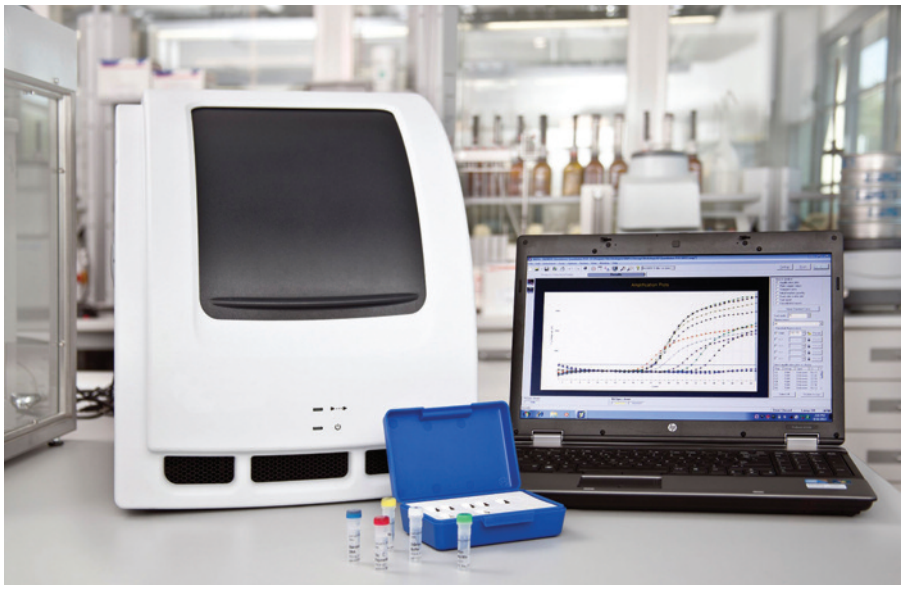
# Fish identification: what are you really buying?

***by the technical team, R-Biopharm Rhone Ltd, Block 10 Todd Campus, West of Scotland Science Park, Acre Road, Glasgow, Scotland G20 0XA, UK.***



For many, food species identification may appear to be a fairly recent problem, but for some this has been very important for thousands of years. Since ancient times, Jews have had to be able to identify certain types of fish to allow them to follow a kosher diet according to Jewish dietary laws.

In much more recent times, the horse meat scandal in 2013 highlighted how relatively easy it is for certain food suppliers to abuse systems and supply chains to substitute lesser quality and value products for more expensive products, increasing their profit margins in the process. This sparked a huge enquiry and a massive increase in the volume of meat species identification testing.

Since then, the food industry has looked at what other types of food could be mislabelled. Filleted fish are often impossible to identify visually without their tell-tale skins, fins and other identifying characteristics.

This means that the only way to identify them is through molecular methods. The results of eating mislabelled fish can cause serious health conditions and even be fatal for some people.

## The scale of the problem

The international organisation, Oceana, published a report in 2013 as a result of a two year investigation into seafood fraud in the US.

Over a third of the seafood samples they analysed were mislabelled. Snapper and tuna were most frequently mislabelled of all the species tested. Sushi restaurants had the worst record for mislabelled fish at 74%.

This problem has been shown to occur internationally with studies in numerous countries highlighting very similar issues.

In Canada, a study of fish sold in grocery stores found that 34 of 153 fish samples were mislabelled. Scientists from the University College Dublin in 2012 showed that 7% of cod products in the UK were mislabelled and the researchers were extending their investigations to other products including hake, tuna and monkfish.

The Australian government found that 23% of fish samples were mislabelled there.

Fishing is now a global industry and tests across Europe have identified fish species, which had never previously been commercialised, in the food chain.

One of the lead researchers in the University College Dublin study found that cod had been substituted by fish such as Vietnamese Pangasius.

While the majority of fish mislabelling is due to fraud, some instances are down to the difference between labelling laws around the world.

There are numerous differences between EU and US laws. For example, the mixing of two fish species in a tin may be against European Law, but a tin of 'light tuna' can legally contain a number of tuna species in the US.

In December 2014 European legislation 1379/2013 came fully into force. It specifies that the mandatory information required for the consumer or caterers is 'the commercial designation of the species and its scientific name'.

## Available technologies

For vertebrate animal species identification, different immunological methods exist. It is possible to use ELISA or lateral flow to detect specific proteins in the meat as these types of systems show an analytical sensitivity of approximately 1%.

However, processed food is problematic as the proteins are denatured by the heat and chemical elements within that process.

Mass spectrometry is a highly complex analytical system that detects the

*Continued on page 8*

*Continued from page 7*
fingerprints of specific cleaved peptide fragments. This is a much more sensitive method but only a few highly specialised laboratories are able to do this.

DNA is considered to be the most accurate analytical target and provides a reproducible means of differentiating one species from another. It generally remains unaltered in cooked and processed foods.

This precise method of testing is required because, aside from the potentially highly dangerous health risks associated with some people eating the wrong type of fish, this is also fraud.

The repercussions of this fraud grows with each person or company that purchases the mislabelled fish as it makes its way through the supply chain.

## Qualitative real-time PCR

Qualitative real-time PCR is the method of choice for fish species detection more than for land living species. Real-time PCR is an established technology in food analysis, widely used for GMO, allergen, pathogen and clinical analysis. Qualitative real-time PCR enables a highly sensitive and specific identification of the animal species in the presence of meat from other species. The nucleus and the mitochondria in each fish cell contain the entire genetic code and out of this blueprint specific information can be amplified.

The released fluorescent dye of the probe will enhance an exponentially growing optical signal. In a real-time thermocycler device this process is performed automatically in closed tubes or plates and after an hour, or 35-45 cycles, the analyst may detect the signal indicating that the specific DNA was present in the sample.

The in silico design of fish species primers is dependent on suitable public databases like GenBank or BOLD. Furthermore, validation requires real samples that include the species of interest and exclude related but different fish species.

The databases are not free from errors and some fish species are protected, so the development of new tests takes longer than, for example, land-living animals. A test for black and white halibut is already available. New tests for species like cod, tuna and haddock are expected to be available this year.

## In-house testing

R-Biopharm offers comprehensive DNA based detection methods for food and agricultural analysis through its company, Congen Biotechnologie GmbH, an innovative biotechnology company specialising in molecular analysis.

The laboratory has been accredited by DAR (Germany Accreditation Council) in accordance with DIN EN ISO/IEC 17025:2005. The service offers a rapid turnaround from receipt of the sample and also an extremely sensitive detection limit depending on matrix and DNA preparation.

There is also a wide range of PCR test kits available for in-house testing, sold under the SureFood and SureFast brand names worldwide. They integrate effective sample preparation with high quality real-time PCR (both qualitative and quantitative).

They cover all relevant applications in the fields of animal and plant speciation as well as allergens, GMOs and pathogen detection. A broad selection of kits are available for next day delivery. ■