

codex alimentarius commission



FOOD AND AGRICULTURE
ORGANIZATION
OF THE UNITED NATIONS

WORLD
HEALTH
ORGANIZATION



JOINT OFFICE: Viale delle Terme di Caracalla 00153 ROME Tel: 39 06 57051 www.codexalimentarius.net Email: codex@fao.org Facsimile: 39 06 5705 4593

Agenda Item 2

CX/GP 09/25/2-Add.1

JOINT FAO/WHO FOOD STANDARDS PROGRAMME CODEX COMMITTEE ON GENERAL PRINCIPLES

Twenty-fifth Session

Paris, France, 30 March – 3 April 2009

MATTERS ARISING FROM THE CODEX ALIMENTARIUS COMMISSION AND OTHER CODEX COMMITTEES

Addendum 1

Committee on Methods of Analysis and Sampling

The Committee on Methods of Analysis and Sampling completed its review of the *Working Instructions for the Implementation of the Criteria Approach in Codex* in the Procedural Manual in order to provide guidelines for the Identification of Relevant Analytical Methods. The Appendix provides some amendments to the current section (adopted by the 31st Session of the Commission) and a new section **GUIDELINES FOR ESTABLISHING NUMERIC VALUES FOR METHOD CRITERIA AND/OR ASSESSING METHODS FOR COMPLIANCE THEREOF**.

PROPOSED AMENDMENTS TO THE PROCEDURAL MANUAL**WORKING INSTRUCTIONS FOR THE IMPLEMENTATION OF
THE CRITERIA APPROACH IN CODEX**

(To add the following note above Table 1)

Note: These criteria are applicable to fully validated methods except for methods such as PCR and ELISA, which requires other set of criteria.

In Table 1

Precision should be described as

$RSD_R \leq 2$. $PRSD_R$

Add the following text at the end of the section

GUIDELINES FOR ESTABLISHING NUMERIC VALUES FOR METHOD CRITERIA AND/OR ASSESSING METHODS FOR COMPLIANCE THEREOF.**1. RECOMMENDATIONS FOR ESTABLISHING NUMERIC VALUES FOR METHOD CRITERIA**

Only the provision for the commodity along with its ML (maximum level, minimum level, normative level or concentration range) is needed when establishing numeric values for method criteria.

Note: These criteria are applicable to fully validated methods except for methods such as PCR and ELISA, which requires other set of criteria.

1.1 The applicability

The method has to be applicable to the particular analyte(s)/provision(s) in the specified matrix/ commodity or food category. For horizontal methods the relevant food categories should have been tested. Furthermore, it should have been shown that the method is applicable for concentrations levels around the specified ML, i.e. the ML should be within the validated range.

- For $ML \geq 10^{-7}$, the minimum applicable range should be: $ML \pm 3s_R$
- For $ML < 10^{-7}$, the minimum applicable range should be: $ML \pm 2s_R$

The minimum applicable concentration range should correspond to an interval containing a large fraction of the expected variation (due to measurement uncertainty) in the results around the specified limit (ML). For collaboratively validated methods the expected variation would be the reproducibility standard deviation (s_R) multiplied with a coverage factor. A coverage factor of 2 corresponds to a confidence level of approx. 95%, and a coverage factor of 3 corresponds to a confidence level about 99%. As 99% is often used as an action level in control charts, a coverage factor of 3 is recommended for concentration ratios at or above 10^{-7} , (≥ 0.1 mg/kg). For concentrations lower than 0.1 mg/kg, a coverage factor of 2 is recommended, as a coverage factor of 3 would make it hard to find applicable methods for certain analytes/provisions due to the low level.

Calculation of the minimum applicable range for specified MLs:

The minimum applicable range can be estimated based on the Horwitz/Thompson equation for reproducibility standard deviation, s_R .

1.1.1: For concentration ratios $\geq 10^{-7}$ (≥ 0.1 mg/kg) the Horwitz' equation is applied:

$$PRSD_R (\%) = 100 \cdot s_R/c = 2C^{-0.1505}$$

where

$PRSD_R$ is the "predicted" relative standard deviation,

s_R is the predicted standard deviation
 c is the concentration of interest, which here is the ML and
 C is the concentration ratio, i.e. the concentration ratio of ML (C_{ML})

By rearranging the equation with respect of s_R , the following equation is obtained:

$$s_R = \frac{c \cdot 2C^{-0.1505}}{100} = \frac{ML \cdot 2 \cdot C_{ML}^{-0.1505}}{100}$$

Example 1: ML = 0.1 mg/kg, $C_{ML} = 10^{-7}$:

$$0.1 \pm 3 \cdot s_R = 0.1 \pm 3 \cdot \frac{0.1 \cdot 2 \cdot (0.0000001)^{-0.1505}}{100} = 0.1 \pm 0.07 \text{ mg/kg}$$

The minimum applicable range for a ML of 0.1 mg/kg is then 0.03 to 0.17 mg/kg

Example 2: For a ML of 1 mg/kg (i.e. 10^{-6}):

$$1.0 \pm 3 \cdot s_R = 1.0 \pm 3 \cdot \frac{1.0 \cdot 2 \cdot (0.000001)^{-0.1505}}{100} = 1.0 \pm 0.48 \text{ mg/kg}$$

The minimum applicable range for ML of 1 mg/kg is then 0.5 to 1.5 mg/kg

1.1.2: For concentration ratios $< 10^{-7}$, the Thompson theory is applied, i.e. $PRSD_R = 22\%$ and hence $s_R = 0.22 \cdot ML$

Example 3: ML = 0.01 mg/kg (i.e. 10^{-8}):

$$0.01 \pm 2 \cdot s_R = 0.01 \pm 2 \cdot (0.22 \cdot ML) = 0.01 \pm 0.44 \cdot 0.01 = 0.01 \pm 0.0044 \text{ mg/kg}$$

The minimum applicable range for a ML of 0.01 mg/kg is then 0.006 to 0.014 mg/kg.

In table 1, a number of minimum applicable concentration ranges for specified MLs are given.

Table 1: Recommended criteria for minimum application range for specified MLs

ML (mg/kg)	0.01	0.02	0.05	0.1	1	10	100
Lower level:	0.006	0.011	0.028	0.03	0.52	6.6	76
Upper level: *	0.014	0.029	0.072	0.17	1.48	13.3	124

* Upper level will seldom be the limiting factor like the lower level.

1.2 Limit of Detection (LOD) and limit of Quantification (LOQ)

As an alternative to establishing minimum applicable range, the criteria could be numeric values for LOD and LOQ.

The numeric value for the limit of detection (LOD), should be:

- no more than 1/10 of the specified ML for levels at or above 0.1 mg/kg, and
- no more than 1/5 of the specified ML for levels below 0.1 mg/kg.

The numeric value for the limit of quantification (LOQ) should be:

- no more than 1/5 of the specified ML for levels at or above 0.1 mg/kg, and
- no more than 2/5 of the specified ML for levels below 0.1 mg/kg.

1.3 The method precision, derived from collaborative method performance studies

The precision should be expressed as the obtained relative reproducibility standard deviation (RSD_R) obtained from collaborative method performance studies, which is compared to the predicted relative reproducibility standard deviation ($PRSD_R$)

According to Horwitz, the ratio between the found and the predicted value should be ≤ 2 (known as the HorRat value), this is also applicable for Thompson equation of $PRSD_R = 22\%$:

$$\frac{RSD_R}{PRSD_R} \leq 2 \Leftrightarrow RSD_R \leq 2 \cdot PRSD_R$$

The numeric values for the precision given in table 2 are also based on the Horwitz/Thompson equation. For some analyses, using advanced techniques, a better precision can be obtained.

Table 2. Precision requirement at different concentrations based on the Horwitz/Thompson equation.

	Thompson	Horwitz equation ($2C^{-0.1505}$)							
Concentration ratio (C)	$< 10^{-7}$	10^{-7}	10^{-6}	10^{-5}	10^{-4}	10^{-3}	10^{-2}	10^{-1}	1
Concentration unit	< 0.1 mg/kg	0.1 mg/kg	1 mg/kg	10 mg/kg	0.1 g/kg	1 g/kg	10 g/kg	100 g/kg	1000 g/kg
$PRSD_R$ (%)	22	22	16	11	8	6	4	3	2
$RSD_R \leq 2 \cdot PRSD_R$ (%)	≤ 44	≤ 44	≤ 32	≤ 22	≤ 16	≤ 12	≤ 8	≤ 6	≤ 4

$PRSD_R$ = predicted value for relative standard deviation of reproducibility.

RSD_R = found value for the relative standard deviation of reproducibility in a collaborative study.

1.4 Recovery

Evaluation and estimation of recovery is included in the method validation. Whether or not recovery is of relevance depends on the method procedure.

1.5 Trueness

For the evaluation of trueness preferably appropriate certified reference materials (CRMs) should be analysed and demonstrated to give the certified value (allowing for measurement uncertainty) is achieved.

1.6 Examples on how to establish criteria for a provision

In order to illustrate how to set criteria for a provision the following example is used:

According to Codex Standard 1993-1995, Rev 2-2006, General Standard for contaminants and toxins in food, the ML for lead in fruit juices is 0.05 mg/kg. According to the recommendations for obtaining numeric values for the characteristics based on the ML, the criteria would be those in table 3:

Table 3. Recommendation for numeric criteria values for lead in fruit juice

Applicability:	Lead
Analyte:	Juice
Matrix/provision:	0.05 mg/kg
ML:	
Lower level of min. application range:	≤ 0.03 mg/kg (= ML - $2s_{SR}$ = 0.05 mg/kg - $0.44 \cdot 0.05$ mg/kg). See Table 1
LOD:	≤ 0.01 mg/kg (= ML $\cdot 1/5$ = 0.05 mg/kg $\cdot 1/5$)
LOQ:	≤ 0.02 mg/kg (= ML $\cdot 2/5$ = 0.05 mg/kg $\cdot 2/5$)
Precision:	For concentration at 0.05 mg/kg, the $RSD_R \leq 44\%$, See Table 2
Recovery:	The method procedure does not include an extraction step and hence recovery is of no relevance.
Trueness:	Use of CRM.

2. METHOD CRITERIA AT DIFFERENT MLs (MAXIMUM LEVEL, MINIMUM LEVEL, NORMATIVE LEVEL OR CONCENTRATION RANGE)

In table 4 examples on method criteria are given for certain MLs.

Table 4: Method criteria for MLs at increasing orders of magnitude.

ML unit	0.001 mg/kg	0.01 mg/kg	0.1 mg/kg	1 mg/kg	10 mg/kg	100 mg/kg	1 g/kg	10 g/kg
Concentration ratio of ML (C_{ML})	10^{-9}	10^{-8}	10^{-7}	10^{-6}	10^{-5}	10^{-4}	10^{-3}	10^{-2}
Minimum applicable Range	From 0.0006 to 0.0014 (mg/kg)	From 0.006 to 0.014 (mg/kg)	From 0.03 to 0.17 (mg/kg)	From 0.52 to 1.48 (mg/kg)	From 6.6 to 13.3 (mg/kg)	From 76 to 124 (mg/kg)	From 0.83 to 1.2 (g/kg)	From 8.8 to 11 (g/kg)
LOD (\leq mg/kg)	0.0002	0.002	0.01	0.1	1	10	100	1000
LOQ (\leq mg/kg)	0.0004	0.004	0.02	0.2	2	20	200	2000
RSD_R (\leq %)	44	44	44	32	22	16	12	8
Recovery (%) *	40 - 120	60 - 115	80 - 110	80-110	80 - 110	90 - 107	95 - 105	97 - 103

* Other guidelines are available for expected recovery ranges in specific areas of analysis. In cases where recoveries have been shown to be a function of the matrix other specified requirements may be applied.

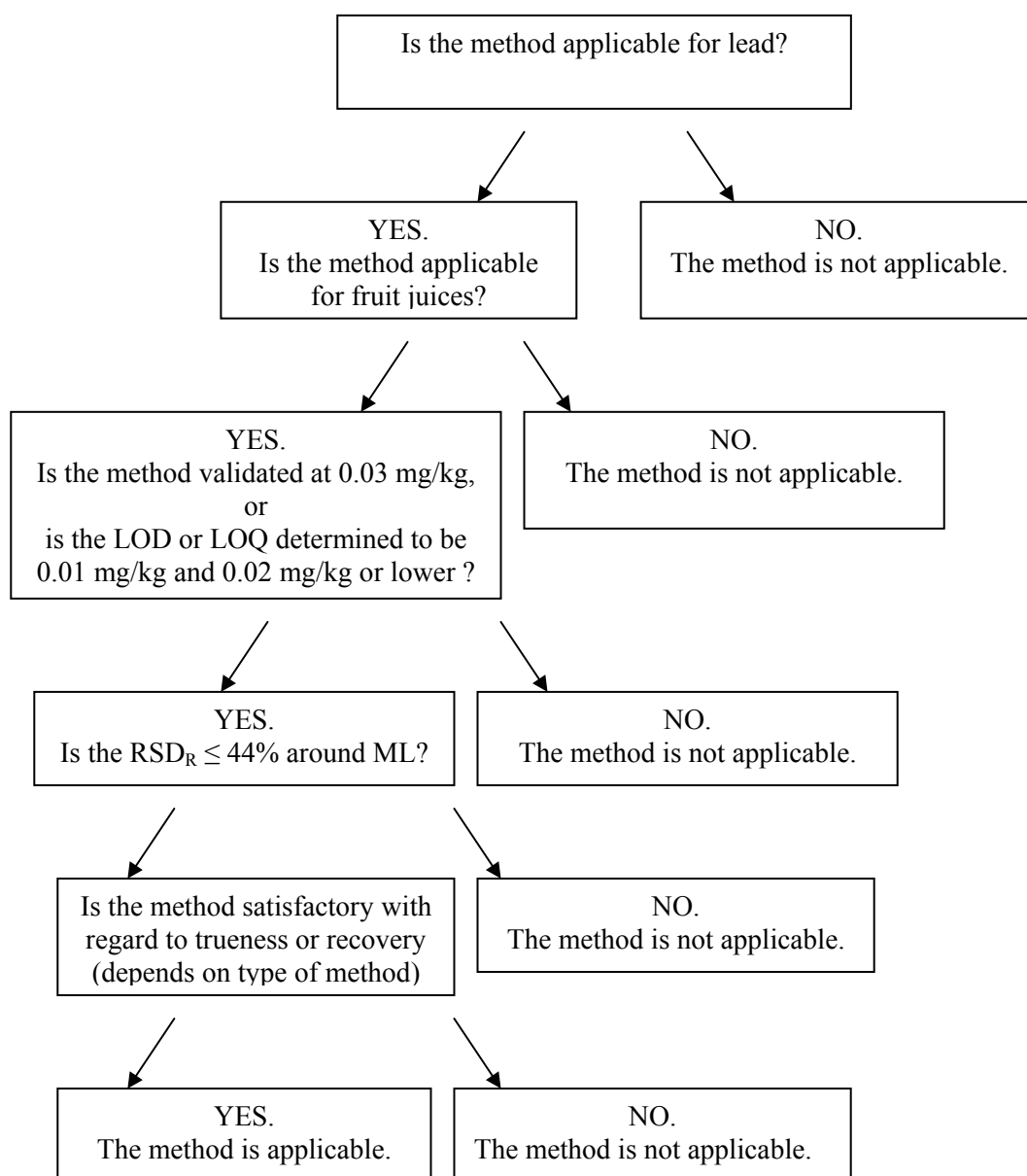
2.1 How to elucidate a method's compliance with the criteria.

To review a method for possible compliance with the established criteria, the method performance characteristics have to be assessed. The result of a method performance study is available in the method and/or published in an international journal.

2.1.1 Example on assessing methods for compliance

Continuing the example above on lead in fruit juice, having ML of 0.05 mg/kg, the methods considered should be able to quantify lead in fruit juice as low as 0.03 mg/kg, with a precision, $PRSD_R$ of 22%, the RSD_R obtained from the method performance study should then not be higher than 44% (corresponding to a 95% confidence interval).

When assessing a method for compliance, the following steps should be considered:



In order to find appropriate methods for this purpose, information are collected on methods for determination of lead. (As this is an example in the Procedural Manual, the methods' identification is omitted):

Table 5: Collaboratively validated methods for analysis of lead

Method No	Applicability	Principle	Assessed level (mg/kg)	LOD (mg/kg)	RSD _R (%)	Applicable Yes/No and why
1	All foods	Flame AAS	2.2 – 29		4.9-36	NO Flame AAS will not be able to detect at 0.05 mg/kg
2	All Foods (Chicken, apple)	Anodic stripping voltammetry	0.03-2.8	0.03	17-106	NO The RSD _R is 106% (not <44%) at 0.03 mg/kg
3	Sugars	GF-AAS	0.03-0.50		12-30	YES Even if the applicability does not say Juice (or all foods) it should be considered applicable as fruit juice contains a lot of sugar. The precision is satisfactory.
4	Fats and Oils	GF-AAS	0.018-0.090		5.9-30	NO The method describes sample prep. for fats and oils only.
5	Natural mineral water	AAS	0.0197-0.977	< 0.01	2.8-4.2	NO The method describes sample prep. for water only.
6	All foods	GF-AAS after dry ashing	0.045-0.25	< 0.01	26-40	NO The lowest validated level is not low enough, however as the technique is GF-AAS, it should be applicable for 0.03 mg/kg.
7	All foods except oils, fats and extremely fatty products.	AAS after microwave oven digestion under pressure.	0.005-1.62	0.014	26-44	YES Validation level and RSD _R are ok
8	All foods	ICP-MS after pressure digestion	0.013-2.45	< 0.01	8-47	YES Validation level and RSD _R are ok for levels of 0.03 mg/kg and above.

AAS = Atomic Absorption Spectrometry

GF-AAS = Graphite Furnace Atomic Absorption Spectrometry

ICP-MS = Inductive Coupled Plasma - Mass Spectrometry

Conclusion: Methods No. 3, 7 and 8 are found applicable for the determination of lead in fruit juices for the given ML of 0.05 mg/kg. Assessing methods for compliance requires knowledge about the methods; sample preparation, procedures and instrumentation. Thus the methods cannot be “judged” by numeric values for the criteria alone.