

Supplementary material

Fish DNA Sensors for Authenticity Assessment—Application to Sardine Species Identification

Myrto Kakarelidou¹, Panagiotis Christopoulos¹, Alexis Conides², Despina P. Kalogianni^{1,*} and Theodore K. Christopoulos^{1,3,*}

¹ Analytical/Bioanalytical Chemistry & Nanotechnology Group, Department of Chemistry, University of Patras, Rio, 26504 Patras, Greece; myrtokakarelidou@gmail.com (M.K.); pkchristop@gmail.com (P.C.)

² Hellenic Centre for Marine Research, Institute for Marine Biological Resources, 46.7 km Athens-Sounion, Anavyssos, 19013 Attika, Greece; conides@hcmr.gr

³ Institute of Chemical Engineering Sciences, Foundation for Research and Technology Hellas (FORTH/ICE-HT), Platani, 26504 Patras, Greece

* Correspondence: kalogian@upatras.gr (D.P.K.); tchrist@upatras.gr (T.K.C.)

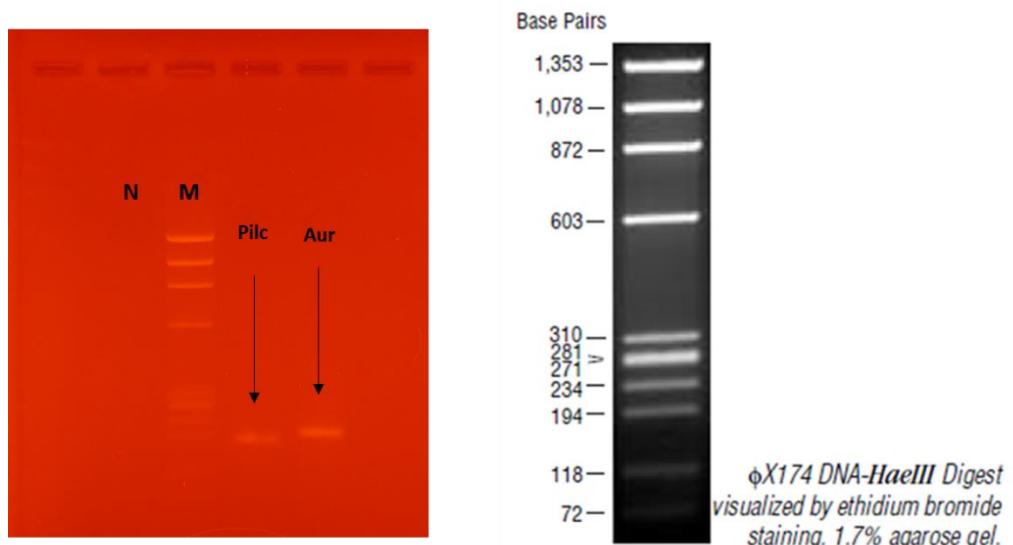


Figure S1. Electropherogram of PCR products for *S. pilchardus* with *S. aurita*. N: negative, M: DNA marker (ϕ X174 DNA HaeIII Digest on the right), Pilc: *S. pilchardus* and Aur: *S. aurita*.

Mixtures of PCR products

% Content of S. aurita in S. pilchardus

DNA sensor with S. pilchardus probe

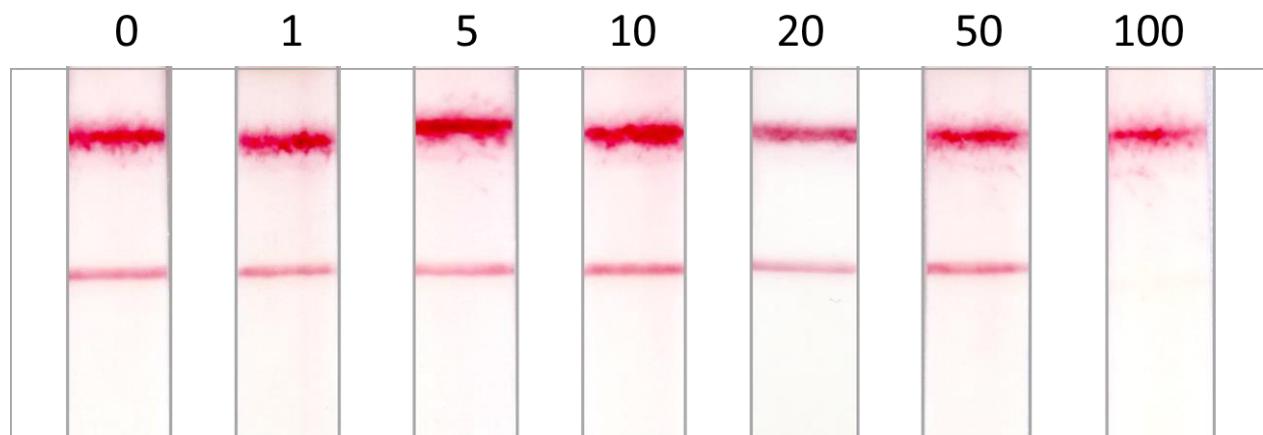


Figure S2. Mixtures of PCR products. The mixtures contained 0 – 100 % PCR product from *S. aurita* in PCR product from *S. pilchardus* with a total amount of 100 fmol on the strip.

Mixtures of processed samples

% Content of S. aurita in S. pilchardus

DNA sensor with S. pilchardus probe

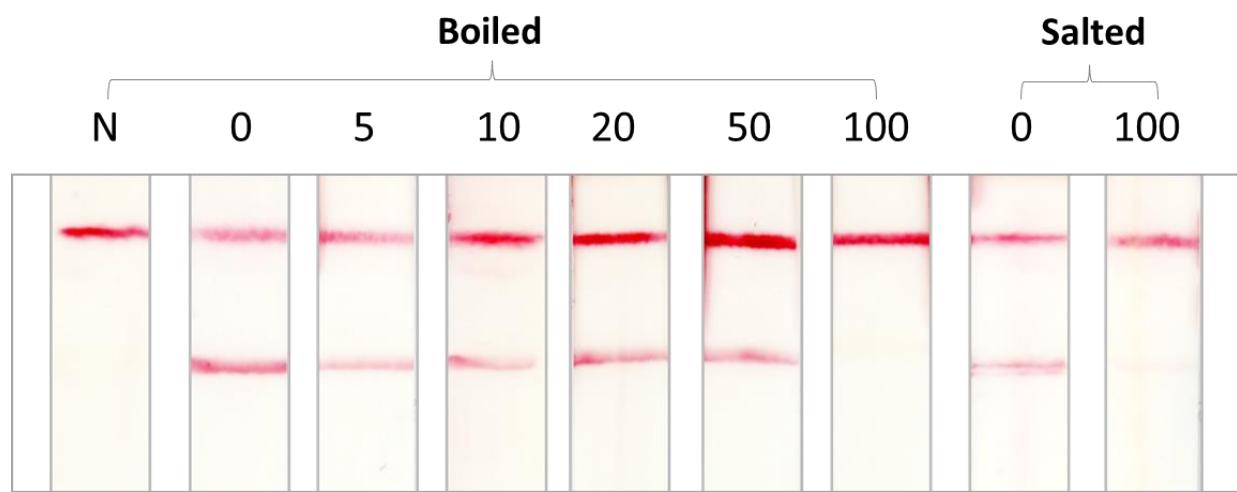


Figure S3. Mixtures of processed samples. The mixtures contained 0 – 100 % of *S. aurita* (tissue) in *S. pilchardus*.

Table S1. Comparison of methods for sardines' adulteration detection

| Method | Species examined | LOD | Discrimination capability | Quantitation capability | Ref. |
|--|---|-----|--|-------------------------|-----------|
| PCR-RFLP and phylogenetic analysis | <i>S. pilchardus</i> <i>S. aurita</i> <i>S. melanostictus</i> <i>S. caeruleus</i> <i>S. maderensis</i> | - | Low | - | 12 |
| PCR-RFLP and DNA sequencing | <i>S. pilchardus</i> <i>S. aurita</i> <i>S. brasiliensis</i> <i>S. sagax</i> <i>S. caeruleus</i> | - | Low for PCR-RFLP and high for DNA sequencing | ✓ for DNA sequencing | 13 |
| PCR-RFLP and phylogenetic analysis | <i>S. pilchardus</i> <i>S. aurita</i> | - | Low | - | 14 |
| DNA sequencing and phylogenetic analysis | <i>S. pilchardus</i> <i>S. aurita</i> | 5% | High | ✓ | 6 |
| DNA sequencing and phylogenetic analysis | <i>S. pilchardus</i> <i>S. aurita</i> <i>S. sagax</i> <i>S. caeruleus</i> <i>S. melanostictus</i> <i>S. maderensis</i> <i>S. longiceps</i> | - | High | ✓ | 15 |
| DNA sequencing and phylogenetic analysis | <i>S. pilchardus</i> <i>S. aurita</i> <i>S. longiceps</i> <i>S. lemuroides</i> <i>S. brasiliensis</i> <i>S. gibbose</i> <i>S. jussieu</i> <i>S. fimbriata</i> <i>S. tawilis</i> | - | High | ✓ | 16 |
| PCR and agarose gel electrophoresis | <i>S. pilchardus</i> | - | Medium | - | 17 |
| PCR and agarose gel electrophoresis | <i>S. pilchardus</i> <i>S. aurita</i> <i>S. maderensis</i> | - | Medium | - | 18 |
| PCR and agarose gel electrophoresis | <i>S. pilchardus</i> | - | Medium | - | 19 |
| Exon-primed intron-crossing (EPIC) PCR with acrylamide gel electrophoresis | <i>S. pilchardus</i> | - | Medium | - | 20 |
| Real-time PCR / SYBR Green | <i>S. pilchardus</i> | - | Medium | ✓ | 21 |
| Real-time PCR / Taqman probes | <i>S. pilchardus</i> <i>S. aurita</i> | - | High | ✓ | 5 |
| Real-time PCR / Taqman probes | <i>S. pilchardus</i> | - | High | ✓ | 22 |
| Real-time PCR / Melting curve analysis | <i>S. pilchardus</i> | - | High | ✓ | 23 |
| Real-time PCR / High-resolution Melting curve analysis (HRM) | <i>S. pilchardus</i> | - | High | ✓ | 24 |
| DNA sensor | <i>S. pilchardus</i> <i>S. aurita</i> | 5% | High | - | This work |