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Supplemental information

Care4Rare Canada: Outcomes from a decade of network science for rare disease gene discovery

Kym M. Boycott, Taila Hartley, Kristin D. Kernohan, David A. Dyment, Heather Howley, A. Micheil Innes, Francois P. Bernier, Michael Brudno, and Care4Rare Canada Consortium

Supplemental Information

Content

Box S1. Matchmaking ends a 19-year diagnostic odyssey by disentangling two novel rare diseases

The importance of global matchmaking for RD gene discovery, and ultimately diagnostic clarity for all affected families, cannot be overstated. In one striking example, we identified, in parallel, candidate variants in novel genes in a single individual. A baby boy presented with marked hypotonia and nystagmus shortly after birth. Brain MRI showed lack of myelination, and a likely clinical diagnosis of Pelizaeus-Merzbacher-like disease (MIM 312080) was given although *PLP1* (MIM 300401) testing was negative. A gastrostomy tube was placed at 5 months of age for a failure to thrive. Around that time, he developed intermittent episodes of choreoathetosis, initially thought to be seizures, evolving to dystonic arm posturing in his teen years. He has severe intellectual disability. Following research exome sequencing in 2015 as part of the Care4Rare project, we identified *de novo* variants in two genes that had never been associated with human disease, *TMEM106B* [exon 8; c.754G>A p.(Asp252Asn); NM_001134232.2] (MIM 613413) and *USP7* [exon 14; c.1406T>G p.(Val469Gly); NM_001286457.2] (MIM 602519). Both genes were entered into the Matchmaker Exchange data sharing platform. We matched immediately for *USP7* to an international cohort of 16 individuals with autism, intellectual disability, epilepsy, and feeding difficulties (MIM 616863).[1] No affected individuals, however, exhibited hypomyelination and so we kept our submission for *TMEM106B* active within MME. One year later, we matched to another proband with hypomyelination and the same *de novo* mutation in *TMEM106B*. Two years following our initial review of the exome data, we published a cohort of four affected individuals from Canada, Australia, Germany, and the Netherlands with the same recurrent *TMEM106B* mutation (MIM 617964).[2] Global matchmaking made it possible to disentangle the co-occurrence of two different RDs for this individual, while discovering two new RDs and characterizing their clinical spectrum, so that other families will not have to wait 19 years for diagnostic clarity. The family is now an active member of the USP7 Foundation, a non-profit organization dedicated to supporting families and research for this RD.

Box S2. Querying databases for variants in candidate genes facilitates RD gene discovery

Two French Canadian sibships from the Care4Rare project had a strikingly similar and complex clinical presentation characterized by hereditary spastic paraplegia (HSP), intellectual disability, and cerebral anomalies yet remained without a diagnosis following research exome sequencing in 2015. Re-analysis in 2020 with the latest Care4Rare bioinformatics pipeline revealed a shared homozygous candidate variant in *ABHD16A* (MIM 142620) that was missed by our earlier bioinformatics pipeline. We immediately submitted the gene to MME but no matches were returned. In 2021, we queried the data in Genomics4RD, and quickly identified two probands with rare, deleterious looking biallelic variants in *ABHD16A* and symptoms of complex HSP: one family who remained without a diagnosis and whose data had not yet been re-analyzed, and a second family whose data had just been returned from a US-based clinical laboratory. A subsequent query of rare variants in Geno2MP quickly identified two additional families from the USA. These findings add *ABHD16A* to the growing list of genes where deleterious variants cause early-onset autosomal recessive complicated HSP through the dysregulation of lipid metabolism (MIM 619735).[3] This discovery highlights the power of querying databases for variants in candidate genes in genomic data from families with unsolved RD.

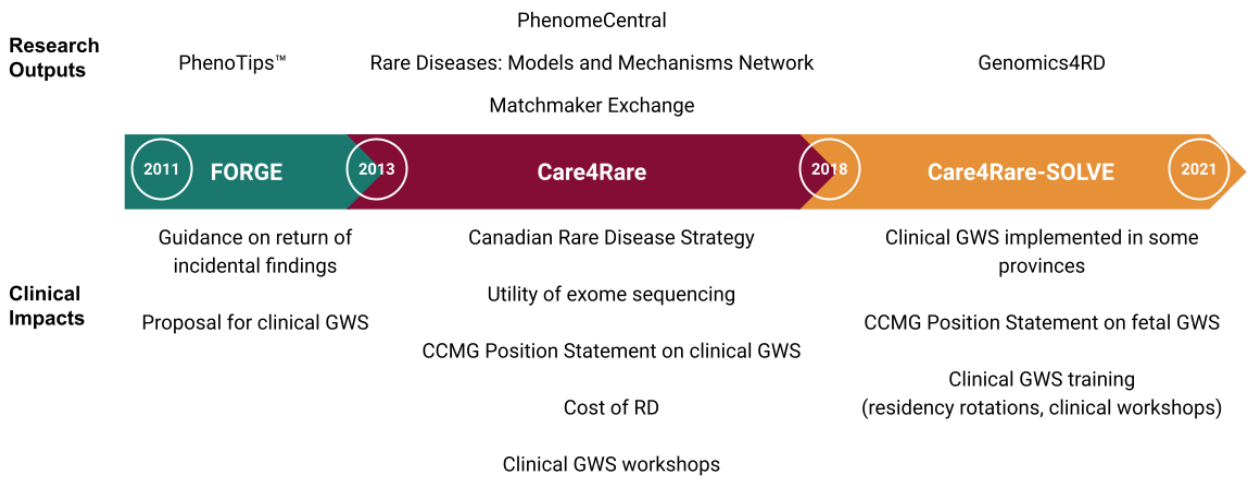

Figure S1. Research outputs and clinical impacts resulting from a decade of the Care4Rare Canada Consortium. In addition to its considerable efforts in rare disease (RD) diagnosis (n=623) and gene discovery (n=203), the Care4Rare Canada Consortium identified and responded to the needs of the RD research and clinical communities. Through all eras, the Consortium delivered key technologies, evidence, resources, and guides to support RD diagnosis and discovery. For the research community, the Care4Rare Consortium delivered technologies to support RD data capture (PhenoTips™) and sharing (PhenomeCentral, Matchmaker Exchange, Genomics4RD), as well as facilitated functional characterization research of novel genes in model organisms by establishing the Canadian Rare Diseases: Models and Mechanisms Network. For the clinical community, the Consortium (in partnership with key stakeholders) paved the way for clinical implementation of genome-wide sequencing (GWS) by publishing evidence of its diagnostic utility and the costs of RD; contributing to a national RD strategy, developing clinical guidelines and position statements; and building capacity through curriculum development and workshops. Ten years later, Canada remains an international leader in RD discovery and GWS is available as a publicly funded test for complex RD diagnosis in most regions of Canada.

Eras	FORGE	Care4Rare	SOLVE
General inclusion criteria	• Assessment by a clinical geneticist or subspecialist practicing in genetics who is a member of Care4Rare Canada Consortium • Suspected monogenic disease • Standard-of-care testing in their province/territory completed • No molecular diagnosis or compelling VUS • Consented using the Care4Rare Research Ethics Board-approved protocol • Samples available and follow-up possible • Family members available, including deep-phenotype and samples		
Population	• Children • At least one affected individual from Canada available for a particular project	Children and adults	Children and adults
Era specific criteria	• Highly recognizable syndrome • Family history of recurrence • Known consanguinity • Isolated community	FORGE criteria plus single families	Genomic data only AND/OR Multi-omics data

Table S1. Inclusion Criteria for Participation in Care4Rare across the Eras

Group	RD Characteristics	Reason(s) for Remaining Unsolved	Technologies Proposed
1	Recognizable RD with no or limited genetic heterogeneity (e.g., Tuberous sclerosis)	Pathogenic variation is undetectable with current technologies	Genome sequencing RNA sequencing +/-Deep sequencing
2	Recognizable RD where the molecular etiology has not yet been elucidated (e.g. PHACES, as reviewed by Boycott et al[4])	Molecular etiology is not yet known and is undetectable using current approaches OR Non-Mendelian inheritance OR Non-genetic	Large-scale data sharing Multi-omics Novel computational approaches
3	RD with high degree of genetic heterogeneity (e.g., cerebellar ataxia)	Pathogenic variant in a known gene that is missed by current diagnostic technologies or clinical panel OR Pathogenic variant in a novel gene	Re-analysis Genome sequencing RNA sequencing
4	RD associated with a constellation of features without a name	Phenotypic expansion of a known disease OR Novel gene-disease association OR More than one genetic diagnosis OR Non-Mendelian inheritance OR Non-genetic	Re-analysis Multi-omics Large-scale data sharing

Table S3. Overview of a Research Protocol for RDs Remaining Unsolved Following Exome Sequencing
Multi-omics refers to genome and RNA sequencing data, lipidomics, metabolomics, and/or epigenomics in any combination

Care4Rare Canada Consortium: Current and past leadership teams

FORGE Canada project: Finding of Rare Disease Genes in Canada

Kym Boycott (lead; University of Ottawa), Jan Friedman (co-lead; University of British Columbia), Jacques Michaud (co-lead; University of Montreal), Denise Avard (McGill University), Francois Bernier (University of Calgary), Michael Brudno (University of Toronto), Dennis Bulman (University of Ottawa), Bridget Fernandez (Memorial University of Newfoundland), Steven Jones (BC Cancer Agency Genome Sciences Centre), Bartha Knoppers (McGill University), Jacek Majewski (McGill University), Janet Marcadier (Children's Hospital of Eastern Ontario Research Institute), Marco Marra (BC Cancer Agency Genome Sciences Centre), Alexandre Montpetit (McGill Applied Genomics Innovation Core), Andrew Orr (Dalhousie University), Francois Rousseau (University of Laval), Mark Samuels (University of Montreal), Steven Scherer (The Centre for Applied Genomics), Richard Wintle (The Centre for Applied Genomics).

Care4Rare project: Enhanced Care for Rare Genetic Diseases in Canada

Kym Boycott (lead; University of Ottawa), Alex MacKenzie (co-lead; University of Ottawa), Genevieve Bernard (McGill University), Francois Bernier (University of Calgary), Bernard Brais (McGill University), Michael Brudno (University of Toronto), Dennis Bulman (University of Ottawa), David Dyment (University of Ottawa), Roy Gravel (University of Calgary), Micheil Innes (University of Calgary), Bartha Knoppers (McGill University), Jacek Majewski (McGill University), Deborah Marshall (University of Calgary), Christopher McMaster (Dalhousie University), Jacques Michaud (University of Montreal), Mark Samuels (University of Montreal), Barbara von Tigerstrom (University of Saskatchewan).

Care4Rare SOLVE project: Care4Rare Canada: Harnessing Multi-omics to Deliver Innovative Diagnostic Care for Rare Genetic Diseases in Canada

Kym Boycott (lead; University of Ottawa), Michael Brudno (co-lead, University of Toronto), Francois Bernier (co-lead, University of Calgary), Clara van Karnebeek (co-lead, University of British Columbia), James Dowling (University of Toronto), David Dyment (CHEO Research Institute), Robin Hayeems (University of Toronto), Micheil Innes (University of Calgary), Kristin Kernohan (University of Ottawa), Bartha Knoppers (McGill University), Ryan Lamont (University of Calgary), Anna Lehman (University of British Columbia), Christian Marshall (University of Toronto), Deborah Marshall (University of Calgary), Roberto Mendoza (University of Toronto), Sara Mostafavi (University of British Columbia), Jillian Parboosingh (University of Calgary).

Care4Rare Canada Administrative Team

Project management: Taila Hartley (Operations Director, University of Ottawa; current), Elisabeth Soubry (Project Manager, University of Ottawa; current), Meryl Akers (Project Manager, University of Toronto; current), Brenda McInnes (Project Manager, Univesity of Calgary; current), Meriam Waqas (Project Manager, University of British Columbia; current), Hanh Dao (Administrative Assistant, University of Ottawa; current), Grace Ediae (Project Manager, University of Ottawa; past), Chandree Beaulieu (Project Manager, University of Ottawa; past), Janet Marcadier (Project Manager, University of Ottawa; past).

Research Administration: Heather Howley (Research Funding Development Officer, current), Laura Minaker (Senior Finance Officer, current), Samira Chamaa (Manager of Grants and Awards, current).

Members of the Care4Rare Canada Consortium

Iman Abumansour, Nassima Addour, Sohnee Ahmed, Ebba Alkhunaizi, Judith Allanson, Kim Amburgey, Ingrid Ambus, Laura Arbour, Christine Armour, Linlea Armstrong, Taryn Athey, Setareh Ashtiani, Billie Au, Gudren Aubertin, Rita Aul, Lauren Badalato, Steven Baker, Tugce Balci, Torben Bech-Hansen, Karen Bedard, Melanie Bedford, Steffany Bennett, Samuel Berkovic, Genevieve Bernard, Priya Bhola, Neal Boerkoel, Brittney Bosse, Danielle Bourque, Sarah Bowdin, Lauren Brady, Tamara Braid, Bernard Brais, Paula Brna, Lina Buelvas, Dennis Bulman, Oana Caluseriu, Craig Campbell, Maggie Campbell, Philippe Campeau, Melissa Carter, Michelle Caudle, Inara Chacon, Lauren Chad, Pranesh Chakraborty, Erin Chamberlain, Alicia Chan, Caitlin Chang, Marisa Chard, Stephanie Chieffo, Caitlin Chisholm, David Chitayat, Tillie Chiu, Bernie Chodirker, Karen Chong, Albert Chudley, Mireille Cloutier, Ronni Cohn, Samantha Coliacovo, Ashley Collier, Melissa Cornthwaite, Patrick Cossette, Gregory Costain, Gina Cowing, Alessandra Cumming, Lauren Currie, Donna Cushing, Daniela D'Agostino, Dawn Danielson, Isabelle De Bie, Nomazulu Dlamini, Asif Doja, James Dowling, Saunya Dover, Eric Dupas, Lucie Dupuis, Sarah Dyack, David Dyment, Alison Eaton, Hanna Faghfoury, Sandra Farrell, Bridget Fernandez, Patrick Ferreira, Rachel Ferrier, Isabel Filges, Cynthia Forster-Gibson, Zoey Freedman, Jan Friedman, Patrick Frosk, Charlotte Fung, Michael Geraghty, Brenda Gerull, William Gibson, Meredith Gillespie, Jane Gillis, Cathy Gilpin, Nicole Gojska, Elaine Goh, Claire Goldsmith, Hernan Gonorazky, Sharan Goobie, Robert Gow, Sara Gracie, Gail Graham, Jane Green, Cheryl Greenberg, Eyal Grunebaum, Andrea Guerin, Elie Haddad, Farah Hassan, Elise Heon, Stacy Hewson, Christina Honeywell, Gabriella Horvath, Rebecca

Hough, Cathy Huculak, Stacey Hume, Peter Humphreys, Teresa Hunt, Alasdair Hunter, Stephanie Huynh, Michal Inbar-Feigenberg, Angela Inglis, Micheil Innes, Nada Jabado, Shailly Jain, Leslie Colvin James, Rebekah Jobling, Jack Jung, Roman Jurencak, Rita Kadida, Peter Kannu, Natalya Karp, Kathryn Keely, Kristin Kernohan, Aneal Khan, Raymond Kim, Courtney Kiss, Robert Koenekoop, Nataly Laflamme, Sylvie Langlois, Julie Lauzon, Jessica Lawrence, Joanna Lazier, Anna Lehman, Ordan Lehmann, Edmond Lemire, Gabrielle Lemire, Chumei Li, Matt Lines, Brian Lowry, Ian MacDonald, Karen MacDonald, Sara MacKay, Alex Mackenzie, Yolanda MacKinnon, Linda MacLaren, Joanna MacLean, Melissa MacPherson, Bruno Maranda, Julien Marcadier, Sandra Marles, Nicole Martin, Andre Mattman, Anna Matveev, Ashish Marwaha, Barbara McGillivray, Brenda McInnes, Margaret McKinnon, Ross McLeod, Christopher McMaster, Hugh McMillan, Kirsten Meagher, Jariwala Mehul, Roberto Mendoza, Saadet Mercimek-Andrews, Leanne Mercer, Wendy Meschino, Aziz Mhanni, Jacques Michaud, Kamiar Mireskandari, Burcin Morali, Chantal Morel, Stephen Mosca, Alexio Muise, Jillian Murphy, Mitzi Murray, Melanie Napier, Marjan Nezarati, Karen Niederhoffer, Sarah Nikkel, Graeme Nimmo, Malgorzata Nowaczyk, Laura Nunez, Margaret O'Riley, Andrew Orr, Matthew Osmond, Sandhya Parkash, Millan Patel, Melanie Paterson, Larissa Peck, Lynette Penny, Helene Perras, Renee Perrier, Anne Pham-huy, Chitra Prasad, Magda Price, Josee Provost, Julian Raiman, Julie Richer, Christie Riddell, Carleigh Robertson, Marie-Eve Robinson, Johane Robitaille, Maian Roifman, Samantha Rojas, David Rosenblatt, Alison Rusnak, Maha Saleh, Mark Samuels, Parkash Sandhya, Sarah Sawyer, Andreas Schulze, Kim Seath, Sheiva Shaari, Priyana Sharma, Eric Shoubridge, Komudi Siriwardena, Sandra Sirrs, Victoria Siu, David Skidmore, Carter Snead, Renata Sobiesiak, Neal Sondheimer, Taylor Speziale, Rebecca Sparkes, Myriam Srour, Sylvia Stockler, Julia Su, Oksana Suchowersky, Anna Szuto, Marta Szybowska, Julia Tagoe, Mark Tarnopolsky, Deborah Terespolsky, Maryann Thomas, Alan Tinmouth, Eva Tomiak, Clara van Karnebeek, Anthony Vandersteen, Kalene Van Engelen, Lea Velsher, Sunita Venkateswaran, Krista Vincent, Jagdeep Walia, Karin Wallace, Jodi Warman, Jonathan Wasserman, Rosanna Weksberg, Grace Yoon, Dana Young, Andrea Yu, Farah Zahir, Jessica Zambonin

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Web Resources

Justin's Odyssey: A journey in rare disease data sharing - YouTube, https://www.youtube.com/watch?v=IDEhdzvlR4I)

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