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Letter to the Editor

Mycobacterium tuberculosis infection in a blue-fronted amazon parrot (Amazona aestiva aestiva)

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Psittacine birds are the only avian species known to become infected with Mycobacterium tuberculosis and tuberculosis-infected humans are often suspected to be the source of infections (Washko et al., 1998). In this report a case of an adult female blue-fronted amazon (Amazona aestiva aestiva) is presented. The parrot developed after several weeks of general illness exophytic skin proliferations predominantly on the head (Fig. 1) and also a sublingual nodule. The bird was euthanised and tuberculosis was diagnosed using pathomorphological, bacteriological and molecular approaches. The two owners who used to feed the bird with pre-chewed food, had suffered from pulmonary tuberculosis and had completed anti-tuberculosis treatment 15 and 6 months, respectively, before admission of the bird to a veterinary clinic.

On *post mortem* examination the amazon was found to be emaciated. The skin proliferations could easily be broken off. Impression smears of the lesions revealed acid-fast bacteria. Histologically, the skin proliferations were characterised by extensive hyperkeratosis and multiple underlying intradermal granulomas. In the tuberculous lesions with central necroses which were surrounded by Langhans and epithelioid cells and peripheral fibroplasia, acid-fast bacteria were detected. Furthermore, numerous granulomas were present in the sublingual nodule, the conjunctiva of the right eye, the lungs, liver, spleen, kidneys, and under the serosal surface of the gizzard. *Mycobacterium tuberculosis* was cultured from several tissues of the bird and identified by molecular methods (Kamerbeek et al., 1997; Rodriguez et al., 1995). The isolates of the bird's owners were kindly provided by a local laboratory. Identical spoligotyping patterns, MIRU codes, and RFLP-profiles of the avian and human *M. tuberculosis* isolates were obtained indicating strongly a zoonotic link.

The spoligotyping pattern found, belonged to the Haarlem Type 3 as deposited in the spoligotyping database SpolDB4 (Brudey et al., 2006). The Haarlem lineage represents approximately 25% of the M. tuberculosis isolates in Europe including Germany, and was also detected in diverse non-European countries. RFLP typing was performed on the basis of the insertion sequence (IS) 6110 (Van Soolingen et al., 1994). IS6110-RFLP, has up to now been the most widespread method used in tuberculosis molecular epidemiology. RFLP analysis revealed that the avian lung isolate and the human isolates possessed identical profiles (Fig. 2). The profile of the avian liver isolate differed from the other isolates by one single additional band. This is not unusual and demonstrates that the evolutionary clock regarding this mobile element may go rather fast (Glynn et al., 2004). Both methods, RFLP analysis and spoligotyping, are complex and time-consuming since they combine PCR, southern blotting and DNA-DNA hybridization. Mycobacterial interspersed repetitive units (MIRU) typing is a PCR-based more and more commonly used subtyping method and was conducted targeting 12



Fig. 1. Hyperkeratotic vertucous skin proliferations (arrow) on the head of the blue-fronted amazon parrot (*Amazona aestiva aestiva*) due to cutaneous tuberculosis.

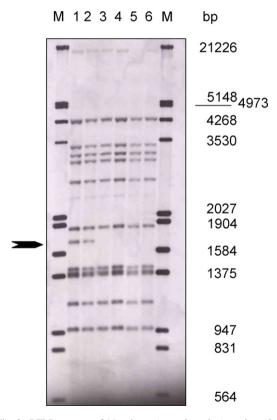


Fig. 2. RFLP patterns of *Mycobacterium tuberculosis* strains originating from the blue-fronted amazon parrot (*Amazona aestiva aestiva*) and both of its owners. Note that the avian liver isolate shows an additional band (arrow). Lanes 1 and 2: avian isolate, liver; lanes 3 and 4: avian isolate, lung; lane 5: human isolate, owner 1; lane 6: human isolate, owner 2; M: molecular weight marker; bp: base pairs.

single genomic loci using primers previously described (Mazars et al., 2001; Prodinger et al., 2005).

These loci are well known for their discriminatory capacity towards *M. tuberculosis* isolates of global origin and other members of the MTC and the identity of the avian and human patterns strongly indicates the close genetic relationship of the isolates. The pattern was similar to MIRU patterns of other *M. tuberculosis* isolates from different European and non-European countries listed in the database (http://ibl.fr/mirus/mirus.html).

With regard to anamnestic and pathomorphological facts, the reported case reveals remarkable parallels to a disseminated *M. tuberculosis* infection in a greenwinged macaw (Washko et al., 1998). In both cases the tuberculosis-infected owners used to feed the parrot from their mouths. The proliferative cutaneous lesions on the head may be due to transmission of the agent by aerosols (Hinshaw, 1933). The extreme hyperkeratotic and verrucosal appearance, as well as the histopathological morphology of the tubercles, suggests a parallel to "tuberculosis verrucosa cutis (TVC)", a form of cutaneous tuberculosis in humans following inoculation of *M. tuberculosis* into the skin of previously sensitized patients possessing an intact immune system (Barbagallo et al., 2002) perhaps by its own infective exudates.

This report is to our knowledge the first one of a *M*. *tuberculosis* infection of human and bird where the zoonotic link was supported by molecular typing of the respective bacterial isolates.

In those cases, the infected pet has to be considered to be a health risk and potential source of re-infection of its owners.

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