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Occurrence of *Anisakis* spp. larvae in products made of herring (C*lupea harengus*)

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INTRODUCTION. Herrings are the third most commercialized species in the European Union (EUMOFA report, 2017) and common hosts of third stage *Anisakis* spp. larvae (Levsen & Lunestad, 2010, Vet. Parasitol, 171:247–253). Different kind of ready to eat (RTE) herring products are available on the market. Although these products undergo technological processes able to kill viable parasites, exposure to dead larvae may cause allergic reactions (Audicana & Kennedy, 2008, Clin. Microbiol. Rev, 21:360-379). Given the scarcity of available data on anisakids in RTE herrings sold on the Italian market, the aim of this study was to assess the occurrence of *Anisakis* spp. larvae in various kind of such products.
MATERIALS AND METHODS. 120 products consisting of 50 smoked whole specimens and 70 filleted products (25 smoked, 30 smoked and marinated, 15 canned) were sampled between 2016 and 2018. In the case of whole herrings, viscera and muscle were visually inspected and separately digested using Trichineasy® (CTSV srl, Brescia). Filleted products, including marinating liquid if present, were also visually inspected and digested. Nematodes collected during visual inspection and after digestion were identified to genus level, counted and stored. A subsample (N=150) was molecularly identified targeting the *COII* gene. The positivity rate and the larval density per gram (number of larvae per g of examined tissue), both at muscle and visceral level, when present, were calculated; differences between whole and filleted products were investigated by Chi-square and Kruskal-Wallis tests.
RESULTS AND CONCLUSIONS. At least one *Anisakis* spp. larva was found in 56 products (46.7%), with a total of 1715 dead larvae collected (range 0-172 larvae/product). The majority (1559, 91%) were found in the viscera of 49 of the 50 whole herrings (98%). Interestingly, a highly significant difference ($p<0.0001$) was observed between the positivity rate and larval density of the remaining 156 larvae found at muscle level, as 149 larvae were found in the muscle of 31 whole herrings (positivity rate 62%, 0.022 larval density/g) while only 7 larvae were found in the 70 filleted products (positivity rate 10.7%, 0.001 larval density/g). Larvae were molecularly identified as *A. simplex*. Although no live larvae were found, dead visible larvae represent a defect and make the product unfit for human consumption (Reg. (EC) No 178/2002). Especially in the case of heavy infections, larvae may be evident and cause consumers' rejection. In addition, the allergenic potential of dead larvae in sensitized subjects is debated (Daschner et al., 2012, Trends Parasitol, 28:9-15). The significant difference between muscle tissue of whole and filleted herrings, likely due to differences in the production process, might result in different level of exposure depending on consumers' preferences.