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### Genotyping anisakid nematodes using HRM analysis

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Anisakids are parasitic nematodes belonging to the super family Ascaridoidea. The presence of infective larvae in fish intended for human consumption cause economic and medical problems. Among anisakids, *Anisakis simplex sensu lato* and *Pseudoterranova decipiens* s. l are the species mainly responsible for human anisakidosis. Rarely, *Contracaecum* sp. and *Hysterothylacium* sp. (Raphidascaridae Family) have been found in association with accidental found in gastric/intestinal tract, even if the latter is commonly considered not pathogenic to humans. The aim of the study was to develop a new molecular approach to differentiate and identify anisakids based on the High Resolution Melting (HRM) of a selected molecular marker (nuclear ribosomal ITS). By now, only *A. simplex* complex was discriminated using HRM. Representative species of *Anisakis*, *Pseudoterranova*, *Contracaecum* and *Hysterothylacium* were tested to verify the presence of diagnostic profiles to differentiate nematodes at genus and species level. HRM profiles of *Anisakis simplex* s. l revealed Tm of 78.0-78.2 °C for *A. simplex* s. s., of 79.4-79.5 °C for *A. pegreffii* and of 78.4-78.6 °C for hybrids. Members of *P. decipiens* complex were tested with *Anisakis* spp., *Contracaecum osculatum* and *Hysterothylacium aduncum*. Results revealed no amplification for all *Anisakis* spp and three HRM profiles: One showed Tm of 79.4-79.5 °C for *P. decipiens sensu stricto* and *P. krabbei*; one showed 79.9-80.0 °C for *P. azarasi* and *P. cattani*; one showed 79.1-79.2 °C for *C. osculatum* and *H. aduncum*. Preliminary results obtained indicate the usefulness of such genotyping approach, potentially functional for screening in basic research area, food industry and diagnostics.

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