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Occurrence of the oribatid mite *Trhypochthoniellus longisetus longisetus* (Acari: Trhypochthoniidae) on tilapia *Oreochromis niloticus*

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ABSTRACT: Mites as parasites infesting fish have been described in a few case reports involving *Histiostoma anguillarum*, *H. papillata*, and *Schwiebea estradai*. We describe the unexpected occurrence of oribatid mites of the genus *Trhypochthoniellus* on farmed tilapia *Oreochromis niloticus*. The fish had mites on the skin, fins, and gills, as well as in the mouth. The morphological characteristics of the mites, observed by optical and scanning electron microscopy, were consistent with those described for *T. longisetus longisetus*. All stages of development were observed, suggesting that the mites were able to actively reproduce on fish.

KEY WORDS: Fish parasite · Mite · Oribatida · *Trhypochthoniellus longisetus* · Tilapia · *Oreochromis niloticus* · Aquatic animal health

INTRODUCTION

Many fishes in wild populations have parasites, but most do not represent a serious health threat. However, farmed fishes are subjected to environmental stress, which makes them more sensitive than wild fishes to parasitic diseases. Fish parasites belong to many different taxonomic groups, including Protozoa, Monogenea, Trematoda (Digenea), Cestoidea, Nematoda, Annelida, Crustacea, Mollusca, and Acarina. Very little is known about mite (Acarina) infestation of fishes, and there are few records listed in journals. These studies indicate, however, that massive infestation, although infrequent, has negative effects on fish health (Fain & Belpaire 1985, Fain & Ferrando 1990, Halliday & Collins 2002).

The habitat and behavior of fishes do not contribute to mite infestation, and some authors consider mites to be unusual parasites of fish, whereas for others, mites are not fish parasites at all (Halliday & Collins 2002). However, under certain environmental conditions, mites may proliferate and colonize weak or stressed fishes, attaching to the mucous membranes and causing serious damage. Several genera of mites have been isolated from the skin, gills, and esophagus of fishes in Europe, Australia, and North America (Heckmann 2003), in some cases associated with high fish mortality. In one report, *Histiostoma papillata* (Histiostomatidae, Astigmata) attacked and apparently killed juvenile Murray cod *Maccullochella peelii peelii* held in a recirculation aquaculture system (Halliday & Collins 2002). Likewise, *H. anguillarum* was described as a mite that parasitizes the gills of young farmed freshwater eels *Anguilla anguilla* (Fain & Belpaire 1985). Mortality was also high in trout *Salmo trutta fario* from a fish farm and a river; these fish had *Schwiebea* (*Jacoti-*

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etta) *estradai* mites on the skin, gills, and in the intestine (Fain & Ferrando 1990). In the present work, we describe, for the first time, the occurrence of the oribatid mite *Trhypochthoniellus longisetus longisetus* on farmed tilapia *Oreochromis niloticus*. This oribatid mite is an aquatic species that lives in a wide range of freshwater habitats (from bogs to aquariums) commonly associated with vegetable organic material or aquatic plants (Lange & Tolstikov 1999, Kuriki 2005). Specimens of *Trhypochthoniellus* sp. have also been previously found in the gills of juvenile Atlantic salmon *Salmo salar* from a river system (Hare & Burt 1975).

MATERIALS AND METHODS

Farmed tilapia were held at the Animal Production Field Station of the Polytechnic University of Madrid, Spain. Tilapia were held in a recirculation aquaculture system consisting of 16 fiberglass tanks interconnected with a solids separator and biofilter. Each tank (120 l capacity) was properly aerated and kept at a temperature (±SD) of 21.8 ± 0.7°C and photoperiod of 12 h light:12 h dark. At the time of disease and our investigation, there were 10 fish tank^{-1}, with an average (±SD) weight of 56.5 ± 13.4 g, and fish were fed twice a day with pelleted commercial feed (32% protein, Dibaq). The physical and biological properties of the water were not considered to be outside the normal ranges for tilapia (oxygen >6 ppm, pH 7 to 8, ammonia and nitrites <0.01 ppm, and nitrates <150 ppm).

Mites collected from the skin of the diseased fish were examined using a Nikon eclipse E-400 optical microscope for morphological data. For this purpose, 16 mites were mounted in polyvinyl alcohol (PVA) on microscope slides. For examination of mite taxonomic characteristics, samples were prepared and mounted for scanning electron microscopy (SEM) on a JEOL JSM 6400 (see Fig. 2C,D). The mites were pre-fixed in 3% glutaraldehyde in phosphate buffer (pH 7.4) for 3 h, washed twice in phosphate buffer (pH 7.4), and dehydrated in an alcohol series (50, 70, 80, 90, and 100%) for 15 min. Further routine treatments were processed for SEM at the 'Luis Bru' Electronic Microscope Laboratory at Complutense University, Madrid.

RESULTS AND DISCUSSION

Significant signs of external parasitism, anomalous swimming, and irritation were detected in 4 of the tanks. In 1 tank, 2 fish had died, while most of the remaining fish were alive but lethargic. These fish were taken to Complutense University for diagnosis.

Examination of the affected fish revealed the presence of numerous mites attached to the body surface, fins, gills, and inside the mouth, with up to 15 mites cm^{-2} in some areas (Fig. 1). Of the 16 mites analyzed, a total of 10 adults were identified as females (average size 523 × 294 µm) . Of the 6 immature mites, 2 were identified as larvae, which are hexapods (average size, 222 × 104 µm), and 4 were identified as nymphs (average size, 443 × 140 µm). The high numbers of mites on fish mucus and the presence of all developmental stages, including gravid females (Fig. 2A,B) and free eggs, suggest that mites were reproducing on the fish. The number of eggs ranged from 1 to 4 female^{-1}. Morphological data for dorsal and ventral sides were obtained by SEM (Fig. 2C,D). The mites were identified as the oribatid mite *Trhypochthoniellus longisetus*

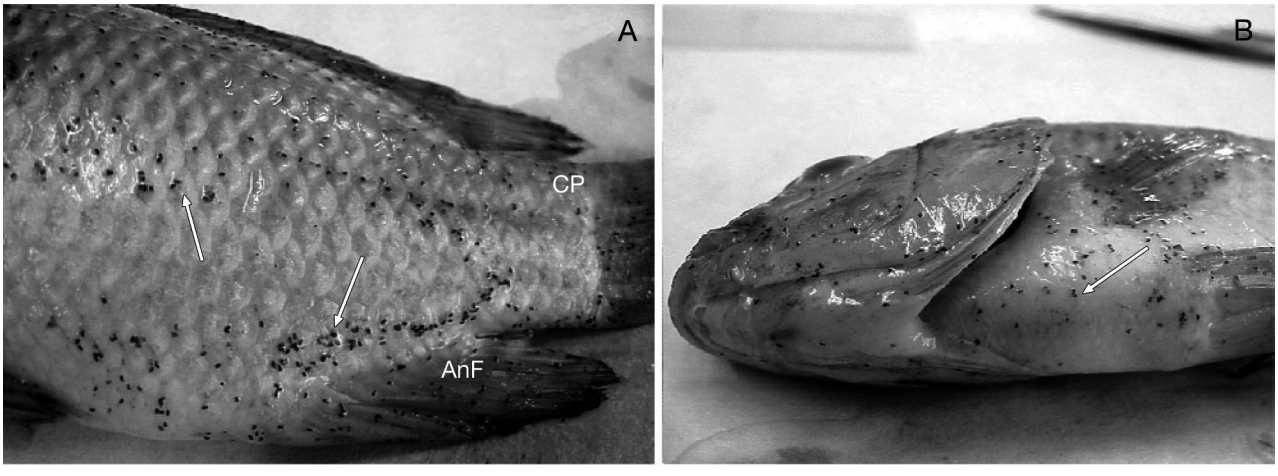

Fig. 1. *Trhypochthoniellus longisetus longisetus* on *Oreochromis niloticus*. Gross appearance of mites (arrows) on the skin of tilapia. (A) Lateral and (B) ventral view. AnF = anal fin, CP = caudal peduncle

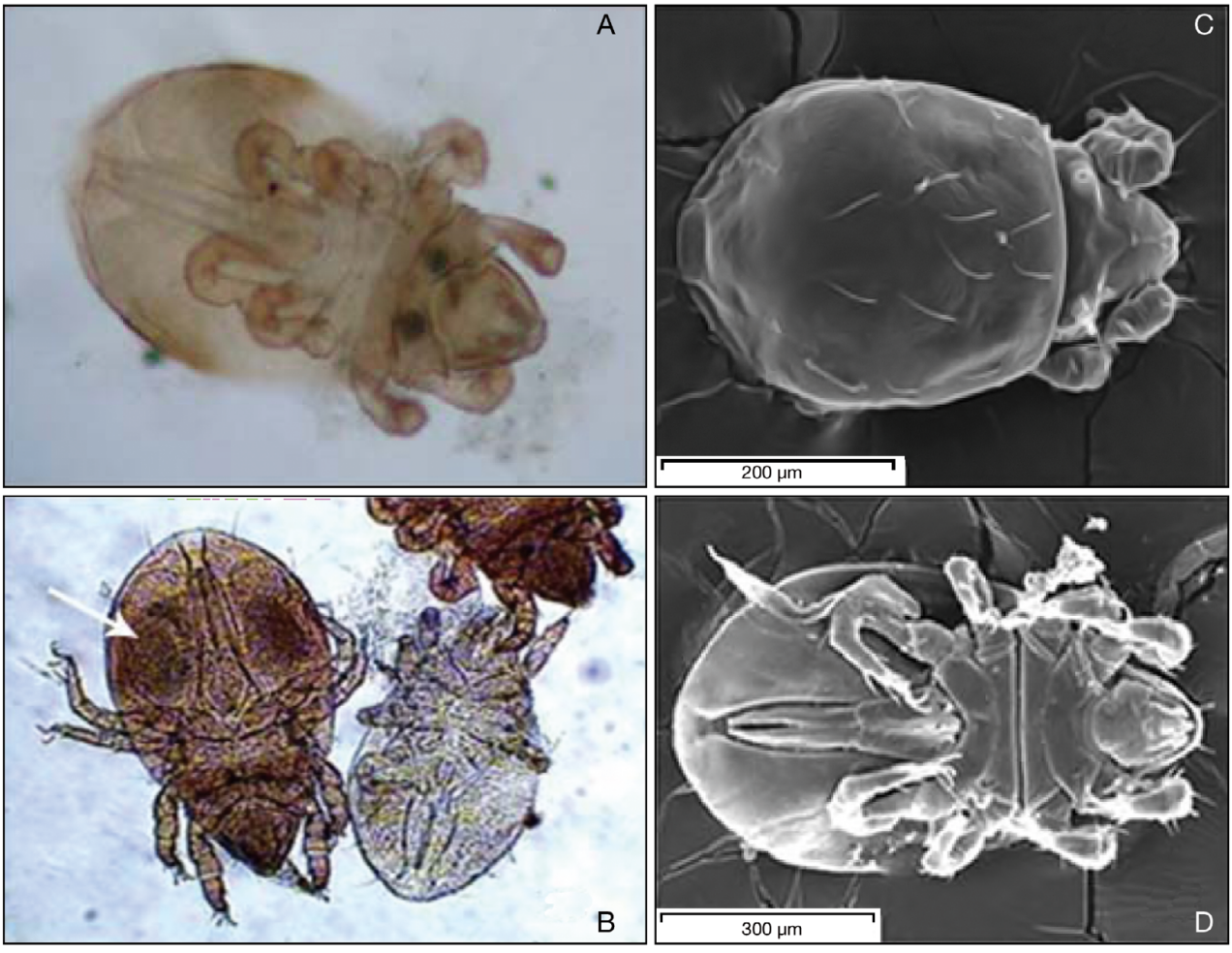

Fig. 2. *Trhypochthoniellus longisetus longisetus* from *Oreochromis niloticus*. Light micrographs of cleared, slide-mounted specimens of mites from fish skin: (A) adult female mite in ventral position and (B) adult female mite with developing eggs (arrow) and 1 nymph. Scanning electron micrographs of mites in (C) dorsal and (D) ventral positions. Scale bars show the original magnification

longisetus based on taxonomic characteristics originally described for this species (Berlese 1904, Aoki 1964, Colloff & Halliday 1998), and on the setae formula of the epimeral region 3-1-3-2 (Fig. 2C,D) subsequently described for this mite (Weigmann 1999, Kuriki 2005).

According to the mite classification by Subías (2010), the genus *Trhypochthoniellus* (Willmann, 1928) comprises 7 species, including *T. longisetus,* which in turn includes 3 subspecies: one with a wide distribution, *T. longisetus longisetus* (Holarctic, Australian, Ethiopian, and Neotropical), and 2 that are more geographically restricted (*T. longisetus brasiliensis* and *T. longisetus canadensis*). *T. longisetus* is a parthenogenetic species, so there are no males, as we observed in our samples of mites. The subspecies *T. longisetus longisetus* (junior synonym of *T. crassus* [Warburton & Pearce 1905]; *Hydronothrus crispus* [Aoki 1964]; *Hydronothrus aquariorum* [Fain & Lambrechts 1987]; *Trhypochthonius setosus* [Willmann 1928]; in addition to other synonymous names, Subías 2010) is commonly found in vegetation at the edge of lakes and ponds, where it seems to feed primarily on cellulose of living green plants or decaying vegetation, such as dead leaves of *Hibiscus tiliaceus* (Aoki 1964). This oribatid mite is widespread, extremely common in aquatic habitats that are rich in organic material, such as some natural freshwater, mires, high-mounted drinking-water tanks, and swimming pools (Tagami et al. 1992, Kuriki 2005, Perotti et al. 2009). In fact, because it is a common contaminant in indoor pools in some countries, such as Japan, it has recently been considered to be an allergen of public health significance (Robinson 2005).

Owing to their small size and great plasticity, mites can adapt to a wide range of vertebrate hosts (from fishes to mammals) and habitats (Fain 1994). Mites have evolved and adapted to almost every habitat on land and they are found in every kind of freshwater. Their adaptation to aquatic conditions evolved independently in different taxa: Mesostigmata, Astigmata, Oribatida, and Prostigmata (Proctor 2004, Weigmann & Deichsel 2006, Schatz & Behan-Pelletier 2008). Ecologically, mites belonging to the Oribatida subgroup are primarily terrestrial, predominantly free-living detritivores and fungivores, and only about 1% of all known species (some species of a few oribatid genera, including *Trhypochthoniellus*) are truly aquatic (Schatz & Behan-Pelletier 2008). Aquatic oribatids are present in standing and running water, but all are non-swimming, and have diets similar to their terrestrial relatives (Proctor 2009).

Oribatid mites have previously been associated with parasitism of fishes (Hare & Burt 1975, Choudhury et al. 2004), but, to our knowledge, the present work is the first case that describes infestation of farmed tilapia by oribatid mites (*Trhypochthoniellus longisetus longisetus*). It is not clear how these mites reached the freshwater, but once they had, the recirculating system provided an ideal environment for mite growth: warmth, nutrient-rich water, and abundant surface-hosts. The fact that all stages of development, including free eggs, were observed could indicate that the mites were able to actively reproduce on fish, and suggests an adaptation of mite to host. Probably, these mites were feeding on bacterial slime, fungal remains or spores, and yeast growth on the surface of the fish or attached to the fish mucus. Although it seems very unlikely that mites would feed directly on a corpse, there is at least 1 report of *T. longisetus longisetus* associated with the necrotic skin of discus fish *Symphysodon* sp. (Fain & Lambrechts 1987); however, the ingestion of animal debris by oribatid mites is rare and could be considered accidental. In the present study, *T. longisetus longisetus* seems to be an opportunistic feeder that reached high densities on the fish, having negative effects on the health of these animals. Heavily infested fish may become seriously injured because of the alterations in the fins and mucus and may even suffer mechanical asphyxia as a result of the infestation of the gills.

CONCLUSIONS

Trhypochthoniellus longisetus longisetus has been found, for the first time, on farmed tilapia. All stages of development were observed in the mites analyzed, suggesting that they were able to actively reproduce on fish. Although the presence of this mite on tilapia is not part of its normal biological life cycle, the unexpected high proliferation of this mite could have negative effects on fish health.

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