



**Agenda Item 11**

**CX/FFP 14/33/13**

**JOINT FAO/WHO FOOD STANDARDS PROGRAMME  
CODEX COMMITTEE ON FISH AND FISHERY PRODUCTS**

**Thirty-third Session  
Bergen, Norway  
17 – 21 February 2014**

**DISCUSSION PAPER ON NITROGEN FACTORS FOR THE CHEMICAL ANALYSIS METHOD LISTED IN THE STANDARD FOR QUICK FROZEN FISH STICKS (FISH FINGERS), FISH PORTIONS AND FISH FILLETS – BREADED OR IN BATTER (CODEX STAN 166-1989)**

*Prepared by the United States of America in cooperation with the United Kingdom and New Zealand*

**BACKGROUND**

1. The *Codex Standard for Quick Frozen Fish Sticks (Fish Fingers), Fish Portions and Fish Fillets – Breaded or in Batter (STAN 166-1989)* (Fish Stick Standard), has the following provision in the Labelling Section:

In addition to Sections 2, 3, 7, and 8 of the *General Standard for the Labelling of Prepackaged Foods (CODEX STAN 1-1985)* the following specific provisions apply:...

*6.1.3 The proportion of fish content should be declared on the label...*

2. The *Estimation of Fish Content* section of the Standard lists the *Codex-Adopted Method (AOAC Method 996.15)* as the primary method, and two alternative methods used in special situations, the *Chemical Analysis Method (Nitrogen Factor End Product Method)*, and the *Rapid Method Used during Production*.

3. At the 26<sup>th</sup> CCFFP Session (2003), the *Discussion Paper on Fish Content in Fish Sticks (CX/FFP 03/14)*<sup>1</sup> presented arguments in support of, and in opposition to, using chemical analysis to determine fish content. Subsequently it was agreed to include the chemical analysis in the Standard according to Appendix VII of the 26<sup>th</sup> *Session Report (ALINORM 04/27/18)*<sup>2</sup>.

4. The chemical method for determination of fish content is based on the level of nitrogen (a marker of protein) in the product. The term “nitrogen factor”, as used in the Fish Stick Standard, is the percentage nitrogen by weight expected in a particular fish ingredient.

5. A table of interim whitefish nitrogen factors was included in the Standard (26<sup>th</sup> CCFFP Session), and at the 31<sup>st</sup> CCFFP Session (2011) a nitrogen factor for tilapia was added. At the 32<sup>nd</sup> CCFFP Session (2012)<sup>3</sup>, it was agreed to add nitrogen factors for South Atlantic hake (CX/FFP 12/32/9)<sup>4</sup>; however there was disagreement with the methods used to determine the nitrogen factors and discussion about the need for different nitrogen factors, and their effectiveness for determining fish content. *After some discussion, the Committee agreed that a discussion paper would be prepared by the United States of America, the United Kingdom and New Zealand with assistance from other interested members and observers, for discussion at*

<sup>1</sup> CX/FFP 03/14, Discussion Paper on Fish Content in Fish Sticks

Link: [ftp://ftp.fao.org/codex/meetings/CCFFP/CCFFP26/fp03\\_14e.pdf](ftp://ftp.fao.org/codex/meetings/CCFFP/CCFFP26/fp03_14e.pdf)

<sup>2</sup> Report of the Twenty-Sixth Session of the Codex Committee on Fish and Fishery Products (13-17 October 2003)

Link: [ftp://ftp.fao.org/codex/Reports/Alinorm04/al04\\_18e.pdf](ftp://ftp.fao.org/codex/Reports/Alinorm04/al04_18e.pdf)

<sup>3</sup> Report of the Thirty-Second Session of the Codex Committee on Fish and Fishery Products (1-5 October 2012)

Link: [ftp://ftp.fao.org/codex/Reports/Reports\\_2013/REP13\\_FFPe.pdf](ftp://ftp.fao.org/codex/Reports/Reports_2013/REP13_FFPe.pdf)

<sup>4</sup> Proposed Draft Amendment to the Standard for Quick Frozen Fish Sticks (Nitrogen Factor for South Atlantic Hake)

Link: [ftp://ftp.fao.org/codex/meetings/ccffp/ccffp32/fp32\\_09e.pdf](ftp://ftp.fao.org/codex/meetings/ccffp/ccffp32/fp32_09e.pdf)

*its next session. It was agreed that the discussion paper should address the usefulness of nitrogen factors; and the need to review, as appropriate, the list of existing nitrogen factors contained in the Table of the Standard.*

6. Members of the drafting group did not come to agreement on the details of a long discussion paper; therefore this paper briefly outlines the areas discussed that lead to agreed recommendations for consideration by CCFFP.

## **DISCUSSION**

### Codex Procedure

7. The acceptance procedure for proposed new “nitrogen factors” is time consuming because the studies are presented to the CCFFP and discussed over one or two sessions. Once agreed, the proposed amendment is published in the Meeting Report and approved by the Codex Alimentarius Commission before the Fish Stick Standard can be amended and published on the Codex website. If there are species or harvest areas specific to certain Codex member countries, then they have the option of providing the data for a new nitrogen factor directly to CCFFP for consideration. This implies a long term programme of work for the CCFFP to revise and add new nitrogen factors to the Standard. If the studies were performed in a uniform manner and already published in a peer reviewed scientific journal, this discussion could be briefer, or may not need to occur.

### Use of methods

#### *Relationship between methods:*

8. It is understood that the AOAC gravimetric method and the chemical nitrogen method measure different things (fish core content versus nitrogen content) and have different purposes, which is why they are both listed in the Standard. The fish ingredient (core) is estimated on a weight basis using the gravimetric method. The nitrogen method is used when there is reason to doubt the composition of the fish core (i.e., it may not be 100% fish).

9. At the 31<sup>st</sup> CCFFP Session (2011), the wording of Section 7.4 of the Standard was changed *to better clarify when to use the different methods*. There was no discussion or intent to change the agreed relationship between the methods as indicated in the original wording from the 26<sup>th</sup> Session (2003), and endorsed by CCMAS and CCFL:

#### *Estimation of fish Content*

*According to AOAC Method 966.15. In cases where there is some remaining doubts over the composition of the fish core then the method of analysis as outlined below could be used, i.e. as a reference method...*

#### *Checking of fish content by chemical analysis...*

10. However, the 31<sup>st</sup> Session revision adds ambiguity about method use, and further revision or return to previously agreed wording is needed.

#### *Natural variance of fish nitrogen content:*

11. The natural levels of protein (and nitrogen) in fish flesh fluctuate depending on factors such as season and location of catch, size, spawning cycles, and nutrition; and in the case of farmed fish, on the culture conditions. Currently, the nitrogen factors in the Standard have a +/- 10% allowance for natural nitrogen variation; however, it was identified that this allowance does not adequately cover all the natural variance observed, and it was suggested that the allowance should be statistically based and should cover 2 standard errors about the mean (approximately 95% of the expected distribution).

12. It was also suggested that the Standard should indicate that any suspect results from the chemical method should trigger in plant verification of the fish content on a recipe basis. It was identified that this is how the chemical method is currently used in practice.

#### *AOAC method adjustment factors:*

13. The 23<sup>rd</sup> Session of the CCFFP agreed to include adjustment factors for the AOAC Official Method (ALINORM 99/18, paragraph 13):

*13. The Committee agreed to include in the standard a reference to AOAC Method 996.15 with an adjustment factor of 2% for raw breaded and batter-dipped products; 4% for precooked products, subject to endorsement by the CCMAS.*

14. These factors adjust for moisture migration from the fish core into the breading, and should be included with the AOAC method listed in the Standard.

#### Methodology for determining nitrogen factors

15. It is agreed that loss or addition of water during processing has a significant effect on nitrogen levels. However, there are different viewpoints on if nitrogen factors should be determined from dry fresh fillets, or from processed fillet and mince blocks. One viewpoint is that the nitrogen factor of a 'fish ingredient' should recognise the necessary use of water for the hygienic preparation of fish and seafood and the unavoidable pick-up of some processing water. The other viewpoint is that processed fish with unavoidable water pick-up should not be recognized as 'fish ingredient', but as 'fish ingredient' and 'water ingredient', and that the nitrogen factor should be based on dry fish in order to more accurately determine the total amount of "water ingredient".

16. Nitrogen factors based on processed fish are appropriate to use to distinguish added water above that which is unavoidable during processing under GMPs. Nitrogen factors based on unprocessed fresh fillets, which have not been exposed to water or ice, are appropriate to use to estimate total added water.

17. The Fish Stick Standard currently lists some nitrogen factors based on fresh fillets with a ~8% reduction to allow for loss during processing, and other nitrogen factors based directly on processed fish blocks that underwent icing and washing under good manufacturing practices. We recommend also listing nitrogen factors based on dry fresh fillets. The methodology to determine "dry" nitrogen factors can be more easily standardized because the variance in wet processing at different locations is excluded.

18. The studies for the existing nitrogen factors looked at each species separately, however the range in values for different species show significant overlap. This overlap raises the question if it may be possible to reduce the number of nitrogen factors, or number of new nitrogen factors, if the individual nitrogen factors are not statistically different.

19. The collection of samples for the determination of a nitrogen factor should take into account the natural variables discussed in the Natural variance of fish nitrogen content section (above); as well as the variables in handling and processing for nitrogen factors based on processed products.

## **RECOMMENDATIONS**

### Codex Procedure

20. In order to make more efficient use of CCFPP session time, the Drafting Group recommends to retain the principle of the chemical analysis method (nitrogen factor method) in the Standard, but remove the table of nitrogen factors from the Standard and, instead, make reference to an external reference source. The external reference source would contain a published up-to-date table of nitrogen factors. The nitrogen factors would be based on studies published in peer reviewed journals, performed with appropriate uniform procedure. The table would be inclusive of species, harvest areas and dates, type of nitrogen factor (e.g., dry fillet, minced block), sample type (e.g., one fillet, 250g of block), number of samples, standard deviation, and study citation. Possibilities for external reference sources include:

1. The UK's Royal Society of Chemistry-Analytical Methods Committee Technical Brief on Nitrogen Factors. Publicly available on the following link:

(<http://www.rsc.org/Membership/Networking/InterestGroups/Analytical/AMC/TechnicalBriefs.asp>)

2. The FAO/WHO website, if they are able to maintain such information.

### Use of methods

21. The Drafting Group recommends that the use of methods be further clarified in the Standard. Annex A includes the proposed draft revision of Section 7.4 for consideration by the Committee.

22. Instead of the +/- 10% allowance currently listed with the Table of nitrogen factors, it is recommended that the uncertainty of each nitrogen factor should be taken into account by users from the statistical data presented with the published nitrogen factor (e.g., 2 standard errors about the mean).

23. It is recommended that the agreed adjustment factors for moisture migration be included with the AOAC method in the Standard, and these are included in the proposed amended draft (Annex A).

Methodology for determining nitrogen factors

24. The Drafting Group recommends that the appropriate procedure used to determine nitrogen factors for use with the Fish Stick Standard should be documented for use by CCFFP or third parties. Annex B includes an initial Proposed Draft Uniform Procedure to Determine Nitrogen Factors

25. We recommend that “dry” nitrogen factors, determined from freshly caught fish fillets that are not exposed to ice or fresh water and handled to minimize moisture loss, should be included with processed fish nitrogen factors; and we recommend that “dry” nitrogen factors (unprocessed) and “wet” (processed) nitrogen factors should be clearly differentiated in tables of nitrogen factors.

Possible further work

26. CCFFP may consider further work to:

1. Refine the Draft Uniform Procedure to Determine Nitrogen Factors
2. Arrange the format for publishing the list of nitrogen factors
3. Analyse current nitrogen factor data to determine standard errors, and other relevant statistical information.
4. Analyse the statistical validity of a single “dry” nitrogen factor for groups of species (i.e., whitefish).

**Annex A – Proposed Revision of Fish Stick Standard (Section 7.4)****7.4 ESTIMATION OF FISH CONTENT**Codex-Adopted Method**AOAC Method 996.15. (End Product Method)**

Calculation:

$$\% \text{ Fish Content} = (\text{Wd/Wb}) \times 100 + \text{Adjustment Factor}^*$$

Wd = weight of debattered and/or debreaded test unit

Wb = weight of battered and/or breaded test unit

\*Raw Breaded Frozen Coated Fish and Fishery Products: 2.0%

\*Batter-dipped Frozen Coated Fish and Fishery Products: 2.0%

\*Precooked Frozen Coated Fish and Fishery Products: 4.0%

Reference: J. AOAC Int. 80, 1235(1997)

Other Methods**(1) Chemical Analysis Method (Nitrogen Factor End-Product Method)**

Appropriate in cases where there is reason to doubt the composition of the fish core (i.e., appears to contain non-fish ingredients). This method requires confirmation with the Codex-Adopted Method, or with Method #2 (Rapid Method Used during Production) in conjunction with investigation at the processing plant when determining product compliance with the labelling provisions in this Standard. This method should trigger in-factory investigation (e.g. raw ingredient recipe checks) when suspect products are identified.

The percentage fish content, corrected for the non-fish flesh nitrogen contributed by the carbohydrate coating, is calculated as follows.

$$\% \text{ Fish} = \frac{(\% \text{ total nitrogen} - \% \text{ non-fish flesh nitrogen}) \times 100}{\text{Nitrogen factor}^*}$$

\*appropriate N (nitrogen) factor for the species of fish ingredient used.

The non-fish flesh nitrogen is calculated as follows:

$$\% \text{ non-fish flesh nitrogen} = \% \text{ carbohydrate} \times 0.02$$

Where the carbohydrate is calculated by difference:

$$\% \text{ carbohydrate} = 100 - (\% \text{ water} + \% \text{ fat} + \% \text{ protein} + \% \text{ ash})$$

References:

Determination of nitrogen: ISO 937:1978

Determination of moisture: ISO 1442:1997

Determination of total fat: ISO 1443:197

Determination of ash: ISO 936:1978

[Average nitrogen factors to be used for fish flesh for specific fish species used as raw material for the product can be found at the following website:

UK Royal Society of Chemistry – Analytical Methods Committee Technical Brief. Electronic link:

<http://www.rsc.org/Membership/Networking/InterestGroups/Analytical/AMC/TechnicalBriefs.asp>

Or

FAO/WHO. Electronic link:]

The uncertainty of each nitrogen factor should be taken into account from the statistical data presented with the published nitrogen factor (e.g., 2 standard errors about the mean).

## **(2) Determination of Fish Content During Production**

The fish content of a fish finger (fish stick) is calculated by using the following equation:

$$\% \text{ Fish Content} = \frac{\text{Weight of in-going fish}}{\text{Weight of final product}} \times 100$$

For most products therefore, the fish ingredient weight is that of the raw ingredient. Any figure placed or declared on a product label would be a typical quantity reflecting the producer's normal manufacturing variations, in accordance with good manufacturing practice.

### **Annex B – Draft Uniform Procedure to Determine Nitrogen Factors for Use With the Chemical Method Listed in the Codex Fish Stick Standard (for further elaboration)**

Nitrogen factors may be determined from unprocessed fish and/or processed fish produced by GHP/GMP depending on the intended use of the nitrogen factor. The natural levels of nitrogen in fresh fish from the sea or farm can have a wide spread (large standard deviation) because of environmental factors. Natural nitrogen variation, and variation due to processing methods, should be taken into account when judging whether further investigation is necessary.

Fish samples are analysed in duplicate for nitrogen, fat, moisture and ash, usually running a reference material with every batch of samples. If possible, more than one laboratory is used and samples are randomised to prevent bias. The raw data of all the samples should be available in order that the mean nitrogen value and a standard error/deviation can be verified.

In most European laboratories, the method now used is the more rapid nitrogen determination of Dumas, whereas previously the method used was Kjeldahl. In some laboratories Kjeldahl is still used. There is a difference between the two methods as the Dumas method measures the non-protein nitrogen as well as the protein nitrogen, and hence gives higher results than the Kjeldahl method<sup>5</sup>. The Dumas is higher by a factor of 1.014.

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<sup>5</sup> M. Thompson, L. Owen, K. Wilkinson, R. Wood and A. Damant, A Comparison of the Kjeldahl and Dumas Methods for the Determination of Protein in Foods, using Data from a Proficiency Testing Scheme, Analyst, 2002, 127, 1666–1668.