Fresenius Z Anal Chem (1988) 332:533–538

Fresenius Zeitschrift für
Analytische
Chemie

BCR reference materials for food and agricultural analysis: an overview

J. J. Belliardo and P. J. Wagstaffe

Community Bureau of Reference (BCR), Commision of the European Communities, 200 Rue de la Loi, B-1049 Bruxelles, Belgium

BCR-Referenzmaterialien für die Lebensmittel- und landwirtschaftliche Analyse – Ein Überblick

Summary. The BCR is a research programme of the European Communities having the broad aim of improving accuracy and measurement harmony in areas of concern at Community level. Much of the work is directly relevant to the removal of trading and other obstacles as is necessary for the achievement of a single European market by 1992.

The presentation provides an overview of the sector of the programme concerned with Reference Materials (RMs) for food and agricultural analyses.

Summarises are given of activities for inorganic, trace organic and nutritional analyses, for calibration of rapid methods (NMR, NIR, XRF and Immuno-assay techniques) and technological measurements (industrial quality of cereals, food colour, water-sorption and detection of fraudulent addition of sugar during wine making by NMR). An alphabetic listing of analytes and properties studied during the programme is given.

Introduction

The role of RMs in analytical chemistry is now well established specially for calibration and verification of the accuracy of methods, and there should be no need to convince participants to this symposium of their usefulness.

Unfortunately however, judging by the modest rate of sale and the large number of potential user laboratories, a very serious educational task remains.

The major problem facing RM producers results from the virtually unlimited variety of analyses obtained in combining analytes, concentrations and matrices, that is to say, the problem of fulfilling the priority needs.

Nowhere is this more true than in the field of food and agriculture where no less than 4,000 methods of analysis have been counted.

The approach adopted by the BCR for the selection of new projects has been essentially the pragmatic one of concentrating on topics where there are known to be problems in practice. The mere existence of a method, an analyte or of a regulation is not automatic justification for a reference material or an intercomparison project.

Offprint requests to: J. J. Belliardo

The objective of this presentation is to give an overview of BCR activities in the area of food and agriculture although it is emphasized that it forms only a part of a whole measurement programme, which also includes environmental, biomedical, industrial problems and physical properties.

No more than an outline of the various activities is now given in the short time available, however, a number of projects will be presented in detail by other participants during the course of this symposium.

Some facts and figures

As many of you know, BCR is a Community research programme with the general objective of improving accuracy and thereby measurement harmony.

The programme is essentially one of collaboration and the role of the BCR is to bring together the experience of European laboratories.

Table 1 presents some facts and figures concerning the last programme (1983–1987).

Of particular importance is the number of intercomparison excercises. These are undertaken with several objectives in view:

- to establish the state of the art,
- to identify and eliminate weaknesses in methods (reduction of sources of error),
- to pool European experience.

Intercomparisons are run in groups of typically 12–15 experienced laboratories and are repeated as necessary until the major source of error in the various methods are reduced to acceptable levels.

Where there is sufficient economic justification, the projects may lead to the preparation of reference materials which allow any user to compare his results with the reference values established by the BCR groups thus linking laboratories to a common scale of accuracy.

The major areas of the sub-programme are reviewed in the following.

Table 1
BCR RM Activities for food and agriculture (1983–1987) – Some facts and figures

Projects	27
CRMs issued	17
Certified properties	75
No. of measurements	6000
Indicative values	60
Intercomparisons	36
RMs in preparation	30
Participating laboratories	300

Table 2. Reference materials for elemental analysis in food and feed with certified and indicative values

CRM	189	191	063	150	151	184	185	186	273	274
Type	Wholemeal flour	Brown bread	Skim milk powder, natural	Skim milk powder, spiked	Skim milk powder, spiked	Bovine muscle	Bovine liver	Pig kidney	Single cell protein	Single cell protein
As (ng/g)	(18)	(23)				(26)	24	63		132
Ca (mg/g)	(0.52)	(0.41)	12.6			(0.150)	(0.131)	(0.295)	11.97	
Cd (ng/g)	71.3	28.4	2.9	21.8	101	13	198	2710		30
Cl (mg/g)	(0.7)	(16.5)	10.7			(2.0)	(2.9)	(9.4)		
Co (ng/g)										3
Cr (ng/g)	(57–73)	(68–360)				(76–153)	(47–124)	(58–142)		
Cu (µg/g)	6.4	2.6	0.545	2.23	5.23	2.36	189	31.9		13.1
F (µg/g)										(17.6)
Fe (µg/g)	68.3	40.7	2.06	11.8	50.1	79	214	299	156	
Hg (ng/g)	(1)	(2)	1.0	9.4	101	2.6	44	1970		
I (ng/g)				1290	5350	(40)	(105)	(145)		(21)
K (mg/g)	(6.3)	(3.1)	17.8			(16.6)	(11.2)	(12.6)	2.22	
Mg (mg/g)	(1.9)	(0.5)	1.12			(1.020)	(0.634)	(0.829)	(2.72)	
Mn (µg/g)	63.3	20.3	(226)	(236)	(223)	0.334	9.3	8.5		51.9
N (mg/g)			58.8						121.6	
N (mg/g)[a]			58.3						(121)	
Na (mg/g)	(0.04)	(10)	4.57			(2.0)	(2.1)	(7.1)	(0.044)	
Ni (µg/g)	(0.38)	(0.44)	(11.2)	(61.5)	(56)	(0.27)	(1.4)	(0.42)		0.3
P (mg/g)			10.4			(8.3)	(11.7)	(12.2)	26.8	
Pb (µg/g)	0.379	0.187	0.104	1.0	2.002	0.239	0.501	0.306		44
S (mg/g)									(10.2)	
Se (µg/g)	0.132		(88)	(127)	(125)	0.183	0.446	10.3		1.03
Zn (µg/g)	56.5	19.5	(42)	(49)	(50)	166	142	128		427

[a] Kjeldahl
Values in brackets are not certified

Elemental analysis

Historically, most attention has been given to the preparation of reference materials for trace and minor elements analysis.

Although there are now some 50 RMs available from various sources for elemental analyses of food, there remain some notable gaps with respect to certain matrices and elements.

Table 2 summarises the CRMs available currently from the BCR and gives the indicative and certified contents. It is to be noted that the indicative values are generally based on the results of 3 or more laboratories but no uncertainty can be given. Details of individual results are given in the certification report which is supplied with the reference materials [1–5].

CRMs with very low contents of specific elements provide an invaluable means of exploring some of the most vexing analytical problems, namely:

- limits of detection and determination,
- analytical background (blanks) including contamination from laboratory atmospheres and reagents.

Table 3 presents several biological CRMs (Nos. 63, 184, 185, 191 and 274) which are suitable for such studies for the elements As, Cd, Hg, Pb and Se.

Table 3. Low level reference materials for investigation of laboratory backgrounds and limits of determination in trace element analysis

Study of { Blank values / Limit of detection/determination / Laboratory contamination }

CRM	Contents ng/g				
	As	Cd	Hg	Pb	Se
63 Milk powder	–	2.9	1.0	104	–
184 Bovine muscle	(26)	13	2.6	*	183
185 Bovine liver	24	*	*	*	*
191 Wholemeal bread	(23)	28.4	(2)	*	(25)
274 Single cell protein	*	30	–	44	*

* Also certified; – not determined

Future developments

Future developments will be orientated towards:

- completion of the range of food matrices e.g. vegetables, fish and beverages,
- ensuring that European needs for mixed diet RMs are fulfilled,
- elements or inorganic analyses of increasing interest such as Al, Sn and nitrate and, speciation particularly where bio-availability is of importance in human and animal nutrition.

Trace organic analysis

General

Trace organic analysis presents almost all the problems which can be encountered in accurate quantitation. The

Table 4. Idealised requirements for trace organic analysis

Steps	Requirements (ideal)
Calibration	– High purity, stable calibrants – Linear in range of interest – Valid in the presence of co-extractives
Blanks	– High signal-to-background ratio
Extraction	– Efficient (80% – 100%) – Repeatable
Clean-up, concentration	– No losses of analytes
Final determination	– Specific, sensitive
General	– Absence of contamination

Table 5. BCR Activities for organic contaminants in food and agricultural analysis

Contaminant	BCR activity
Aflatoxin M_1 in milk-powder	4 CRMs available (No. 282, 283, 284 and 285)
Aflatoxin B_1 in animal feed	5 CRMs under preparation
Deoxynivalenol in wheat/maize	4 CRMs under preparation
PCBs in fish oil	2 CRMs available (No. 349 and 350)
Organo-chlorine pesticides in milk-powder	2 CRMs available 1988
PAHs in edible oil/kale	2 RMs under preparation
Genotoxic compounds (IQ, MeIQ, MeIQx) in processed food	Preliminary analytical studies
Shell fish toxins	Toxin RMs and preliminary studies planned
Illegal hormones in animal tissues and urine	Contaminated and blank material under preparation
Veterinary drugs in milk, eggs and meat	Next BCR programme

critical steps and minimum requirements may be summarised as in Table 4.

In practice, the analyst can only approximate to these ideals. Although recovery experiments (spiking) may show that the added compound is fully recovered, there is no certainty that the endogenous compounds are present in the same readily accessible form (e.g. bound to protein or dissolved in the lipid phase).

Biological CRMs again provide a convenient means of allowing laboratories to investigate the reliability of their methods. Table 5 summarises subjects where BCR has undertaken projects for organic contaminants in food.

Practical

The practical problems of preparing homogeneous, stable and representative biological materials are considerable but usually they can be overcome e.g. by lyophilisation, grinding and seiving to produce uniform, stable powders.

The problem of assigning a reliable reference value is generally much more difficult.

Table 6. Edible oil and fat RMs

	162 Soya-maize oil	163 Beef-pig fat	164[a] Milk-fat
FA profile	*	*	*
Butyric acid	–	–	*
Triglycerides	–	–	(*)
Sterol profile	(*)	–	(*)
Cholesterol[b]	(*)	(*)	(*)
Unsap. matter	(*)	–	–
2-position FA	–	(*)	–
PUFAs	(*)	–	–
Tocopherols	(*)	–	–
I_2 Value	(*)	–	–

[a] To be issued in 1988
[b] Ongoing study
(*) Indicative values
* Certified values

In general, projects involve a series of intercomparisons of methods in the more experienced laboratories. The intercomparisons often lead to considerable improvement in the accuracy of the methods as illustrated by mycotoxin projects [6–8].

Future developments

Amongst the many problems in trace-organic analysis which must be tackled in the next BCR programme, the areas of residues of veterinary drugs, prohibited growth promoting compounds and agricultural chemicals will demand much attention in view of the Community legislation and the obligation on Member States to carry out routine monitoring.

Nutritional analysis

Many nutritional properties are routinely measured and are widely considered to present no difficulties. However, judging by the results of interlaboratory trials, there is no such animal as a property which cannot be badly measured by someone. This is clearly demonstrated in the results of the determination of common elements in orange juice as undertaken by 20 laboratories in the context of a quality assurance programme for the fruit juice industry [9].

Reference materials are thus just as important for the routine and so-called easy measurements as for the more demanding analyses where the analysts exercise more care.

Edible oils and fats analysis

Three RMs have been prepared, covering the primary needs for vegetable oil (RM 162), animal fat (RM 163) and milk-fat (RM 164). The properties for which certified and indicative values are given are summarised in Table 6 [10, 11].

Major nutritional properties and elements

This project responds specifically to the needs for quality assurance of relatively simple properties, the objective being to provide a series of low cost materials covering the basic needs of most nutritional laboratories.

Five RMs have been prepared and will undergo certification during 1988. The materials and properties of interest are presented in Table 7.

Future trends

Increasing attention will be given to problems of vitamin analysis, and amino acids the latter being of growing importance in animal nutrition.

Rapid methods of analysis

Rapid methods based on NMR, NIR, XRF and immunoassay find increasing application in the factory situation where the high sample through-put is incompatible with the relatively slow, traditional analytical methods.

In cases where the results find direct application for CAP levies or subventions, verification that toxins are absent at prohibited levels or because of contractual obligations, e.g. oil content and glucosinolates [12] in rapeseed, aflatoxin B_1 in animal feed [8], it is essential that the results given by the rapid methods are equivalent to those obtained by the 'official' methods, however, the latter are defined.

Several BCR projects address these needs. The principle is presented schematically in Fig. 1 and examples are given in Table 8.

Other topics

Not all topics fall neatly into the above sub-divisions.

Although of primary importance for assessing the quality of products in food industry and intra-community trade, the following measurements are mentioned only briefly to complete this review:

- Rheological and technological properties of cereals, including farinograph, alveograph and extensograph determinations following international norms (ICC, ISO), used to assess the trading quality of cereals. Because the results are instrument-dependent, there is a need for an appropriate intercalibration mechanism consisting of a reference laboratory network for improving the comparability of results.
- H/D isotopic ratio at natural abundance levels by quantitative NMR: Detection of fraudulent addition of sugar during wine making has been made possible by Nuclear Magnetic Resonance (NMR) spectrometry. A set of reference ethanols of different natural origin is currently in preparation for calibration and alignment of NMR spectrometers. The method will be described in detail later in the Symposium [13].
- Colour measurement: A project is underway to produce red ceramic tiles for the calibration of colorimeters used for determination of tomato paste colour.
- Water sorption isotherms reference material: Accurate definition of water sorption isotherms is of considerable importance to processors of food and agricultural products. The water activity subgroup of COST 90 (Physical Properties of food) [14] has developed a method for determining sorption isotherms and showed that microcrystalline cellulose (MCC) could serve as a stable and homogeneous reference material. BCR has undertaken the preparation of a batch of MCC which will be issued as a CRM by the end of 1988.

Table 7. RMs for Major nutritional properties

	Total N protein	Total carbohydrate	Total fat	Individual sugars	Dietary fibre
Full cream milk powder	*	*	*	*	
Wholemeal flour	*	*	*	*	*
White flour	*	*	*		*
Haricot vert	*	*	*		*
Pork muscle	*	*	*		

* ASH and major elements – Ca, K, Na, P...

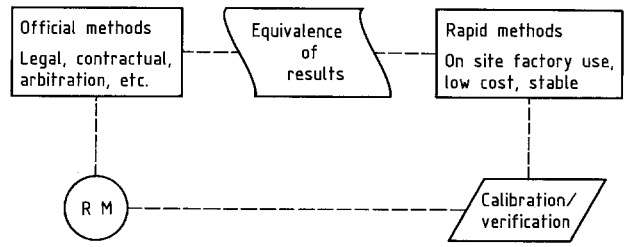

Fig. 1. Calibration of rapid methods via certified reference materials

Table 8. Calibration of rapid methods: examples of RM projects under development

RMs	"Official method"	Rapid method
Total glucosinolate (rapeseed)	GLC, HPLC	Enzymatic XRF, NIR
Oil content of seeds (rapeseed)	Solvent extraction	NMR
Aflatoxin B_1 Animal feed Peanuts for human consumption	HPLC, TLC	Immunoassay

Conclusion

The measurement problems covered in the part of the BCR programme on food and agriculture during the period 1983–1987, are given in Table 9.

Further details on current work and on the next (88–92) BCR programme may be obtained by contacting the BCR in Brussels who would also welcome proposals for collaborative projects.

Acknowledgements. The authors warmly recognise the support and collaboration of the many European Scientists who have made this work possible and to whom the authors dedicate this paper.

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Table 9. Summary of current BCR RM activities for food and agricultural measurements (certified and indicative values)[a]

Property	Activity	RMs
Aflatoxin B_1	In animal feed + peanut butter RMs	Available 1989
Aflatoxin M_1	In 4 milk powder RMs	Available*
Amino acids	In fruit juice, intercomparison	Under development
Arsenic	In meats, cereals and single cell protein RMs	Available*
Butyric acid	In milk fat RM	Available 1988
Cadmium	In milk, meat, cereal and single cell protein RMs	Available*
Calcium	In milk, meat, cereals and single cell protein	Available*
Carbohydrate	(Total) in RM for nutritional properties	Under development
Cholesterol	See 'sterols'	Available*
Chromium	In meat and cereal RMs	Available*
Chloride	In milk, meat and cereal RMs	Available*
Copper	In milk, meat, cereal and single cell protein RMs	Under development
Colour	Tiles for tomato paste (colorimeter calibration)	Available 1989
Deoxynivalenol	In cereals, intercomparisons and RMs under development	Under development
Dietary fibre	In cereals, legumes, intercomparisons and RMs	Under development
Fat (total)	In RMs for nutritional properties	Available*
Fatty acid profile	In vegetable oil and animal fat RMs in milk fat RM	Available 1989
Fatty acid	At 2 position in vegetable oil	Available*
Glucosinolates	Total and individual in rapeseed intercomparisons + RMs	Available 1988
Genotoxic compounds (IQ, MeIQ, MeIQx)	In processed food (intercomparisons)	Under development
Hormones	(Illegal growth promotors) in urine and muscle, intercomparisons + RMs	Available 1989
Iodide	In milk	Available*
Iodine value	In vegetable oil	Available*
Iron	In milk, meat and cereals	Available*
Lead	In milk, meat, cereals	Available*
Magnesium	In milk, meat, cereals	Available*
Nickel	In meats	Available*
Nitrogen	(Total) in milk and single cell protein	Available*
Nitrogen	(Kjeldahl) in milk and single cell protein	Available*
Oil content	Off rapeseed, intercomparisons + RMs	Under development
Polychlorinated biphenyls (PCBs)	In fish oils	Available*
Polycyclic aromatic hydrocarbons (PAH)	In food oil and vegetables, intercomparison + RMs	Under development
Potassium	In milk, meat, cereal and single cell protein RMs	Available*
Protein	(Total) in RMs for nutritional properties	Under development
Selenium	In meat, cereal and single cell protein	Available*
Sodium	In milk, meat, cereal and single cell protein	Available*
Shell fish toxins	In mussels, intercomparisons + RMs	Under development
Sterol (profile)	In vegetable oil	Available*
Sterol (individual)	In vegetable oil, animal fat and butter, intercomparisons + RMs	Under development
Sugars	In fruit juice and RMs for nutritional properties, intercomparison and RMs	Under development
Technological properties	In common wheat flour for bread-making	Under development
Tocopherols	In vegetable oil	Available*
Triglyceride	In milk fat	Available 1988
Unsaponifiable matter	In vegetable oil	Available*
Water sorption isotherm	In microcrystalline cellulose	Available 1988
Zinc	In milk, meat, cereal and single cell protein	Available*

[a] See current catalogue for details of individual certified and indicative properties
* Certification reports are given in references with EUR no.

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Received June 9, 1988