ANIMAL GENETICS Immunogenetics, Molecular Genetics and Functional Genomics

SHORT COMMUNICATION

doi: 10.1111/age.12825

A homozygous *ADAMTS2* nonsense mutation in a Doberman Pinscher dog with Ehlers Danlos syndrome and extreme skin fragility

J. A. Jaffey*, G. Bullock†, E. Teplin‡, J. Guo†, N. A. Villani†, T. Mhlanga-Mutangadura†, R. D. Schnabel§, L. A. Cohn¶ and G. S. Johnson†

*Department of Small Animal Medicine and Surgery, Midwestern University, Glendale, 85308, AZ, USA. †Department of Veterinary Pathobiology, University of Missouri, Columbia, 65211, MO, USA. ‡South Willamette Veterinary Clinic, 55 Emerald Parkway, Creswell, 97426, OR, USA. §Division of Animal Sciences, University of Missouri, Columbia, 65211, MO, USA. ¶Department of Veterinary Medicine and Surgery, University of Missouri, Columbia, 65211, MO, USA.

Summary

An eight-week old Doberman Pinscher was diagnosed with Ehlers Danlos syndrome based on the dog's hyper-mobile carpal, tarsal and stifle joints and abnormal skin. The skin was loose and hyper-elastic with several wounds and large atrophic scars. The dog was euthanized after a severe degloving injury from minimal trauma. A whole-genome sequence, generated with DNA from the dog's blood, contained a rare, homozygous C-to-T transition at position 2408978 on chromosome 11. This transition is predicted to alter the *ADAMTS2* transcript (*ADAMTS2*:c.769C>T) and encode a nonsense mutation (p.Arg257-Ter). Biallelic *ADAMTS2* mutations have caused a type of Ehlers Danlos syndrome known as dermatosparaxis in other species.

Keywords connective tissue disease, dermatosparaxis, whole genome sequence

Hereditary connective-tissue disorders that feature tissue fragility, skin hyper-extensibility and articular hyper-mobility are classified as types of Ehlers Danlos syndrome (EDS) (Malfait *et al.* 2017). Dermatosparaxis is a recessively inherited type of EDS. Individuals with dermatosparaxis exhibit extreme skin fragility and a characteristic ultrastructure in which cross-sections of collagen fibrils from the dermis have a distinct hieroglyphic-like appearance (Holbrook & Byers 1982; Brady *et al.* 2017). Dermatosparaxis was first described in cattle (O'Hara *et al.* 1970) and subsequently diagnosed in sheep, cats and dogs (Fjolstad & Helle 1974; Holbrook *et al.* 1980; Holbrook & Byers 1982). The first descriptions of human dermatosparaxis appeared in 1992 (Nusgens *et al.* 1992; Smith *et al.* 1992). Dermatosparaxis results from a deficiency of procollagen I N-proteinase, the enzyme that excises the N-terminal propeptide from the α-chains of structural collagens (Lenaers *et al.* 1971). Removal of the N-terminal propeptides allows mature trimeric collagen α-chains to assemble into highly organized, fully functional fibrils that provide tensile strength to skin and other tissues (Bekhouche & Colige 2015). In 1999, homozygous truncating mutations in *ADAMTS2* were reported to cause bovine and human dermatosparaxis (Colige *et al.* 1999). Since then, additional homozygous or compound heterozygous *ADAMTS2* variants have been identified as causes of human dermatosparatic EDS (Van Damme *et al.* 2016). In addition, homozygous *ADAMTS2* mutations have been reported to cause dermatosparaxis in sheep (Zhou *et al.* 2012; Monteagudo *et al.* 2015).

We received the clinical history and a blood sample from the subject, an 8-week-old male Doberman Pinscher that was presented to South Willamette Veterinary Clinic for evaluation of cutaneous wounds. The physical examination identified pain, hyper-mobility and moderate effusion in the carpal, tarsal and stifle joints. In addition, bilateral ocular chemosis and elevation of the nictitating membranes were noted. The skin had several wounds in various stages of healing, and several small, atrophic scars from previous wounds that had healed by secondary intention were apparent. The ventral abdomen had a fresh linear 6-cm-long wound. The skin was noticeably loose and hyper-elastic (Fig. 1).

The subject had four littermates. Two were deceased at parturition and one appeared healthy but unexpectedly died under sedation for a routine ear cropping. The other sibling was euthanized because of severe skin fragility, persistent wounds and painful joints. The sire and dam were clinically unremarkable.

Address of correspondence

G. S. Johnson, Veterinary Pathobiology, University of Missouri, 201 Connaway Hall, Columbia, MO, USA.
E-mail: JohnsonGS@missouri.edu

Accepted for publication 16 April 2016

Figure 1 The subject with lacerations and atrophic scars (white arrows) and with hyper-extensible skin (black arrow).

Since birth, the subject had developed many lacerations caused by minimal trauma. The joint effusion and pain were intermittent. Because of the numerous wounds at the time of evaluation, the dog was treated with tramadol and amoxicillin/clavulanic acid. The wounds were cleaned, and a wrap was applied to the laceration along the ventral abdomen. Five days later, after walking into the corner of a kitchen cabinet, the dog suffered a severe degloving injury to the skin on the dorsum and was subsequently euthanized.

DNA from the subject's blood was submitted to the University of Missouri DNA Core Facility for library construction and 2× 150-bp paired-end whole-genome sequencing in an Illumina NextSeq sequencer. Appendix S1 contains details about the analysis of the resulting 19-fold average coverage whole-genome sequence, including Sequence Read Archive accession numbers. Only one candidate for causality was identified when the variant calls were filtered to retain rare variants predicted to alter the amino acid sequence of the polypeptides encoded by 19 genes associated with human EDS (Malfait *et al*. 2017). This candidate was a homozygous C-to-T transition at position 2408978 on chromosome 11 (11:2408978C>T). The 11:2408978T allele was absent from 92 other canine whole-genome sequences in our collection. According to Ensembl annotation for canine *ADAMTS2* (ENSCAFT00000000511), the 11:2408978C>T transition is reflected in the mature *ADAMTS2* transcript (*ADAMTS2*: c.769C>T), where it converts an arginine codon to a termination codon and predicts the truncation of the encoded polypeptide (p.Arg257Ter).

Thus, we have identified a rare homozygous truncating *ADAMTS2* variant as the apparent cause of the fatal EDS in the subject, indicating that this dog had the dermatosparaxis subtype of EDS. Unfortunately, no biopsies or postmortem tissues were retained, so independent confirmation of this diagnosis is not possible. An earlier review included an example of collagen fibrils with hieroglyphic-like ultrastructure from the dermis of dog with dermatosparaxis; however, no clinical information about this dog was provided (Holbrook & Byers 1982). Other canine EDS case reports that described dogs with extremely friable skin may have been cases of .(Arlein 1947; Wall 1947) The *ADAMTS2* nonsense mutation is the first reported molecular genetic cause for canine EDS.

Acknowledgement

This study was supported in part by a Clinician Scientist Award to L.A. Cohn from the Committee on Research, College of Veterinary Medicine, University of Missouri.

References

Arlein M.S. (1947) Generalized acute cutaneous asthenia in a dog. *Journal of the American Veterinary Medical Association* **111**, 52.

Bekhouche M. & Colige A. (2015) The procollagen N-proteinases ADAMTS2, 3 and 14 in pathophysiology. *Matrix Biology*, **44–46**, 46–53.

Brady A.F., Demirdas S., Fournel-Gigleux S. *et al*. (2017) The Ehlers-Danlos syndromes, rare types. *American Journal of Medical Genetics Part C: Seminars in Medical Genetics* **175**, 70–115.

Colige A., Sieron A.L., Li S.W. *et al*. (1999) Human Ehlers-Danlos syndrome type VII C and bovine dermatosparaxis are caused by mutations in the procollagen I N-proteinase gene. *American Journal of Human Genetics* **65**, 308–17.

Fjolstad M. & Helle O. (1974) A hereditary dysplasia of collagen tissues in sheep. *Journal of Pathology* **112**, 183–8.

Holbrook K.A. & Byers P.H. (1982) Structural abnormalities in the dermal collagen and elastic matrix from the skin of patients with inherited connective tissue disorders. *Journal of Investigative Dermatology* **79**(Suppl 1), 7s–16s.

Holbrook K.A., Byers P.H., Counts D.F. & Hegreberg G.A. (1980) Dermatosparaxis in a Himalayan cat: II. Ultrastructural studies of dermal collagen. *Journal of Investigative Dermatology* **74**, 100–4.

Lenaers A., Ansay M., Nusgens B.V. & Lapiere C.M. (1971) Collagen made of extended -chains, procollagen, in genetically-defective dermatosparaxic calves. *European Journal of Biochemistry* **23**, 533–43.

Malfait F., Francomano C., Byers P. *et al*. (2017) The 2017 international classification of the Ehlers-Danlos syndromes. *American Journal of Medical Genetics Part C: Seminars in Medical Genetics* **175**, 8–26.

Monteagudo L.V., Ferrer L.M., Catalan-Insa E., Savva D., McGuffin L.J. & Tejedor M.T. (2015) *In silico* identification and three-dimensional modelling of the missense mutation in *ADAMTS2* in a sheep flock with dermatosparaxis. *Veterinary Dermatology* **26**, 49–52.

Nusgens B.V., Verellen-Dumoulin C., Hermanns-Le T., De Paepe A., Nuytinck L., Pierard G.E. & Lapiere C.M. (1992) Evidence for a relationship between Ehlers-Danlos type VII C in humans and bovine dermatosparaxis. *Nature Genetics* **1**, 214–7.

O'Hara P.J., Read W.K., Romane W.M. & Bridges C.H. (1970) A collagenous tissue dysplasia of calves. *Laboratory Investigation* **23**, 307–14.

Smith L.T., Wertelecki W., Milstone L.M., Petty E.M., Seashore M.R., Braverman I.M., Jenkins T.G. & Byers P.H. (1992) Human

dermatosparaxis: a form of Ehlers-Danlos syndrome that results from failure to remove the amino-terminal propeptide of type I procollagen. *American Journal of Human Genetics* **51**, 235–44.

Van Damme T., Colige A., Syx D. *et al.* (2016) Expanding the clinical and mutational spectrum of the Ehlers-Danlos syndrome, dermatosparaxis type. *Genetics in Medicine* **18**, 882–91.

Wall R.D. (1947) Congenital defect of the skin. *North American Veterinarian* **28**, 166–8.

Zhou H., Hickford J.G. & Fang Q. (2012) A premature stop codon in the *ADAMTS2* gene is likely to be responsible for dermatosparaxis in Dorper sheep. *Animal Genetics* **43**, 471–3.

Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.
Appendix S1 Supporting information.

