina Sperimentale, Università degli Studi di Milano**
- Prof. Rita Maiavacca – **UOS Laboratorio di Biochimica, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico**
- Dr. Fabio Triulzi **-** UOC Neuroradiologia - **Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico**
- Dr. Alessandro Sillani **-** UOC Neuroradiologia - **Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico**
- Dott.ssa Clara Sina **-** UOC Neuroradiologia - **Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico**

- Dr. Umberto Giovanella – **CNR –ISMAC**
- Dr. Anna Villa - **Human Genome Lab, Humanitas Clinical and Research Center, Milano**
- Prof. Agostino Cortelezzi – **UOC Ematologia1 e Centro Trapianti di Midollo, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano**
- Prof. Enzo Nisoli, **Center for Study and Research on Obesity, Department of Medical Biotechnology and Translational Medicine, University of Milan, Milan, Italy;**
- Prof. Dario Parazzoli**, Imaging Facility IFOM Foundation – The FIRC Institute of Molecular Oncology Foundation, Milan, Italy**
- Prof. Stefano Campaner**, Center for Genomic Science of IIT@SEMM; Istituto Italiano di Tecnologia (IIT); Milan, Italy**
- Prof. Thierry Vanderdriessche – **VRIJE Universiteit Brussel**
- Dr. Laura Porretti – **Clinical Chemistry and Microbiology Laboratory, Flow Cytometry and Experimental Hepatology Service, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano**
- Prof. Giorgio Roberto Merlo – **Dept. Biotecnologie Molecolari Scienze della Salute, Università degli Studi di Torino**
- Prof. Luciano Conti – **Centro di Biologia Integrata CIBIO, Università degli Studi di Trento**
- Prof. Alessandro Quattrone**, Director of CiBio, University of Trento**
- Prof. Elena Cattaneo**, Department of Biosciences and Centre for Stem cell Research, Università degli Studi di Milano**
- Prof. Giovanna Cantarella**, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano**
- Prof. Mauro Pluderi **, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano**
- Prof. Nadia Grimoldi**, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico di Milano**
- Prof. Paolo Vezzoni**,Unità Operativa di Supporto (UOS) dell'Istituto di Ricerca Genetica e Biomedica (IRGB) del CNR**.
- Prof. Marina Bouchè**, Unit of Histology, and IIM, Sapienza University, DAHFMO, Rome, Italy**
- Prof. Davide Gabellini, **Dulbecco Telethon Institute and Division of Regenerative Medicine, San Raffaele Scientific Institute, Milan**
- Prof. Franco Rustichelli**, Dipartimento di Scienze Cliniche e Odontostomatologiche, Sezione di Biochimica, Biologia e Fisica, Università Politecnica delle Marche, Ancona, Italy**
- Prof. Silvia Della Bella**, Lab of Clinical and Experimental Immunology, Humanitas Clinical and Research Center, Rozzano (MI), Italy, Department of Medical Biotechnologies and Translational Medicine, University of Milan, Milan, Italy**
- Prof. Aldo Pagano**, Department of Experimental Medicine,UniversityofGenoa,Genoa,Italy, IRCCS Azienda Ospedaliera Universitaria SanMartino-IST, Genoa, Italy**
- Prof. Francesco Meinardi **, Università di Milano Bicocca**

- Prof. Jose F Rodriguez-Matas-, **Department "Giulio Natta"Politecnico di Milano, Italy**
- Prof. Jerry Mendell – **Nationwide Children's Hospital, Columbus, USA**
- Prof. Leonard Petrucelli – **Department of Neurology, Mayo Clinic, Florida, USA**
- Prof. Simone Guglielmetti - **Faculty of Agriculture and Food Sciences, University of Milan, Italy**
- Dr Barbara Cassani - **Humanitas Clinical and Research Center, National Research Council IRGB UOS Milan**
- Dr. Domenico Aquino – **Istituto Besta, Milan**
- Prof. C. Covino – **ALEMBIC, Ospedale San Raffele**
- Prof. G. Pajardi – **Dip. Scienze Cliniche e di Comunità, Università degli Studi di Milano**
- Dr Y. D'Alessandra **– Unità di Immunologia e genomica funzionale, Centro Cardiologico Monzino IRCCS**
- Prof. S. Biressi **–CIBIO, Università degli Studi di Trento**
- Prof.ssa M. Raimondi **- Dipartimento di Chimica, Materiali e Ingegneria Chimica "Giulio Natta", Politecnico di Milano**
- Prof.ssa M.G. Bruzzone **– UOC Neuroradiologia, Istituto Neurologico Besta**
- Prof. T. Patridge **– Children's National Medical Center, University of Washington, USA**
- Prof F. Rossi **- University of British Columbia**
- Prof. Rudnicki - **Ottawa Hospital Research Institute**
- Prof A. Caplan – **Case Western Researve University**
- Prof. E. Hoffman - **George Washington University School of Medicine**
- Prof. E. MacNally – **Northwestern University**
- Prof. T. Rando – **Stanford University**
- Prof Blau - **Harvard University**
- Prof. Robert Bryson-Richardson - **School of Biological Sciences, Monash University, Melbourne, Australia**
- Dott.ssa Cinzia Gellera – **IRCCS Istituto C. Besta, Milano**
- Dr. Franco Taroni – **IRCCS Istituto C. Besta, Milano**
- Prof. Giuseppe Lauria– **IRCCS Istituto C. Besta, Milano**
- Dott.ssa Giacomina Rossi – **IRCCS Istituto C. Besta, Milano**
- Dr. Fabrizio Tagliavini– **IRCCS Istituto C. Besta, Milano**
- Dr. Fabio Blandini – **IRCCS Istituto Mondino Pavia**

- Dr. Cristina Cereda – **IRCCS Istituto Mondino Pavia**
- Dr. Christian Lunetta – **Centro Clinico Nemo**
- Dott.ssa Valeria Sansone – **Centro Clinico Nemo**
- Dr. Luca Persani **– Dipartimento di Endocrinologia IRCCS Istituto Auxologico Italiano**
- Prof.ssa Palma Finelli **– Laboratorio di Citogenetica IRCCS Istituto Auxologico Italiano**
- Dott.ssa daniela Giardino **– Laboratorio di Citogenetica IRCCS Istituto Auxologico Italiano**
- Dr. Luigi Sironi **– Dipartimento di Farmacologia Università di Milano – CEND**
- Dr. A.E. Rigamonti **– Dipartimento di Farmacologia Università di Milano – CEND**
- Prof. Massimo Filippi – **Neuroimaging Research Unit and Department of Neurology, Istitute of Experimental Neurology – San Raffaele Scientific Institute, Milan**
- Dott.ssa Federica Agost**a** – **Neuroimaging Research Unit and Department of Neurology, Istitute of Experimental Neurology San Raffaele Scientific Institute, Milan**
- Prof. Giancarlo Comi **- Neuroimaging Research Unit and Department of Neurology, Istitute of Experimental Neurology San Raffaele Scientific Institute, Milan San Raffaele Milano**
- Prof. Andrea Falini **– Division of Neuroscience and Department of Neuroradiology, Vita-Salute University and San Raffaele Scientific Institute, Milan**
- Dr. Emanuele Buratti – **Laboratory of Molecular Pathology International Centre for Genetic Engineering and Biotechnology (ICGEB), Trieste**
- Dr. Francesco Bifari **– Dipartimento di Biotecnologie Mediche e Medicina Traslazionale, Università degli Studi di Milano**
- Dr. Marco Feligioni **– Centro EBRI Roma**
- Prof. Emanuele Borgonovo **– Dipartimento di Scienze Decisionali Università Commerciale "L. Bocconi" Milano**
- Dott.ssa Raffaella Piccarreta **- Dipartimento di Scienze Decisionali Università Commerciale "L. Bocconi" Milano**
- Prof. Robert H. Brown – **University of Massachussetts Medical School, Department of Neurology Worcester, MA , USA**
- Prof. Markus Weber **– Dipartimento di Neurologia Università di St. Gallen, Svizzera**
- Prof. Sharon Abrahams **– EUAN Mac Donald Centre for Motor Neurone Disease Research University of Edimburg – UK**
- Prof C. Becchio – **IIT Genova**
- Dott. FM Santorelli- **Fondazione Stella Maris, Pisa**
- Dott IAN HARDING - , **Monash Institute of Cognitive and Clinical Neurosciences, Monash University, Melbourne, Australia**

- Prof . Dallapiccola- **IRCCS Bambino Gesù Roma**
- Prof E. I Rugarli- **Institute for Genetics CECAD Research Center**
- Joseph-Stelzmann **University of Cologne -Str. 26 50931 Köln Germany**
- Prof Luca De Gioia - **Department of Biotechnology and Biosciences, University of Milan-Bicocca, 20126 Milan, Italy.**
- Prof. Mario Clerici **-Department of Physiopathology and Transplantation, University of Milan, 20090 Milan, Italy. and Don C. Gnocchi Foundation ONLUS, IRCCS, 20148 Milan, Italy.**
- Edward J Hollox - **Department of Genetics, University of Leicester, Leicester LE2 1TE, UK**
- Prof Giuseppe Bianchi **- Nephrology and Dialysis Unit, San Raffaele Scientific Institute, University Vita Salute San Raffaele, Milan, Ita**
- Nasser M. Al-Daghri - **Biomarker research program, Biochemistry Department, College of Science, King Saud University, Riyadh 11451, Kingdom of Saudi Arabia (KSA) and Prince Mutaib Chair for Biomarkers of Osteoporosis, Biochemistry Department, College of science, King Saud University, Riyadh, KSA**
- Dott. Franca Guerini - **Don C. Gnocchi Foundation ONLUS, IRCCS, 20100 Milan, Italy**
- Dott. Mara Biasin **- Department of Biomedical and Clinical Sciences, University of Milan, 20157 Milan, Italy**
- Prof. Roberto de Franchis **- IBD Unit, Chair of Gastroenterology, Luigi Sacco University Hospital, 20157 Milan**
- Dott. Sergio Lo Caputo - **S. Maria Annunziata Hospital, 50122 Florence, Italy**
- Dott. Rosanna Asselta - **Dipartimento di Biotecnologie Mediche e Medicina Traslazionale, Università degli Studi di Milano, Milano, Italy.**
- Dott. Juan Antonio Pineda **- Infectious Diseases and Microbiology Clinical Unit. Valme Hospital, Seville, Spain**
- Dott. Antonio Rivero-Juarez - **Maimonides Institut for Biomedical Research (IMIBIC)-Reina Sofia Universitary Hospital-University of Cordoba, Spain**
- Dott. Antonio Caruz - **Immunogenetics Unit, Department of Experimental Biology, University of Jaen, Jaen, Spain**
- Dott. Manuel Comabella - **Hospital Universitari Vall d´Hebron (HUVH). Barcelona, Spain**
- Dott. Matteo Fumagalli - **UCL Genetics Institute, Department of Genetics, Evolution and Environment, University College London, Gower Street, London WC1E 6BT, United Kingdom**
- Dott. Matteo Cereda - **Department of Experimental Oncology, European Institute of Oncology (IEO), 20139 Milan, Italy**
- Prof. Jernej Ule **- Department of Molecular Neuroscience, UCL Institute of Neurology, Queen Square, London WC1N 3BG, UK**

Molecular Neurobiology
https://doi.org/10.1007/s12035-019-1554-x

Advances, Challenges, and Perspectives in Translational Stem Cell Therapy for Amyotrophic Lateral Sclerosis

Elena Abati[1] · Nereo Bresolin[1,2] · Giacomo Comi[1,2] · Stefania Corti[1,2,3]

Received: 29 December 2018 / Accepted: 13 March 2019

Abstract
Finding an effective therapeutic approach is a primary goal for current and future research for amyotrophic lateral sclerosis (ALS), a fatal neurological disease characterized by degeneration and loss of upper and lower motor neurons. Transplantation approaches based on stem cells have been attempted in virtue of their potential to contrast simultaneously different ALS pathogenic aspects including either the replacement of lost cells or the protection of motor neurons from degeneration and toxic microenvironment. Here, we critically review the recent translational research aimed at the assessment of stem cell transplantation safety and feasibility in the treatment of ALS. Most of these efforts aim to exert a neuroprotective action rather than cell replacement. Critical aspects that emerge in these studies are the need for the identification of the most effective therapeutic cell source (mesenchymal stem cells, immune, or neural stem cells), the definition of the optimal injection site (cortical area, spinal cord, or muscles) with a suitable administration protocol (local or systemic injection), and the analysis of therapeutic mechanisms, which are necessary steps in order to overcome the hurdles posed by previous in vivo human studies. New perspectives will also be offered by the increasing number of induced pluripotent stem cell-based therapies that are now being tested in clinical trials. A thorough analysis of recently completed trials is the foundation for continued progress in cellular therapy for ALS and other neurodegenerative disorders.

Keywords Amyotrophic lateral sclerosis · Motor neuron · Mesenchymal stem cells · Regulatory T cells · Neural stem cells · Induced pluripotent stem cells · Stem cell transplantation

Introduction

Amyotrophic lateral sclerosis (ALS) is a devastating neurodegenerative disorder which provokes the progressive degeneration of upper and lower motor neurons [1]. The disease starts with insidious focal muscle weakness, frequently in one hand, and then disseminates relentlessly to affect most skeletal muscles, leading to complete paralysis. Death occurs approximately 3–5 years after symptoms onset, mainly because of respiratory failure [2]. So far, no therapy was shown to provide a substantial clinical benefit for ALS patients. Up to now, FDA has approved only two treatments, riluzole, which prolongs median survival by about only 2 to 3 months [3] and edaravone, which slightly reduces the rate of decline in the early stages of disease [4–6].

Since ALS still represents a devastating disease with a significant impact on patients, caregivers, and society, effective treatments are urgently needed. The lack of therapeutic tools could be ascribed, at least in part, to incomplete knowledge of the pathogenetic basis of ALS motor neuronal degeneration. In this context, why is stem cell therapy so fascinating and potentially useful for ALS? Stem cell transplantation could potentially tackle the multifaceted and largely unknown ALS disease pathogenesis through multiple mechanisms, such as by replacing lost or diseased cells, by introducing factors that will provide neuroprotective effects or by modulating the pathogenetic pathways linked to toxic microenvironment [7]. Regarding paracrine delivery, growth factors have been shown to exert neuroprotective effects when delivered in a variety of motor neuron models [8]. However,

✉ Stefania Corti
stefania.corti@unimi.it

[1] Department of Pathophysiology and Transplantation (DEPT), Dino Ferrari Centre, Neuroscience Section, University of Milan, Milan, Italy

[2] Neurology Unit, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

[3] Department of Pathophysiology and Transplantation (DEPT), Dino Ferrari Centre, Neuroscience Section, Neurology Unit, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy

Published online: 26 March 2019

Received: 2 September 2019 | Revised: 31 October 2019 | Accepted: 9 December 2019

DOI: 10.1111/tid.13236

CASE REPORT AND REVIEW OF THE LITERATURE

WILEY

Herpes Simplex virus type 2 myeloradiculitis with a pure motor presentation in a liver transplant recipient

Elena Abati[1] | Delia Gagliardi[1] | Daniele Velardo[2] | Megi Meneri[2] | Giorgio Conte[3] | Claudia Cinnante[3] | Nereo Bresolin[1,2] | Giacomo Comi[1,2] | Stefania Corti[1,2]

[1]Department of Pathophysiology and Transplantation (DEPT), Dino Ferrari Centre, Neuroscience Section, University of Milan, Milan, Italy

[2]Neurology Unit, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

[3]Neuroradiology Unit, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

Correspondence
Stefania Corti, Department of Pathophysiology and Transplantation (DEPT), Dino Ferrari Centre, Neuroscience Section, Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy.
Email: stefania.corti@policlinico.mi.it

Abstract

In this case report, we describe the first PCR-confirmed case of HSV2 myeloradiculitis with a purely motor presentation, occurring in a 68-year-old liver transplant recipient. The patient reported ascending weakness with no sensory nor sphincteric symptoms, thereby resembling acute demyelinating inflammatory neuropathy, or Guillain-Barré syndrome. HSV2 was detected in cerebrospinal fluid by PCR, and the patient was successfully treated with intravenous Acyclovir.

KEYWORDS

ascending weakness, Elsberg syndrome, Herpes simplex virus type 2, HSV2 infection, liver transplant, myeloradiculitis

1 | INTRODUCTION

Elsberg syndrome is an extremely rare lumbosacral infectious disorder characterized by involvement of the *cauda equina* and associated with Herpes simplex virus type 2 (HSV2) reactivation or, infrequently, primary infection.[1,2] Only 12 PCR-confirmed cases have been reported to date,[2-6] and they all featured sphincterial and/or sensorimotor symptoms, while a purely motor presentation is not typical and occurs rarely. HSV2 disseminated infection is known to occur in transplant recipients as a result of reactivation of dormant virus or, rarely, in a donor-derived fashion, although a neurologic involvement has never been described.[7,8] We hereby describe the case of a 68-year-old immunocompromised man with progressive motor impairment due to HSV2 central nervous system (CNS) infection.

2 | CLINICAL CASE

The patient was admitted to our Emergency Department for a six-day history of progressive limb weakness. He reported a recent gastrointestinal illness with watery diarrhea, which lasted 5 days and disappeared after treatment with metronidazole. Six days after the resolution of the diarrhea, he started to experience progressive weakness of the lower limbs, with impairment in walking and climbing stairs, associated with lumbar pain and fatigue. He denied both sensory and autonomic or sphincteric deficits. Four days later, weakness of the hands appeared.

His medical history was relevant for a liver transplant following acute HBV hepatitis at the age of 60. Seven years later, he was diagnosed with post-transplant large B-cell intestinal lymphoma and treated with jejunoileal resection. Since then, the patient had been receiving immunosuppressive treatment with tacrolimus and mofetil mycophenolate, and antiviral treatment with anti-HbS immunoglobulins and entecavir.

Upon admittance, physical examination was unremarkable while neurological examination showed symmetric upper and lower limb weakness with proximal predominance, steppage gait, normal sensory testing, and no sphincterial involvement. The remaining neurological examination, including cranial nerves, muscle tone, deep tendon reflexes (DTRs), and plantar cutaneous response, was normal. Blood tests were normal, except for leukocytosis (12.000 wbc/mm3) and elevated C-reactive protein (6 mg/dL).

Abati and Gagliardi equally contributed to the work.

Molecular Neurobiology
https://doi.org/10.1007/s12035-019-1533-2

Disease Modeling and Therapeutic Strategies in CMT2A: State of the Art

Kordelia Barbullushi[1] · Elena Abati[1] · Federica Rizzo[1] · Nereo Bresolin[1,2] · Giacomo P. Comi[1,2] · Stefania Corti[1,2]

Received: 13 November 2018 / Accepted: 19 February 2019

Abstract

Mitofusin 2 (MFN2) is a protein of the mitochondrial outer membrane that belongs to a family of highly conserved dynamin-related GTPases. It is implicated in several intracellular pathways; however, its main role is the regulation of mitochondrial dynamics, in particular mitochondrial fusion. Mutations in *MFN2* are associated with Charcot–Marie–Tooth disease type 2A (CMT2A), a neurological disorder characterized by a wide spectrum of clinical features, primarily a motor sensory neuropathy. The cellular and molecular mechanisms by which *MFN2* mutations lead to neuronal degeneration are largely unknown, and there is currently no cure for patients. Here, we present the most recent in vitro and in vivo models of CMT2A and the more promising therapeutic approaches under development. These models and therapies may represent relevant tools for the study and recovery of defective mitochondrial dynamics that seem to play a significant role in the pathogenesis of other more common neurodegenerative diseases.

Keywords Mitofusin2 · Charcot–Marie–Tooth disease type 2 · Hereditary neuropathies · Mitochondrial diseases · Molecular therapy · Gene therapy · Mitofusin agonists

Abbreviation

CMT	Charcot–Marie–Tooth disease
MCNV	Motor nerve conduction velocity
MNs	Motor neurons
SNs	Sensory neurons
IPSCs	Induced pluripotent stem cells
ASO	Antisense oligonucleotide
RNAi	RNA interference
CRISPR	Clustered regularly interspersed short palindromic repeats
Cas9	Caspase 9
KO	Knockout

Introduction

Charcot–Marie–Tooth 2A (CMT2A) is an axonal peripheral neuropathy that belongs to the group of Charcot–Marie–Tooth diseases (CMT) [1]. CMT are part of a wide range of inherited sensory motor neuropathies with different clinical presentations and various genetic causes. The main clinical features include distal weakness, sensory loss, gait impairment, and foot deformities in the context of a very heterogeneous disease [2]. The first classification of CMT is based on the inheritance mode (autosomal dominant, autosomal recessive, and X-linked); however, the most used classification relies on the motor nerve conduction velocity (MNCV), thereby distinguishing a demyelinating form with a MNCV < 38 m/s (CMT1), an axonal form with a MNCV > 38 m/s (CMT2), and an intermediate form with a MNCV 25–45 m/s [3].

CMT disease is the most common inherited neuropathy and affects 1 in 2500 individuals [4]. CMT1A presents the highest prevalence and is mainly related to mutations in the gene of peripheral myelin protein 22 (*PMP22*) [5]. With respect to CMT2, the most frequent form is CMT2A (33% of CMT2 cases) [6], which is caused by mutations in the mitofusin 2 (*MFN2*) gene. Noteworthy, CMT2A accounts

✉ Stefania Corti
stefania.corti@unimi.it

[1] Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Via Francesco Sforza 35, 20122 Milan, Italy

[2] Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurology Unit, Via Francesco Sforza 35, 20122 Milan, Italy

Published online: 04 March 2019

Molecular Neurobiology
https://doi.org/10.1007/s12035-019-01820-5

Molecular Approaches for the Treatment of Pompe Disease

Anita Sofia Bellotti[1] · Luca Andreoli[1] · Dario Ronchi[1] · Nereo Bresolin[1,2] · Giacomo P. Comi[1,3] · Stefania Corti[1,2]

Received: 26 June 2019 / Accepted: 23 October 2019

Abstract
Glycogen storage disease type II (GSDII, Pompe disease) is a rare metabolic disorder caused by a deficiency of acid alpha-glucosidase (GAA), an enzyme localized within lysosomes that is solely responsible for glycogen degradation in this compartment. The manifestations of GSDII are heterogeneous but are classified as early or late onset. The natural course of early-onset Pompe disease (EOPD) is severe and rapidly fatal if left untreated. Currently, one therapeutic approach, namely, enzyme replacement therapy, is available, but advances in molecular medicine approaches hold promise for even more effective therapeutic strategies. These approaches, which we review here, comprise splicing modification by antisense oligonucleotides, chaperone therapy, stop codon readthrough therapy, and the use of viral vectors to introduce wild-type genes. Considering the high rate at which innovations are translated from bench to bedside, it is reasonable to expect substantial improvements in the treatment of this illness in the foreseeable future.

Keywords GSDII · Pompe disease · Alpha-glucosidase (GAA) · Therapy · Gene therapy · Molecular therapy · Antisense oligonucleotides

Introduction

Pompe Disease

Glycogenosis type II (GSDII), or Pompe disease, is a rare autosomal recessive disease caused by a deficiency of the enzyme solely responsible for glycogen degradation within lysosomes: acid maltase or acid alpha-glucosidase (GAA). Over time, the progressive accumulation of glycogen alters cellular architecture, causing a loss of function and eventually necrosis. Although it has long been considered a disease that mainly affects striated muscular tissue with a disproportionate involvement of respiratory muscles, GSDII is multisystemic: glycogen accumulates in all tissues and organs, particularly in the skeletal muscle, central nervous system, heart and brain (the latter are almost exclusively affected by the early-onset form of the disease), causing not only a reduction in motor function and important respiratory deficits, the main cause of death in patients with Pompe disease, but also arrhythmias, dysphagia, incontinence, gastrointestinal symptoms, and several other problems [1–3].

Pompe disease has been documented in most ethnicities, with an incidence ranging from 1:14,000 (in African populations and African-Americans) [4] to 1:238,000 in Europe [5]. The advent of newborn screening (NBS) and the increasing availability of genetic testing for at-risk patients contribute to more precocious diagnosis and are uncovering the real incidence of the disease; for example, in Taiwan, the global incidence of GSDII based on NBS programs is estimated to be 1:17,000 [6].

Patients are divided into two main groups based on the age at disease onset: early-onset Pompe disease (EOPD) and late-onset Pompe disease (LOPD); however, presentation is extremely varied and correlates only partially with residual enzyme activity levels and the mutations carried by patients [7]. Early-onset Pompe disease (EOPD) is defined as GSDII arising before 12 months of age and more typically manifests during the first two months of life; patients present with severe muscular hypotonia and hypertrophic cardiomyopathy, which

✉ Stefania Corti
Stefania.corti@unimi.it

[1] Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Milan, Italy

[2] Neurology Unit, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

[3] Neuromuscular and Rare Diseases Unit, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

Published online: 12 November 2019

Received: 22 April 2019 | Revised: 14 September 2019 | Accepted: 10 November 2019

DOI: 10.1111/jcmm.14874

REVIEW

WILEY

Spinal muscular atrophy with respiratory distress type 1: Clinical phenotypes, molecular pathogenesis and therapeutic insights

Matteo Saladini[1] | Monica Nizzardo[2] | Alessandra Govoni[2] | Michela Taiana[2] | Nereo Bresolin[1,2] | Giacomo P. Comi[1,3] | Stefania Corti[1,2]

[1]Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Milan, Italy

[2]Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurology Unit, Milan, Italy

[3]Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neuromuscular and rare diseases unit, Milan, Italy

Correspondence
Stefania Corti, Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurology Unit, Via Francesco Sforza 35, 20122, Milan, Italy.
Email: stefania.corti@unimi.it

Funding information
This work was partially supported by the Italian Ministry of Health (MoH) with grants to MN, NB, GPC and N. B. Cariplo grant to MN is gratefully acknowledged. Grant Number:GR-2016-02362377

Abstract

Spinal muscular atrophy with respiratory distress type 1 (SMARD1) is a rare autosomal recessive neuromuscular disorder caused by mutations in the *IGHMBP2* gene, which encodes immunoglobulin μ-binding protein 2, leading to progressive spinal motor neuron degeneration. We review the data available in the literature about SMARD1. The vast majority of patients show an onset of typical symptoms in the first year of life. The main clinical features are distal muscular atrophy and diaphragmatic palsy, for which permanent supportive ventilation is required. No effective treatment is available yet, but novel therapeutic approaches, such as gene therapy, have shown encouraging results in preclinical settings and thus represent possible methods for treating SMARD1. Significant advancements in the understanding of both the SMARD1 clinical spectrum and its molecular mechanisms have allowed the rapid translation of preclinical therapeutic strategies to human patients to improve the poor prognosis of this devastating disease.

KEYWORDS
gene therapy, IGHMBP2, motor neuron disease, Spinal muscular atrophy with respiratory distress type 1

1 | INTRODUCTION

Autosomal recessive spinal muscular atrophy with respiratory distress type 1 (SMARD1) is a form of spinal muscular atrophy with severe diaphragmatic involvement that causes respiratory distress. This condition is due to autosomal recessive mutations in the *IGHMBP2* gene, which is located on chromosome 11q13.2-q13.4.[1,2] Mellins, considering this mutation a variant of spinal muscular atrophy (SMA) 5q with respiratory onset, provided the first description of this condition in 1974, and it was not recognized as a separate clinical entity until 1996.[3,4] The actual prevalence of SMARD1 is unknown, but diaphragmatic paralysis is observed in approximately 1% of patients with an early onset of the clinical features of spinal muscle atrophy and an estimated incidence of 1/100 000.[5] The main clinical feature is the onset of respiratory distress requiring mechanical ventilation between the ages of 6 weeks and 6 months. The clinical symptoms rapidly progress in the first years of life, with distal limb muscular atrophy extending to proximal regions. The overall prognosis is poor, and progressive autonomic nervous system dysfunction also develops in association with the progressive worsening of motor functions in affected children. In fact, there are no approved treatments for SMARD1.[6]

PLOS ONE

RESEARCH ARTICLE

Long-term natural history data in Duchenne muscular dystrophy ambulant patients with mutations amenable to skip exons 44, 45, 51 and 53

Claudia Brogna[1,2‡], Giorgia Coratti[1,2‡], Marika Pane[2‡], Valeria Ricotti[3,4], Sonia Messina[5,6], Adele D'Amico[7], Claudio Bruno[8], Gianluca Vita[6], Angela Berardinelli[9], Elena Mazzone[2], Francesca Magri[10], Federica Ricci[11], Tiziana Mongini[11], Roberta Battini[12,13], Luca Bello[14], Elena Pegoraro[14], Giovanni Baranello[15], Stefano C. Previtali[16], Luisa Politano[17], Giacomo P. Comi[10], Valeria A. Sansone[18], Alice Donati[19], Enrico Bertini[7], Francesco Muntoni[3], Nathalie Goemans[20], Eugenio Mercuri[1,2]*, on behalf on the International DMD group[¶]

1 Pediatric Neurology, Department of Woman and Child Health and Public Health, Child Health Area, Università Cattolica del Sacro Cuore, Rome, Italy, **2** Centro Clinico Nemo, Fondazione Policlinico Universitario Agostino Gemelli IRCCS, Rome, Italy, **3** Dubowitz Neuromuscular Centre, UCL & Great Ormond Street Hospital, London, United Kingdom, **4** NIHR Great Ormond Street Hospital Biomedical Research Centre, London, United Kingdom, **5** Department of Clinical and Experimental Medicine, University of Messina, Messina, Italy, **6** Nemo SUD Clinical Centre, University Hospital "G. Martino", Messina, Italy, **7** Department of Neurosciences, Unit of Neuromuscular and Neurodegenerative Disorders, Bambino Gesù Children's Hospital, Rome, Italy, **8** Center of Myology and Neurodegenerative Disorders, Istituto Giannina Gaslini, Genoa, Italy, **9** Child Neurology and Psychiatry Unit, "Casimiro Mondino" Foundation, Pavia, Italy, **10** Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Dino Ferrari Centre, Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy, **11** Neuromuscular Center, AOU Città della Salute e della Scienza, University of Torino, Torino, Italy, **12** Department of Developmental Neuroscience, Stella Maris Institute, Pisa, Italy, **13** Department of Clinical and Experimental Medicine, University of Pisa, Pisa, Italy, **14** Department of Neurosciences, University of Padua, Padua, Italy, **15** Fondazione IRCCS Istituto Neurologico Carlo Besta, Milan, Italy, **16** Division of Neuroscience, IRCCS San Raffaele Scientific Institute, Milan, Italy, **17** Dipartimento di Medicina Sperimentale, Seconda Università di Napoli, Napoli, Italy, **18** The NEMO Center in Milan, Neurorehabilitation Unit, University of Milan, ASST Niguarda Hospital, Milan, Italy, **19** Metabolic Unit, A. Meyer Children's Hospital, Florence, Italy, **20** Department of Child Neurology, University Hospitals Leuven, Leuven, Belgium

‡ These authors are Joint First Authors on this work.
¶ Membership of the International DMD group is provided in the Acknowledgments.
* eugeniomaria.mercuri@unicatt.it

OPEN ACCESS

Citation: Brogna C, Coratti G, Pane M, Ricotti V, Messina S, D'Amico A, et al. (2019) Long-term natural history data in Duchenne muscular dystrophy ambulant patients with mutations amenable to skip exons 44, 45, 51 and 53. PLoS ONE 14(6): e0218683. https://doi.org/10.1371/journal.pone.0218683

Editor: Antonio Musaro, University of Rome La Sapienza, ITALY

Received: March 29, 2019

Accepted: June 6, 2019

Published: June 25, 2019

Data Availability Statement: All relevant data are within the manuscript.

Funding: The study was partially founded by the Italian Telethon (GUP 15011) to EM and partially funded by the Association Française contre les Myopathies (AFM) for the collection and analysis of the data from the patients recruited in London (Grant N. 17100). This study was also funded by the International MDEX Consortium for the participation to the AFM funded natural history

Abstract

Introduction

The aim of this international collaborative effort was to report 36-month longitudinal changes using the 6MWT in ambulant patients affected by Duchenne muscular dystrophy amenable to skip exons 44, 45, 51 or 53.

Materials and methods

Of the 92 patients included in the study, 24 had deletions amenable to skip exon 44, 27 exon 45, 18 exon 51, and 28 exon 53. Five patients with a single deletion of exon 52 were counted in both subgroups skipping exon 51 and 53.

frontiers in Neurology

CASE REPORT
published: 13 August 2019
doi: 10.3389/fneur.2019.00823

Ophthalmoplegia Due to Miller Fisher Syndrome in a Patient With Myasthenia Gravis

Roberta Brusa[1†], Irene Faravelli[2†], Delia Gagliardi[2], Francesca Magri[1], Filippo Cogiamanian[3], Domenica Saccomanno[1], Claudia Cinnante[4], Eleonora Mauri[2], Elena Abati[2], Nereo Bresolin[2], Stefania Corti[2] and Giacomo Pietro Comi[2]*

[1] Neurology Unit, Department of Pathophysiology and Transplantation, Dino Ferrari Center, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, [2] Neurology Unit, Department of Pathophysiology and Transplantation, Dino Ferrari Center, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy, [3] Neuropathophysiology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, [4] Neuroradiology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

OPEN ACCESS

Edited by:
Angelo Schenone,
University of Genoa, Italy

Reviewed by:
Carmelo Rodolico,
University of Messina, Italy
Antonino Uncini,
G. d'Annunzio University of Chieti and Pescara, Italy

***Correspondence:**
Giacomo Pietro Comi
giacomo.comi@unimi.it

[†] These authors have contributed equally to this work

Specialty section:
This article was submitted to Neuromuscular Diseases, a section of the journal Frontiers in Neurology

Received: 14 June 2019
Accepted: 17 July 2019
Published: 13 August 2019

Citation:
Brusa R, Faravelli I, Gagliardi D, Magri F, Cogiamanian F, Saccomanno D, Cinnante C, Mauri E, Abati E, Bresolin N, Corti S and Comi GP (2019) Ophthalmoplegia Due to Miller Fisher Syndrome in a Patient With Myasthenia Gravis. Front. Neurol. 10:823. doi: 10.3389/fneur.2019.00823

Here, we describe a 79-year-old man, admitted to our unit for worsening diplopia and fatigue, started a few weeks after an episode of bronchitis and flu vaccination. Past medical history includes myasthenia gravis (MG), well-controlled by Pyridostigmine, Azathioprine, and Prednisone. During the first days, the patient developed progressive ocular movement abnormalities up to complete external ophthalmoplegia, severe limb and gait ataxia, and mild dysarthria. Deep tendon reflexes were absent in lower limbs. Since not all the symptoms were explainable with the previous diagnosis of myasthenia gravis, other etiologies were investigated. Brain MRI and cerebrospinal fluid analysis were normal. Electromyography showed a pattern of predominantly sensory multiple radiculoneuritis. Suspecting Miller Fisher syndrome (MFS), the patient was treated with plasmapheresis with subsequent clinical improvement. Antibodies against GQ1b turned out to be positive. MFS is an immune-mediated neuropathy presenting with ophthalmoplegia, ataxia, and areflexia. Even if only a few cases of MFS overlapping with MG have been described so far, the coexistence of two different autoimmune disorders can occur. It is always important to evaluate possible differential diagnosis even in case of known compatible diseases, especially when some clinical features seem atypical.

Keywords: myasthenia gravis, Miller Fisher syndrome, autoimmune diseases, ophthalmoplegia, GQ1b

BACKGROUND

Myasthenia gravis (MG) is an autoimmune disorder of the neuromuscular junction characterized by fatigability and fluctuating weakness of voluntary muscles, leading to symptoms such as diplopia, palpebral ptosis, dysphagia, dyspnea, or limb weakness. Disease exacerbations can be triggered by concomitant infections, vaccination, or administration of certain antibiotics. Antibodies against acetylcholine receptor (AchR), muscle-specific kinase (MuSK), and low-density lipoprotein receptor-related protein 4 (LRP4) may be found on serum. Treatment generally involves symptomatic therapy with acetylcholinesterase inhibitors and immunosuppression, despite initiation of high doses of steroids can precipitate symptoms.

cells

MDPI

Review

iPSCs-Based Neural 3D Systems: A Multidimensional Approach for Disease Modeling and Drug Discovery

Gianluca Costamagna, Luca Andreoli, Stefania Corti and Irene Faravelli *

Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, 20122 Milan, Italy; gianluca.costamagna@unimi.it (G.C.); luca.andreoli95@gmail.com (L.A.); stefania.corti@unimi.it (S.C.)

* Correspondence: irene.faravelli@unimi.it; Tel.: +39-02-5503-3817; Fax: +39-02-5503-3800

Received: 30 September 2019; Accepted: 11 November 2019; Published: 14 November 2019

Abstract: Induced pluripotent stem cells (iPSCs)-based two-dimensional (2D) protocols have offered invaluable insights into the pathophysiology of neurological diseases. However, these systems are unable to reproduce complex cytoarchitectural features, cell-cell and tissue-tissue interactions like their in vivo counterpart. Three-dimensional (3D)-based culture protocols, though in their infancy, have offered new insights into modeling human diseases. Human neural organoids try to recapitulate the cellular diversity of complex tissues and can be generated from iPSCs to model the pathophysiology of a wide spectrum of pathologies. The engraftment of iPSCs into mice models and the improvement of differentiation protocols towards 3D cultures has enabled the generation of more complex multicellular systems. Consequently, models of neuropsychiatric disorders, infectious diseases, brain cancer and cerebral hypoxic injury can now be investigated from new perspectives. In this review, we consider the advancements made in modeling neuropsychiatric and neurological diseases with iPSC-derived organoids and their potential use to develop new drugs.

Keywords: brain organoids; neurological disorders; iPSCs; drug discovery; disease modeling; neural chimera

1. Introduction

Recent technological advances achieved in stem cell research have provided unprecedented means to study the nervous system, both in vitro and in vivo. The enthusiasm for stem cell-based technologies rose with the development of embryonic stem cells (ESCs) cultures, followed by human-induced pluripotent stem cells (iPSCs) and, lately, by ESCs- and iPSCs-derived three-dimensional (3D) culture systems.

Human ESC lines were first isolated in 1998 [1] and differentiation protocols towards multiple tissues were soon designed, aiming to eventually develop allogeneic cell-based therapies to several degenerative diseases. As for neural disease modeling, ESCs were successfully differentiated to neural precursors [2] and many neuronal subtypes, e.g., dopaminergic neurons [3] and motor neurons [4], as well as astrocytes [5], oligodendrocytes [6] and microglia [7]. However, ESCs advantages were offset by the need of genetic manipulation to introduce disease-relevant mutations and their limited supply [8].

Human iPSCs reprogrammed from patients' somatic cells such as fibroblasts and blood cells [9–11] have given new stimuli in many fields of neurobiology: they provided researchers with patient-derived human stem cells offering a more scalable supply for culturing systems and the theoretical possibility of personalized autologous therapies for a wide spectrum of diseases [12]. Moreover, iPSCs can be differentiated into cells able to recapitulate the hallmarks of pathological cells and tissues to develop disease models and test new potential therapies [13]. Many neural diseases have already been

Brain Research xxx (xxxx) xxxx

Contents lists available at ScienceDirect

Brain Research

journal homepage: www.elsevier.com/locate/brainres

Review

Back to the origins: Human brain organoids to investigate neurodegeneration

I. Faravelli[1], G. Costamagna[1], S. Tamanini, S. Corti*

Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

HIGHLIGHTS

- Human organoids allow researchers to investigate brain development and pathology.
- Late disease onset could be linked to alterations during brain development.
- Brain organoids can be used to model neurodegenerative disorders.
- Organoid maturation can be enhanced by *in vivo* transplantation or *in vitro* patterning.

ARTICLE INFO

Keywords:
Brain organoids
Neurological disorders
Neurodegeneration
Disease modelling

ABSTRACT

Neurodegenerative disorders represent a high burden in terms of individual, social and economical resources. No ultimate therapy has been established so far; human brain morphology and development can not be entirely reproduced by animal models, and genomic, metabolic and biochemical differences might contribute to a limited predictive power for human translation. Thus, the development of human brain organoid models holds a wide potential to investigate the range of physiological and pathological features that characterise the early onset of the degeneration. Moreover, central nervous system development has gained a crucial role in the study of the pathogenesis of neurodegenerative disorders. Premature alterations during brain maturation have been related to late disease manifestations; genetic mutations responsible for neurodegeneration have been found in genes highly expressed during neural development. Elucidating the mechanisms triggering neuronal susceptibility to degeneration is crucial for pathogenetic studies and therapeutic discoveries. In the present work, we provide an overview on the current applications of human brain organoids towards studies of neurodegenerative diseases, with a survey on the recent discoveries and a closing discussion on the present challenges and future perspectives.

1. Introduction

Neurodegenerative diseases are a clinically and pathologically heterogeneous group of disorders that affects a specific subset of neurons and whose progression is nowadays inevitable (Przedborski et al., 2003). They can be classified according to the clinical phenotype or to the area most predominantly affected. Alzheimer's disease (AD), frontotemporal dementia (FTD), Parkinson's disease (PD) and Huntington's disease (HD) have a devastating impact on patients and families, representing a high burden in terms of individual, social and economical resources. No ultimate therapy has been established so far, although some of these conditions benefit the availability of drugs slightly able to modify the natural history. These disorders share some common features and pathogenetic mechanisms, which have been identified in protein misfolding and aggregation, altered RNA homeostasis, inflammation and involvement of non-neuronal cells and hindered lysosomal functioning (Katsnelson et al., 2016). No unique mechanism seems to be primarily causative of neurodegeneration, suggesting that the complex synergy of different pathways could play a role. Reliable human *in vitro* models are precious tools for the discovery of specific pathogenic mechanisms and potential therapeutic approaches.

Brain organoid cultures were implemented in 2013 by Lancaster

* Corresponding author at: Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza 35, 20122 Milan, Italy.
E-mail address: stefania.corti@unimi.it (S. Corti).
[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.brainres.2019.146561
Received 2 February 2019; Received in revised form 25 October 2019; Accepted 18 November 2019
0006-8993/

Journal of Neurology (2019) 266:953–959
https://doi.org/10.1007/s00415-019-09219-x

ORIGINAL COMMUNICATION

Muscle pain in mitochondrial diseases: a picture from the Italian network

Massimiliano Filosto[1] · Stefano Cotti Piccinelli[1] · Costanza Lamperti[2] · Tiziana Mongini[3] · Serenella Servidei[4] · Olimpia Musumeci[5] · Paola Tonin[6] · Filippo Maria Santorelli[7] · Costanza Simoncini[8] · Guido Primiano[4] · Liliana Vercelli[3] · Anna Rubegni[7] · Anna Galvagni[1] · Maurizio Moggio[9] · Giacomo Pietro Comi[10] · Valerio Carelli[11] · Antonio Toscano[5] · Alessandro Padovani[1] · Gabriele Siciliano[8] · Michelangelo Mancuso[8]

Received: 9 November 2018 / Revised: 14 December 2018 / Accepted: 28 January 2019 / Published online: 2 February 2019

Abstract

Muscle pain may be part of many neuromuscular disorders including myopathies, peripheral neuropathies and lower motor neuron diseases. Although it has been reported also in mitochondrial diseases (MD), no extensive studies in this group of diseases have been performed so far. We reviewed clinical data from 1398 patients affected with mitochondrial diseases listed in the database of the "Nation-wide Italian Collaborative Network of Mitochondrial Diseases", to assess muscle pain and its features. Muscle pain was present in 164 patients (11.7%). It was commonly observed in subjects with chronic progressive external ophthalmoplegia (cPEO) and with primary myopathy without cPEO, but also—although less frequently—in multisystem phenotypes such as MELAS, MERFF, Kearns Sayre syndrome, NARP, MNGIE and Leigh syndrome. Patients mainly complain of diffuse exercise-related muscle pain, but focal/multifocal and at rest myalgia were often also reported. Muscle pain was more commonly detected in patients with mitochondrial DNA mutations (67.8%) than with nuclear DNA changes (32.2%). Only 34% of the patients showed a good response to drug therapy. Interestingly, patients with nuclear DNA mutations tend to have a better therapeutic response than patients with mtDNA mutations. Muscle pain is present in a significant number of patients with MD, being one of the most common symptoms. Although patients with a myopathic phenotype are more prone to develop muscle pain, this is also observed in patients with a multi system involvement, representing an important and disabling symptom having poor response to current therapy.

Keywords Mitochondrial diseases · Muscle pain · Myalgia · cPEO · Mitochondrial myopathy

✉ Massimiliano Filosto
massimiliano.filosto@unibs.it

[1] Center for Neuromuscular Diseases, Unit of Neurology, ASST Spedali Civili and University of Brescia, Brescia, Italy

[2] Unit of Medical Genetics and Neurogenetics, Fondazione IRCCS Istituto Neurologico 'Carlo Besta', Milan, Italy

[3] Department of Neurosciences Rita Levi Montalcini, University of Torino, Torino, Italy

[4] UOC Neurofisiopatologia Fondazione Policlinico Universitario A. Gemelli IRCCS, Istituto di Neurologia Università Cattolica del Sacro Cuore, Roma, Italy

[5] Department of Clinical and Experimental Medicine, UOC di Neurologia e Malattie Neuromuscolari, University of Messina, Messina, Italy

[6] Neurological Clinic, University of Verona, Verona, Italy

[7] Unit of Molecular Medicine, IRCCS Foundation Stella Maris, Pisa, Italy

[8] Neurological Clinic, University of Pisa, Pisa, Italy

[9] Neuromuscular and Rare Diseases Unit, Department of Neuroscience, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, Milan, Italy

[10] Neurology Unit, Neuroscience Section, Department of Pathophysiology and Transplantation, Dino Ferrari Centre, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy

[11] IRCCS Institute of Neurological Sciences of Bologna, Bellaria Hospital, Bologna, Italy

Experimental Neurology 321 (2019) 113041

Contents lists available at ScienceDirect

Experimental Neurology

journal homepage: www.elsevier.com/locate/yexnr

Research Paper

CSF transplantation of a specific iPSC-derived neural stem cell subpopulation ameliorates the disease phenotype in a mouse model of spinal muscular atrophy with respiratory distress type 1

Giulia Forotti[a,1], Monica Nizzardo[a,1], Monica Bucchia[b], Agnese Ramirez[b], Elena Trombetta[c], Stefano Gatti[d], Nereo Bresolin[a,b], Giacomo Pietro Comi[a,b,2], Stefania Corti[a,b,*,2]

[a] Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
[b] Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Italy
[c] Flow Cytometry Service, Analysis Laboratory, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
[d] Center for Surgical Research, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

ARTICLE INFO

Keywords:
Spinal muscular atrophy with respiratory distress type 1
Neural stem cells
Transplantation

ABSTRACT

Spinal muscular atrophy with respiratory distress type 1 (SMARD1) is a genetic motor neuron disease affecting infants. This condition is caused by mutations in the *IGHMBP2* gene and currently has no cure. Stem cell transplantation is a potential therapeutic strategy for motor neuron diseases such as SMARD1, exerting beneficial effects both by replacing cells and by providing support to endogenous motor neurons. In this work, we demonstrate that human induced pluripotent stem cell (iPSC)-derived neural stem cells (NSCs) selected for the expression of specific markers, namely, Lewis X, CXCR4 and beta 1 integrin, and pretreated with neurotrophic factors and apoptosis/necroptosis inhibitors were able to effectively migrate and engraft into the host parenchyma after administration into the cerebrospinal fluid in a SMARD1 mouse model. We were able to detect donor cells in the ventral horn of the spinal cord and observe improvements in neuropathological features, particularly preservation of the integrity of the motor unit, that were correlated with amelioration of the SMARD1 disease phenotype in terms of neuromuscular function and lifespan. This minimally invasive stem cell approach can confer major advantages in the context of cell-mediated therapy for patients with neurodegenerative diseases.

1. Introduction

Spinal muscular atrophy with respiratory distress type I (SMARD1) is an infantile autosomal recessive genetic disease caused by mutations in the *IGHMBP2* gene, which encodes immunoglobulin-μ-binding protein 2 (IGHMBP2); the mutations reduce the level of this ubiquitous protein (Grohmann et al., 2001; Guenther et al., 2009; Jankowsky et al., 2011). SMARD1 is the second most common form of spinal muscular atrophy after spinal muscular atrophy 5q (SMA 5q). SMARD1 has a very early onset, usually within the first months of life; patients present

Abbreviations: AAV, adeno-associated virus; ALS, atrophic lateral sclerosis; BDNF, brain-derived neurotrophic factor; BTX, bungarotoxin; CSF, cerebrospinal fluid; CXCR4, C-X-C chemokine receptor type 4; DMEM/F12, Dulbecco Modified Eagles Medium/F12; EGF, epidermal growth factor; FACS, fluorescent-activated cell sorting; FGF-2, fibroblast growing factor; GDNF, glial cell-derived neurotrophic factor; GFP, green fluorescent protein; ICV, intracerebroventricular; IGF1, insulin-like growth factor 1; IGHMBP2, immunoglobulin-μ-binding protein 2; iPSCs, induced pluripotent stem cells; LeX, LewisX; MACS, magnetic-activated cell sorting; MAP 2, microtubule-associated protein 2; MNs, motor neurons; Neaa, non-essential aminoacid; NeuN, neuronal nuclei; NF-M, neurofilament-M; NGS, normal goat serum; nmd-mice, neuromuscular degeneration mice; NMJ, neuromuscular junction; NOD/SCID mice, nonobese diabetic/severe combined immunodeficiency mice; NSCs, neural stem cells; OCT4, octamer-binding transcription factor 4; Olig2, oligodendrocyte transcription actor; Pax6, Paired Box 6; SDF-1, stromal cell derived factor-1; SMA5q, spinal muscular atrophy 5q; SMARD1, spinal muscular atrophy with respiratory distress type 1; SSEA-4, stage-specific embryonic antigen-4; wt, wild-type; b-FGF, basic fibroblast growth factor

* Corresponding author at: Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza 35, 20122 Milan, Italy.
E-mail address: stefania.corti@unimi.it (S. Corti).
[1] Co-first authors.
[2] Co-senior authors.

https://doi.org/10.1016/j.expneurol.2019.113041
Received 19 March 2019; Received in revised form 21 July 2019; Accepted 20 August 2019
Available online 21 August 2019
0014-4886/

frontiers in Neurology

CASE REPORT
published: 31 January 2019
doi: 10.3389/fneur.2019.00038

Can Intestinal Pseudo-Obstruction Drive Recurrent Stroke-Like Episodes in Late-Onset MELAS Syndrome? A Case Report and Review of the Literature

Delia Gagliardi[1], Eleonora Mauri[1], Francesca Magri[1], Daniele Velardo[2], Megi Meneri[2], Elena Abati[1], Roberta Brusa[1], Irene Faravelli[1], Daniela Piga[2], Dario Ronchi[1], Fabio Triulzi[3], Lorenzo Peverelli[4], Monica Sciacco[4], Nereo Bresolin[1,2], Giacomo Pietro Comi[1,2], Stefania Corti[1,2,5†] and Alessandra Govoni[2,5*†]*

[1] Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), Dino Ferrari Centre, University of Milan, Milan, Italy, [2] Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, [3] Neuroradiology Unit, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, [4] Neuromuscular and Rare Disease Unit, Department of Neuroscience, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy, [5] Neurology Unit, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), Dino Ferrari Centre, University of Milan, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

OPEN ACCESS

Edited by:
Clara van Karnebeek, University Medical Center Amsterdam, Netherlands

Reviewed by:
Curtis R. Coughlin II, School of Medicine, University of Colorado, United States
Ramona Salvarinova, British Columbia Children's Hospital, Canada

***Correspondence:**
Stefania Corti
stefania.corti@unimi.it
Alessandra Govoni
alessandra.govoni@policlinico.mi.it

†These authors have contributed equally to this work

Specialty section:
This article was submitted to Neurogenetics, a section of the journal Frontiers in Neurology

Received: 13 November 2018
Accepted: 11 January 2019
Published: 31 January 2019

Citation:
Gagliardi D, Mauri E, Magri F, Velardo D, Meneri M, Abati E, Brusa R, Faravelli I, Piga D, Ronchi D, Triulzi F, Peverelli L, Sciacco M, Bresolin N, Comi GP, Corti S and Govoni A (2019) Can Intestinal Pseudo-Obstruction Drive Recurrent Stroke-Like Episodes in Late-Onset MELAS Syndrome? A Case Report and Review of the Literature. Front. Neurol. 10:38. doi: 10.3389/fneur.2019.00038

Mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes (MELAS) syndrome is a maternally inherited mitochondrial disorder that is most commonly caused by the m. 3243A>G mutation in the MT-TL1 mitochondrial DNA gene, resulting in impairment of mitochondrial energy metabolism. Although childhood is the typical age of onset, a small fraction (1–6%) of individuals manifest the disease after 40 years of age and usually have a less aggressive disease course. The clinical manifestations are variable and mainly depend on the degree of heteroplasmy in the patient's tissues and organs. They include muscle weakness, diabetes, lactic acidemia, gastrointestinal disturbances, and stroke-like episodes, which are the most commonly observed symptom. We describe the case of a 50-year-old male patient who presented with relapsing intestinal pseudo-obstruction (IPO) episodes, which led to a late diagnosis of MELAS. After diagnosis, he presented several stroke-like episodes in a short time period and developed a rapidly progressive cognitive decline, which unfortunately resulted in his death. We describe the variable clinical manifestations of MELAS syndrome in this atypical and relatively old patient, with a focus on paralytic ileus and stroke-like episodes; the first symptom may have driven the others, leading to a relentless decline. Moreover, we provide a brief revision of previous reports of IPO occurrence in MELAS patients with the m.3243A>G mutation, and we investigate its relationship with stroke-like episodes. Our findings underscore the importance of recognizing gastrointestinal disturbance to prevent neurological comorbidities.

Keywords: MELAS, mitochondrial disorders, intestinal pseudo-obstruction, stroke-like episodes, gastrointestinal disturbance

International Journal of
Molecular Sciences

MDPI

Review

Diagnostic and Prognostic Role of Blood and Cerebrospinal Fluid and Blood Neurofilaments in Amyotrophic Lateral Sclerosis: A Review of the Literature

Delia Gagliardi [1], Megi Meneri [2], Domenica Saccomanno [2], Nereo Bresolin [1,2], Giacomo Pietro Comi [1,3] and Stefania Corti [1,2,*]

1 Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, 20122 Milan, Italy
2 Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurology Unit, 20122 Milan, Italy
3 Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurmuscular and Rare Diseases Unit, 20122 Milan, Italy
* Correspondence: stefania.corti@unimi.it; Tel.: +39-0255033817

Received: 29 July 2019; Accepted: 23 August 2019; Published: 25 August 2019

Abstract: Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disorder affecting upper and lower motor neurons (MNs) that still lacks an efficacious therapy. The failure of recent therapeutic trials in ALS, other than depending on the poor knowledge of pathogenic mechanisms responsible for MNs loss, is largely due to diagnostic delay and the lack of reliable biomarkers for diagnosis, prognosis and response to pharmacologic intervention. Neurofilaments (Nfs) are neuron-specific cytoskeletal proteins, whose levels increased in biological fluids proportionally to the degree of axonal damage, both in normal and in pathologic conditions, representing potential biomarkers in various neurological disorders, such as motor neuron disorder (MND). Growing evidence has shown that phosphorylated neurofilaments heavy chain (p-NfH) and neurofilaments light chain (NfL) are increased in blood and cerebrospinal fluid (CSF) of ALS patients compared to healthy and neurological controls and are found to correlate with disease progression. In this review, we reported the most relevant studies investigating the diagnostic and prognostic role of Nfs in ALS. Given their reliability and reproducibility, we consider Nfs as promising and useful biomarkers in diagnosis of MND, early patient identification for inclusion in clinical trials, prediction of disease progression, and response to pharmacological intervention, and we suggest the validation of their measurement in clinical activity.

Keywords: amyotrophic lateral sclerosis; neurofilaments; p-NfH; NfL; cerebrospinal fluid; blood; biomarkers

1. Introduction

Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disorder affecting upper and lower motor neurons (MNs) that still lacks an efficacious therapy. In the past decades, several studies have been led to find a molecule able to halt disease progression, but they were unsuccessful. The incomplete knowledge of pathophysiological mechanisms underlying ALS and the lack of available and reliable biomarkers for diagnosis, disease progression, prognosis, and response to pharmacologic intervention has limited the clinical experience about motor neuron disorders (MND) and has probably contributed to the failure of recent therapeutic trials in ALS.

Received: 14 July 2018 | Accepted: 29 September 2018

DOI: 10.1111/jcmm.13976

REVIEW

WILEY

MicroRNAs as regulators of cell death mechanisms in amyotrophic lateral sclerosis

Delia Gagliardi | Giacomo P. Comi | Nereo Bresolin | Stefania Corti

Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy

Correspondence
Stefania Corti, Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy.
Email: stefania.corti@unimi.it

Funding information
EU Community JPND DAMNDPATHS; Agenzia di Ricerca per la Sclerosi Laterale Amiotrofica smallRNALS; regione lombardia TRANS-ALS; Ministero della Salute, Grant/Award Number: RF-2013023555764

Abstract

Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disorder affecting upper and lower motor neurons (MNs), resulting in paralysis and precocious death from respiratory failure. Although the causes of ALS are incompletely understood, the role of alterations in RNA metabolism seems central. MicroRNAs (miRNAs) are noncoding RNAs implicated in the regulation of gene expression of many relevant physiological processes, including cell death. The recent model of programmed cell death (PCD) encompasses different mechanisms, from apoptosis to regulated necrosis (RN), in particular necroptosis. Both apoptosis and necroptosis play a significant role in the progressive death of MNs in ALS. In this review, we present key research related to miRNAs that modulate apoptosis and RN pathways in ALS. We also discuss whether these miRNAs represent potential targets for therapeutic development in patients.

KEYWORDS
amyotrophic lateral sclerosis, apoptosis, microRNAs, motor neurons, necroptosis, therapy

1 | INTRODUCTION

Amyotrophic lateral sclerosis (ALS) is a fatal neurological disease characterized by the progressive degeneration of both upper and lower motor neurons (MNs), which results in muscle paralysis and ultimately death from respiratory failure within a median of 3 years.[1] At present, no effective therapy is available for ALS. Riluzole and Edaravone are the sole two drugs approved for ALS but only modestly slow disease progression.[1,2]

Patients affected by idiopathic ALS and without a family history are classified as sporadic (sALS). A fraction of cases (approximately 10%) are familial (fALS), because of mutations in genes involved in a wide range of cellular functions, encompassing roles in motor neuronal and non-neuronal cells.[3,4]

Since the first identification, in 1993, of superoxide dismutase 1 (SOD1) gene mutations as being responsible for some forms of fALS,[5] many other genes linked to ALS have been identified.[4] The most frequent altered gene in both sALS and fALS is *C9ORF72*.[6] Other relevant genes are *TAR DNA-binding protein (TARDBP)*, encoding for TDP-43, and *fused in sarcoma (FUS)*, which encode for proteins implicated in the regulation of RNA processing and expression.[4] Intracellular inclusions of TDP-43 are the main pathological substrate of sporadic and familial forms of ALS, except for those associated with *SOD1* mutations.[7-9]

Understanding the etiology of ALS and the factors that influence its progression is crucial for the implementation of effective therapeutic strategies that are urgently needed. Although specific causes leading to ALS are unknown, different cellular mechanisms were proposed to mediate MN degeneration, such as glutamate excitotoxicity, mitochondrial dysfunction, protein aggregation, proteasomal and autophagic dysfunction, neuroinflammation, altered axonal transport, and impaired RNA metabolism.[1] In this context, the role of alterations of RNA metabolism seems particularly central, especially considering that *TDP43* and *FUS* are key components of coding and noncoding RNA processing.

Cell Calcium 85 (2020) 102110

Contents lists available at ScienceDirect

Cell Calcium

journal homepage: www.elsevier.com/locate/ceca

STIM1 R304W in mice causes subgingival hair growth and an increased fraction of trabecular bone

Thilini H. Gamage[a,1], Emma Lengle[a,1], Gjermund Gunnes[b], Helen Pullisaar[c], Asbjørn Holmgren[a], Janne E. Reseland[d], Else Merckoll[e], Stefania Corti[f,g], Masahiro Mizobuchi[h], Raul J. Morales[i], Leonidas Tsiokas[j], Geir E. Tjønnfjord[k], Rodrigo S. Lacruz[l], Staale P. Lyngstadaas[d], Doriana Misceo[a,2], Eirik Frengen[a,*,2]

[a] Department of Medical Genetics, Oslo University Hospital and University of Oslo, Oslo, Norway
[b] Faculty of Veterinary Medicine, Norwegian University of Life Sciences, Norway
[c] Department of Orthodontics, Institute of Clinical Dentistry, University of Oslo, Oslo, Norway
[d] Department of Biomaterials, Institute of Clinical Dentistry, University of Oslo, Oslo, Norway
[e] Department of Radiology and Nuclear Medicine, Oslo University Hospital, Oslo, Norway
[f] Neurology Unit, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
[g] Neuroscience Section, Department of Pathophysiology and Transplantation, Dino Ferrari Centre, University of Milan, Milan, Italy
[h] Department of Neurology, Nakamura Memorial Hospital, Sapporo, Japan
[i] CHRU, Hôpital Gui de Chauliac, Montpellier, France
[j] Department of Cell Biology, University of Oklahoma Health Sciences Center, Oklahoma, USA
[k] Department of Haematology, Oslo University Hospital and Institute of Clinical Medicine, University of Oslo, Oslo, Norway
[l] Department of Basic Science and Craniofacial Biology, New York University College of Dentistry, New York, USA

ARTICLE INFO

Keywords:
Abnormal bone architecture
Ectopic hair growth
Skeletal defects
STIM1 R304W
Stormorken syndrome

ABSTRACT

Calcium signaling plays a central role in bone development and homeostasis. Store operated calcium entry (SOCE) is an important calcium influx pathway mediated by calcium release activated calcium (CRAC) channels in the plasma membrane. Stromal interaction molecule 1 (STIM1) is an endoplasmic reticulum calcium sensing protein important for SOCE.

We generated a mouse model expressing the STIM1 R304W mutation, causing Stormorken syndrome in humans. $Stim1^{R304W/R304W}$ mice showed perinatal lethality, and the only three animals that survived into adulthood presented with reduced growth, low body weight, and thoracic kyphosis. Radiographs revealed a reduced number of ribs in the $Stim1^{R304W/R304W}$ mice. Microcomputed tomography data revealed decreased cortical bone thickness and increased trabecular bone volume fraction in $Stim1^{R304W/R304W}$ mice, which had thinner and more compact bone compared to wild type mice. The $Stim1^{R304W/+}$ mice showed an intermediate phenotype. Histological analyses showed that the $Stim1^{R304W/R304W}$ mice had abnormal bone architecture, with markedly increased number of trabeculae and reduced bone marrow cavity. Homozygous mice showed STIM1 positive osteocytes and osteoblasts. These findings highlight the critical role of the gain-of-function (GoF) STIM1 R304W protein in skeletal development and homeostasis in mice. Furthermore, the novel feature of bilateral subgingival hair growth on the lower incisors in the $Stim1^{R304W/R304W}$ mice and 25 % of the heterozygous mice indicate that the GoF STIM1 R304W protein also induces an abnormal epithelial cell fate.

1. Introduction

Calcium signaling plays an important role in bone development and homeostasis [1]. One important Ca^{2+} entry pathway in non-excitable cells is the store-operated Ca^{2+} entry (SOCE) [2,3]. SOCE is mediated by stromal interaction molecule 1 (STIM1), a single-pass transmembrane protein, which resides in the endoplasmic reticulum (ER) membrane and senses ER Ca^{2+} levels [4]. When ER Ca^{2+} stores are low,

* Corresponding author.
E-mail address: eirik.frengen@medisin.uio.no (E. Frengen).
[1] These authors contributed equally to this work.
[2] These authors contributed equally to this work.

https://doi.org/10.1016/j.ceca.2019.102110
Received 13 July 2019; Received in revised form 5 November 2019; Accepted 11 November 2019
Available online 13 November 2019

Parkinsonism and Related Disorders 63 (2019) 66–72

Contents lists available at ScienceDirect

Parkinsonism and Related Disorders

journal homepage: www.elsevier.com/locate/parkreldis

Neurofascin (*NFASC*) gene mutation causes autosomal recessive ataxia with demyelinating neuropathy

Edoardo Monfrini[a,b], Letizia Straniero[c,d], Sara Bonato[a,b], Giacomo Monzio Compagnoni[a,b], Andreina Bordoni[a,b], Robertino Dilena[e], Paola Rinchetti[a,b], Rosamaria Silipigni[f], Dario Ronchi[a,b], Stefania Corti[a,b], Giacomo P. Comi[a,b], Nereo Bresolin[a,b], Stefano Duga[c,d], Alessio Di Fonzo[a,b,*]

[a] Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurology Unit, Milan, Italy
[b] Dino Ferrari Center, Neuroscience Section, Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy
[c] Department of Biomedical Sciences, Humanitas University, Pieve Emanuele, Milan, Italy
[d] Humanitas Clinical and Research Center, Rozzano, Milan, Italy
[e] Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurofisiopatologia Pediatrica, UOC Neurofisiopatologia, Milan, Italy
[f] Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Laboratory of Medical Genetics, Milan, Italy

ARTICLE INFO

Keywords:
Neurofascin
NFASC
Hereditary Ataxia
Neuropathy
Nodopathy

ABSTRACT

Introduction: Neurofascin, encoded by *NFASC*, is a transmembrane protein that plays an essential role in nervous system development and node of Ranvier function. *Anti*-Neurofascin autoantibodies cause a specific type of chronic inflammatory demyelinating polyneuropathy (CIDP) often characterized by cerebellar ataxia and tremor. Recently, homozygous *NFASC* mutations were recently associated with a neurodevelopmental disorder in two families.

Methods: A combined approach of linkage analysis and whole-exome sequencing was performed to find the genetic cause of early-onset cerebellar ataxia and demyelinating neuropathy in two siblings from a consanguineous Italian family. Functional studies were conducted on neurons from induced pluripotent stem cells (iPSCs) generated from the patients.

Results: Genetic analysis revealed a homozygous p.V1122E mutation in *NFASC*. This mutation, affecting a highly conserved hydrophobic transmembrane domain residue, led to significant loss of Neurofascin protein in the iPSC-derived neurons of affected siblings.

Conclusions: The identification of *NFASC* mutations paves the way for genetic research in the developing field of nodopathies, an emerging pathological entity involving the nodes of Ranvier, which are associated for the first time with a hereditary ataxia syndrome with neuropathy.

1. Introduction

Neurofascin, a member of the L1 immunoglobulin cell adhesion molecule family (L1CAM) family, is a transmembrane protein, encoded by *NFASC*, an evolutionarily conserved gene located on chromosome 1q32, and ubiquitously expressed in human tissues [1]. Neurofascin protein is abundant in the adult central nervous system, especially in the cerebellum and peripheral nerves [2,3]. Moreover, Neurofascin plays a pivotal role in the development and function of the axon initial segment (AIS) and the nodes of Ranvier [4,5]. Both the AIS and node of Ranvier contain a high density of sodium voltage-gated channels, necessary for action potential generation and propagation, which are clustered by interactions with several cytoskeletal and scaffolding proteins, including Neurofascin. Another recognized role of Neurofascin is to connecting the extracellular matrix and glial cells (Schwann cells and oligodendroglia) with the intracellular skeleton of neurons [6–8].

NFASC transcripts undergo extensive alternative splicing and protein isoforms are differentially expressed in the central and peripheral nervous systems in a developmental- and cell type-specific manner. In the adult mouse, Neurofascin has two principal isoforms: the neuronal isoform (138 kDa), known as NF186, and the glial isoform (133 kDa), known as NF155. These proteins are highly glycosylated with observed molecular weights on Western blot of 186 kDa and 155 kDa respectively [9,10]. Similarly, adult human nervous tissues have two principal Neurofascin isoforms with molecular weights of 186 kDa and 150 kDa.

Here, we describe two affected siblings from a consanguineous

* Corresponding author. Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza 35, 20122, Milan, Italy.
E-mail address: alessio.difonzo@policlinico.mi.it (A. Di Fonzo).

https://doi.org/10.1016/j.parkreldis.2019.02.045
Received 2 October 2018; Received in revised form 10 January 2019; Accepted 25 February 2019

frontiers in Neurology

ORIGINAL RESEARCH
published: 27 February 2019
doi: 10.3389/fneur.2019.00160

Lipomatosis Incidence and Characteristics in an Italian Cohort of Mitochondrial Patients

Olimpia Musumeci[1†], Emanuele Barca[2†], Costanza Lamperti[3], Serenella Servidei[4], Giacomo Pietro Comi[5], Maurizio Moggio[6], Tiziana Mongini[7], Gabriele Siciliano[8], Massimiliano Filosto[9], Elena Pegoraro[10], Guido Primiano[4], Dario Ronchi[5], Liliana Vercelli[7], Daniele Orsucci[8], Luca Bello[10], Massimo Zeviani[11], Michelangelo Mancuso[8] and Antonio Toscano[1]*

[1] Department of Clinical and Experimental Medicine, UOC Neurologia e Malattie Neuromuscolari, University of Messina, Messina, Italy, [2] Department of Neurology, Columbia University Medical Center, New York, NY, United States, [3] UO of Medical Genetics and Neurogenetics, The Foundation "Carlo Besta" Institute of Neurology-IRCCS, Milan, Italy, [4] UOC Neurofisiopatologia, Fondazione Policlinico Universitario A. Gemelli IRCCS, Istituto di Neurologia Università Cattolica del Sacro Cuore, Rome, Italy, [5] Neurology Unit, Neuroscience Section, Department of Pathophysiology and Transplantation, Dino Ferrari Centre, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy, [6] Neuromuscular and Rare Diseases Unit, Department of Neuroscience, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, Milan, Italy, [7] Department of Neurosciences Rita Levi Montalcini, University of Torino, Torino, Italy, [8] Department of Clinical and Experimental Medicine, Neurological Institute, University of Pisa, Pisa, Italy, [9] Unit of Neurology, Center for Neuromuscular Diseases, ASST Spedali Civili and University of Brescia, Brescia, Italy, [10] Department of Neurosciences, University of Padova, Padova, Italy, [11] Mitochondrial Biology Unit, Medical Research Council, Cambridge, United Kingdom

OPEN ACCESS

Edited by:
Margherita Milone, Mayo Clinic, United States

Reviewed by:
Lee-Jun Wong, Baylor College of Medicine, United States
Filippo M. Santorelli, Fondazione Stella Maris (IRCCS), Italy

***Correspondence:**
Olimpia Musumeci
omusumeci@unime.it

[†]These authors have contributed equally to this work

Specialty section:
This article was submitted to Neuromuscular Diseases, a section of the journal Frontiers in Neurology

Received: *10 January 2019*
Accepted: *07 February 2019*
Published: *27 February 2019*

Citation:
Musumeci O, Barca E, Lamperti C, Servidei S, Comi GP, Moggio M, Mongini T, Siciliano G, Filosto M, Pegoraro E, Primiano G, Ronchi D, Vercelli L, Orsucci D, Bello L, Zeviani M, Mancuso M and Toscano A (2019) Lipomatosis Incidence and Characteristics in an Italian Cohort of Mitochondrial Patients. Front. Neurol. 10:160. doi: 10.3389/fneur.2019.00160

Lipomas have often been associated with mtDNA mutations and were mainly observed in patients with mutation in mitochondrial tRNAlysine which is also the most frequent mutation associated with MERRF. Up to date, no systematic studies have been developed in order to assess the incidence of lipomas in large cohorts of mitochondrial patients.The aim of this study is to analyze the incidence and characteristics of lipomas among an Italian cohort of patients with mitochondrial diseases. A retrospective, database-based study (Nation-wide Italian Collaborative Network of Mitochondrial Diseases) of patients with lipomas was performed. A total of 22 (1.7%) patients with lipomas have been identified among the 1,300 mitochondrial patients, enrolled in the Italian database. In about 18% multiple systemic lipomatosis (MSL) was the only clinical manifestation; 54% of patients showed a classical MERRF syndrome. Myopathy, alone or in association with other symptoms, was found in 27% of patients. Lactate was elevated in all the 12 patients in which was measured. Muscle biopsy was available in 18/22 patients: in all of them mitochondrial abnormalities were present. Eighty six percent had mutations in mtDNA coding for tRNA lysine. In most of patients, lipomas were localized along the cervical-cranial-thoracic region. In 68% of the patients were distributed symmetrically. Only two patients had lipomas in a single anatomical site (1 in right arm and 1 in gluteus maximum). MSL is often overlooked by clinicians in patients with mitochondrial diseases where the clinical picture could be dominated by a severe multi-systemic involvement. Our data confirmed that MSL is a rare sign of mitochondrial disease with a strong association between multiple lipomas and lysine tRNA mutations. MSL could be considered, even if rare, a red flag for mitochondrial disorders, even in patients with an apparently isolated MSL.

Keywords: multiple symmetrical lipomatosis, MERRF, mitochondrial myopathy, madelung's disease, brown fat

Human induced pluripotent stem cell models for the study and treatment of Duchenne and Becker muscular dystrophies

Ther Adv Neurol Disord

2019, Vol. 12: 1–28

DOI: 10.1177/1756285619833478

Daniela Piga, Sabrina Salani, Francesca Magri, Roberta Brusa, Eleonora Mauri, Giacomo P. Comi, Nereo Bresolin and Stefania Corti

Abstract: Duchenne and Becker muscular dystrophies are the most common muscle diseases and are both currently incurable. They are caused by mutations in the dystrophin gene, which lead to the absence or reduction/truncation of the encoded protein, with progressive muscle degeneration that clinically manifests in muscle weakness, cardiac and respiratory involvement and early death. The limits of animal models to exactly reproduce human muscle disease and to predict clinically relevant treatment effects has prompted the development of more accurate *in vitro* skeletal muscle models. However, the challenge of effectively obtaining mature skeletal muscle cells or satellite stem cells as primary cultures has hampered the development of *in vitro* models. Here, we discuss the recently developed technologies that enable the differentiation of skeletal muscle from human induced pluripotent stem cells (iPSCs) of Duchenne and Becker patients. These systems recapitulate key disease features including inflammation and scarce regenerative myogenic capacity that are partially rescued by genetic and pharmacological therapies and can provide a useful platform to study and realize future therapeutic treatments. Implementation of this model also takes advantage of the developing genome editing field, which is a promising approach not only for correcting dystrophin, but also for modulating the underlying mechanisms of skeletal muscle development, regeneration and disease. These data prove the possibility of creating an accurate Duchenne and Becker *in vitro* model starting from iPSCs, to be used for pathogenetic studies and for drug screening to identify strategies capable of stopping or reversing muscular dystrophinopathies and other muscle diseases.

Keywords: 3D models, Becker muscular dystrophy, cellular differentiation, Duchenne muscular dystrophy, iPSC models

Received: 21 July 2018; revised manuscript accepted: 27 November 2018.

Correspondence to:
Stefania Corti
Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Via Francesco Sforza 35, 20122, Milan, Italy
stefania.corti@unimi.it

Daniela Piga
Sabrina Salani
Francesca Magri
Roberta Brusa
Eleonora Mauri
Giacomo P. Comi
Nereo Bresolin
Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy

Introduction

Duchenne (DMD) and Becker (BMD) muscular dystrophies, also known as dystrophinopathies, are the most prevalent muscle diseases, with incidences of 1:5000 and 1:18000 live born males, respectively.[1–3] They are characterized by progressive weakness and muscle degeneration. Dystrophinopathies are X-linked genetic disorder caused by mutations in the *DMD* gene, which leads to the loss (DMD) or severe reduction/truncation (BMD) of the full length dystrophin protein.[1–3] This protein is essential, both to connect the cytoskeleton with the basal lamina and to mediate signaling pathways; indeed, its absence produces membrane destabilization and subsequent muscle degeneration.[4,5] Over time, the damaged fibers are not regenerated effectively and are then replaced by fat and fibrotic tissue, which causes progressive weakness with muscular atrophy and eventual death. Generally, the symptoms of DMD begin in early childhood with a rapid progression and death in early

doi:10.1093/brain/awy330 BRAIN 2019: 142; 276–294 | 276

BRAIN
A JOURNAL OF NEUROLOGY

Key role of SMN/SYNCRIP and RNA-Motif 7 in spinal muscular atrophy: RNA-Seq and motif analysis of human motor neurons

Federica Rizzo,[1,*] Monica Nizzardo,[1,*] Shikha Vashisht,[2] Erika Molteni,[2] Valentina Melzi,[1] Michela Taiana,[1] Sabrina Salani,[3] Pamela Santonicola,[4] Elia Di Schiavi,[4] Monica Bucchia,[1] Andreina Bordoni,[1] Irene Faravelli,[1] Nereo Bresolin,[1,2] Giacomo Pietro Comi,[1,2] Uberto Pozzoli[3] and Stefania Corti[1,2]

*These authors contributed equally to this work.

Spinal muscular atrophy is a motor neuron disorder caused by mutations in *SMN1*. The reasons for the selective vulnerability of motor neurons linked to SMN (encoded by *SMN1*) reduction remain unclear. Therefore, we performed deep RNA sequencing on human spinal muscular atrophy motor neurons to detect specific altered gene splicing/expression and to identify the presence of a common sequence motif in these genes. Many deregulated genes, such as the neurexin and synaptotagmin families, are implicated in critical motor neuron functions. Motif-enrichment analyses of differentially expressed/spliced genes, including neurexin2 (*NRXN2*), revealed a common motif, motif 7, which is a target of SYNCRIP. Interestingly, SYNCRIP interacts only with full-length SMN, binding and modulating several motor neuron transcripts, including SMN itself. SYNCRIP overexpression rescued spinal muscular atrophy motor neurons, due to the subsequent increase in SMN and their downstream target *NRXN2* through a positive loop mechanism and ameliorated SMN-loss-related pathological phenotypes in *Caenorhabditis elegans* and mouse models. SMN/SYNCRIP complex through motif 7 may account for selective motor neuron degeneration and represent a potential therapeutic target.

1 Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Milan, Italy
2 Scientific Institute IRCCS E. MEDEA, Computational Biology, Bosisio Parini, Lecco, Italy
3 Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
4 Institute of Bioscience and BioResources, IBBR, CNR, Naples, Italy

Correspondence to: Prof. Stefania Corti
Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza 35, 20122 Milan Italy
E-mail: stefania.corti@unimi.it.

Keywords: motor neurons; RNA sequencing; *NRXN2*; *SYNCRIP*; *SMN1*

Abbreviations: AAV9 = adenovirus-associated vector serotype 9; DAVID = database for annotation, visualization, and integrated discovery; GO = gene ontology; hnRNP = heterogeneous nuclear ribonucleoprotein; iPSC = induced pluripotent stem cell; NMJ = neuromuscular junction; SMA = spinal muscular atrophy; SMN = survival motor neuron; RBP = RNA binding protein; RNA-Seq = RNA sequencing

Received January 20, 2018. Revised October 23, 2018. Accepted November 3, 2018. Advance Access publication January 15, 2019

ANNALS of Clinical and Translational Neurology
Open Access
ANA American Neurological Association since 1875

BRIEF COMMUNICATION

Novel mutations in DNA2 associated with myopathy and mtDNA instability

Dario Ronchi[1,2,*], Changwei Liu[3,*], Leonardo Caporali[4,*], Daniela Piga[1], Hongzhi Li[3], Francesca Tagliavini[4], Maria Lucia Valentino[4,5], Maria Teresa Ferrò[6], Paola Bini[7], Li Zheng[3], Valerio Carelli[4,5,†], Binghui Shen[3,†] & Giacomo Pietro Comi[2,8,†]

[1]Neurology Unit, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
[2]Dino Ferrari Center, Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy
[3]Department of Cancer Genetics and Epigenetics, Beckman Research Institute City of Hope, Duarte, California
[4]Istituto delle Scienze Neurologiche di Bologna, UOC Clinica Neurologica, Bologna, Italy
[5]Dipartimento di Scienze Biomediche e Neuromotorie (DIBINEM), Università di Bologna, Bologna, Italy
[6]Neurology Unit, Ospedale Maggiore, Crema, Italy
[7]IRCCS "C. Mondino" Foundation, National Neurological Institute, Pavia, Italy
[8]Department of Neuroscience, Neuromuscular and Rare Diseases Unit, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan,20122, Italy

Correspondence
Giacomo Pietro Comi, Department of Pathophysiology and Transplantation, Neuroscience Section, University of Milan IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza 35, 20122 Milan, Italy. Tel: +39 0255033817; Fax: +39 0250320430; E-mail: giacomo.comi@unimi.it and Binghui Shen, Department of Cancer Genetics and Epigenetics Beckman Research Institute, City of Hope 1500 East Duarte Road, Duarte, CA 91010-3000. Tel: +626-301-8879; Fax: +626-301-8280; E-mail: bshen@coh.org and Valerio Carelli, Istituto delle Scienze Neurologiche di Bologna, UOC Clinica Neurologica, Bologna, Italy Dipartimento di Scienze Biomediche e Neuromotorie (DIBINEM), Università di Bologna, Bologna, Italy. Tel: +39 051 4966747; Fax: +39 051 209 2751; E-mail: valerio.carelli@unibo.it

Funding Information
This work was supported by Fondazione Cariplo (Project 2014-1010 to D.R.), Italian Telethon (Grant GSP16001 to G.P.C.) and the Italian Ministry of Health (Ricerca Corrente to V.C.). The work done in the Shen laboratory was supported by R01CA085344 to B.H.S.

Received: 31 May 2019; Revised: 12 August 2019; Accepted: 13 August 2019

Annals of Clinical and Translational Neurology **2019; 6(9): 1893–1899**

Abstract

The maintenance of mitochondrial DNA (mtDNA) relies on proteins encoded by nuclear genes. Mutations in their coding sequences result in heterogenous clinical presentations featuring mtDNA instability in affected tissues. DNA2 is a multi-catalytic protein involved in the removal of single strand DNA during mtDNA replication or Long Patch Base Excision Repair pathway. We have previously described *DNA2* mutations in adult patients affected with familial and sporadic forms of mitochondrial myopathy. Here we describe four novel probands presenting with limb weakness associated with novel *DNA2* molecular defects. Biochemical assays were established to investigate the functional effects of these variants.

cells

MDPI

Review

GBA, Gaucher Disease, and Parkinson's Disease: From Genetic to Clinic to New Therapeutic Approaches

Giulietta M. Riboldi [1,2,*] and Alessio B. Di Fonzo [2,3]

[1] The Marlene and Paolo Fresco Institute for Parkinson's and Movement Disorders, NYU Langone Health, New York, NY 10017, USA
[2] Dino Ferrari Center, Neuroscience Section, Department of Pathophysiology and Transplantation, University of Milan, 20122 Milan, Italy; alessio.difonzo@policlinico.mi.it
[3] Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurology Unit, 20122 Milan, Italy
* Correspondence: giulietta.riboldi@nyulangone.org or giulietta.riboldi@unimi.it; Tel.: +1-929-455-5652

Received: 21 March 2019; Accepted: 16 April 2019; Published: 19 April 2019

Abstract: Parkinson's disease (PD) is the second most common degenerative disorder. Although the disease was described more than 200 years ago, its pathogenetic mechanisms have not yet been fully described. In recent years, the discovery of the association between mutations of the *GBA* gene (encoding for the lysosomal enzyme glucocerebrosidase) and PD facilitated a better understating of this disorder. *GBA* mutations are the most common genetic risk factor of the disease. However, mutations of this gene can be found in different phenotypes, such as Gaucher's disease (GD), PD, dementia with Lewy bodies (DLB) and rapid eye movements (REM) sleep behavior disorders (RBDs). Understanding the pathogenic role of this mutation and its different manifestations is crucial for geneticists and scientists to guide their research and to select proper cohorts of patients. Moreover, knowing the implications of the *GBA* mutation in the context of PD and the other associated phenotypes is also important for clinicians to properly counsel their patients and to implement their care. With the present review we aim to describe the genetic, clinical, and therapeutic features related to the mutation of the *GBA* gene.

Keywords: glucocerebrosidase; Parkinson's disease; Gaucher's disease; Lewy Body Dementia; REM sleep behavior disorders

1. Introduction

GBA is a gene located on chromosome 1 (1q21) encoding for the glucocerebrosidase (GCase), a lysosomal enzyme involved in the metabolism of glucosylceramide. The mutation of this gene has been classically associated with Gaucher's disease, a systemic disorder with a variable degree of involvement of the central nervous system. Surprisingly, about 14 years ago it was observed that mutations in this same gene were associated with an increased incidence of Parkinson's disease (PD), in both Gaucher's patients as well as asymptomatic carriers [1–4]. PD is the second most common neurodegenerative disorder, affecting 2–3% of the world population over the age of 65 [5]. It is caused by the progressive loss of dopaminergic neurons in the substantia nigra. Classically it presents with a combination of bradykinesia, rigidity, resting tremor, and postural instability. However, a list of non-motor features, such as hyposmia, constipation, urinary symptoms, orthostatic hypotension, anxiety, depression, impaired sleep, and cognitive impairment can present as well in various degrees [5]. Since the first observations of *GBA* and PD, their association has been extensively explored. Different hypotheses have been formulated to explain the causative role of this mutation in PD [6]. First of all, GCase is part of the endolysosomal pathway, which seems to be particularly crucial in the pathogenesis

Current Neurology and Neuroscience Reports (2018) 18:37
https://doi.org/10.1007/s11910-018-0847-1

MOVEMENT DISORDERS (S FOX, SECTION EDITOR)

Genetics of Movement Disorders and the Practicing Clinician; Who and What to Test for?

Alessio Di Fonzo[1] · Edoardo Monfrini[1] · Roberto Erro[2]

Abstract

Purpose of Review This review aims to provide the basic knowledge on the genetics of hypokinetic and hyperkinetic movement disorders to guide clinicians in the decision of "who and what to test for?"

Recent Findings In recent years, the identification of various genetic causes of hypokinetic and hyperkinetic movement disorders has had a great impact on a better definition of different clinical syndromes. Indeed, the advent of next-generation sequencing (NGS) techniques has provided an impressive step forward in the easy identification of genetic forms. However, this increased availability of genetic testing has challenges, including the ethical issue of genetic testing in unaffected family members, "commercially" available home testing kits and the increasing number and relevance of "variants of unknown significance."

Summary The emergent role of genetic factors has important implications on clinical practice and counseling. As a consequence, it is fundamental that practicing neurologists have a proper knowledge of the genetic background of the diseases and perform an accurate selection of who has to be tested and for which gene mutations.

Keywords Genetics · Movement disorders · Next-generation sequencing · Parkinson's disease · Dystonia · Chorea

Introduction

In the last 20 years, the identification of genetic causes of movement disorders has had a significant impact on the comprehension of pathological mechanisms and on a better definition of different clinical syndromes. Moreover, the advent of next-generation sequencing (NGS) techniques has provided an impressive step forward in the ease of identification of several genetic forms of hypokinetic and hyperkinetic movement disorders [1].

The emergent role of genetic factors also has important implications for clinical practice and counseling.

This article is part of the Topical Collection on *Movement Disorders*

✉ Roberto Erro
rerro@unisa.it

[1] IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Dino Ferrari Center, Neuroscience Section, Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy

[2] Neurodegenerative disease center (CEMAND), Department of Medicine, Surgery and Dentistry "Scuola Medica Salernitana", University of Salerno, Baronissi, SA, Italy

Genetic testing is a very complex subject, which involves clinicians, geneticists, patients, and their families. Clinical genetic testing in patients or relatives can serve a variety of purposes. If causal or symptomatic treatments depend on the molecular diagnosis, the purpose of genetic analysis is evident (e.g. chelating therapy in Wilson's disease). However, even if there are no clear therapeutic consequences, a patient can still benefit from genetic diagnosis. For example, defining the molecular etiology of the disease can be a reassuring final explanation of the signs and symptoms for the patient and can put an end to a long and painful series of medical visits and expensive diagnostic tests. Life or family planning could be another reasonable cause to perform a genetic test, in particular if a highly penetrant dominant or X-linked pattern of inheritance is present. In the clinical setting, genetic testing should be performed only if there is a clear and informed wish of the patient, always following genetic counseling. Therefore, patients or people- at-risk should be comprehensively informed and counseled about the possibility of transmission, penetrance, expressivity, and available therapeutic solutions [2].

It is important to highlight that genetic analysis in the research setting—with or without disclosure of the test results—can be performed in every patient who gives appropriate

Published online: 23 May 2018

Parkinsonism in diseases predominantly presenting with dystonia

Alessio Di Fonzo[a], Giulia Franco[a], Paolo Barone[b] and Roberto Erro[b,*]

[a]Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurology Unit, Milan, Italy; Dino Ferrari Center, Neuroscience Section, Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy
[b]Department of Medicine, Surgery and Dentistry "Scuola Medica Salernitana", University of Salerno, Baronissi, SA, Italy
*Corresponding author: E-mail: rerro@unisa.it

Contents

International Review of Neurobiology, Volume 149
ISSN 0074-7742
https://doi.org/10.1016/bs.irn.2019.10.007

Neurological Sciences (2019) 40:2163–2169
https://doi.org/10.1007/s10072-019-03944-x

ORIGINAL ARTICLE

Validation of the Italian version of carers' quality-of-life questionnaire for parkinsonism (PQoL Carer) in progressive supranuclear palsy

Marina Picillo[1] · Sofia Cuoco[1] · Marianna Amboni[1] · Francesco Paolo Bonifacio[2] · Antonino Bruno[3] · Fabio Bruschi[4] · Arianna Cappiello[1] · Rosa De Micco[2] · Anna De Rosa[5] · Francesca Di Biasio[6] · Francesca Elifani[7] · Roberto Erro[1] · Margherita Fabbri[8] · Marika Falla[9,10] · Giulia Franco[11] · Daniela Frosini[3] · Sebastiano Galantucci[12] · Giulia Lazzeri[11] · Luca Magistrelli[13,14] · Maria Chiara Malaguti[15] · Anna Vera Milner[13] · Brigida Minafra[4] · Enrica Olivola[7] · Andrea Pilotto[16] · Cristina Rascunà[17] · Maria Cristina Rizzetti[18] · Tommaso Schirinzi[19] · Barbara Borroni[16] · Roberto Ceravolo[3] · Alessio Di Fonzo[11] · Leonardo Lopiano[8] · Roberta Marchese[6] · Nicola B Mercuri[19] · Nicola Modugno[7] · Alessandra Nicoletti[17] · Alessandro Padovani[16] · Gabriella Santangelo[20] · Alessandro Stefani[19] · Alessandro Tessitore[2] · Maria Antonietta Volontè[12] · Roberta Zangaglia[4] · Mario Zappia[17] · Paolo Barone[1]

Received: 1 April 2019 / Accepted: 20 May 2019 / Published online: 12 June 2019

Abstract

Progressive supranuclear palsy (PSP) is a rare, rapidly progressive, neurodegenerative disease characterized by falls and ocular movement disturbances. Caring for a partner or relative who suffers from PSP entails a strenuous and demanding task, usually lasting for years that affects carers' everyday life routines and emotional and social well-being. The 26-item Parkinsonism Carers QoL (PQoL Carer) is a self-administered, concise instrument evaluating the quality of life of caregivers of patients with atypical parkinsonism (both PSP and multiple system atrophy). Here, the PQoL Carer was translated into Italian and validated in 162 carers of PSP patients (54.3% women; mean age (standard deviation), 62.4 (15.4)) diagnosed according to the Movement Disorder Society criteria and recruited in 16 third-level movement disorders centers participating in the Neurecanet project. The mean PQoL total score was 40.66 ± 19.46. The internal consistency was excellent (Cronbach's alpha = 0.941); corrected item-total correlation was > 0.40 for all the items. A correlation with other health-related quality of life measures as well as with behavioral assessments was shown suggesting adequate convergent validity of the scale. PQoL also correlated with patients' severity of disease. The discriminant validity of the scale was evidenced by its capacity to differentiate between carers with varying levels of self-reported health ($p < 0.001$). In conclusion, the Italian version of the PQoL Carer is an easy, consistent, and valid tool for the assessment of the quality of life in carers of PSP patients.

Keywords Progressive supranuclear palsy · Quality of life · Caregiver · Carer · Clinical trials

Abbreviations

EQ-5D	Euroqol 5D
EQ-VAS	Euroqol visual analogue scale
HADS	Hospital Anxiety Depression Scale
HADS-A	Hospital Anxiety Depression Scale – anxiety
HADS-D	Hospital Anxiety Depression scale – depression
HR-QoL	Health-related quality of life
MDS	Movement Disorders Society
PSP	Progressive supranuclear palsy
PSP-RS	Progressive Supranuclear Palsy rating scale
PQoL Carers	Carers' quality-of-life questionnaire for parkinsonism

✉ Paolo Barone
pbarone@unisa.it

Extended author information available on the last page of the article

Introduction

Progressive supranuclear palsy (PSP) is a rare, rapidly progressive, neurodegenerative disease characterized by falls and supranuclear vertical palsy with a prevalence of about 6 per 100,000 and associated with reduced life expectancy,

[1] Center for Neurodegenerative Diseases (CEMAND), Department of Medicine, Surgery and Odontoiatry, University of Salerno, 84131 Salerno, Italy

[2] Department of Advanced Medical and Surgical Sciences, University of Campania "Luigi Vanvitelli", Naples, Italy

[3] Dipartimento di Medicina Clinica e Sperimentale, Università di Pisa, Pisa, Italy

[4] Parkinson's Disease and Movement Disorders Unit, IRCCS Mondino Foundation, Pavia, Italy

[5] Department of Neurosciences and Reproductive and Odontostomatological Sciences, Federico II University, Naples, Italy

[6] IRCCS Policlinico San Martino, Genoa, Italy

[7] IRCCS Neuromed, Pozzilli, Italy

[8] Department of Neuroscience "Rita Levi Montalcini", University of Turin, Via Cherasco 15, 10124 Torino, Italy

[9] Department of Neurology, General Hospital of Bolzano, Bolzano, Italy

[10] CIMec and CeRIN, University of Trento, Rovereto, Italy

[11] IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Dino Ferrari Center, Neuroscience Section, Department of Pathophysiology and Transplantation, University of Milan, 20122 Milan, Italy

[12] Dipartimento Neurologico, IRCCS Ospedale San Raffaele – Milano, Milan, Italy

[13] Movement Disorders Centre, Neurology Unit, Department of Translational Medicine, University of Piemonte Orientale, Novara, Italy

[14] PhD Program in Clinical and Experimental Medicine and Medical Humanities, University of Insubria, Varese, Italy

[15] UO Neurologia, Ospedale Santa Chiara Trento, Azienda Provinciale per i Servizi Sanitari Provincia Autonoma di Trento, Trento, Italy

[16] Centre for Neurodegenerative Disorders, Neurology Unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy

[17] Department G.F. Ingrassia, Section of Neurosciences, University of Catania, Catania, Italy

[18] S. Isidoro Hospital - FERB Onlus, Trescore Balneario, Bergamo, Italy

[19] Centro Parkinson, Dip. Medicina dei Sistemi, Policlinico Tor Vergata, Rome, Italy

[20] Department of Psychology, University of Campania "Luigi Vanvitelli", Viale Ellittico 31, 81100 Caserta, Italy

Neurological Sciences (2019) 40:2587–2594
https://doi.org/10.1007/s10072-019-04010-2

ORIGINAL ARTICLE

Validation of the Italian version of the PSP Quality of Life questionnaire

Marina Picillo[1] · Sofia Cuoco[1] · Marianna Amboni[1] · Francesco Paolo Bonifacio[2] · Fabio Bruschi[3] · Immacolata Carotenuto[1] · Rosa De Micco[2] · Anna De Rosa[4] · Eleonora Del Prete[5] · Francesca Di Biasio[6] · Francesca Elifani[7] · Roberto Erro[1] · Margherita Fabbri[8] · Marika Falla[9,10] · Giulia Franco[11] · Daniela Frosini[5] · Sebastiano Galantucci[12] · Giulia Lazzeri[11] · Luca Magistrelli[13,14] · Maria Chiara Malaguti[15] · Anna Vera Milner[13] · Brigida Minafra[3] · Enrica Olivola[7] · Andrea Pilotto[16] · Cristina Rascunà[17] · Maria Cristina Rizzetti[18] · Tommaso Schirinzi[19] · Barbara Borroni[16] · Roberto Ceravolo[5] · Alessio Di Fonzo[11] · Roberta Marchese[6] · Nicola B. Mercuri[19] · Nicola Modugno[7] · Alessandra Nicoletti[17] · Alessandro Padovani[16] · Gabriella Santangelo[20] · Alessandro Stefani[19] · Alessandro Tessitore[2] · Maria Antonietta Volontè[12] · Roberta Zangaglia[3] · Mario Zappia[17] · Maurizio Zibetti[8] · Paolo Barone[1]

Received: 29 March 2019 / Accepted: 10 July 2019 / Published online: 26 July 2019

Abstract

Background Progressive supranuclear palsy (PSP) is a rare rapidly progressive, neurodegenerative disease characterized by falls and ocular movement disturbances. The use of health-related quality of life (HR-QoL) measures allows assessing changes in health status induced by therapeutic interventions or disease progress in neurodegenerative diseases. The PSP-QoL is a 45-item, self-administered questionnaire designed to evaluate HR-QoL in PSP.

Methods and Results Here, the PSP-QoL was translated into Italian and validated in 190 PSP (96 women and 94 men; mean age ± standard deviation, 72 ± 6.5; mean disease duration, 4.2 ± 2.3) patients diagnosed according to the Movement Disorder Society criteria and recruited in 16 third level movement disorders centers participating in the Neurecanet project. The mean PSP-QoL total score was 77.8 ± 37 (physical subscore, 46.5 ± 18.7; mental subscore, 33.6 ± 19.2). The internal consistency was high (Cronbach's alpha = 0.954); corrected item-total correlation was > 0.40 for the majority of items. The significant and moderate correlation of the PSP-QoL with other HR-QoL measures as well as with motor and disability assessments indicated adequate convergent validity of the scale. Gender and geographic location presented a significant impact on the PSP-QoL in our sample with women and patients from the South of Italy scoring higher than their counterparts.

Conclusion In conclusion, the Italian version of the PSP-QoL is an easy, reliable and valid tool for assessment of HR-QoL in PSP.

Keywords Parkinsonism · Progressive supranuclear palsy · Quality of life · Clinical trials

Abbreviations

EQ-5D	The EuroQoL questionnaire
EQ-VAS	The EuroQoL Visual Analogue Scale
HADS	The Hospital Anxiety and Depression Scale
HR-QoL	Health-related Quality of Life
MDS	Movement Disorder Society
MoCA	Montreal Cognitive Assessment
PSP	Progressive supranuclear palsy
PSP-QoL	Progressive Supranuclear Palsy Quality of Life Questionnaire
PSP-RS	Progressive Supranuclear Palsy Rating Scale
S&E	The Schwab and England Scale
VAS	Visual Analogue Scale

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s10072-019-04010-2) contains supplementary material, which is available to authorized users.

✉ Paolo Barone
pbarone@unisa.it

Extended author information available on the last page of the article

Affiliations

Marina Picillo[1] · Sofia Cuoco[1] · Marianna Amboni[1] · Francesco Paolo Bonifacio[2] · Fabio Bruschi[3] · Immacolata Carotenuto[1] · Rosa De Micco[2] · Anna De Rosa[4] · Eleonora Del Prete[5] · Francesca Di Biasio[6] · Francesca Elifani[7] · Roberto Erro[1] · Margherita Fabbri[8] · Marika Falla[9,10] · Giulia Franco[11] · Daniela Frosini[5] · Sebastiano Galantucci[12] · Giulia Lazzeri[11] · Luca Magistrelli[13,14] · Maria Chiara Malaguti[15] · Anna Vera Milner[13] · Brigida Minafra[3] · Enrica Olivola[7] · Andrea Pilotto[16] · Cristina Rascunà[17] · Maria Cristina Rizzetti[18] · Tommaso Schirinzi[19] · Barbara Borroni[16] · Roberto Ceravolo[5] · Alessio Di Fonzo[11] · Roberta Marchese[6] · Nicola B. Mercuri[19] · Nicola Modugno[7] · Alessandra Nicoletti[17] · Alessandro Padovani[16] · Gabriella Santangelo[20] · Alessandro Stefani[19] · Alessandro Tessitore[2] · Maria Antonietta Volontè[12] · Roberta Zangaglia[3] · Mario Zappia[17] · Maurizio Zibetti[8] · Paolo Barone[1]

[1] Center for Neurodegenerative Diseases (CEMAND), Department of Medicine, Surgery and Odontoiatry, University of Salerno, 84131 Salerno, Italy

[2] Department of Advanced Medical and Surgical Sciences, University of Campania "Luigi Vanvitelli", Naples, Italy

[3] Parkinson's Disease and Movement Disorders Unit, IRCCS Mondino Foundation, Pavia, Italy

[4] Department of Neurosciences and Reproductive and Odontostomatological Sciences, Federico II University, Naples, Italy

[5] Dipartimento di Medicina Clinica e Sperimentale, Università di Pisa, Pisa, Italy

[6] IRCCS Policlinico San Martino, Genoa, Italy

[7] IRCCS Neuromed, Pozzilli, Italy

[8] Department of Neuroscience "Rita Levi Montalcini", University of Turin, via Cherasco 15, 10124 Torino, Italy

[9] Department of Neurology, General Hospital of Bolzano, Bolzano, Italy

[10] CIMec and CeRIN, University of Trento, Rovereto, Italy

[11] IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Dino Ferrari Center, Neuroscience Section, Department of Pathophysiology and Transplantation, University of Milan, 20122 Milan, Italy

[12] Dipartimento Neurologico, IRCCS Ospedale San Raffaele, Milan, Italy

[13] Movement Disorders Centre, Neurology Unit, Department of Translational Medicine, University of Piemonte Orientale, Novara, Italy

[14] PhD Program in Clinical and Experimental Medicine and Medical Humanities, University of Insubria, Varese, Italy

[15] UO Neurologia, Ospedale Santa Chiara Trento, Azienda provinciale per i servizi sanitari provincia autonoma di Trento, Trento, Italy

[16] Centre for Neurodegenerative Disorders, Neurology Unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy

[17] Department G.F. Ingrassia, Section of Neurosciences, University of Catania, Catania, Italy

[18] S. Isidoro Hospital - FERB Onlus, Trescore Balneario, Bergamo, Italy

[19] Centro Parkinson, Dip. Medicina dei Sistemi, Policlinico Tor Vergata, Rome, Italy

[20] Department of Psychology, University of Campania "Luigi Vanvitelli", Viale Ellittico 31, 81100 Caserta, Italy

Psychosomatics 2019:60:278–288

Original Research Article

The Neuroanatomy of Somatoform Disorders: A Magnetic Resonance Imaging Study

Giuseppe Delvecchio, Ph.D.[1], Maria Gloria Rossetti, M.Sc.[1], Elisabetta Caletti, Psy.D., Andrea Arighi, M.D., Daniela Galimberti, Ph.D., Paola Basilico, M.D., Matteo Mercurio, Ph.D., Riccardo Paoli, M.D., Claudia Cinnante, M.D., Fabio Triulzi, M.D., A. Carlo Altamura, M.D., Elio Scarpini, M.D., Paolo Brambilla, M.D., Ph.D.

Background: *Somatoform disorders (SDs) are a heterogeneous group of psychiatric syndromes characterized by common symptoms, which may mimic a physical condition but they are not explained by a medical condition. Although the biologic nature of this disorder has been widely accepted, the neuroanatomical correlates characterizing SDs are still inconclusive.* **Objective:** *This study aims to explore gray matter (GM) volume alterations in SD patients compared to healthy controls and their possible association with clinical and cognitive measures.* **Method:** *We used voxel-based morphometry to examine regional GM volumes in 20 inpatients with SDs and 24-matched healthy controls. Only for SD patients, we employed multiple instruments to assess psychopathology and cognitive functioning, which were then used to explore their association with GM volume deficits.* **Results:** *Compared to healthy controls, SD patients showed GM volume reductions in the hypothalamus, left fusiform gyrus, right cuneus, left inferior frontal gyrus, left posterior cingulate, and right amygdala ($p < 0.05$, cluster Family Wise Error corrected). Additionally, in SD, Symptom Checklist-90-Phobia and Hamilton Depressive Rating Scale scores negatively correlated with specific fronto-temporoparietal regions whereas Symptom Checklist-90-Sleep scores positively correlated with anterior cingulate cortex. Lastly, the Boston Naming Test negatively correlated with fronto-temporoparietal and striatal volumes whereas Free and Cued Selective Reminding Test and Stroop scores positively correlated with superior temporal gyrus and cuneus, respectively (all $p < 0.05$, cluster Family Wise Error corrected).* **Conclusion:** *Our results suggest that SDs might be characterized by selective impairments in specific cortico-limbic regions associated to two overlapping circuits, the neuromatrix of pain and the emotion regulation system.*
(Psychosomatics 2019; 60:278–288)

Key words: MRI, gray matter, somatoform disorder, cognition, clinical profile.

INTRODUCTION

Somatoform disorders (SDs) are a heterogeneous group of psychiatric syndromes, such as somatization disorder (SOD), pain disorder, and conversion disorder (CD), characterized by physical symptoms that cannot be explained through objective examinations or instrumental diagnostic investigations, which ultimately lead to a decrement of quality of life.[1]

In recent years, neuroimaging studies have driven their attention toward the investigation of biological markers that might explain the clinical presentation

[1] These authors contributed equally to this article.

Received May 31, 2018; revised July 10, 2018; accepted July 13, 2018.
From the University of Milan (G.D., A.A., D.G., P.B., M.M., F.T., A.C.A., E.S., P.B.), Department of Pathophysiology and Transplantation, Milan, Italy; Department of Neurosciences (M.G.R.), Biomedicine and Movement Sciences, Section of Psychiatry and Clinical Psychology - University of Verona, Italy; Fondazione IRCCS Ca' Granda.Ospedale Maggiore Policlinico (E.C., A.A., D.G., P.B., M.M., R.P., C.C., F.T., A.C.A., E.S.), Milan, Italy; "Dino Ferrari" Center (A.A., D.G., P.B., M.M., E.S.), Milan, Italy; Scientific Institute IRCCS "E. Medea" (P.B.), Bosisio Parini (Lc), Italy
Send correspondence and reprint requests to Paolo Brambilla, M.D., Ph.D., Department of Pathophysiology and Transplantation, University of Milan, via F. Sforza 35, 20122 Milan, Italy; e-mail: paolo.brambilla1@unimi.it

MULTIPLE SCLEROSIS JOURNAL MSJ

Original Research Paper

CSF β-amyloid predicts prognosis in patients with multiple sclerosis

Anna M Pietroboni, Michela Caprioli, Tiziana Carandini, Marta Scarioni, Laura Ghezzi, Andrea Arighi, Sara Cioffi, Claudia Cinnante, Chiara Fenoglio, Emanuela Oldoni, Milena A De Riz, Paola Basilico, Giorgio G Fumagalli, Annalisa Colombi, Giovanni Giulietti, Laura Serra, Fabio Triulzi, Marco Bozzali, Elio Scarpini and Daniela Galimberti

Multiple Sclerosis Journal

2019, Vol. 25(9) 1223–1231

DOI: 10.1177/1352458518791709

Abstract

Background: The importance of predicting disease progression in multiple sclerosis (MS) has increasingly been recognized, and hence reliable biomarkers are needed.

Objectives: To investigate the prognostic role of cerebrospinal fluid (CSF) amyloid beta$_{1–42}$ (Aβ) levels by the determination of a cut-off value to classify patients in slow and fast progressors. To evaluate possible association with white matter (WM) and grey matter (GM) damage at early disease stages.

Methods: Sixty patients were recruited and followed up for 3–5 years. Patients underwent clinical assessment, brain magnetic resonance imaging (MRI; at baseline and after 1 year), and CSF analysis to determine Aβ levels. T1-weighted volumes were calculated. T2-weighted scans were used to quantify WM lesion loads.

Results: Lower CSF Aβ levels were observed in patients with a worse follow-up Expanded Disability Status Scale (EDSS; $r=-0.65$, $p<0.001$). The multiple regression analysis confirmed CSF Aβ concentration as a predictor of patients' EDSS increase ($r=-0.59$, $p<0.0001$). Generating a receiver operating characteristic curve, a cut-off value of 813 pg/mL was determined as the threshold able to identify patients with worse prognosis (95% confidence interval (CI): 0.690–0.933, $p=0.0001$). No differences in CSF tau and neurofilament light chain (NfL) levels were observed ($p>0.05$).

Conclusion: Low CSF Aβ levels may represent a predictive biomarker of disease progression in MS.

Keywords: Biomarkers, MRI, multiple sclerosis

Date received: 4 April 2018; revised: 21 June 2018; accepted: 9 July 2018

Correspondence to:
AM Pietroboni
Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Via F. Sforza 35, 20122 Milan, Italy.
pb.anna@libero.it

Anna M Pietroboni
Michela Caprioli
Tiziana Carandini
Marta Scarioni
Laura Ghezzi
Andrea Arighi
Claudia Cinnante
Milena A De Riz
Paola Basilico
Annalisa Colombi
Fabio Triulzi
Elio Scarpini
Daniela Galimberti
Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy/University of Milan, Dino Ferrari Center, Milan, Italy

Sara Cioffi
Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

Chiara Fenoglio
University of Milan, Dino Ferrari Center, Milan, Italy

Emanuela Oldoni
Laboratory for Neuroimmunology, Department of Neurosciences, KU Leuven, Leuven, Belgium

Giorgio G Fumagalli
Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy/University of Milan, Dino Ferrari Center, Milan, Italy/Department of Neurosciences, Psychology, Drug Research and Child Health (NEUROFARBA), University of Florence, Florence, Italy/University of Milan, Milan, Italy

Giovanni Giulietti
Laura Serra
Neuroimaging Laboratory, IRCCS Santa Lucia Foundation, Rome, Italy

Introduction

Multiple sclerosis (MS) is the most common chronic inflammatory disease of the central nervous system (CNS). Although traditionally regarded as a white matter (WM) demyelinating disease, axonal loss is critically involved in MS pathophysiology since early clinical stages.[1,2] The mechanisms underlying axonal damage, however, are not entirely clear and no reliable prognostic biomarker of disease progression is currently available.

Magnetic resonance imaging (MRI) is an invaluable tool for the diagnostic work-up of MS patients.[3,4] However, no strong correlation has been found between conventional MRI measures and clinical outcomes of progression.[4,5]

When taking into account the hypothesis of neurodegeneration as a major contributor to MS disability, β-amyloid$_{1–42}$ (Aβ) has recently become an interesting candidate for its putative role in this process. Amyloid precursor protein (APP) has been detected in MS plaques with a higher APP immunoreactivity in actively demyelinating than in chronic lesions, thus indicating a modification of APP metabolism across disease stages.[6] Moreover, APP was found upregulated in both acute and chronic MS lesions and has been regarded as a sensitive marker of axonal damage.[7,8] Reduced cerebrospinal fluid (CSF) Aβ levels have already been reported in MS patients[9–12] although the interpretation of these findings remains controversial.[13] In addition, a previous study on the mouse model of MS suggested a possible protective role of increased serum Aβ levels.[14] On

European Journal of Nuclear Medicine and Molecular Imaging (2019) 46:280–287
https://doi.org/10.1007/s00259-018-4182-1

ORIGINAL ARTICLE

Amyloid PET as a marker of normal-appearing white matter early damage in multiple sclerosis: correlation with CSF β-amyloid levels and brain volumes

Anna M. Pietroboni[1,2,3] · Tiziana Carandini[1,2,3] · Annalisa Colombi[1,2,3] · Matteo Mercurio[1] · Laura Ghezzi[1,2,3] · Giovanni Giulietti[4] · Marta Scarioni[1,2,3] · Andrea Arighi[1,2,3] · Chiara Fenoglio[2] · Milena A. De Riz[1,2,3] · Giorgio G. Fumagalli[1,2,3,5] · Paola Basilico[1,2,3] · Maria Serpente[2] · Marco Bozzali[4,6] · Elio Scarpini[1,2,3] · Daniela Galimberti[1,2,3] · Giorgio Marotta[1,2]

Received: 24 July 2018 / Accepted: 25 September 2018 / Published online: 21 October 2018

Abstract

Purpose The disease course of multiple sclerosis (MS) is unpredictable, and reliable prognostic biomarkers are needed. Positron emission tomography (PET) with β-amyloid tracers is a promising tool for evaluating white matter (WM) damage and repair. Our aim was to investigate amyloid uptake in damaged (DWM) and normal-appearing WM (NAWM) of MS patients, and to evaluate possible correlations between cerebrospinal fluid (CSF) β-amyloid$_{1-42}$ (Aβ) levels, amyloid tracer uptake, and brain volumes.

Methods Twelve MS patients were recruited and divided according to their disease activity into active and non-active groups. All participants underwent neurological examination, neuropsychological testing, lumbar puncture, brain magnetic resonance (MRI) imaging, and ^{18}F-florbetapir PET. Aβ levels were determined in CSF samples from all patients. MRI and PET images were co-registered, and mean standardized uptake values (SUV) were calculated for each patient in the NAWM and in the DWM. To calculate brain volumes, brain segmentation was performed using statistical parametric mapping software. Nonparametric statistical analyses for between-group comparisons and regression analyses were conducted.

Results We found a lower SUV in DWM compared to NAWM ($p < 0.001$) in all patients. Decreased NAWM-SUV was observed in the active compared to non-active group ($p < 0.05$). Considering only active patients, NAWM volume correlated with NAWM-SUV ($p = 0.01$). Interestingly, CSF Aβ concentration was a predictor of both NAWM-SUV ($r = 0.79$; $p = 0.01$) and NAWM volume ($r = 0.81$, $p = 0.01$).

Conclusions The correlation between CSF Aβ levels and NAWM-SUV suggests that the predictive role of β-amyloid may be linked to early myelin damage and may reflect disease activity and clinical progression.

Keywords PET · Amyloid tracer · Florbetapir · Multiple sclerosis · Amyloid · White matter

Anna M. Pietroboni, Tiziana Carandini, Daniela Galimberti, and Giorgio Marotta contributed equally to this work.

✉ Anna M. Pietroboni
pb.anna@libero.it

1 Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Via F. Sforza 35, 20122 Milan, Italy

2 University of Milan, Milan, Italy

3 Dino Ferrari Center, Milan, Italy

4 Neuroimaging Laboratory, IRCCS Santa Lucia Foundation, Rome, Italy

5 Department of Neurosciences, Psychology, Drug Research and Child Health (NEUROFARBA), University of Florence, Florence, Italy

6 Department of Neuroscience, Brighton and Sussex Medical School, University of Sussex, Brighton, UK

Introduction

Multiple sclerosis (MS) is the most common chronic inflammatory disease of the central nervous system (CNS) [1], whose demyelination is the pathological hallmark. MS is characterized by inflammation, axonal damage, and neurodegeneration [2]. The factors that promote spontaneous remyelination or determine axonal and neuronal loss remain poorly understood [3], and currently there are no reliable prognostic biomarkers of disease progression.

Magnetic resonance imaging (MRI) is the most widely used technique for identifying the demyelinating lesions, especially in the white matter (WM). However, no strong correlation exists between conventional MRI measures, such as T2- and T1-weighted lesion loads, and risk of disease progression,

Neuroimage
E L S E V I E R

Spatiotemporal analysis for detection of pre-symptomatic shape changes in neurodegenerative diseases: Initial application to the GENFI cohort

Claire Cury, Stanley Durrleman, [...], and Genetic FTD Initiative, GENFI

Additional article information

Associated Data

Supplementary Materials

Abstract

Brain atrophy as measured from structural MR images, is one of the primary imaging biomarkers used to track neurodegenerative disease progression. In diseases such as frontotemporal dementia or Alzheimer's disease, atrophy can be observed in key brain structures years before any clinical symptoms are present. Atrophy is most commonly captured as volume change of key structures and the shape changes of these structures are typically not analysed despite being potentially more sensitive than summary volume statistics over the entire structure.

In this paper we propose a spatiotemporal analysis pipeline based on Large Diffeomorphic Deformation Metric Mapping (LDDMM) to detect shape changes from volumetric MRI scans. We applied our framework to a cohort of individuals with genetic variants of frontotemporal dementia and healthy controls from the Genetic FTD Initiative (GENFI) study. Our method, take full advantage of the LDDMM framework, and relies on the creation of a population specific average spatiotemporal trajectory of a relevant brain structure of interest, the thalamus in our case. The residuals from each patient data to the average spatiotemporal trajectory are then clustered and studied to assess when presymptomatic mutation carriers differ from healthy control subjects.

We found statistical differences in shape in the anterior region of the thalamus at least five years before the mutation carrier subjects develop any clinical symptoms. This region of the thalamus has been shown to be predominantly connected to the frontal lobe, consistent with the pattern of cortical atrophy seen in the disease.

Keywords: Shape analysis, Clustering, Computational anatomy, Thalamus, Spatiotemporal geodesic regression, Parallel transport

Graphical abstract

[i]Dept. of Pathophysiology and Transplantation, "Dino Ferrari" Center, University of Milan, Fondazione C Granda, IRCCS Ospedale Maggiore Policlinico, Milan, Italy
[j]Cognitive Neurology Research Unit, Sunnybrook Health Sciences Centre, Hurvitz Brain Sciences Research Program, Sunnybrook Research Institute, Department of Medicine, University of Toronto, Canada
[k]Tanz Centre for Research in Neurodegenerative Diseases, University of Toronto, Canada
[l]University of Cambridge, United Kingdom
[m]Karolinska Institutet, Stockholm, Sweden
[n]Karolinska Institutet, Department NVS, Center for Alzheimer Research, Division of Neurogeriatrics, Sweden
[o]Department of Geriatric Medicine, Karolinska University Hospital, Stockholm, Sweden
[p]Instituto Neurologico Carlo Besta, Milan, Italy
[q]IRCCS San Giovanni di Dio Fatebenefratelli Brescia, Italy
[r]Université Laval, Quebec, Canada
[s]University of Western Ontario, Ontario, Canada
[t]Faculdade de Medicina, Universidade de Lisboa, Portugal
[u]Department of Neurosciences, Psychology, Drug Research and Child Health (NEUROFARBA), University of Florence, Florence, Italy
[v]IRCCS Don Gnocchi, Firenze, Italy
Claire Cury: moc.liamg@orp.yruc.erialc
*Corresponding author. VISAGES team, IRISA-Inria Bretagne Atlantique, Rennes, France. moc.liamg@orp.yruc.erialc
[1]List of consortium members in appendix.
Received 2018 Aug 5; Revised 2018 Nov 15; Accepted 2018 Nov 30.

References

Beg M.F., Miller M.I., Trouvé A., Younes L. Computing large deformation metric mappings via geodesic flows of diffeomorphisms. Int. J. Comput. Vis. 2005;61(2):139–157. [Google Scholar]
Behrens T.E.J., Johansen-Berg H., Woolrich M.W., Smith S.M., Wheeler-Kingshott C. a. M., Boulby P.A., Barker G.J., Sillery E.L., Sheehan K., Ciccarelli O., Thompson A.J., Brady J.M., Matthews P.M. Non-invasive mapping of connections between human thalamus and cortex using diffusion imaging. Nat. Neurosci. 2003;6(7):750–757. [PubMed] [Google Scholar]
Benzinger T.L.S., Blazey T., Jack C.R. Regional variability of imaging biomarkers in autosomal dominant Alzheimer's disease. Proc. Natl. Acad. Sci. Unit. States Am. 2013;110(47) E4502–9. [PMC free article] [PubMed] [Google Scholar]
Boccardi M., Bresciani L., Geroldi C., Beltramello A., Frisoni G.B., Laakso M.P. Clinical characteristics of frontotemporal patients with symmetric brain atrophy. Eur. Arch. Psychiatr. Clin. Neurosci. 2002;252(5):235–239. [PubMed] [Google Scholar]
Bocchetta M., Gordon E., Cardoso M.J., Modat M., Ourselin S., Warren J.D., Rohrer J.D. Thalamic atrophy in frontotemporal dementia - not just a C9orf72 problem. Neuroimage Clin. 2018;18:675–681. [PMC free article] [PubMed] [Google Scholar]
Cardoso M.J., Leung K., Modat M., Keihaninejad S., Cash D., Barnes J., Fox N.C., Ourselin S., for ADNI STEPS: similarity and truth estimation for propagated segmentations and its application to hippocampal segmentation and brain parcelation. Med. Image Anal. 2013;17(6):671–684. [PubMed] [Google Scholar]
Cash D.M., Bocchetta M., Thomas D.L., Dick K.M., van Swieten J.C., Borroni B., Galimberti D., Masellis M., Tartaglia M.C., Rowe J.B., Graff C., Tagliavini F., Frisoni G.B., Laforce J., Robert,

Neurobiology of Aging 77 (2019) 169–177

Contents lists available at ScienceDirect

Neurobiology of Aging

journal homepage: www.elsevier.com/locate/neuaging

Functional network resilience to pathology in presymptomatic genetic frontotemporal dementia

Timothy Rittman [a,*], Robin Borchert [a], Simon Jones [a], John van Swieten [b], Barbara Borroni [c], Daniela Galimberti [d], Mario Masellis [e,f,g], Maria Carmela Tartaglia [h], Caroline Graff [i,j], Fabrizio Tagliavini [k], Giovanni B. Frisoni [l,m], Robert Laforce Jr [n], Elizabeth Finger [o], Alexandre Mendonça [p], Sandro Sorbi [q,r], Jonathan D. Rohrer [s], James B. Rowe [a], The Genetic Frontotemporal Dementia Initiative (GENFI)

[a] Department of Clinical Neurosciences, University of Cambridge, Cambridge, UK
[b] Alzheimercentrum, Erasmus Medical Center, Rotterdam, the Netherlands
[c] Department of Clinical and Experimental Sciences, University of Brescia, Italy
[d] Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, University of Milan, Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Milan, Italy
[e] Cognitive Neurology Research Unit, Sunnybrook Health Sciences Centre, Toronto, Canada
[f] Hurvitz Brain Sciences Research Program, Sunnybrook Research Institute, Toronto, Canada
[g] Department of Medicine, University of Toronto, Toronto, Canada
[h] Tanz Centre for Research in Neurodegenerative Diseases, University of Toronto, Toronto, Canada
[i] Department NVS, Center for Alzheimer Research, Division of Neurogeriatrics, Karolinska Institutet, Stockholm, Sweden
[j] Department of Geriatric Medicine, Karolinska University Hospital, Stockholm, Sweden
[k] Istituto Neurologico Carlo Besta, Milan, Italy
[l] Department of Psychiatry, University Hospitals and University of Geneva, Geneva, Switzerland
[m] Neuroimaging and Epidemiology Unit, IRCCS San Giovanni di Dio Fatebenefratelli Brescia, Brescia, Italy
[n] Faculty of Medicine, Université Laval, Quebec, Canada
[o] Department of Clinical Neurological Sciences, University of Western Ontario, Ontario, Canada
[p] Faculdade de Medicina, Universidade de Lisboa, Lisboa, Portugal
[q] Department of Neurosciences, Psychology, Drug Research and Child Health (NEUROFARBA), University of Florence, Florence, Italy
[r] IRCCS Don Gnocchi, Florence, Italy
[s] Dementia Research Centre, Department of Neurodegenerative Disease, UCL Institute of Neurology, Queen Square, London, UK

ARTICLE INFO

Article history:
Received 22 February 2018
Received in revised form 23 December 2018
Accepted 24 December 2018
Available online 4 January 2019

Keywords:
Frontotemporal dementia
Genetics
Connectivity
Functional imaging
Cognition

ABSTRACT

The presymptomatic phase of neurodegenerative diseases are characterized by structural brain changes without significant clinical features. We set out to investigate the contribution of functional network resilience to preserved cognition in presymptomatic genetic frontotemporal dementia. We studied 172 people from families carrying genetic abnormalities in C9orf72, MAPT, or PGRN. Networks were extracted from functional MRI data and assessed using graph theoretical analysis. We found that despite loss of both brain volume and functional connections, there is maintenance of an efficient topological organization of the brain's functional network in the years leading up to the estimated age of frontotemporal dementia symptom onset. After this point, functional network efficiency declines markedly. Reduction in connectedness was most marked in highly connected hub regions. Measures of topological efficiency of the brain's functional network and organization predicted cognitive dysfunction in domains related to symptomatic frontotemporal dementia and connectivity correlated with brain volume loss in frontotemporal dementia. We propose that maintaining the efficient organization of the brain's functional network supports cognitive health even as atrophy and connectivity decline presymptomatically.

1. Introduction

Many neurodegenerative dementias begin their neuropathology years or even decades before the onset of symptoms. The evidence of presymptomatic pathology comes from changes in structural

* Corresponding author at: Department of Clinical Neurosciences, Cambridge Biomedical Campus, Herchel Smith Building, Robinson Way, Cambridge CB2 0SZ, UK. Tel: +44 0 1223 331515.
E-mail address: tr332@medschl.cam.ac.uk (T. Rittman).

https://doi.org/10.1016/j.neurobiolaging.2018.12.009

ARTICLES
https://doi.org/10.1038/s41588-019-0358-2
nature genetics

Genetic meta-analysis of diagnosed Alzheimer's disease identifies new risk loci and implicates Aβ, tau, immunity and lipid processing

Risk for late-onset Alzheimer's disease (LOAD), the most prevalent dementia, is partially driven by genetics. To identify LOAD risk loci, we performed a large genome-wide association meta-analysis of clinically diagnosed LOAD (94,437 individuals). We confirm 20 previous LOAD risk loci and identify five new genome-wide loci (*IQCK*, *ACE*, *ADAM10*, *ADAMTS1*, and *WWOX*), two of which (*ADAM10*, *ACE*) were identified in a recent genome-wide association (GWAS)-by-familial-proxy of Alzheimer's or dementia. Fine-mapping of the human leukocyte antigen (HLA) region confirms the neurological and immune-mediated disease haplotype HLA-DR15 as a risk factor for LOAD. Pathway analysis implicates immunity, lipid metabolism, tau binding proteins, and amyloid precursor protein (APP) metabolism, showing that genetic variants affecting APP and Aβ processing are associated not only with early-onset autosomal dominant Alzheimer's disease but also with LOAD. Analyses of risk genes and pathways show enrichment for rare variants ($P = 1.32 \times 10^{-7}$), indicating that additional rare variants remain to be identified. We also identify important genetic correlations between LOAD and traits such as family history of dementia and education.

Our previous work identified 19 genome-wide-significant common variant signals in addition to *APOE* that influence risk for LOAD (onset age > 65 years)[1]. These signals, combined with 'subthreshold' common variant associations, account for ~31% of the genetic variance of LOAD[2], leaving the majority of genetic risk uncharacterized[3]. To search for additional signals, we conducted a GWAS meta-analysis of non-Hispanic Whites (NHW) by using a larger Stage 1 discovery sample (17 new, 46 total datasets; $n = 21{,}982$ cases, 41,944 cognitively normal controls) from our group, the International Genomics of Alzheimer's Project (IGAP) (composed of four consortia: Alzheimer Disease Genetics Consortium (ADGC), Cohorts for Heart and Aging Research in Genomic Epidemiology Consortium (CHARGE), The European Alzheimer's Disease Initiative (EADI), and Genetic and Environmental Risk in AD/Defining Genetic, Polygenic and Environmental Risk for Alzheimer's Disease Consortium (GERAD/PERADES) (Supplementary Tables 1 and 2, and Supplementary Note). To sample both common and rare variants (minor allele frequency (MAF) ≥ 0.01 and MAF < 0.01, respectively), we imputed the discovery datasets by using a 1,000 Genomes reference panel consisting of 36,648,992 single-nucleotide polymorphisms (SNPs), 1,380,736 insertions/deletions, and 13,805 structural variants. After quality control, 9,456,058 common variants and 2,024,574 rare variants were selected for analysis. Genotype dosages were analyzed within each dataset, and then combined with meta-analysis (Supplementary Fig. 1 and Supplementary Tables 1–3).

Results

Meta-analysis of Alzheimer's disease GWAS. The Stage 1 discovery meta-analysis produced 12 loci with genome-wide significance ($P \leq 5 \times 10^{-8}$) (Table 1), all of which are previously described[1,4–11]. Genomic inflation factors (λ) were slightly inflated (λ median = 1.05; λ regression = 1.09; see Supplementary Figure 2 for a quantile–quantile (QQ) plot); however, univariate linkage disequilibrium score (LDSC) regression[12,13] estimates indicated that the majority of this inflation was due to a polygenic signal, with the intercept being close to 1 (1.026, s.e.m. = 0.006). The observed heritability (h^2) of LOAD was estimated at 0.071 (0.011) using LDSC. Stage 1 meta-analysis was first followed by Stage 2, using the I-select chip we previously developed in Lambert et al.[1] (including 11,632 variants, $n = 18{,}845$; Supplementary Table 4) and finally Stage 3A ($n = 11{,}666$) or Stage 3B ($n = 30{,}511$) (for variants in regions not well captured in the I-select chip) (see Supplementary Figure 1 for the workflow). The final sample was 35,274 clinical and autopsy-documented Alzheimer's disease cases and 59,163 controls.

Meta-analysis of Stages 1 and 2 produced 21 genome-wide-significant associations ($P \leq 5 \times 10^{-8}$) (Table 1 and Fig. 1), 18 of which were previously reported as genome-wide significant in Lambert et al.[1]. Three other signals were not initially described in the initial IGAP GWAS: the rare R47H *TREM2* coding variant previously reported by others[7,8,14]; *ECHDC3* (rs7920721; NC_000010.10: g.11720308A>G), which was recently identified as a potential genome-wide-significant Alzheimer's disease risk locus in several studies[15–17], and *ACE* (rs138190086; NC_000017.10: g.61538148G>A) (Supplementary Figs. 3 and 4). In addition, seven signals showed suggestive association with $P < 5 \times 10^{-7}$ (closest genes: *ADAM10, ADAMTS1, ADAMTS20, IQCK*, *MIR142/TSPOAP1-AS1*, *NDUFAF6,* and *SPPL2A*) (Supplementary Figs. 5–11). Stage 3A and meta-analysis of all three stages for these nine signals (excluding the *TREM2* signal; see Supplementary Table 5 for the variant list) identified five genome-wide-significant loci. In addition to *ECHDC3*, this included four new genome-wide Alzheimer's disease risk signals at *IQCK, ADAMTS1, ACE,* and *ADAM10* (Table 2). *ACE* and *ADAM10* were previously reported as Alzheimer's disease candidate genes[18–22] but were not replicated in some subsequent studies[23–25]. A recent GWAS using family history of Alzheimer's disease or dementia as a proxy[26] also identified these two risk loci, suggesting that while use of proxy Alzheimer's disease/dementia cases introduces less sensitivity and specificity for true Alzheimer's disease signals overall in comparison to clinically diagnosed Alzheimer's disease, proxy studies can identify disease-relevant associations. Two of the four other signals approached genome-wide significance: *miR142/TSPOAP1-AS1* ($P = 5.3 \times 10^{-8}$) and *NDUFAF6* ($P = 9.2 \times 10^{-8}$) (Table 2). Stage 3A also extended the analysis of two loci (*NME8* and *MEF2C*) that were previously genome-wide significant in our 2013 meta-analysis. These loci were

A full list of authors and affiliations appears at the end of the paper.

[1]John P. Hussman Institute for Human Genomics, University of Miami Miller School of Medicine, Miami, FL, USA. [2]Inserm, U1167, RID-AGE-Risk Factors and Molecular Determinants of Aging-Related Diseases, Lille, France. [3]Institut Pasteur de Lille, Lille, France. [4]Univ. Lille, U1167-Excellence Laboratory LabEx DISTALZ, Lille, France. [5]Division of Psychological Medicine and Clinical Neurosciences, MRC Centre for Neuropsychiatric Genetics and Genomics, Cardiff University, Cardiff, UK. [6]UK Dementia Research Institute at Cardiff, Cardiff University, Cardiff, UK. [7]Cardiovascular Health Research Unit, Department of Medicine, University of Washington, Seattle, WA, USA. [8]Department of Biostatistics and Epidemiology/Center for Clinical Epidemiology and Biostatistics, University of Pennsylvania Perelman School of Medicine, Philadelphia, PA, USA. [9]Centre National de Recherche en Génomique Humaine, Institut de Biologie François Jacob, CEA, Université Paris-Saclay, and LabEx GENMED, Evry, France. [10]Department of Epidemiology, Erasmus Medical Center, Rotterdam, the Netherlands. [11]Penn Neurodegeneration Genomics Center, Department of Pathology and Laboratory Medicine, University of Pennsylvania Perelman School of Medicine, Philadelphia, PA, USA. [12]Framingham Heart Study, Framingham, MA, USA. [13]Department of Neurology, Boston University School of Medicine, Boston, MA, USA. [14]Neurodegenerative Brain Diseases Group, Center for Molecular Neurology, VIB, Antwerp, Belgium. [15]Laboratory for Neurogenetics, Institute Born-Bunge, University of Antwerp, Antwerp, Belgium. [16]Icelandic Heart Association, Kopavogur, Iceland. [17]Research Center and Memory Clinic of Fundació ACE, Institut Català de Neurociències Aplicades-Universitat Internacional de Catalunya, Barcelona, Spain. [18]Centro de Investigación Biomédica en Red de Enfermedades Neurodegenerativas, Instituto de Salud Carlos III, Madrid, Spain. [19]Department of Biostatistics, Boston University School of Public Health, Boston, MA, USA. [20]Institute of Biomedicine, University of Eastern Finland, Kuopio, Finland. [21]Department of Neurology, Kuopio University Hospital, Kuopio, Finland. [22]Taub Institute on Alzheimer's Disease and the Aging Brain, Department of Neurology, Columbia University, New York, NY, USA. [23]Gertrude H. Sergievsky Center, Columbia University, New York, NY, USA. [24]Department of Neurology, Columbia University, New York, NY, USA. [25]UMR 894, Center for Psychiatry and Neuroscience, Inserm, Université Paris Descartes, Paris, France. [26]Institute of Human Genetics, University of Bonn, Bonn, Germany. [27]Department of Genomics, Life & Brain Center, University of Bonn, Bonn, Germany. [28]Division of Medical Genetics, University Hospital and Department of Biomedicine, University of Basel, Basel, Switzerland. [29]School of Biotechnology, Dublin City University, Dublin, Ireland. [30]Department of Family Medicine, University of Washington, Seattle, WA, USA. [31]Department of Epidemiology, University of Washington, Seattle, WA, USA. [32]Dementia Research Centre, Department of Neurodegenerative Disease, UCL Institute of Neurology, London, UK. [33]Department of Biostatistics, University of Michigan, Ann Arbor, MI, USA. [34]Faculty of Medicine, University of Iceland, Reykjavik, Iceland. [35]Brown Foundation Institute of Molecular Medicine, University of Texas Health Sciences Center at Houston, Houston, TX, USA. [36]Section of Neuroscience and Clinical Pharmacology, Department of Biomedical Sciences, University of Cagliari, Cagliari, Italy. [37]UK Dementia Research Institute at UCL, Department of Neurodegenerative Disease, UCL Institute of Neurology, London, UK. [38]Neurology Service and CIBERNED, 'Marqués de Valdecilla' University Hospital (University of Cantabria and IDIVAL), Santander, Spain. [39]Department of Basic and Clinical Neuroscience, Institute of Psychiatry, Psychology and Neuroscience, King's College London, London, UK. [40]Department of Immunology, Hospital Universitario Doctor Negrín, Las Palmas de Gran Canaria, Spain. [41]Department of Neurology, Medical School, Aristotle University of Thessaloniki, Thessaloniki, Greece. [42]Department of Medicine, University of Washington, Seattle, WA, USA. [43]Normandie University, UNIROUEN, Inserm U1245, and Rouen University Hospital, Department of Neurology, Department of Genetics and CNR-MAJ, Normandy Center for Genomic and Personalized Medicine, Rouen, France. [44]Department of Neurodegenerative Disease, MRC Prion Unit at UCL, Institute of Prion Diseases, London, UK. [45]Centre for Public Health, University of Iceland, Reykjavik, Iceland. [46]Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, Neurodegenerative Diseases Unit, Milan, Italy. [47]University of Milan, Centro Dino Ferrari, Milan, Italy. [48]Clinical Division of Neurogeriatrics, Department of Neurology, Medical University Graz, Graz, Austria. [49]Institute for Medical Informatics, Statistics and Documentation, Medical University of Graz, Graz, Austria. [50]Institute for Computational Biology, Department of Population & Quantitative Health Sciences, Case Western Reserve University, Cleveland, OH, USA. [51]Department of Psychiatry and Psychotherapy, University of Erlangen-Nuremberg, Erlangen, Germany. [52]Program in Medical and Population Genetics, Broad Institute, Cambridge, MA, USA. [53]Department of Pharmacology and Neuroscience, University of North Texas Health Science Center, Fort Worth, TX, USA. [54]Laboratory for Neurochemistry and Behavior, Institute Born-Bunge, University of Antwerp, Antwerp, Belgium. [55]Department of Neurology and Memory Clinic, Hospital Network Antwerp, Antwerp, Belgium. [56]Department of Psychiatry and Psychotherapy, University Hospital, Saarland, Germany. [57]Department of Psychiatry, University of Texas Southwestern Medical Center, Dallas, TX, USA. [58]Laboratory for Cognitive Neurology, Department of Neurology, University Hospital and University of Leuven, Leuven, Belgium. [59]Department of Neurology, Johns Hopkins University, Baltimore, MD, USA. [60]National Alzheimer's Coordinating Center, University of Washington, Seattle, WA, USA. [61]Department of Psychiatry, Martin Luther University Halle-Wittenberg, Halle, Germany. [62]Department of Epidemiology, Harvard T.H. Chan School of Public Health, Harvard University, Boston, MA, USA. [63]Department of Psychiatry, Massachusetts General Hospital/Harvard Medical School, Boston, MA, USA. [64]Department of Psychiatry, University of Oxford, Oxford, UK. [65]Laboratory of Epidemiology and Population Sciences, National Institute on Aging, Bethesda, MD, USA. [66]Alzheimer's Disease and Memory Disorders Center, Baylor College of Medicine, Houston, TX, USA. [67]Division of Clinical Neurosciences, School of Medicine, University of Southampton, Southampton, UK. [68]Section of Computational Biomedicine, Department of Medicine, Boston University School of Medicine, Boston, MA, USA. [69]Office of Strategy and Measurement, University of North Texas Health Science Center, Fort Worth, TX, USA. [70]C.S. Kubik Laboratory for Neuropathology, Massachusetts General Hospital, Charlestown, MA, USA. [71]Theme Aging, Unit for Hereditary Dementias, Karolinska University Hospital-Solna, Stockholm, Sweden. [72]Karolinska Institutet, Department of Neurobiology, Care Sciences and Society, Alzheimer Research Center, Division of Neurogeriatrics, Solna, Sweden. [73]German Center for Neurodegenerative Diseases, Bonn, Germany. [74]Department of Psychiatry and Psychotherapy, University of Bonn, Bonn, Germany. [75]Department of Pathology and Laboratory Medicine, Indiana University, Indianapolis, IN, USA. [76]Department of Public Health and Caring Sciences/Geriatrics, Uppsala University, Uppsala, Sweden. [77]Department for Neurodegenerative Diseases and Geriatric Psychiatry, University Hospital Bonn, Bonn, Germany. [78]Department of Surgery, University of Texas Southwestern Medical Center, Dallas, TX, USA. [79]Institute for Medical Informatics, Biometry and Epidemiology, University Hospital of Essen, University Duisburg-Essen, Essen, Germany. [80]Department of Psychiatry, University of Pittsburgh, Pittsburgh, PA, USA. [81]Department of Human Genetics, University of Pittsburgh, Pittsburgh, PA, USA. [82]Alzheimer's Disease Research Center, University of Pittsburgh, Pittsburgh, PA, USA. [83]Institute of Genetics, Queen's Medical Centre, University of Nottingham, Nottingham, UK. [84]Department of Neurology, Albert Einstein College of Medicine, Bronx, NY, USA. [85]Section of Neuroscience, DIMEC-University of Parma, Parma, Italy. [86]FERB-Alzheimer Center, Gazzaniga (Bergamo), Italy. [87]Department of Pathology, University of Washington, Seattle, WA, USA. [88]Elderly and Psychiatric Disorders Department, Medical University of Lodz, Lodz, Poland. [89]Mercer's Institute for Research on Aging, St. James's Hospital and Trinity College, Dublin, Ireland. [90]St. James's Hospital and Trinity College, Dublin, Ireland. [91]Kaiser Permanente Washington Health Research Institute, Seattle, WA, USA. [92]A.I. Virtanen Institute for Molecular Sciences, University of Eastern Finland, Kuopio, Finland. [93]Departments of Medicine, Geriatrics, Gerontology and Neurology, University of Mississippi Medical Center, Jackson, MS, USA. [94]Interdisciplinary Department of Medicine, Geriatric Medicine and Memory Unity, University of Bari, Bari, Italy. [95]Department of Neurology, University of Washington, Seattle, WA, USA. [96]Department of Geriatrics, Center for Aging Brain, University of Bari, Bari, Italy. [97]Fundació per la Recerca Biomèdica i Social Mútua Terrassa, Terrassa, Barcelona, Spain. [98]Memory Disorders Unit, Department of Neurology, Hospital Universitari Mutua de Terrassa, Terrassa, Barcelona, Spain. [99]Department of Internal Medicine, Erasmus University Medical Center, Rotterdamt, the Netherlands. [100]Netherlands Consortium on Health Aging and National Genomics Initiative, Leiden, the Netherlands. [101]Department of Neurology, Mayo Clinic, Rochester, MN, USA. [102]CHU Lille, Memory Center of Lille (Centre Mémoire de Ressources et de Recherche), Lille, France. [103]Department of Clinical and Behavioral Neurology, Experimental Neuropsychobiology Laboratory, IRCCS Santa Lucia Foundation, Rome, Italy. [104]School of Public Health, Human Genetics Center, University of Texas Health Science Center at Houston, Houston, TX, USA. [105]Human Genome

NeuroImage 189 (2019) 645–654

Contents lists available at ScienceDirect

NeuroImage

journal homepage: www.elsevier.com/locate/neuroimage

The inner fluctuations of the brain in presymptomatic Frontotemporal Dementia: The chronnectome fingerprint☆

Enrico Premi [a,b], Vince D. Calhoun [c], Matteo Diano [d,e], Stefano Gazzina [a], Maura Cosseddu [a], Antonella Alberici [a], Silvana Archetti [f], Donata Paternicò [a], Roberto Gasparotti [g], John van Swieten [h], Daniela Galimberti [i], Raquel Sanchez-Valle [j], Robert Laforce Jr. [k], Fermin Moreno [l], Matthis Synofzik [m], Caroline Graff [n], Mario Masellis [o], Maria Carmela Tartaglia [p], James Rowe [q], Rik Vandenberghe [r], Elizabeth Finger [s], Fabrizio Tagliavini [t], Alexandre de Mendonça [u], Isabel Santana [v], Chris Butler [w], Simon Ducharme [x], Alex Gerhard [y], Adrian Danek [z], Johannes Levin [z], Markus Otto [aa], Giovanni Frisoni [ab,ac], Stefano Cappa [ab], Sandro Sorbi [ad,ae], Alessandro Padovani [a], Jonathan D. Rohrer [af], Barbara Borroni [a,*], on behalf Genetic FTD Initiative, GENFI

[a] Centre for Neurodegenerative Disorders, Neurology Unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy
[b] Stroke Unit, Azienda Socio Sanitaria Territoriale Spedali Civili, Spedali Civili Hospital, Brescia, Italy
[c] The Mind Research Network, Albuquerque USA, Department of Electrical and Computer Engineering, University of New Mexico, Albuquerque, USA
[d] Department of Psychology, University of Turin, Turin, Italy
[e] Department of Medical and Clinical Psychology, CoRPS – Center of Research on Psychology in Somatic Diseases, Tilburg University, the Netherlands
[f] Biotechnology Laboratory, Department of Diagnostic, Spedali Civili Hospital, Brescia, Italy
[g] Neuroradiology Unit, University of Brescia, Italy
[h] Department of Neurology, Erasmus Medical Center, Rotterdam, Netherlands
[i] Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, University of Milan, Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Milan, Italy
[j] Neurology Department, Hospital Clinic, Institut d'Investigacions Biomèdiques, Barcelona, Spain
[k] Clinique Interdisciplinaire de Mémoire, Département des Sciences Neurologiques, CHU de Québec, and Faculté de Médecine, Université Laval, QC, Canada
[l] Department of Neurology, Hospital Universitario Donostia, San Sebastian, Gipuzkoa, Spain
[m] Department of Cognitive Neurology, Center for Neurology and Hertie-Institute for Clinical Brain Research, Tübingen, Germany
[n] Karolinska Institutet, Department NVS, Center for Alzheimer Research, Division of Neurogenetics, Sweden
[o] LC Campbell Cognitive Neurology Research Unit, Sunnybrook Research Institute, Toronto, ON, Canada
[p] Toronto Western Hospital, Tanz Centre for Research in Neurodegenerative Disease, Toronto, ON, Canada
[q] Department of Clinical Neurosciences, University of Cambridge, Cambridge, UK
[r] Laboratory for Cognitive Neurology, Department of Neurosciences, KU Leuven, Leuven, Belgium
[s] Department of Clinical Neurological Sciences, University of Western Ontario, London, ON, Canada
[t] Fondazione Istituto di Ricovero e Cura a Carattere Scientifico Istituto Neurologico Carlo Besta, Milan, Italy
[u] Faculty of Medicine, University of Lisbon, Lisbon, Portugal
[v] Neurology Department, Centro Hospitalar e Universitário de Coimbra, Portugal
[w] Department of Clinical Neurology, University of Oxford, Oxford, UK
[x] Department of Neurology and Neurosurgery, McGill University, Montreal, Quebec, Canada
[y] Institute of Brain, Behaviour and Mental Health, The University of Manchester, Withington, Manchester, UK
[z] Neurologische Klinik und Poliklinik, Ludwig-Maximilians-Universität, Munich, German Center for Neurodegenerative Diseases (DZNE), Munich, Germany
[aa] Department of Neurology, University Hospital Ulm, Ulm, Germany
[ab] Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS) Istituto Centro San Giovanni di Dio Fatebenefratelli, Brescia, Italy
[ac] Memory Clinic and LANVIE-Laboratory of Neuroimaging of Aging, University Hospitals and University of Geneva, Geneva, Switzerland
[ad] Department of Neuroscience, Psychology, Drug Research and Child Health, University of Florence, Florence, Italy
[ae] Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS) "Don Gnocchi", Florence, Italy
[af] Dementia Research Centre, UCL Institute of Neurology, UK

☆ List of GENFI Consortium Members in Appendix.

* Corresponding author. Neurology Unit, University of Brescia, Piazza Spedali Civili 1, Brescia, 25125, Italy.
E-mail address: bborroni@inwind.it (B. Borroni).

https://doi.org/10.1016/j.neuroimage.2019.01.080
Received 17 October 2018; Received in revised form 27 January 2019; Accepted 31 January 2019
Available online 1 February 2019
1053-8119/

Journal of Alzheimer's Disease 67 (2019) 985–993
DOI 10.3233/JAD-180715
IOS Press

A Case with Early Onset Alzheimer's Disease, Frontotemporal Hypometabolism, ApoE Genotype ε4/ε4 and C9ORF72 Intermediate Expansion: A Treviso Dementia (TREDEM) Registry Case Report

Maurizio Gallucci[a,*], Carola Dell'Acqua[a], Cristina Bergamelli[a], Chiara Fenoglio[b], Maria Serpente[b], Daniela Galimberti[b,c], Vittorio Fiore[d], Stefano Medea[d], Michele Gregianin[d] and Maria Elena Di Battista[a]
[a]*Cognitive Impairment Center, Local Health Autority n.2 Marca Trevigiana, Treviso, Italy*
[b]*University of Milan, Dino Ferrari Center, Milan, Italy*
[c]*Fondazione IRCCS Ca' Granda, Ospedale Policlinico, Neurodegenerative Disease Unit, Milan, Italy*
[d]*Nuclear Medicine Unit, Local Health Autority n.2 Marca Trevigiana, Treviso, Italy*

Handling Associate Editor: Beatrice Arosio

Accepted 28 November 2018

Abstract. We report the case of a woman firstly referred to our Memory Clinic at the age of 61, following the development of cognitive complaints and difficulties in sustained attention. The investigation that was performed showed: predominant executive dysfunctions at the neuropsychological evaluation, with mild, partial and stable involvement of the memory domain; cortical and subcortical atrophy with well-preserved hippocampal structures at MRI; marked fronto-temporal and moderate parietal hypometabolism from ^{18}F-FDG PET study with a sparing of the posterior cingulate and precuneus; positivity of amyloid-β at ^{18}F-Flutemetamol PET; an hexanucleotide intermediate repeats expansion of *C9ORF72* gene (12//38 repeats) and ApoE genotype ε4/ε4. The patient was diagnosed with probable early onset frontal variant of Alzheimer's disease (AD), presenting with a major executive function impairment. The lack of specific areas of brain atrophy, as well as the failure to meet the clinical criteria for any frontotemporal dementia, drove us to perform the aforementioned investigations, which yielded our final diagnosis. The present case highlights the need to take into consideration a diagnosis of frontal variant of AD when the metabolic and the clinical picture are somehow dissonant.

Keywords: ApoE, *C9ORF72* HREs, early onset Alzheimer's disease, FDG PET, TREDEM Registry

INTRODUCTION

Alzheimer's disease (AD) is the most common form of progressive neurodegenerative syndrome, typically occurring in middle or late life [1]. The classical clinical phenotype comprises of episodic memory impairment together with deficits in other cognitive domains. Based on the evidence of peculiar presentations of AD, the term "atypical AD" has been postulated, indicating less usual but well described phenotypes that occur with AD pathology hallmarks [2,3]. According to the most recent criteria, these

*Correspondence to: Maurizio Gallucci, MD, Cognitive Impairment Center, Local Health Authority n.2 Marca Trevigiana, Treviso, Italy. E-mail: maurizio.gallucci@aulss2.veneto.it.

Dalmasso et al. *Translational Psychiatry* (2019)9:55
https://doi.org/10.1038/s41398-019-0394-9

Translational Psychiatry

ARTICLE Open Access

Transethnic meta-analysis of rare coding variants in *PLCG2*, *ABI3*, and *TREM2* supports their general contribution to Alzheimer's disease

Maria Carolina Dalmasso[1], Luis Ignacio Brusco[2,3,4], Natividad Olivar[2,3], Carolina Muchnik[5], Claudia Hanses[6], Esther Milz[7], Julian Becker[6], Stefanie Heilmann-Heimbach[8,9], Per Hoffmann[8,9,10], Federico A. Prestia[1], Pablo Galeano[1], Mariana Soledad Sanchez Avalos[11], Luis Eduardo Martinez[4], Mariana Estela Carulla[4], Pablo Javier Azurmendi[5], Cynthia Liberczuk[2], Cristina Fezza[5], Marcelo Sampaño[2], Maria Fierens[2], Guillermo Jemar[2], Patricia Solis[12], Nancy Medel[12], Julieta Lisso[12], Zulma Sevillano[12], Paolo Bosco[13], Paola Bossù[14], Gianfranco Spalletta[15], Daniela Galimberti[16], Michelangelo Mancuso[17], Benedetta Nacmias[18], Sandro Sorbi[18,19], Patrizia Mecocci[20], Alberto Pilotto[21], Paolo Caffarra[22,23], Francesco Panza[24], Maria Bullido[25,26,27], Jordi Clarimon[26,28], Pascual Sánchez-Juan[29], Eliecer Coto[30], Florentino Sanchez-Garcia[31], Caroline Graff[32,33], Martin Ingelsson[34], Céline Bellenguez[35,36,37], Eduardo Miguel Castaño[1], Claudia Kairiyama[4], Daniel Gustavo Politis[4], Silvia Kochen[12], Horacio Scaro[11], Wolfgang Maier[38,39], Frank Jessen[38,40], Carlos Alberto Mangone[2], Jean-Charles Lambert[35,36,37], Laura Morelli[1] and Alfredo Ramirez[7,39]

Abstract

Rare coding variants in *TREM2*, *PLCG2*, and *ABI3* were recently associated with the susceptibility to Alzheimer's disease (AD) in Caucasians. Frequencies and AD-associated effects of variants differ across ethnicities. To start filling the gap on AD genetics in South America and assess the impact of these variants across ethnicity, we studied these variants in Argentinian population in association with ancestry. *TREM2* (rs143332484 and rs75932628), *PLCG2* (rs72824905), and *ABI3* (rs616338) were genotyped in 419 AD cases and 486 controls. Meta-analysis with European population was performed. Ancestry was estimated from genome-wide genotyping results. All variants show similar frequencies and odds ratios to those previously reported. Their association with AD reach statistical significance by meta-analysis. Although the Argentinian population is an admixture, variant carriers presented mainly Caucasian ancestry. Rare coding variants in *TREM2*, *PLCG2*, and *ABI3* also modulate susceptibility to AD in populations from Argentina, and they may have a European heritage.

Correspondence: Laura Morelli (lmorelli@leloir.org.ar) or Alfredo Ramirez (alfredo.ramirez@uk-koeln.de)
[1]Laboratory of Amyloidosis and Neurodegeneration, Fundación Instituto Leloir-IIBBA-CONICET, Ciudad Autónoma de Buenos Aires (C.A.B.A.), Buenos Aires, Argentina
[2]Centro de Neuropsiquiatría y Neurología de la Conducta (CENECON), Facultad de Medicina, Universidad de Buenos Aires (UBA), C.A.B.A, Buenos Aires, Argentina
Full list of author information is available at the end of the article.
These authors contributed equally: Maria Carolina Dalmasso, Luis Ignacio Brusco, Laura Morelli, Alfredo Ramirez

Introduction

Alzheimer's disease (AD) is the most common form of dementia, and has an estimated genetic component of 60–80%[1]. Over the last decade, more than 20 loci containing common genetic variants (minor allele frequency (MAF) >5%) have been associated with AD[2]. The advent of new genetic sequencing technologies has enabled the identification of several rare variants (MAF <1%) with

Cytokine 116 (2019) 115–119

Contents lists available at ScienceDirect

Cytokine

journal homepage: www.elsevier.com/locate/cytokine

Inflammatory expression profile in peripheral blood mononuclear cells from patients with Nasu-Hakola Disease

D. Galimberti[a,b,*], C. Fenoglio[a], L. Ghezzi[a,b], M. Serpente[a], M. Arcaro[b], M. D'Anca[a], M. De Riz[b], A. Arighi[b], G.G. Fumagalli[b,c], A.M. Pietroboni[b], L. Piccio[d], E. Scarpini[a,b]

[a] University of Milan, Centro Dino Ferrari, Milan, Italy
[b] Fondazione IRCCS Cà Granda, Ospedale Maggiore Policlinico, Neurodegenerative Diseases Unit, Milan, Italy
[c] Department of Neurosciences, Psychology, Drug Research and Child Health (NEUROFARBA), University of Florence, Florence, Italy
[d] Department of Neurology, Washington University School of Medicine, St Louis, MO, USA

ARTICLE INFO

Keywords:
Nasu-Hakola Disease (NHD)
Triggering Receptor Expressed on Myeloid cells 2 (TREM2)
Inflammation
Cytokines
Chemokines
Expression
Peripheral Blood Mononuclear Cells (PBMC)

ABSTRACT

Homozygous mutations in Triggering Receptor Expressed on Myeloid cells 2 gene (*TREM2*) are one of the major causes of Nasu Hakola Disease (NHD). We analysed Peripheral Blood Mononuclear Cells (PBMC) profile of 164 inflammatory factors in patients with NHD carrying the *TREM2* Q33X mutation as compared with heterozygous and wild type individuals.

Several molecules related to bone formation and angiogenesis were altered in NHD compared to non-carriers: Bone Morphogenetic Protein (BMP)-1 mRNA levels were significantly increased in PBMC (2.32 fold-increase; $P = 0.01$), as were Transforming Growth Factor Beta (TGFB)3 levels (1.51 fold-increase; $P = 0.02$). Conversely, CXCL5 and Pro Platelet Basic Protein (PPBP) were strongly downregulated (−28.26, −9.85 fold-decrease over non-carriers, respectively, $P = 0.01$), as well as Platelet Factor 4 Variant 1 (PF4V1; −41.44, $P = 0.03$).

Among other inflammatory factors evaluated, Interleukin (IL)-15 and Tumor Necrosis Factor Superfamily Member (TNFSF)4 mRNA levels were decreased in NHD as compared with non-carriers (−2.25 and −3.87 fold-decrease, $P = 0.01$ and 0.001, respectively).

In heterozygous individuals, no significant differences were observed, apart from IL-15 mRNA levels, that were decreased at the same extent as NHD (−2.05 fold-decrease over non-carriers, $P = 0.002$).

We identified a signature in PBMC from patients with NHD consisting of strongly decreased mRNA levels of CXCL5, PPBP, PF4V1, mildly decreased IL-15 and TNFSF4 and mildly increased BMP-1 and TGFB3.

1. Introduction

Nasu-Hakola Disease (NHD), also known as Polycystic Lipomembranous Osteodysplasia with Sclerosing Leukoencephalopathy (PLOSL), is a recessively inherited rare disorder characterized by a combination of pre-senile frontal dementia and systemic bone cysts formation, leading to pathological fractures of the wrists and ankles after microtrauma [1–3]. NHD culminates in a profound dementia and death occurs by the age of 50. It is caused by mutations in Triggering Receptor Expressed on Myeloid cells 2 gene (*TREM2*) or TYRO protein tyrosine kinase binding protein (*TYROBP*), also known as DNAX-activating protein of 12 kDa (*DAP12*). These genes encode different domains of the same receptor signaling protein, involved in the activation of the immune response (namely TREM2/TYROBP signaling cascade). TREM2/TYROBP signaling is essential for the development of osteoclasts and dendritic cells [4] as well as for microglia activation, phagocytosis and survival [5].

TREM2 possesses an immunoglobulin superfamily domain [6] and is expressed in myeloid cells including microglia, monocyte-derived dendritic cells and macrophages [7]. It plays important roles in innate and adaptive immunity [8]. TREM2 promotes survival, proliferation and remodeling of the actin cytoskeleton, which regulates adhesion and migration [9]. TREM2 is involved in key signaling events related to the immune response and the phagocytic activity of microglia [10].

In mice models, TREM2 was found to be essential for microglia-mediated synaptic refinement during the early stages of brain development. In fact, the absence of *TREM2* resulted in impaired synapse elimination, accompanied by enhanced excitatory neurotransmission and reduced long-range functional connectivity [11]. Heterozygous variants of *TREM2* increase the risk of Alzheimer's disease (AD) and are involved in other neurodegenerative diseases (see [12] 2018 for review).

* Corresponding author.
E-mail address: daniela.galimberti@unimi.it (D. Galimberti).

https://doi.org/10.1016/j.cyto.2018.12.024
Received 19 September 2018; Received in revised form 14 November 2018; Accepted 31 December 2018
Available online 25 January 2019

Journal of Alzheimer's Disease 67 (2019) 1173–1179
DOI 10.3233/JAD-180723
IOS Press

Short Communication

Monozygotic Twins with Frontotemporal Dementia Due To Thr272fs *GRN* Mutation Discordant for Age At Onset

Giorgio Giulio Fumagalli[a,b,c,*], Luca Sacchi[a,b], Paola Basilico[a,b], Andrea Arighi[a,b], Tiziana Carandini[a,b], Marta Scarioni[a,b], Annalisa Colombi[a,b], Anna Pietroboni[a,b], Laura Ghezzi[a,b], Chiara Fenoglio[a,b], Maria Serpente[a,b], Marianna D'anca[a,b], Marina Arcaro[a,b], Matteo Mercurio[a,b], Fabio Triulzi[a,b], Elisa Scola[a,b], Giorgio Marotta[b], Elio Scarpini[a,b] and Daniela Galimberti[a,b]
[a]*University of Milan, "Dino Ferrari" Center, Milan, Italy*
[b]*Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy*
[c]*Department of Neurosciences, University of Florence, Psychology, Drug Research and Child Health (NEUROFARBA), Florence, Italy*

Handling Associate Editor: Laura Serra

Accepted 18 December 2018

Abstract. We report the case of two monozygotic twins with Thr272fs mutation in progranulin gene. Both patients developed frontotemporal dementia with 5 years difference in age at onset (Twin 1:73 years, Twin 2:68 years), with early behavioral, language, dysexecutive, and memory problems. They had the same formal education (5 years), but while Twin 1 dedicated more to social and leisure activity, Twin 2 worked all her life. At neuroimaging (MRI for Twin 1 and CT for Twin 2), they both showed asymmetric atrophy with left predominance. The two were discordant for total tau levels in cerebrospinal fluid, neuropsychological testing, and smoking habits. The description of the twins can help identify environmental factors that influence the onset and phenotype of frontotemporal dementia.

Keywords: Age at onset, atrophy, cerebrospinal fluid, frontotemporal dementia, progranulin (*GRN*), twins

INTRODUCTION

Frontotemporal dementia (FTD) due to mutations in progranulin gene (*GRN*) is a heterogeneous disease with a wide spectrum of phenotypic variability and age at onset, even within the same family [1–3]. The most frequent clinical conditions are behavioral variant frontotemporal dementia (bvFTD) and primary progressive aphasia with some cases presenting with corticobasal syndrome [4], Alzheimer's disease, and Lewy body dementia. Neuroradiologically it is characterized by white matter hyperintensities [5, 6] and asymmetric atrophy, which can even be identified using visual rating scales [7]. It is known from literature that cognitive reserve, calculated with level of education and occupation, modulates functional connectivity in GRN carriers [8] and recently it has been demonstrated that leisure time activities mitigate the burden in FTD [9]. Higher occupation levels were associated with a more severe hypometabolism in prefrontal cortex [10].

*Correspondence to: Giorgio G. Fumagalli, Department of Neurosciences, Psychology, Drug Research and Child Health (NEUROFARBA), University of Florence, Fondazione Ca' Granda, IRCCS Ospedale Maggiore Policlinico, Via F. Sforza 35, 20122, Milan, Italy. E-mail: giorgiofumagalli@hotmail.com.

doi:10.1093/brain/awz039 BRAIN 2019: 142; 1108–1120 | 1108

BRAIN
A JOURNAL OF NEUROLOGY

Cerebral perfusion changes in presymptomatic genetic frontotemporal dementia: a GENFI study

Henri J.M.M. Mutsaerts,[1,*] Saira S. Mirza,[1,*] Jan Petr,[2] David L. Thomas,[3] David M. Cash,[3] Martina Bocchetta,[3] Enrico de Vita,[3] Arron W.S. Metcalfe,[1] Zahra Shirzadi,[1] Andrew D. Robertson,[1] Maria Carmela Tartaglia,[4,5,6] Sara B. Mitchell,[1,6,7] Sandra E. Black,[1,6,7] Morris Freedman,[6,8] David Tang-Wai,[5,6] Ron Keren,[5] Ekaterina Rogaeva,[4] John van Swieten,[9] Robert Laforce Jr,[10] Fabrizio Tagliavini,[11] Barbara Borroni,[12] Daniela Galimberti,[13] James B. Rowe,[14] Caroline Graff,[15] Giovanni B. Frisoni,[16] Elizabeth Finger,[17] Sandro Sorbi,[18] Alexandre de Mendonça,[19] Jonathan D. Rohrer,[3] Bradley J. MacIntosh[1] and Mario Masellis[1,6,7,20] on behalf of the GENetic Frontotemporal dementia Initiative (GENFI)[†]

[*]These authors contributed equally to this work.
[†]Appendix 1.

Genetic forms of frontotemporal dementia are most commonly due to mutations in three genes, *C9orf72*, *GRN* or *MAPT*, with presymptomatic carriers from families representing those at risk. While cerebral blood flow shows differences between frontotemporal dementia and other forms of dementia, there is limited evidence of its utility in presymptomatic stages of frontotemporal dementia. This study aimed to delineate the cerebral blood flow signature of presymptomatic, genetic frontotemporal dementia using a voxel-based approach. In the multicentre GENetic Frontotemporal dementia Initiative (GENFI) study, we investigated cross-sectional differences in arterial spin labelling MRI-based cerebral blood flow between presymptomatic *C9orf72*, *GRN* or *MAPT* mutation carriers ($n = 107$) and non-carriers ($n = 113$), using general linear mixed-effects models and voxel-based analyses. Cerebral blood flow within regions of interest derived from this model was then explored to identify differences between individual gene carrier groups and to estimate a timeframe for the expression of these differences. The voxel-based analysis revealed a significant inverse association between cerebral blood flow and the expected age of symptom onset in carriers, but not non-carriers. Regions included the bilateral insulae/orbitofrontal cortices, anterior cingulate/paracingulate gyri, and inferior parietal cortices, as well as the left middle temporal gyrus. For all bilateral regions, associations were greater on the right side. After correction for partial volume effects in a region of interest analysis, the results were found to be largely driven by the *C9orf72* genetic subgroup. These cerebral blood flow differences first appeared approximately 12.5 years before the expected symptom onset determined on an individual basis. Cerebral blood flow was lower in presymptomatic mutation carriers closer to and beyond their expected age of symptom onset in key frontotemporal dementia signature regions. These results suggest that arterial spin labelling MRI may be a promising non-invasive imaging biomarker for the presymptomatic stages of genetic frontotemporal dementia.

1 Hurvitz Brain Sciences Program, Sunnybrook Research Institute, University of Toronto, Toronto, Canada
2 PET Center, Institute of Radiopharmaceutical Cancer Research, Helmholtz-Zentrum Dresden-Rossendorf, Dresden, Germany
3 Institute of Neurology, University College London, London, UK
4 Tanz Centre for Research in Neurodegenerative Diseases, University of Toronto, Toronto, Canada

Received October 14, 2018. Revised December 14, 2018. Accepted January 4, 2019. Advance Access publication March 7, 2019

5 Memory Clinic, University Health Network, Toronto, Canada
6 Division of Neurology, Department of Medicine, Sunnybrook Health Sciences Centre, University of Toronto, Toronto, Canada
7 L.C. Campbell Cognitive Neurology Research Unit, Sunnybrook Health Sciences Centre, Toronto, Canada
8 Baycrest Centre for Geriatric Care, Toronto, Canada
9 Department of Neurology, Erasmus Medical Center, Rotterdam, The Netherlands
10 Clinique Interdisciplinaire de Mémoire (CIME), Département des Sciences Neurologiques, CHU de Québec, Faculté de médecine, Université Laval, Québec, Canada
11 Fondazione Istituto di Ricovero e Cura a Carattere Scientifico, Milan, Italy
12 Department of Medical and Experimental Sciences, University of Brescia, Brescia, Italy
13 Centro Dino Ferrari, Fondazione Ca' Granda IRCCS Ospedale Policlinico, University of Milan, Milan, Italy
14 Department of Clinical Neurosciences, University of Cambridge, Cambridge, UK
15 Department of Geriatric Medicine, Karolinska Institutet, Stockholm, Sweden
16 IRCCS San Giovanni di Dio Fatebenefratelli, Brescia, Italy
17 Department of Clinical Neurological Sciences, University of Western Ontario, London, Canada
18 Department of Neuroscience, Psychology, Drug Research and Child Health, University of Florence, Florence, Italy
19 Neurology Department, Faculty of Medicine of Lisbon, Portugal
20 Cognitive and Movement Disorders Clinic, Sunnybrook Health Sciences Centre, Toronto, Canada

Correspondence to: Mario Masellis, MSc, MD, PhD
Cognitive and Movement Disorders Clinic, Sunnybrook Health Sciences Centre
2075 Bayview Avenue, Room A4 55
Toronto, Ontario, Canada M4N 3M5
E-mail: mario.masellis@sunnybrook.ca

Keywords: genetic frontotemporal dementia; arterial spin labelling; cerebral blood flow; presymptomatic biomarker

Abbreviations: ASL = arterial spin labelling; GENFI = GENetic Frontotemporal dementia Initiative

Introduction

Frontotemporal dementia encompasses a pathologically heterogeneous group of neurodegenerative diseases, characterized clinically by prominent behavioural and/or language disruption. Frontotemporal dementia significantly impacts patients and their families during the prime of their lives when individuals have responsibilities to their careers, raising children and social interactions (Neary *et al.*, 2005; Onyike and Diehl-Schmid, 2013). It is highly heritable, with an autosomal dominant family history documented in about one-third of people with the disease (Rohrer *et al.*, 2009). Several mutations across three genes (*C9orf72*, *GRN* and *MAPT*) make up the majority of genetic frontotemporal dementia (Seelaar *et al.*, 2011*b*; Warren *et al.*, 2013). The study of presymptomatic mutation carriers compared to non-carriers affords a unique opportunity to understand more about the natural history of genetic frontotemporal dementia during the preclinical phases.

One priority in frontotemporal dementia research is to develop imaging biomarkers (Neary *et al.*, 2005; Rohrer *et al.*, 2013). It is hoped that such biomarkers of genetic frontotemporal dementia could identify those at highest risk of transitioning into the clinical phase, the clinical subtype that will develop, and the age when symptoms will appear. Furthermore, these biomarkers may allow for longitudinal monitoring of disease progression, as well as evaluating efficacy of potential disease-modifying drugs (Cenik *et al.*, 2011; Bateman *et al.*, 2012). Indeed, brain changes have been demonstrated with imaging in presymptomatic/early stage genetic Alzheimer's disease (Bateman *et al.*, 2012). Our structural MRI work on presymptomatic/early stage genetic frontotemporal dementia, as well as that of others, has previously shown early brain changes, particularly in the fronto-insular-temporal regions (Rohrer *et al.*, 2015; Lee *et al.*, 2017; Bertrand *et al.*, 2018). A potential functional imaging biomarker is arterial spin labelling (ASL) perfusion MRI. Increasing evidence suggests that perfusion changes in frontotemporal dementia may be more extensive than structural brain changes early on in the disease course (Olm *et al.*, 2016). ASL allows non-invasive imaging of cerebral blood flow *in vivo* (Alsop *et al.*, 2015). In contrast to other perfusion imaging techniques, ASL uses blood water as an endogenous tracer, which is safe and ideal for longitudinal imaging (Alsop *et al.*, 2015).

Several studies have shown that ASL cerebral blood flow can distinguish symptomatic frontotemporal dementia cases from controls and other dementias (Hu *et al.*, 2010; Shimizu *et al.*, 2010; Binnewijzend *et al.*, 2014; Olm *et al.*, 2016; Steketee *et al.*, 2016). To our knowledge, only one study conducted at a single site has examined the use of ASL perfusion MRI in contrasting presymptomatic *MAPT* and *GRN* mutation carriers ($n = 34$ combined) to non-carriers ($n = 31$) (Dopper *et al.*, 2016). Whilst they did not find any cross-sectional differences in perfusion between carriers and non-carriers at baseline, they demonstrated a significant reduction in perfusion of the frontal pole, superior frontal gyrus, paracingulate gyrus, posterior (mid)cingulate gyrus, precuneus, and thalamus among *GRN* carriers compared to non-carriers on 2

RESEARCH PAPER

Education modulates brain maintenance in presymptomatic frontotemporal dementia

Stefano Gazzina,[1] Mario Grassi,[2] Enrico Premi,[3] Maura Cosseddu,[4] Antonella Alberici,[1] Silvana Archetti,[5] Roberto Gasparotti,[6] John Van Swieten,[7] Daniela Galimberti,[8,9] Raquel Sanchez-Valle,[10] Robert Jr Laforce,[11] Fermin Moreno,[12] Matthis Synofzik,[13,14] Caroline Graff,[15] Mario Masellis,[16] Maria Carmela Tartaglia,[17] James B Rowe,[18] Rik Vandenberghe,[19] Elizabeth Finger,[20] Fabrizio Tagliavini,[21] Alexandre de Mendonça,[22] Isabel Santana,[23] Christopher R Butler,[24] Simon Ducharme,[25,26] Alex Gerhard,[27] Adrian Danek,[28] Johannes Levin,[28] Markus Otto,[29] Giovanni Frisoni,[30,31] Sandro Sorbi,[32,33] Alessandro Padovani,[1] Jonathan D Rohrer,[34] Barbara Borroni,[1] on behalf of the Genetic FTD Initiative, GENFI

For numbered affiliations see end of article.

Correspondence to
Professor Barbara Borroni, Centre for Neurodegenerative Disorders, Neurology Unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy; bborroni@inwind.it

Received 20 January 2019
Revised 30 April 2019
Accepted 1 May 2019
Published Online First 10 June 2019

ABSTRACT

Objective Cognitively engaging lifestyles have been associated with reduced risk of conversion to dementia. Multiple mechanisms have been advocated, including increased brain volumes (ie, brain reserve) and reduced disease progression (ie, brain maintenance). In cross-sectional studies of presymptomatic frontotemporal dementia (FTD), higher education has been related to increased grey matter volume. Here, we examine the effect of education on grey matter loss over time.

Methods Two-hundred twenty-nine subjects at-risk of carrying a pathogenic mutation leading to FTD underwent longitudinal cognitive assessment and T1-weighted MRI at baseline and at 1 year follow-up. The first principal component score of the graph-Laplacian Principal Component Analysis on 112 grey matter region-of-interest volumes was used to summarise the grey matter volume (GMV). The effects of education on cognitive performances and GMV at baseline and on the change between 1 year follow-up and baseline (slope) were tested by Structural Equation Modelling.

Results Highly educated at-risk subjects had better cognition and higher grey matter volume at baseline; moreover, higher educational attainment was associated with slower loss of grey matter over time in mutation carriers.

Conclusions This longitudinal study demonstrates that even in presence of ongoing pathological processes, education may facilitate both brain reserve and brain maintenance in the presymptomatic phase of genetic FTD.

To cite: Gazzina S, Grassi M, Premi E, *et al*. *J Neurol Neurosurg Psychiatry* 2019;**90**:1124–1130.

INTRODUCTION

Frontotemporal dementia (FTD) is a neurodegenerative disorder characterised by executive dysfunction, personality changes and language impairment, along with atrophy of frontal and temporal lobes.[1 2] FTD has a strong genetic background with autosomal dominant inheritance in around a third of patients. Mutations in *Microtubule-Associated Protein Tau* (*MAPT*), *Granulin* (*GRN*) and *chromosome 9 open reading frame 72* (*C9orf72*) genes are proven major causes of genetic FTD, accounting for 10% to 20% of all FTD cases.[3]

There is wide variation in the age at onset within genes and within families with the same mutation, and possible disease modifiers have been recently reported. Identification of disease modifiers is key to correctly select subjects, reduce heterogeneity and increase statistical power of analysis of clinical trials, to stage presymptomatic disease and to enable long-term care planning in at-risk subjects.

Genetic variations within *Transmembrane Protein 106B* (*TMEM106B*) have been suggested to modulate disease onset in frontotemporal lobar degeneration due to transactive response (TAR) DNA binding protein 43 proteinopathy,[4 5] and more recently, *glial cell line-derived neurotrophic factor (GDNF) Family Receptor Alpha 2* (*GFRA2*) polymorphism and *C6orf10/LOC101929163* locus have been further implied in affecting the onset in *GRN* and *C9orf72* mutation carriers, respectively.[6 7]

Along with non-modifiable genetic determinants, modifiable factors that modulate brain structure and function have been identified. For example, educational attainment contributes to resilience against brain damage in neurodegenerative disorders including Alzheimer's disease and FTD,[8 9] in symptomatic and presymptomatic disease stages. In particular, it has been shown that higher educational achievements are associated with greater grey matter volumes in presymptomatic subjects carrying pathogenic FTD mutations.[10] These findings corroborated previous studies in healthy individuals, in which life exposures, such as educational and occupational attainments and engagement in leisure and social activities, have been associated with decreased risk of developing dementia[11 12] and with greater brain volumes.[13 14]

These results argue that education, a proxy measure of brain reserve, may postpone FTD symptom onset; however, these findings cannot give any information on the role of educational attainment in counteracting the effect of the

Our results seem to confirm the latter hypothesis, showing that higher education confers higher grey matter volumes and greater brain maintenance over time. Additionally, as previously reported,[10 30] *TMEM106B* genotype significantly modulates grey matter volume at baseline in mutation carriers.

These findings are in line with previous longitudinal studies demonstrating that reserve proxies are associated with reduced rate of hippocampal atrophy,[31 32] reduced rate of brain hypometabolism[33] and cerebrospinal fluid biomarkers changes[33] in healthy agers and Alzheimer's disease.

One intriguing aspect of brain maintenance is that it may reflect differences in the accumulation of pathology-related changes.[34 35] Such demonstration in FTD requires in vivo pathological markers (ie, tau or TDP-43 tracers), which are not currently available.[36] This neuroprotective effect may be related to changes at the molecular level, such as increased levels of neurotrophic factors[37] and glutamate neurotransmission,[38] or at the cellular level, with increased neurogenesis,[39] synaptogenesis[40] and angiogenesis,[41] and might be able to go beyond the underlying pathogenic mechanisms related to the specific mutation (*GRN, C9ORF72, MAPT*) or to specific proteinopathy (ie, TDP-43 or tau).

Interestingly, as previously reported,[10] years of education had a significant effect on grey matter volume even in mutation non-carriers, supporting the idea of a generalisable beneficial effect of education. Conversely, in the present work, we did not find any effect of education on brain maintenance in mutation non-carriers, but we recognise that this could be likely due to the low variance of grey matter volume within 1 year follow-up in healthy subjects. However, longer follow-up is necessary to draw definitive conclusions.

Regarding cognition, higher education led to better cognitive performances at baseline, but not to significant effects on cognitive decline. This effect was comparable in mutation carriers and mutation non-carriers; of note, in subjects without pathogenic mutations, the beneficial effect of education on cognitive performances was greater than in mutation carriers.

We acknowledge that this study entails some limitations. Despite that education represents an environmental factor, it is often immutable because acquired in childhood/young adulthood. Thus, the present results do not allow to directly conclude that interventional trials could delay disease onset. However, education is known to influence professional attainment, which has been already proven a proxy measure of reserve in FTD.[9 42] Also, we chose MMSE as a global measure of cognition, acknowledging that MMSE is affected only close to disease onset[17] and that it does not represent the best measure of severity even in symptomatic phases.[43] Thus, the effect of more sensitive neuropsychological tests[17] has to be evaluated in future studies, especially to assess changes of cognitive performances over time. Moreover, we could not test the effect of educational attainment in each mutation due to low sample number: larger samples are needed to address this issue. Last, due to the observational nature of the study, data on possible confounders, such as concomitant vascular risk factors, were not available. However, in a recent large-scale Mendelian randomisation study of the related condition, that is amyotrophic lateral sclerosis, the authors confirmed educational attainment to be an important modulator based on genetics.[44]

In conclusion, these findings extend our knowledge of the reserve theory, demonstrating that in presymptomatic FTD the rate of atrophy was influenced by the educational level, with reduced grey matter loss in more educated subjects. Thus, even in presence of an ongoing pathological process, presymptomatic FTD subjects still maintain a high-performing reserve like in healthy brains, virtually turning back the clock of the disease natural history. The demonstration that differences in early lifestyle may slow down later disease progression suggests that even in monogenic disorders, outcomes are not wholly determined from birth, and this opens exciting perspectives for eventually delaying symptom onset. Future confirmatory studies assessing the role of other reserve proxies and their effect on longitudinal brain changes in symptomatic monogenic and sporadic FTD are needed.

Author affiliations
[1]Centre for Neurodegenerative Disorders, Neurology Unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy
[2]Department of Brain and Behavioral Science, Medical and Genomic Statistics Unit, University of Pavia, Pavia, Italy
[3]Stroke Unit, Neurology Unit, Spedali Civili Hospital, Brescia, Italy
[4]Neurology Unit, Spedali Civili Hospital, Brescia, Italy
[5]Biotechnology Laboratory, Department of Diagnostics, Spedali Civili Hospital, Brescia, Italy
[6]Neuroradiology Unit, University of Brescia, Brescia, Italy
[7]Department of Neurology, Erasmus Medical Center, Rotterdam, Netherlands
[8]Centro Dino Ferrari, University of Milan, Milan, Italy
[9]Neurodegenerative Diseases Unit, Fondazione IRCSS Ca' Granda, Ospedale Maggiore Policlinico, Milan, Italy
[10]Neurology Department, Hospital Clinic, Institut d'Investigacions Biomèdiques, Barcelona, Spain
[11]Clinique Interdisciplinaire de Mémoire, Département des Sciences Neurologiques, CHU de Québec, and Faculté de Médecine, Université Laval, Québec, Quebec, Canada
[12]Department of Neurology, Hospital Universitario Donostia, San Sebastian, Gipuzkoa, Spain
[13]Department of Neurodegenerative Diseases, Hertie-Institute for Clinical Brain Research & Center of Neurology, University of Tübingen, Tübingen, Germany
[14]German Center for Neurodegenerative Diseases (DZNE), Tübingen, Germany
[15]Karolinska Institutet, Department NVS, Center for Alzheimer Research, Division of Neurogenetics, Stockholm, Sweden
[16]LC Campbell Cognitive Neurology Research Unit, Sunnybrook Research Institute, Toronto, Ontario, Canada
[17]Toronto Western Hospital, Tanz Centre for Research in Neurodegenerative Disease, Toronto, Ontario, Canada
[18]Department of Clinical Neurosciences, University of Cambridge, Cambridge, United Kingdom
[19]Department of Neurosciences, Laboratory for Cognitive Neurology, KU Leuven, Leuven, Belgium
[20]Department of Clinical Neurological Sciences, University of Western Ontario, London, Ontario, Canada
[21]Fondazione Istituto di Ricovero e Cura a Carattere Scientifico Istituto Neurologico Carlo Besta, Milan, Italy
[22]Faculty of Medicine, University of Lisbon, Lisbon, Portugal
[23]Neurology Department, Centro Hospitalar e Universitário de Coimbra, Coimbra, Portugal
[24]Department of Clinical Neurology, University of Oxford, Oxford, United Kingdom
[25]Department of Psychiatry, McGill University Health Centre, McGill University, Montreal, Quebec, Canada
[26]McConnell Brain Imaging Centre, Montreal Neurological Institute, McGill University, Montreal, Quebec, Canada
[27]Institute of Brain, Behaviour and Mental Health, The University of Manchester, Withington, Manchester, United Kingdom
[28]Neurologische Klinik und Poliklinik, Ludwig-Maximilians-Universität, Munich, German Center for Neurodegenerative Diseases (DZNE), Munich, Germany
[29]Department of Neurology, University Hospital Ulm, Ulm, Germany
[30]Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS) Istituto Centro San Giovanni di Dio Fatebenefratelli, Brescia, Italy
[31]Memory Clinic and LANVIE-Laboratory of Neuroimaging of Aging, University Hospitals and University of Geneva, Geneva, Switzerland
[32]Department of Neuroscience, Psychology, Drug Research and Child Health, University of Florence, Florence, Italy
[33]Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS) "Don Gnocchi", Florence, Italy
[34]Dementia Research Centre, UCL Institute of Neurology, London, United Kingdom

Acknowledgements James B Rowe is supported by the Wellcome Trust (103838) and the Cambridge NIHR BiomedicalResearch Centre.

ABCA1- and ABCG1-mediated cholesterol efflux capacity of cerebrospinal fluid is impaired in Alzheimer's disease

Cinzia Marchi,*,[1] Maria Pia Adorni,*,[1] Paolo Caffarra,[†,§] Nicoletta Ronda,*,[3] Marco Spallazzi,[†] Federica Barocco,[†,2] Daniela Galimberti,,[††] Franco Bernini,*,[3] and Francesca Zimetti***

Department of Food and Drug* and Department of Medicine and Surgery, Section of Neurology,[†] University of Parma, Parma, Italy; Alzheimer Center,[§] Briolini Hospital, Gazzaniga, Bergamo, Italy; Department of Biomedical, Surgical and Dental Sciences,** Dino Ferrari Center, University of Milano, Milano, Italy; and Neurodegenerative Diseases Unit,[††] Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Milano, Italy

ORCID IDs: 0000-0002-9386-9260 (C.M.); 0000-0002-4810-6862 (M.A.); 0000-0003-2246-5223 (P.C.); 0000-0001-8891-1340 (N.R.); 0000-0002-8091-2063 (M.S.); 0000-0002-5010-6167 (D.G.); 0000-0002-2576-7983 (F.B.); 0000-0002-6665-263X (F.Z.)

Abstract HDL-like particles in human cerebrospinal fluid (CSF) promote the efflux of cholesterol from astrocytes toward the neurons that rely on this supply for their functions. We evaluated whether cell cholesterol efflux capacity of CSF (CSF-CEC) is impaired in Alzheimer's disease (AD) by analyzing AD ($n = 37$) patients, non-AD dementia (non-AD DEM; $n = 16$) patients, and control subjects ($n = 39$). As expected, AD patients showed reduced CSF Aβ 1-42, increased total and phosphorylated tau, and a higher frequency of the apoε4 genotype. ABCA1- and ABCG1-mediated CSF-CEC was markedly reduced in AD (−73% and −33%, respectively) but not in non-AD DEM patients, in which a reduced passive diffusion CEC (−40%) was observed. Non-AD DEM patients displayed lower CSF apoE concentrations (−24%) compared with controls, while apoA-I levels were similar among groups. No differences in CSF-CEC were found by stratifying subjects for apoε4 status. ABCG1 CSF-CEC positively correlated with Aβ 1-42 ($r = 0.305$, $P = 0.025$), while ABCA1 CSF-CEC inversely correlated with total and phosphorylated tau ($r = -0.348$, $P = 0.018$ and $r = -0.294$, $P = 0.048$, respectively). The CSF-CEC impairment and the correlation with the neurobiochemical markers suggest a pathophysiological link between CSF HDL-like particle dysfunction and neurodegeneration in AD.— Marchi, C., M. P. Adorni, P. Caffarra, N. Ronda, M. Spallazzi, F. Barocco, D. Galimberti, F. Bernini, and F. Zimetti. **ABCA1- and ABCG1-mediated cholesterol efflux capacity of cerebrospinal fluid is impaired in Alzheimer's disease. *J. Lipid Res.* 2019.** 60: **1449–1456.**

Supplementary key words ATP-binding cassette A1 • ATP-binding cassette G1 • apolipoproteins • apolipoprotein A-I • apolipoprotein E • apolipoprotein E4

Dysregulation of cholesterol homeostasis in the CNS has been associated with various neurodegenerative disorders, including Parkinson's, Huntington's, and Alzheimer's disease (AD) (1). Evidence supporting this relationship derives, for example, from recent genomic-wide association studies that have identified several loci involved in lipid metabolism among the AD-susceptible genes (2, 3). For example, the ε4 allele of the *APOE* gene encoding apoE is undoubtedly the most strong genetic risk factor, but recently other genes have been identified such as *BIN1*, *CLU*, *PICALM*, *ABCA7*, *ABCA1*, *ABCG1*, and *SORL1* (4).

However, the exact mechanisms linking cholesterol homeostasis derangement and AD pathogenesis are far from being understood and conflicting data have been released, describing both increased, decreased, or no change of cholesterol levels in different brain sections and the cerebrospinal fluid (CSF) of AD patients compared with control subjects (5).

Approximately 30% of the total body cholesterol is present in the brain, where it plays a crucial role in the synaptogenesis and maintenance of neuronal plasticity and function (6). The brain relies on endogenous local cholesterol synthesis because it is isolated from other body compartments by the blood-brain barrier (7, 8). While cholesterol synthesis in neurons and glial cells is very high during embryogenesis, adult neurons progressively lose this capacity and most exclusively rely on cholesterol produced from other

This work was partially supported by funding for basic activities related to research from the Italian National Agency for the Evaluation of Universities and Research Institutes (F.Z.).

Manuscript received 15 November 2018 and in revised form 25 May 2019.

Published, JLR Papers in Press, June 5, 2019
DOI https://doi.org/10.1194/jlr.P091033

Abbreviations: AD, Alzheimer's disease; Aβ, amyloid β; CEC, cholesterol efflux capacity; CHO, Chinese hamster ovary; CSF, cerebrospinal fluid; DEM, dementia.

[1] C. Marchi and M. P. Adorni contributed equally to this article.
[2] Present address of F. Barocco: Alzheimer Center, Briolini Hospital, Gazzaniga, Bergamo, Italy.
[3] To whom correspondence should be addressed.
e-mail: fbernini@unipr.it (F.B.); nicoletta.ronda@unipr.it (N.R.)
The online version of this article (available at http://www.jlr.org) contains a supplement.

Journal of Alzheimer's Disease 69 (2019) 663–669
DOI 10.3233/JAD-190119
IOS Press

Short Communication

Cerebrospinal Fluid Level of Aquaporin4: A New Window on Glymphatic System Involvement in Neurodegenerative Disease?

Andrea Arighi[a,b,c,*], Andrea Di Cristofori[d], Chiara Fenoglio[a,b,c], Stefano Borsa[e], Marianna D'Anca[a,b,c], Giorgio Giulio Fumagalli[a,b,c,f], Marco Locatelli[b,e], Giorgio Carrabba[e], Anna Margherita Pietroboni[a,b,c], Laura Ghezzi[a,b,c], Tiziana Carandini[a,b,c], Annalisa Colombi[a,b,c], Marta Scarioni[a,b,c], Milena Alessandra De Riz[a,b,c], Maria Serpente[a,b,c], Paolo Maria Rampini[e], Elio Scarpini[a,b,c] and Daniela Galimberti[a,c,g]

[a]*Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, UOSD Neurologia – Malattie Neurodegenerative, Milan, Italy*
[b]*Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy*
[c]*Centro Dino Ferrari, Milan, Italy*
[d]*Azienda Socio Sanitaria Territoriale – Monza, Ospedale San Gerardo, Monza, Italy – U.O. Neurochirurgia*
[e]*Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, UOC Neurochirurgia, Milan, Italy*
[f]*Department of Neurosciences, Psychology, Drug Research and Child Health (NEUROFARBA), University of Florence, Florence, Italy*
[g]*Department of Biomedical, Surgical and Dental Sciences, University of Milan, Milan, Italy*

Handling Associate Editor: Marco Bozzali

Accepted 2 April 2019

Abstract. Aquaporin4 (AQP4) is a water channel protein located at astrocyte foot processes that plays a role in glymphatic system, a highly organized fluid transport pathway which seems to be involved in Alzheimer's disease (AD) and normal pressure hydrocephalus (NPH) pathophysiology. Cerebrospinal fluid (CSF) AQP4 levels were determined in 11 patients with AD, 10 patients with NPH, and 9 controls. We found significantly reduced AQP4 in AD patients, a trend in reduction in NPH patients, and a correlation between AQP4 and amyloid-β CSF levels. This study indicates the potential role of AQP4 and glymphatic system in neurodegenerative diseases pathophysiology.

Keywords: Alzheimer's disease, Aquaporin 4, glymphatic system, normal pressure hydrocephalus

INTRODUCTION

Lymphatic vasculature represents a second circulation that accounts for the clearance of interstitial fluid (ISF) with its constituent proteins and other solutes not absorbed across postcapillary venules [1, 2]. Human cerebral parenchyma is devoid of lymphatic vessels and transport of ISF was traditionally attributed only to diffusion, but several historical observations have revealed a lymphatic-like system in the rodent brain [3–5].

Photon microscopic studies of cerebrospinal fluid (CSF) flow, carried out starting from 2012 in live murine brain, have indicated the existence of the

*Correspondence to: Andrea Arighi, MD, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza, Milan, Italy. Tel.: +39 02 55038710; E-mail: andrea.arighi@policlinico.mi.it.

Journal of Alzheimer's Disease 69 (2019) 839–847
DOI 10.3233/JAD-181298
IOS Press

Overlap Between Frontotemporal Dementia and Dementia with Lewy Bodies: A Treviso Dementia (TREDEM) Registry Case Report

Maurizio Gallucci[a,*], Carola Dell'Acqua[a], Franco Boccaletto[b], Chiara Fenoglio[c], Daniela Galimberti[c,d] and Maria Elena Di Battista[a]
[a]*Cognitive Impairment Center, Local Health Authority n.2 Marca Trevigiana, Treviso, Italy*
[b]*Nuclear Medicine Unit, Local Health Authority n.2 Marca Trevigiana, Treviso, Italy*
[c]*University of Milan, Dino Ferrari Center, Milan, Italy*
[d]*Fondazione IRCCS Ca' Granda, Ospedale Policlinico, Neurodegenerative Disease Unit, Milan, Italy*

Handling Associate Editor: Beatrice Arosio

Accepted 2 April 2019

Abstract. In the present work, we report the case of a patient presenting signs of Lewy body dementia (DLB) and frontotemporal dementia (FTD) throughout different phases of the disease. In January 2017, a 79-year-old right-handed living man was admitted to our Memory Clinic for the presence of behavioral disturbances and progressive cognitive decline. For the previous six years, he was monitored by other Neurological Clinics for the onset of extrapyramidal features. Indeed, through the first phase of the disease (2011–2014), the patient predominantly showed: extrapyramidal features, initial cognitive decline, sleep disturbances, and visual hallucinations, together with a reduced dopamine transporter uptake in basal ganglia at the DATscan, suggesting a diagnosis of DLB. In a second phase (2015–2017), while his extrapyramidal features remained substantially stable, his cognitive profile deteriorated, with an additional development of severe behavioral and neuropsychiatric disturbances. Again, a subsequent DATscan study was positive and slightly worse than the preceding one; however, the ^{18}F-FDG PET showed reduced metabolic activity in the frontal and temporal lobes, with the occipital regions left spared. Genetic analysis revealed a hexanucleotide expansion in *C9ORF72* (6//38 repeats; ITALSGEN NV <30). In conclusion, we report the case of a patient presenting, firstly, with probable DLB and, in a second phase, with predominant bvFTD features with stable parkinsonism. Even though some clinical and neuropsychological aspects can co-exist in different neurodegenerative diseases, we find such a significant intersection of clinical features to be fairly atypical. Moreover, what is challenging to define is whether the two clinical phenotypes are somehow lying on a continuum, or if they are two individual entities.

Keywords: *C9ORF72*, dementia with Lewy bodies, diagnosis, frontotemporal dementia, neuroimaging, overlap, Progranulin, TREDEM

INTRODUCTION

Frontotemporal dementia (FTD) is a clinical term referring to a cluster of disorders that affect the frontal and temporal lobe causing a collection of diverse symptoms including: personality change (apathy, disinhibition, loss of insight and emotional control), language impairments, loss of semantics, executive dysfunction, and overall cognitive deterioration [1]. FTD consists of a spectrum of clinical syndromes: behavioral variant FTD (bvFTD) [2] and primary progressive aphasia (PPA) variants [3]. These variants

*Correspondence to: Maurizio Gallucci, MD, Cognitive Impairment Center, Local Health Authority n.2 Marca Trevigiana, Treviso, Italy. E-mail: maurizio.gallucci@aulss2.veneto.it.

OPEN ACCESS

RESEARCH PAPER

Clinical value of cerebrospinal fluid neurofilament light chain in semantic dementia

Lieke H H Meeter,[1] Rebecca M E Steketee,[2] Dina Salkovic,[1] Maartje E Vos,[1] Murray Grossman,[3] Corey T McMillan,[3] David J Irwin,[3] Adam L Boxer,[4] Julio C Rojas,[4] Nicholas T Olney,[5] Anna Karydas,[5] Bruce L Miller,[4] Yolande A L Pijnenburg,[6] Frederik Barkhof,[7,8] Raquel Sánchez-Valle,[9,10] Albert Lladó,[9,10] Sergi Borrego-Ecija,[9,10] Janine Diehl-Schmid,[11] Timo Grimmer,[11] Oliver Goldhardt,[11] Alexander F Santillo,[12] Oskar Hansson,[12] Susanne Vestberg,[13] Barbara Borroni,[14] Alessandro Padovani,[14] Daniela Galimberti,[15,16] Elio Scarpini,[15,17] Jonathan D Rohrer,[18] Ione O C Woollacott,[18] Matthis Synofzik,[19,20] Carlo Wilke,[19,20] Alexandre de Mendonca,[21] Rik Vandenberghe,[22,23] Luisa Benussi,[24] Roberta Ghidoni,[24] Giuliano Binetti,[24,25] Wiro J Niessen,[26,27] Janne M Papma,[1] Harro Seelaar,[1] Lize C Jiskoot,[1] Frank Jan de Jong,[1] Laura Donker Kaat,[1,28] Marta Del Campo,[29] Charlotte E Teunissen,[29] Esther E Bron,[26] Esther Van den Berg,[1] John C Van Swieten[1]

► Additional material is published online only. To view please visit the journal online (http://dx.doi.org/10.1136/jnnp-2018-319784).

For numbered affiliations see end of article.

Correspondence to
Dr John C Van Swieten, Alzheimer Center and Department of Neurology, Erasmus MC, Rotterdam 3000 CA, The Netherlands; j.c.vanswieten@erasmusmc.nl

Portions of this study have been presented in abstract form at the 4th Congress of the European Academy of Neurology on 17 June 2018 and at the 11th International Conference on Frontotemporal Dementias on 12 November 2018.

Received 8 October 2018
Revised 12 February 2019
Accepted 10 April 2019
Published Online First 23 May 2019

Check for updates

To cite: Meeter LHH, Steketee RME, Salkovic D, *et al*. *J Neurol Neurosurg Psychiatry* 2019;**90**:997–1004.

ABSTRACT

Background Semantic dementia (SD) is a neurodegenerative disorder characterised by progressive language problems falling within the clinicopathological spectrum of frontotemporal lobar degeneration (FTLD). The development of disease-modifying agents may be facilitated by the relative clinical and pathological homogeneity of SD, but we need robust monitoring biomarkers to measure their efficacy. In different FTLD subtypes, neurofilament light chain (NfL) is a promising marker, therefore we investigated the utility of cerebrospinal fluid (CSF) NfL in SD.

Methods This large retrospective multicentre study compared cross-sectional CSF NfL levels of 162 patients with SD with 65 controls. CSF NfL levels of patients were correlated with clinical parameters (including survival), neuropsychological test scores and regional grey matter atrophy (including longitudinal data in a subset).

Results CSF NfL levels were significantly higher in patients with SD (median: 2326 pg/mL, IQR: 1628–3593) than in controls (577 (446–766), p<0.001). Higher CSF NfL levels were moderately associated with naming impairment as measured by the Boston Naming Test (r_s=−0.32, p=0.002) and with smaller grey matter volume of the parahippocampal gyri (r_s=−0.31, p=0.004). However, cross-sectional CSF NfL levels were not associated with progression of grey matter atrophy and did not predict survival.

Conclusion CSF NfL is a promising biomarker in the diagnostic process of SD, although it has limited cross-sectional monitoring or prognostic abilities.

INTRODUCTION

Semantic dementia (SD) is a sporadic neurodegenerative disorder characterised by loss of semantic knowledge, impaired naming and word comprehension, with preserved speech production.[1] Compared with other disorders in the frontotemporal lobar degeneration (FTLD) spectrum, SD is relatively homogeneous because of the typical clinical presentation, the neuroimaging signature of asymmetrical anteroinferior temporal atrophy and the typical pathology of type C FTLD with TAR DNA binding protein 43 kDa inclusions (FTLD-TDP).[1–3] This homogeneity provides opportunities for the development of disease-modifying agents, for which reliable biomarkers are essential to measure their efficacy.

A promising biomarker in frontotemporal dementia (FTD) is neurofilament light chain (NfL), a major component of the neuronal cytoskeleton involved in axonal and dendritic growth, signalling and transport.[4] Previous studies have demonstrated elevated cerebrospinal fluid (CSF) NfL levels across the FTLD spectrum which are associated with disease severity, brain atrophy and survival.[5–11] Moreover, CSF and serum NfL levels are strongly correlated, enabling repeated measurements in serum to assess disease progression or treatment response.[5 12] Small series have shown high CSF and serum NfL concentrations exclusively in the group of patients with SD,[7 13–15] but a larger cohort may be needed to detect associations with clinical variables. Another interesting question in this context is whether high NfL levels are also associated with survival in SD, considering that SD is a relatively slow progressive disease.[16 17]

In a large series of patients with SD from 14 different centres, we investigated our hypothesis that CSF NfL levels are elevated compared with controls and correlate with disease severity, atrophy and clinical progression in SD.

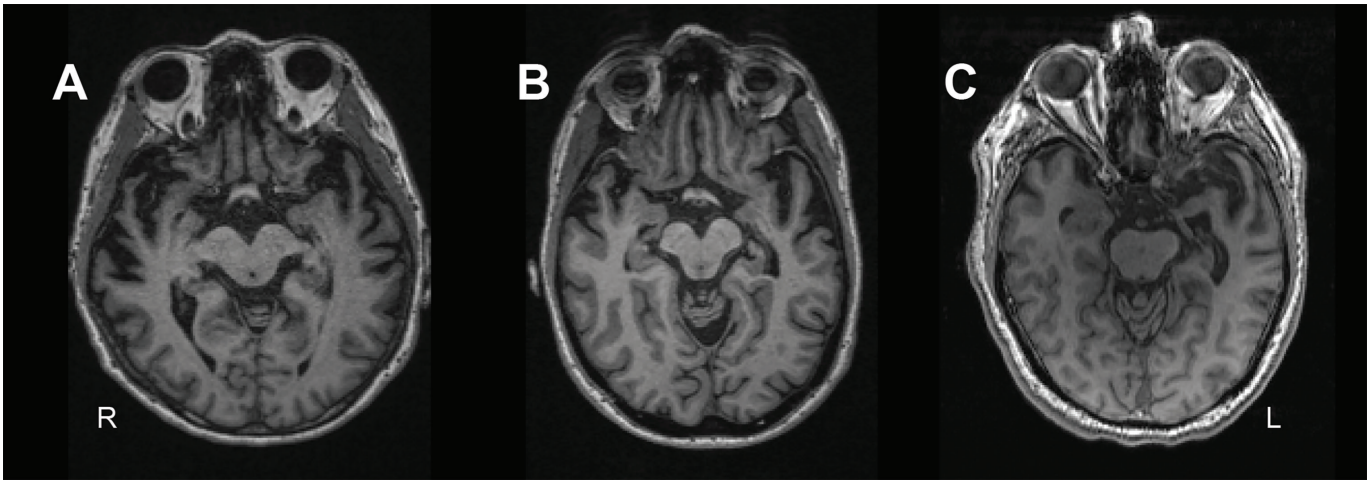


Figure 4 Temporal pole atrophy in patients. Transversal T1-weighted MR images of patients representative for the distribution of dominant anterior temporal pole atrophy within the sample. From left to right: patient with lower quartile (A), median (B) and upper quartile (C) anterior temporal pole grey matter volumes of the dominant side.

excluding data from scanners on which less than three datasets were acquired and correcting for scanner in the analysis. Additionally, we did not study different primary progressive aphasia subtypes and did not include serum—which is likely to replace CSF measurements in the near future. The lack of longitudinal NfL data did not allow us to draw conclusions about NfL and clinical or imaging markers over time; it is conceivable that a plateau phase or even a decrease of NfL may occur over time.

In conclusion, our results show that cross-sectional elevated CSF NfL may be a useful biomarker for the neurodegenerative process in SD, which could lead to the use of CSF NfL as a diagnostic biomarker. However, the use of CSF NfL to monitor disease progression in SD remains debatable, since we only found a moderate association with language deterioration and atrophy and no relation of CSF NfL with survival (in a relatively short follow-up time). This is in contrast with previous reports on NfL in other neurodegenerative diseases[5 6 10 11] and should thus be taken into account when interpreting studies that combined different FTLD subtypes. Recently, it has become clear that NfL in blood and CSF strongly correlate,[5 12] facilitating longitudinal monitoring. More longitudinal multicentre studies are needed to assess how serial NfL levels fluctuate over time in relation to longitudinal clinical and imaging changes in SD (and other FTLD subpopulations) and thereby their potential utility.

Author affiliations
[1]Alzheimer Center and Department of Neurology, Erasmus MC, Rotterdam, The Netherlands
[2]Department of Radiology and Nuclear Medicine, Erasmus MC, Rotterdam, Zuid-Holland, The Netherlands
[3]Penn FTD Center, Department of Neurology, University of Pennsylvania Perelman School of Medicine, Philadelphia, Pennsylvania, USA
[4]Neurology, Memory and Aging Center University of California San Francisco, San Francisco, California, USA
[5]Neurology, University of California San Francisco Memory and Aging Center, San Francisco, California, USA
[6]Alzheimer Center and Department of Neurology, Amsterdam Neuroscience, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, The Netherlands
[7]Department of Radiology and Nuclear Medicine, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, The Netherlands
[8]Neurology and Healthcare Engineering, University College London Medical School, London, UK
[9]Department of Neurology, Hospital Clinic de Barcelona, Barcelona, Catalunya, Spain
[10]Institut d'Investigacions Biomèdiques August Pi i Sunyer, Barcelona, Spain
[11]Department of Psychiatry and Psychotherapy, Klinikum rechts der Isar, Technical University of Munich, School of Medicine, Munich, Germany
[12]Clinical Memory Research Unit, Department of Clinical Sciences, Lund University, Lund, Sweden
[13]Psychology, Lund University, Lund, Sweden
[14]Centre for Ageing Brain and Neurodegenerative Disorders, Neurology Unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy
[15]Neurodegenerative Diseases Unit, Fondazione IRCCS Ca' Granda, Ospedale Policlinico, Milan, Italy
[16]Biomedical, Surgical and Dental Sciences, University of Milan, Centro Dino Ferrari, Milan, Italy
[17]Pathophysiology and Transplantation, University of Milan, Centro Dino Ferrari, Milan, Italy
[18]Dementia Research Centre, Department of Neurodegenerative Diseases, UCL Institute of Neurology, London, UK
[19]Department of Neurodegenerative Diseases, Hertie Institute for Clinical Brain Research, Tübingen, Germany
[20]German Center for Neurodegenerative Diseases (DZNE), Tübingen, Germany
[21]Institute of Molecular Medicine and Faculty of Medicine, University of Lisbon, Lisbon, Portugal
[22]Department of Neurology, University Hospital Leuven, Leuven, Belgium
[23]Laboratory for Cognitive Neurology, Department of Neurosciences, KU Leuven, Leuven, Vlaanderen, Belgium
[24]Molecular Markers Laboratory, IRCCS Istituto Centro San Giovanni di Dio Fatebenefratelli, Brescia, Italy
[25]MAC Memory Clinic, IRCCS Istituto Centro San Giovanni di Dio Fatebenefratelli, Brescia, Italy
[26]Biomedical Imaging Group Rotterdam, Departments of Medical Informatics and Radiology & Nuclear Medicine, Erasmus MC, Rotterdam, Zuid-Holland, The Netherlands
[27]Imaging Physics, Applied Sciences, Delft University of Technology, Delft, The Netherlands
[28]Department of Clinical Genetics, Leids Universitair Medisch Centrum, Leiden, Zuid-Holland, The Netherlands
[29]Neurochemistry Laboratory, Department of Clinical Chemistry, Amsterdam Neuroscience, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, The Netherlands

Correction notice This article has been corrected since it first published online. The open access licence type has been amended.

Acknowledgements JDR would like to thank Lucy L. Russell and Martina Bocchetta for their help in collecting clinical, imaging and CSF data. AdM would like to thank Carolina Maruta for her invaluable neuropsychological aid and Catarina Beatriz Ferreira for her assistance in the preparation of the samples.

Contributors LHHM and RMES: study concept and design, acquisition of data, analysis and interpretation of data, writing and revising the manuscript. DS: analysis and interpretation of data, revising the manuscript. MV: acquisition of data, analysis and interpretation of data, writing and revising the manuscript. EEB: study concept and design, acquisition of data, critical revision of manuscript for intellectual content and interpretation of data. EvdB: study concept and design, acquisition of data, critical revision of manuscript for intellectual content and interpretation of data. JCvS: study concept and design, acquisition of data, analysis and interpretation of

Journal of Psychiatric Research 114 (2019) 17–23

Contents lists available at ScienceDirect

Journal of Psychiatric Research

journal homepage: www.elsevier.com/locate/jpsychires

Exploring the role of BDNF DNA methylation and hydroxymethylation in patients with obsessive compulsive disorder

Claudio D'Addario[a,b,*,1], Fabio Bellia[a,1], Beatrice Benatti[c], Benedetta Grancini[c], Matteo Vismara[c], Mariangela Pucci[a], Vera De Carlo[c], Caterina Viganò[c], Daniela Galimberti[f,g], Chiara Fenoglio[f,g], Elio Scarpini[f,g], Mauro Maccarrone[h,i], Bernardo Dell'Osso[c,d,e,**]

[a] University of Teramo, Bioscience, Teramo, Italy
[b] Karolinska Institutet, Department of Clinical Neuroscience, Stockholm, Sweden
[c] University of Milan, Department of Biomedical and Clinical Sciences Luigi Sacco, Milano, Italy
[d] CRC "Aldo Ravelli", University of Milan, Milano, Italy
[e] Department of Psychiatry and Behavioral Sciences, Stanford University, CA, USA
[f] University of Milan, Dino Ferrari Center, Milan, Italy
[g] Fondazione IRCCS Ca' Granda, Ospedale Policlinico, Neurodegenerative Diseases Unit, Milan, Italy
[h] Department of Medicine, Campus Bio-Medico University of Rome, Rome, Italy
[i] Fondazione IRCCS Santa Lucia, Unit of Lipid Neurochemistry, Rome, Italy

ARTICLE INFO

Keywords:
BDNF
Obsessive compulsive disorder
DNA methylation
DNA hydroxymethylation

ABSTRACT

Obsessive-compulsive disorder (OCD) is a clinically heterogeneous neuropsychiatric condition associated with profound disability, whose susceptibility, stemming from genetic and environmental factors that intersect with each other, is still under investigation. In this perspective, we sought to explore the transcriptional regulation of Brain Derived Neurotrophic Factor (BDNF), a promising candidate biomarker in both development and etiology of different neuropsychiatric conditions, in peripheral blood mononuclear cells from OCD patients and healthy controls. In particular, we focused on *BDNF* gene expression and interrogated in depth DNA methylation and hydroxymethylation at gene promoters (exons I, IV and IX) in a sample of OCD patients attending a tertiary OCD Clinic to receive guidelines-recommended treatment, and matched controls. Our preliminary data showed a significant increase in *BDNF* gene expression and a significant correlation with changes in the two epigenetic modifications selectively at promoter exon I, with no changes in the other promoters under study. We can conclude that transcriptional regulation of *BDNF* in OCD engages epigenetic mechanisms, and can suggest that this is likely evoked by the long-term pharmacotherapy. It is important to underline that many different factors need to be taken into account (i.e. age, sex, duration of illness, treatment), and thus further studies are mandatory to investigate their role in the epigenetic regulation of *BDNF* gene. Of note, we provide unprecedented evidence for the importance of analyzing 5-hydroxymethylcytosine levels to correctly evaluate 5-methylcytosine changes.

1. Introduction

Obsessive-Compulsive disorder (OCD) is a condition with frequent early onset and chronic course (Dell'Osso et al., 2013) characterized by recurrent, unwanted, time-consuming obsessive and compulsive behaviors that cause distress and/or impairment (Milad and Rauch, 2012). The World Health Organization classifies OCD as one of the 10 most disabling conditions for decreased quality of life and loss of income, with a lifetime prevalence of 2–3% of the general population (Milad and Rauch, 2012).

Abbreviations: **OCD**, Obsessive-Compulsive Disorder; **BDNF**, Brain Derived Neurotrophic Factor; **SNP**, Single Nucleotide Polymorphism; **5 mC**, 5-methylcytosine; **5hmC**, 5-hydroxymethylcytosine; **TET**, Ten-Eleven-Translocation proteins; **BD**, Bipolar Disorder; **MDD**, Major Depressive Disorder; **PBMCs**, Peripheral Blood Mononuclear Cells; **GAPDH**, Glyceraldehyde 3-phosphate dehydrogenase; **ß-ACT**, beta actin; **BS**, Bisulfite conversion; **oxBS**, oxidative Bisulfite conversion

* Corresponding author. University of Teramo, Bioscience, Teramo, Italy
** Corresponding author. Department of Biomedical and Clinical Sciences Luigi Sacco, Milano, Italy.
E-mail addresses: cdaddario@unite.it (C. D'Addario), bernardo.dellosso@unimi.it (B. Dell'Osso).
[1] equal contribution.

https://doi.org/10.1016/j.jpsychires.2019.04.006
Received 3 December 2018; Received in revised form 20 March 2019; Accepted 5 April 2019

Massaccesi et al. Immunity & Ageing (2019) 16:7
https://doi.org/10.1186/s12979-019-0147-x

Immunity & Ageing

SHORT REPORT Open Access

Lag-time in Alzheimer's disease patients: a potential plasmatic oxidative stress marker associated with ApoE4 isoform

Luca Massaccesi[1], Emanuela Galliera[1,2*], Daniela Galimberti[3,4], Chiara Fenoglio[3,4], Marina Arcaro[4], Giancarlo Goi[5], Alessandra Barassi[6] and Massimiliano Marco Corsi Romanelli[1,7]

Abstract

In the brain, Oxidative Stress (OS) contribute to structural and functional changes associated with vascular aging, such as endothelial dysfunction, extracellular matrix degradation, resulting in age-related reduced vasodilatation in response to agonists. For this reason, OS is considered a key factor in Alzheimer's Disease (AD) development and recent evidence correlated oxidative stress with vascular lesion in the pathogenesis of AD, but the mechanism still need to be fully clarified.

The etiology of AD is still not completely understood and is influenced by several factors including Apolipoprotein E (ApoE) genotype. In particular, the Apo ε4 isoform is considered a risk factor for AD development.

This study was aimed to evaluate the possible relationship between three plasmatic OS marker and Apo ε4 carrier status. Plasmatic soluble receptor for advanced glycation end products (sRAGE) levels, plasma antioxidant total defenses (by lag-time method) and plasmatic Reactive Oxygen species (ROS) levels were evaluated in 25 AD patients and in 30 matched controls. ROS were significantly higher while plasma antioxidant total defenses and sRAGE levels were significantly lower in AD patients compared to controls. In AD patients lag-time values show a significant positive linear correlation with sRAGE levels and a (even not significant) negative correlation with ROS levels. Lag-time is significantly lower in ε4 carrier ($N = 13$) than in ε4 non-carrier ($N = 12$). Our result confirms the substantial OS in AD. Lag-time levels showed a significant positive correlation with sRAGE levels and a significant association with ε4 carrier status suggesting that plasmatic lag-time evaluation can be considered as a potential useful OS risk marker in AD.

Keywords: Oxidative stress, Vascular dysfunction, Age-associated diseases, Alzheimer's disease

Introduction

Alzheimer disease (AD) accounts for the largest proportion of dementia diseases in the older population [1]. Recent evidences indicated that vascular dysfunction and damage are linked to cerebrovascular disorders in the elderly and increase significantly AD incidence [2]. The vascular endothelium is a major target of oxidative stress (OS) caused by Reactive Oxygen species (ROS), which play a critical role in the pathophysiology of vascular disease. ROS are important regulators of the inflammatory response: on one hand, at low concentration they act as regulators of cell growth and activity in the inflammatory process, on the other hand, at high concentration they have deleterious effects on cells and tissues [3]. The oxidative stress results from an imbalance between ROS and antioxidant molecules, resulting in an excess of ROS leading to cell injury and death and it is commonly associated with ageing process and age-related degenerative disorders [4]. Compared to other organs, the brain is more vulnerable to oxidative stress due to its high rate of oxygen consumption [4]. In the brain, OS also contribute to structural and functional changes associated with vascular aging, such as endothelial dysfunction, extracellular matrix degradation, resulting in age-related reduced vasodilatation in response to agonists [5]. For this

* Correspondence: emanuela.galliera@unimi.it
[1]Department of Biomedical Sciences for Health, Università degli Studi di Milano, Milan, Italy
[2]IRCCS Galeazzi Orthopaedic Institute, Milan, Italy
Full list of author information is available at the end of the article

Ethics approval and consent to participate
The study was carried out in accordance with recommendation of ethical committee of Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico (approval number: 441/2016) All subjects gave written informed consent in accordance with the Declaration of Helsinki.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

Publisher's Note

Author details
[1]Department of Biomedical Sciences for Health, Università degli Studi di Milano, Milan, Italy. [2]IRCCS Galeazzi Orthopaedic Institute, Milan, Italy. [3]Department of Pathophysiology and Transplantation, Università degli Studi di Milano, Centro "Dino Ferrari", Milan, Italy. [4]U.O.S.D. Neurologia-Malattie Neurodegenerative, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy. [5]Department of Biomedical, Surgical and Dental Sciences, Università degli Studi di Milano, Milan, Italy. [6]Department of Health's Science, Università degli Studi di Milano, Milan, Italy. [7]U.O.C SMEL-1 Patologia Clinica IRCCS Policlinico San Donato, San Donato, Milan, Italy.

Received: 17 December 2018 Accepted: 14 March 2019
Published online: 01 April 2019

References

1. Di Marco LY, Venneri A, Farkas E, Evans PC, Marzo A, Frangi AF. Vascular dysfunction in the pathogenesis of Alzheimer's disease--a review of endothelium-mediated mechanisms and ensuing vicious circles. Neurobiol Dis. 2015 Oct;82:593–606.
2. Kalaria RN. The role of cerebral ischemia in Alzheimer's disease. Neurobiol Aging. 2000 Mar-Apr;21(2):321–30.
3. Chen Q, Wang Q, Zhu J, Xiao Q, Zhang L. Reactive oxygen species: key regulators in vascular health and diseases. Br J Pharmacol. 2018 Apr;175(8):1279–92.
4. Aliev G, Smith MA, Seyidov D, Neal ML, Lamb BT, Nunomura A, et al. The role of oxidative stress in the pathophysiology of cerebrovascular lesions in Alzheimer's disease. Brain Pathol. 2002 Jan;12(1):21–35.
5. Carvalho C, Moreira PI. Oxidative stress: a major player in cerebrovascular alterations associated to neurodegenerative events. Front Physiol. 2018;9:806.
6. Bonet-Costa V, Pomatto LC, Davies KJ. The Proteasome and Oxidative Stress in Alzheimer's Disease. Antioxid Redox Signal. 2016;25(16):886–901. https://doi.org/10.3389/fphys.2018.00806.
7. Pratico D, Clark CM, Liun F, Rokach J, Lee VY, Trojanowski JQ. Increase of brain oxidative stress in mild cognitive impairment: a possible predictor of Alzheimer disease. Arch Neurol. 2002;59(6):972–6.
8. Zhao Y, Zhao B. Oxidative stress and the pathogenesis of Alzheimer's disease. Oxidative Med Cell Longev. 2013:316523. https://doi.org/10.1155/2013/316523.
9. Tamaoka A, Miyatake F, Matsuno S, Ishii K, Nagase S, Sahara N, et al. Apolipoprotein E allele-dependent antioxidant activity in brains with Alzheimer's disease. Neurology. 2000 Jun 27;54(12):2319–21.
10. Badia MC, Lloret A, Giraldo E, Dasi F, Olaso G, Alonso MD, et al. Lymphocytes from young healthy persons carrying the ApoE4 allele overexpress stress-related proteins involved in the pathophysiology of Alzheimer's disease. J Alzheimers Dis. 2013;33(1):77–83. https://doi.org/10.3233/JAD-2012-120973.
11. Trotti R, Carratelli M, Barbieri M, Micieli G, Bosone D, Rondanelli M, et al. Oxidative stress and a thrombophilic condition in alcoholics without severe liver disease. Haematologica. 2001;86(1):85–91.
12. Cervato G, Viani P, Cazzola R, Cestaro B. A fluorescence method for the determination of plasma susceptibility to lipid peroxidation. Clin Biochem. 1999 Apr;32(3):171–7.
13. Wevers NR, de Vries HE. Morphogens and blood-brain barrier function in health and disease. Tissue Barriers. 2015;4(1):e1090524. https://doi.org/10.1080/21688370.2015.1090524.
14. Lopez N, Tormo C, De Blas I, Llinares I, Alom J. Oxidative stress in Alzheimer's disease and mild cognitive impairment with high sensitivity and specificity. J Alzheimers Dis. 2013;33(3):823–9. https://doi.org/10.3233/JAD-2012-121528.
15. Chico L, Simoncini C, Lo Gerfo A, Rocchi A, Petrozzi L, Carlesi C, et al. Oxidative stress and APO E polymorphisms in Alzheimer's disease and in mild cognitive impairment. Free Radic Res. 2013 Aug;47(8):569–76. https://doi.org/10.3109/10715762.2013.804622.
16. Cestaro B, Gandini R, Viani P, Maraffi F, Cervato G, Montalto C, et al. Fluorescence-determined kinetics of plasma high oxidability in diabetic patients. Biochem Mol Biol Int. 1994 Apr;32(5):983–94.
17. Atamer A, Kocyigit Y, Ecder SA, Selek S, Ilhan N, Ecder T, et al. Effect of oxidative stress on antioxidant enzyme activities, homocysteine and lipoproteins in chronic kidney disease. J Nephrol. 2008 Nov-Dec;21(6):924–30.
18. Cheignon C, Tomas M, Bonnefont-Rousselot D, Faller P, Hureau C, Collin F. Oxidative stress and the amyloid beta peptide in Alzheimer's disease. Redox Biol. 2018 Apr;14:450–64. https://doi.org/10.1016/j.redox.2017.10.014.
19. Collin F, Cheignon C, Hureau C. Oxidative stress as a biomarker for Alzheimer's disease. Biomark Med. 2018 Mar;12(3):201–3. https://doi.org/10.2217/bmm-2017-0456.
20. de Leeuw FA, Peeters CFW, Kester MI, Harms AC, Struys EA, Hankemeier T, et al. Blood-based metabolic signatures in Alzheimer's disease. Alzheimers Dement (Amst). 2017;8:196–207. https://doi.org/10.1016/j.dadm.2017.07.006.
21. Markesbery WR. Oxidative stress hypothesis in Alzheimer's disease. Free Radic Biol Med. 1997;23(1):134–47.
22. Drenth H, Zuidema SU, Krijnen WP, Bautmans I, van der Schans C, Hobbelen H. Association between advanced glycation end-products and functional performance in Alzheimer's disease and mixed dementia. Int Psychogeriatr. 2017 Sep;29(9):1525–34. https://doi.org/10.1017/S1041610217000886.
23. Ajith TA, Vinodkumar P. Advanced Glycation end products: association with the pathogenesis of diseases and the current therapeutic advances. Curr Clin Pharmacol. 2016;11(2):118–27.
24. Xie J, Mendez JD, Mendez-Valenzuela V, Aguilar-Hernandez MM. Cellular signalling of the receptor for advanced glycation end products (RAGE). Cell Signal. 2013 Nov;25(11):2185–97. https://doi.org/10.1016/j.cellsig.2013.06.013.
25. Walker D, Lue LF, Paul G, Patel A, Sabbagh MN. Receptor for advanced glycation endproduct modulators: a new therapeutic target in Alzheimer's disease. Expert Opin Investig Drugs. 2015;24(3):393–9. https://doi.org/10.1517/13543784.2015.1001490.
26. Juranek J, Ray R, Banach M, Rai V. Receptor for advanced glycation end-products in neurodegenerative diseases. Rev Neurosci. 2015;26(6):691–8. https://doi.org/10.1515/revneuro-2015-0003.
27. Uttara B, Singh AV, Zamboni P, Mahajan RT. Oxidative stress and neurodegenerative diseases: a review of upstream and downstream antioxidant therapeutic options. Curr Neuropharmacol. 2009 Mar;7(1):65–74.
28. Sultana R, Baglioni M, Cecchetti R, Cai J, Klein JB, Bastiani P, et al. Lymphocyte mitochondria: toward identification of peripheral biomarkers in the progression of Alzheimer disease. Free Radic Biol Med. 2013 Dec;65:595–606. https://doi.org/10.1016/j.freeradbiomed.2013.08.001.
29. Swomley AM, Butterfield DA. Oxidative stress in Alzheimer disease and mild cognitive impairment: evidence from human data provided by redox proteomics. Arch Toxicol. 2015 Oct;89(10):1669–80. https://doi.org/10.1007/s00204-015-1556-z.
30. Garcia-Blanco A, Baquero M, Vento M, Gil E, Bataller L, Chafer-Pericas C. Potential oxidative stress biomarkers of mild cognitive impairment due to Alzheimer disease. J Neurol Sci. 2017 Feb 15;373:295–302. https://doi.org/10.1016/j.jns.2017.01.020.
31. Kamat PK, Kalani A, Rai S, Swarnkar S, Tota S, Nath C, et al. Mechanism of oxidative stress and synapse dysfunction in the pathogenesis of Alzheimer's disease: understanding the therapeutics strategies. Mol Neurobiol. 2016 Jan;53(1):648–61. https://doi.org/10.1007/s12035-014-9053-6.
32. Guidi I, Galimberti D, Lonati S, Novembrino C, Bamonti F, Tiriticco M, et al. Oxidative imbalance in patients with mild cognitive impairment and Alzheimer's disease. Neurobiol Aging. 2006 Feb;27(2):262–9.
33. Miyata M, Smith JD. Apolipoprotein E allele-specific antioxidant activity and effects on cytotoxicity by oxidative insults and beta-amyloid peptides. Nat Genet. 1996 Sep;14(1):55–61.

CAMs shed light on one of the major debates in EAE models and MS, which has centred around how lymphocytes are primed to target myelin[4,5]. Peripheral monocytes and dendritic cells invade the CNS early in the inflammatory progression of EAE and MS, which means that many antigen-presenting cells (including the central myeloid cells, CAMs and microglia) within the brain could potentially prime and recruit T cells and B cells. Compared with homeostatic CAMs, disease-associated CAMs upregulated genes required for MHC class II presentation of antigens to lymphocytes, suggesting that disease-associated CAMs might be functionally responsible for this priming. To test this hypothesis, the progression of EAE was assessed in a transgenic mouse in which MHC class II presentation was abolished in all CAMs and microglia. Unexpectedly, abolishing the antigen-presenting capacity of these cell types had no effect on disease course or severity. Only when MHC class II machinery was conditionally deleted from both central and peripheral myeloid cells did mice become resistant to EAE. Lymphocytes invading the CNS also showed significantly longer duration of contact with peripherally derived monocytes than with tissue-resident macrophages. This finding suggests that the peripherally invading myeloid cells are crucial for recruiting T cells, and that the robust expression of antigen-presenting machinery in disease-associated CAMs is redundant.

In many fields, the application of scRNA-seq has revolutionized our understanding of the complex and dynamic nature of cellular processes[6]. In the past, profiling of myeloid cells was performed in bulk. That approach, although providing insights into the common biology occurring collectively in a group of cells, collapsed the heterogeneity of the response of individual cells to a particular stimulus. The work of Jordão et al.[3] confirms the vastness of this heterogeneity by demonstrating just how distinct myeloid cells in the brain can be, not just indicated by their cell type (that is, microglia, CAMs or monocyte-derived cells), but on the basis of their location and disease state. Using this single-cell data set, we can now define markers that might be more relevant to classifying a distinct cellular process. For example, Jordão et al. demonstrate that classic microglial markers such as *Tmem119* are dynamic in their expression between different microglial subclusters, but that other targets such as *Olfml3* and *Sparc* might serve as more universal microglial markers. Additionally, *Ms4a7* was expressed by all CAMs across all brain regions and stages of inflammation and might be a robust marker for studying relevant biology in this population of cells.

Beyond illustrating the complexity underlying the immune response in EAE, this work also sets the stage for the development of novel, targeted disease-modifying therapies (DMTs) for use in inflammatory demyelinating diseases such as MS[7]. Prior DMTs have utilized hammer-like approaches to target inflammation broadly, in the hope of stemming the course of autoimmune destruction. From this work, we have gained evidence that although monocyte-derived myeloid cells are crucial for antigen presentation early in the disease course, they diminish in numbers during the chronic stage of inflammation. Concomitantly, microglia undergo clonal expansion over the course of disease and are the predominant myeloid cell type in the parenchyma during chronic inflammation. Therapies aimed at countering inflammation at early versus late stages during the demyelinating process must, therefore, target different cellular processes and responses. Armed with the single-cell data for each of the relevant cell types, future efforts can target specific cells and minimize broad off-target effects.

Important questions still remain unanswered. For instance, multiple mouse models of EAE exist that utilize different antigens to elicit inflammation or use different toxic agents that damage myelin, and debate is ongoing regarding which models have the most translational relevance. Application of this single-cell approach to the different models could ascertain the commonality in immune responses within these disparate modalities of inciting demyelination in mice. Ultimately, application of single-cell profiling to brain biopsy samples or post-mortem tissue from patients with MS could enable definitive mapping of conserved cellular immune responses in mice and humans, which would greatly accelerate the development of therapeutic interventions for inflammatory conditions that affect the CNS.

*Ravikiran M. Raju[1] and Li-Huei Tsai[2]**

[1]Division of Newborn Medicine, Boston Children's Hospital, Harvard Medical School, Boston, MA, USA.

[2]Picower Institute for Learning and Memory, Massachusetts Institute of Technology, Cambridge, MA, USA.

**e-mail: lhtsai@mit.edu*

https://doi.org/10.1038/s41582-019-0165-5

1. Dendrou, C. A., McVean, G. & Fugger, L. Neuroinflammation — using big data to inform clinical practice. *Nat. Rev. Neurol.* **12**, 685–698 (2016).
2. Amor, S. et al. Identification of epitopes of myelin oligodendrocyte glycoprotein for the induction of experimental allergic encephalomyelitis in SJL and Biozzi AB/H mice. *J. Immunol.* **153**, 4349–4356 (1994).
3. Jordão, M. J. C. et al. Single-cell profiling identifies myeloid cell subsets with distinct fates during neuroinflammation. *Science* **363**, eaat7554 (2019).
4. King, I. L., Dickendesher, T. L. & Segal, B. M. Circulating Ly-6C$^+$ myeloid precursors migrate to the CNS and play a pathogenic role during autoimmune demyelinating disease. *Blood* **113**, 3190–3197 (2009).
5. Tompkins, S. M. et al. De novo central nervous system processing of myelin antigen is required for the initiation of experimental autoimmune encephalomyelitis. *J. Immunol.* **168**, 4173–4183 (2002).
6. Wang, Y. & Navin, N. E. Advances and applications of single-cell sequencing technologies. *Mol. Cell* **58**, 598–609 (2015).
7. Glatigny, S. & Bettelli, E. Experimental autoimmune encephalomyelitis (EAE) as animal models of multiple sclerosis (MS). *Cold Spring Harb. Perspect. Med.* **8**, a028977 (2018).

Competing interests
The authors declare no competing interests.

NEURODEGENERATIVE DISEASE

Genetic risk factors and role of immune dysfunction in FTLD

Daniela Galimberti

A new study has identified novel genes involved in sporadic frontotemporal lobar degeneration with neuronal inclusions of TAR DNA-binding protein 43. These findings might enable the elucidation of pathogenic mechanisms of the disease and have implications for the identification of potential therapeutic targets.

Refers to Pottier, C. et al. Genome-wide analyses as part of the international FTLD-TDP whole-genome sequencing consortium reveals novel disease risk factors and increases support for immune dysfunction in FTLD. *Acta Neuropathol.* https://doi.org/10.1007/s00401-019-01962-9 (2019).

Frontotemporal lobar degeneration (FTLD) with neuronal inclusions of TAR DNA-binding protein 43 (FTLD-TDP) is the most common pathological subtype of FTLD[1]. Five distinct FTLD-TDP subtypes (types A–E) have been described on the basis of different neuronal and cytoplasmic distributions of TDP aggregates[2]. In the past few years, many monogenic autosomal dominant causes of FTLD have been identified. The majority

of these mutations are located in the *MAPT*, progranulin (*GRN*) and *C9orf72* genes[3], and *GRN* and *C9orf72* mutations have been associated with TDP pathology[2]. Most FTLD cases, with or without TDP pathology, remain genetically unexplained, but a new study by Pottier et al.[1] has attempted to remedy this situation by identifying novel genetic risk factors for FTLD.

FTLD is an umbrella term for a highly heterogeneous group of clinical syndromes[4]. The most common clinical presentation is behavioural variant frontotemporal dementia (bvFTD), which is characterized by behavioural disturbances, disinhibition, lack of empathy, and aggressiveness or, more rarely, apathetic mood. Other typical presentations involve language deficits, and include the agrammatic and semantic variants of primary progressive aphasia. Nevertheless, the disease can present with other nontypical syndromes, including bvFTD with motor neuron disease, progressive supranuclear palsy or corticobasal syndrome. Memory impairment can also occur, thus mimicking Alzheimer disease, as can psychoses, leading to a psychiatric diagnosis. The age at disease onset is heterogeneous, although in the majority of cases symptoms manifest in the presenile (<65 years) age group.

In light of the above observations, diagnosis and future treatment of FTLD are extremely challenging as it is difficult to predict the pathology on the basis of symptoms. The development of disease-modifying treatments that affect the underlying pathology requires improved understanding of the pathogenic mechanisms involved in the deposition of different proteins in the brain, and the identification of biomarkers that can predict the underlying pathology during a patient's life.

The study by Pottier and colleagues[1] was recently published in *Acta Neuropathologica* and involved 1,154 unrelated patients with FTLD-TDP from 23 sites in Europe, North America and Australia (one patient per family) for whom a DNA sample had been conserved after death. Considering the vast phenotypic heterogeneity of FTLD-TDP, the pathological diagnosis was considered sufficient for inclusion, irrespective of the clinical diagnosis that each patient had received.

First, patients were screened for the two most common types of mutation associated with TDP pathology, namely *C9orf72* repeat expansions and pathogenic *GRN* mutations. Of 1,134 white patients, 289 (25.5%) had repeat expansions in *C9orf72* and 157 (13.8%) had *GRN* mutations, so together these mutations explained the disease in nearly 40% of the white patient population. *TBK1* mutations and other rare monogenic causes of FTLD-TDP accounted for 0.9% and 1.2% of cases, respectively, in this patient population.

To identify genetic factors contributing to the disease in patients with FTLD-TDP without a known gene mutation, Pottier et al. performed whole-genome sequencing on 554 cases with sufficient DNA quality and quantity available. Among these individuals, differences in disease onset and course were observed depending on the TDP subtype classification. Patients with the type A pathological subtype had the oldest onset of disease, whereas patients categorized as having subtype C had the longest survival after onset.

> “differences in disease onset and course were observed depending on the TDP subtype classification”

Analysis of whole-genome sequencing data showed that the strongest signal was found in *DPP6*, which encodes a type II transmembrane protein exclusively expressed in neurons. An additional 12 loci showing a suggestive association were identified, but two failed the replication analysis. Moreover, a meta-analysis combining the discovery and replication stages resulted in the identification of three FTLD-TDP risk loci with genome-wide significance: one new locus in *DPP6* and two known loci in *UNC13A* and *HLA-DQA2*. Additional genes regulated by *TBK1* were also found to have genome-wide significance, highlighting the importance of this gene, which is well known for its role in the autophagy pathway, in FTLD-TDP.

Notably, all of the FTLD-TDP risk loci that Pottier et al. identified were in genes involved in the regulation of innate immunity pathways. Moreover, a variant in *UNC13A* has been shown to act as a phenotypic modifier in patients with amyotrophic lateral sclerosis (ALS), by increasing the risk of frontotemporal cortical atrophy and impaired cognitive performance, reminiscent of the FTLD clinical presentation[5].

The implications of these findings are manifold. First, from a clinical point of view, a large proportion of white patients with FTLD-TDP carry mutations in *C9orf72* or *GRN*. With this knowledge, the pathology of FTLD-TDP could be predicted during the lives of patients harbouring such mutations, and these individuals could be involved in clinical trials of compounds that can interfere with TDP pathology, once available. In carriers of specific mutations, different therapeutic strategies could be pursued, such as progranulin replacement in *GRN* mutation carriers, or antisense RNA in *C9orf72* repeat expansion carriers.

Second, genetic analysis should be considered, independent of clinical phenotype, in patients with mild FTLD-TDP symptoms, particularly those with a positive family history of dementia, ALS or psychiatric conditions. Evaluation of plasma levels of progranulin is a useful tool for identifying patients who should be screened further by sequencing *GRN*[6].

Last, the findings from this study strongly implicate the immune pathway in FTLD-TDP pathogenesis, supporting previous data suggesting involvement of neuroinflammation and immune dysfunction in FTLD[7]. Moreover, microglial activation is a hallmark of FTLD, and both *GRN* and *C9orf72* mutations have been extensively linked with neuroinflammation and microglial activation[8].

In conclusion, studies that contribute to our knowledge of the genetic background of sporadic FTLD, such as the one by Pottier et al., are extremely important for determining the pathogenic mechanisms that underlie the disease, and might in turn result in the identification of therapeutic targets.

Daniela Galimberti[1,2]

[1]*Department of Biomedical, Surgical and Dental Sciences, University of Milan, Centro Dino Ferrari, Milan, Italy.*

[2]*Neurodegenerative Diseases Unit, Fondazione IRCCS Ca' Granda, Ospedale Policlinico, Milan, Italy.*

e-mail: daniela.galimberti@unimi.it

https://doi.org/10.1038/s41582-019-0173-5

1. Pottier, C. et al. Genome-wide analyses as part of the international FTLD-TDP whole-genome sequencing consortium reveals novel disease risk factors and increases support for immune dysfunction in FTLD. *Acta Neuropathol.* https://doi.org/10.1007/s00401-019-01962-9 (2019).
2. Lee, E. B. et al. Expansion of the classification of FTLD-TDP: distinct pathology associated with rapidly progressive frontotemporal degeneration. *Acta Neuropathol.* **134**, 65–78 (2017).
3. Fenoglio, C. et al. Role of genetics and epigenetics in the pathogenesis of Alzheimer's disease and frontotemporal dementia. *J. Alzheimers Dis.* **62**, 913–932 (2018).
4. Bang, J., Spina, S. & Miller, B. L. Frontotemporal dementia. *Lancet* **386**, 1672–1682 (2015).
5. Placek, K. et al. UNC3A polymorphism contributes to frontotemporal disease in sporadic amyotrophic lateral sclerosis. *Neurobiol. Aging* **73**, 190–199 (2018).
6. Galimberti, D. et al. Progranulin plasma levels predict the presence of GRN mutations in asymptomatic subjects and do not correlate with brain atrophy: results from the GENFI study. *Neurobiol. Aging* **62**, 245.e9–245.e12 (2018).
7. McCauley, M. E. & Baloh, R. H. Inflammation in ALS/FTD pathogenesis. *Acta Neuropathol.* https://doi.org/10.1007/s00401-018-1933-9 (2019).
8. Haukedal, H. & Freude, K. Implications of microglia in amyotrophic lateral sclerosis and frontotemporal dementia. *J. Mol. Biol.* https://doi.org/10.1016/j.jmb.2019.02.004 (2019).

Competing interests
The author declares no competing interests.

cells

MDPI

Article

Untangling Extracellular Proteasome-Osteopontin Circuit Dynamics in Multiple Sclerosis

Chiara Dianzani [1], Domizia Vecchio [2], Nausicaa Clemente [2], Annalisa Chiocchetti [2], Filippo Martinelli Boneschi [3,4], Daniela Galimberti [5,6], Umberto Dianzani [2], Cristoforo Comi [2,7], Michele Mishto [8,9,†,*] and Juliane Liepe [10,†,*]

1 Department of Drug Science and Technology, University of Turin, 10126 Torino, Italy; chiara.dianzani@unito.it
2 Interdisciplinary Research Centre of Autoimmune Diseases (IRCAD), University of Piemonte Orientale, Amedeo Avogadro, 28100 Novara, Italy; domizia.vecchio@gmail.com (D.V.); nausicaa.clemente@med.uniupo.it (N.C.); annalisa.chiocchetti@med.uniupo.it (A.C.); umberto.dianzani@med.uniupo.it (U.D.); cristoforo.comi@med.uniupo.it (C.C.)
3 Department of Biomedical Sciences for Health, University of Milan, 20122 Milan, Italy; filippo.martinelli@unimi.it
4 MS Research Unit and Department of Neurology, IRCCS Policlinico San Donato, San Donato Milanese, 20097 Milan, Italy
5 Department of Biomedical, Surgical and Dental Sciences, University of Milan, "Dino Ferrari" Centre, 20100 Milano, Italy; daniela.galimberti@unimi.it
6 Fondazione IRCCS Cà Granda, Ospedale Maggiore Policlinico, 20100 Milano, Italy
7 Department of Translational Medicine, Section of Neurology, University of Piemonte Orientale, 28100 Novara, Italy
8 Centre for Inflammation Biology and Cancer Immunology (CIBCI) & Peter Gorer Department of Immunobiology, King's College London, SE1 1UL London, UK
9 Institute for Biochemistry, Charité–Universitätsmedizin Berlin, corporate member of Freie Universität Berlin, Humboldt-Universität zu Berlin, and Berlin Institute of Health, Institut für Biochemie, Germany, 10117 Berlin, Germany
10 Max-Planck-Institute for Biophysical Chemistry, 37077 Göttingen, Germany
* Correspondence: michele.mishto@kcl.ac.uk (M.M.); jliepe@mpibpc.mpg.de (J.L.); Tel.: +44-(0)20-7848-6907 (M.M.); +49-(0)551-201-1471 (J.L.)
† These authors equally contributed.

Received: 14 January 2019; Accepted: 18 March 2019; Published: 20 March 2019

Abstract: The function of proteasomes in extracellular space is still largely unknown. The extracellular proteasome-osteopontin circuit has recently been hypothesized to be part of the inflammatory machinery regulating relapse/remission phase alternation in multiple sclerosis. However, it is still unclear what dynamics there are between the different elements of the circuit, what the role of proteasome isoforms is, and whether these inflammatory circuit dynamics are associated with the clinical severity of multiple sclerosis. To shed light on these aspects of this novel inflammatory circuit, we integrated in vitro proteasome isoform data, cell chemotaxis cell culture data, and clinical data of multiple sclerosis cohorts in a coherent computational inference framework. Thereby, we modeled extracellular osteopontin-proteasome circuit dynamics during relapse/remission alternation in multiple sclerosis. Applying this computational framework to a longitudinal study on single multiple sclerosis patients suggests a complex interaction between extracellular proteasome isoforms and osteopontin with potential clinical implications.

Keywords: immunoproteasome; chemotaxis; computational modelling; system biology

Carandini et al. Alzheimer's Research & Therapy (2019) 11:84
https://doi.org/10.1186/s13195-019-0543-7

Alzheimer's Research & Therapy

RESEARCH Open Access

Testing the 2018 NIA-AA research framework in a retrospective large cohort of patients with cognitive impairment: from biological biomarkers to clinical syndromes

Tiziana Carandini[1,2*], Andrea Arighi[1,2], Luca Sacchi[1,2], Giorgio G. Fumagalli[1,2,3], Anna M. Pietroboni[1,2], Laura Ghezzi[1,2], Annalisa Colombi[1,2], Marta Scarioni[1,2], Chiara Fenoglio[2], Milena A. De Riz[1,2], Giorgio Marotta[1], Elio Scarpini[1,2†] and Daniela Galimberti[1,2†]

Abstract

Background: According to the 2018 NIA-AA research framework, Alzheimer's disease (AD) is not defined by the clinical consequences of the disease, but by its underlying pathology, measured by biomarkers. Evidence of both amyloid-β (Aβ) and phosphorylated tau protein (p-tau) deposition—assessed interchangeably with amyloid-positron emission tomography (PET) and/or cerebrospinal fluid (CSF) analysis—is needed to diagnose AD in a living person. Our aim was to test the new NIA-AA research framework in a large cohort of cognitively impaired patients to evaluate correspondence between the clinical syndromes and the underlying pathologic process testified by biomarkers.

Methods: We retrospectively analysed 628 subjects referred to our centre in suspicion of dementia, who underwent CSF analysis, together with neuropsychological assessment and neuroimaging, and were diagnosed with different neurodegenerative dementias according to current criteria, or as cognitively unimpaired. Subjects were classified considering CSF biomarkers, and the prevalence of normal, AD-continuum and non-AD profiles in each clinical syndrome was calculated. The positivity threshold of each CSF biomarker was first assessed by receiver operating characteristic analysis, using Aβ-positive/negative status as determined by amyloid-PET visual reads. The agreement between CSF and amyloid-PET data was also evaluated.

Results: Among patients with a clinical diagnosis of AD, 94.1% were in the AD-continuum, whereas 5.5% were classified as non-AD and 0.4% were normal. The AD-continuum profile was found also in 26.2% of frontotemporal dementia, 48.6% of Lewy body dementia, 25% of atypical parkinsonism and 44.7% of vascular dementia. Biomarkers' profile did not differ in amnestic and not amnestic mild cognitive impairment. CSF Aβ levels and amyloid-PET tracer binding negatively correlated, and the concordance between the two Aβ biomarkers was 89%.

(Continued on next page)

* Correspondence: tizianacarandini@gmail.com
†Elio Scarpini and Daniela Galimberti contributed equally to this work.
[1]Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Via F. Sforza 35, 20122 Milan, Italy
[2]Dino Ferrari Center, University of Milan, Milan, Italy
Full list of author information is available at the end of the article

ARTICLE OPEN ACCESS

Ventricular volume expansion in presymptomatic genetic frontotemporal dementia

Tamara P. Tavares, Derek G.V. Mitchell, PhD, Kristy Coleman, MSc, Christen Shoesmith, MD, Robert Bartha, PhD, David M. Cash, PhD, Katrina M. Moore, John van Swieten, MD, Barbara Borroni, MD, Daniela Galimberti, PhD, Maria Carmela Tartaglia, MD, James Rowe, MD, PhD, Caroline Graff, MD, PhD, Fabrizio Tagliavini, MD, Giovanni Frisoni, MD, Stefano Cappa, MD, Robert Laforce, Jr., MD, PhD, Alexandre de Mendonça, MD, Sandro Sorbi, MD, Garrick Wallstrom, PhD, Mario Masellis, MD, PhD, Jonathan D. Rohrer, MD, PhD, and Elizabeth C. Finger, MD, on behalf of the Genetic FTD Initiative, GENFI

Correspondence
Dr. Finger
elizabeth.finger@lhsc.on.ca

Neurology® 2019;93:e1699-e1706. doi:10.1212/WNL.0000000000008386

Abstract

Objective

To characterize the time course of ventricular volume expansion in genetic frontotemporal dementia (FTD) and identify the onset time and rates of ventricular expansion in presymptomatic FTD mutation carriers.

Methods

Participants included patients with a mutation in *MAPT*, *PGRN*, or *C9orf72*, or first-degree relatives of mutation carriers from the GENFI study with MRI scans at study baseline and at 1 year follow-up. Ventricular volumes were obtained from MRI scans using FreeSurfer, with manual editing of segmentation and comparison to fully automated segmentation to establish reliability. Linear mixed models were used to identify differences in ventricular volume and in expansion rates as a function of time to expected disease onset between presymptomatic carriers and noncarriers.

Results

A total of 123 participants met the inclusion criteria and were included in the analysis (18 symptomatic carriers, 46 presymptomatic mutation carriers, and 56 noncarriers). Ventricular volume differences were observed 4 years prior to symptom disease onset for presymptomatic carriers compared to noncarriers. Annualized rates of ventricular volume expansion were greater in presymptomatic carriers relative to noncarriers. Importantly, time-intensive manually edited and fully automated ventricular volume resulted in similar findings.

Conclusions

Ventricular volume differences are detectable in presymptomatic genetic FTD. Concordance of results from time-intensive manual editing and fully automatic segmentation approaches support its value as a measure of disease onset and progression in future studies in both presymptomatic and symptomatic genetic FTD.

From the Graduate Program in Neuroscience and Brain and Mind Institute (T.P.T., D.G.V.M., E.C.F.) and Departments of Clinical Neurological Sciences (C.S., E.C.F.) and Medical Biophysics (R.B.), Robarts Research Institute, Schulich School of Medicine and Dentistry, University of Western Ontario; Parkwood Institute (K.C., E.C.F.), Lawson Health Research Institute, London, Canada; Dementia Research Centre, Department of Neurodegenerative Disease (D.M.C., K.M.M., J.D.R.), UCL Institute of Neurology, Queen Square; Centre for Medical Image Computing (D.M.C.), University College London, UK; Department of Neurology (J.v.S.), Erasmus Medical Center, Rotterdam, the Netherlands; Neurology Unit, Department of Clinical and Experimental Sciences (B.B.), University of Brescia; Department of Pathophysiology and Transplantation (D.G.), "Dino Ferrari" Center, University of Milan, Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Italy; Toronto Western Hospital (M.C.T.), Tanz Centre for Research in Neurodegenerative Disease, Canada; Department of Clinical Neurosciences (J.R.), University of Cambridge, UK; Department NVS (C.G.), Center for Alzheimer Research, Division of Neurogenetics, Karolinska Institutet, Sweden; Fondazione Istituto di Ricovero e Cura a Carattere Scientifico Istituto Neurologico Carlo Besta (F.T.), Milan; Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS) Istituto Centro San Giovanni di Dio Fatebenefratelli (G.F., S.C.), Brescia, Italy; Memory Clinic and LANVIE-Laboratory of Neuroimaging of Aging (G.F.), University Hospitals and University of Geneva, Switzerland; Clinique Interdisciplinaire de Mémoire, Département des Sciences Neurologiques (R.L.), CHU de Québec, and Faculté de Médecine, Université Laval, Canada; Faculty of Medicine (A.d.M.), University of Lisbon, Portugal; Department of Neuroscience, Psychology, Drug Research and Child Health (S.S.), University of Florence, and the IRCCS Foundazione Don Carlo Gnocchi (S.S.), Florence, Italy; Statistics & Data Corporation (G.W.), Tempe, AZ; and LC Campbell Cognitive Neurology Research Unit (M.M.), Department of Medicine, Division of Neurology, Sunnybrook Health Sciences Centre, Sunnybrook Research Institute, Toronto, Canada.

Go to Neurology.org/N for full disclosures. Funding information and disclosures deemed relevant by the authors, if any, are provided at the end of the article.

Coinvestigators are listed at links.lww.com/WNL/A987.

The Article Processing Charge was funded by CIHR grant to E. Finger and M. Masellis.

frontiers in Aging Neuroscience

REVIEW
published: 28 August 2019
doi: 10.3389/fnagi.2019.00232

Exosome Determinants of Physiological Aging and Age-Related Neurodegenerative Diseases

Marianna D'Anca[1], Chiara Fenoglio[1], Maria Serpente[1], Beatrice Arosio[2], Matteo Cesari[2,3], Elio Angelo Scarpini[1,4] and Daniela Galimberti[4,5]*

[1]Department of Pathophysiology and Transplantation, Dino Ferrari Center, Faculty of Medicine and Surgery, University of Milan, Milan, Italy, [2]Department of Clinical Sciences and Community Health, Faculty of Medicine and Surgery, University of Milan, Milan, Italy, [3]Geriatrics Unit, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, Milan, Italy, [4]Neurodegenerative Diseases Unit, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, Milan, Italy, [5]Department of Biomedical, Surgical and Dental Sciences, Dino Ferrari Center, Faculty of Medicine and Surgery, University of Milan, Milan, Italy

Aging is consistently reported as the most important independent risk factor for neurodegenerative diseases. As life expectancy has significantly increased during the last decades, neurodegenerative diseases became one of the most critical public health problem in our society. The most investigated neurodegenerative diseases during aging are Alzheimer disease (AD), Frontotemporal Dementia (FTD) and Parkinson disease (PD). The search for biomarkers has been focused so far on cerebrospinal fluid (CSF) and blood. Recently, exosomes emerged as novel biological source with increasing interest for age-related neurodegenerative disease biomarkers. Exosomes are tiny Extracellular vesicles (EVs; 30–100 nm in size) released by all cell types which originate from the endosomal compartment. They constitute important vesicles for the release and transfer of multiple (signaling, toxic, and regulatory) molecules among cells. Initially considered with merely waste disposal function, instead exosomes have been recently recognized as fundamental mediators of intercellular communication. They can move from the site of release by diffusion and be retrieved in several body fluids, where they may dynamically reflect pathological changes of cells present in inaccessible sites such as the brain. Multiple evidence has implicated exosomes in age-associated neurodegenerative processes, which lead to cognitive impairment in later life. Critically, consolidated evidence indicates that pathological protein aggregates, including Aβ, tau, and α-synuclein are released from brain cells in association with exosomes. Importantly, exosomes act as vehicles between cells not only of proteins but also of nucleic acids [DNA, mRNA transcripts, miRNA, and non-coding RNAs (ncRNAs)] thus potentially influencing gene expression in target cells. In this framework, exosomes could contribute to elucidate the molecular mechanisms underneath neurodegenerative diseases and could represent a promising source of biomarkers. Despite the involvement of exosomes in age-associated neurodegeneration, the study of exosomes and their genetic cargo in physiological aging and in neurodegenerative diseases is still in its infancy. Here, we review, the current knowledge on protein and ncRNAs cargo of exosomes in normal aging and in age-related neurodegenerative diseases.

Keywords: exosomes, aging, non-coding RNA, Alzheimer's disease, frontotemporal dementia, Parkinson's disease

OPEN ACCESS

Edited by:
Gjumrakch Aliev, GALLY International Biomedical Research, United States

Reviewed by:
Fatah Kashanchi, George Mason University, United States
Safikur Rahman, Yeungnam University, South Korea

****Correspondence:***
Chiara Fenoglio
chiara.fenoglio@unimi.it

Received: *18 April 2019*
Accepted: *13 August 2019*
Published: *28 August 2019*

Citation:
D'Anca M, Fenoglio C, Serpente M, Arosio B, Cesari M, Scarpini EA and Galimberti D (2019) Exosome Determinants of Physiological Aging and Age-Related Neurodegenerative Diseases. Front. Aging Neurosci. 11:232. doi: 10.3389/fnagi.2019.00232

Serum neurofilament light chain in genetic frontotemporal dementia: a longitudinal, multicentre cohort study

*Emma L van der Ende, Lieke H Meeter, Jackie M Poos, Jessica L Panman, Lize C Jiskoot, Elise G P Dopper, Janne M Papma, Frank Jan de Jong, Inge M W Verberk, Charlotte Teunissen, Dimitris Rizopoulos, Carolin Heller, Rhian S Convery, Katrina M Moore, Martina Bocchetta, Mollie Neason, David M Cash, Barbara Borroni, Daniela Galimberti, Raquel Sanchez-Valle, Robert Laforce Jr, Fermin Moreno, Matthis Synofzik, Caroline Graff, Mario Masellis, Maria Carmela Tartaglia, James B Rowe, Rik Vandenberghe, Elizabeth Finger, Fabrizio Tagliavini, Alexandre de Mendonça, Isabel Santana, Chris Butler, Simon Ducharme, Alex Gerhard, Adrian Danek, Johannes Levin, Markus Otto, Giovanni B Frisoni, Stefano Cappa, Yolande A L Pijnenburg, Jonathan D Rohrer, John C van Swieten, on behalf of the Genetic Frontotemporal dementia Initiative (GENFI)**

Summary

Background Neurofilament light chain (NfL) is a promising blood biomarker in genetic frontotemporal dementia, with elevated concentrations in symptomatic carriers of mutations in *GRN*, *C9orf72*, and *MAPT*. A better understanding of NfL dynamics is essential for upcoming therapeutic trials. We aimed to study longitudinal NfL trajectories in people with presymptomatic and symptomatic genetic frontotemporal dementia.

Methods We recruited participants from 14 centres collaborating in the Genetic Frontotemporal Dementia Initiative (GENFI), which is a multicentre cohort study of families with genetic frontotemporal dementia done across Europe and Canada. Eligible participants (aged ≥18 years) either had frontotemporal dementia due to a pathogenic mutation in *GRN*, *C9orf72*, or *MAPT* (symptomatic mutation carriers) or were healthy at-risk first-degree relatives (either presymptomatic mutation carriers or non-carriers), and had at least two serum samples with a time interval of 6 months or more. Participants were excluded if they had neurological comorbidities that were likely to affect NfL, including cerebrovascular events. We measured NfL longitudinally in serum samples collected between June 8, 2012, and Dec 8, 2017, through follow-up visits annually or every 2 years, which also included MRI and neuropsychological assessments. Using mixed-effects models, we analysed NfL changes over time and correlated them with longitudinal imaging and clinical parameters, controlling for age, sex, and study site. The primary outcome was the course of NfL over time in the various stages of genetic frontotemporal dementia.

Findings We included 59 symptomatic carriers and 149 presymptomatic carriers of a mutation in *GRN*, *C9orf72*, or *MAPT*, and 127 non-carriers. Nine presymptomatic carriers became symptomatic during follow-up (so-called converters). Baseline NfL was elevated in symptomatic carriers (median 52 pg/mL [IQR 24–69]) compared with presymptomatic carriers (9 pg/mL [6–13]; p<0·0001) and non-carriers (8 pg/mL [6–11]; p<0·0001), and was higher in converters than in non-converting carriers (19 pg/mL [17–28] *vs* 8 pg/mL [6–11]; p=0·0007; adjusted for age). During follow-up, NfL increased in converters (*b*=0·097 [SE 0·018]; p<0·0001). In symptomatic mutation carriers overall, NfL did not change during follow-up (*b*=0·017 [SE 0·010]; p=0·101) and remained elevated. Rates of NfL change over time were associated with rate of decline in Mini Mental State Examination (*b*=–94·7 [SE 33·9]; p=0·003) and atrophy rate in several grey matter regions, but not with change in Frontotemporal Lobar Degeneration-Clinical Dementia Rating scale score (*b*=–3·46 [SE 46·3]; p=0·941).

Interpretation Our findings show the value of blood NfL as a disease progression biomarker in genetic frontotemporal dementia and suggest that longitudinal NfL measurements could identify mutation carriers approaching symptom onset and capture rates of brain atrophy. The characterisation of NfL over the course of disease provides valuable information for its use as a treatment effect marker.

Funding ZonMw and the Bluefield project.

Introduction

Frontotemporal dementia is a common cause of young-onset dementia and is characterised by progressive behavioural or language changes, or both.[1,2] Autosomal dominant inheritance is present in 20–30% of cases, most commonly owing to mutations in granulin (*GRN*), chromosome 9 open reading frame 72 (*C9orf72*), or microtubule-associated protein tau (*MAPT*).[3] With upcoming therapeutic trials, biomarkers are needed to identify the appropriate time to start treatment, probably in the preclinical stage, and as surrogate endpoints to measure treatment effect.

Neurofilament light chain (NfL), a constituent of the axonal cytoskeleton, is a promising diagnostic and prognostic blood biomarker in genetic frontotemporal dementia, with low concentrations in presymptomatic mutation

Lancet Neurol 2019; 18: 1103–11

See Comment page 1070

*Members are listed in the appendix (pp 17–18)

Department of Neurology and Alzheimer Center, Erasmus Medical Center Rotterdam, Rotterdam, Netherlands (E L van der Ende MD, L H Meeter MD, J M Poos MSc, J L Panman MSc, L C Jiskoot PhD, E G P Dopper MD, J M Papma PhD, F J de Jong MD, Prof J C van Swieten MD); **Department of Radiology, Leiden University Medical Center, Leiden, Netherlands** (J L Panman); **Dementia Research Institute, Department of Neurodegenerative Disease, UCL Queen Square Institute of Neurology, London, UK** (L C Jiskoot, C Heller MSc, R S Convery MSc, K M Moore BSc, M Bocchetta PhD, M Neason MSc, D M Cash PhD, J D Rohrer MD); **Neurochemistry Laboratory, Department of Clinical Chemistry, Amsterdam Neuroscience, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, Netherlands** (I M W Verberk MSc, Prof C Teunissen PhD); **Department of Biostatistics, Erasmus Medical Center Rotterdam, Rotterdam, Netherlands** (Prof D Rizopoulos PhD); **Centre for Neurodegenerative Disorders, Neurology unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy** (B Borroni MD); **Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurodegenerative Diseases Unit, Milan, Italy** (D Galimberti PhD); **University of Milan, Centro Dino Ferrari, Milan, Italy** (D Galimberti); **Hospital Clinic de Barcelona, IDIBAPS, University of Barcelona, Barcelona, Spain**

NeuroImage: Clinical 24 (2019) 102077

Contents lists available at ScienceDirect

NeuroImage: Clinical

journal homepage: www.elsevier.com/locate/ynicl

White matter hyperintensities in progranulin-associated frontotemporal dementia: A longitudinal GENFI study

Carole H. Sudre[a,b,c], Martina Bocchetta[b], Carolin Heller[b], Rhian Convery[b], Mollie Neason[b], Katrina M. Moore[b], David M. Cash[b,c], David L. Thomas[b], Ione O.C. Woollacott[b], Martha Foiani[b], Amanda Heslegrave[b], Rachelle Shafei[a], Caroline Greaves[b], John van Swieten[d], Fermin Moreno[e], Raquel Sanchez-Valle[f], Barbara Borroni[g], Robert Laforce Jr[h], Mario Masellis[i], Maria Carmela Tartaglia[j], Caroline Graff[k], Daniela Galimberti[l,m], James B. Rowe[n], Elizabeth Finger[o], Matthis Synofzik[p], Rik Vandenberghe[q], Alexandre de Mendonça[r], Fabrizio Tagliavini[s], Isabel Santana[t], Simon Ducharme[u], Chris Butler[v], Alex Gerhard[w], Johannes Levin[x], Adrian Danek[x], Giovanni B. Frisoni[y], Sandro Sorbi[z], Markus Otto[a1], Henrik Zetterberg[b], Sebastien Ourselin[a], M. Jorge Cardoso[a,b,c], Jonathan D. Rohrer[b,*], on behalf of GENFI[1]

[a] School of Biomedical Engineering and Imaging Sciences, King's College London, UK
[b] Dementia Research Centre, Department of Neurodegenerative Disease, UCL Queen Square Institute of Neurology, London WC1N 3BG, UK
[c] Centre for Medical Image Computing, University College London, UK
[d] Department of Neurology, Erasmus Medical Centre, Rotterdam, Netherlands
[e] Cognitive Disorders Unit, Department of Neurology, Donostia University Hospital, San Sebastian, Gipuzkoa, Spain
[f] Alzheimer's disease and Other Cognitive Disorders Unit, Neurology Service, Hospital Clínic, Institut d'Investigacións Biomèdiques August Pi I Sunyer, University of Barcelona, Barcelona, Spain
[g] Centre for Neurodegenerative Disorders, Neurology Unit, Department of Clinical and Experimental Sciences, University of Brescia, Brescia, Italy
[h] Clinique Interdisciplinaire de Mémoire, Département des Sciences Neurologiques Université Laval Québec, Québec, Canada
[i] Sunnybrook Health Sciences Centre, Sunnybrook Research Institute, University of Toronto, Toronto, Canada
[j] Tanz Centre for Research in Neurodegenerative Diseases, University of Toronto, Toronto, Canada
[k] Department of Geriatric Medicine, Karolinska University Hospital-Huddinge, Stockholm, Sweden
[l] University of Milan, Centro Dino Ferrari, Milan, Italy
[m] Fondazione IRCCS Ca' Granda, Ospedale Policlinico, Neurodegenerative Diseases Unit, Milan, Italy
[n] Department of Clinical Neurosciences, University of Cambridge, Cambridge, UK
[o] Department of Clinical Neurological Sciences, University of Western Ontario, London, Ontario Canada
[p] Department of Neurodegenerative Diseases, Hertie-Institute for Clinical Brain Research and Center of Neurology, University of Tübingen, Tübingen, Germany
[q] Laboratory for Cognitive Neurology, Department of Neurosciences, KU Leuven, Leuven, Belgium
[r] Faculty of Medicine, University of Lisbon, Lisbon, Portugal
[s] Fondazione Istituto di Ricovero e Cura a Carattere Scientifico Istituto Neurologica Carlo Besta, Milano, Italy
[t] Faculty of Medicine, University of Coimbra, Coimbra, Portugal
[u] Department of Psychiatry, McGill University Health Centre, McGill University, Montreal, Québec, Canada
[v] Department of Clinical Neurology, University of Oxford, Oxford, UK
[w] Faculty of Medical and Human Sciences, Institute of Brain, Behaviour and Mental Health, University of Manchester, Manchester, UK
[x] Department of Neurology, Ludwig-Maximilians-University, Munich, Germany
[y] Instituto di Recovero e Cura a Carattere Scientifico Istituto Centro San Giovanni di Dio Fatebenefratelli, Brescia, Italy
[z] Department of Neuroscience, Psychology, Drug Research, and Child Health, University of Florence, Florence, Italy
[a1] Department of Neurology, University of Ulm, Ulm, Germany

Abbreviations: CSF, Cerebrospinal fluid; FTD, Frontotemporal dementia; GENFI, GENetic Frontotemporal dementia Initiative; GFAP, Glial Fibrillary Acidic Protein; GM, Grey Matter; GRN, Progranulin; MRI, Magnetic Resonance Imaging; WM, White Matter; WMH, White Matter Hyperintensity

* Corresponding author.
E-mail address: j.rohrer@ucl.ac.uk (J.D. Rohrer).

[1] List of GENFI consortium authors:

https://doi.org/10.1016/j.nicl.2019.102077
Received 17 July 2019; Received in revised form 3 September 2019; Accepted 4 November 2019
Available online 06 November 2019

Journal of Alzheimer's Disease 68 (2019) 1401–1414
DOI 10.3233/JAD-181253
IOS Press

Expert Opinion

Timely Detection of Mild Cognitive Impairment in Italy: An Expert Opinion

Angelo Bianchetti[a,b,c], Nicola Ferrara[b,c,d], Alessandro Padovani[e], Elio Scarpini[f,g], Marco Trabucchi[b,c,h] and Stefania Maggi[i,*]
[a]*Dipartimento di Medicina e Riabilitazione, Istituto Clinico S. Anna-Gruppo San Donato, Brescia, Italy*
[b]*Italian Society of Gerontology and Geriatrics (SIGG), Florence, Italy*
[c]*Italian Association of Psychogeriatrics (AIP), Brescia, Italy*
[d]*Dipartimento di Scienze Mediche Traslazionali, Università degli Studi di Napoli "Federico II" Naples, Italy*
[e]*Neurology Unit, Dipartimento Scienze Cliniche e Sperimentali, Università degli Studi di Brescia, Brescia, Italy*
[f]*Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy*
[g]*Università di Milano, Centro Dino Ferrari, Milan, Italy*
[h]*University of "Tor Vergata", Rome, Italy*
[i]*CNR, Neuroscience Institute, Aging Branch, Padua, Italy*

Accepted 4 February 2019

Abstract. Mild cognitive impairment (MCI) generally evolves in a gradually progressive decline in memory and non-memory cognitive domains that may eventually decay to dementia. This process might be preventable by improving early detection of the MCI syndrome followed by proper and timely interventions. The aim of this work was providing helpful indications for a standardized early diagnosis of MCI, mainly focusing on the Italian elderly population. We reviewed here MCI epidemiology and classification, as well as the most recent advancements in early detection of the patient with MCI in the Italian scenario. Specialist centers in connection with general practitioners (GPs) have been established across the country and designated as Centers for Cognitive Disorders and Dementia (CDCD). CDCDs are dedicated to the diagnosis and management of patients for all forms of dementia across all the complex staging spectrum. New tools were made available by the advancements of imaging techniques and of the research on biomarkers, leading to novel approaches based on the combination of imaging and biomarker detection, to improve accuracy and effectiveness in the early diagnosis of MCI. Moreover, patient genotyping, alone or in combination with other techniques, was also revealed as a promising method in evaluating and preventing MCI progression. We recommend the introduction of all these novel tools in the diagnostic practice of the specialist centers and that further efforts and resources are spent into the research of the most effective techniques and biomarkers to be introduced as first-level tests into the practice of early diagnosis of MCI.

Keywords: Biomarkers, early diagnosis, genotyping, imaging, mild cognitive impairment, neuropsychological assessment

Mild cognitive impairment (MCI) embodies a disease spectrum including impairment in both memory and non-memory cognitive domains that may further worsen to dementia [1]. However, subjects with MCI can remain stable or even revert to normal cognitive status [2]. In the past, MCI has been defined mainly focusing on amnesia. Some criteria were developed for the definition of MCI based on the presence of isolated memory deficits [3, 4], while according to type and number of the impaired

*Correspondence to: Stefania Maggi, MD, PhD, CNR – Neuroscience Institute, Aging Branch, Via Giustiniani 2, 35128 Padova, Italy. Tel.: +39 049 8211746; Fax: +39 049 8211818; E-mail: stefania.maggi@in.cnr.it.

ISSN 1387-2877/19/$35.00

Aging Clinical and Experimental Research (2019) 31:101–107
https://doi.org/10.1007/s40520-018-1032-8

ORIGINAL ARTICLE

The instruments used by the Italian centres for cognitive disorders and dementia to diagnose mild cognitive impairment (MCI)

Federica Limongi[1] · Marianna Noale[1] · Angelo Bianchetti[2,3,4] · Nicola Ferrara[3,4,5] · Alessandro Padovani[6] · Elio Scarpini[7,8] · Marco Trabucchi[3,4,9] · Stefania Maggi[1] · for the MCI Working Group

Received: 8 June 2018 / Accepted: 27 August 2018 / Published online: 3 September 2018

Abstract

Aims The purpose of this study was to examine the tools used in Italy to diagnose mild cognitive impairment (MCI).

Methods In collaboration with the Luigi Amaducci Research Consortium, the Italian Network of Alzheimer Evaluation Units prepared a questionnaire to describe how MCI is diagnosed in the Italian Centres for cognitive disorders and dementia (CCDD).

Results Most of the ninety-two CCDDs participating in the survey were located in hospitals (54.7%); large percentages were coordinated by neurologists (50.8%) and geriatricians (44.6%). Almost all (98.5%) used the Mini Mental State Examination to diagnose MCI; the Clock Drawing Test was also frequently used (83.9%). Other neuropsychological, imaging and biomarker tests were utilized less frequently and a wide diversity in the instruments used was noted.

Conclusions According to the results, diagnoses of MCI are based on a multitude of instruments, with major differences in the clinical assessment of geriatricians and neurologists. Standardized testing protocols, validated instruments and cut-off points need to be identified and adopted by the CCDDs for assessing MCI.

Keywords Mild cognitive impairment · Diagnosis tools · Italian centres for cognitive disorders and dementia

Introduction

Mild cognitive impairment (MCI) is an intermediate stage of cognitive impairment that often, but not always, represents a prodromal phase of dementia. In accordance with the international criteria based on the Key Symposium held in 2003, MCI is considered a clinical syndrome that has different aetiologies and clinical profiles including impairment not only in memory but also in other cognitive domains [1, 2]. New criteria for dementia recently published by the American Psychiatric Association in the fifth edition of the diagnostic and statistical manual for mental disorders (DSM-5) identified a pre-dementia stage of cognitive impairment defined as mild neurocognitive disorder (NCD). Sharing many features with MCI, the condition represents an initial phase of cognitive impairment that precedes the major neurocognitive disorder that corresponds to outright dementia [3].

Objective cognitive impairment, defined as poor performance on one or more neuropsychological tests or batteries, can refer to any cognitive domain such as executive functions, attention, language, memory and visuospatial skills. Depending on the type and number of cognitive domains

Federica Limongi and Marianna Noale contributed equally to the study.

✉ Marianna Noale
marianna.noale@in.cnr.it

1 CNR, Neuroscience Institute, Aging Branch, Via Giustiniani 2, 35128 Padua, Italy

2 Dipartimento di Medicina e Riabilitazione, Istituto Clinico S.Anna—Gruppo San Donato, Brescia, Italy

3 Società Italiana di Geriatria e Gerontologia (SIGG), Florence, Italy

4 Associazione Italiana di Psicogeriatria (AIP), Brescia, Italy

5 Dipartimento di Scienze Mediche Traslazionali, Università degli Studi di Napoli "Federico II", Naples, Italy

6 Neurology Unit, Dipartimento Scienze Cliniche e Sperimentali, Università degli Studi di Brescia, Brescia, Italy

7 Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

8 Università di Milano, Centro Dino Ferrari, Milan, Italy

9 University of "Tor Vergata", Rome, Italy

MULTIPLE SCLEROSIS JOURNAL | MSJ

Original Research Paper

The loss of macular ganglion cells begins from the early stages of disease and correlates with brain atrophy in multiple sclerosis patients

Multiple Sclerosis Journal

2019, Vol. 25(1) 31–38

DOI: 10.1177/1352458517740214

Anna M Pietroboni, Laura Dell'Arti, Michela Caprioli, Marta Scarioni, Tiziana Carandini, Andrea Arighi, Laura Ghezzi, Giorgio G Fumagalli, Milena A De Riz, Paola Basilico, Annalisa Colombi, Eleonora Benatti, Fabio Triulzi, Elio Scarpini, Francesco Viola and Daniela Galimberti

Abstract

Background: The importance of neurodegeneration in multiple sclerosis (MS) is increasingly well recognized.

Objectives: To evaluate retinal pathology using optical coherence tomography (OCT) and to investigate possible associations between retinal layers' thickness and specific patterns of gray matter volume in patients with a new diagnosis of MS.

Methods: A total of 31 patients underwent OCT scans and brain magnetic resonance imaging. In total, 30 controls underwent the same OCT procedure. The association between focal cortical volume and OCT measurements was investigated with voxel-based morphometry (VBM).

Results: Compared to controls, patients' macular retinal nerve fiber layer (mRNFL), macular ganglion cell layer (mGCL), macular inner plexiform layer (mIPL), and macular ganglion cell-inner plexiform layer (mGCIPL) thickness were significantly reduced ($p=0.0009$, $p=0.0003$, $p=0.0049$, and $p=0.0007$, respectively). Peripapillary RNFL (pRNFL) and temporal sector pRNFL (T-pRNFL) did not show any significant changes, although there was a trend toward T-pRNFL thinning ($p=0.0254$). VBM analysis showed that mGCIPL and pRNFL were significantly correlated with the volume reduction of occipital-parietal cortex ($p<0.005$).

Conclusion: mRNFL, mGCL, and mIPL are significantly reduced in MS patients without concomitant pRNFL thinning. These retinal changes show a significant association with cortical regions that are known to be important for visuospatial performance.

Keywords: Multiple sclerosis, MRI, functional MRI, outcome measurement, atrophy, axonal loss

Date received: 14 June 2017; revised: 6 September 2017; accepted: 7 October 2017

Correspondence to:
AM Pietroboni
Neurodegenerative Disease Unit, Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, University of Milan, Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Via F. Sforza 35, Milan 20122, Italy.
pb.anna@libero.it

Anna M Pietroboni
Michela Caprioli
Marta Scarioni
Tiziana Carandini
Andrea Arighi
Laura Ghezzi
Giorgio G Fumagalli
Milena A De Riz
Paola Basilico
Annalisa Colombi
Elio Scarpini
Daniela Galimberti
Neurodegenerative Disease Unit, Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, University of Milan, Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Milan, Italy

Laura Dell'Arti
Eleonora Benatti
Francesco Viola
Ophthalmological Unit, Department of Clinical Sciences and Community Health, University of Milan, Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Milan, Italy

Fabio Triulzi
Neuroradiology Unit, Department of Pathophysiology and Transplantation, University of Milan, Fondazione Cà Granda, IRCCS Ospedale Maggiore Policlinico, Milan, Italy

Introduction

Multiple sclerosis (MS) is the most common chronic inflammatory disease of the central nervous system (CNS), where demyelination, axonal degeneration/loss, and gliosis are the hallmarks of disease.[1]

Although MS is traditionally regarded as a white matter (WM) demyelinating disease, in which axonal loss occurs late during the disease progression, there is evidence indicating that axonal and neuronal loss may be present since early stages of the disease.[1] Moreover, these neurodegenerative aspects play a major role in determining accumulation of permanent physical and cognitive disabilities.[2,3]

Imaging surrogates, such as brain atrophy on magnetic resonance imaging (MRI) and retinal nerve fiber layer (RNFL) thinning on optical coherence tomography (OCT), can be used for prognostic purposes in MS. As mentioned by Galetta et al.,[4] the earliest application of OCT technology in MS was reported by Parisi et al.[5] in 1999. In this study, the RNFL thickness of the eye with a history of optic neuritis (ON) was shown to be reduced compared to the unaffected

RESEARCH ARTICLE SUMMARY

HUMAN GENOMICS

Multiple sclerosis genomic map implicates peripheral immune cells and microglia in susceptibility

International Multiple Sclerosis Genetics Consortium*†

INTRODUCTION: Multiple sclerosis (MS) is an inflammatory and degenerative disease of the central nervous system (CNS) that often presents in young adults. Over the past decade, certain elements of the genetic architecture of susceptibility have gradually emerged, but most of the genetic risk for MS remained unknown.

RATIONALE: Earlier versions of the MS genetic map had highlighted the role of the adaptive arm of the immune system, implicating multiple different T cell subsets. We expanded our knowledge of MS susceptibility by performing a genetic association study in MS that leveraged genotype data from 47,429 MS cases and 68,374 control subjects. We enhanced this analysis with an in-depth and comprehensive evaluation of the functional impact of the susceptibility variants that we uncovered.

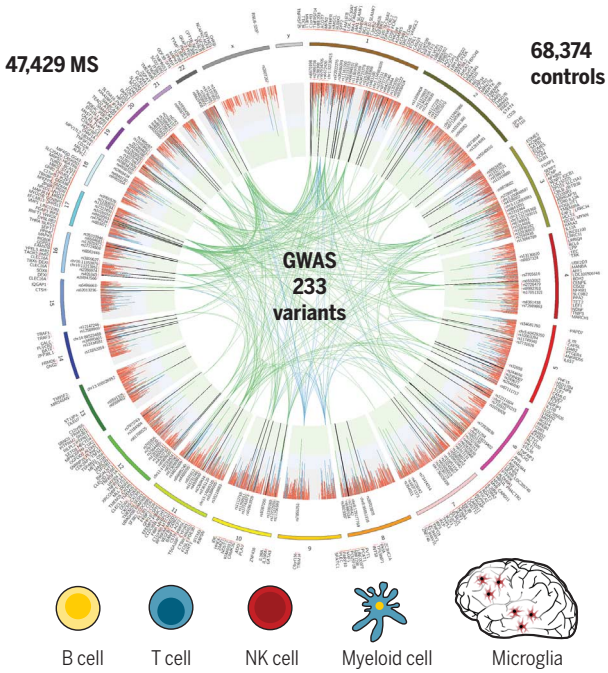
RESULTS: We identified 233 statistically independent associations with MS susceptibility that are genome-wide significant. The major histocompatibility complex (MHC) contains 32 of these associations, and one, the first MS locus on a sex chromosome, is found in chromosome X. The remaining 200 associations are found in the autosomal non-MHC genome. Our genome-wide partitioning approach and large-scale replication effort allowed the evaluation of other variants that did not meet our strict threshold of significance, such as 416 variants that had evidence of statistical replication but did not reach the level of genome-wide statistical significance. Many of these loci are likely to be true susceptibility loci. The genome-wide and suggestive effects jointly explain ~48% of the estimated heritability for MS.

Using atlases of gene expression patterns and epigenomic features, we documented that enrichment for MS susceptibility loci was apparent in many different immune cell types and tissues, whereas there was an absence of enrichment in tissue-level brain profiles. We extended the annotation analyses by analyzing new data generated from human induced pluripotent stem cell–derived neurons as well as from purified primary human astrocytes and microglia, observing that enrichment for MS genes is seen in human microglia, the resident immune cells of the brain, but not in astrocytes or neurons. Further, we have characterized the functional consequences of many MS susceptibility variants by identifying those that influence the expression of nearby genes in immune cells or brain. Last, we applied an ensemble of methods to prioritize 551 putative MS susceptibility genes that may be the target of the MS variants that meet a threshold of genome-wide significance. This extensive list of MS susceptibility genes expands our knowledge more than twofold and highlights processes relating to the development, maturation, and terminal differentiation of B, T, natural killer, and myeloid cells that may contribute to the onset of MS. These analyses focus our attention on a number of different cells in which the function of MS variants should be further investigated.

ON OUR WEBSITE

Read the full article at http://dx.doi.org/10.1126/science.aav7188

Using reference protein-protein interaction maps, these MS genes can also be assembled into 13 communities of genes encoding proteins that interact with one another; this higher-order architecture begins to assemble groups of susceptibility variants whose functional consequences may converge on certain protein complexes that can be prioritized for further evaluation as targets for MS prevention strategies.



The MS genetic map implicates microglia as well as multiple different peripheral immune cell populations in the onset of the disease. We list some of the immune cells in which we found an excess of MS susceptibility genes, implicating these cells as contributing to the earliest events that trigger MS. The sample size of our genome-wide association study is listed along with a circus plot illustrating main results.

CONCLUSION: We report a detailed genetic and genomic map of MS susceptibility, one that explains almost half of this disease's heritability. We highlight the importance of several cells of the peripheral and brain resident immune systems—implicating both the adaptive and innate arms—in the translation of MS genetic risk into an auto-immune inflammatory process that targets the CNS and triggers a neurodegenerative cascade. In particular, the myeloid component highlights a possible role for microglia that requires further investigation, and the B cell component connects to the narrative of effective B cell–directed therapies in MS. These insights set the stage for a new generation of functional studies to uncover the sequence of molecular events that lead to disease onset. This perspective on the trajectory of disease onset will lay the foundation for developing primary prevention strategies that mitigate the risk of developing MS. ■

*The list of authors and affiliations is available in the full article online.
†Corresponding author. Email: pld2115@cumc.columbia.edu
Cite this article as International Multiple Sclerosis Genetics Consortium, *Science* **365**, eaav7188 (2019). DOI: 10.1126/science.aav7188

Berkeley (contact R. Harris, rharris@berkeley.edu, for researchers who meet the criteria for access to confidential data. Please reference the manuscript title and corresponding author in your communication). Corresponding summary statistics for these three GWAS studies (ANZGENE, Rotterdam, and Berkeley) are available upon request.

SUPPLEMENTARY MATERIALS

science.sciencemag.org/content/365/6460/eaav7188/suppl/DC1
Materials and Methods
Supplementary Text
Consortium Members
Figs. S1 to S40
Tables S1 to S53
Data Files S1 to S4
References (*43*–*69*)

The International Multiple Sclerosis Genetics Consortium Nikolaos A Patsopoulos[1,2,3,4], Sergio E. Baranzini[5], Adam Santaniello[5], Parisa Shoostari[4,6,7]*, Chris Cotsapas[4,6,7], Garrett Wong[1,3], Ashley H. Beecham[8], Tojo James[9], Joseph Replogle[2,3,4,10], Ioannis S. Vlachos[1,3,4], Cristin McCabe[4], Tune H. Pers[11], Aaron Brandes[4], Charles White[4,10], Brendan Keenan[12], Maria Cimpean[10], Phoebe Winn[10], Ioannis-Pavlos Panteliadis[1,4], Allison Robbins[10], Till F. M. Andlauer[13,14,15], Onigiusz Zarzycki[1,4], Bénédicte Dubois[16], An Goris[16], Helle Bach Søndergaard[17], Finn Sellebjerg[17], Per Soelberg Sorensen[17], Henrik Ullum[18], Lise Wegner Thørner[18], Janna Saarela[19], Isabelle Cournu-Rebeix[20], Vincent Damotte[20,21], Bertrand Fontaine[20,22], Lena Guillot-Noel[20], Mark Lathrop[23,24,25], Sandra Vukusic[26,27,28], Achim Berthele[14,15], Viola Pongratz[14,15], Dorothea Buck[14,15], Christiane Gasperi[14,15], Christiane Graetz[15,29], Verena Grummel[14,15], Bernhard Hemmer[14,15,30], Muni Hoshi[14,15], Benjamin Knier[14,15], Thomas Korn[14,15,30], Christina M. Lill[15,31,32], Felix Luessi[15,31], Mark Mühlau[14,15], Frauke Zipp[15,31], Efthimios Dardiotis[33], Cristina Agliardi[34], Antonio Amoroso[35], Nadia Barizzone[36], Maria D. Benedetti[37,38], Luisa Bernardinelli[39], Paola Cavalla[40], Ferdinando Clarelli[41], Giancarlo Comi[41,42], Daniele Cusi[43], Federica Esposito[41,44], Laura Ferrè[44], Daniela Galimberti[45,46], Clara Guaschino[41,44], Maurizio A. Leone[47], Vittorio Martinelli[44], Lucia Moiola[44], Marco Salvetti[48,49], Melissa Sorosina[41], Domizia Vecchio[50], Andrea Zauli[41], Silvia Santoro[41], Nicasio Mancini[51], Miriam Zuccalà[52], Julia Mescheriakova[53], Cornelia van Duijn[53,54], Steffan D. Bos[55], Elisabeth G. Celius[55,56], Anne Spurkland[57], Manuel Comabella[58], Xavier Montalban[58], Lars Alfredsson[59], Izaura L. Bomfim[60], David Gomez-Cabrero[60,61,62], Jan Hillert[60], Maja Jagodic[60], Magdalena Lindén[60], Fredrik Piehl[60], Ilijas Jelčić[63,64], Roland Martin[63,64], Mirela Sospedra[63,64], Amie Baker[65], Maria Ban[66], Clive Hawkins[66], Pirro Hysi[67], Seema Kalra[68], Fredrik Karpe[68], Jyoti Khadake[69], Genevieve Lachance[67], Paul Molyneux[67], Matthew Neville[68], John Thorpe[70], Elizabeth Bradshaw[10], Stacy J. Caillier[5], Peter Calabresi[71], Bruce A. C. Cree[5], Anne Cross[72], Mary Davis[73], Paul W. I. de Bakker[2,3,4]†, Silvia Delgado[74], Marieme Dembele[71], Keith Edwards[75], Kate Fitzgerald[71], Irene Y. Frohlich[10], Pierre-Antoine Gourraud[5,76], Jonathan L Haines[77], Hakon Hakonarson[78,79], Dorlan Kimbrough[3,80], Noriko Isobe[5,81], Ioanna Konidari[8], Ellen Lathi[82], Michelle H. Lee[10], Taibo Li[83], David An[83], Andrew Zimmer[83], Lohith Madireddy[5], Clara P. Manrique[8], Mitja Mitrovic[4,6,7], Marta Olah[10], Ellis Patrick[10,84,85], Margaret A. Pericak-Vance[8], Laura Piccio[71], Cathy Schaefer[86], Howard Weiner[87], Kasper Lage[82], ANZgene, IIBDGC, WTCCC2, Alastair Compston[64], David Hafler[4,88], Hanne F. Harbo[54,55], Stephen L. Hauser[5], Graeme Stewart[89], Sandra D'Alfonso[90], Georgios Hadjigeorgiou[33], Bruce Taylor[91], Lisa F. Barcellos[92], David Booth[93], Rogier Hintzen[94], Ingrid Kockum[9], Filippo Martinelli-Boneschi[41,42], Jacob L. McCauley[8], Jorge R. Oksenberg[5], Annette Oturai[16], Stephen Sawcer[62], Adrian J. Ivinson[93], Tomas Olsson[9], Philip L. De Jager[4,10]

[1]Systems Biology and Computer Science Program, Ann Romney Center for Neurological Diseases, Department of Neurology, Brigham & Women's Hospital, Boston, MA 02115, USA. [2]Division of Genetics, Department of Medicine, Brigham & Women's Hospital, Harvard Medical School, Boston, MA, USA. [3]Harvard Medical School, Boston, MA 02115, USA. [4]Broad Institute of Harvard and Massachusetts Institute of Technology, Cambridge, MA, USA. [5]Department of Neurology, University of California at San Francisco, Sandler Neurosciences Center, 675 Nelson Rising Lane, San Francisco, CA 94158, USA. [6]Department of Neurology, Yale University School of Medicine, New Haven, CT 06520, USA. [7]Department of Genetics, Yale School of Medicine, New Haven, CT 06520, USA. [8]John P. Hussman Institute for Human Genomics, University of Miami, Miller School of Medicine, Miami, FL 33136, USA. [9]Department of Clinical Neuroscience, Karolinska Institutet, Stockholm, Sweden. [10]Center for Translational and Computational Neuroimmunology, Multiple Sclerosis Center, Department of Neurology, Columbia University Medical Center, New York, NY, USA. [11]The Novo Nordisk Foundation Center for Basic Metabolic Research, Faculty of Health and Medical Sciences, University of Copenhagen, Copenhagen, 2100, Denmark. [12]Center for Sleep and Circadian Neurobiology, University of Pennsylvania Perelman School of Medicine, Philadelphia, PA. [13]Max Planck Institute of Psychiatry, 80804 Munich, Germany. [14]Department of Neurology, Klinikum rechts der Isar, Technical University of Munich, 81675 Munich, Germany. [15]German competence network for multiple sclerosis. [16]KU Leuven Department of Neurosciences, Laboratory for Neuroimmunology, Herestraat 49 bus 1022, 3000 Leuven, Belgium. [17]Danish Multiple Sclerosis Center, Department of Neurology, Rigshospitalet, University of Copenhagen, Section 6311, 2100 Copenhagen, Denmark. [18]Department of Clinical Immunology, Rigshospitalet, University of Copenhagen, Section 2082, 2100 Copenhagen, Denmark. [19]Institute for Molecular Medicine Finland, University of Helsinki, Helsinki, Finland. [20]ICM-UMR 1127, INSERM, Sorbonne University, Hôpital Universitaire Pitié-Salpêtrière 47 Boulevard de l'Hôpital, F-75013 Paris. [21]UMR1167 Université de Lille, Inserm, CHU Lille, Institut Pasteur de Lille. [22]CRM-UMR974 Department of Neurology Hôpital Universitaire Pitié-Salpêtrière 47 Boulevard de l'Hôpital F-75013 Paris. [23]Commissariat à l'Energie Atomique, Institut Genomique, Centre National de Génotypage, Evry, France. [24]Fondation Jean Dausset - Centre d'Etude du Polymorphisme Humain, Paris, France. [25]McGill University and Genome Quebec Innovation Center, Montreal, Canada. [26]Hospices Civils de Lyon, Service de Neurologie, sclérose en plaques, pathologies de la myéline et neuro-inflammation, F-69677 Bron, France. [27]Observatoire Français de la Sclérose en Plaques, Centre de Recherche en Neurosciences de Lyon, INSERM 1028 et CNRS UMR 5292, F-69003 Lyon, France. [28]Université de Lyon, Université Claude Bernard Lyon 1, F-69000 Lyon, France; Eugène Devic EDMUS Foundation against multiple sclerosis, F-69677 Bron, France. [29]Focus Program Translational Neuroscience (FTN), Rhine Main Neuroscience Network (rmn2), Johannes Gutenberg University-Medical Center, Mainz, Germany. [30]Munich Cluster for Systems Neurology (SyNergy), 81377 Munich, Germany. [31]Department of Neurology, Focus Program Translational Neuroscience (FTN), and Immunology (FZI), Rhine-Main Neuroscience Network (rmn2), University Medical Center of the Johannes Gutenberg University Mainz, Mainz, Germany. [32]Genetic and Molecular Epidemiology Group, Institute of Neurogenetics, University of Luebeck, Luebeck, Germany. [33]Neurology Department, Neurogenetics Lab, University Hospital of Larissa, Greece. [34]Laboratory of Molecular Medicine and Biotechnology, Don C. Gnocchi Foundation ONLUS, IRCCS S. Maria Nascente, Milan, Italy. [35]Department of Medical Sciences, Torino University, Turin, Italy. [36]Department of Health Sciences and Interdisciplinary Research Center of Autoimmune Diseases (IRCAD), University of Eastern Piedmont, Novara, Italy. [37]Centro Regionale Sclerosi Multipla, Neurologia B, AOUI Verona, Italy. [38]Fondazione IRCCS Cà Granda, Ospedale Maggiore Policlinico, Italy. [39]Medical Research Council Biostatistics Unit, Robinson Way, Cambridge CB2 0SR, UK. [40]MS Center, Department of Neuroscience, A.O. Città della Salute e della Scienza di Torino and University of Turin, Torino, Italy. [41]Laboratory of Human Genetics of Neurological complex disorder, Institute of Experimental Neurology (INSPE), Division of Neuroscience, San Raffaele Scientific Institute, Via Olgettina 58, 20132, Milan, Italy. [42]Department of Biomedical Sciences for Health, University of Milan, Milan, Italy. [43]University of Milan, Department of Health Sciences, San Paolo Hospital and Filarete Foundation, viale Ortles 22/4, 20139 Milan, Italy. [44]Department of Neurology, Institute of Experimental Neurology (INSPE), Division of Neuroscience, San Raffaele Scientific Institute, Via Olgettina 58, 20132, Milan, Italy. [45]Neurology Unit, Department of Pathophysiology and Transplantation, University of Milan, Dino Ferrari Center, Milan, Italy. [46]Fondazione IRCCS Ca' Granda, Ospedale Policlinico, Milan, Italy. [47]Fondazione IRCCS Casa Sollievo della Sofferenza, Unit of Neurology, San Giovanni Rotondo (FG), Italy. [48]Center for Experimental Neurological Therapies, Sant'Andrea Hospital, Department of Neurosciences, Mental Health and Sensory Organs, Sapienza University, Rome, Italy. [49]Istituto Neurologico Mediterraneo (INM) Neuromed, Pozzilli, Isernia, Italy. [50]Department of Neurology, Ospedale Maggiore, Novara, Italy. [51]Laboratory of Microbiology and Virology, University Vita-Salute San Raffaele, Hospital San Raffaele, Milan, Italy. [52]Department of Health Sciences and Interdisciplinary Research Center of Autoimmune Diseases (IRCAD), University of Eastern Piedmont, Novara, Italy. [53]Department of Neurology, Erasmus MC, Rotterdam, Netherlands. [54]Nuffield Department of Population Health, Big Data Institute, University of Oxford, Li Ka Shing Centre for Health Information and Discovery, Old Road Campus, Oxford OX3 7LF, UK. [55]Department of Neurology, Institute of Clinical Medicine, University of Oslo, Norway. [56]Department of Neurology, Oslo University Hospital, Oslo, Norway. [57]Institute of Basic Medical Sciences, University of Oslo, Oslo, Norway. [58]Servei de Neurologia-Neuroimmunologia, Centre d'Esclerosi Múltiple de Catalunya (Cemcat), Institut de Recerca Vall d'Hebron (VHIR), Hospital Universitari Vall d'Hebron, Spain. [59]Institute of Environmental Medicine, Karolinska Institutet, Stockholm, Sweden. [60]Department of Clinical Neuroscience, Karolinska Institutet, Stockholm, Sweden. [61]Translational Bioinformatics Unit, NavarraBiomed, Complejo Hospitalario de Navarra (CHN), Universidad Pública de Navarra (UPNA), IdiSNA, Pamplona, Navarra, Spain. [62]Mucosal and Salivary Biology Division, King's College, London Dental Institute, London, UK. [63]Neuroimmunology and MS Research (nims), Neurology Clinic, University Hospital Zurich, Frauenklinikstrasse 26, 8091 Zurich, Switzerland. [64]Department of Neuroimmunology and MS Research, Neurology Clinic, University Hospital Zürich, Frauenklinikstrasse 26, 8091 Zürich, Switzerland. [65]University of Cambridge, Department of Clinical Neurosciences, Addenbrooke's Hospital, BOX 165, Hills Road, Cambridge CB2 0QQ, UK. [66]Keele University Medical School, University Hospital of North Staffordshire, Stoke-on-Trent ST4 7NY, UK. [67]Department of Twin Research and Genetic Epidemiology, King's College London, London, SE1 7EH, UK. [68]NIHR Oxford Biomedical Research Centre, Diabetes and Metabolism Theme, OCDEM, Churchill Hospital, Oxford UK. [69]NIHR BioResource, Box 299,University of Cambridge and Cambridge University Hospitals NHS Foundation Trust Hills Road, Cambridge CB2 0QQ, UK. [70]Department of Neurology, Peterborough City Hospital, Edith Cavell Campus, Bretton Gate, Peterborough PE3 9GZ, UK. [71]Department of Neurology, Johns Hopkins University School of medicine, Baltimore MD. [72]Multiple sclerosis center, Department of neurology, School of medicine, Washington University St Louis, St Louis MO. [73]Center for Human Genetics Research, Vanderbilt University Medical Center, 525 Light Hall, 2215 Garland Avenue, Nashville, TN 37232, USA. [74]Multiple Sclerosis Division, Department of Neurology, University of Miami, Miller School of Medicine, Miami, FL 33136, USA. [75]MS Center of Northeastern NY 1205 Troy Schenectady Rd, Latham, NY 12110, USA. [76]Université de Nantes, INSERM, Centre de Recherche en Transplantation et Immunologie, UMR 1064, ATIP-Avenir, Equipe 5, Nantes, France. [77]Population and Quantitative Health Sciences, Department of Epidemiology and Biostatistics, Case Western Reserve University, 10900 Euclid Avenue, Cleveland, OH 44106-4945 USA. [78]Center for Applied Genomics, The Children's Hospital of Philadelphia, 3615 Civic Center Blvd., Philadelphia, PA 19104, USA. [79]Department of Pediatrics, The Perelman School of Medicine, University of Pennsylvania, Philadelphia PA, USA. [80]Department of Neurology, Brigham & Women's Hospital, Boston, 02115 MA, USA. [81]Departments of Neurology and Neurological Therapeutics, Neurological Institute, Graduate School of Medical Sciences, Kyushu University, 3-1-1 Maidashi, Higashi-ku, Fukuoka City, Fukuoka 812-8582 Japan. [82]The Elliot Lewis Center, 110 Cedar St, Wellesley MA, 02481, USA. [83]Broad Institute of Harvard University and MIT, Cambridge, 02142 MA, USA. [84]School of Mathematics and Statistics, University of Sydney, Sydney, NSW 2006, Australia. [85]Westmead Institute for Medical Research, University of Sydney, Westmead, NSW 2145, Australia. [86]Kaiser Permanente Division of Research, Oakland, CA, USA. [87]Ann Romney Center for Neurological Diseases, Department of Neurology, Brigham & Women's Hospital, Boston, 02115 MA, USA. [88]Departments of Neurology and Immunobiology, Yale University School of Medicine, New Haven, CT 06520, USA. [89]Westmead Millennium Institute, University of Sydney, New South Wales, Australia. [90]Department of Health Sciences and Interdisciplinary Research Center of Autoimmune Diseases (IRCAD), University of Eastern Piedmont, Novara, Italy. [91]Menzies Research Institute Tasmania, University of Tasmania, Australia. [92]UC Berkeley School of Public Health and Center for Computational Biology, USA. [93]Westmead Millennium Institute, University of Sydney, New South Wales, Australia. [94]Department of Neurology and Department of Immunology, Erasmus MC, Rotterdam, Netherlands. [95]UK Dementia Research Institute, University College London, Gower Street, London WC1E 6BT, UK.
*Present address: Center for Computational Medicine, Peter Gilgan Centre for Research and Learning, Hospital for Sick Children (SickKids), Toronto, ON M5G 0A4, Canada.
†Present address: Vertex Pharmaceuticals, 50 Northern Avenue, Boston, MA 02210, USA.

13 October 2018; accepted 6 August 2019
10.1126/science.aav7188

ANZJP

Australian & New Zealand Journal of Psychiatry
1–11
DOI: 10.1177/0004867418815976

Structural and metabolic cerebral alterations between elderly bipolar disorder and behavioural variant frontotemporal dementia: A combined MRI-PET study

Giuseppe Delvecchio[1], Gian Mario Mandolini[1,2], Andrea Arighi[1,2,3], Cecilia Prunas[1,2], Carlo Massimo Mauri[2], Anna M Pietroboni[1,2,3], Giorgio Marotta[2], Claudia Maria Cinnante[2], Fabio Maria Triulzi[1,2], Daniela Galimberti[1,2,3], Elio Scarpini[1,2,3], Alfredo Carlo Altamura[1,2] and Paolo Brambilla[1,4]

Abstract

Background: Elderly bipolar disorder (BD) and behavioural variant of frontotemporal dementia (bvFTD) may exhibit similar symptoms and both disorders are characterized by selective abnormalities in cortical and subcortical regions that are associated with cognitive and emotional impairments. We aimed to investigate common and distinct neural substrates of BD and bvFTD by coupling, for the first time, magnetic resonance imaging (MRI) and positron emission tomography (PET) techniques.

Methods: 3-Tesla MRI and 18 fluorodeoxyglucose–PET scans were acquired for 16 elderly BD patients, 23 bvFTD patients with mild cognitive impairments and 68 healthy controls (48 for PET and 20 for MRI analyses).

Results: BD and bvFTD patients exhibit a different localization of grey matter reductions in the lateral prefrontal cortex, with the first group showing grey matter decrease in the ventrolateral prefrontal cortex and the latter group showing grey matter reductions in the dorsolateral prefrontal cortex as well as unique grey matter and metabolic alterations within the orbitofrontal cortex. The bvFTD group also displayed unique volumetric shrinkage in regions within the temporo-parietal network together with greater metabolic impairments within the temporal cortex and more extensive volumetric and metabolic abnormalities within the limbic lobe. Finally, while the BD group showed greater grey matter volumes in caudate nucleus, bvFTD subjects displayed lower metabolism.

Conclusion: This MRI-PET study explored, for the first time to the best of our knowledge, structural and functional abnormalities in bvFTD and elderly BD patients, with the final aim of identifying the specific biological signature of these disorders, which might have important implications not only in prevention but also in differential diagnosis and treatment.

Keywords

Bipolar disorder, frontotemporal dementia, grey matter, positron emission tomography, magnetic resonance imaging

[1]Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy
[2]Department of Neurosciences and Mental Health, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
[3]'Dino Ferrari' Center, Milan, Italy
[4]Department of Psychiatry and Behavioural Sciences, UT Houston Medical School, Houston, TX, USA

Corresponding author:
Paolo Brambilla, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza 35, 20122 Milan, Italy.
Email: paolo.brambilla1@unimi.it

ORIGINAL ARTICLE

Value of insoluble PABPN1 accumulation in the diagnosis of oculopharyngeal muscular dystrophy

V. Galimberti[a], R. Tironi[a], A. Lerario[a], M. Scali[a], R. Del Bo[b,c], C. Rodolico[d], T. Brizzi[d,e], S. Gibertini[f], L. Maggi[f], M. Mora[f], A. Toscano[d], G. P. Comi[b,c], M. Sciacco[a], M. Moggio[a] and L. Peverelli[a]

[a]*Neuromuscular and Rare Disease Unit, Department of Neuroscience, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milano;* [b]*Dino Ferrari Centre, Neuroscience Section, Department of Pathophysiology and Transplantation (DEPT), University of Milan, Milano;* [c]*Neurology Unit, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, Milano;* [d]*Neurology and Neuromuscular Diseases Unit, Department of Clinical and Experimental Medicine, University of Messina, Messina;* [e]*DIBIMIS University of Palermo, Palermo; and* [f]*Neuromuscular Diseases and Neuroimmunology Unit, Fondazione IRCCS Istituto Neurologico Carlo Besta, Milano, Italy*

Keywords:
oculopharyngeal musclular dystrophy, PABPN1 accumulations, PABPN1 immunofluorescence, rimmed vacuoles, tubulofilamentous intranuclear inclusions

Received 4 August 2019
Accepted 18 November 2019

European Journal of Neurology 2019, **0:** 1–7

doi:10.1111/ene.14131

Background and purpose: The aim was to assess the value of insoluble PABPN1 muscle fibre nuclei accumulation in the diagnosis of atypical cases of oculopharyngeal muscular dystrophy (OPMD).

Methods: Muscle biopsies from a selected cohort of 423 adult patients from several Italian neuromuscular centres were analysed by immunofluorescence: 30 muscle biopsies of genetically proven OPMD, 30 biopsies from patients not affected by neuromuscular disorders, 220 from genetically undiagnosed patients presenting ptosis or swallowing disturbances, progressive lower proximal weakness and/or isolated rimmed vacuoles at muscle biopsy and 143 muscle biopsies of patients affected by other neuromuscular diseases.

Results: The detection of insoluble nuclear PABPN1 accumulation is rapid, sensitive (100%) and specific (96%). The revision of our cohort allowed us to discover 23 new OPMD cases out of 220 patients affected with nonspecific muscle diseases.

Conclusions: Oculopharyngeal muscular dystrophy is often misdiagnosed leading to diagnosis delay, causing waste of time and resources. A great number of these cases present symptoms and histological findings frequently overlapping with other muscle diseases, i.e. inclusion body myositis and progressive external ophthalmoplegia. PABPN1 nuclear accumulation is a reliable method for diagnostic purposes and it is safe and useful in helping pathologists and clinicians to direct genetic analysis in the case of suspected OPMD, even when clinical and histological clues are deceptive.

Introduction

Oculopharyngeal muscular dystrophy (OPMD) is a late-onset muscle disease, clinically characterized by ptosis, with or without ophthalmoparesis, dysphagia and proximal weakness. Inheritance can be autosomal dominant or recessive [1–3]. OPMD is caused by a GCN repeat expansion in the poly-A binding protein nuclear 1 gene (*PABPN1*) (14q11.2–q13), which leads to an expanded polyalanine tract in the N-terminal of the PABPN1 protein. Normal repeat size is (GCN)10, and (GCN)11–18 is considered pathological. Poly-A expansion in PABPN1 favours the accumulation of toxic, insoluble protein deposits in the nuclei of muscle cells [4]. The point mutation c.35G>C in *PABPN1* can also affect disease phenotype [1].

Oculopharyngeal muscular dystrophy is often overlooked and diagnosed after considerable delay, prolonging discomfort for patients and their relatives and wasting time and resources [6,7]. Histopathological findings of muscle biopsies from affected patients are overall nonspecific; however, rimmed vacuoles and oxidative stain alterations along with scattered

Correspondence: Lorenzo Peverelli, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Padiglione Ex convitto suore piano - 1, Via Francesco Sforza 35 20122, Milano, Italia (tel.: +39 0255036504; fax: +39 0255033827; e-mail: lorenzo.peverelli84@gmail.com).

Received: 10 September 2018 | Revised: 31 January 2019 | Accepted: 5 February 2019

DOI: 10.1002/humu.23729

RESEARCH ARTICLE

WILEY Human Mutation HGVS HUMAN GENOME VARIATION SOCIETY

Clinical-genetic features and peculiar muscle histopathology in infantile *DNM1L*-related mitochondrial epileptic encephalopathy

Daniela Verrigni[1*] | Michela Di Nottia[1*] | Anna Ardissone[2,3*] | Enrico Baruffini[4*] | Alessia Nasca[5] | Andrea Legati[5] | Emanuele Bellacchio[6] | Gigliola Fagiolari[7] | Diego Martinelli[8] | Lucia Fusco[9] | Domenica Battaglia[10] | Giulia Trani[1] | Gianmarco Versienti[5] | Silvia Marchet[5] | Alessandra Torraco[1] | Teresa Rizza[1] | Margherita Verardo[1] | Adele D'Amico[1] | Daria Diodato[1] | Isabella Moroni[2] | Costanza Lamperti[5] | Stefania Petrini[11] | Maurizio Moggio[7] | Paola Goffrini[4] | Daniele Ghezzi[5,12†] | Rosalba Carrozzo[1†] | Enrico Bertini[1†]

[1]Department of Neurosciences, Unit of Muscular and Neurodegenerative Disorders, Laboratory of Molecular Medicine, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy

[2]Department of Clinical Neurosciences, Child Neurology Unit, Fondazione IRCCS Istituto Neurologico Carlo Besta, Milan, Italy

[3]Department of Molecular and Translational Medicine DIMET, University of Milan-Bicocca, Milan, Italy

[4]Department of Chemistry, Life Sciences and Environmental Sustainability, University of Parma, Parma, Italy

[5]Department of Molecular Neurogenetics, Unit of Medical Genetics and Neurogenetics, Fondazione IRCCS Istituto Neurologico Carlo Besta, Milan, Italy

[6]Genetics and Rare Diseases, Research Division, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy

[7]Dino Ferrari Centre, Unit of Neuromuscular and Rare Disorders, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Università of Milano, Milan, Italy

[8]Division of Metabolism, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy

[9]Neurophysiology Unit, Department of Neuroscience, Bambino Gesu' Children's Hospital, Rome, Italy

[10]Department of Child Neurology and Psychiatry, Catholic University, Rome, Italy

[11]Scientific Direction, Research Laboratories, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy

[12]Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy

Correspondence
Enrico Bertini, Unit of Muscular and Neurodegenerative Disorders, Laboratory of Molecular Medicine, Bambino Gesù Children's Hospital, IRCCS, Viale di San Paolo 15, 00146 Rome, Italy.
Email: bertini@opbg.net

Funding information
Ministero dell~Istruzione, dell~Università e della Ricerca, Grant/Award Number: FIR2013

Abstract

Mitochondria are highly dynamic organelles, undergoing continuous fission and fusion. The *DNM1L* (dynamin-1 like) gene encodes for the DRP1 protein, an evolutionary conserved member of the dynamin family, responsible for fission of mitochondria, and having a role in the division of peroxisomes, as well. DRP1 impairment is implicated in several neurological disorders and associated with either de novo dominant or compound heterozygous mutations. In five patients presenting

*Daniela Verrigni, Michela Di Nottia, Anna Ardissone, and Enrico Baruffini contributed equally to this work.

†Daniele Ghezzi, Rosalba Carrozzo, and Enrico Bertini contributed equally as senior authors.

Acta Myologica • 2018; XXXVII: p. 257-262

Immune-mediated necrotizing myopathy due to statins exposure

Luisa Villa[1 2], Alberto Lerario[1], Sonia Calloni[3], Lorenzo Peverelli[1], Caterina Matinato[4], Federica de Liso[4], Ferruccio Ceriotti[4], Roberto Tironi[1], Monica Sciacco[1], Maurizio Moggio[1], Fabio Triulzi[3] and Claudia Cinnante[3]

[1] Neuromuscular and Rare Disease Unit, Department of Neuroscience, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Italy; [2] Neuromuscular Disease Centre, Department of Clinical Neurosciences, University Hospital of Nice (CHU), France; [3] Neuroradiology Unit, Fondazione IRCCS Ca Granda Ospedale Maggiore Policlinico, Milan, Italy; [4] Laboratorio analisi, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

Statin-induced necrotizing autoimmune myopathy (IMNM) is an autoimmune disorder induced by anti-3-hydroxy-3-methylglutaryl-coenzyme-A reductase (anti-HMGCR) antibodies. We performed a retrospective clinical, histological, and radiological evaluation of 5 patients with a 3-year therapeutic follow-up. All patients used statins and then experienced proximal weakness that persisted after drug cessation. Muscle biopsies revealed a primary necrotizing myopathy without inflammatory infiltrates. All patients required immunomodulant combination therapy to achieve clinical remission. Magnetic resonance imaging (MRI) showed the presence of edema in the medial gastrocnemius, posterior and central loggia of the thigh, posterior loggia of the arm, and the infraspinatus and subscapularis muscles, as well as extensive inflammation of the subcutaneous tissues and muscolaris fasciae. Serum analysis, muscle biopsy, and MRI are fundamental for IMNM diagnosis and follow-up. The growing use of statins in the general population raises the importance of acquaintance with this disease in clinical practice.

Key words: HMGCR autoantibodies, muscular MRI, necrotizing myopathy

Introduction

Inflammatory myopathies constitute a heterogeneous group of disorders targeting skeletal muscle. Different inflammatory myopathies vary with regards to prognosis and response to pharmacological therapy. Immune-mediated necrotizing myopathy (IMNM) is a recently recognized category of idiopathic inflammatory myopathy. The autoimmune nature of IMNM is suggested by its frequent association with two specific autoantibodies: 3-hydroxy-3-methylglutaryl-CoA reductase (HMGCR) and signal recognition particle (SRP) (1). Among patients using statins, the estimated IMNM incidence rate is 2-3 per 100,000 patients, with increased risk among patients over 50 years of age (2, 3).

Histological characteristics of IMNM include the presence of necrotic fibers without inflammatory cell infiltrates. The underlying pathogenesis remains unclear, but statins appear to play a major role. Statins can trigger the expression of anti-HMGCR antibodies. This induces muscle synthesis of HMGCR enzyme, which is normally poorly expressed in mature muscle cells, potentially maintaining inflammatory activity even after statin discontinuation (4-6). First-line treatment of IMNM involves steroids, which is generally effective although steroid treatment usually must be administered in combination with other immunosuppressive agents (9, 10).

Over the last decade, muscle magnetic resonance imaging (MRI) has become a very useful tool in the diagnosis and follow-up of patients with myopathies. Muscle MRI provides information regarding skeletal muscle structure and function, such as the presence of edema and/or fatty infiltration, and it is a good technique for monitoring disease progression (7). To date, only one study has analyzed the muscle involvement pattern in patients with IMNM, reporting widespread muscle involvement and a trend towards atrophy and fatty replacement (8). The predominantly involved muscles are the lateral obturators, glutei, and the thigh medial and posterior compartment (8).

Address for correspondence: Alberto Lerario, Neuromuscular and Rare Disease Unit, Department of Neuroscience, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, University of Milan, via F. Sforza 35, 20122 Milan, Italy. E-mail: alberto.lerario@policlinico.mi.it

observed in 3 of 4 patients. Post-treatment MRI revealed complete resolution of edema. The grades of fatty replacement remained largely unchanged.

Discussion

Statin use has consistently increased in the last twenty years and has led to more frequent toxic neuromuscular complications, usually self-limiting after drug discontinuation. Quite often, however, statin use induces an autoimmune reaction and causes the development of an aggressive IMNM. The disease is quite rare and, also due to the lack of a validated commercial diagnostic kit, often still under-diagnosed.

In our study all patients had taken atorvastatin. However, due to the small number of examined patients, further studies are certainly needed to validate this association. The 5 patients affected with statin-induced IMNM with serum positivity for anti-HMGCR antibodies and typical pattern of severe necrotizing muscular biopsy, showing acute weakness of trunk flexor and limb girdle muscles, whereas the bulbar muscles were generally spared. Axial involvement is not common among other types of inflammatory myopathies (15). Indeed, in statin-induced myopathies, MRI imaging reportedly shows involvement of the dorsal muscle groups of both the thighs (8, 16). Compared to others toxic and drug-induced myopathies, MRI imaging in our IMNM patients revealed more extensive edema and a trend towards fatty muscle replacement. These findings are in agreement with the recent literature (8). Moreover, extending the study to leg, arm and shoulder girdle, we found a new pattern recognition involving also the medial gastrocnemius and, at the arm level, triceps and deltoid followed by infraspinatus and subscapularis. We also observed inflammation of the subcutaneous tissues and of muscolaris fasciae of both arms and legs, which has not been previously reported to our knowledge (Fig. 2). These findings suggest that patients affected with IMNM may exhibit a wider systemic inflammatory response that is not limited to skeletal muscles.

These new MRI findings allow to improve the differential diagnosis between IMNM myopathy and other inflammatory myopathies and to distinguish IMNM from the toxic myopathy related to statin intake. This distinction is important because toxic myopathy usually improves and then resolves following interruption of statin intake; conversely, IMNM myopathy progressively worsens even after statin suspension and causes a very severe, sometimes hardly or non-reversible, damage to the muscles. Ultimately, muscle MRI may be a useful tool to monitor the evolution of muscle disease over time, but it can be used also as a fist-line screening to identify the most affected muscle and therefore increase the diagnostic accuracy of the skeletal muscle biopsy.

Appropriate therapeutic control is tricky in this type of inflammatory myopathy. Each patient required several immunosuppressive treatments before achieving clinical control. Indeed, all patients were initially treated with IV steroids, followed by high-dose oral steroid therapy, with concomitant superimposition of additional immunosuppressive drugs. Moreover all patients achieved normalization of CK levels and improved muscle strength, albeit with different drug associations and time intervals until response. In two patients, tapering attempts were followed by immediate increase of CK levels that required drug restoration.

In our present study, we aimed to provide a systematic comprehension of this currently under-diagnosed disease in terms of both diagnostic tools and therapeutic options, and to help define the MRI pattern for IMNM recognition to improve its diagnosis.

Acknowledgements

The authors wish to thank the “Associazione Amici del Centro Dino Ferrari, University of Milan”; The Biobank of Skeletal Muscle, Peripheral Nerve, DNA and Cell Cultures, which is a member of the Telethon Network of Genetic Biobanks, funded by Telethon Italy; and the Eurobiobank Network. Representatives of these networks gave their consent to be named.

References

1. Arouche-Delaperche L, Allenbach Y, Amelin D, et al. Pathogenic role of anti-signal recognition protein and anti-3-Hydroxy-3-methylglutaryl-CoA reductase antibodies in necrotizing myopathies: myofiber atrophy and impairment of muscle regeneration in necrotizing autoimmune myopathies. Ann Neurol 2017;81:538-48.
2. Floyd JS, Brody JA, Tiniakou E, et al. Absence of anti-HMG-CoA reductase autoantibodies in severe self-limited statin-related myopathy. Muscle Nerve 2016;54:142-4.
3. Mammen AL, Chung T, Christopher-Stine L, et al. Autoantibodies against 3-hydroxy-3-methylglutaryl-coenzyme A reductase in patients with statin-associated autoimmune myopathy. Arthritis Rheum 2011;63:713-21.
4. Albayda J, Mammen AL. Is statin-induced myositis part of the polymyositis disease spectrum? Curr Rheumatol Rep 2014;16:433.
5. Christopher-Stine L. Statin myopathy: an update. Curr Opin Rheumatol 2006;18:647-53.
6. Christopher-Stine L, Casciola-Rosen LA, Hong G, et al. A novel autoantibody recognizing 200-kd and 100-kd proteins is associated with an immune-mediated necrotizing myopathy. Arthritis Rheum 2010;62:2757-66.

frontiers in Psychology

ORIGINAL RESEARCH
published: 11 October 2019
doi: 10.3389/fpsyg.2019.02265

A Novel Approach for Investigating Parkinson's Disease Personality and Its Association With Clinical and Psychological Aspects

Laura Carelli[1], Federica Solca[2], Silvia Torre[1], Jacopo Pasquini[1], Claudia Morelli[1], Rita Pezzati[3,4], Francesca Mancini[1], Andrea Ciammola[1], Vincenzo Silani[1,2†] and Barbara Poletti[1†]*

[1] Department of Neurology and Laboratory of Neuroscience, Italian Auxological Institute (IRCCS), Milan, Italy, [2] Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, University of Milan, Milan, Italy, [3] Department of Business Economics, Health and Social Care, University of Applied Sciences and Arts of Southern Switzerland, Manno, Switzerland, [4] Centro Terapia Cognitiva, Como, Italy

OPEN ACCESS

Edited by:
Gianluca Castelnuovo,
Catholic University of the Sacred Heart, Italy

Reviewed by:
Barbara Colombo,
Champlain College, United States
Agostino Brugnera,
University of Bergamo, Italy
Cristina Zarbo,
University of Bergamo, Italy

***Correspondence:**
Laura Carelli
l.carelli@auxologico.it

† These authors have contributed equally to this work

Specialty section:
This article was submitted to Psychology for Clinical Settings, a section of the journal Frontiers in Psychology

Received: 06 May 2019
Accepted: 23 September 2019
Published: 11 October 2019

Citation:
Carelli L, Solca F, Torre S, Pasquini J, Morelli C, Pezzati R, Mancini F, Ciammola A, Silani V and Poletti B (2019) A Novel Approach for Investigating Parkinson's Disease Personality and Its Association With Clinical and Psychological Aspects. Front. Psychol. 10:2265. doi: 10.3389/fpsyg.2019.02265

Objective: A complex relationship between neuropsychiatric symptoms, personality traits and neurochemical changes in patients with Parkinson's disease (PD) has been highlighted in the past several decades. In particular, a specific Parkinson personality with obsessive traits has been described. However, despite the great amount of anecdotal evidence, this aspect, together with its neurobiological, psychological and clinical correlates, are still not clearly defined. Therefore, we performed a case-control study in order to investigate the presence and rate of obsessive personality traits in PD patients within the theoretical framework of cognitive-constructivist model. Moreover, the relationship between PD personality and clinical, psychological and quality of life (QoL) aspects in PD were investigated.

Methods: Fifty-one non-demented patients with probable or possible PD (not demented) were recruited at the inpatient-outpatient San Luca Hospital, IRCCS Istituto Auxologico Italiano. Control group was composed by forty-eight age- and education-matched healthy volunteers. Patients underwent a neurological investigation including Unified PD Rating Scale (UPDRS), Modified Hoehn and Yahr and Schwab and England staging scales. The following psychological questionnaires were administered to the overall sample: Personal Meaning Questionnaire (PMQ), State-Trait Anxiety Inventory-Form Y (STAI-Y), Beck Depression Inventory (BDI), Symptom Check List-90 (SCL-90), Short-Form Health Survey-36 (SF-36).

Results: No significant differences in personality styles were observed in PD patients and controls, with a prevalence of phobic personal meaning organization (PMO) in both groups. However, PD patients showed more anxiety, depression and obsessive-compulsive (OC) symptoms than controls at the psychological questionnaires, as well as poorer QoL levels. The intensity of personality traits, and in particular for the obsessive personality style, were negatively associated with QoL and positively with disease severity. No significant relationships were observed between personality and other clinical aspects, such as side of onset and disease duration.

Received: 25 March 2019 | Revised: 6 June 2019 | Accepted: 1 August 2019

DOI: 10.1002/ccd.28445

ORIGINAL STUDIES

WILEY

Carotid artery stenting is safe and effective for symptomatic patients with acute coronary syndrome

Renato Casana[1,2] | Valerio Stefano Tolva[3] | Andrea Odero Jr[1] | Chiara Malloggi[2] | Vincenzo Silani[4,5] | Gianfranco Parati[6,7]

[1]Istituto Auxologico Italiano, IRCCS, Department of Surgery, Milan, Italy

[2]Istituto Auxologico Italiano, IRCCS, Laboratory of Research in Vascular Surgery, Milan, Italy

[3]Department of Vascular and Endovascular Surgery, Policlinico Di Monza, Monza, Italy

[4]Istituto Auxologico Italiano, IRCCS, Department of Neurology-Stroke Unit and Laboratory of Neuroscience, Ospedale San Luca, Milan, Italy

[5]Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, Università degli Studi di Milano, Milan, Italy

[6]Istituto Auxologico Italiano, IRCCS, Department of Cardiovascular, Neural and Metabolic Sciences, San Luca Hospital, Milan, Italy

[7]Department of Medicine and Surgery, Università di Milano-Bicocca, Monza, Italy

Correspondence
Renato Casana, Istituto Auxologico Italiano, IRCCS, Department of surgery Via Mercalli, 30, 20122, Milano, Italy.
Email: r.casana@auxologico.it

Funding information
CARDIMAUX project

Abstract

Background: Patients with symptomatic carotid stenosis recently treated with percutaneous transluminal coronary angioplasty (PTCA) for acute coronary syndrome (ACS) are always classified as at high risk for surgery, given that they are required uninterrupted dual antiplatelet therapy. In this regard, carotid artery stenting (CAS) may represent a valid alternative.

Objective: The purpose of this study is to overview CAS outcomes in symptomatic patients with and without ACS.

Methods: One hundred fifty-one consecutive symptomatic patients who underwent CAS between 2010 and 2017 in a single institution were included in this study, of which 66 (43.7%) were identified as having ACS. All patients were followed-up with carotid duplex ultrasound scan and a neurological assessment of symptoms status at 30-day postprocedure and at 3, 6, and 12 months, with annual follow-up after that for 3 years.

Results: Among symptomatic ACS patients, common risks factors were active smoking, metabolic syndrome, diabetes, and hypertension. In the short-term follow-up, no significant differences were observed among rates of death, stroke, myocardial infarction (MI), and restenosis, between patients with and without ACS. Mean clinical follow-up was 28.2 (12.3) months. In the long-term follow-up, higher rates of death and MI were recorded in patients with ACS (death: 11.4% vs. 5.4%, $p = .04$; MI: 11.4% vs. 3.6%, $p = .02$), owing to the complexity of these patients.

Conclusions: This single-center study suggested that CAS is a safe and effective treatment for patients with symptomatic carotid artery stenosis, who recently underwent PTCA for ACS, requiring uninterrupted dual antiplatelet therapy.

KEYWORDS

acute myocardial infarction/STEMI, carotid artery disease, coronary artery disease, percutaneous coronary intervention, stenting technique

1 | INTRODUCTION

Acute coronary syndrome (ACS) refers to a spectrum of conditions compatible with acute myocardial ischemia and/or infarction (MI) that are usually due to an abrupt reduction in coronary blood flow.[1] A key branch point is the rise and/or fall of cardiac troponin values (cTn) with at least one value above the 99th percentile upper reference limit (URL) that identifies the myocardial injury. Criteria for acute myocardial infarction regards clinical evidence of (at least one): symptoms of myocardial ischemia; new ischemic electrocardiogram (ECG)

Original Article

Vascular

Vascular
0(0) 1–9

DOI: 10.1177/1708538119836312
journals.sagepub.com/home/vas
SAGE

Three-year outcomes after carotid artery revascularization: Gender-related differences

Renato Casana[1,2], Chiara Malloggi[2], Valerio Stefano Tolva[3], Andrea Odero Jr[1], Richard Bulbulia[4,5], Alison Halliday[6], Vincenzo Silani[7,8] and Gianfranco Parati[9,10]

Abstract

Objectives: Carotid artery stenosis is thought to cause up to 10% of ischemic strokes. Historically, carotid artery endarterectomy has shown a higher risk of perioperative adverse events for women. More recent trials reported conflicting results regarding the benefit of carotid artery endarterectomy and carotid artery stenting for men and women. The aim of the present retrospective study was to investigate the influence of gender on the short- (30 days) and long-term (3 years) outcomes of carotid artery endarterectomy and carotid artery stenting in a single centre.

Methods: From 2010 to 2017, 912 consecutive symptomatic and asymptomatic patients who underwent carotid artery endarterectomy (389, 42.7%) or carotid artery stenting (523, 57.3%) in a single institution had been evaluated to determine the influence of sex (540 men, 59.2%, vs. 372 women, 40.8%) on the outcomes after both revascularization procedures during three years of follow-up. The primary endpoint was the incidence of death, stroke, myocardial infarction, and restenosis in the short-term follow-up. The secondary endpoint was the incidence of death, stroke, myocardial infarction, and restenosis in the long-term follow-up.

Results: Mean clinical follow-up was 21.1 (16.1) months. Women had internal and common carotid artery diameters significantly smaller with respect to men. For peri-procedural outcomes, women undergoing carotid artery stenting had a higher risk of moderate (50–70%) restenosis (6 women, 2.9%, vs. 3 men, 1.0%). For long-term outcomes, women undergoing carotid artery endarterectomy had a higher rate of moderate restenosis (16 women, 16.3%, vs. 11 men, 7.6%). No significant differences in long-term outcomes were observed between men and women undergoing carotid artery stenting, even after stratification for baseline risk factors.

Conclusions: Contrary to previous reports, from this single-centre study, long-term risk of events seems to be higher in women who underwent carotid artery endarterectomy than in those who underwent carotid artery stenting, while fewer differences were observed in men.

Keywords

Carotid endarterectomy, complications, gender, mortality, restenosis, stenting

[1]Istituto Auxologico Italiano, IRCCS, Centro Chirurgia Vascolare, Auxologico Capitanio, Milano, Italy
[2]Istituto Auxologico Italiano, IRCCS, Laboratorio Sperimentale di Ricerche di Chirurgia Vascolare, Milano, Italy
[3]Dipartimento di Chirurgia Vascolare, Policlinico Di Monza, Monza, Italy
[4]Clinical Trial Service Unit, Nuffield Department of Population Health, University of Oxford, Oxford, UK
[5]MRC Population Health Research Unit, Nuffield Department of Population Health, University of Oxford, Oxford, UK
[6]Nuffield Department of Surgical Sciences, University of Oxford, Oxford, UK
[7]Istituto Auxologico Italiano, IRCCS, Dipartimento di Neurologia e Stroke Unit e Laboratorio di Ricerche di Neuroscienze, Ospedale San Luca, Milano, Italy
[8]Dipartimento di Fisiopatologia Medico-Chirurgica e dei Trapianti, Centro 'Dino Ferrari', Università degli Studi di Milano, Milano, Italy
[9]Istituto Auxologico Italiano, IRCCS, Dipartimento di Scienze Cardiovascolari, Neurologiche, Metaboliche, Ospedale San Luca, Milano, Italy
[10]Dipartimento di Medicina e Chirurgia, Università di Milano-Bicocca, Milano, Italy

Corresponding author:
Renato Casana, Istituto Auxologico Italiano, IRCCS Centro Chirurgia Vascolare, Auxologico Capitanio, Via Mercalli, 30, 20122 Milano, Italy.
Email: r.casana@auxologico.it

frontiers in Neurology

CASE REPORT
published: 06 November 2019
doi: 10.3389/fneur.2019.01124

A Novel Mutation of *GFAP* Causing Adult-Onset Alexander Disease

Andrea Ciammola[1†], Davide Sangalli[1†], Jenny Sassone[2,3*], Barbara Poletti[1], Laura Carelli[1], Paolo Banfi[4], Gabriele Pappacoda[4], Isabella Ceccherini[5], Alice Grossi[5], Luca Maderna[1], Monica Pingue[1,2], Floriano Girotti[1‡] and Vincenzo Silani[1,6‡]*

[1] Department of Neurology and Laboratory of Neuroscience, Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS), Istituto Auxologico Italiano, Milan, Italy, [2] Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS), San Raffaele Scientific Institute, Milan, Italy, [3] Vita-Salute San Raffaele University, Milan, Italy, [4] Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS), Fondazione Don Carlo Gnocchi, Milan, Italy, [5] Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS), Istituto Giannina Gaslini, Genoa, Italy, [6] Department of Pathophysiology and Transplantation, Dino Ferrari Center, University of Milan, Milan, Italy

Alexander disease (AxD) is a rare, autosomal dominant neurological disorder. Three clinical subtypes are distinguished based on age at onset: infantile (0–2 years), juvenile (2–13 years), and adult (>13 years). The three forms differ in symptoms and prognosis. Rapid neurological decline with a fatal course characterizes the early-onset forms, while symptoms are milder and survival is longer in the adult forms. Currently, the sole known cause of AxD is mutations in the *GFAP* gene, which encodes a type III intermediate filament protein that is predominantly expressed in astrocytes. A wide spectrum of *GFAP* mutations comprising point mutations, small insertions, and deletions is associated with the disease. The genotype-phenotype correlation remains unclear. The considerable heterogeneity in severity of disease among individuals carrying identical mutations suggests that other genetic or environmental factors probably modify age at onset or progression of AxD. Describing new cases is therefore important for establishing reliable genotype-phenotype correlations and revealing environmental factors able to modify age at onset or progression of AxD. We report the case of a 54-year-old Caucasian woman, previously diagnosed with ovarian cancer and treated with surgery and chemotherapy, who developed dysarthria, ataxia, and spastic tetraparesis involving mainly the left side. Cerebral and spinal magnetic resonance imaging (MRI) revealed a peculiar tadpole-like atrophy of the brainstem. Genetic analysis of the *GFAP* gene detected a heterozygous mutation in exon 1 (c.219G>C), resulting in an amino acid exchange from methionine to isoleucine at codon 73 (p.M73I). The expression of this mutant *in vitro* affected the formation of the intermediate filament network. Thus, we have identified a new *GFAP* mutation in a patient with an adult form of AxD.

Keywords: Alexander disease, GFAP-glial fibrillary acidic protein, leukodystrophy, gene mutation, adult onset

OPEN ACCESS

Edited by:
Clara van Karnebeek, University Medical Center Amsterdam, Netherlands

Reviewed by:
Liena Elbaghir Omer Elsayed, University of Khartoum, Sudan
Kenya Nishioka, Juntendo University, Japan

***Correspondence:**
Andrea Ciammola
a.ciammola@auxologico.it
Jenny Sassone
sassone.jenny@hsr.it

†Co-first authors

‡Co-last authors

Specialty section:
This article was submitted to Neurogenetics, a section of the journal Frontiers in Neurology

Received: *18 June 2019*
Accepted: *08 October 2019*
Published: *06 November 2019*

Citation:
Ciammola A, Sangalli D, Sassone J, Poletti B, Carelli L, Banfi P, Pappacoda G, Ceccherini I, Grossi A, Maderna L, Pingue M, Girotti F and Silani V (2019) A Novel Mutation of GFAP Causing Adult-Onset Alexander Disease. Front. Neurol. 10:1124. doi: 10.3389/fneur.2019.01124

BACKGROUND

Alexander disease (AxD) is an autosomal dominant neurological disorder (OMIM #203450) caused by mutations in the glial fibrillary acidic protein (*GFAP*) gene located on chromosome 17q21.31. AxD is a leukodystrophy characterized by the progressive accumulation in astrocytes of GFAP aggregates, called Rosenthal fibers, which are spread over the subpial, perivascular, and subependymal regions of the cortex and white matter. These inclusions impair cytoskeleton formation and astrocyte survival and functioning, ultimately resulting in demyelination (1).

BBA - Gene Regulatory Mechanisms 1862 (2019) 194413

Contents lists available at ScienceDirect

BBA - Gene Regulatory Mechanisms

journal homepage: www.elsevier.com/locate/bbagrm

TDP-43 and NOVA-1 RNA-binding proteins as competitive splicing regulators of the schizophrenia-associated *TNIK* gene

Valentina Gumina[a,b], Claudia Colombrita[a], Claudia Fallini[c], Patrizia Bossolasco[a], Anna Maria Maraschi[a], John E. Landers[c], Vincenzo Silani[a,b,d], Antonia Ratti[a,e,*]

[a] Istituto Auxologico Italiano, IRCCS, Department of Neurology-Stroke Unit and Laboratory of Neuroscience, Via Zucchi 18, 20095, Cusano Milanino, Milan, Italy
[b] Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, Università degli Studi di Milano, Via F. Sforza 35, 20122 Milan, Italy
[c] Department of Neurology, University of Massachusetts Medical School, 368 Plantation Street, ASC 6-1053, Worcester, MA 01605, USA
[d] "Aldo Ravelli" Center for Neurotechnology and Experimental Brain Therapeutics, Università degli Studi di Milano, Via A. di Rudinì 8, 20142 Milan, Italy
[e] Department of Medical Biotechnology and Translational Medicine, Università degli Studi di Milano, Via Fratelli Cervi 93, 20090, Segrate, Milan, Italy

ARTICLE INFO

Keywords:
TNIK
RNA-binding protein
Splicing
TDP-43
NOVA-1
Schizophrenia

ABSTRACT

The RNA-binding protein TDP-43, associated to amyotrophic lateral sclerosis and frontotemporal dementia, regulates the alternative splicing of several genes, including the skipping of *TNIK* exon 15. *TNIK*, a genetic risk factor for schizophrenia and causative for intellectual disability, encodes for a Ser/Thr kinase regulating negatively F-actin dynamics.

Here we show that in the human adult nervous system TNIK exon 15 is mostly included compared to the other tissues and that, during neuronal differentiation of human induced pluripotent stem cells and of human neuroblastoma cells, *TNIK* exon 15 inclusion increases independently of TDP-43 protein content. By studying the possible molecular interplay of TDP-43 with brain-specific splicing factors, we found that the neuronal NOVA-1 protein competitively inhibits both TDP-43 and hnRNPA2/B1 skipping activity on *TNIK* by means of a RNA-dependent interaction and that this competitive mechanism is common to other TDP-43 RNA targets. We also show that the TNIK protein isoforms including/excluding exon 15 differently regulate cell spreading in non-neuronal cells and neuritogenesis in primary cortical neurons.

Our data suggest a complex regulation between the ubiquitous TDP-43 and the neuron-specific NOVA-1 splicing factors in the brain that may help better understand the pathobiology of both neurodegenerative diseases and schizophrenia.

1. Introduction

Alternative splicing is a very active process in the mammalian brain contributing to the high complexity of the nervous system proteome [1]. During neurodevelopment, a tight spatio-temporal regulation of alternative splicing is crucial for the proper development of the different brain areas, depending on the combinatorial control of both ubiquitous and neuron-specific splicing factors [2,3]. In developing neurons, NOVA protein family members regulate the alternative splicing of genes important for axon guidance, synaptic architecture and activity [4,5]. These RNA-binding proteins (RBP) are predominantly expressed in neuronal cells, with NOVA-1 particularly enriched in the hindbrain and ventral spinal cord and NOVA-2 in the hippocampus and neocortex [6].

Among the ubiquitous splicing factors, TDP-43 is of particular interest as it is associated to the neurodegenerative diseases amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD), where it forms pathological aggregates in the cytoplasm of affected neuronal cells with the concomitant reduction of its nuclear localization [7,8] and splicing activity [9]. In the nucleus TDP-43 binds to a wide array of genes identified by high throughput approaches such as CLIP and mainly regulates their splicing [10]. TDP-43 was shown to bind to *TNIK* intron 15 and to promote the skipping of the alternative exon 15 [11]. Upon TDP-43 knock-down in human neuroblastoma cells to mimic a

Abbreviations: RBP, RNA-binding proteins; ALS, amyotrophic lateral sclerosis; FTD, frontotemporal dementia; GCK, germinal center kinase; CNH, citron homology; iPSC, induced pluripotent stem cell; IPSC-N, iPSC-derived neurons; NSC, neural stem cells; EB, embryoid bodies; UV-CLIP, UV Cross-linking and immunoprecipitation

* Corresponding author at: Istituto Auxologico Italiano, IRCCS, Department of Neurology-Stroke Unit and Laboratory of Neuroscience, Via Zucchi 18, 20095, Cusano Milanino, Milan, Italy.

E-mail addresses: Claudia.Fallini@umassmed.edu (C. Fallini), a.maraschi@auxologico.it (A.M. Maraschi), John.Landers@umassmed.edu (J.E. Landers), vincenzo@silani.com (V. Silani), antonia.ratti@unimi.it (A. Ratti).

https://doi.org/10.1016/j.bbagrm.2019.194413
Received 20 March 2019; Received in revised form 31 July 2019; Accepted 31 July 2019
Available online 02 August 2019
1874-9399/

International Journal of
Molecular Sciences

MDPI

Article

Inter-Species Differences in Regulation of the Progranulin–Sortilin Axis in TDP-43 Cell Models of Neurodegeneration

Valentina Gumina [1], Elisa Onesto [1,†], Claudia Colombrita [1], AnnaMaria Maraschi [1], Vincenzo Silani [1,2,3] and Antonia Ratti [1,4,*]

1 Istituto Auxologico Italiano, IRCCS, Department of Neurology-Stroke Unit and Laboratory of Neuroscience, Via Zucchi 18, 20095 Cusano Milanino, Milan, Italy; valegumina@gmail.com (V.G.); elisa.onesto.eo@axxam.com (E.O.); claudiacolombrita@hotmail.com (C.C.); a.maraschi@auxologico.it (A.M.); vincenzo@silani.com (V.S.)
2 Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, Università degli Studi di Milano, Via F. Sforza 35, 20122 Milan, Italy
3 "Aldo Ravelli" Center for Neurotechnology and Experimental Brain Therapeutics, Università degli Studi di Milano, Via A. di Rudinì 8, 20142 Milan, Italy
4 Department of Medical Biotechnology and Translational Medicine, Università degli Studi di Milano, Via Fratelli Cervi 93, 20090 Segrate, Milan, Italy
* Correspondence: antonia.ratti@unimi.it; Tel.: +39-02-619113045
† Current adress: Axxam Spa OpenZone, via A. Meucci 3, 20091 Bresso, Milan, Italy.

Received: 22 October 2019; Accepted: 20 November 2019; Published: 22 November 2019

Abstract: Cytoplasmic aggregates and nuclear depletion of the ubiquitous RNA-binding protein TDP-43 have been described in the autoptic brain tissues of amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTLD) patients and both TDP-43 loss-of-function and gain-of-function mechanisms seem to contribute to the neurodegenerative process. Among the wide array of RNA targets, TDP-43 regulates progranulin (*GRN*) mRNA stability and sortilin (*SORT1*) splicing. Progranulin is a secreted neurotrophic and neuro-immunomodulatory factor whose endocytosis and delivery to the lysosomes are regulated by the neuronal receptor sortilin. Moreover, *GRN* loss-of-function mutations are causative of a subset of FTLD cases showing TDP-43 pathological aggregates. Here we show that TDP-43 loss-of-function differently affects the progranulin–sortilin axis in murine and human neuronal cell models. We demonstrated that although TDP-43 binding to *GRN* mRNA occurs similarly in human and murine cells, upon TDP-43 depletion, a different control of sortilin splicing and protein content may determine changes in extracellular progranulin uptake that account for increased or unchanged secreted protein in murine and human cells, respectively. As targeting the progranulin–sortilin axis has been proposed as a therapeutic approach for *GRN*-FTLD patients, the inter-species differences in TDP-43-mediated regulation of this pathway must be considered when translating studies from animal models to patients.

Keywords: progranulin; sortilin; TDP-43; ALS; FTLD

1. Introduction

The ubiquitous TDP-43 RNA-binding protein (RBP) is the major component of the pathological aggregates described in the autoptic brain tissues of the majority of amyotrophic lateral sclerosis (ALS) patients and of a subset of frontotemporal lobar dementia (FTLD) cases [1,2]. Pathological TDP-43 aggregates are prevalently localized in the cytoplasm of affected neurons, which typically also show a concomitant depletion of TDP-43 protein from the nucleus [1,2]. The pathobiology associated to

BMJ Open

Proteostasis and ALS: protocol for a phase II, randomised, double-blind, placebo-controlled, multicentre clinical trial for colchicine in ALS (Co-ALS)

Jessica Mandrioli,[1] Valeria Crippa,[2] Cristina Cereda,[3] Valentina Bonetto,[4] Elisabetta Zucchi,[5] Annalisa Gessani,[1] Mauro Ceroni,[6,7] Adriano Chio,[8] Roberto D'Amico,[9] Maria Rosaria Monsurrò,[10] Nilo Riva,[11] Mario Sabatelli,[12,13,14] Vincenzo Silani,[15,16] Isabella Laura Simone,[17] Gianni Sorarù,[18] Alessandro Provenzani,[19] Vito Giuseppe D'Agostino,[19] Serena Carra,[20] Angelo Poletti[2]

To cite: Mandrioli J, Crippa V, Cereda C, *et al*. Proteostasis and ALS: protocol for a phase II, randomised, double-blind, placebo-controlled, multicentre clinical trial for colchicine in ALS (Co-ALS). *BMJ Open* 2019;**9**:e028486. doi:10.1136/bmjopen-2018-028486

► Prepublication history for this paper is available online. To view these files, please visit the journal online (http://dx.doi.org/10.1136/bmjopen-2018-028486).

SC and AP contributed equally.

Received 12 December 2018
Revised 25 February 2019
Accepted 10 April 2019

For numbered affiliations see end of article.

Correspondence to
Dr Jessica Mandrioli;
mandrioli.jessica@aou.mo.it

ABSTRACT

Introduction Disruptions of proteasome and autophagy systems are central events in amyotrophic lateral sclerosis (ALS) and support the urgent need to find therapeutic compounds targeting these processes. The heat shock protein B8 (HSPB8) recognises and promotes the autophagy-mediated removal of misfolded mutant SOD1 and TDP-43 fragments from ALS motor neurons (MNs), as well as aggregating species of dipeptides produced in C9ORF72-related diseases. In ALS-SOD1 mice and in human ALS autopsy specimens, HSPB8 is highly expressed in spinal cord MNs that survive at the end stage of disease. Moreover, the HSPB8–BAG3–HSP70 complex maintains granulostasis, which avoids conversion of dynamic stress granules (SGs) into aggregation-prone assemblies. We will perform a randomised clinical trial (RCT) with colchicine, which enhances the expression of HSPB8 and of several autophagy players, blocking TDP-43 accumulation and exerting crucial activities for MNs function.

Methods and analysis Colchicine in amyotrophic lateral sclerosis (Co-ALS) is a double-blind, placebo-controlled, multicentre, phase II RCT. ALS patients will be enrolled in three groups (placebo, colchicine 0.01 mg/day and colchicine 0.005 mg/day) of 18 subjects treated with riluzole; treatment will last 30 weeks, and follow-up will last 24 weeks. The primary aim is to assess whether colchicine decreases disease progression as measured by ALS Functional Rating Scale - Revised (ALSFRS-R) at baseline and at treatment end. Secondary aims include assessment of (1) safety and tolerability of Colchicine in patiets with ALS; (2) changes in cellular activity (autophagy, protein aggregation, and SG and exosome secretion) and in biomarkers of disease progression (neurofilaments); (3) survival and respiratory function and (4) quality of life. Preclinical studies with a full assessment of autophagy and neuroinflammation biomarkers in fibroblasts, peripheral blood mononuclear cells and lymphoblasts will be conducted in parallel with clinic assessment to optimise time and resources.

Ethics and dissemination The study protocol was approved by the Ethics Committee of Area Vasta Emilia Nord and by Agenzia Italiana del Farmaco (EUDRACT N.2017-004459-21) based on the Declaration of Helsinki. This research protocol was written without patient involvement. Patients' association will be involved in disseminating the study design and results. Results will be presented during scientific symposia or published in scientific journals.

Trial registration number EUDRACT 2017-004459-21; NCT03693781; Pre-results.

Strengths and limitations of this study

- Amyotrophic lateral sclerosis (ALS) is a rare and devastating disease without an effective treatment so far; this urgent gap can be filled with a multicentre randomised controlled trial (RCT) that can give reliable data on candidate molecules.
- Colchicine in ALS is going to be a randomised clinical trial using a drug (colchicine) that potentially targets multiple complex mechanisms involved in ALS, such as autophagy and inflammation, with the potential of slowing disease progression, but the possible limitation that we might not be able to understand which specific process prevaricates in disease initiation and then progression.
- The absence of reliable biomarkers for disease progression and drug efficacy is an important target to be addressed in ALS; the study will give inference to possible biomarkers of disease progression (neurofilaments and neuroinflammation biomarkers) and will allow in vivo search of colchicine-driven modifications of disease (study of autophagy process, protein aggregation, exosomes and mRNA) compared to placebo arm, which can widen knowledge about ALS pathogenesis.
- Colchicine is an already approved drug, with known pharmacokinetics and safety, which is already available, and therefore there is significant possibility of transferability to patients and of a rapid translation to daily clinics.
- This short study will optimise time and resources to get reliable information for a following larger phase III RCT.

Patients' association will be involved in disseminating the study design at patients' enrolment beginning, to allow patients to participate in it; patients' association will be involved in study results dissemination not only to participants but to the entire patient communities (eg, by website information)

Author affiliations
[1]Department of Neurosciences, St. Agostino Estense Hospital, Azienda Ospedaliero Universitaria di Modena, Modena, Italy
[2]Dipartimento di Scienze Farmacologiche e Biomolecolari, Centro di Eccellenza sulle Malattie Neurodegenerative, Università degli Studi di Milano, Milano, Italy
[3]Genomics and Post-Genomics Center, IRCCS Mondino Foundation, Pavia, Italy
[4]Laboratory of Translational Biomarkers, Istituto di Ricerche Farmacologiche Mario Negri IRCCS, Milano, Italy
[5]Department of Biomedical, Metabolic and Neural Sciences, University of Modena and Reggio Emilia, Modena, Italy
[6]Department of Brain and Behavioral Sciences, University of Pavia, Pavia, Italy
[7]Department of General Neurology, Rare Diseases Unit, IRCCS Mondino Foundation, Pavia, Italy
[8]"Rita Levi Montalcini" Departmentof Neurosciences, ALS Centre, University of Turin and Azienda Ospedaliero Universitaria Città della Salute e della Scienza, Turin, Italy
[9]Department of Diagnostic, Clinical and Public Health Medicine, University of Modena and Reggio Emilia, Modena, Italy
[10]Dipartimento ad attività integratedi Medicina Interna e Specialistica, Azienda Ospedaliero Universitaria "L. Vanvitelli", Napoli, Italy
[11]Department of Neurology, Institute of Experimental Neurology (INSPE), Division of Neuroscience, IRCCS San Raffaele Scientific Institute, Vita-Salute San Raffaele University, Milan, Italy
[12]Neuromuscular Omni Centre (NEMO), Fondazione Serena Onlus, Policlinico A. Gemelli IRCCS, Roma, Italy
[13]Istituto di Neurologia, Università Cattolica del Sacro Cuore, Roma, Italy
[14]UOC di Neurologia, Dipartimento di Scienze dell'invecchiamento, Neurologiche, ortopediche e della testa collo, Fondazione Policlinico Universitario A. Gemelli IRCCS, Roma, Italy
[15]Department of Neurology-Stroke Unitand Laboratory of Neuroscience, Istituto Auxologico Italiano IRCCS, Milan, Italy
[16]Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, Università degli Studi di Milano, Milano, Italy
[17]Department of Basic Medical Sciences, Neurosciences and Sense Organs, University of Bari, Bari, Italy
[18]Department of Neurosciences, University of Padua, Padua, Italy
[19]Centre of Integrative Biology (CIBIO), University of Trento, Trento, Italy
[20]Centre for Neuroscience and Nanotechnology, Department of Biomedical, Metabolic and Neural Sciences, University of Modena and Reggio Emilia, Modena, Italy

Acknowledgements The authors thank the Co-ALS investigators group: Department of Neuroscience, St. Agostino Estense Hospital, Azienda Ospedaliero Universitaria di Modena, Modena, Italy (JM, N Fini and AG); Dipartimento di Scienze Farmacologiche e Biomolecolari, Centro di Eccellenza sulle Malattie Neurodegenerative, Università degli Studi di Milano (APo and VC); Genomics and Post-Genomics Center, 'C. Mondino' National Neurological Institute, Pavia, Italy (CC, O Pansarasa and S Gagliardi); Laboratory of Translational Biomarkers, Istituto di Ricerche Farmacologiche (Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS)), M. Negri, Milan, Italy (VB, L Pasetto and S Luotti); Department of Biomedical, Metabolic and Neural Sciences, University of Modena and Reggio Emilia, Modena, Italy (A Fasano and EZ); Unit of Statistics, Department of Diagnostic, Clinical and Public Health Medicine, University of Modena and Reggio Emilia, Modena, Italy (RD and R Vicini); 'Rita Levi Montalcini' Department of Neurosciences, ALS Centre, University of Turin and Azienda Ospedaliero UniversitariaCittà della Salute e della Scienza, Turin, Italy (AC, A Calvo, C Moglia, U Manera, G Fuda, A Canosa and A Ilardi); Department of Neurosciences, University of Padua, Padua,Italy (GS and M Gizzi); Istituto Auxologico Italiano, IRCCS, University of Milan Medical School, Milan, Italy (VS, N Ticozzi, A Doretti, C Morelli and G Dimirtzidis); 'C. Mondino' National Neurological Institute, University of Pavia, Pavia, Italy (MC, CC, L Diamanti, O Pansarasa and S Gagliardi); Department of Basic Medical Sciences, Neurosciences and Sense Organs, University of Bari, Bari, Italy (ILS, E D'Errico, EA Introna and I Tempesta); Department of Neurology, Institute of Experimental Neurology (INSPE), Division of Neuroscience, IRCCS San Raffaele Scientific Institute, Vita-Salute San Raffaele University, Milan, Italy (NR); Istituto di Neurologia, Università Cattolica del Sacro Cuore, Rome, Italy; UOC di Neurologia, Dipartimento di Scienze dell'invecchiamento, neurologiche, ortopediche e della testa collo, Fondazione Policlinico Universitario A. Gemelli IRCCS, Rome, Italy; Neuromuscular OmniCentre, Fondazione Serena onlus, Policlinico A. Gemelli IRCCS (MS and A Conte); Dipartimento ad attività integrate di Medicina Interna e Specialistica, Azienda Ospedaliero Universitaria 'L. Vanvitelli', Napoli, Italy (MRM, F Trojsi and D Ricciardi); Laboratory of Molecular Biology, University of Modena and Reggio Emilia, Modena, Italy (SC); and Centre for Integrative Biology, University of Trento, Povo (TN), Italy (VGD and AP).

Contributors Conceptualization: JM, EZ, VB, SC, CC, VC, RD, APr, VGD and APo. Data curation: JM, EZ, AG, VB, SC, CC, MC, AC, VC, RD, MRM, NR, MS, VS, ILS, GS and APo. Formal analysis: JM, EZ, AG, VB, SC, CC, VC, RD, APr, VGD and APo. Funding acquisition: JM, VB, SC, CC, RD, MC, AC, MRM, NR, MS, VS, ILS, GS and APo. Investigation: JM, EZ, AG, MC, AC, MRM, NR, MS, VS, ILS and GS. Methodology: JM, EZ, AG, VB, SC, CC, VC, RD, APr, VGD and APo. Project administration: JM and APo. Resources: JM and APo. Software: RD. Supervision: JM, VB, SC, CC, VC, RD, APr, VGD and APo. Validation: JM and RD. Visualization: JM and APo. Writing—original draft: JM and EZ. Writing—review and editing: JM, EZ, AG, VB, SC, CC, MC, AC, VC, RD, MRM, NR, MS, VS, ILS, GS, APr, VGD and APo.

Funding This is an academic, independent clinical research project supported by Agenzia Italiana del Farmaco, grant number 2016-02364678. Participating units and patients will not be paid.

Competing interests None declared.

Patient consent for publication Obtained.

Ethics approval Comitato Etico Area Vasta Emilia Nord approval ID: 366/2018/FARM/AOUMO—Co-ALS.

Provenance and peer review Not commissioned; externally peer reviewed.

REFERENCES

1. Taylor JP, Brown RH, Cleveland DW. Decoding ALS: from genes to mechanism. *Nature* 2016;539:197–206.
2. Calvo A, Moglia C, Lunetta C, *et al*. Factors predicting survival in ALS: a multicenter Italian study. *J Neurol* 2017;264:54–63.
3. Miller RG, Mitchell JD, Moore DH. Riluzole for amyotrophic lateral sclerosis (ALS)/motor neuron disease (MND). *Cochrane Database Syst Rev* 2012:CD001447.
4. Writing GroupEdaravone (MCI-186) ALS 19 Study Group. Safety and efficacy of edaravone in well defined patients with amyotrophic lateral sclerosis: a randomised, double-blind, placebo-controlled trial. *Lancet Neurol* 2017;16:505–12.
5. Mitsumoto H, Brooks BR, Silani V. Clinical trials in amyotrophic lateral sclerosis: why so many negative trials and how can trials be improved? *Lancet Neurol* 2014;13:1127–38.
6. van den Berg LH, Sorenson E, Gronseth G, *et al*. Revised Airlie House consensus guidelines for design and implementation of ALS clinical trials. *Neurology* 2019;92:e1610–23.
7. Thomas M, Alegre-Abarrategui J, Wade-Martins R. RNA dysfunction and aggrephagy at the centre of an amyotrophic lateral sclerosis/frontotemporal dementia disease continuum. *Brain* 2013;136:1345–60.
8. Iguchi Y, Eid L, Parent M, *et al*. Exosome secretion is a key pathway for clearance of pathological TDP-43. *Brain* 2016;139:3187–201.
9. Buchan JR, Kolaitis RM, Taylor JP, *et al*. Eukaryotic stress granules are cleared by autophagy and Cdc48/VCP function. *Cell* 2013;153:1461–74.
10. Chitiprolu M, Jagow C, Tremblay V, *et al*. A complex of C9ORF72 and p62 uses arginine methylation to eliminate stress granules by autophagy. *Nat Commun* 2018;9:2794.
11. Zhao W, Beers DR, Bell S, *et al*. TDP-43 activates microglia through NF-κB and NLRP3 inflammasome. *Exp Neurol* 2015;273:24–35.
12. Cadwell K. Crosstalk between autophagy and inflammatory signalling pathways: balancing defence and homeostasis. *Nat Rev Immunol* 2016;16:661–75.

Amyotrophic Lateral Sclerosis and Frontotemporal Degeneration

ISSN: 2167-8421 (Print) 2167-9223 (Online) Journal homepage: https://www.tandfonline.com/loi/iafd20

CSF angiogenin levels in amyotrophic lateral Sclerosis-Frontotemporal dementia spectrum

C. Morelli, C. Tiloca, C. Colombrita, A. Zambon, D. Soranna, A. Lafronza, F. Solca, L. Carelli, B. Poletti, A. Doretti, F. Verde, L. Maderna, N. Ticozzi, A. Ratti & V. Silani

To cite this article: C. Morelli, C. Tiloca, C. Colombrita, A. Zambon, D. Soranna, A. Lafronza, F. Solca, L. Carelli, B. Poletti, A. Doretti, F. Verde, L. Maderna, N. Ticozzi, A. Ratti & V. Silani (2019): CSF angiogenin levels in amyotrophic lateral Sclerosis-Frontotemporal dementia spectrum, Amyotrophic Lateral Sclerosis and Frontotemporal Degeneration

To link to this article: https://doi.org/10.1080/21678421.2019.1704016

PROVISIONAL BEST PRACTICES GUIDELINES FOR THE EVALUATION OF BULBAR DYSFUNCTION IN AMYOTROPHIC LATERAL SCLEROSIS

GARY L. PATTEE, MD,[1] EMILY K. PLOWMAN, PHD/CCC-SLP,[2] KENDREA L. (FOCHT) GARAND, PHD, CSCD/CCC-SLP,[3] JOHN COSTELLO, MA/CCC-SLP,[4] BENJAMIN RIX BROOKS, MD,[5] JAMES D. BERRY, MD, MPH,[6] RICHARD A. SMITH, MD,[7] NAZEM ATASSI, MD,[6] JENNIFER L. CHAPIN, MA/CCC-SLP,[2] YANA YUNUSOVA, PHD/CCC-SLP,[8] COURTNEY E. MCILDUFF, MD,[9] EUFROSINA YOUNG, MD,[10] ERIC A. MACKLIN, PHD,[11] EDUARDO R. LOCATELLI, MD, MPH,[12] VINCENZO SILANI, MD,[13] DARAGH HEITZMAN, MD,[14] JAMES WYMER, MD, PHD,[15] STEPHEN A. GOUTMAN, MD, MS,[16] DEBORAH F. GELINAS, MD,[17] BRIDGET PERRY, PHD/CCC-SLP, MS,[4] PAIGE NALIPINSKI, MA/CCC-SLP,[18] KAILA STIPANCIC, MA/CCC-SLP,[4] MEGHAN O'BRIEN, MS/CCC-SLP,[4] STACEY L. SULLIVAN, MS/CCC-SLP,[18] ERIK P. PIORO, MD, PHD,[19] GISELLA GARGIULO, MD, PHD,[20] JORDAN R. GREEN, PHD/CCC-SLP,[4] and CONTRIBUTING MEMBERS OF THE NEALS BULBAR SUBCOMMITTEE

[1] Neurology Associates P.C, Lincoln, NE, U.S.A.
[2] Department of Speech, Language and Hearing Sciences, University of Florida, Gainesville, FL, U.S.A.
[3] Department of Speech Pathology and Audiology, University of South Alabama, Mobile, AL, U.S.A.
[4] Speech and Feeding Disorders Lab, MGH Institute of Health Professions, Charlestown, MA, U.S.A., ALS Augmentative Communication Program, Boston /children's Hospital, Boston, MA, U.S.A.
[5] Carolinas Medical Center, Charlotte, NC, U.S.A.
[6] MGH Department of Neurology, Boston, MA, U.S.A.
[7] Center for Neurologic Study, San Diego, CA, U.S.A.
[8] Department of Speech-Language Pathology, University of Toronto, Toronto, ON, CANADA
[9] Beth Israel Deaconess Medical Center, Boston, MA, U.S.A.
[10] State University of New York, Department of Neurology, Syracuse, NY, U.S.A.
[11] MGH Biostatistics Center, Boston, MA, U.S.A.
[12] Holy Cross Hospital, Department of Neurology, Fort Lauderdale, FL, U.S.A.
[13] Istituto Auxologico Italiano, IRCCS, Department of Neurology and Laboratory of Neuroscience, Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, Università degli studi di Milano, Milan 20122, Italy
[14] Texas Neurology, P.A, Dallas, TX, U.S.A.
[15] University of Florida, Rehabilitation Science, Gainesville, FL, U.S.A.
[16] University of Michigan, Department of Neurology, Ann Arbor, MI, U.S.A.
[17] UNC Health Care, Chapel Hill, NC, U.S.A.
[18] MGH Speech Language Pathology, Boston, MA, U.S.A.
[19] Cleveland Clinic, Department of Neurology, Cleveland, OH, U.S.A.
[20] National Scientific and Technical Research Council, Buenos Aires, Argentina

Accepted 2 January 2019

ABSTRACT: *Introduction:* Universally established comprehensive clinical bulbar scales objectively assessing disease progression in amyotrophic lateral sclerosis (ALS) are currently lacking. The goal of this working group project is to design a best practice set of provisional bulbar ALS guidelines, available for immediate implementation within all ALS clinics. *Methods:* ALS specialists across multiple related disciplines participated in a series of clinical bulbar symposia, intending to identify and summarize the currently accepted best practices for the assessment and management of bulbar dysfunction in ALS *Results:* Summary group recommendations for individual speech, Augmentative and Alternative Communication (AAC), and swallowing sections were achieved, focusing on the optimal proposed level of care within each domain. *Discussion:* We have identified specific clinical recommendations for each of the 3 domains of bulbar functioning, available for incorporation within all ALS clinics. Future directions will be to establish a formal set of bulbar guidelines through a methodological and evidence-based approach.

Muscle Nerve **000**: 000–000, 2019

Abbreviations: AAC, Alternative and Augmentative Communication; ALS, amyotrophic lateral sclerosis; ALS-FRS-R, ALS functional rating scale-revised; CNS-BFS, Center for Neurologic Study Bulbar Function Scale; CPF, cough peak flow; FVC, forced vital capacity; NEALS, Northeast ALS Consortium; PBA, pseudobulbar affect; SLP, speech-language pathologist; SVC, slow vital capacity; VFSS, Videofluoroscopic Swallow Study.

Key words: Bulbar; Speech; Swallowing; Guidelines; AAC
Funding: Cytokinetics Pharmaceuticals, Center for Neurologic Study (CNS)
Conflicts of Interest: The authors declare no financial conflicts of interest for the work presented in this article.

Correspondence to: Gary L. Pattee, M.D. 2631 South 70 Street Lincoln, NE 68506; glpattee@gmail.com

Published online 00 Month 2019 in Wiley Online Library (wileyonlinelibrary.com). DOI 10.1002/mus.26408

Bulbar dysfunction in amyotrophic lateral sclerosis (ALS) significantly contributes to reductions in quality of life, social withdrawal, malnutrition, dehydration, aspiration pneumonia, and early mortality. A globally recognized working metric, incorporating reliable clinical assessment scales to monitor bulbar disease progression in ALS, has yet to be achieved. Previous attempts aimed at achieving this metric have fallen short of a satisfactory and comprehensive protocol.[1–5] The Northeast ALS (NEALS) bulbar subcommittee has recently completed a bulbar practice survey regarding current practice patterns within participating sites, which identified significant inconsistencies involving the assessment and management of bulbar dysfunction in ALS.[6] These survey results revealed an urgent need to design and incorporate a best practice set of provisional guidelines, intended to comprehensively assess and monitor bulbar dysfunction across clinical sites. In an attempt to

REVIEW

Sexuality and intimacy in ALS: systematic literature review and future perspectives

Barbara Poletti,[1] Laura Carelli,[1] Federica Solca,[2] Rita Pezzati,[3,4] Andrea Faini,[5] Nicola Ticozzi,[1,2] Hiroshi Mitsumoto,[6] Vincenzo Silani[1,2]

ABSTRACT

Several features of amyotrophic lateral sclerosis (ALS) impact on sexuality and intimate relationship; however, the issue has received poor attention so far. We performed a systematic literature review in order to provide an up-to-date account of sexuality in ALS. References were identified by searches of PubMed, Web of Science, Scopus and PsycINFO (1970–2017, English literature). The following were the key terms: 'sexual' OR 'sexuality' OR 'intimacy' OR 'marital' AND 'ALS' OR 'Amyotrophic Lateral Sclerosis' OR 'Motor Neuron Disease' OR 'MND'. Titles and abstracts were screened for relevance and a full-text analysis was performed on the selected articles. Studies were included if they referred to sexual well-being/activities/functions or intimate relationship between patients and their partners and management of such topic by clinicians. Eligibility assessment was performed independently by two reviewers. A thematic and level of evidence classification of studies was performed. Studies' design, objectives, measurements and outcomes were summarised. Thirty articles were included and four topics were identified: intimacy in the dyads; sexual activities in patients and with their partners; sexual function disturbances; and sexuality and cognitive-behavioural alterations. The quality of the studies varies, with globally poor level of evidence. Some sexuality issues have been only sparsely addressed, such as gender-related differences, same-sex relationships and sexual activities other than intercourse. Sexuality in ALS is still not adequately considered by clinicians and researchers. We present preliminary recommendations for improving sexuality and intimacy at any ALS multidisciplinary clinics.

INTRODUCTION

Sexuality in patients with amyotrophic lateral sclerosis (ALS) has received poor attention so far. One possible reason is that ALS represents a severe and progressive neurological disorder leading clinicians and researchers to focus on critical features exerting an effect on treatment and survival, such as movement, respiratory and nutritional aspects. Moreover, patients themselves often express the feeling that when everyday survival is an issue, talking about sexuality is rather embarrassing and an inappropriate subject.[1] However, even if not directly affecting survival, sexual activities are well recognised as an important component of daily life and of intimate relationships; they have significant impact on emotional well-being and therefore, indirectly, on disease-related aspects as observed in other disorders, both neurological or due to other aetiologies.[2 3] Another possible explanation for the poor consideration of sexuality issue in ALS is that this condition affects the motor system, causing skeletal muscle weakness, but it does not directly involve sexual functions. However, there are several aspects of ALS disease and progression that could influence sexuality and intimate relationship, involving both physical, cognitive-behavioural, emotional and psychosocial dimensions. In fact, both physical weakness and psychological features (ie, poor self-esteem, change in one's body image, depression) can indirectly impact sexuality. Moreover, the presence of contrasting representations of disease limitations and consequences among patients and their partners could impact on desire and willingness to engage in sexual activities.[4 5] Another reason to investigate sexuality in ALS is related to the presence of possible ALS-frontotemporal spectrum alterations, previously described and also involving disinhibition and inappropriate sexual behaviour, as observed in other neurodegenerative disorders.[6 7]

Starting from the observation that sexuality plays a crucial role in personal well-being, and from the inadequate consideration of this aspect in ALS care, we aimed to collect and summarise existing information about sexuality issues in ALS. The objectives of the present systematic review were twofold: to provide an up-to-date account of sexuality aspects and changes in ALS and to highlight both available and missing information/practices in literature and clinical care. As an outcome, we intended to provide preliminary recommendations for improving sexuality in patients with ALS and their partners within ALS multidisciplinary clinics. Specifically, the following are our questions: Is there evidence of an adequate consideration of sexuality-related topics among both researchers and clinicians in literature and clinical practice? Are patients' and spouses' needs and quality of life (QoL) issues related to sexuality and intimacy taken into account, or is most clinicians' attention focused on alterations or dysfunctions from a clinical/pathological point of view? Then, if both patients' and spouses' point of views are considered in the literature, is there any difference between them concerning sexuality interest or satisfaction?

METHODS

Eligibility criteria

Patients with ALS, together with their carers and health professionals, were considered as study participants. Both inpatients and outpatients were considered, of any age and at any disease stage. Due to the limited consideration of sexuality issues in the literature, studies of any type (experimental, observational, single-case studies and literature

► Additional material is published online only. To view please visit the journal online (http://dx.doi.org/10.1136/jnnp-2018-319684).

[1]Department of Neurology and Laboratory of Neuroscience, Istituto Auxologico Italiano, IRCCS, Milan, Italy
[2]Department of Pathophysiology and Transplantation, 'Dino Ferrari' Center, Università degli Studi di Milano, Milan, Italy
[3]University of Applied Sciences and Arts of Southern Switzerland, Manno, Switzerland
[4]Centro Terapia Cognitiva, Como, Italy
[5]Department of Cardiovascular, Neural and Metabolic Sciences, Istituto Auxologico Italiano, IRCCS, Milan, Italy
[6]Eleanor and Lou Gehrig ALS Center, Department of Neurology, Columbia University Medical Center, New York City, New York, USA

Correspondence to
Dr Vincenzo Silani, Department of Neurology and Laboratory of Neuroscience, Istituto Auxologico Italiano, IRCCS, Milan 20149, Italy; vincenzo.silani@unimi.it

BP and LC contributed equally. HM and VS contributed equally.

Received 20 September 2018
Revised 5 November 2018
Accepted 5 November 2018

To cite: Poletti B, Carelli L, Solca F, *et al*. *J Neurol Neurosurg Psychiatry* Epub ahead of print: [*please include* Day Month Year]. doi:10.1136/jnnp-2018-319684

frontiers in Neuroscience

ORIGINAL RESEARCH
published: 17 May 2019
doi: 10.3389/fnins.2019.00485

Comparative Analysis of *C9orf72* and Sporadic Disease in a Large Multicenter ALS Population: The Effect of Male Sex on Survival of *C9orf72* Positive Patients

Francesca Trojsi[1†], Mattia Siciliano[1,2†], Cinzia Femiano[1], Gabriella Santangelo[2], Christian Lunetta[3,4], Andrea Calvo[5], Cristina Moglia[5], Kalliopi Marinou[6], Nicola Ticozzi[7,8], Christian Ferro[9], Carlo Scialò[10], Gianni Sorarù[11], Amelia Conte[12], Yuri M. Falzone[13], Rosanna Tortelli[14], Massimo Russo[4,15], Valeria Ada Sansone[3,16], Adriano Chiò[5], Gabriele Mora[6], Vincenzo Silani[7,8], Paolo Volanti[9], Claudia Caponnetto[10], Giorgia Querin[11], Mario Sabatelli[12,17], Nilo Riva[13], Giancarlo Logroscino[14], Sonia Messina[4,15], Antonio Fasano[18], Maria Rosaria Monsurrò[1], Gioacchino Tedeschi[1] and Jessica Mandrioli[18]*

[1] Department of Advanced Medical and Surgical Sciences, MRI Research Center SUN-FISM, University of Campania "Luigi Vanvitelli", Naples, Italy, [2] Department of Psychology, Università degli Studi della Campania "L. Vanvitelli", Naples, Italy, [3] NEuroMuscular Omnicentre (NEMO), Serena Onlus Foundation, Milan, Italy, [4] NEMO Sud Clinical Center for Neuromuscular Diseases, Aurora Onlus Foundation, Messina, Italy, [5] ALS Center, "Rita Levi Montalcini" Department of Neuroscience, University of Torino, Turin, Italy, [6] Department of Neurorehabilitation–ALS Center, IRCCS Scientific Clinical Institute Maugeri, Milan, Italy, [7] Department of Neurology and Laboratory of Neuroscience, IRCCS Istituto Auxologico Italiano, Milan, Italy, [8] Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, University of Milan, Milan, Italy, [9] Neurorehabilitation Unit/ALS Center, Scientific Clinical Institutes (ICS) Maugeri, IRCCS, Messina, Italy, [10] Department of Neurosciences, Rehabilitation, Ophthalmology, Genetics, Maternal, and Child Health (DINOGMI), University of Genova, IRCCS AOU San Martino-IST, Genova, Italy, [11] Department of Neurosciences, Neuromuscular Center, University of Padova, Padua, Italy, [12] NEuroMuscular Omnicentre (NEMO), Serena Onlus Foundation–Pol. A. Gemelli Foundation, Rome, Italy, [13] Department of Neurology, Institute of Experimental Neurology (INSPE), Division of Neuroscience, San Raffaele Scientific Institute, Milan, Italy, [14] Department of Clinical Research in Neurology, University of Bari "A. Moro", at Pia Fondazione "Card. G. Panico", Lecce, Italy, [15] Department of Clinical and Experimental Medicine, University of Messina, Messina, Italy, [16] Department of Biomedical Sciences for Health, University of Milan, Milan, Italy, [17] Department of Geriatrics, Neurosciences and Orthopedics, Institute of Neurology, Catholic University of Sacred Heart, Rome, Italy, [18] Department of Neuroscience, S. Agostino-Estense Hospital and University of Modena and Reggio Emilia, Modena, Italy

OPEN ACCESS

Edited by:
Henry Houlden,
University College London,
United Kingdom

Reviewed by:
Sulev Kõks,
University of Tartu, Estonia
Amber L. Southwell,
University of Central Florida,
United States

***Correspondence:**
Francesca Trojsi
francesca.trojsi@unicampania.it

[†]These authors have contributed equally to this work

Specialty section:
This article was submitted to Neurogenomics, a section of the journal Frontiers in Neuroscience

Received: 26 February 2019
Accepted: 29 April 2019
Published: 17 May 2019

Citation:
Trojsi F, Siciliano M, Femiano C, Santangelo G, Lunetta C, Calvo A, Moglia C, Marinou K, Ticozzi N, Ferro C, Scialò C, Sorarù G, Conte A, Falzone YM, Tortelli R, Russo M, Sansone VA, Chiò A, Mora G, Silani V, Volanti P, Caponnetto C, Querin G, Sabatelli M, Riva N, Logroscino G, Messina S, Fasano A, Monsurrò MR, Tedeschi G and Mandrioli J (2019) Comparative Analysis of C9orf72 and Sporadic Disease in a Large Multicenter ALS Population: The Effect of Male Sex on Survival of C9orf72 Positive Patients. Front. Neurosci. 13:485. doi: 10.3389/fnins.2019.00485

We investigated whether the *C9orf72* repeat expansion is associated with specific clinical features, comorbidities, and prognosis in patients with amyotrophic lateral sclerosis (ALS). A cohort of 1417 ALS patients, diagnosed between January 1, 2009 and December 31, 2013 by 13 Italian ALS Referral Centers, was screened for the *C9orf72* repeat expansion, and the analyses were performed comparing patients carrying this expansion (ALS-C9Pos) to those negative for this and other explored ALS-related mutations (ALS without genetic mutations, ALSwoGM). Compared to the ALSwoGM group, ALS-C9Pos patients ($n = 84$) were younger at disease onset, at the first clinical observation and at diagnosis ($p < 0.001$). After correcting for these differences, we found that ALS-C9Pos patients had higher odds of bulbar onset, diagnosis of frontotemporal dementia (FTD) and family history of ALS, FTD, and Alzheimer's disease and had lower odds of spinal onset, non-invasive ventilation, hypertension and psychiatric diseases than

Published Ahead of Print on March 8, 2019 as 10.1212/WNL.0000000000007242

ARTICLE OPEN ACCESS

Revised Airlie House consensus guidelines for design and implementation of ALS clinical trials

Leonard H. van den Berg, MD, PhD, Eric Sorenson, MD, Gary Gronseth, MD, Eric A. Macklin, PhD, Jinsy Andrews, MD, Robert H. Baloh, MD, PhD, Michael Benatar, MD, PhD, James D. Berry, MD, Adriano Chio, MD, Philippe Corcia, MD, PhD, Angela Genge, MD, Amelie K. Gubitz, PhD, Catherine Lomen-Hoerth, MD, PhD, Christopher J. McDermott, MD, Erik P. Pioro, MD, PhD, Jeffrey Rosenfeld, MD, PhD, Vincenzo Silani, MD, Martin R. Turner, MBBS, PhD, Markus Weber, MD, Benjamin Rix Brooks, MD, Robert G. Miller, MD, and Hiroshi Mitsumoto, MD, DSc, for the Airlie House ALS Clinical Trials Guidelines Group

Correspondence
Dr. van den Berg
L.H.vandenBerg@umcutrecht.nl

Neurology® 2019;92:e1-e14. doi:10.1212/WNL.0000000000007242

Abstract

Objective

To revise the 1999 Airlie House consensus guidelines for the design and implementation of preclinical therapeutic studies and clinical trials in amyotrophic lateral sclerosis (ALS).

Methods

A consensus committee comprising 140 key members of the international ALS community (ALS researchers, clinicians, patient representatives, research funding representatives, industry, and regulatory agencies) addressed 9 areas of need within ALS research: (1) preclinical studies; (2) biological and phenotypic heterogeneity; (3) outcome measures; (4) disease-modifying and symptomatic interventions; (5) recruitment and retention; (6) biomarkers; (7) clinical trial phases; (8) beyond traditional trial designs; and (9) statistical considerations. Assigned to 1 of 8 sections, committee members generated a draft set of guidelines based on a "background" of developing a (pre)clinical question and a "rationale" outlining the evidence and expert opinion. Following a 2-day, face-to-face workshop at the Airlie House Conference Center, a modified Delphi process was used to develop draft consensus research guidelines, which were subsequently reviewed and modified based on comments from the public. Statistical experts drafted a separate document of statistical considerations (section 9).

Results

In this report, we summarize 112 guidelines and their associated backgrounds and rationales. The full list of guidelines, the statistical considerations, and a glossary of terms can be found in data available from Dryad (appendices e-3–e-5, doi.org/10.5061/dryad.32q9q5d). The authors prioritized 15 guidelines with the greatest potential to improve ALS clinical research.

Conclusion

The revised Airlie House ALS Clinical Trials Consensus Guidelines should serve to improve clinical trial design and accelerate the development of effective treatments for patients with ALS.

From the Department of Neurology (L.H.v.d.B.), Brain Center Rudolf Magnus, University Medical Center Utrecht, the Netherlands; Department of Neurology (E.S.), Mayo Clinic, Rochester, MN; Department of Neurology (G.G.), University of Kansas Medical Center, Kansas City; Department of Medicine (E.A.M.), Massachusetts General Hospital, Biostatistics Center, Harvard Medical School, Boston; Department of Neurology (J.A., H.M.), Columbia University, Eleanor and Lou Gehrig ALS Center, New York, NY; Department of Neurology (R.H.B.), Cedars-Sinai Medical Center, Los Angeles, CA; Department of Neurology (M.B.), University of Miami, FL; Neurological Clinical Research Institute (J.D.B.), Massachusetts General Hospital, Boston; Rita Levi Montalcini Department of Neuroscience (A.C.), University of Torino, Italy; Centre Constitutif SLA (P.C.), Université de Tours, France; Department of Neurology (A.G.), Clinical Research Unit, Montreal Neurological Institute, Neurosurgery, McGill University, Montreal, Canada; National Institute of Neurological Disorders and Stroke (A.K.G.), National Institutes of Health, Bethesda, MD; ALS Center (C.L.-H.), University of California San Francisco; Department of Neuroscience (C.J.M.), Sheffield Institute for Translational Neuroscience, University of Sheffield, UK; Department of Neurology (E.P.P.), Section of ALS & Related Disorders, Cleveland Clinic, OH; Department of Neurology (J.R.), The Center for Restorative Neurology, Loma Linda University School of Medicine, CA; Department of Neurology and Laboratory of Neuroscience (V.S.), Istituto Auxologico Italiano, IRCCS, Milan; Department of Pathophysiology and Transplantation (V.S.), "Dino Ferrari" Centre, Università degli Studi di Milano, Milan, Italy; Nuffield Department of Clinical Neurosciences (M.R.T.), University of Oxford, UK; Neuromuscular Diseases Unit/ALS Clinic (M.W.), Kantonsspital St. Gallen, Switzerland; Carolinas Neuromuscular/ALS-MDA Care Center (B.R.B.), Charlotte; Department of Neurology (B.R.B.), Carolinas Medical Center, University of North Carolina School of Medicine, Charlotte; Forbes Norris ALS Treatment and Research Center (R.G.M.), California Pacific Medical Center San Francisco; and Department of Neurosciences (R.G.M.), Stanford University, CA.

Go to Neurology.org/N for full disclosures. Funding information and disclosures deemed relevant by the authors, if any, are provided at the end of the article.

The Article Processing Charge was funded by Columbia University.

Coinvestigators are listed in the appendix at the end of the article.

RESEARCH PAPER

Neurofilament light chain in serum for the diagnosis of amyotrophic lateral sclerosis

Federico Verde,[1,2] Petra Steinacker,[1] Jochen H Weishaupt,[1] Jan Kassubek,[1] Patrick Oeckl,[1] Steffen Halbgebauer,[1] Hayrettin Tumani,[1] Christine A F von Arnim,[1] Johannes Dorst,[1] Emily Feneberg,[1,3] Benjamin Mayer,[4] Hans-Peter Müller,[1] Martin Gorges,[1] Angela Rosenbohm,[1] Alexander E Volk,[5] Vincenzo Silani,[2] Albert C Ludolph,[1] Markus Otto[1]

[1]Department of Neurology, University of Ulm, Ulm, Germany
[2]Department of Neurology and Laboratory of Neuroscience, IRCCS Istituto Auxologico Italiano, and Department of Pathophysiology and Transplantation, 'Dino Ferrari' Center, Università degli Studi di Milano, Milan, Italy
[3]Nuffield Department of Clinical Neurosciences, University of Oxford, Oxford, UK
[4]Institute of Epidemiology and Medical Biometry, University of Ulm, Ulm, Germany
[5]Institute of Human Genetics, University Medical Center Hamburg-Eppendorf, Hamburg, Germany

Correspondence to
Professor Markus Otto, Department of Neurology, University of Ulm, Ulm 89081, Germany; markus.otto@uni-ulm.de

Received 25 April 2018
Revised 3 July 2018
Accepted 24 July 2018
Published Online First 11 October 2018

To cite: Verde F, Steinacker P, Weishaupt JH, *et al*. *J Neurol Neurosurg Psychiatry* 2019;**90**:157–164.

ABSTRACT

Objective To determine the diagnostic and prognostic performance of serum neurofilament light chain (NFL) in amyotrophic lateral sclerosis (ALS).

Methods This single-centre, prospective, longitudinal study included the following patients: 124 patients with ALS; 50 patients without neurodegenerative diseases; 44 patients with conditions included in the differential diagnosis of ALS (disease controls); 65 patients with other neurodegenerative diseases (20 with frontotemporal dementia, 20 with Alzheimer's disease, 19 with Parkinson's disease, 6 with Creutzfeldt-Jakob disease (CJD)). Serum NFL levels were measured using the ultrasensitive single molecule array (Simoa) technology.

Results Serum NFL levels were higher in ALS in comparison to all other categories except for CJD. A cut-off level of 62 pg/mL discriminated between ALS and all other conditions with 85.5% sensitivity (95% CI 78% to 91.2%) and 81.8% specificity (95% CI 74.9% to 87.4%). Among patients with ALS, serum NFL correlated positively with disease progression rate (r_s=0.336, 95% CI 0.14 to 0.506, p=0.0008), and higher levels were associated with shorter survival (p=0.0054). Serum NFL did not differ among patients in different ALS pathological stages as evaluated by diffusion-tensor imaging, and in single patients NFL levels were stable over time.

Conclusions Serum NFL is increased in ALS in comparison to other conditions and can serve as diagnostic and prognostic biomarker. We established a cut-off level for the diagnosis of ALS.

INTRODUCTION

Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disease causing demise of motor neurons of the cerebral cortex, brainstem and spinal cord. This leads to progressive paralysis ending with death from respiratory failure after a median of 30 months. Up to 10% of patients with ALS are also affected by frontotemporal dementia (FTD),[1] while 5%–10% of ALS cases are familial and caused by mutations in known genes.[2] The diagnosis of ALS is primarily clinical; fluid biomarkers have not yet entered clinical practice, but they are urgently needed for diagnosis, prognosis, patient stratification in clinical trials and monitoring of drug effects.[3]

The most promising biomarkers in ALS are the light chain and phosphorylated heavy chain of neurofilaments (NFL and pNFH, respectively), cytoskeletal proteins of large myelinated axons of neurons and therefore axonal impairment markers.[4] NFL and pNFH are elevated in the cerebrospinal fluid (CSF) of patients with ALS relative to controls, with diagnostic sensitivities and specificities >80%; they also correlate with disease progression rate and survival.[5–7] Likewise, blood NF levels are higher in ALS in comparison to controls, but to date they have not been studied in a systematical comparison between ALS and several neurological control groups.[8–11] Therefore, we evaluated the usefulness of serum NFL in the diagnosis of ALS, as well as its role as prognostic biomarker and its stability over time.

METHODS

Participants and clinical characterisation

This prospective longitudinal study included 283 patients investigated in the Department of Neurology of Ulm University Hospital, Germany, between 2010 and 2016. Patients were subdivided as follows: 124 patients (74 (59.7%) male (M) and 50 (40.3%) female (F)) with ALS; 50 patients (23 (46%) M and 27 (54%) F) admitted to a neurological inpatient clinic but without a final diagnosis of degenerative or inflammatory central nervous system (CNS) disease (non-neurodegenerative controls); 44 patients (32 (72.7%) M and 12 (27.3%) F) with conditions included in the differential diagnosis of ALS (disease controls); 20 patients (11 (55%) M and 9 (45%) F) with FTD; 20 patients (8 (40%) M and 12 (60%) F) with Alzheimer's disease (AD); 19 patients (12 (63.2%) M and 7 (36.8%) F) with Parkinson's disease (PD); and 6 patients (4 (66.7%) M and 2 (33.3%) F) with Creutzfeldt-Jakob disease (CJD).

Patients with ALS were selected according to availability of blood samples and MRI examinations taken at the same time points. Patients of all other disease categories were selected according to a random criterion.

Patients with ALS had a diagnosis of definite or probable ALS according to the revised El Escorial criteria[12]; among them, four also had concomitant FTD. Disease severity was expressed by the score

Neurochemical biomarkers in amyotrophic lateral sclerosis

Federico Verde[a], *Vincenzo Silani*[a,b], *and Markus Otto*[c]

Purpose of review
The diagnosis of amyotrophic lateral sclerosis (ALS) still relies mainly on clinical criteria. In present review we will provide an overview of neurochemical ALS biomarkers, which are in the most advanced position on the way towards inclusion into the clinical work-up.

Recent findings
The field of ALS neurology still lacks a neurochemical marker for routine clinical use. However, this is urgently needed, because it would help in diagnosis, prognostic stratification, and monitoring of drug response. Despite this lack of a routinely used biomarker, in the last decade significant progress has been made in the field. In particular, two molecules have been extensively studied – the light chain and the phosphorylated form of the heavy chain of neurofilaments, NFL and pNFH, respectively – which have demonstrated a high diagnostic performance and promising prognostic value and are therefore ready to be introduced into the clinical scenario. On the other hand, we still lack a neurochemical cerebrospinal fluid or blood biomarker reflecting TDP-43 pathology.

Summary
Neurofilaments seem to be ready for clinical use in the early and differential diagnosis of ALS. We also highlight still unresolved issues which deserve further investigation.

Keywords
ALS, CHIT1, dipeptide repeats, neurofilaments, TDP-43

INTRODUCTION

Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disease affecting motor neurons. A total of 5–10% of cases are familial, usually with autosomal dominant inheritance, because of mutations in more than 25 genes, mainly *C9orf72*, *SOD1*, *TARDBP*, *FUS*, and *TBK1* [1]. About 15% of patients have comorbid frontotemporal dementia (FTD), which shares genetic, neuropathological, and clinical commonalities with ALS [2].

The diagnosis of ALS is mainly clinical [3] and a neurochemical marker for routine use is still missing. However, this is urgently needed, because it would help in diagnosis and prognosis and demonstration of target engagement and detection of drug effects in clinical trials [4].

In this review we will provide an overview of the most promising ALS biomarkers, focusing mainly on neurofilaments because they occupy the most advanced position in the pipeline leading to introduction into the clinical field, and highlighting still unsolved issues and points which deserve further investigation in future studies.

NEUROFILAMENTS

Neurofilaments are intermediate filaments and are the major component of the cytoskeleton of large myelinated axons. They are composed of different subunits: light (NFL), intermediate (NFM), heavy (NFH), and α-internexin in the central nervous system (CNS) or peripherin in the peripheral nervous system (PNS) [5].

Because of their abundance and structural role in the axons of motor neurons, neurofilaments are an optimal candidate biomarker for ALS. Here, they are supposed to rise in the cerebrospinal fluid (CSF)

[a]Department of Neurology and Laboratory of Neuroscience, IRCCS Istituto Auxologico Italiano, [b]Department of Pathophysiology and Transplantation, 'Dino Ferrari' Center, Università degli Studi di Milano, Milan, Italy and [c]Department of Neurology, University of Ulm, Ulm, Germany

Correspondence to Markus Otto, Department of Neurology, Ulm University Hospital, Oberer Eselsberg 45, D-89081 Ulm, Germany.
Tel: +49 731 500 63010; fax: +49 731 500 63012;
e-mail: markus.otto@uni-ulm.de

Curr Opin Neurol 2019, 32:747–757
DOI:10.1097/WCO.0000000000000744

Neurological Sciences (2019) 40:1469–1473
https://doi.org/10.1007/s10072-019-03834-2

BRIEF COMMUNICATION

PON1 is a disease modifier gene in amyotrophic lateral sclerosis: association of the Q192R polymorphism with bulbar onset and reduced survival

Federico Verde[1] · Cinzia Tiloca[1] · Claudia Morelli[1] · Alberto Doretti[1] · Barbara Poletti[1] · Luca Maderna[1] · Stefano Messina[1] · Davide Gentilini[2,3] · Isabella Fogh[1,4] · Antonia Ratti[1,5] · Vincenzo Silani[1,6,7] · Nicola Ticozzi[1,6]

Received: 4 November 2018 / Accepted: 11 March 2019 / Published online: 22 March 2019

Abstract

Introduction Previous studies have associated single-nucleotide polymorphisms (SNPs) in the gene encoding the detoxifying enzyme paraoxonase 1 (*PON1*) to the risk of sporadic ALS. Here, we aimed to assess the role of the coding rs662 (Q192R) SNP as a modifier of ALS phenotype.

Materials and methods We genotyped a cohort of 409 patients diagnosed with ALS at our Center between 2002 and 2009 (269 males and 140 females; mean age at onset, 58.3 ± 37.5 years).

Results We found *PON1* to be a disease modifier gene in ALS, with the minor allele G associated both with bulbar onset (30.9% vs. 24.6%, $p = 0.013$) and independently with reduced survival (OR = 1.38, $p = 0.012$) under a dominant model. No association was found with gender or age at onset.

Discussion As this SNP is known to modify the detoxifying activity of paraxonase 1 with respect to different substrates as well as other activities of the protein, we hypothesize that the identified association might reflect specific motor neuron vulnerability to certain exogenous toxic substances metabolized less efficiently by the 192R alloenzyme, or to detrimental endogenous pathophysiological processes such as oxidative stress. Further exploration of this possible metabolic susceptibility could deepen our knowledge of ALS pathomechanisms.

Keywords ALS · Motor neurons · Paraoxonase · *PON1* · SNP · Toxicity

Introduction

Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disease affecting upper and lower motor neurons and leading to death from progressive paralysis in a median time of 3–5 years [1]. ALS displays wide variation in phenotype and survival, with older age at onset and bulbar onset representing established negative prognostic factors [2]. The vast majority of ALS cases are sporadic (sALS), whereas 5–10% are familial (fALS), caused by genetic mutations. The etiology of sALS is not clearly understood, presumably involving an interaction between genetic and environmental factors [1].

Vincenzo Silani and Nicola Ticozzi jointly supervised this work.

✉ Federico Verde
fdrc.verde@gmail.com

1 Department of Neurology - Stroke Unit and Laboratory of Neuroscience, Istituto Auxologico Italiano, IRCCS, Piazzale Brescia 20, 20149 Milan, Italy

2 Unit of Bioinformatics and Genomic Statistics, Istituto Auxologico Italiano, IRCCS, Via Zucchi 18, 20095 Cusano Milanino, Italy

3 Department of Brain and Behavioral Sciences, University of Pavia, Pavia, Italy

4 Department of Basic and Clinical Neuroscience, Maurice Wohl Clinical Neuroscience Institute, King's College London, London, UK

5 Department of Medical Biotechnology and Translational Medicine, Università degli Studi di Milano, via Vanvitelli 32, 20129 Milan, Italy

6 Department of Pathophysiology and Transplantation, "Dino Ferrari" Center, Università degli Studi di Milano, Via Francesco Sforza 35, 20122 Milan, Italy

7 "Aldo Ravelli" Center for Neurotechnology and Experimental Brain Therapeutics, Università degli Studi di Milano, via Festa del Perdono 7, 20122 Milan, Italy

European Journal of Case Reports in Internal Medicine

Myalgia, Obtundity and Fever in a Patient with a Prosthesis

Valeria Di Stefano[1], Margherita Migone De Amicis[2], Cecilia Bonino[3], Natalia Scaramellini[1], Yvan Torrente[4], Stefania Piconi[5], Francesca Minonzio[2]

[1]Department of Internal Medicine, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico and Department of Clinical Sciences and Community Health, University of Milan, Milan, Italy

[2]Department of Internal Medicine, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, Milan, Italy

[3]Department of Emergency Medicine, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, University of Milan, Milan, Italy

[4]Stem Cell Laboratory, Department of Pathophysiology and Transplantation, University of Milan, Unit of Neurology, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Centro Dino Ferrari, Milan, Italy

[5]First Division of Infectious Diseases Unit, University of Milan, Ospedale L. Sacco, Milan, Italy

Doi: 10.12890/2019_001021 - European Journal of Case Reports in Internal Medicine - © EFIM 2019

Received: 16/12/2018
Accepted: 24/12/2018
Published: 08/02/2019

How to cite this article: Di Stefano V, Migone De Amicis M, Bonino C, Scaramellini N, Torrente Y, Piconi S, Minonzio F. Myalgia, obtundity and fever in a patient with a prosthesis. *EJCRIM* 2019;**6**: doi:10.12890/2019_001021.

Conflicts of Interests: The Authors declare that there are no competing interests.

ABSTRACT

Objective: We describe a rare case of group G streptococcus (GGS) sepsis complicated by bacterial toxin myopathy.
Case: A 65-year-old man, with a history of infection of his shoulder prosthesis, presented with multiorgan failure and notable myalgia likely caused by toxins. The patient was treated successfully with antibiotics and prosthesis removal.
Conclusion: This case suggests infection by GGS should be considered in a patient presenting with myalgia associated with sepsis.

LEARNING POINTS

- Infection by GGS should be considered in a patient presenting with myalgia associated with sepsis.
- The differential diagnosis in this case included a neurological condition (meningitis or atypical Guillain–Barré syndrome) and sepsis with myopathy induced by bacterial toxins.
- Group G streptococcus (GGS) infection in a prosthetic shoulder was successfully treated with antibiotics and prosthesis removal.

KEYWORDS

Group G streptococcus, streptococcal toxic shock syndrome, myopathy, myalgia, myositis

CASE DESCRIPTION

We present the case of a 65-years old patient hospitalized in February 2018 for fever, alteration of consciousness and arthromyalgia. He had a history of right shoulder replacement after traumatic fracture in 2011 and a diagnosis of persistent prosthesis infection and dislocation since 2015. Prosthesis replacement had been recommended by orthopaedics but had not been carried out.
In 2018, because of high persistent fever and diffuse pain, the patient was admitted to the Emergency Room and then to our Internal Medicine Unit. For some days before hospitalization, he had been taking non-steroidal anti-inflammatory drugs for lumbar pain without improvement.
On physical examination, the patient was obtunded, although responsive to verbal stimuli, and inclined to sleep. He presented normal vital parameters except for tachycardia and fever (BP 120/70 mmHg, HR 120/min, RR 18/min, O^2 Sat 93%, temperature 38°C). He complained of diffuse pain, mostly in the limbs, exacerbated by minimal movement and allodynia. Skin inspection demonstrated migratory skin erythema

Article

SOURCE DATA | TRANSPARENT PROCESS | OPEN ACCESS

EMBO Molecular Medicine

Blockade of IGF2R improves muscle regeneration and ameliorates Duchenne muscular dystrophy

Pamela Bella[1], Andrea Farini[1], Stefania Banfi[2], Daniele Parolini[3], Noemi Tonna[4], Mirella Meregalli[1], Marzia Belicchi[1], Silvia Erratico[5], Pasqualina D'Ursi[6], Fabio Bianco[4], Mariella Legato[1], Chiara Ruocco[7], Clementina Sitzia[8], Simone Sangiorgi[9], Chiara Villa[1], Giuseppe D'Antona[10], Luciano Milanesi[6], Enzo Nisoli[7], PierLuigi Mauri[6] & Yvan Torrente[1,*]

Abstract

Duchenne muscular dystrophy (DMD) is a debilitating fatal X-linked muscle disorder. Recent findings indicate that IGFs play a central role in skeletal muscle regeneration and development. Among IGFs, insulinlike growth factor 2 (IGF2) is a key regulator of cell growth, survival, migration and differentiation. The type 2 IGF receptor (IGF2R) modulates circulating and tissue levels of IGF2 by targeting it to lysosomes for degradation. We found that IGF2R and the store-operated Ca^{2+} channel CD20 share a common hydrophobic binding motif that stabilizes their association. Silencing CD20 decreased myoblast differentiation, whereas blockade of IGF2R increased proliferation and differentiation in myoblasts via the calmodulin/calcineurin/NFAT pathway. Remarkably, anti-IGF2R induced CD20 phosphorylation, leading to the activation of sarcoplasmic/endoplasmic reticulum Ca^{2+}-ATPase (SERCA) and removal of intracellular Ca^{2+}. Interestingly, we found that IGF2R expression was increased in dystrophic skeletal muscle of human DMD patients and *mdx* mice. Blockade of IGF2R by neutralizing antibodies stimulated muscle regeneration, induced force recovery and normalized capillary architecture in dystrophic *mdx* mice representing an encouraging starting point for the development of new biological therapies for DMD.

Keywords DMD; IGF2; IGF2R; muscle regeneration; muscular dystrophy
Subject Categories Musculoskeletal System; Pharmacology & Drug Discovery
DOI 10.15252/emmm.201911019 | Received 18 June 2019 | Revised 29 October 2019 | Accepted 30 October 2019 | Published online 2 December 2019
EMBO Mol Med (2020) 12: e11019

Introduction

Duchenne muscular dystrophy (DMD) is a devastating X-linked disease characterized by progressive muscle weakness and caused by a lack of dystrophin protein in the sarcolemma of muscle fibres (Emery, 2002). Impaired muscle regeneration with exhaustion of the satellite cell pool is a major hallmark of DMD. Members of the insulin-like growth factor (IGF) family are secreted during muscle repair and promote muscle regeneration and hypertrophy. Among the IGFs, IGF1 signalling has been extensively characterized for its capacity to promote the proliferation and differentiation of satellite cells, regulate muscle hypertrophy and ameliorate the features of muscular dystrophy (Florini *et al*, 1996; Barton *et al*, 2002; Zanou & Gailly, 2013). Nevertheless, little is known about the role of IGF2 in skeletal muscle development and regeneration *in vivo*. *In vitro* studies have shown that the IGF2 protein plays a role in a later step of myoblast differentiation (Florini *et al*, 1991; Wilson *et al*, 2003; Ge *et al*, 2011). Interestingly, it was previously shown that there is a link between the *Myod* and *Igf2* genes in myoblast cell culture (Montarras *et al*, 2005). Further studies suggested that IGF2, by binding to the IGF1 receptor, activates the Akt pathway and downstream targets of *Myod*, although the exact mechanisms underlying these processes have not been identified (Wilson & Rotwein, 2006, 2007). IGF2 signalling is regulated by IGF-binding proteins, which sequester circulating IGF2; the IGF2 receptor (IGF2R), which reduces IGF2 bioactivity (Brown *et al*, 2009); and the insulin receptor and IGF1 receptor, both of which can be activated by IGF2 (Livingstone, 2013). The extracytoplasmic region of IGF2R has three binding sites: one for IGF2 in domain 11 and two for Man-6-P in domains 3, 5 and 9 (Dahms *et al*, 1993; Reddy *et al*, 2004; Williams

1 Stem Cell Laboratory, Department of Pathophysiology and Transplantation, Unit of Neurology, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Centro Dino Ferrari, Universitá degli Studi di Milano, Milan, Italy
2 Hematology Department Fondazione IRCCS, Department of Oncology and Hemato-oncology, Istituto Nazionale dei Tumori, Universitá degli Studi di Milano, Milan, Italy
3 Thermo Fisher Scientific, Life Technologies Italia, Monza, Italy
4 Neuro-Zone s.r.l., Open Zone, Milano, Italy
5 Novystem Spa, Milan, Italy
6 Institute of Technologies in Biomedicine, National Research Council (ITB-CNR), Milan, Italy
7 Department of Medical Biotechnology and Translational Medicine, Center for Study and Research on Obesity, Milan University, Milan, Italy
8 UOC SMEL-1, Scuola di Specializzazione di Patologia Clinica e Biochimica Clinica, Università degli Studi di Milano, Milan, Italy
9 Neurosurgery Unit, Department of Surgery, ASST Lariana-S. Anna Hospital, Como, Italy
10 Department of Public Health, Experimental and Forensic Medicine, Pavia University, Pavia, Italy
*Corresponding author. Tel: +39 0255 033874; E-mail: yvan.torrente@unimi.it

Stem Cell Research 40 (2019) 101544

Contents lists available at ScienceDirect

Stem Cell Research

journal homepage: www.elsevier.com/locate/scr

Lab resource: Stem Cell Line

Establishment of a Duchenne muscular dystrophy patient-derived induced pluripotent stem cell line carrying a deletion of exons 51–53 of the dystrophin gene (CCMi003-A)

Rovina Davide[a], Castiglioni Elisa[a], Farini Andrea[b], Bellichi Marzia[b], Gervasini Cristina[c], Paganini Stefania[d], Di Segni Marina[d], Santoro Rosaria[a], Torrente Yvan[b], Pompilio Giulio[a,e,f], Gowran Aoife[a,*]

[a] Centro Cardiologico Monzino-IRCCS, Unit of Vascular Biology and Regenerative Medicine, Milan, Italy
[b] Stem Cell Laboratory, Department of Pathophysiology and Transplantation, University of Milan, Unit of Neurology, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Centro Dino Ferrari, Milan, Italy
[c] Medical Genetics, Department of Health Sciences, Università degli Studi di Milano, Milan, Italy
[d] Laboratory of Medical Genetics, Fondazione IRCCS Ca' Grande, Ospedale Maggiore Policlinico, Milan, Italy
[e] Centro Cardiologico Monzino-IRCCS, Department of Cardiac Surgery, Centro Cardiologico Monzino IRCCS, Milan, Italy
[f] Department of Clinical Sciences and Community Health, University of Milan, Italy

A B S T R A C T

Duchenne's muscular dystrophy (DMD) is a neuromuscular disorder affecting skeletal and cardiac muscle function, caused by mutations in the dystrophin (*DMD*) gene. Dermal fibroblasts, isolated from a DMD patient with a reported deletion of exons 51 to 53 in the DMD gene, were reprogrammed into induced pluripotent stem cells (iPSCs) by electroporation with episomal vectors containing the reprogramming factors: OCT4, SOX2, LIN28, KLF4, and L-MYC. The obtained iPSC line showed iPSC morphology, expression of pluripotency markers, possessed trilineage differentiation potential and was karyotypically normal.

Resource utility

This iPSC line will be helpful to the study of disease mechanisms underlying muscular dystrophy and to screen novel compounds with potential therapeutic effects.

Resource details

Duchenne Muscular dystrophy (DMD) is an X-linked neuromuscular disorder affecting skeletal and cardiac muscle function caused by mutations in the dystrophin (*DMD*) gene (D'Amario et al., 2018). Dystrophin is localized under the sarcolemma and is connected to the dystrophin-associated protein complex (DAPC) that normally anchors the cortical actin cytoskeleton and the plasma membrane to the extracellular matrix (ECM) (Constantin, 2014). *DMD* mutations cause deficiency in full-length dystrophin protein expression (427 kDa) which lead to a general disorganization of the DAPC. Myofibers lacking dystrophin and a disorganized DAPC are sensitive to mechanical damage and have deregulated signaling which activates cell death (Farini et al., 2019; Nanni et al., 2016).

Following institutional ethical committee approval and patient informed consent, dermal fibroblasts were isolated by explant culture of a skin biopsy obtained from a 10-year-old male with DMD. To protect privacy, no identifying patient information is included. Subsequent to the skin biopsy the patient lost ambulation at age 13 years. Cardiac monitoring revealed the development of left ventricular dysfunction at 18 years. The patient's electrocardiogram while normal at biopsy age but at last exam showed increased RS in V1 and deep narrow Q waves in left precordial leads. The ejection fraction (EF) was normal at time of biopsy but had decreased to 40% when last measured by echocardiography. The shortening fraction (SF) followed a similar pattern and was 20% when last determined.

Patient fibroblasts (FBS DMD3) were reprogramed into induced pluripotent stem cells (iPSCs) by electroporation with episomal vectors encoding human L-MYC, LIN28, SOX2, KLF4, OCT3/4 and cultured under feeder-free defined conditions. Following 25 days of reprograming iPSC colonies were selected for manual isolation and expanded to P5–10.

The clonal iPSC line described in this publication was named CCMi003-A, and entered iPSC characterization by evaluating: iPSC pluripotent cell morphology (Fig. 1A) and expression of pluripotency marker SSEA4 by immunocytochemistry (Fig. 1B). FACS analyzes

* Corresponding author at: Centro Cardiologico Monzino-IRCCS, Unit of Regenerative Medicine, Milan, Italy.
E-mail address: agowran@ccfm.it (G. Aoife).

https://doi.org/10.1016/j.scr.2019.101544
Received 19 June 2019; Received in revised form 6 August 2019; Accepted 19 August 2019
Available online 20 August 2019

The American Journal of Pathology, Vol. 189, No. 2, February 2019

ELSEVIER

The American Journal of
PATHOLOGY
ajp.amjpathol.org

MUSCULOSKELETAL PATHOLOGY

Fibrosis Rescue Improves Cardiac Function in Dystrophin-Deficient Mice and Duchenne Patient–Specific Cardiomyocytes by Immunoproteasome Modulation

Andrea Farini,* Aoife Gowran,† Pamela Bella,* Clementina Sitzia,‡ Alessandro Scopece,† Elisa Castiglioni,† Davide Rovina,† Patrizia Nigro,† Chiara Villa,* Francesco Fortunato,§ Giacomo Pietro Comi,§ Giuseppina Milano,†¶ Giulio Pompilio,‖**†† and Yvan Torrente*

From the Stem Cell Laboratory, Department of Pathophysiology and Transplantation, Universitá degli Studi di Milano, Unit of Neurology, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Centro Dino Ferrari, Milan, Italy; the Unit of Vascular Biology and Regenerative Medicine,† Centro Cardiologico Monzino-IRCCS, Milan, Italy; the UOC SMEL-1,‡ Scuola di Specializzazione di Patologia Clinica e Biochimica Clinica, Università degli Studi di Milano, Milan, Italy; the Neurology Unit,§ Neuroscience Section, Department of Pathophysiology and Transplantation, Dino Ferrari Centre, IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Milan, Italy; the Laboratory of Cardiovascular Research,¶ Department of Surgery and Anesthesiology, University Hospital of Lausanne, Lausanne, Switzerland; the Unit of Vascular Biology and Regenerative Medicine,‖ the Department of Cardiac Surgery,** Centro Cardiologico Monzino-IRCCS, Milan, Italy; and the Department of Clinical Sciences and Community Health,†† University of Milan, Milan, Italy

Accepted for publication October 10, 2018.

Address correspondence to Yvan Torrente, Ph.D., M.D., Stem Cell Laboratory, Department of Pathophysiology and Transplantation, Università degli Studi di Milano, Unit of Neurology, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Centro Dino Ferrari, via F. Sforza 35, 20122 Milan, Italy. E-mail: yvan.torrente@unimi.it.

Patients affected by Duchenne muscular dystrophy (DMD) develop a progressive dilated cardiomyopathy characterized by inflammatory cell infiltration, necrosis, and cardiac fibrosis. Standard treatments consider the use of β-blockers and angiotensin-converting enzyme inhibitors that are symptomatic and unspecific toward DMD disease. Medications that target DMD cardiac fibrosis are in the early stages of development. We found immunoproteasome dysregulation in affected hearts of *mdx* mice (murine animal model of DMD) and cardiomyocytes derived from induced pluripotent stem cells of patients with DMD. Interestingly, immunoproteasome inhibition ameliorated cardiomyopathy in *mdx* mice and reduced the development of cardiac fibrosis. Establishing the immunoproteasome inhibition–dependent cardioprotective role suggests the possibility of modulating the immunoproteasome as new and clinically relevant treatment to rescue dilated cardiomyopathy in patients with DMD. *(Am J Pathol 2019, 189: 339–353; https://doi.org/10.1016/j.ajpath.2018.10.010)*

Skeletal myopathy and muscular dystrophy progression are commonly associated with cardiac dysfunctions and a consequent high mortality attributable to heart failure.[1–3] In particular, patients with Duchenne muscular dystrophy (DMD) present with early diastolic dysfunction and myocardial fibrosis that turn into a dilated cardiomyopathy, complicated by heart failure and arrhythmia.[4] Even though recent improvements in the management of respiratory insufficiency have improved the lifespan and overall prognosis of patients with DMD, sudden deaths attributable to heart failure negatively affect their quality of life. Prompt treatment and early detection of cardiomyopathy represent the requirements for successful cardioprotective therapies that block or at least slow the processes of cardiac remodeling and heart failure.[3] Unfortunately, the current treatments for dilated cardiomyopathy are still inadequate because a deep understanding of the specific mechanisms underlying DMD-attributable heart failure is

A.F. and A.G. contributed equally to this work.

Supported by Italian Ministry of Health Conte Capitale 2015 (G.P.), Fondazione Umberto Veronesi (A.G.), and Fondazione IEO CCM (D.R.).

Disclosures: None declared.

https://doi.org/10.1016/j.ajpath.2018.10.010

frontiers in Neurology

CLINICAL TRIAL
published: 23 July 2019
doi: 10.3389/fneur.2019.00755

Preliminary Evidences of Safety and Efficacy of Flavonoids- and Omega 3-Based Compound for Muscular Dystrophies Treatment: A Randomized Double-Blind Placebo Controlled Pilot Clinical Trial

Clementina Sitzia[1†], Mirella Meregalli[1], Marzia Belicchi[1], Andrea Farini[1], Maddalena Arosio[2], Denise Bestetti[3], Chiara Villa[1], Luca Valenti[4], Paolo Brambilla[5] and Yvan Torrente[1]*

[1] Stem Cell Laboratory, Unit of Neurology, Department of Pathophysiology and Transplantation, Centro Dino Ferrari, Università degli Studi di Milano, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, [2] Service of Physiotherapy, San Raffaele Scientific Institute, Milan, Italy, [3] Bianchi Bonomi Haemophilia and Thrombosis Center, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, [4] Department of Pathophysiology and Transplantation, Department of Transfusion Medicine and Hepatology, Translational Medicine, Università degli Studi di Milano, Fondazione IRCCS Ca' Granda, Milan, Italy, [5] Department of Laboratory Medicine, Desio Hospital, University Milano Bicocca, Milan, Italy

Background: Nutritional compounds can exert both anti-inflammatory and anti-oxidant effects. Since these events exacerbate the pathophysiology of muscular dystrophies, we investigated nutraceutical supplementation as an adjuvant therapy in dystrophic patients, to low costs and easy route of administration. Moreover, this treatment could represent an alternative therapeutic strategy for dystrophic patients who do not respond to corticosteroid treatment.

Objective: A 24 weeks randomized double-blind placebo-controlled clinical study was aimed at evaluating the safety and efficacy of daily oral administration of flavonoids- and omega3-based natural supplement (FLAVOMEGA) in patients affected by muscular dystrophy with recognized muscle inflammation.

Design: We screened 60 patients diagnosed for Duchenne (DMD), Facioscapulohumeral (FSHD), and Limb Girdle Muscular Dystrophy (LGMD). Using a computer-generated random allocation sequence, we stratified patients in a 2:1:1 ratio (DMD:FSHD:LGMD) to one of two treatment groups: continuous FLAVOMEGA, continuous placebo. Of 29 patients included, only 24 completed the study: 15 were given FLAVOMEGA, 14 placebo.

Results: FLAVOMEGA was well tolerated with no reported adverse events. Significant treatment differences in the change from baseline in 6 min walk distance (6MWD; secondary efficacy endpoint) ($P = 0.033$) and in isokinetic knee extension ($P = 0.039$) (primary efficacy endpoint) were observed in LGMD and FSHD subjects. Serum CK levels (secondary efficacy endpoint) decreased in all FLAVOMEGA treated groups with significant difference in DMD subjects ($P = 0.039$).

OPEN ACCESS

Edited by:
Edoardo Malfatti, INSERM U1179 Handicap neuromusculaire: Physiopathologie, Biothérapie et Pharmacologie appliquées (END-ICAP), France

Reviewed by:
Rosanna Cardani, Policlinico San Donato (IRCCS), Italy
Massimiliano Filosto, Civil Hospital of Brescia, Italy

***Correspondence:**
Yvan Torrente
yvan.torrente@unimi.it

†Present Address:
Clementina Sitzia, Residency program in Clinical Pathology and Clinical Biochemistry, University of Milan, Milan, Italy

Specialty section:
This article was submitted to Neuromuscular Diseases, a section of the journal Frontiers in Neurology

Received: *26 March 2019*
Accepted: *01 July 2019*
Published: *23 July 2019*

Citation:
Sitzia C, Meregalli M, Belicchi M, Farini A, Arosio M, Bestetti D, Villa C, Valenti L, Brambilla P and Torrente Y (2019) Preliminary Evidences of Safety and Efficacy of Flavonoids- and Omega 3-Based Compound for Muscular Dystrophies Treatment: A Randomized Double-Blind Placebo Controlled Pilot Clinical Trial. Front. Neurol. 10:755. doi: 10.3389/fneur.2019.00755