JOURNAL OF FORENSIC SCIENCES

J Forensic Sci, September 2011, Vol. 56, No. 5
doi: 10.1111/j.1556-4029.2011.01856.x
Available online at: onlinelibrary.wiley.com

Authors' Response

Sir,

We thank Dr. De-Giorgio and his colleagues for acknowledging our manuscript entitled "Sudden and unexpected death in three cases of Ehlers-Danlos syndrome (EDS) type IV," by Shields et al. (1). We have asked Melanie Pepin, MS, CGC, Genetic Counselor, and Peter H. Byers, MD, Professor of Pathology and Medicine (Medical Genetics) at the Collagen Diagnostic Laboratory, University of Washington, Seattle, WA (http://www.uwcdl.org) for their expert insight to help us to address the issues raised in the Letter to the Editor written by Dr. De-Giorgio and colleagues.

In their Letter to the Editor of the *Journal of Forensic Sciences*, the authors state that the sensitivity of genetic testing for EDS type IV is 61% and cite Perdu et al. (2) as the source of this estimate. In the Perdu et al. article, the statement cites the study of the natural history of EDS type IV (3) as the source for the sensitivity quoted. The Pepin et al. (3) article, however, does not address the sensitivity of genomic sequencing of *COL3A1* or of collagen screening in its ability to identify individuals with EDS type IV. In the METHODS section of the article (Study Subjects), there were 220 index cases. All index cases had EDS type IV confirmed by biochemical studies of cultured fibroblasts. In the RESULTS section of the article (Correlation between Genotype and Phenotype), the causative mutation was reported in 135 (61% of all the 220 index patients). The mutation had not been searched for in the remaining 85 index cases. Thus, the 61% is not a measure of sensitivity but of the proportion of patients tested.

For ease and speed of analysis, we and all other clinical laboratories now sequence the coding regions and flanking intronic domains of the *COL3A1* gene as the first step in diagnostic testing. Rarely, we have encountered an individual in whom the clinical phenotype is strikingly consistent with EDS type IV, but in whom the *COL3A1* genomic sequencing is normal. In such instances, single or multiple exon duplication/deletions have been identified by the analysis of type III procollagen produced by cultured fibroblasts, cDNA sequence analysis, or multiplex ligation-dependent probe amplification or array-based studies of genomic DNA. Even after added testing, there remains a small number of individuals with an EDS type IV clinical phenotype in whom a *COL3A1* mutation is not found. The clinical features of EDS type IV and Loeys-Dietz syndrome overlap, and some individuals in whom we do not find mutations in *COL3A1* have a mutation in *TGFBR1* or *TGFBR2*.

We do not have a direct measure of the sensitivity of sequence analysis of the *COL3A1* gene to confirm the diagnosis of EDS type IV when suspected on clinical grounds. Overall, we think that genomic sequence analysis is the most sensitive technique to identify individuals with EDS type IV and find mutations in greater than 95% of affected individuals. It will miss some small deletions/duplications (they represent less than 3% of all mutations identified by all methods) and whole gene deletions (which we have not encountered, even in those with haploinsufficiency).

In their Letter to the Editor, Dr. De-Giorgio and colleagues also state "genetic tests should be reserved for cases in which the classic signs and symptoms of type IV EDS are not present or are so mild that they are not clinically evident." We do not agree with this sentiment, for several reasons. First, appropriate counseling of surviving family members depends on the correct diagnosis. In a small group of individuals that succumb to arterial rupture, the underlying mutations can be in the *TGFBR1* and *TGFBR2* genes (Loeys-Dietz syndrome), which can mimic EDS type IV. Mutations in other genes can also produce similar outcomes. Because the management of the disorder in affected relatives of the deceased individuals may differ, the correct diagnosis is essential to clinical care. Second, if a mutation is identified, accurate genetic tests of all at-risk family members can be provided in a rapid and inexpensive fashion to avoid expensive and repeated screening evaluation. Finally, the absence of a mutation in the *COL3A1* gene will direct attention to other loci that may unearth the causative mutations. Fundamentally, it is important to test all individuals in which the clinical presentation suggests an underlying genetic cause of arterial rupture because this is the only way to confirm the diagnosis and provide the resources with which to test at-risk family members. Our clinical diagnostic skill often falls short (4).

References

1. Shields LBE, Rolf CM, Davis GJ, Hunsaker JC. Sudden and unexpected death in three cases of Ehlers-Danlos syndrome type IV. J Forensic Sci 2010;55:1641–5.
2. Perdu J, Boutouyrie P, Lahlou-Laforêt K, Khau Van Kien P, Denarié N, Mousseaux E, et al. Vascular Ehlers-Danlos syndrome. Presse Med 2006;35:1864–75.
3. Pepin M, Schwarze U, Superti-Furga A, Byers PH. Clinical and genetic features of Ehlers-Danlos syndrome type IV, the vascular type. N Engl J Med 2000;342:673–80.
4. Ong KT, Perdu J, De Backer J, Bozec E, Collignon P, Emmerich J et al. Effect of celiprolol on prevention of cardiovascular events in vascular Ehlers-Danlos syndrome: a prospective randomised, open, blinded-endpoints trial. Lancet 2010;376(9751):1476–84.

Melanie Pepin,[1] M.S., C.G.C.; Peter H. Byers,[1] M.D.;
Lisa B.E. Shields,[2] M.D.; Cristin M. Rolf,[3,4] M.D.;
Gregory J. Davis,[3,4] M.D.; and John C. Hunsaker,[3,4] M.D., J.D.
[1]Collagen Diagnostic Laboratory, Department of Pathology, Box 357470, University of Washington, Seattle, WA 98195-7470.
[2]Norton Neuroscience Institute, 210 East Gray Street, Suite 1102, Louisville, KY 40202.
[3]Office of the Associate Chief Medical Examiner, 100 Sower Boulevard, Suite 202, Frankfort, KY 40601-8272.
[4]Department of Pathology and Laboratory Medicine, University of Kentucky College of Medicine, Lexington, KY.
E-mail: mpepin@u.washington.edu