

Towards the Harmonisation of Analytical Methods for Food Additives and Food Processing Toxicants

Introduction

The presentation is based on the work done by the MoniQA WG on Food Additives and Processing Toxicants

Specific topics were selected based on the criteria :

- Legislative regulations and limits
- Technological need, benefit and health risk of an additive and severity of a processing toxicant
- Broad use or frequency of occurrence

Legislative regulations

- **Directive 89/107/EEC** defines the substances which may be used as additives and the conditions under which they may be used.
- **Directive 94/36/EC** defines the substances that can be used as colours, specifies the conditions and foodstuffs in which colours can be added and the permissible limits of each colour.
- **Directive 94/35/EC** defines the substances that can be used as sweeteners, specifies the foodstuffs in which sweeteners can be added and the permissible limits.
- **Directive 95/2/EC** specifies the permitted preservatives, antioxidants and other additives, the foodstuffs in which they may be added and the permissible dose.

Legislative regulations

- All food additives must comply with the purity criteria laid down by three other directives (***Directive 95/45/EC*** for colours, ***Directive 95/31/EC*** for sweeteners, ***Directive 2000/63/EC*** for additives other than colours and sweeteners)
- ***Commission Recommendation 2007/331/EC*** concerns the monitoring of acrylamide levels in foodstuffs.
- ***Commission Regulation No 1881/2006*** concerns contaminants including PAHs and specifies maximum levels for certain foodstuffs.

Legislative regulations

Currently the EFSA and the European Commission are dealing with a legislation package consisting of 4 new regulations

- regulation of “common authorization procedure” will conduce to the free movement of food within the community

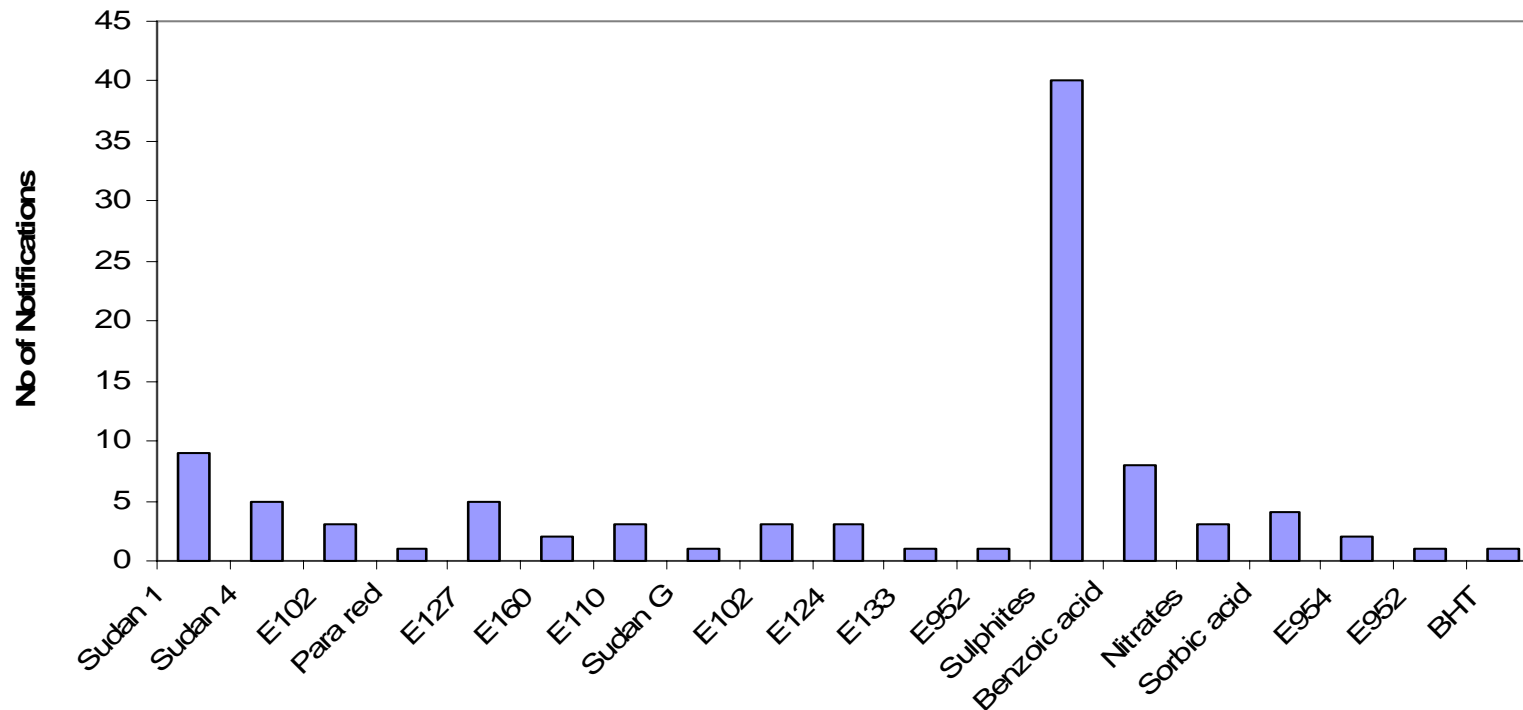
- regulation concerning additives
 - listing the safe to use additives including the technological need of their use and advantages and benefits to consumers
 - introducing new labeling rules regarding azo dyes
 - food additives which might have been changed by nanotechnologies

Legislative regulations

- regulation concerning flavourings
 - establishing stricter conditions on the use of term “natural flavouring”
 - setting new maximum levels for “naturally occurring undesirable substances”
- regulation concerning food enzymes
 - setting rules for evaluation, approval and control of enzymes used in food processes.

RASFF Data

During the first semester of 2008, the Rapid Alert System for Food and Feed (RASFF), received 96 notifications of unauthorised use or high levels of food additives.



Technological need, benefits, and health risks

□ Colours

- restore lost or faded colour of the foodstuff
 - function as a supplement to the natural colouring of a foodstuff so as to avoid batch-to-batch colour variations
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- ❖ *Natural or synthetic*
 - ❖ Synthetic dyes show several advantages (high stability to light, oxygen and pH; colour uniformity; low microbiological contamination; low production cost) over natural ones
 - ❖ The most common synthetic food colourants are azo dyes, which are characterized by the presence of a chromophoric azo-group

Technological need, benefits, and health risks

□ Colours

- Most of the azo dyes are determined as carcinogenic
 - *Sudan (I-IV) dyes, Auremine and Rhodamine are not permitted*
 - ✓ Sudan I is considered to be a genotoxic carcinogen and classified as a category 3 carcinogen by the International Agency for Research on Cancer
 - *Tartrazine (E 102), Sunset yellow (E 110), Carmoisine (E 122), Amaranth (E 123), Black PN (E 151), Ponceau 4R (E 124) and Chocolate brown HT (E 155) are permitted as azo food colors*

Technological need, benefits, and health risks

□ Sweeteners

- *Production of dietetic and low colorie foodstuffs*
- *Prolonging the shelf-life of the product*
- ❖ *Common (nutritive) sweeteners: polyols*
- ❖ *Intense (non nutritive) sweeteners: e.g. salt of aspartame and acesulfame*

Technological need, benefits, and health risks

□ Sweeteners

- The safety of aspartame has been a controversial issue. However the AFC Panel of EFSA evaluated a new long-term study regarding the carcinogenicity of aspartame and concluded that there is no need to further review the safety of aspartame nor to revise the previously established Acceptable Daily Intake (ADI) for aspartame (40 mg/kg body weight).

Technological need, benefits, and health risks

Preservatives

Benzoic acid and sorbic acid

- Antimicrobial activity
- Present low toxicity, however if abused can threaten consumer's health.

Technological need, benefits, and health risks

Preservatives

☐ **Sulphites**

- Antioxidant activity
- Inhibition of enzymic and chemical reactions
- Antimicrobial activity
- Bleaching activity
- High intake
 - stomach irritation, abdominal pains and vomiting
 - Symptoms similar to those of allergic reaction (eg. anaphylactic reaction)

Technological need, benefits, and health risks

Preservatives

☐ **Sulphates**

- Not used as additive.
 - Present in water supplies and not effectively removed by treatment processes due to its solubility and stability in water.
 - Non toxic, however laxative effect has been observed in the case of higher than 500mg/l of sulphate in water or drinks.
- Apart from sulphites, sulphates may also affect the total sulphur intake, which should not exceed the ADI.

Technological need, benefits, and health risks

Preservatives

□ **Nitrites**

- Antimicrobial activity
- Can be converted to carcinogenic nitrosamines in foodstuffs and through the human digestive system
- Has been linked to methaemoglobinemia (decreased ability of blood to carry the vital oxygen around the body)
- *Major contributors to nitrite dietary intake:* cured, dried meat products ~50mg/kg, cured bacon ~175mg/kg

□ **Nitrates**

- Antimicrobial activity (uncertain)
- More stable and less toxic than nitrites

Technological need, benefits, and health risks

□ Antioxidants

- *Delay/inhibit lipid oxidation*
 - *flavor degradation/rancidity*
 - *shelf life limitation*
 - *loss of nutritive value*

Technological need, benefits, and health risks

□ Antioxidants

➤ *Potential health risks*

- BHA has been evaluated by IARC as possible carcinogen
- BHT might have the ability to affect allergic diseases in humans (such as allergic rhinitis and asthma) while long-term exposure to BHT is capable of inducing oxidative and metabolic alterations similar to some pathological disorders
- TBHQ has also been accounted for allergenic properties and carcinogenicity
- Ethoxyquin has been shown to cause several unfavourable side effects in animals fed with diets containing this compound, but also adverse effects have been observed in people exposed to EQ

Food processing toxicants –formation and health risk

□ Trans fatty acids

- from *cis* unsaturated fatty acids during biohydrogenation by rumen microorganisms
 - during the hydrogenation process of vegetable and fish oils
- Major contributors to trans fatty acid intake: stick margarines, shortenings, bakery products, French fries and others processed foods made by using partially hydrogenated oils.
- trans fatty acid intake
- increase of plasma LDL-cholesterol and the decrease of HDL-cholesterol
 - increase risk of cardiovascular disease

Food processing toxicants –formation and health risk

□ Acrylamide

- cooking practices -generated primarily in fried, grilled, and baked products at temperatures above 120 °C.
- highest concentrations occur in starchy foods (e.g.potato chips, french fries, bread, processed cereals).
- possible risk to public health remains unclear.

Food processing toxicants –formation and health risk

□ N-nitrosamines

- reaction of nitrosating agents (nitrite added as preservative in meat, nitrite produced by bacterial reduction of nitrate in beer) with secondary amines.
 - smoking and drying processes (e.g.dried milk and some cheeses)
 - reactions of some disinfectants and from some ion exchange resins (in water)
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- In most cases are strongly carcinogenic,
 - ✓ N-nitrosodimethylamine (dimethylnitrosamine, NDMA) is the most frequently occurring
 - Levels of N-nitrosamines in food are not regulated directly by law

Status of Analytical Methods

□ Colours

- *E 102, E 110, E 124, E 127 and E 133* : AOAC official (column chromatography).
- *Sudan*: GB/T standard, FSA standard (mostly HPLC).
- Need for
 - ✓ European or international standard methods
 - ✓ Rapid, screening methods for raw materials and food products

Status of Analytical Methods

□ Sweeteners

- *acesulfame-K, aspartame, saccharin, cyclamate* : EN standards for single and multi-component methods (HPLC, spectroscopy)
- *cyclamate and saccharin* : AOAC officials (colourimetry, LC, differential pulse polarography)
- Need for
 - ✓ rapid methods, which can be used for screening purposes ?

Status of Analytical Methods

Preservatives

□ **Sorbic acid, Benzoic acid**

- AOAC- officials (spectroscopy, GC)
- national standards (e.g.GB/T-HPLC)
- Need for
 - ✓ improvement of the recovery of some methods

Status of Analytical Methods

Preservatives

□ Sulphites

- EN ISO standards (Monier-Williams, Enzymatic)
- AOAC official (Monier-Williams, Spectroscopy, Differential Pulse polarography, FIA)
- Need for
 - ✓ modification for other food matrixes (e.g. cabbage, dried garlic and onions, etc.)

Status of Analytical Methods

Preservatives

□ Nitrites/Nitrates

- EN ISO standards (Spectrometry, FIA)
- AOAC officials (colorimetry, modified Jones reduction method, xylenol method)
- national MSZ standards (Spectroscopy, HPLC)
- test kits providing semi-quantitative estimates of nitrate-N content.
 - Need for
 - ✓ improvement and validation of rapid methods

Status of Analytical Methods

□ Antioxidants

- *BHA, BHT, PG, THBP, TBHQ, NDGA*: AOCS official method (HPLC)
- *BHA, BHT*: GB/T standard method (GC), IUPAC official method (steam distillation and colourimetry)
- Need for
 - ✓ development of validated methods for fat containing food -standardization of the recovery (extraction/pretreatment)

Status of Analytical Methods

□ Trans fatty acids

- EN standard method (direct GC)
 - AOAC official methods (direct -with prior fractionation- GC, IR spectrometry, and capillary GC-IR spectrophotometry)
 - AOCS official methods (direct GC, silver-ion exchange HPLC, IR spectrometry, attenuated total reflection IR spectroscopy (ATR-FTIR))
- Need for
- ✓ improvement of existing methods (overlapping between *trans* and *cis* fatty acids , co-elution of *trans* isomers with major saturated and unsaturated fatty acids which can lead to underestimation of the *trans* fatty acids content)
 - ✓ Development of simple, rapid and cheap method for identification and quantification of all kind (geometrical and positional) of *trans* fatty acids in one run of analysis.

Status of Analytical Methods

□ Acrylamide

- there are no official methods
- most widely used methods are GC-MS and LC-MS/MS.
- need for
 - ✓ International or European official methods

□ N-nitrosamines

- AOAC official (GC-Thermal Energy Analyser)- used extensively to determine volatile N-nitrosamines
- GB/T standard methods (GC -TEA and GC-MS)
- need for
 - ✓ *Development, testing and validation of*
 - *GC-MS methods*
 - *novel rapid methods*
 - *LC-MS methods (determination of non-volatile nitrosamines)*

Towards Harmonization

Horizontal Issues

□ Measurement Uncertainty-Recovery

- Main sources of measurement uncertainty for food additive analysis
 - sample effects / interference of other components of the matrix
 - instrument effects / instrument response may be affected by other components of the matrix
 - measurement conditions
- Recovery
 - pre-treatment of a complex sample may lead to partial loss of the analyte

□ Sampling

- no legislative requirements regarding sampling of food additives
- sampling procedure of acrylamide is outlined by ***Commission Recommendation 2007/331/EC***

Towards Harmonization

Challenges in Food additives and processing toxicants analysis

- Modification of existing official or development of new method that can be applied to additional food material
- Development of methods where a lack of official method is encountered
- Development of
 - rapid, screening and/or high throughput methods
 - methods for multi-component analysis
 - methods using state of the art equipment which can provide higher accuracy and selectivity
- Development of harmonization guidelines to validate the above methods

Towards Harmonization

Future activities for the WG

- Survey of the method performance criteria of the standard and official methods to define possible missing issues
- Identification of promising methods where a lack of official method is encountered
- Identification of promising rapid, screening, and multi-component analysis methods
- Development of harmonized guidelines to validate the above methods