The Ehlers-Danlos Society

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Whole genome sequencing in a cat with Ehlers–Danlos syndrome.

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Highlights:

- Whole genome sequencing (WGS) was performed on a cat with Ehlers–Danlos syndrome (EDS).
- WGS identified a homozygous 3-bp deletion in *COL6A1* that is one of the collagen-encoding genes, and this variant was absent in other 194 cats' WGS database, generated by 99 Lives Cat Genome Sequencing Initiative.
- Transcription analysis of *COL6A1* confirmed an in-frame 3-bp deletion predicted to delete one residue (c.1677_1679delCAA; p.N560del).
- Although further validation is needed, our result might offer new insights into the spectrum of EDS.

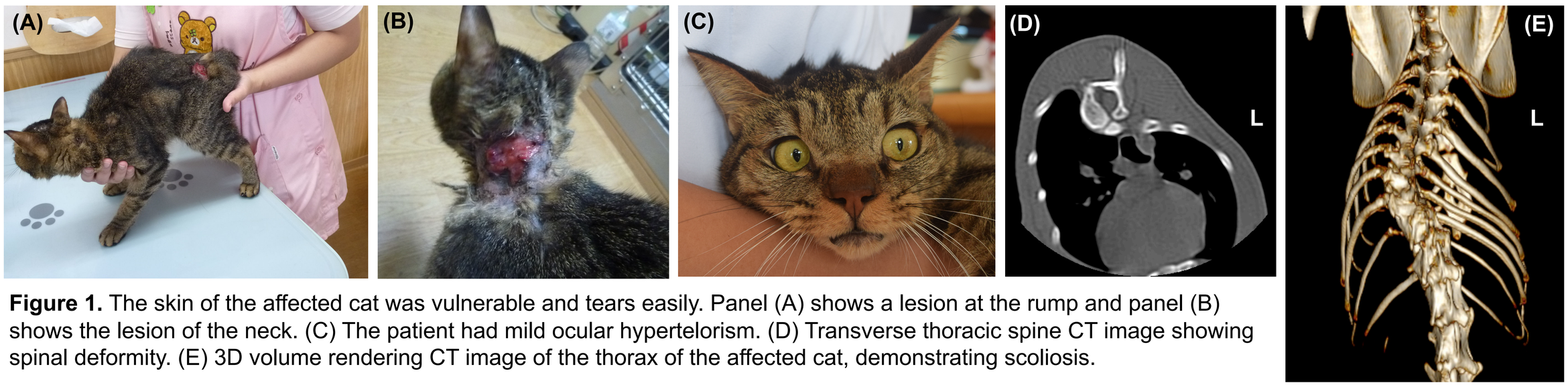

Figure 1. The skin of the affected cat was vulnerable and tears easily. Panel (A) shows a lesion at the rump and panel (B) shows the lesion of the neck. (C) The patient had mild ocular hypertelorism. (D) Transverse thoracic spine CT image showing spinal deformity. (E) 3D volume rendering CT image of the thorax of the affected cat, demonstrating scoliosis.

Case Signalment:

- Domestic short-haired cat
- An unknown age (a suspected one-year-old) (stray cat)
- Intact female
- No pedigree information

Case Presentation:

- Since **the skin was vulnerable and tears easily** (shown in Figure 1), this cat was diagnosed with EDS.
- Routine laboratory tests, such as CBC and serum chemistry, were unremarkable.
- Radiography, computed tomography (CT), and magnetic resonance imaging were performed, and **spinal deformities (scoliosis, thoracolumbar transitional vertebrae)** were identified.
- The aim of this study is to reveal the causal variant of feline EDS using WGS.

Materials and Methods / Result

Figure 2. The flow chart of the analysis.

HiSeq X Ten (illumina)

Aligned to felis catus 9.0

Feline WGS database (195 cats)

Variant filtering using VarSeq (Golden Helix)

62 unique homozygous variants

193 unique Heterozygous variants

unique variants in the collagen-encoding genes & known candidate genes for human EDS[1]

- ✓ *COL6A1* (p.N560del)
- ✓ *COL17A1* (p.A559V)
- ✓ *COL22A1* (p.M1534I)

Pathogenicity prediction by PROVEAN[2]

Only ***COL6A1* (c.1677_1679delCAA; p.N560del)** was predicted to be deleterious

A normal cat

A skin tissue sample

The EDS affected cat

Cultured fibroblast cells

RNA extraction, Reverse transcription PCR, & Sanger sequencing

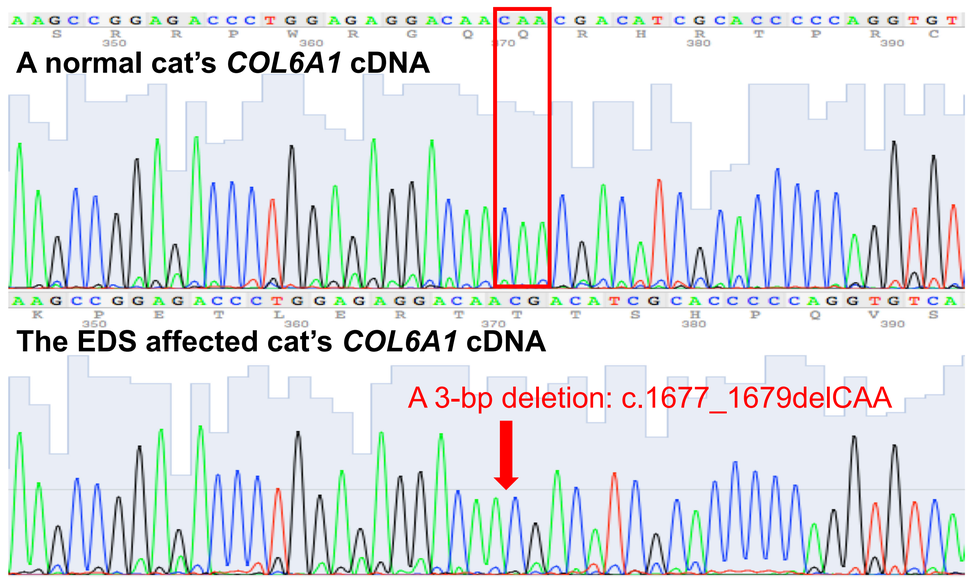

Figure 3. Sanger sequencing chromatograms of a normal cat (upper) and the affected cat (lower). A homozygous 3-bp deletion (c.1677_1679delCAA) is detected in the affected cat (lower) (The 3-bp deletion is absent in the normal cat as surrounded by a red square in the upper panel).

Comparison of coding sequences of *COL6A1* between a normal and the affected cat

- A normal cat : 3084 nucleotide (1027 amino acids)
- The EDS cat : 3081 nucleotide (1026 amino acids)

Discussion and Conclusions :

- An in-frame 3-bp deletion predicted to cause p.N560del in *COL6A1* was identified in a cat with EDS using WGS and the following cDNA analysis, and this variant was absent in other 194 cats.
- Mutations in *COL6A1* are known to cause Bethlem myopathy and Ullrich congenital muscular dystrophy in humans, [3] and some of them show skin abnormalities. [4]
- Furthermore, connective tissue abnormalities found in Ullrich congenital muscular dystrophy support phenotypic overlap with EDS. [5]
- Mutations/variants in *COL6A1* are also known to cause or be associated with abnormal ossification, [6,7] which may suggest the association of the variant we found with the spinal deformities in this cat.
- Although further validation is needed, our result might offer new insights into the spectrum of EDS.

References

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