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Protocol for the isolation of processed animal proteins from insects in feed and their identification by microscopy

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12 Abstract

13 Insect processed animal proteins (PAPs) constitute a new alternative source of proteins in feed. In
14 2017, a closed list of insect species was authorized on the European market for use in aquafeed
15 production. Authenticity and contamination controls will have to be set up by authorities and feed
16 actors and supported by adequate detection methods, which are lacking. The present paper presents
17 an original isolation and detection protocol for insect material. The protocol, based on sedimentation
18 by a mixture of petroleum ether and tetrachloroethylene to concentrate insect particles, was
19 developed and tested on a series of ten different aquafeeds fortified at 1 % w/w with four different
20 commercially available insect meals (from *H. illucens*, *T. molitor*, *G. assimilis* and *A. diaperinus*). The
21 results showed that this sedimentation protocol combined with light microscopic observation was
22 adequate for insect detection and more efficient than the current official method. Morphological key
23 features for reliable characterization of insect PAPs were also investigated. Structural details of
24 cuticular fragments, such as sensilla and tracheolar structures, combined with patterns of muscle

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