II

(Non-legislative acts)

REGULATIONS

COMMISSION IMPLEMENTING REGULATION (EU) 2016/635

of 22 April 2016

amending the Annex to Regulation (EC) No 2870/2000 as regards certain reference methods for the analysis of spirit drinks

THE EUROPEAN COMMISSION,

Having regard to the Treaty on the Functioning of the European Union,

Having regard to Regulation (EC) No 110/2008 of the European Parliament and of the Council of 15 January 2008 on the definition, description, presentation, labelling and the protection of geographical indications of spirit drinks and repealing Council Regulation (EEC) No 1576/89 ([1]), and in particular Article 28(2) thereof,

Whereas:

(1) Commission Regulation (EC) No 2870/2000 ([2]) lists and describes the reference methods for the analysis of spirit drinks. However, some of the methods listed in the Annex to that Regulation, among which, the methods for the determination of volatile acidity and total sugars in spirit drinks, are not yet described.

(2) The methods for the determination of volatile acidity and total sugars in certain spirit drinks have been subjected to two international validation studies that were conducted in accordance with internationally agreed procedures and their method performance parameters have been found to be acceptable. The studies were carried out as part of a research project under the European Commission (EC) framework IV standards measurements and testing (SMT) programme. The description of those methods should therefore be included in the Annex to Regulation (EC) No 2870/2000.

(3) Regulation (EC) No 110/2008 lays down requirements for some categories of spirit drinks to be aged in wood and provides that others may undergo such ageing. Analysis of the principal compounds coming from wood can be helpful when considering if a sample is consistent with the definition corresponding to the relevant category of spirit drink. The International Organisation of Vine and Wine (OIV) has recognised a method of analysis for the determination of those compounds in its Resolution OIV/OENO 382A/2009. The recognition of the method was based on data obtained from an international method-performance study on different spirit drinks carried out following internationally-agreed procedures. This method and its description should therefore be added to the Union reference methods for the analysis of spirit drinks set out in the Annex to Regulation (EC) No 2870/2000.

(4) Regulation (EC) No 2870/2000 should therefore be amended accordingly.

(5) The measures provided for in this Regulation are in accordance with the opinion of the Committee for Spirit Drinks,

([1]) OJ L 39, 13.2.2008, p. 16.
([2]) Commission Regulation (EC) No 2870/2000 of 19 December 2000 laying down Community reference methods for the analysis of spirit drinks (OJ L 333, 29.12.2000, p. 20).

HAS ADOPTED THIS REGULATION:

Article 1

The Annex to Regulation (EC) No 2870/2000 is amended in accordance with the Annex to this Regulation.

Article 2

This Regulation shall enter into force on the third day following that of its publication in the *Official Journal of the European Union*.

This Regulation shall be binding in its entirety and directly applicable in all Member States.

Done at Brussels, 22 April 2016.

For the Commission
The President
Jean-Claude JUNCKER

ANNEX

The Annex to Regulation (EC) No 2870/2000 is amended as follows:

(1) The table of contents is amended as follows:

(a) in points III.3 and VIII, the term '(p.m.)' is deleted;

(b) the following point is added:

'X. Determination of wood compounds: furfural, 5-hydroxymethylfurfural, 5-methylfurfural, vanillin, syringaldehyde, coniferaldehyde, sinapaldehyde, gallic acid, ellagic acid, vanillic acid, syringic acid and scopoletin.'

(2) In Chapter III the following part is added:

'III.3. DETERMINATION OF VOLATILE ACIDITY OF SPIRIT DRINKS

1. **Scope**

The method has been validated in an interlaboratory study for rum, brandy, marc and fruit spirits, at levels ranging from 30 mg/l to 641 mg/l.

2. **Normative references**

ISO 3696: 1987 Water for analytical use — Specifications and test methods.

3. **Definitions**

3.1. Volatile acidity is calculated by deducting the fixed acidity from the total acidity.

3.2. Total acidity is the sum of titratable acidities.

3.3. Fixed acidity is the acidity of the residue left after evaporating the spirit to dryness.

4. **Principle**

The total acidity and fixed acidity are determined by titration or by potentiometry.

5. **Reagents and materials**

During the analysis, unless otherwise stated, use only reagents of recognised analytical grade and water of at least grade 3 as defined in ISO 3696:1987.

5.1. 0,01 M sodium hydroxide solution (NaOH)

5.2. Mixed indicator solution:

Weigh 0,1 g of indigo carmine and 0,1 g of phenol red.

Dissolve in 40 ml water and make up to 100 ml with ethanol.

6. **Apparatus and equipment**

Indirect laboratory apparatus, grade A glassware and the following:

6.1. Water pump

6.2. Rotary evaporator or ultrasonic bath

6.3. Equipment for potentiometric titration (optional).

7. **Sampling and samples**

Samples are stored at room temperature prior to analysis.

8. **Procedure**

8.1. Total acidity

8.1.1. Preparation of sample

The spirit is irradiated with ultrasonic (ultrasonication) or stirred two minutes under a vacuum to rid it of carbon dioxide if required.

8.1.2. Titration

Pipette 25 ml of the spirit into a 500 ml Erlenmeyer flask.

Add about 200 ml of cooled boiled distilled water (prepared fresh daily) and 2-6 drops of the mixed indicator solution (5.2).

Titrate with the 0,01 M sodium hydroxide solution (5.1) until the yellow-green colour changes to violet in the case of colourless spirits, the yellow-brown colour to red-brown in the case of brown-coloured spirits respectively.

The titration may also be carried out by potentiometry, to pH 7,5.

Let n_1 ml be the volume of the 0,01 M sodium hydroxide solution added.

8.1.3. Calculation

The total acidity (TA) expressed in milliequivalents per l of spirit is equal to $0,4 \times n_1$.

The total acidity (TA′) expressed in mg of acetic acid per l of spirit is equal to $24 \times n_1$.

8.2. Fixed acidity

8.2.1. Preparation of sample

Evaporate 25 ml of the spirit to dryness:

Pipette 25 ml of the spirit into a flat-bottomed cylindrical evaporating dish 55 mm in diameter. During the first hour of evaporation the evaporating dish is placed on the lid of a boiling water bath so that the liquid will not boil, as this could lead to losses through splattering.

Complete the drying by placing the evaporating dish in a drying oven at 105 °C for two hours. Allow the evaporating dish to cool in a desiccator.

8.2.2. Titration

Dissolve the residue left after evaporating with cooled boiled distilled water (prepared fresh daily) and make up to a volume to circa 100 ml and add 2-6 drops of the mixed indicator solution (5.2).

Titrate with the 0,01 M sodium hydroxide solution (5.1).

The titration may also be carried out by potentiometry, to pH 7,5.

Let n_2 ml be the volume of the 0,01 M sodium hydroxide solution added.

8.2.3. Calculation

The fixed acidity (FA) expressed in milliequivalents per l of spirit is equal to $0,4 \times n_2$.

The fixed acidity (FA) expressed in mg of acetic acid per l of spirit is equal to $24 \times n_2$.

9. **Calculation of volatile acidity**

9.1. Expression in milliequivalents per l:

Let:

TA = total acidity in milliequivalents per l

FA = fixed acidity in milliequivalents per l

Volatile acidity, VA, in milliequivalents per l is equal to:

TA − FA.

9.2. Expression in mg of acetic acid per l:

Let:

TA′ = total acidity in mg of acetic acid per l

FA′ = fixed acidity in mg of acetic acid per l

Volatile acidity, VA, in mg of acetic acid per l is equal to:

TA′ − FA′.

9.3. Expression in g of acetic acid per hl of pure 100 % vol. alcohol is equal to: $\frac{TA' - FA'}{A} \times 10$

where A is the alcoholic strength by volume of the spirit drink.

10. **Method performance characteristics (Precision)**

10.1. Statistical results of the interlaboratory test

The following data were obtained from an international method performance study carried out to internationally agreed procedures [1] [2].

Year of interlaboratory test	**2000**
Number of laboratories	18
Number of samples	6

Samples	A	B	C	D	E	F
Number of laboratories retained after eliminating outliers	16	18	18	14	18	18
Number of outliers (laboratories)	2			4		
Number of accepted results	32	36	36	28	36	36
Mean value ($\bar{x}$) (mg/L)	272* 241*	30	591* 641*	46	107	492
Repeatability standard deviation, s_r (mg/l)	8,0	3,6	15,0	3,7	6,7	8,5
Repeatability relative standard deviation, RSDr (%)	3,1	11,8	2,4	8,0	6,2	1,7
Repeatability limit, r (mg/l)	23	10	42	10	19	24
Reproducibility standard deviation, s_R (mg/l)	8,5	8,4	25,0	4,55	13,4	24,4
Reproducibility relative standard deviation, RSD_R (%)	3,3	27,8	4,1	9,9	12,5	5,0
Reproducibility limit, R (mg/l)	24	23	70	13	38	68

Sample types:

A Plum spirit; split level *

B Rum I; blind duplicates

C Rum II; split level *

D Slivovitz; blind duplicates

E Brandy; blind duplicates

F Marc spirit; blind duplicates.

([1]) "Protocol for the design, conduct and interpretation of method-performance studies", Horwitz, W. (1995) *Pure and Applied Chemistry*, 67, 332-343.

([2]) Horwitz, W. (1982) *Analytical Chemistry*, 54, 67A-76A.'

(3) The following Chapter VIII is inserted:

'VIII. **TOTAL SUGARS**

1. **Scope**

The HPLC–RI method is applicable for the determination of total sugars (expressed as invert sugar) in spirit drinks, with the exclusion of liqueurs containing egg and milk products.

The method has been validated in an interlaboratory study for pastis, distilled anis, cherry liqueur, crème de (followed by the name of a fruit or the raw material used) and crème de cassis, at levels ranging from 10,86 g/l to 509,7 g/l. However, linearity of the instrument response was proven for the concentration range 2,5 g/l to 20,0 g/l.

This method is not intended for determining low levels of sugars.

2. **Normative references**

ISO 3696:1987 Waters for analytical use — Specifications and test methods.

3. **Principle**

High-performance liquid chromatography assays of sugar solutions, in order to determine their glucose, fructose, sucrose, maltose and lactose concentrations.

This method uses an alkylamine stationary phase and differential refractometry detection and is given as an example. The use of anion exchange resins as stationary phase would also be possible.

4. **Reagents and materials**

4.1. Glucose (CAS 50-99-7), at least 99 % pure.

4.2. Fructose (CAS 57-48-7), at least 99 % pure.

4.3. Sucrose (CAS 57-50-1), at least 99 % pure.

4.4. Lactose (CAS 5965-66-2), at least 99 % pure.

4.5. Maltose monohydrate (CAS 6363-53-7), at least 99 % pure.

4.6. Pure acetonitrile (CAS 75-05-8) for HPLC analysis.

4.7. Distilled or demineralised water, preferably microfiltered.

4.8. Solvents (example)

The elution solvent is composed of:

75 parts by volume of acetonitrile (4.6),

25 parts by volume of distilled water (4.7).

Pass helium through at a slow rate for 5-10 minutes prior to use to degas.

If the water being used has not been microfiltered, the solvent should be filtered with a filter for organic solvents with a pore size less than or equal to 0,45 µm.

4.9. Ethanol absolute (CAS 64-17-5).

4.10. Ethanol solution (5 %, v/v).

4.11. Preparation of stock standard solution (20 g/l)

Weigh 2 g each of the sugars to be analysed (4.1 to 4.5), transfer them without loss to a 100 ml volumetric flask. (NB 2,11 g of maltose monohydrate is equivalent to 2 g of maltose).

Adjust to 100 ml with a 5 % vol. alcohol solution (4.10), shake and store at around + 4 °C. Prepare a new stock solution once a week.

4.12. Preparation of working standard solutions (2,5, 5,0, 7,5, 10,0 and 20,0 g/L)

Dilute the stock solution, 20 g/l (4.11) appropriately with a 5 % vol. alcohol solution (4.10) to give five working standards of 2,5, 5,0, 7,5, 10,0 and 20,0 g/l. Filter with a filter of a pore size less than or equal to 0,45 µm (5.3).

5. **Apparatus and Equipment**

5.1. HPLC system capable of achieving baseline resolution of all of the sugars.

5.1.1. High-performance liquid chromatograph with a six-way injection valve fitted with a 10 µl loop or any other device, whether automatic or manual, for the reliable injection of microvolumes.

5.1.2. Pumping system enabling one to achieve and maintain a constant or programmed rate of flow with great precision.

5.1.3. Differential refractometer.

5.1.4. Computational integrator or recorder, the performance of which is compatible with the rest of the set-up.

5.1.5. Pre-column:

It is recommended that a suitable pre-column is attached to the analytical column.

5.1.6. Column (example):

Material:	stainless steel or glass.
Internal diameter:	2-5 mm.
Length:	100-250 mm (depending on the packing particle size), for example, 250 mm if the particles are 5 µm in diameter.
Stationary phase:	alkylamine functional groups bonded to silica, maximum particle size 5 µm.

5.1.7. Chromatography conditions (example):

Elution solvent (4.8), flow rate: 1 ml/minute.

Detection: Differential refractometry.

To make certain that the detector is perfectly stable, it should be switched on a few hours before use. The reference cell must be filled with the elution solvent.

5.2. Analytical balance accurate to 0,1 mg.

5.3. Filtration set-up for small volumes using a 0,45 µm micromembrane.

6. **Sample storage**

On receipt, samples are to be stored at room temperature prior to analysis.

7. **Procedure**

7.1. PART A: Sample preparation

7.1.1. Shake the sample.

7.1.2. Filter the sample through a filter with a pore size less than or equal to 0,45 µm (5.3).

7.2. PART B: HPLC

7.2.1. Determination

Inject 10 µl of the standard solutions (4.12) and samples (7.1.2). Perform the analysis under suitable chromatography conditions, for example those described above.

7.2.2. Should any peak of a sample have a greater area (or height) than the corresponding peak in the most concentrated standard, then the sample should be diluted with distilled water and reanalysed.

8. Calculation

Compare the two chromatograms obtained for the standard solution and spirit. Identify the peaks by their retention times. Measure their areas (or heights) to calculate the concentrations by the external standard method. Take into account any dilutions made to the sample.

The final result is the sum of sucrose, maltose, lactose, glucose and fructose, expressed as invert sugar in g/l.

Invert sugar is calculated as the sum of all monosaccharides and reducing disaccharides present, plus the stoichiometric amount of glucose and fructose calculated from the sucrose present.

Invert sugar (g/l) = glucose (g/l) + fructose (g/l) + maltose (g/l) + lactose (g/l) + (sucrose (g/l) × 1,05).

1,05 = (molecular weight of fructose + molecular weight of glucose)/molecular weight of sucrose.

9. Method performance characteristics (precision)

9.1. Statistical results of the interlaboratory test

The following data were obtained from an international method performance study carried out to internationally agreed procedures [1] [2].

Year of interlaboratory test 2000

Number of laboratories 24

Number of samples 8

[1] "Protocol for the design, conduct and interpretation of method-performance studies", Horwitz, W. (1995) *Pure and Applied Chemistry*, 67, 332-343.

[2] Horwitz, W. (1982) *Analytical Chemistry*, 54, 67A-76A.

Table 1

Fructose, glucose, maltose

Analyte	Fructose		Glucose			Maltose	
Samples (× 2)	Crème de Cassis	Standard (50 g/l)	Aniseed-flavoured spirit drink	Crème de Cassis	Standard (50 g/l)	Aniseed-flavoured spirit drink	Standard (10 g/l)
Mean value (g/l)	92,78	50,61	15,62	93,16	50,06	15,81	9,32
No of labs without outliers	21	22	21	23	19	21	22
Repeatability standard deviation, s_r, (g/l)	2,34	2,12	0,43	3,47	1,01	0,48	0,54

Analyte	Fructose		Glucose			Maltose	
Samples (× 2)	Crème de Cassis	Standard (50 g/l)	Aniseed-flavoured spirit drink	Crème de Cassis	Standard (50 g/l)	Aniseed-flavoured spirit drink	Standard (10 g/l)
Repeatability relative standard deviation, RSD_r (%)	2,53	4,2	2,76	3,72	2,03	3,02	5,77
Repeatability limit, r (g/l) ($r = 2,8 \times s_r$)	6,56	5,95	1,21	9,71	2,84	1,34	1,51
Reproducibility standard deviation, s_R (g/l)	7,72	3,13	0,84	9,99	2,7	0,88	1,4
Reproducibility relative standard deviation, RSD_R (%)	8,32	6,18	5,37	10,72	5,4	5,54	15,06
Reproducibility limit, R (g/l) (R = 2,8 × sR)	21,62	8,76	2,35	27,97	7,57	2,45	3,93

Table 2

Sucrose

Analyte	Sucrose					
Samples	Pastis	Ouzo	Cherry liqueur	Crème de Menthe	Crème de Cassis	Standard (100 g/l)
Mean value (g/l)	10,83	29,2 19,7 (*)	103,33	349,96	319,84	99,83
No of labs without outliers	19	19	20	18	18	18
Repeatability standard deviation, s_r (g/l)	0,09	0,75	2,17	5,99	4,31	1,25
Repeatability relative standard deviation, RSD_r (%)	0,81	3,07	2,1	1,71	1,35	1,25
Repeatability limit, r (g/l) ($r = 2,8 \times s_r$)	0,25	2,1	6,07	16,76	12,06	3,49
Reproducibility standard deviation, s_R (g/l)	0,79	0,92	4,18	9,94	16,11	4,63
Reproducibility relative standard deviation, RSD_R (%)	7,31	3,76	4,05	2,84	5,04	4,64
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	2,22	2,57	11,7	27,84	45,12	12,97

(*) split level.

Table 3

Total Sugars

(*Note:* this data was calculated for total sugars, not invert sugar as defined in Section 8 above.)

Samples	Pastis	Ouzo	Aniseed-flavoured spirit drink	Cherry liqueur	Crème de Menthe	Crème de Cassis	Standard (220 g/l)
Mean value (g/l)	10,86	29,2 19,7 (*)	31,59	103,33	349,73	509,69	218,78
No of Labs without outliers	20	19	20	20	18	18	19
Repeatability standard deviation, s_r (g/l)	0,13	0,75	0,77	2,17	5,89	5,59	2,71
Repeatability relative standard deviation, RSD_r (%)	1,16	3,07	2,45	2,1	1,69	1,1	1,24
Repeatability limit, r (g/l) ($r = 2,8 \times s_r$)	0,35	2,1	2,17	6,07	16,5	15,65	7,59
Reproducibility standard deviation s_R (g/l)	0,79	0,92	1,51	4,18	9,98	14,81	8,53
Reproducibility relative standard deviation, RSD_R (%)	7,25	3,76	4,79	4,04	2,85	2,91	3,9
Reproducibility limit R (g/l) ($R = 2,8 \times s_R$)	2,21	2,57	4,24	11,7	27,94	41,48	23,89

(*) split level.'

(4) The following Chapter X is added:

'X. **DETERMINATION OF THE FOLLOWING WOOD COMPOUNDS IN SPIRIT DRINKS BY HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC): FURFURAL, 5-HYDROXYMETHYLFURFURAL, 5-METHYLFURFURAL, VANILLIN, SYRINGALDEHYDE, CONIFERALDEHYDE, SINAPALDEHYDE, GALLIC ACID, ELLAGIC ACID, VANILLIC ACID, SYRINGIC ACID AND SCOPOLETIN**

1. **Scope**

The method pertains to the determination of furfural, 5-hydroxymethylfurfural, 5-methylfurfural, vanillin, syringaldehyde, coniferaldehyde, sinapaldehyde, gallic acid, ellagic acid, vanillic acid, syringic acid and scopoletin, by high-performance liquid chromatography.

2. **Normative reference**

Analytical method recognised by the General Assembly of the International Organisation of Vine and Wine (OIV) and published by OIV under the reference *OIV-MA-BS-16: R2009*.

3. **Principle**

Determination by high-performance liquid chromatography (HPLC), with detection by ultraviolet spectrophotometry at several wavelengths and by spectrofluorimetry.

4. **Reagents**

The reagents must be of analytical quality. The water used must be distilled water or water of at least equivalent purity. It is preferable to use microfiltered water with a resistivity of 18,2 M Ω.cm.

4.1. 96 % vol. alcohol.

4.2. HPLC-quality methanol (Solvent B).

4.3. Acetic acid diluted to 0,5 % vol. (Solvent A).

4.4. Mobile phases: (given as an example only).

Solvent A (0,5 % acetic acid) and solvent B (pure methanol). Filter through a membrane (porosity 0,45 µm). Degas in an ultrasonic bath, if necessary.

4.5. Reference standards of 99 % minimum purity: furfural, 5-hydroxymethyl furfural, 5-methylfurfural, vanillin, syringaldehyde, coniferaldehyde, sinapaldehyde, gallic acid, ellagic acid, vanillic acid, syringic acid and scopoletin.

4.6. Reference solution: the standard substances are dissolved in a 50 % vol. aqueous-alcoholic solution. The final concentrations in the reference solution should be of the order of:

furfural: 5 mg/l; 5-hydroxymethyl furfural: 10 mg/l; 5-methylfurfural 2 mg/l; vanillin: 5 mg/l; syringaldehyde: 10 mg/l; coniferaldehyde: 5 mg/l; sinapaldehyde: 5 mg/l; gallic acid: 10 mg/l; ellagic acid: 10 mg/l; vanillic acid: 5 mg/l; syringic acid: 5 mg/l; scopoletin: 0,5 mg/l.

5. **Apparatus**

Standard laboratory apparatus

5.1. A high-performance liquid chromatograph capable of functioning in binary gradient mode and equipped with:

5.1.1. A spectrophotometric detector capable of measuring at wavelengths from 260 to 340 nm. It is however preferable to work with a multiple wavelength detector with a diode array or similar, in order to confirm the purity of the peaks.

5.1.2. A spectrofluorimetric detector — excitation wavelength: 354 nm, emission wavelength: 446 nm (for the trace determination of scopoletin; which is also detectable at 313 nm by spectrophotometry).

5.1.3. An injection device capable of introducing 10 or 20 µl (for example) of the test sample.

5.1.4. A high-performance liquid chromatography column, RP C18 type, 5 µm maximum particle size.

5.2. Syringes for HPLC.

5.3. Device for membrane-filtration of small volumes.

5.4. Integrator-computer or recorder with performance compatible with the entire apparatus, and in particular, it must have several acquisition channels.

6. **Procedure**

6.1. Preparation of the solution to be injected

The reference solution and the spirit drink are filtered, if necessary, through a membrane with a maximum pore diameter of 0,45 µm.

6.2. Chromatographic operating conditions: carry out the analysis at ambient temperature by means of the equipment described in (5.1) and using the mobile phases (4.4) with a flow of approximately 0,6 ml per minute following the gradient below (given as an example only)

Time: 0 min 50 min 70 min 90 min

solvent A (water-acid): 100 % 60 % 100 % 100 %

solvent B (methanol): 0 % 40 % 0 % 0 %

Note that in certain cases this gradient should be modified to avoid co-elutions.

6.3. Determination

6.3.1. Inject the reference standards separately, then mixed.

Adapt the operating conditions so that the resolution factors of the peaks of all the compounds are equal to at least 1.

6.3.2. Inject the sample as prepared in 6.1.

6.3.3. Measure the area of the peaks in the reference solution and the spirit drink and calculate the concentrations.

7. **Expression of results**

Express the concentration of each constituent in mg/l.

8. **Performance characteristics of the method (precision)**

The following data were obtained in 2009 from an international method-performance study on a variety of spirit drinks, carried out following internationally-agreed procedures ([1]) ([2]).

8.1. Furfural

Analyte	Furfural					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	15	15	15	15	15	15
No of results accepted (laboratories)	14	12	13	14	13	13
Mean value (mg/l)	2,9	1,2	1,7	10,6	15,3	13,9
Repeatability standard deviation, s_r (mg/l)	0,04	0,05	0,04	0,18	0,23	0,20
Repeatability relative standard deviation, RSD_r (%)	1,4	4,5	2,3	1,7	1,5	1,5

Analyte	Furfural					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,1	0,2	0,1	0,5	0,6	0,6
Reproducibility standard deviation, s_R (mg/l)	0,24	0,18	0,09	1,4	0,49	0,69
Reproducibility relative standard deviation, RSD_R (%)	8	15	5	13	3	5
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,7	0,5	0,3	3,8	1,4	1,9

8.2. 5-Hydroxymethylfurfural

Analyte	5-Hydroxymethylfurfural					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	16	16	16	16	16	16
No of results accepted (laboratories)	14	14	14	14	14	14
Mean value (mg/l)	5,0	11,1	9,4	33,7	5,8	17,5
Repeatability standard deviation, s_r (mg/l)	0,09	0,09	0,09	0,42	0,07	0,13
Repeatability relative standard deviation, RSD_r (%)	1,7	0,8	1,0	1,3	1,2	0,8
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,2	0,3	0,3	1,2	0,2	0,4
Reproducibility standard deviation, s_R (mg/l)	0,39	1,01	0,50	4,5	0,4	1,6
Reproducibility relative standard deviation, RSD_R (%)	8	9	5	13	7	9
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	1,1	2,8	1,4	12,5	1,1	4,6

8.3. 5-Methylfurfural

Analyte	5-Methylfurfural					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	11	11	11	11	11	11
No of results accepted (laboratories)	11	11	8	11	10	11
Mean value (mg/l)	0,1	0,2	0,1	0,5	1,7	0,8
Repeatability standard deviation, s_r (mg/l)	0,01	0,01	0,02	0,02	0,03	0,07
Repeatability relative standard deviation, RSD_r (%)	10,7	6,1	13,6	4,7	2,0	10,0
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,0	0,0	0,1	0,1	0,1	0,2
Reproducibility standard deviation, s_R (mg/l)	0,03	0,04	0,03	0,18	0,20	0,26
Reproducibility relative standard deviation, RSD_R (%)	35	18	22	39	12	35
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,1	0,1	0,1	0,5	0,6	0,7

8.4. Vanillin

Analyte	Vanillin					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	16	15	16	16	16	16
No of results accepted (laboratories)	16	15	16	16	16	16
Mean value (mg/l)	0,5	0,2	1,2	1,2	3,2	3,9
Repeatability standard deviation, s_r (mg/l)	0,03	0,02	0,06	0,11	0,11	0,09

Analyte	Vanillin					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
Repeatability relative standard deviation, RSD_r (%)	6,8	9,6	4,6	8,9	3,5	2,3
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,1	0,1	0,2	0,3	0,3	0,3
Reproducibility standard deviation, s_R (mg/l)	0,09	0,06	0,18	0,27	0,41	0,62
Reproducibility relative standard deviation, RSD_R (%)	19	25	15	22	13	16
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,3	0,2	0,5	0,8	1,2	1,7

8.5. Syringaldehyde

Analyte	Syringaldehyde					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	16	15	16	16	16	16
No of results accepted (laboratories)	13	13	13	12	14	13
Mean value (mg/l)	1,0	0,2	4,8	3,2	10,5	9,7
Repeatability standard deviation, s_r (mg/l)	0,03	0,02	0,04	0,08	0,10	0,09
Repeatability relative standard deviation, RSD_r (%)	2,6	8,1	0,8	2,6	0,9	0,9
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,1	0,1	0,1	0,2	0,3	0,3
Reproducibility standard deviation, s_R (mg/l)	0,08	0,07	0,23	0,19	0,39	0,43
Reproducibility relative standard deviation, RSD_R (%)	8	33	5	6	4	4
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,2	0,2	0,7	0,5	1,1	1,2

8.6. Coniferaldehyde

Analyte	Coniferaldehyde					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	13	12	13	12	13	13
No of results accepted (laboratories)	12	12	13	12	13	13
Mean value (mg/l)	0,2	0,2	0,6	0,8	4,6	1,3
Repeatability standard deviation, s_r (mg/l)	0,02	0,02	0,03	0,03	0,09	0,06
Repeatability relative standard deviation, RSD_r (%)	9,2	9,8	4,6	4,3	1,9	4,5
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,04	0,04	0,07	0,09	0,24	0,16
Reproducibility standard deviation, s_R (mg/l)	0,04	0,04	0,11	0,18	0,38	0,25
Reproducibility relative standard deviation, RSD_R (%)	23	27	21	23	8	19
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,1	0,1	0,3	0,5	1,1	0,7

8.7. Sinapaldehyde

Analyte	Sinapaldehyde					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	14	14	14	14	15	14
No of results accepted (laboratories)	14	13	12	13	13	12
Mean value (mg/l)	0,3	0,2	0,2	1,6	8,3	0,3
Repeatability standard deviation, s_r (mg/l)	0,02	0,01	0,02	0,06	0,14	0,03

Analyte	Sinapaldehyde					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
Repeatability relative standard deviation, RSD_r (%)	7,5	4,6	11,2	3,7	1,6	11,4
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,06	0,03	0,06	0,17	0,38	0,08
Reproducibility standard deviation, s_R (mg/l)	0,09	0,05	0,08	0,20	0,81	0,18
Reproducibility relative standard deviation, RSD_R (%)	31	27	46	13	10	73
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,2	0,2	0,2	0,6	2,3	0,5

8.8. Gallic acid

Analyte	Gallic acid					
Sample	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	16	15	16	16	16	16
No of results accepted (laboratories)	15	14	16	16	16	16
Mean value (mg/l)	1,2	0,4	2,0	6,1	7,3	21,8
Repeatability standard deviation, s_r (mg/l)	0,07	0,04	0,06	0,18	0,18	0,60
Repeatability relative standard deviation, RSD_r (%)	6,1	8,1	2,9	3,0	2,4	2,8
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,2	0,1	0,2	0,5	0,5	1,7
Reproducibility standard deviation, s_R (mg/l)	0,43	0,20	0,62	3,3	2,2	7,7
Reproducibility relative standard deviation, RSD_R (%)	36	47	31	53	30	35
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	1,2	0,6	1,7	9,1	6,2	21,7

8.9. Ellagic acid

Analyte	Ellagic acid					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	7	7	7	7	7	7
No of results accepted (laboratories)	7	7	7	7	7	6
Mean value (mg/l)	3,2	1,0	9,5	13	13	36
Repeatability standard deviation, s_r (mg/l)	0,20	0,16	0,30	0,41	0,95	0,34
Repeatability relative standard deviation, RSD_r (%)	6,3	16	3,2	3,2	7,4	1,0
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,6	0,4	0,9	1,1	2,7	1,0
Reproducibility standard deviation, s_R (mg/l)	1,41	0,42	4,0	5,0	4,9	14
Reproducibility relative standard deviation, RSD_R (%)	44	43	42	39	39	40
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	4,0	1,2	11	14	14	40

8.10. Vanillic acid

Analyte	Vanillic acid					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	15	15	15	15	15	15
No of results accepted (laboratories)	12	11	14	14	15	14
Mean value (mg/l)	0,2	0,2	1,5	0,8	2,4	2,7
Repeatability standard deviation, s_r (mg/l)	0,03	0,04	0,03	0,10	0,13	0,21

Analyte	Vanillic acid					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
Repeatability relative standard deviation, RSD_r (%)	14,2	16,5	2,3	12,6	5,3	7,7
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,1	0,1	0,1	0,3	0,4	0,6
Reproducibility standard deviation, s_R (mg/l)	0,06	0,05	0,51	0,2	1,22	0,70
Reproducibility relative standard deviation, RSD_R (%)	28	20	35	31	51	26
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,2	0,1	1,4	0,7	3,4	2,0

8.11. Syringic acid

Analyte	Syringic acid					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	16	15	16	16	16	16
No of results accepted (laboratories)	16	15	15	15	16	15
Mean value (mg/l)	0,4	0,2	2,5	1,4	3,4	4,8
Repeatability standard deviation, s_r (mg/l)	0,03	0,02	0,06	0,13	0,08	0,11
Repeatability relative standard deviation, RSD_r (%)	6,7	12,6	2,3	9,0	2,3	2,3
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,1	0,1	0,2	0,4	0,2	0,3
Reproducibility standard deviation, s_R (mg/l)	0,08	0,05	0,29	0,26	0,43	0,67
Reproducibility relative standard deviation, RSD_R (%)	19	29	11	18	13	14
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,2	0,1	0,8	0,7	1,2	1,9

8.12. Scopoletin

Analyte	Scopoletin					
Samples	Whisky	Brandy	Rum	Cognac 1	Bourbon	Cognac 2
No of laboratories participating	10	10	10	10	10	10
No of results accepted (laboratories)	9	8	9	8	8	8
Mean value (mg/l)	0,09	0,04	0,11	0,04	0,65	0,15
Repeatability standard deviation, s_r (mg/l)	0,0024	0,0008	0,0018	0,0014	0,0054	0,0040
Repeatability relative standard deviation, RSD_r (%)	2,6	2,2	1,6	3,3	0,8	2,7
Repeatability limit, r (mg/l) ($r = 2,8 \times s_r$)	0,007	0,002	0,005	0,004	0,015	0,011
Reproducibility standard deviation, s_R (mg/l)	0,01	0,01	0,03	0,01	0,09	0,02
Reproducibility relative standard deviation, RSD_R (%)	15	16	23	17	15	15
Reproducibility limit, R (g/l) ($R = 2,8 \times s_R$)	0,04	0,02	0,07	0,02	0,26	0,06

(1) "Protocol for the design, conduct and interpretation of method-performance studies", Horwitz, W. (1995) *Pure and Applied Chemistry*, 67, 332-343.

(2) Horwitz, W. (1982) *Analytical Chemistry*, 54, 67A-76A.'