



# 2016 世界头足类渔业学术大会

2016' International Conference on Cephalopod Biology and Sustainable Fisheries

头足类渔业可持续发展  
Sustainable Development of Cephalopod Fisheries



## 会议手册 HAND BOOK

支持单位：联合国粮农组织 头足类国际咨询委员会 浙江省自然科学基金委员会

主办单位：中国水产学会 浙江海洋大学

Supported by Food and Agriculture Organization of the United Nations (FAO)

Cephalopod International Advisory Council (CIAC)

Zhejiang Provincial Natural Science Foundation (ZJNSF)

Hosted by China Society of Fisheries Zhejiang Ocean University

中国浙江舟山群岛新区

Zhoushan Archipelago New Area, Zhejiang, China

2016年11月18日-20日



**Dr. Lara Tinacci**

**Dr. Lara Tinacci**

Department of Veterinary Sciences, University of Pisa

### Biography

March-Sept. 2015: Six month Research fellowship in “Molecular biology methods for the control of commercial and health issues in ethnic seafood sold on Italian market.” Department of Veterinary Sciences, University of Pisa

Jan. 2011-Dec. 2014: Research and study activity on: “Development of Molecular biology methods for fish identification and fish fraud control and prevention.” Department of Veterinary Sciences, University of Pisa

April-Oct. 2010: Six month post degree scholarship for “Fish species identification by means of Molecular techniques.” Department of Veterinary Sciences; University of Pisa

February 2008-October 2009: Training in Immunology, basic principles and training in Immunoassay and protein electrophoresis technique for the development of ELISA test to be applied for the control of illegal hormones treatment in bovine and buffalo

### Title

Beyond tentacles: improving cephalopod species identification and traceability through DNA based methods

### Abstract

Cephalopods are short-lived organisms, characterised by rapid growth rates. In the last decades, due to the environmental changes, their worldwide biomass is dramatically increased. Their high nutritional quality, comparable to that of fish, has led to a significant increase of international market demand. Squids represent the great majority of the yearly global catches followed by octopus and cuttlefish. Spain, Italy, Japan and US are the largest consumers and importers. Major market suppliers include Asian, North African, US and South American countries. Cephalopods are generally commercialized as fresh or frozen whole or sliced products (rings and arms, tubes and wings) or used for the production of multispecies seafood products such as minced burgers or surimi. In order to ensure seafood traceability, the European Union (EU) has established some mandatory label declaration (commercial and scientific name, production method, catching or farming area, category of fishing gear (Reg. (EU) N. 1379/2013). In particular, each EU member state is in charge to produce and update official lists of commercial designations applying the principle one species one name. Moreover, due to the high content in tropomyosin, cephalopods are included in the list of allergens and must be declared on the product label (Reg. (EU) N. 1169/2011). The ascertained worldwide increase of seafood falsification attests the importance of verifying seafood identity to prevent both economic loss and health concerns due to fraudulent species substitutions. In this perspective DNA-based methodologies represent a valuable tool for food traceability and are particularly useful for seafood identification especially in processed product where the external key morphological features have been removed. In this study a



preliminary revision of the literature regarding the application of DNA based methods for cephalopods identification has been conducted to assess the state of the art. Moreover, the results of three different studies conducted in our lab are presented. The conducted revision shows how DNA based studies have been developed, initially for taxonomic purposes, since early 1990s by the application of FINS (Forensically Informative Nucleotide Sequencing) technique targeting several mitochondrial and nuclear genes. Lately, analytical tools were also applied to inspection and traceability purposes. RFLP based techniques were set for the rapid detection of a limited number of species. Given the rapid growth of global cephalopod market and the increased number of commercialized species, RFLP has been abandoned in favor of universal techniques (FINS and DNA barcoding) mainly addressed to 16S rRNA, Cyt b, COI genes analysis. These methods are currently used for species identification and labelling check of both fresh and processed seafood but present major limits when applied to complex multispecies products. Next Generation Sequencing technology (NGS), only recently introduced for the species detection in processed meat and fish, may represent a valid alternative tool since it produces distinct molecular information for all the DNA populations contained in the original food mixture. The first work aimed at evaluating the approach of several EU countries with respect to the draw up and the update of the official lists of commercial seafood designation. A comparative analysis of the official lists published by the four major consumers and producers of cephalopod among EU countries was set in order to verify the accuracy of the data reported. The second study, conducted with the collaboration of a local Ministerial Board Inspection Post, aimed at verifying the applicability of the DNA barcoding technique as a Physical Check tool within the ordinary inspective procedures and at evaluating the mislabeling rate of seafood imported from Third countries. The third study aimed at assessing the efficiency of universal primer pairs currently applied for amplification of the major DNA fragments targeted in seafood identification. The study was conducted as a preliminary step to the development of a Next Generation Sequencing (NGS) based method for species discrimination in multispecies products such as surimi or fish balls made of a combination of fish and cephalopods. Overall, results highlight a general lack of harmonization among the analyzed lists and several shortcomings in the denominations. Cephalopods were the most mislabeled products: a mislabeling rate of 40,8% highlights major limits in the traceability system of supplier countries. 16SrRNA primer pairs showed the highest coverage capacity on the annealing region and a standard efficiency even between phylogenetically distant orders, confirming the gene universality and its suitability for the NGS analysis finalized to seafood species detection. In conclusion, given the lack of harmonization in cephalopods denominations and the high mislabeling rates for this kind of seafood, applying DNA based methods for the physical check procedures by both exporting operators and official inspectors would significantly improve seafood traceability and reduce falsification phenomena. Besides, the upgrading of analytical tools designed for the analysis of complex processed food matrices is needed. In this perspective NGS techniques could effectively become a turning point in the food inspection field, overcoming the limits of the standard DNA barcoding in the detection of multispecies matrices.