

Fish content and QUID

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Introduction

This note is intended to give guidance to seafood buyers on the chemical composition of seafood products so that they may understand the risks of product extension or dilution that can happen in their upstream supply chains and they are able to develop effective analytical mitigation measures to protect themselves against potential acts of fraud.

Soak and injection processes, sometimes using salt (sodium chloride) or possibly using other chemical additives can reduce the analytical fish content of seafood significantly and there is anecdotal cause for concern that the use of production technologies and 'magic powders' to enhance the water retention capabilities of fish muscle to produce low fish content product is becoming increasingly common. In some international markets there is an acceptance of this practice as a means of value engineering product to meet low price points, and manufacturers of primary processed fish products are able to deliberately produce to specifications targeting fish contents as low as 60% to meet this demand.

The increasing presence of low fish content product in the international marketplace represents a risk to businesses that are seeking to buy 100% natural, untreated product. The unwitting purchase of treated product by these businesses could result in the sale of undeclared water to the consumer and the presence of undeclared (possibly unlicensed) food additives. Given that food manufacturers have legal obligations under Article 22 of the Food Information to Consumers Regulation EU 1169/2011, relating to the quantities of ingredients used in the manufacture or preparation of food, it is important that any business selling food to the consumer fully understands the true compositional nature of their product so that their food labels are legal, demonstrably accurate and not misleading to the public.

Basic fish muscle composition

The science of understanding the classes of substances present in food is generally referred to as 'proximate composition' – this aggregates all of the individual sub-components, such as different types of proteins or fats into their five higher level orders: proteins, water, fats, minerals and carbohydrates. Seafood has been studied at a compositional level for many years, so thankfully there is a wealth of reference data to draw upon when analysing and comparing products to assess whether or not they meet typically expected parameters.

The three principal proximate components of fish flesh are:

- Water
- Fat
- Protein

However, no two fish are alike and the ratios of these components do vary between species and even between different animals of the same species. There can also be seasonal variations within a fish population during the year, for instance because of the natural changes that happen to fish muscle during spawning and migration cycles. When protein and fat levels become naturally depleted because of the energy demands of spawning or migration, they will typically be replaced with water, and vice versa once the cycle is completed and typical levels are restored. By observing these interactions throughout the lifecycle, it is possible to develop benchmark average values and to observe the typical ranges of variability.

As examples:

	Atlantic cod	Herring
Water	78 – 83%	60 – 80%
Fat	0.1 – 0.9%	0.4 – 22.0%
Protein	15.0 – 19.0%	16.0 – 19.0%

Reference data for water, fat and protein levels for a number of commercial fish species can be found from several different sources;

FAO web page: <http://www.fao.org/wairdocs/tan/x5916e/x5916e01.htm>

UK national reference data is available in McCance & Widdowson's 'composition of foods integrated dataset' on the nutrient content of the UK food supply. This can be used by food manufacturers for reference as the data is nationally recognised as typical of the analysed products that are listed. Data is available to download and includes a wide range of seafood species and product formats:

<https://www.gov.uk/government/publications/composition-of-foods-integrated-dataset-cofid>

Farmed fish do not undertake migrations and they are typically harvested prior to any morphological changes that would be triggered by their spawning cycle, so they are less susceptible to this natural, cyclical variance in proximate composition. However, these fish may also exhibit variations for other reasons, particularly in their protein and fat levels in accordance with different feed regimes and market specifications.

Fish that have been treated with chemical additives or which have been badly processed will lose a measurable amount of protein in percentage terms either through dilution (through treatment) or through the loss of soluble protein (through for example, the use of excessive water in processing). In turn, this becomes manifest as an apparent low fish content with a corresponding increase in the water content when subject to laboratory analysis.

Because it is possible to artificially manipulate the balance of these three components during processing – typically through the addition of water, it is important that seafood buyers have an understanding of the natural levels and the typical ranges of variability of these three components in order to be assured that they are buying product that is in accordance with their specification.

How do we test analytical fish content?

The concept of Analytical Fish Content is based on the principle of using nitrogen as a proxy for fish protein content, so the product must first be analysed in a laboratory to determine the amount of nitrogen present. There are two main laboratory methods currently used to determine nitrogen levels, Kjeldahl and Dumas.

Kjeldahl method – this traditional analytical method uses ‘wet chemistry’ to liberate nitrogen as ammonium sulphate by heating a substance with sulphuric acid.

Dumas method – in the Dumas method, a substance is heated to very high temperatures to release carbon dioxide, water and nitrogen. The Dumas method has a number of advantages over Kjeldahl; it does not require the use of toxic chemicals, it is relatively fast and the process can be fully automated. Because of these advantages, Dumas has largely replaced the Kjeldahl method in popular usage in the UK, although Kjeldahl is still widely used in international laboratories. Because the Dumas method determines total Nitrogen including inorganic fractions like nitrite and nitrate, and the Kjeldahl method only recovers organic nitrogen and ammonia, the different methods will generate different results for N levels. It is important therefore, that these analyses are conducted by laboratories that have achieved accreditation for the appropriate methodology (such as that provided by the United Kingdom Accreditation Service, UKAS).

Near Infrared Reflectance (NIR) spectroscopy - this is useful as a real-time quality inspection method for the analysis of nitrogen in a sample. NIR spectroscopy is performed by equipment that measures the interaction between a sample and infrared light that has been dispersed into individual wavelengths, usually by a prism. For many years, this technology has been widely used to evaluate the protein strength of wheat flour and with the correct calibration curves, it can be used to detect Nitrogen in seafood products and determine analytical fish content.

Calculating analytical fish content

Analytical fish content is the estimate of fish content based on the amount of Nitrogen (N) as determined by chemical analysis. Nitrogen is one of the easiest parameters to test for when measuring protein and, consequently, analytical fish content.

In the calculation of analytical fish content, it is important to understand which chemical method has been used to determine the nitrogen level (Kjeldahl or Dumas) because the two methods generate slightly different results. Most laboratories in the UK use the rapid Dumas method for nitrogen determination. Dumas measures the non-protein nitrogen as well as the protein nitrogen, and hence gives slightly higher results (by a factor of 1.014). This is important for seafood because of its higher non-protein nitrogen content than meat.

Fish content is calculated thus:

- a) Stubbs and More calculation – this simple method is widely accepted and is easily understood

$$\% \text{ Fish Content} = \left(\frac{N - 0.02}{N \text{ factor}} \right) 100$$

The 0.02 adjustment applies only if there is known to be carbohydrate present, as in the flour in batter mixes and breadcrumbs used on coated fish - it represents the nitrogen present in these carbohydrates. If the sample is known to be raw fish then the 0.02 adjustment is omitted.

- b) Full calculation – this is the more complex calculation method approved by the Royal Society of Chemistry

$$\left(\frac{((N - (100 - X)0.02) - XN)}{N \text{ factor}} \right) 100$$

Where; $X = ((N \times 6.25) + M + A + F)$

M = % Moisture, A = % Ash, F = % Fat, N = % Nitrogen, XN = % non-fish protein

For fish such as farmed salmon with a high fat content, it is permitted to add the fat into the fish content. This only applies when the natural fat content of the sample is found to be >5%.

The nitrogen factor

For many years, the protein content of foods has been determined on the basis of total nitrogen content. Nitrogen content is then multiplied by a factor to arrive at protein content. On the basis of early determinations, the average nitrogen (N) content of proteins was found to be about 16 percent, which led to use of the calculation $N \times 6.25$ ($1/0.16 = 6.25$) to convert nitrogen content into protein content.

This use of a single factor, 6.25, has since been superseded on the basis of two considerations. First, not all nitrogen in foods is found in proteins: it is also contained in variable quantities of other compounds, such as free amino acids, nucleotides, creatine and choline, where it is referred to as non-protein nitrogen (NPN). Second, the nitrogen content of specific amino acids (as a percentage of weight) varies according to the molecular weight of the amino acid and the number of nitrogen atoms it contains (from one to four, depending on the amino acid in question). Based on these facts, and the different amino acid compositions

of various proteins, the nitrogen content of proteins actually varies from about 13 to 19 percent. This would equate to nitrogen conversion factors ranging from 5.26 to 7.69.

In response to these considerations, $N \times 6.25$ has been abandoned and replaced by $N \times$ a factor specific for the food in question.

The value used for the N factor is the mean value of the percentage nitrogen content of the species of fish used. Once the level of nitrogen has been determined, the N Factor is applied to convert the nitrogen content of the product into an analytical fish content figure.

N Factors are used to determine the true meat content for a number of food products such as pork, beef, lamb and chicken and there are different N factors for each species. Because of the number of species involved and their inherent natural variability, it is rather more complex to evolve nitrogen factors for seafood, although a number of these were listed as interim values in the "Code of Practice for the Declaration of Fish Content in Fish Products" which was published in 1998.

Since 1998, there have been a number of efforts to review and add to the published list of N Factors, most recently in 2012 when Defra commissioned the Royal Society of Chemistry (RSC) to evaluate N factors for two increasingly popular species in the UK; Pangasius (*Pangasius hypophthalmus*) and Alaska pollack (*Theragra chalcogramma*), and in 2014 when the RSC Nitrogen Factors Sub-committee published their paper on seafood nitrogen factors (Analytical Methods Committee AMCTB No 62).

See Appendix 1 for the published list of RSC AMCTB No 62 Nitrogen Factors.

Factors are calculated as the mean nitrogen level from a statistically significant number of samples from a single species of known provenance, i.e. not phosphated or subjected to other treatment, and produced according to Good Manufacturing Practice (GMP), which is defined by the Code as 'produced in accordance with operating practices which do not seek to debase the product unnecessarily in relation to composition, quality, form or texture', or as whole unprocessed fish. This methodology gives a norm value taking into account the biological changes in fish muscle caused by spawning condition and of normal processing conditions. These official factors are adopted into CODEX¹ and are internationally applicable and accepted. Because the Nitrogen factor for any species is a mean figure and is based on official sampling of product taken throughout the entire annual spawning cycle and thereby taking account of the natural changes in Nitrogen levels present, it is expected to find some variation around that mean when analysing individual samples.

For species where no published N factor exists, manufacturers may determine proxy values of their own, subject to assurance that the product used to establish that factor is of genuine provenance and has been manufactured in accordance with the principles of GMP. Any proxy nitrogen factor values must be based on

¹ CODEX or 'Codex Alimentarius' (the Food Code) was established by the Food and Agriculture Organisation of the United Nations in 1963, as a global reference point for consumers, food producers and processors, national food control agencies and the international food trade.

the levels found in a statistically significant sample, which reflects the full range of natural muscle condition variability caused by such factors as seasonal spawning changes.

Lower N Factor values are accepted for some products such as minced fish blocks (see Appendix 1 for details), where even under GMP it may be reasonably expected because of the nature of the processes involved, that a higher % of nitrogen will be lost during manufacturing.

Long-term trend analysis of analytical data indicates that it is reasonable to expect this method to produce a mean result of 100% analytical fish content with a +/-10% fluctuation for both IQF fillet and fillet block materials. This evidences the fact that the current N factors used are appropriate and that taken over a significant time period, the analytical fish content of any major commodity can be expected to meet the target average of 100%.

Other analytical indicators of low fish content product

There are a variety of additional indicators of low fish content that can be used – if used, it is recommended that these methods are viewed as screening tests, possibly in conjunction with each other as a presumptive means of detecting added water in raw materials, and that any anomalies are then investigated more thoroughly by the empirical assessment of analytical fish content.

Water Content – the water content of a sample can be measured in a number of ways by a food chemistry analytical laboratory, and given that the main exposure to risk is the reduction of analytical fish content by deliberately increasing fish muscle water-holding capacity, it follows that the measurement of water as a percentage of the product is a good indicator of whether or not the product has been treated. This is especially useful when taken in conjunction with an analytical fish content result as for many white-fleshed fish species the predominant proximate balance is between the two principal components of water and protein.

pH testing – with some slight seasonal variation caused by stress levels during capture or seasonality, fish muscle tends to have a fairly stable pH balance. The post-capture pH of cod and haddock muscle for instance tends to be consistently around 6.7². The water-holding capacity of fish muscle can be increased by shifting the pH either towards acid or alkali. A number of proprietary technical ingredients have been developed to achieve this pH shift during primary processing, including citric acid and sodium bicarbonate based binders which are used as soaks or are injected into the fillets. Atypical pH tests can be used to indicate whether these proprietary binders have been used.

Sodium and chloride – most seafood naturally contains salt (NaCl or sodium chloride), but because brine soaks can be used as a means of enhancing water retention in fish muscle, higher than typical levels of sodium compounds in a product can be seen as an indicator of reduced fish content. Because there are a number of sodium based compounds that have water retaining qualities (not just salt), it is important to test product for sodium chloride and for total sodium levels.

² The Post-mortem pH of Cod and Haddock Muscle and its Seasonal Variation by R Malcolm Love (Torry Research Station), 1978

Polyphosphates

A phosphate is a salt of phosphoric acid; when a number of simple phosphate units are linked to form a more complex structure, this is known as a polyphosphate. The main value of polyphosphates lies in improving the retention of water by the protein in fish. There are concerns that as phosphates can be used to help retain moisture, then by the application of excess phosphates the retention of “added water” can lead to unfair trade practices resulting in economic fraud.

Polyphosphates E450 (Potassium and sodium di-phosphates), E451 (Potassium and sodium tri-phosphates) and E452 (Polyphosphates) are legally permitted in seafood in the EU, although they must be declared as additives, and if the level of water retention achieved exceeds 5% of the product weight, then the relative percentages of fish and water must also be declared.

Testing by ion chromatography can determine the presence of different types of polyphosphates, but for many businesses, this is considered a too specialised and expensive analysis to conduct on a routine basis. Commercial food chemistry laboratories can however conduct colorimetric testing for total phosphate – whilst this method does not distinguish between polyphosphate additions and naturally occurring phosphates, laboratories are able to reasonably determine how much phosphate has been added by comparing their results to literature references for the natural levels of phosphate in seafood. The results may also be viewed in conjunction with other analyses such as total moisture and analytical fish content to determine whether polyphosphates have been added to increase the water retention capacity of the fish.

Detailed advice on laboratory testing methods for polyphosphates can be found in the Seafish briefing note, ‘Review of polyphosphates as additives and testing methods for them in scallops and prawns’:
http://www.seafish.org/media/Publications/SR654_final.pdf

The Quantitative Ingredient Declaration (QUID) issue

QUID is a basic requirement set out in Article 22 of the Food Information to Consumers Regulation EU 1169/2011 relating to the *“indication of the quantity of an ingredient used in the manufacture or preparation of a food.”*

It is important to note that the indication of quantity does not simply refer to the gross weight of the ingredient added (core weight), but to the ingredient’s weight net of added water, including frozen glaze or other water retained within the product for instance, through soaking and injection processes. It is therefore incumbent on the manufacturer to ensure that their ingredient declarations accurately reflect the contents of the packages, and in order to do this, they must understand the true fish content of the materials they are using and in particular, whether they have been diluted through the addition of water. Due diligence in this respect would require that raw materials are empirically tested to ensure that they conform with the expected fish content levels that are required to meet the quantitative ingredient declaration on the pack.

It is accepted that even under Good Manufacturing Practice (GMP) conditions, seafood products will retain levels of technically unavoidable process water during their primary processing. These levels may differ between different processes and product formats, and the manufacturer should seek to establish what the

typical levels of technically unavoidable water are for their product ranges as in some circumstances this may invoke a legal requirement for the declaration of water in the product ingredient listing.

From a legal perspective, manufacturers' due diligence of compliance with QUID is based on their product formulations (recipes) and their 'mixing bowl' records as proof of their conformance to the recipe. Processors who use seafood as an ingredient in value added products such as coated fish or ready meals therefore need to know how much additional water (beyond that which is to be expected under GMP) is retained within the raw material in order for them to be able to fully declare the total amount of water they have added to the recipe.

In cases where the technically unavoidable water addition is greater than 5% of the product weight, and in cases where seafood products have been deliberately 'soaked' to enhance their weight through water absorption, it is mandatory that the added water must be listed in the ingredient declaration, even if the presentation to the consumer is a natural product format.

Regulatory and other monitoring activity

Enforcement authorities are guided by CODEX on this issue. CODEX advises that either strip testing, in which the fish ingredient is physically separated from the other ingredients, or chemical analysis based on Nitrogen may be used to determine fish content.

The UK Trading Standards Office has in the past included analytical fish content based on Nitrogen as part of their routine marketplace surveillance to verify the amount of fish declared to the consumer in the Quantitative Ingredient Declarations (QUID) as stated on the pack. This method is considered the most empirical way to determine fish content (as opposed to strip testing), particularly for value added seafood products that have been inextricably combined with other ingredients such as coatings or sauces.

Given the increased focus on food integrity issues since the horsemeat incident of 2013, some UK and EU retailers have increased their product authenticity surveillance testing regimes. There is some evidence from their suppliers in the marketplace that these major retailers are now placing a greater emphasis on analytical testing rather than traditional factory floor inspection of Process Control evidence as a means of verifying the proximate composition of food products in their own label ranges.

Good practice – practical advice to seafood buyers

In recent years an international market for low fish content fish products has been developing and there are now significant quantities of seafood commodities traded which have been treated to increase their gross weight through enhancing the water-holding capacity of fish muscle. It is entirely possible that such products could be manufactured and sold legally, provided that they comply with EU rules and labelling requirements, but because it may not be possible to tell the difference between treated and untreated materials without conducting laboratory tests, these materials may be entering food chains without the buyers being aware of their low protein levels, and the potential that they may contain food additives.

It is incumbent upon the buyers therefore, to protect themselves against such mis-selling through the application of risk-based due diligence measures. Such measures may include:

- Ensure that you have a formal, legally binding specification agreement with the supplier – this specification should include the analytical fish content of the material to be purchased and an agreement on the Nitrogen factor and assessment method used to verify this
- Unless agreed otherwise, ensure that the technical specification specifically prohibits any soaking, injection or any other form of chemical intervention which reduces the natural ratio of fish protein to water
- Ensure that empirical tests are carried out against a risk-based sampling plan to ensure that purchased raw materials meet the agreed technical specification for proximate composition (bearing in mind natural variability)
- Ensure that Quantitative Ingredient Declarations are calculated on the basis of the true fish content of the raw material used and keep laboratory records to prove that materials conform with this value
- Conduct audits and inspections to ascertain the supplier workshop capability of producing low fish content products and in the case where this is possible, to agree how treated and non-treated products are kept segregated.

Appendix 1 – Royal Society of Chemistry Nitrogen Factors Sub-committee

(AMCTB No 62)

Summary of published nitrogen factors for GMP fish ingredients

Species	N Factor (Kjeldahl)
Cod fillet block	2.75
Minced cod block	2.67
Scampi (Nephrops)	2.45
Atlantic salmon (fat-free) fillet	3.75
Atlantic salmon mince (fat-free)	2.81
Tilapia fillet	2.88
S. Atlantic hake fillet block	2.45
S. Atlantic hake mince block	2.33
Pangasius fillet	2.66
Alaska pollack fillet block	2.64
Alaska pollack mince block	2.47

Interim nitrogen factors awaiting confirmation

Species	N Factor
Coley/saithe	2.69
European hake	2.64
Haddock	2.72
Ling	2.78
Whiting	2.68
White fish mean	2.65

References and further reading

- Code of Practice on the Declaration of Fish Content in Fish Products (1998), UK Association of Frozen Food Producers, BFFF, BRC, BHA, Seafish, LOCTS, Association of Public Analysts, http://www.seafish.org/media/Publications/Fish_Content_CoP.pdf
- Royal Society of Chemistry, Seafood Nitrogen Factors, Analytical Methods, 2014, 6, 4490; http://www.rsc.org/images/TB%2062_tcm18-241512.pdf
- FAO FOOD AND NUTRITION PAPER 77. Food energy - methods of analysis and conversion factors. Report of a Technical Workshop, Rome, 3-6 December 2002; <http://www.fao.org/docrep/006/y5022e/y5022e03.htm>
- Polyphosphates in Fish Processing by A Aitken – Torry Advisory Note no.31 (Revised); <http://www.fao.org/wairdocs/tan/x5909e/x5909e00.htm#Contents>
- McCance and Widdowson's 'composition of foods integrated dataset' on the nutrient content of the UK food supply; <https://www.gov.uk/government/publications/composition-of-foods-integrated-dataset-cofid>
- CODEX Standard for Quick Frozen Fish Sticks (Fish Fingers), Fish Portions and Fish Fillets - Breaded or in Batter *CODEX STAN 166 – 1989*; http://www.seafood.nmfs.noaa.gov/pdfs/frozen_fishsticks_portions_fingers_fillets.pdf

Further information from Seafish

- Review of polyphosphates as additives and testing methods for them in scallops and prawns – http://www.seafish.org/media/Publications/SR654_final.pdf
- Food Information for Consumers - http://www.seafish.org/media/Publications/Seafish_labelling_update_2016_v2_1.pdf

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