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Authors
Sandy, Natascha S
Huysentruyt, Koen
Mulder, Daniel J
et al.

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The Diverse Phenotype of Intestinal Dysmotility Secondary to ACTG2-related Disorders

Natascha S. Sandy[*,†], **Koen Huysentruyt**[*,†,‡], **Daniel J. Mulder**[*], **Neil Warner**[*], **Karen Chong**[§], **Chantal Morel**[‖], **Saleh AlQahtani**[*], **Paul W. Wales**[†,¶], **Martin G. Martin**[#], **Aleixo M. Muise**[*], **Yaron Avitzur**[*,†]

[*]Division of Gastroenterology, Hepatology and Nutrition, The Hospital for Sick Children, University of Toronto, Toronto, Ontario, Canada

[†]Group for Improvement of Intestinal Function and Treatment (GIFT), The Hospital for Sick Children, University of Toronto, Toronto, Ontario, Canada

[‡]Department of Pediatric Gastroenterology, Universitair Ziekenhuis Brussel, vrije Universiteit Brussel (vUB), Brussels, Belgium

[§]The Prenatal Diagnosis and Medical Genetics Program. Mount Sinai Hospital, Toronto, ON

[‖]Cancer Clinical Research Unit (CCRU), Princess Margaret Cancer Centre, The Hospital for Sick Children, University ofToronto, Toronto, Ontario, Canada

[¶]Division of general and thoracic Surgery, The Hospital for Sick Children, University ofToronto, Toronto, Ontario, Canada

Division of Gastroenterology and Nutrition, Department of Pediatrics, Mattel Children's Hospital and the David Geffen School of Medicine, University of California Los Angeles, Los Angeles, CA, USA

Abstract

Background and Aims: The initial description of a heterozygous dominant *ACTG2* variant in familial visceral myopathy was followed by the identification of additional variants in other forms of intestinal dysmotility disorders. we aimed to describe the diverse phenotype of this newly reported and rare disease.

Methods: Report of 4 new patients, and a systematic review of *ACTG2*-related disorders. we analyzed the population frequency and used in silico gene damaging predictions. Genotype-phenotype correlations were explored.

Address correspondence and reprint requests to Yaron Avitzur, MD, Division of Gastroenterology, Hepatology & Nutrition, Hospital for Sick Children, 555 University Avenue, Toronto, ON M5G 1X8, Canada (yaron.avitzur@sickkids.ca).

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Results: One hundred three patients (52% girls), from 14 publications, were included. Twenty-eight unique variants were analyzed, all exceedingly rare, and 27 predicted to be highly damaging. The median Combined Annotation Dependent Depletion (CADD) score was 29.2 (Interquartile range 26.3–29.4). Most patients underwent abdominal surgery (66%), about half required intermittent bladder catheterization (48.5%), and more than half were parenteral nutrition (PN)-dependent (53%). One-quarter of the patients died (25.7%), and 6 required transplant (5.8%). Girls had a higher rate of microcolon ($P = 0.009$), PN dependency ($P = 0.003$), and death/transplant ($P = 0.029$) compared with boys, and early disease onset (<2 years of age) was associated with megacystis-microcolon-intestinal hypoperistalsis syndrome (MMIHS) features. There was no statistical association between disease characteristics and CADD scores.

Conclusions: Damaging *ACTG2* variants are rare, often associated with MMIHS phenotype, and overall have a wide phenotypic variation. Symptoms usually present in the perinatal period but can also appear at a later age. The course of the disease is marked by frequent need for surgical interventions, PN support, and mortality. Poor outcomes are more common among girls with *ACTG2* variants.

Keywords

ACTG2; dysmotility; megacystis-microcolon-intestinal hypoperistalsis; visceral myopathy

The ACTG2 protein (Actin Gamma 2, Smooth Muscle), encoded by a gene that carries the same name, is an evolutionarily conserved and structurally complex protein. It is a member of the actin family, and makes up a critical component of the cytoskeleton of enteric smooth muscle cells. Intestinal dysmotility secondary to *ACTG2* mutation was first reported by Lehtonen et al (1) in 2012 in a large Finnish family with a familial visceral myopathy (FVM), with 8 adults carrying a heterozygous *ACTG2* R148S variant, with an autosomal dominant inheritance, and a striking phenotypic heterogeneity: 3 subjects died, 1 remained dependent on parenteral nutrition (PN), and was listed for small bowel transplantation, whereas the remainder had mild-moderate symptoms and were orally fed. An insight into the molecular mechanism of *ACTG2* disorder was also provided: aberrant actin filaments appeared to aggregate in abnormal intracellular inclusion bodies in the small bowel, esophagus, and bladder of affected patients, leading to reduced muscle contractility (1).

This initial description was followed by the identification of additional *ACTG2* variants in other forms of visceral myopathies, such as megacystis-microcolon-intestinal hypoperistalsis syndrome (MMIHS) (2,3) and Prune belly syndrome (3). It became evident that *ACTG2* variants underly the genetics in an important subset of patients with chronic intestinal pseudoobstruction (CIPO) (4) and dysmotility disorders, particularly in pediatric intestinal pseudoobstruction (PIPO). To date, the diverse phenotype of *ACTG2*-related disorders remains relatively ill-defined and is limited to case reports and small case series. In this context, we aimed to conduct a systematic review of the literature to describe the full spectrum of the *ACTG2* mutation-associated phenotype, to explore genotype-phenotype correlations and to report additional 4 cases, including a novel *ACTG2* variant.

METHODS

We report 4 unrelated cases of patients with CIPO and *ACTG2* mutations followed at our centers. All methods were carried out in accordance with institutions' Research Ethical Board (REB) guidelines and regulations.

Systematic Review

A systematic review on ACTG2-related disorders was conducted and aligned with the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) guidelines (5,6). The protocol was registered on PROSPERO International Prospective Register of Systematic Reviews database (CRD42020177543). The protocol and search strategy can be found at: https://www.crd.york.ac.uk/PROSPEROFILES/177543_PROTOCOL_20200421.pdf.

Original articles describing an *ACTG2* variant(s) and reporting on genetic testing and clinical course were considered eligible. Meeting abstracts, letters to the editor, review papers, and non-English manuscripts were excluded. Two authors (N.S.S. and K.H.) screened the titles and abstracts independently and in duplicate to assess for eligibility. This screening was blinded and performed via Rayyan QCRI application (7). Whenever there was insufficient information available in the abstract, the full text was reviewed. Full-text articles were assessed for the final selection: publications with potential overlapping reports were thoroughly analyzed and data from the most detailed publication was included; finally studies in which available information included less than 50% of available fields for the qualitative synthesis, were excluded.

Relevant information was independently abstracted by the 2 reviewers using a structured case report form. Any discrepancies in study selection or data extraction were resolved through discussion and consensus, and whenever necessary, with the senior author's assistance (Y.A.).

Genetic Analysis

To characterize the genetic impact of the reported *ACTG2* variants, the following in silico damaging predictions were obtained: Combined Annotation Dependent Depletion (CADD) Phred scores (version 1.6) (8,9), minor allele frequencies (from gnomAD version 2.1.1) (10), and a subset of scores (15 scores that provide binary predictions) from the database for Nonsynonymous SNPs' Functional Predictions (dbNSFP version 4.1a) (11,12): DEOGEN2, FATHMM, FATHMM-MKL, FATHMM-XF, LRT, M-CAP, MetaLR, MetaSVM, MutationTaster2, Mutation Assessor, Polyphen2-HDIV, Polyphen2-HVAR, PrimateAI, PROVEAN, and SIFT4G. Variants were summarized using GRCh 38 coordinates. If less than 12 of the dbNSFP scores were not provided for a variant, a score was not reported. The summary of these binary scores and CADD Phred scores is provided in Table 1, Supplemental Digital Content, http://links.lww.com/MPG/C683.

Data Analysis and Synthesis

After an initial qualitative review of all references, a broad number of clinical variables were selected. This was followed by a quantitative review of those variables. Variables were included in the final analysis if they were consistently reported in more than 50% of the publications. The following selected disease characteristics were analyzed: early onset (defined as disease onset before 2 years of age), microcolon, megabladder, need for intermittent bladder catheterization, surgery in a lifetime; selected outcomes were PN dependency, intestinal transplant (IT) or death. Surgery in a lifetime was defined as one of the following procedures: exploratory laparotomy, lysis of adhesions, gastrointestinal (GI) tract resection (stomach, small bowel, or colon), creation and revision of a stoma, insertion of a feeding tube, surgery for volvulus or malrotation, and gallbladder surgery. Central line placement was not included in this category.

Descriptive statistics were used to summarize the data. Univariate analysis was performed using the Mann-Whitney-Wilcoxon test for continuous variables, and Fisher exact test was used for categorical variables (disease characteristics and outcomes). A generalized linear mixed effect modeling with a log link function (the fixed effects terms were CADD scores, and family was added as the random effects term) was used to evaluate the relationship between CADD scores and disease characteristics or outcomes. Models of interaction between CADD and gender were also tested. Lastly, disease characteristics and outcomes were directly compared according to gender and age of disease onset. All statistical tests were 2-sided, and P values <0.05 were considered statistically significant. Analyses were performed using R version 4.0.1 (R Core Team).

RESULTS

Newly Reported Cases

Case 1: a 2-year-old Caucasian boy with a history of refractory constipation starting at 1 year of age. Due to progressive abdominal distension and megabladder, he underwent a combined ileostomy and Mitrofanoff appendicovesicostomy at age 2 years. PN was initiated a month later and a gastrostomy tube was inserted for venting. Following the introduction of cisapride and omeprazole, PN was weaned after 3.5 months. Family history was significant for severe bowel dysmotility without significant bladder involvement in his mother, maternal grandmother, and great-grandmother, as well as mild-moderate constipation in his 2 siblings and uncle. Maternal obstetric history was positive for prolonged labor and life-threatening bleeding and 1 cesarean section. Whole exome sequencing (WES) was performed and revealed a novel and predicted to be damaging ACTG2 variant (c.968C>T, pPro323Leu —described PMID 32084423) in the proband and his affected mother. The variant was validated by Sanger sequencing. Target *ACTG2* testing also confirmed the presence of the variant in the 2 siblings (Fig. 1, Supplemental Digital Content, http://links.lww.com/MPG/C682). Currently, the proband is 9 years old, and fully orally fed.

Case 2: a 4-year-old South Asian girl was referred for a second opinion. She had a prenatal history of megacystis, and in infancy suffered from constipation, and urinary retention with recurrent urinary tract infections (UTIs) despite catheterization. Parents are first-degree

cousins. At 18 months of age, with worsening constipation, abdominal distension, and feeding intolerance, she underwent an ileostomy. Microcolon was present. A gastrostomy tube for feeding and venting was later placed. She had no clinical response to domperidone, or cisapride. *ACTG2* single gene testing *(Blueprint Genetics)* for ACTG2 revealed a variant of uncertain significance (VUS), c.617A>T; p.Glu206Val, predicted to be damaging by in silico tools. Her parents and older sister, unaffected, have not been tested. PN was initiated at 4 years of age following worsening in dysmotility. Currently, she is 5 years old, and requires intravenous fluids to maintain hydration.

Case 3: a Caucasian girl born at 34 weeks presented with feeding intolerance shortly after birth. Prenatally, megacystis was noted. At birth, she underwent exploratory laparotomy and was found to have a perforated appendix, malrotation, microcolon, and megabladder. Postnatally, she was also diagnosed with aortic dilatation. Despite having a gastrostomy for decompression and multiple trials of prokinetic agents (metoclopramide, domperidone, and cisapride), she remained PN-dependent and developed progressive IFALD. She underwent a multivisceral transplant at age 5 months. Bladder catheterization was required thereafter. At 22 years of age, trio WES revealed a de novo ACTG2 variant c.532C>T; p.Arg178Cy. She has 1 unaffected younger sibling and no relevant family history. Currently, she is 23 years old with good graft function.

Case 4: a Caucasian boy was referred at ages 14 years because of severe, chronic constipation. His constipation had insidious onset starting around 6 months of age but became progressively worse. Microcolon or megabladder were not present. He did not respond to promotility drugs. Later, at the age of 18 years, following the development of progressive symptoms of chronic intestinal obstruction, he underwent an ileostomy and jejunostomy tube insertion, and became PN-dependent. Multiple exploratory laparotomies for lysis of adhesions and episodes of bowel obstruction followed the initial surgery. Finally, he underwent an extensive jejunal resection (residual small bowel, 80 cm), and a colectomy followed by symptomatic relief and an increase in enteral feeds. A full-thickness biopsy from the terminal ileum showed aberrant actin expression within muscularis propria; electronic microscopy showed a reduced number of filaments in the smooth muscle cells. Duo WES revealed a c.631C>T (p.Arg2111Ter) *ACTG2* variant. He is the only child of a healthy mother, who tested negative for ACTG2 variants (biological father was not reachable). Currently, he is 23 years old and partially PN-dependent.

Literature Search Results

Sixty-one publications were identified, and 1 study was identified after the search period. Fourteen studies met the inclusion criteria (1,2,4,13–23) for quantitative synthesis (Fig. 2, Supplemental Digital Content, http://links.lww.com/MPG/C682). A total of 103 patients, including the 4 probands reported herein, with 28 unique *ACTG2* variants were included (Fig. 1). Three publications were single case reports (13,21,22), whereas the largest cohort consisted of 33 families in which a molecular diagnosis of *ACTG2* mutation was made; however, clinical data were available for 28 of these probands (23), and 10 of them had been previously described (3).

Genetic Analysis

Among the 28 *ACTG2* mutations identified (Table 1, Supplemental Digital Content, http://links.lww.com/MPG/C683), 27 were single nucleotide variants (SNVs) and 1 was a 2-base pair inframe insertion-deletion (GC>AA); 26 out of the 27 SNVs resulted in missense mutations and 1 caused an immediate stop-gain nonsense mutation. Only 1 variant was found in a large-scale genome aggregation database (gnomAD) and was exceedingly rare. Out of the 15 damaging scores used from dbNSFP that met our analysis criteria (25 of 28 variants), all the *ACTG2* mutations except 1 were predicted to be damaging by 10 or more (66%) of the scores. CADD Phred scores for the reported mutations (27 out of 28) were high: only 2 mutations scored below 20. Amongst the 103 patients included in the analysis, the median CADD score was 29.2 (IQR:26.3–29.4). A density plot of CADD score distribution in the study population is presented in Figure 3, Supplemental Digital Content, http://links.lww.com/MPG/C682. Other *ACTG2* mutations that have been reported in the ClinVar public mutation database, as well novel mutations reported after our literature search period are listed in Table 2, Supplemental Digital Content, http://links.lww.com/MPG/C683.

Clinical Characteristics

Fifty-four patients were girls (52.4%). Prenatal ultrasound findings were reported in 35.9% of patients with megacystis as the most frequent finding. Fifty-nine percentage had early onset of symptoms—before 2 years of age. A wide range of presenting symptoms beyond the typical presentation of PIPO was reported: feeding intolerance, absence of meconium passage, constipation, continuous or episodic abdominal pain, episodic vomiting and/or diarrhea, cholelithiasis, urinary retention, palpable bladder, and recurrent UTIs. Megabladder was more frequently documented than microcolon: 73.7% and 38.8%, respectively. Malrotation or volvulus was reported in 25.2% of cases. Patient demographics and clinical manifestations are summarized in Table 1 and Figure 2.

Sixty-six percentage of the patients underwent abdominal surgery, with ileostomy as the most common procedure. Almost half of the patients required intermittent bladder catheterization (48.5%). Length of PN in patients who were weaned off and pharmacotherapy were not consistently reported. There were some reports of gallbladder disease: cholelithiasis and cholecystectomy, cholecystitis, and a case report of a choledochal cyst (15). A variety of other associated manifestations were reported (listed in Table 3, Supplementary Digital Content, http://links.lww.com/MPG/C683), among which are pyloric stenosis in a 12-year-old (22), aortic dilatation, and obstetric complications (15). A detailed summary of surgical procedures can be found in Table 3, Supplementary Content, http://links.lww.com/MPG/C683.

Outcomes

PN dependency was reported in 53.3% of the cases. Six patients underwent IT (5.8%), 2 were listed for IT and 26.7% died. Cause of death included: infection/sepsis, noncompliance with treatment, malnutrition, intestinal obstruction, and liver failure. The mean age of death was 18 years (median 11; IQR 0.67–34). Treatment modalities and patient outcome are summarized in Table 1.

Genotype-Phenotype Correlation and Analysis According to Gender and Age of Onset

There was no correlation between CADD score and selected disease characteristics or outcome. Further analysis using generalized linear mixed models' methodology, to account for the nested (multiple observations within a family) structure of the data also did not reveal genotype-phenotype correlation. On the other hand, analysis according to gender, showed a statistically significant difference in rates of presence of microcolon (26% vs 48%, $P = 0.009$), PN dependency (60% vs 91%, $P = 003$), and transplant or death (19% vs 40%, $P = 0.029$) suggesting a more severe disease phenotype in girls (Table 2). Finally, when patients were stratified according to disease onset, comparing those with onset before 2years of age (61 patients) to later onset (21 patients), a statistically significant difference in rates of presence of megacystis (90% vs 28%, $P < 0.001$), presence of microcolon (48% vs 0%, $P < 0.001$), surgery in a lifetime (85% vs 48%, $P = 0.002$), and need for intermittent bladder catheterization (78% vs 5%, $P < 0.001$) was noted, with higher rates in patients with early disease onset (Table 2). Of note, 21 patients (20.3%) were not included in this latter analysis, as the age of disease onset was not available.

DISCUSSION

Genetic etiologies for pediatric CIPO have been described in sporadic reports over the last decade, highlighting the importance of genetic analysis in the diagnostic process of PIPO (24). These novel genes include variants not only in the *ACTG2* gene but also in other genes, such as *MYH11, FLNA, MYLK,* and *LMOD1.* Given the increasing number of CIPO cases reported to be secondary to *ACTG2* variants (23,25), and the relative lack of knowledge on the wide clinical spectrum of these disorders, we sought to describe relevant disease characteristics and outcomes through a systematic review and to explore risk factors associated with these disease traits. We noted significant variations in disease onset and characteristics, high morbidity and PN dependency, high mortality, frequent need for IT and a more severe phenotype in girls and children with an early disease onset.

Overall, the *ACTG2* variants reported were exceptionally rare, and predicted to be highly damaging to the *ACTG2* protein by in silico analysis. In addition, the gnomAD variant population database supports a strong intolerance to both missense and loss of function variants in *ACTG2,* in accordance with the type of mutations identified in this review. Milunsky et al (4) have highlighted that most pathogenic variants previously published affect arginine residues, specifically Arg178 or Arg257. A phenotype-genotype correlation has been proposed by Assia Batzir et al (23), suggesting that the recurrent arginine substitutions are the primary driver of disease burden, and proposing a severity spectrum based on the position of arginine residue affected (Arg178>Arg257>Arg40). The significant inter- and intrafamilial phenotypic variability, however, observed suggests that the presence of other risk factors that remain to be elucidated, contribute to poor outcomes. The original report of *ACTG2* mutations by *Lehtonen et al,* as well as the family of Case 1 in our study illustrate well this phenotypic variability. Wei et al (25) analyzed the influence of ethnicity and found that variants affecting Arg178 appeared to be less common in Chinese patients when compared with Caucasians.

MMIHS is often cited as the most severe form of PIPO (26). Before the identification of *ACTG2* as one of the genetic determinant of MMIHS, the role of gender was raised as a potential modifier in MMIHS: a female predominance was reported in the original description of the syndrome (27) and consistently reported afterwards (26,28). It was hypothesized that boys with MMIHS had a more severe phenotype compared with girls (29,30), although some authors questioned this observation as a possible consequence of reporting bias and underdiagnosis of MMIHS in male patients diagnosed with Prune Belly Syndrome (26). In that context, we investigated the role of gender in our systematic review. Overall, the gender distribution among patients with *ACTG2* mutation was similar, whereas microcolon, a hallmark of MMIHS, was more frequently documented in female patients. In addition, girls were more likely to become PN-dependent, die, or undergo transplant.

Although a wide range of phenotypic severity and disease course exists, the potential for severe and life-threatening *ACTG2*-related disease is well described. Whenever patients were stratified according to the age at disease onset, a statistically significant difference was seen in the rates of urinary tract involvement, microcolon,andneedforsurgeryat early onset. These differences could not be solely explained by the deleteriousness of the ACTG2 variant. Furthermore, these differences did not lead to a higher risk for a poor outcome, such as PN dependency, transplant, or death. On the other hand, PN dependency and risk of death and transplant were higher in girls. These findings and the wide phenotypic variety in families with the same mutation support the possibility that disease phenotype is influenced by other factors, such as disease modifiers (eg, female gender) and other yet to be identified factors.

Despite advances in the clinical characterization and insights into the genetics of CIPO, specific treatment strategies for this group of disorders are lacking. Therapy is essentially symptomatic with a limited therapeutic arsenal. Most studies in this systematic review did not report consistently on the medical management. A limited number of cases suggest that promotility drugs may improve symptoms and support reduction or weaning off PN. Often, the refractory nature and severity of symptoms lead to intestinal surgeries, stoma creation, intermittent bladder catheterization, PN dependency, and in selected cases, IT. Of note, a trend towards improvement in patient and graft survival in pediatric IT has been recently documented, with high-volume transplant centers reporting 5- and 10-year graft survival as high as 70% (31–33) and 65% (33), respectively. Particularly important in this context, a recent single center experience, however, showed inferior survival outcome in IT for ‘‘functional’’ intestinal failure (congenital diarrhea syndromes and/or motility disorders) compared with short bowel syndrome (33).

Limitations to our study include the possibility of selection bias in the publications of rare diseases and the fact that after the completion of this review, additional mutations have been described (25,34), highlighting the evolving data generation in *ACTG2*-related disorders. The small number of patients included in each study represents an important challenge for drawing definitive conclusions and conducting statistical analysis in some cases. Lastly, we were unable to establish a model between CADD scores and selected disease characteristics. These points emphasize the value of future research collaborations to better address genotypephenotype correlations.

CONCLUSIONS

In summary, this review shows that *ACTG2* pathogenic variants are often associated with MMIHS phenotype but a wide phenotypic variation is reported. The course of *ACTG2*-related disorders is marked by significant morbidity and high mortality. Presenting symptoms often appear in the first 2 years of life but can also appear at an older age. Genetic testing including *ACTG2* mutations should be a routine diagnostic method in pediatric or adult-onset intestinal pseudo-obstruction, and also considered in milder phenotypes, such as refractory constipation.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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REFERENCES

1. Lehtonen HJ, Sipponen T, Tojkander S, et al. Segregation of a missense variant in enteric smooth muscle actin gamma-2 with autosomal dominant familial visceral myopathy. Gastroenterology 2012;143:1482. [PubMed: 22960657]
2. Thorson W, Diaz-Horta O, Foster J, et al. De novo ACTG2 mutations cause congenital distended bladder, microcolon, and intestinal hypoperistalsis. Hum Genet 2014;133:737–42. [PubMed: 24337657]
3. Wangler MF, Gonzaga-Jauregui C, Gambin T, et al. Heterozygous de novo and inherited mutations in the smooth muscle actin (ACTG2) gene underlie megacystis-microcolon-intestinal hypoperistalsis syndrome. PLoS Genet 2014;10:e1004258. [PubMed: 24676022]
4. Milunsky A, Baldwin C, Zhang X, et al. Diagnosis of chronic intestinal pseudo-obstruction and megacystis by sequencing the ACTG2 gene. J Pediatr Gastroenterol Nutr 2017;65:384–7. [PubMed: 28422808]
5. Moher D, Liberati A, Tetzlaff J, et al. , PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. PLoS Med 2009;6:e1000097. [PubMed: 19621072]
6. Liberati A, Altman DG, Tetzlaff J, et al. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate healthcare interventions: explanation and elaboration. BMJ 2009;339:b2700. [PubMed: 19622552]
7. Ouzzani M, Hammady H, Fedorowicz Z, et al. Rayyan-a web and mobile app for systematic reviews. Syst Rev 2016;5:210. [PubMed: 27919275]
8. Rentzsch P, Witten D, Cooper GM, et al. CADD: predicting the deleteriousness of variants throughout the human genome. Nucleic Acids Res 2019;47:D886–94. [PubMed: 30371827]
9. Kircher M, Witten DM, Jain P, et al. A general framework for estimating the relative pathogenicity of human genetic variants. Nat Genet 2014;46:310–5. [PubMed: 24487276]
10. Karczewski KJ, Francioli LC, Tiao G, et al. The mutational constraint spectrum quantified from variation in 141,456 humans. Nature 2020;581:434–43. [PubMed: 32461654]

11. Liu X, Wu C, Li C, et al. dbNSFP v3.0: a one-stop database of functional predictions and annotations for human nonsynonymous and splice-site SNVs. Hum Mutat 2016;37:235–41. [PubMed: 26555599]
12. Dong C, Wei P, Jian X, et al. Comparison and integration of deleteriousness prediction methods for nonsynonymous SNVs in whole exome sequencing studies. Hum Mol Genet 2015;24:2125–37. [PubMed: 25552646]
13. Holla OL, Bock G, Busk OL, et al. Familial visceral myopathy diagnosed by exome sequencing of a patient with chronic intestinal pseudo-obstruction. Endoscopy 2014;46:533–6. [PubMed: 24777424]
14. Tuzovic L, Tang S, Miller RS, et al. New insights into the genetics of fetal megacystis: ACTG2 mutations, encoding gamma-2 smooth muscle actin in megacystis microcolon intestinal hypoperistalsis syndrome (Berdon Syndrome). Fetal Diagn Ther 2015;38:296–306. [PubMed: 25998219]
15. Klar J, Raykova D, Gustafson E, et al. Phenotypic expansion of visceral myopathy associated with ACTG2 tandem base substitution. Eur J Hum Genet 2015;23:1679–83. [PubMed: 25782675]
16. Moreno CA, Metze K, Lomazi EA, et al. Visceral myopathy: clinical and molecular survey of a cohort of seven new patients and state of the art of overlapping phenotypes. Am J Med Genet A 2016;170:2965–74. [PubMed: 27481187]
17. Matera I, Rusmini M, Guo Y, et al. Variants of the ACTG2 gene correlate with degree of severity and presence of megacystis in chronic intestinal pseudo-obstruction. Eur J Hum Genet 2016;24:1211–5. [PubMed: 26813947]
18. Lu W, Xiao Y, Huang J, et al. Mutation in actin gamma-2 responsible for megacystis microcolon intestinal hypoperistalsis syndrome in 4 Chinese patients. J Pediatr Gastroenterol Nutr 2016;63:624–6. [PubMed: 27007401]
19. Halim D, Hofstra RMW, Signorile L, et al. ACTG2 variants impair actin polymerization in sporadic megacystis microcolon intestinal hypoperistalsis syndrome. Hum Mol Genet 2016;25:571–83. [PubMed: 26647307]
20. Ravenscroft G, Pannell S, O'Grady G, et al. Variants in ACTG2 underlie a substantial number of Australasian patients with primary chronic intestinal pseudo-obstruction. Neurogastroenterol Motil 2018;30:e13371. [PubMed: 29781137]
21. Korgali EU, Yavuz A, Simsek CEC, et al. Megacystis microcolon intestinal hypoperistalsis syndrome in which a different de novo Actg2 gene mutation was detected: a case report. Fetal Pediatr Pathol 2018;37:109–16. [PubMed: 29608093]
22. Collins RRJ, Barth B, Megison S, et al. ACTG2-associated visceral myopathy with chronic intestinal pseudoobstruction, intestinal malrotation, hypertrophic pyloric stenosis, choledochal cyst, and a novel missense mutation. Int J Surg Pathol 2019;27:77–83. [PubMed: 30019982]
23. Assia Batzir N, Kishor Bhagwat P, Larson A, et al. Recurrent arginine substitutions in the ACTG2 gene are the primary driver of disease burden and severity in visceral myopathy. Hum Mutat 2020;41:641–54. [PubMed: 31769566]
24. Thapar N, Saliakellis E, Benninga MA, et al. Paediatric intestinal pseudo-obstruction: evidence and consensus-based recommendations from an ESPGHAN-led expert group. J Pediatr Gastroenterol Nutr 2018;66:991–1019. [PubMed: 29570554]
25. Wei Z, Lu L, Zheng Y, et al. Variants in the enteric smooth muscle actin (−2 cause pediatric intestinal pseudo-obstruction in Chinese patients. J Pediatr Gastroenterol Nutr 2020;72:36–42.
26. Gosemann JH, Puri P. Megacystis microcolon intestinal hypoperistalsis syndrome: systematic review of outcome. Pediatr Surg Int 2011;27:1041–6. [PubMed: 21792650]
27. Berdon WE, Baker DH, Blanc WA, et al. Megacystis-microcolon-intestinal hypoperistalsis syndrome: a new cause of intestinal obstruction in the newborn. Report of radiologic findings in five newborn girls. AJR Am J Roentgenol 1976;126:957–64. [PubMed: 178239]
28. Puri P, Shinkai M. Megacystis microcolon intestinal hypoperistalsis syndrome. Semin Pediatr Surg 2005;14:58–63. [PubMed: 15770589]
29. Köhler M, Pease PW, Upadhyay V. Megacystis-microcolon-intestinal hypoperistalsis syndrome (MMIHS) in siblings: case report and review of the literature. Eur J Pediatr Surg 2004;14:362–7. [PubMed: 15543490]

30. Young ID, McKeever PA, Brown LA, et al. Prenatal diagnosis of the megacystis-microcolon-intestinal hypoperistalsis syndrome. J Med Genet 1989;26:403–6. [PubMed: 2661823]

31. Venick RS, Duggan E, Whatley J. Current status of pediatric intestinal transplantation in the United States. Curr Opin Organ Transplant 2020;25:201–7. [PubMed: 32073484]

32. Raghu VK, Beaumont JL, Everly MJ, et al. Pediatric intestinal transplantation: analysis of the Intestinal Transplant Registry. Pediatr Transplant 2019;23:e13580. [PubMed: 31531934]

33. Kara Balla A, Elsabbagh A, Khan KM, et al. Factors associated with 5- and 10-year survival after intestinal transplantation in infants and children. J Pediatr Gastroenterol Nutr 2020;71:617–23. [PubMed: 33093368]

34. Prathapan KM, King DE, Raghu VK, et al. Megacystis microcolon intestinal hypoperistalsis syndrome: a case series with long-term follow-up and prolonged survival. J Pediatr Gastroenterol Nutr 2021;72:e81–5. [PubMed: 33264186]

What Is Known

- Genetic defects in *ACTG2* cause visceral myopathy.
- *ACTG2* variants are rare and associated with intestinal dysmotility or chronic intestinal pseudo-obstruction.
- Case reports and case series suggest autosomal dominant inheritance.

What is New

- *ACTG2*-related disorders have marked phenotypic variation, variable age of onset, and high morbidity and mortality.
- Poor outcome is associated with a female gender and an early disease onset.
- Genetic testing is a helpful diagnostic method in dysmotility disorders and should include the *ACTG2* gene.

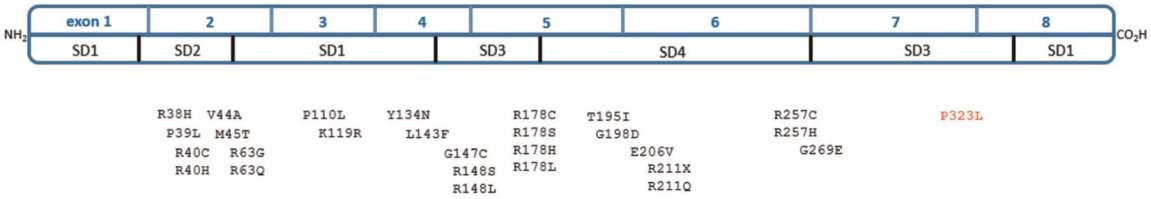

FIGURE 1.

Schematic representation of human gamma enteric smooth muscle actin, a protein encoded by 8 coding exons of the *ACTG2* gene. The solved protein structure of ACTG2 folds into 4 distinct subdomains (SD1, SD2, SD3, and SD4). The positions of heterozygous dominant mutations known to cause chronic intestinal pseudo-obstruction are shown using the NCBI Ref Seq ID NP_001606.1 amino acid numbering. A novel Proline to Leucine variant at amino acid position 323 within subdomain3 first reported in this article is highlighted in red.

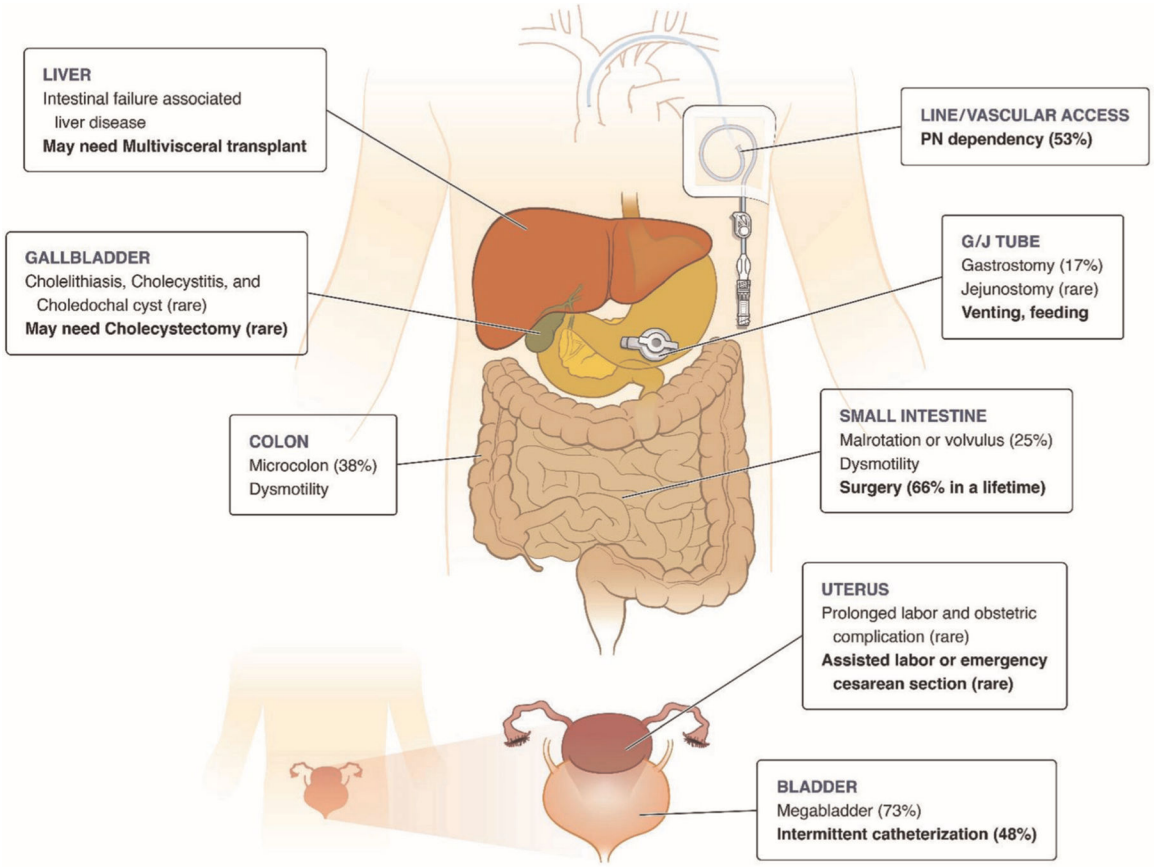

FIGURE 2.

Manifestations of ACTG2-related disorders and possible treatments. The frequency of reported clinical characteristics and interventions found in the systematic review is reported in brackets.

TABLE 1.

Characterization of patients with ACTG2-related disorders: demographics and clinical manifestations, treatment modalities and outcomes

Demographics and clinical manifestations							
Study, year	**Cases**	**Female gender**	**Prenatal findings**	**Early onset**	**Microcolon**	**Mega-bladder**	**Malrotation or volvulus**
Lehtonen et al, 2012	8	3/8	0/8	0/8	0/8	0/8	0/8
Holla et al, 2014	1	1/1	0/1	0/1	0/1	0/1	1/1
Thorson et al, 2014	2	1/2	1/2	2/2	1/2	2/2	2/2
Klar et al, 2015	11	5/11	0/8	5/11	0/11	8/11	7/11
Tuzovic et al, 2015	4	3/4	4/4	4/4	2/4	4/4	0/4
Halim et al, 2016	8	7/8	7/8	7/8	8/8	8/8	5/8
Lu et al, 2016	4	2/4	2/4	4/4	4/4	3/7	0/1
Matera et al, 2016	10	7/10	7/10	2/10	3/10	8/10	6/10
Moreno et al, 2016	4	2/4	4/4	3/4	2/4	4/4	0/4
Milunsky et al, 2017	7	3/7	3/7	7/7	0/7	7/7	2/7
Korgali et al, 2018	1	1/1	1/1	1/1	1/1	1/1	0/1
Ravenscroft et al, 2018	10	2/10	2/10	6/10	1/10	5/10	0/10
Collins et al, 2019	1	0/1	0/1	1/1	0/1	0/1	1/1
Assia Batzir et al, 2020	28*	15/28	4/28	15/28	16/28	23/28	2/28
Present study	4	2/4	1/4	4/4	2/4	3/4	0/4
Total (%)	**103**	**54 (52.4%)**	**37 (35.9%)**	**61 (59.2%)**	**40 (38.8%)**	**76 (73.7%)**	**26 (25.24%)**

Treatment modalities and outcomes							
Study, year]	**Cases**	**Pharmacotherapy**	**Surgery in lifetime**	**Bladder catheterization**	**PN dependency**	**Death**	**Transplant (Tx)**
Lehtonen et al, 2012	8	NR	3/8	0/8	2/8	3/8	1/8 Listed
Holla et al, 2014	1	Metronidazole, cisapride, pancreatic enzymes	1/1	0/1	Not reported	0/1	No
Thorson et al, 2014	2	NR	2/2	2/2	1/2	0/2	1/2 MVTx
Klar et al, 2015	11	NR	5/11	NR	NR	4/11	NR
Tuzovic et al, 2015	4	NR	2/4	4/4	3/4	0/4	NR
Halim et al, 2016	8	NR	5/8	1/8	1/8	7/8	1/8 MVTx
Lu et al, 2016	4	Itopride (1)	3/4	0/4	1/4*	1/4	NR
Matera et al, 2016	10	NR	10/10	3/10	5/10	3/10	1/10 MVTx

Demographics and clinical manifestations							
Study, year	**Cases**	**Female gender**	**Prenatal findings**	**Early onset**	**Microcolon**	**Mega-bladder**	**Malrotation or volvulus**
Moreno et al, 2016	4	NR	4/4	3/4	4/4	2/4	NR (4)
Milunsky et al, 2017	7	NR	5/7	6/7	4/7	3/4	1/7 MVTx
Korgali et al, 2018	1	NR	1/1	1/11	1/1	1/1	0/1
Ravenscroft et al, 2018	10	NR	7/10	3/10	5/10	1/9, 1 TOP	Not reported
Collins et al, 2018	1	Antibiotics, pyridostigmine	1/1	0/1	1/1	0/1	0/1
Assia Batzir et al, 2020	28	Metoclopramide (3), cisapride (5), erythromycin (1)	15/28	24/28	24/28	1/27 1 TOP	1/27 Listed 1/27 MVTx
Present study	4	Cisapride (2), prucalopride (1), octreotide (1), antibiotics (2)	4/4	2/4	3/4	0/4	1/4 MVTx
Total, %	**103**	**NA (descriptive)**	**68 (66%)**	**50 (48.5%)**	**55 (53.3%)**	**26/101 (25.7%) + 2 TOP**	**6 underwent Tx, 2 listed**

Prenatal findings mainly consisted of megacystis and urinary tract dilatation. All documented prenatal findings are detailed in Table 3, Supplementary Digital Content, http://links.lww.com/MPG/C683. Infancy onset was defined as disease onset before 2years of age. MVTx = multivisceral transplant, NR = not reported, TOP = termination pregnancy.

*Assia Batzir et al identified 33 probands but clinical information was available for 28 of those. Including also data from Wangler et al, 2014. The bold font highlights significant p-values.

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TABLE 2.

Disease characteristics and outcomes by gender and by age at disease onset

	Male, *N* = 49	**Female, *N* = 54**	***P* value**
Megacystis	32/44 (73%)	44/53 (83%)	0.3
Microcolon	11/38 (26%)	29/50 (48%)	**0.009**
Infancy onset	27/41 (66%)	34/41 (83%)	0.128
Surgery	31/44 (70%)	37/45 (84%)	0.219
BC	22/46 (52%)	28/46 (61%)	0.295
PN dependency	24/40 (60%)	31/34 (91%)	**0.003**
Transplant or Death	9/48 (19%)	21/53 (40%)	**0.029**
CADD, median (IQR)	29.20 (28.10–30.70)	28.10 (26.20–29.20)	**0.023**
	Early onset*, N = 61	**Late onset**, N = 21**	
Megacystis	54/60 (90%)	5/18 (28%)	**<0.001**
Microcolon	25/52 (48%)	0/17 (0%)	**<0.001**
Surgery	51/60 (85%)	10/21 (48%)	**0.002**
BC	42/54 (78%)	1/20 (5.0%)	**<0.001**
PN dependency	35/47 (74%)	6/12 (50%)	0.2
Transplant or Death	17/59 (29%)	7/21 (33%)	0.8
CADD, median (IQR)	29.20 (26.20, 29.40)	28.10 (28.10, 32.00)	0.5

Fisher exact test used for categoric outcomes. Mann-Whitney-Wilcoxon test for continuous variable. BC = intermittent bladder catheterization, PN = parenteral nutrition.

* Early disease onset defined by onset of symptoms under the age of 2 years.

** Late disease onset defined by onset of symptoms above the age of 2 years. The bold font highlights significant p-values.