Canine Infections with *Onchocerca lupi* Nematodes, United States, 2011–2014

Domenico Otranto, Alessio Giannelli, Maria S. Latrofa, Filipe Dantas-Torres, Nicole Scotty Trumble, Matt Chavkin, Gavin Kennard, Mark L. Eberhard, Dwight D. Bowman

Infections with *Onchocerca lupi* nematodes are diagnosed sporadically in the United States. We report 8 cases of canine onchocercosis in Minnesota, New Mexico, Colorado, and Florida. Identification of 1 cytochrome c oxidase subunit 1 gene haplotype identical to 1 of 5 from Europe suggests recent introduction of this nematode into the United States.

The role of *O. lupi* nematodes as an agent of infection in dogs in the United States has been suspected. However, nematodes were previously identified only as *Onchocerca* sp. in California and Utah (8,9) or as *O. lienalis* in Arizona (10). Recent etiologic delineation of *O. lupi* nematodes in dogs and cats in southwestern states (4,11,12) suggested involvement of this parasite in previous cases.

After the first case report of human ocular onchocercosis caused by *O. lupi* nematodes in Turkey (13), interest in this parasite has been renewed, and additional zoonotic cases have been identified in Turkey, Tunisia, and Iran (14). In addition, this parasite has been extracted from the cervical channel of a 22-month-old child in Arizona (12). Information on the epidemiology and life history of *O. lupi* nematodes is still minimal, and data on its distribution in the United States is limited to 6 case reports (4,11).

We report 8 cases of *O. lupi* nematode infection in dogs from Minnesota, New Mexico, Colorado, and Florida. We also compare cytochrome c oxidase subunit 1 (cox1) gene sequences from 2 nematodes with sequences from parasites in Europe to determine possible recent introduction of this filarioid from Europe to the United States.

The Study

During April 2011–August 2014, a total of 8 privately owned dogs of various ages and sexes were referred to clinical practices in Minnesota (n = 1), New Mexico (n = 4), Colorado (n = 2), and Florida (n = 1) because of different degrees of ocular alterations (Table). At physical examination, nodules were detected in different areas of the eye (Figure 1) and associated with inflammatory reactions ranging from mild scleritis to episcleral swelling and vascular congestion (Table).

All nodules were surgically removed from bulbar conjunctiva or sclera, and white filaria-like parasites were collected and stored in 70% ethanol for morphologic identification. In addition, specimens extracted from 2 dogs (dogs 2 and 3) (Table) were characterized by using molecular techniques. All dogs were treated with macrofilaricides, microfilaricides, antimicrobial drugs, and corticosteroids, which lead to complete resolution of ocular conditions in all except 3 animals (dogs 1, 7, and 8). These 3 dogs had relapses 2, 6, and 12 months, respectively, after surgery.

Nematodes had external, round, transverse ridges and 2 transverse striae per each outer ridge interval, which suggested that they were filarial worms of the genus *Onchocerca*. The ratio between body diameter and distances between ridges (7–10:1) was specific for *O. lupi* nematodes (15). A small piece of nematode was used for molecular...
identification. Genomic DNA was extracted and partial cox1 genes were amplified and sequenced as described (13).

In accordance with clinical signs of nodular ocular lesions and morphologic identification, partial cox1 gene sequence analysis (GenBank accession nos. KP283476 and KP283477) confirmed the identity of the nematode as *O. lupi*, showing 98% nt homology with other sequences of *O. lupi* nematodes in GenBank (KC686701 from Portugal and KC686702 from Greece) and 100% with those derived from dogs and cats from the United States, as well as with a sequence from Greece (EF521409).

Phylogenetic analysis of partial cox1 gene sequences was performed by using the neighbor-joining method and the Kimura 2-parameter model in MEGA5 (http://www.megasoftware.net/). This analysis confirmed that sequences from nematodes examined clustered with *O. lupi* sequences from different areas of the United States (Nevada, California, Colorado, Utah) and with a sequence from Greece (Figure 2). In addition, these sequences were grouped with others from Greece, Hungary and Portugal and formed a paraphyletic clade with other *Onchocerca* species available in GenBank.

**Conclusions**

Our results indicate that a unique haplotype of *O. lupi* nematodes is circulating in the United States and is endemic to the canine population in this country. Although this onchocercid has been implicated as the causative agent of canine onchocercosis in the United States only recently (11), previous cases attributed to *Onchocerca* spp. have been described in dogs from Arizona, California, and Utah (3,8–10). The cases herein reported from Florida, New Mexico, and Minnesota suggest that the distribution of this nematode is probably wider than previously believed. Detection of *O. lupi* nematodes in Englewood, Colorado, confirms a previous report of infection in a dog from Mancos (11).

We identified 1 cox1 haplotype and found that it was identical to all sequences in GenBank from the United States and 1 from Greece. Conversely, up to 5 haplotypes were detected in Greece, Turkey, Iran, and Hungary (7). Genetic variation detected in *O. lupi* nematodes from Europe, Turkey, and Iran, along with isolation of this parasite from the Caucasian wolf, suggests that the infection probably originated in the Old World and was imported into the United States.

The low genetic distance detected for the cox1 gene is evidence of a substantially reduced evolutionary rate, which supports relatively recent divergence among specimens found in the Old World and New World. In addition to recent detection of *O. lupi* nematode infections in the United States, circulation of 1 haplotype could also suggest that a unique vector species occurs in areas of the Old World and New World where the infection has been diagnosed.

Given that all reports above are based on clinical signs, the epidemiology of *O. lupi* nematodes in the United States deserves to be thoroughly investigated. In particular, dogs relocated from disease-endemic areas to new areas should be routinely screened for skin-dwelling microfilariae because these parasites might represent a risk for other animals. In addition, because *O. lupi* nematodes circulate among canine populations, the potential role of dogs as reservoirs for human infection should not be underestimated,

**Table.** Characteristics of 8 dogs infected with *Onchocerca lupi* nematodes, United States

| Dog no. | Age, y/sex | Geographic origin (travel history) | Clinical signs |
|---------|------------|-----------------------------------|----------------|
| 1       | 8/F        | Hollywood, Florida (rescued)      | Conjunctival and relapsing lesions in both eyes |
| 2       | 7/M        | Orofino, Minnesota (Durango, Colorado) | Mild scleritis, proliferative eye lesions |
| 3       | 6/F        | Englewood, Colorado (Farmington, New Mexico) | Episcleral swelling and vascular congestion, squinting, scleral indentation in the temporal fundus |
| 4       | 2/F        | Englewood, Colorado (Farmington, New Mexico) | Inflammatory conjunctival follicles, mild ephora, mild diffuse conjunctival hyperemia, episcleral |
| 5       | 3/M        | Farmington, New Mexico            | Moderate blepharospasm and conjunctival hyperemia |
| 6       | 3/M        | Farmington, New Mexico            | Moderate chemosis, episcleral flocculent mass |
| 7       | 9/F        | Jerez, New Mexico                 | Chronic waxing/waning episcleral mass |
| 8       | 5/M        | Farmington, New Mexico            | Chronic conjunctivitis, superficial keratitis, episcleral mass, chemosis |

**Figure 1.** Subconjunctival nodule on the medial canthus of the right eye of dog 2 (Table), Minnesota, USA. This dog was found to be infected with *Onchocerca lupi* nematodes.

Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 21, No. 5, May 2015
as also inferred by zoonotic cases reported in the United States (12). Finally, further studies are urgently warranted toward improving the diagnosis of O. lupi nematode infections, which will lead to a better appreciation of its distribution and potential risk for human populations.

Dr. Otranto is a professor in the Department of Veterinary Medicine, University of Bari, Valenzano, Italy. His research interests include biology and control of arthropod vector-borne diseases of animals and humans.

References

1. Otranto D, Eberhard ML. Zoonotic helminths affecting the human eye. Parasit Vectors. 2011;4:41. http://dx.doi.org/10.1186/1756-3305-4-41

2. Bowman DD. Heartworms, macrocyclic lactones, and the specter of resistance to prevention in the United States. Parasit Vectors. 2012;5:138. http://dx.doi.org/10.1186/1756-3305-5-138

3. Szent T, Széli Z. Onchocercosis: a newly recognized disease in dogs. Vet Parasitol. 2008;151:1–13. http://dx.doi.org/10.1016/j.vetpar.2007.09.008

4. Labelle AL, Daniels JB, Dix M, Labelle P. Onchocerca lupi causing ocular disease in two cats. Vet Ophthalmol. 2011;14:105–10. http://dx.doi.org/10.1111/j.1463-5224.2011.00911.x

5. Zarfoss MK, Dubielzig RR, Eberhard ML, Schmidt KS. Canine ocular onchocerciasis in the United States: two new cases and a review of the literature. Vet Ophthalmol. 2005;8:51–7. http://dx.doi.org/10.1111/j.1463-5224.2005.00348.x

6. Franchini D, Giannelli A, Di Paola G, Cortes H, Cardoso L, Lia RP, et al. Image diagnosis of zoonotic onchocercosis by Onchocerca lupi. Vet Parasitol. 2014;203:91–5. http://dx.doi.org/10.1016/j.vetpar.2014.03.007

7. Otranto D, Dantas-Torres F, Giannelli A, Latrofa MS, Papadopoulos E, Cardoso L, et al. Zoonotic Onchocerca lupi infection in dogs, Greece and Portugal, 2011–2012. Emerg Infect Dis. 2013;19:200–3. http://dx.doi.org/10.3201/eid1912.130264

8. Orbel TC, Ash LR, Holshuh HJ, Santenelli S. Onchocerciasis in a California dog. Am J Trop Med Hyg. 1991;44:513–7.

9. Gardiner CH, Dick EJ Jr, Meining G, Lozano-Alarcón F, Jackson P. Onchocerciasis in two dogs. J Am Vet Med Assoc. 1993;203:828–30.

10. Eberhard ML, Ortega Y, Dial S, Schiller CA, Sears AW, Greiner E. Ocular Onchocerca infections in two dogs in western United States. Vet Parasitol. 2000;90:333–8. http://dx.doi.org/10.1016/S0304-4017(00)00252-1
11. Labelle AL, Maddox CW, Daniels JB, Lanka S, Eggett TE, Dubielzig RR, et al. Canine ocular onchocercosis in the United States is associated with *Onchocerca lupi*. Vet Parasitol. 2013;193:297–301. http://dx.doi.org/10.1016/j.vetpar.2012.12.002

12. Eberhard ML, Ostovar GA, Chundu K, Hobohm D, Feiz-Erfan I, Mathison BA, et al. Zoonotic *Onchocerca lupi* infection in a 22-month-old child in Arizona: first report in the United States and a review of the literature. Am J Trop Med Hyg. 2013;88:601–5. http://dx.doi.org/10.4269/ajtmh.12-0733

13. Otranto D, Sakru N, Testini G, Gürlü VP, Yakar K, Lia RP, et al. Case report: first evidence of human zoonotic infection by *Onchocerca lupi* (Spirurida, Onchocercidae). Am J Trop Med Hyg. 2011;84:55–8. http://dx.doi.org/10.4269/ajtmh.2011.10-0465

14. Otranto D, Dantas-Torres F, Brianti E, Traversa D, Petrić D, Genchi C, et al. Vector-borne helminths of dogs and humans in Europe. Parasit Vectors. 2013;6:16. http://dx.doi.org/10.1186/1756-3305-6-16

15. Mutafchiev Y, Dantas-Torres F, Giannelli A, Abramo F, Papadopoulos E, Cardoso L, et al. Redescription of *Onchocerca lupi* (Spirurida: Onchocercidae) with histopathological observations. Parasit Vectors. 2013;6:309. http://dx.doi.org/10.1186/1756-3305-6-309

Address for correspondence: Domenico Otranto, Università degli Studi di Bari, Strada Provinciale per Casamassima km 3, Valenzano 70010, Italy; email: domenico.otranto@uniba.it