OIV-MA-AS323-10 Method of determination of phthalates by gas chromatography/ mass spectrometry in wines

Type II/IV methods

1. Scope

This method applies to the detection and assay of phthalates in wines.

2. Principle

The sample is extracted using isohexane. The extract is concentrated by evaporation. The concentrated extract is analysed by gas chromatography/mass spectrometry (GC/MS) with deuterated internal standards.

3. Reagents and materials

Unless otherwise specified, all the reagents used are of recognised analytical quality.

3.1. DMP (dimethyl phthalate) [CAS N°: 131-11-3]
3.2. DnBP (dibutyl phthalate) [CAS N°: 84-74-2]
3.3. DEHP (bis (2-ethylhexyl) phthalate) [CAS N°: 117-81-7]
3.4. BBP (butyl benzyl phthalate) [CAS N°: 85-68-7]
3.5. DINP (di-isononyl phthalate) [CAS N°: 068515-48-0/028553-12-0]
3.6. DIDP (di-isodecyl phthalate) [CAS N°: 068515-49-1/026761-40-0]
3.7. DCHP (dicyclohexyl phthalate) [CAS N°: 84-61-7]
3.8. DEP (diethyl phthalate) [CAS N°: 84-66-2]
3.9. DiBP (di-isobutyl phthalate) [CAS N°: 84-74-2]
3.10. DnOP (di-n-octyl phthalate) [CAS N°: 117-84-0]
3.11. DMP-d4: internal standard [CAS N°: 93951-89-4]
3.12. DEP-d4: internal standard [CAS N°: 93952-12-6]
3.13. DiBP-d4: internal standard [CAS N°: 358730-88-8]
3.14. DnBP-d4: internal standard [CAS N°: 93952-11-5]
3.15. BBP-d4: internal standard [CAS N°: 93951-88-3]
3.16. DCHP-d4: internal standard [CAS N°: 358731-25-6]
3.17. DEHP–d4: internal standard [CAS N°: 93951-87-2]
3.18. DnOP-d4: internal standard [CAS N°: 93952-13-7]
3.19. Isohexane [CAS N°: 107-83-5] and Acetone [CAS N°: 67-64-1]
3.20. Standard solutions

All the volumetric flasks used to prepare the calibration solutions are to be rinsed with acetone then isohexane to avoid any contamination.

3.20.1. Stock solutions

Phthalate - 1 g/L individual solution: for each phthalate weigh 100 mg into a 100 mL flask, dissolve in the isohexane and make up to 100 mL.

DINP-DIDP– 5 g/L individual solution: for each phthalate weigh 500 mg into a 100 mL flask, dissolve in the isohexane and make up to 100 mL.

Internal standard - 0.5 g/L individual solution: deuterated standards are packaged in sealed 25 mg ampoules; for each internal standard, all the contents of the bulb are transferred into a 50 mL volumetric flask; make up to 50 mL with isohexane.

3.20.2. Working solutions

Phthalate 1 mg/L working solution (S1)

Take 100 µL of each 1 g/L and 5g/L stock solution (3.20.1), add the samples to a 100 mL flask, and make up to 100 mL with isohexane.

Phthalate 10 mg/L working solution (S2)

Take 1 mL of each 1 g/L and 5g/L stock solution (3.20.1), add the samples to a 100 mL flask, and make up to 100 mL with isohexane.

Internal standard 10 mg/L working solution (IS)

Take 1 mL of each deuterated standard 0.5 g/L stock solution (3.20.1), add the samples to a 50 mL flask, and make up to 50 mL with isohexane.

3.20.3. Calibration range

The calibration range in isohexane is prepared from the various working solutions (3.20.2), directly into the injection vials that have been heat-treated, rinsed (see § 5.1) and dried under a hood beforehand, according to the following table:

Calibration points	Phthalate concn. (mg/L)*	Vol. of S1 surrogate soln. (µL)	Vol. of S2 surrogate soln. (µL)	Vol. of IS surrogate soln. (µL)	Vol. of isohexane (µL)
C1	0	0	0	50	1000
C2	0,05	50	0	50	950
C3	0,10	100	0	50	900
C4	0,20	200	0	50	800

C5	0,50	0	50	50	950
C6	0,80	0	80	50	920
C7	1,00	0	100	50	900

* to be multiplied by 5 for DINP and DIDP concentrations

4. Equipment

4.1. Glassware and volumetric laboratory equipment:

4.1.1. 50 mL and 100 mL class A volumetric flasks

4.1.2. 50 mL glass centrifuge tubes with stopper

4.1.3. 10 mL glass test tubes with stopper

4.1.4. Micropipettes with variable volumes ranging from 25 µl to 1,000 µl, checked in accordance with ISO 8655-6

4.1.5. Nitrogen flow evaporator

4.2. Analytical balance

4.3. GC-MS System (e.g. Varian 450GC-300MS)

5. Procedure

5.1. Precautions

Due to the presence of phthalates in the laboratory environment, precautions must be taken throughout the analysis of these compounds:

- Avoid any contact with plastic equipment (especially flexible PVC) as much as possible. If this is not possible, make sure there is no contamination.
- Test the solvents used and dedicate bottles of solvent to these analyses.
- Heat-treat all non-volumetric glassware (400°C for at least 2 hours). Rinse all the equipment carefully (with acetone then isohexane).
- Make sure the septums of the injection vials are phthalate-free.
- Before and after each injection, rinse the injection syringe several times.
- If possible, work in a clean room or in a room reserved for these analyses.

5.2. Preparing the samples

Place 12.5 mL of the sample in a 50 mL centrifuge tube. Add 10 mL of isohexane.

Shake vigorously (Vortex mixer) for at least one minute.

Let the mixture decant until the 2 phases have separated (30 minutes in a 50°C ultrasound bath will accelerate the separation). Recover 8 mL of the organic phase and transfer it into a 10 mL test tube. Evaporate under a flow of nitrogen (0.3 bar) at 35°C and avoid continuing to dryness (warning: the temperature must not exceed 40°C)

Resume with 1 mL of isohexane.

Add 50 µl of the 0.01 g/L internal standard solution to each extract.

Transfer into an injection vial.

NOTE: to minimise matrix effects during analysis by GC-MS, a “protective” agent can be added, such as methyl undecanoate [CAS N°: 1731-86-8].

Add 20 µL of this compound is added to each calibration solution and to the extracts from the samples prior to evaporation under a flow of nitrogen.

5.3. Blank test

Prepare a “blank” test by following the procedure described in 5.2 without adding the sample.

5.4. GC/MS analysis

Depending on the apparatus available and its performance, choose between SIM and MRM modes for the mass spectrometry.

For information purposes, analysis conditions are provided in Appendix I and a typical chromatogram is provided in Appendix II.

5.4.1. Calibration

First, carry out several solvent injections (at least 2). Next, inject the standard solutions (3.20.3) in duplicate in increasing order of concentration and end with at least two solvent injections.

Establish a calibration curve for each phthalate:

$$(A_{analyte}/A_{IS}) = f(C_{analyte}/C_{IS})$$

A: peak area

C: concentration

IS: internal standard

Each phthalate is quantified using to the corresponding deuterated standard, with the exception of DINP and DIDP which are quantified using to DnOP-d4.

5.4.2. Analysing the samples

Start the analysis sequence by analysing the "blank" test (5.3).

Then inject the samples prepared (5.2) in duplicate.

Plan solvent injections after potentially highly contaminated samples.

End the series by injecting one or more calibration standards to check any signal drift during the analysis series and to check several solvent injections..

For each injection, measure the area of the identified peaks and internal standards, and use the calibration curve equation (5.4.1) to determine the concentration in the extract analysed.

5.4.3. Expressing the results

For each sample, calculate the average of the results obtained (5.4.2) for both injections.

The results are expressed in mg/L.

6. Quality control

During each analysis series, quality control is provided by the analysis of a wine sample supplemented with phthalates at a concentration level of 0.020 mg/L.

The extract of the sample prepared as per 5.2 is analysed at the beginning of the series, and the results obtained, given in terms of recovery rate, are reflected on a control chart.

7. Method characteristics

The analyses performed in the laboratory, under repeatability and intermediate precision conditions, on a red wine and a white wine supplemented with phthalates at two concentration levels (0.040 mg/L and 0.080 mg/L), gave the following repeatability (CV_r%), intermediate reproducibility (CV_{IP}%), and recovery values:

Phthalates	Recovery %	CV_r%	CV_{IP}%
DMP (dimethyl phthalate)	67	5	8
DEP (diethyl phthalate)	84	8	11
DiBP (di-isobutyl phthalate)	93	7	10

DnBP (dibutyl phthalate)	95	5	7
BBP (butyl benzyl phthalate)	98	5	6
DCHP (dicyclohexyl phthalate)	97	5	7
DEHP (bis(2-ethylhexyl) phthalate)	98	6	7
DnOP (dioctyl phthalate)	98	6	7
DINP (di-isononyl phthalate)	104	7	8
DIDP (di-isodecyl phthalate)	96	8	11

i.e. the following average values for all the phthalates:

Repeatability (given in CV_r%): 6%

Intermediate precision (given in CV_{IP}%): 8%

8. Detection and quantification limits

For each phthalate being analysed for, the detection and quantification limits are provided in the following table:

Phthalates	Quantification limit (mg/L)	Detection limit (mg/L)
DMP (dimethyl phthalate)	0.010	0.004
DEP (diethyl phthalate)	0.010	0.004

DiBP (di-isobutyl phthalate)	0.010	0.004
DnBP (dibutyl phthalate)	0.010	0.004
BBP (butyl benzyl phthalate)	0.010	0.004
DCHP (dicyclohexyl phthalate)	0.010	0.004
DEHP (bis(2-ethylhexyl) phthalate)	0.010	0.004
DnOP (dioctyl phthalate)	0.010	0.004
DINP (di-isononyl phthalate)	0.050	0.020
DIDP (di-isodecyl phthalate)	0.050	0.020

9. References

- FV 1371. DETECTION AND ASSAY OF PHTHALATES IN ALCOHOLIC BEVERAGES. 2011
- FV 1234. QUESTIONS ABOUT PHTHALATES. 2006

Appendix I
(for information)

- Gas chromatography conditions
- VF-5ms type capillary column: 30 m x 0.25 mm internal diameter, film thickness 0.25 µm
- Temperature programming:

For detection in SIM mode:

Oven maintained at 100°C for 1 min; increase to 230°C at a rate of 10°C/min; increase to 270°C at a rate of 10°C/min; maintain for 2 min, increase to 300°C at a rate of 25°C/min; maintain for 8 min.

Note: this programming separates the DEHP and DCHP peaks (which cannot be done with the MRM mode programming)

For detection in MRM mode:

Oven maintained at 80°C for 1 min; increase to 200°C at a rate of 20°C/min; increase to 300°C at a rate of 10°C/min; maintain for 8 min.

Injector: maintained at 150°C for 0.5 min; increase to 280°C at a rate of 200°C/min, in splitless mode at injection

Helium: 1 mL/min at a constant flow rate

Volume injected: 1 µL

Mass spectrometry (MS) conditions

Ionisation in EI mode at 70 eV

Source temperature: 250°C

Transfer line temperature: 300°C

Manifold: 40°C

Phthalate quantification and identification parameters

For an analysis in SIM mode, table 1 provides the quantification ion and the two qualifier ions for each phthalate and its deuterated homologue.

For an analysis in MRM mode, table 2 reflects the quantifying and qualifying transitions for each phthalate and its deuterated homologue.

Note: DIDP and DINP are each a mixture of compounds. Chromatography cannot separate them completely. They are therefore assayed as a "group".

Appendix I

(for information)

Table 1

		Quantification ion m/z	Qualifier ions m/z 1	Qualifier ions m/z 2
DMP	(dimethyl phthalate)	163	77	194
DMP-d4		167	81	198
DEP	(diethyl phthalate)	149	177	222
DEP-d4		153	181	226
DiBP	(di-isobutyl phthalate)	149	167	223
DiBP-d4		153	171	227
DnBP	(dibutyl phthalate)	149	205	223
DnBP-d4		153	209	227
BBP	(butyl benzyl phthalate)	149	91	206
BBP-d4		153	95	210
DCHP	(dicyclohexyl phthalate)	149	167	249
DCHP-d4		153	171	253
DEHP	(bis(2-ethylhexyl) phthalate)	149	167	279
DEHP-d4		153	171	283
DnOP	(dioctyl phthalate)	149	167	279
DnOP-d4		153	171	283
DINP	(di-isononyl phthalate)	149	293	
DIDP	(di-isodecyl phthalate)	149	307	

Table 2

		Quantifying transition	Qualifying transition
DMP	(dimethyl phthalate)	194>163	194>77
DMP-d4		198>167	198>81
DEP	(diethyl phthalate)	177>149	177>93
DEP-d4		181>153	181>97
DiBP	(di-isobutyl phthalate)	223>149	205>149
DiBP-d4		227>153	209>153
DnBP	(dibutyl phthalate)	223>149	205>149
DnBP-d4		227>153	209>153
BBP	(butyl benzyl phthalate)	206>149	149>121
BBP-d4		210>153	153>125
DCHP	(dicyclohexyl phthalate)	249>149	249>93
DCHP-d4		253>153	253>97
DEHP	(bis(2-ethylhexyl) phthalate)	279>149	279>93
DEHP-d4		283>153	283>97
DnOP	(dioctyl phthalate)	279>149	279>93
DnOP-d4		283>153	283>93
DINP	(di-isononyl phthalate)	293>149	
DIDP	(di-isodecyl phthalate)	307>149	

Appendix II

(for information)

GC/MS chromatograms of a phthalate standard solution and deuterated internal standards.

Appendix III

(for information)

Validation of analysis of phthalates in wines

Executive Summary

The Institute for Reference Materials and Measurements (IRMM) organised in close

collaboration with the International Organisation of Vine and Wine (OIV) this collaborative study to validate *Compendium* method OIV-MA-AS323-10:2013 for the determination of ten phthalates in wine by gas chromatography - mass spectrometry (GC-MS).

The design of the method performance study complied with provisions given in ISO 5725-2 and those established by the OIV. The test samples consisted of red wine, white wine, and sweet wine presented as blind duplicates (see Table 1).

The wines were spiked at IRMM, bottled into ampoules, and dispatched to the participants of the validation study.

In addition to the test samples, participants received a deuterated phthalate solution, in order to be able to prepare the internal standard solutions.

The participants of the study were identified by the OIV following a pre-validation study for the method. They comprised laboratories from Europe, Asia, South America and Australia (see Table 2).

The evaluation of the reported results was performed according to ISO 5725-2 and ISO 5725-4, as well as the provisions established by the OIV. Relative standard deviations for reproducibility were mostly within the range of 9% to 71%.

Table 1

Sample	S001	S002	S003	S004	S005	S006
Nature	White wine		Red wine		Sweet wine	

Participants in the study

Table 2: Participants in the study

Analab Chile S.A.	Chile
Animal & Plant & Food Inspection Centre, Tianjin Exit- Entry Inspection and Quarantine Bureau	People's Republic of China
Bureau Interprofessionnel du Cognac	France
Central National de Verificare a Calitatii Productiei Alcoolice	Republic of Moldova
Chemisches und Veterinaeruntersuchungsamt Stuttgart	Germany

Escola Superior de Biotecnologia Universidade Católica Portuguesa	Portugal
Instituto Nacional de Vitivinicultura Departamento de Normas Analiticas Especiales	Argentina
Laboratorio Arbitral Agroalimentario	Spain
Laboratoire DUBERNET	France
Miguel Torres S.A.	Spain
SAILab	Spain
SCL Laboratoire de Bordeaux	France
SCL Laboratoire de Montpellier	France
The Australian Wine Research Institute	Australia

Evaluation of submitted results

The fitness-for-purpose of the calculated reproducibility standard deviation was evaluated. For this purpose, the calculated reproducibility relative standard deviation (RSD_R) was compared to the relative standard deviation derived from the modified Horwitz equation (RSD_{mH},) as proposed by Thompson (Thompson 2000). The latter provides a concentration dependant guidance level for reproducibility.

The agreement with the guidance level of precision was expressed as HORRAT values for reproducibility ($HORRAT_R$).

Evaluation of systematic effects

Laboratories reporting results that, for one or more analytes, exceeded the 1% threshold level of either the Mandel's h or Mandel's k tests were contacted by the organisers and requested to check their reported data and to confirm them if appropriate. Results were excluded from data evaluations if the laboratory did not confirm the correctness of the reported analytical results.

Evaluation of reported results by analyte

Based on the results of the separate analysis of each analyte and according to the reproducibility results, the method should be considered as either type II (DCHP BBP

DBP DIBP DEP) or type IV (DIDP DINP DNOP DEHP DMP).

Table 3: Dimethyl phthalate (DMP)**[1]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006
No. of laboratories that submitted compliant results		11	10	11	10	10	11
Mean	mg/l	0.020	0.073	0.018	0.031	0.053	0.027
Median	mg/l	0.020	0.060	0.018	0.030	0.056	0.028
Assigned value	mg/l	0.030	0.097	0.030	0.049	0.104	0.046
Rel. dev. assign. value		-33.3%	-38.1%	-40.0%	-38.8%	-46.2%	-39.1%
Repeatability s.d.	mg/l	0.003	0.007	0.002	0.006	0.011	0.003
Reproducibility s.d.	mg/l	0.006	0.041	0.007	0.011	0.022	0.009
Rel. repeatability s.d.		9.42%	7.33%	8.04%	13.00%	10.25%	7.09%
Rel. reproducibility s.d.		20.10%	42.40%	23.12%	22.54%	21.10%	19.07%
Modified Horwitz s.d. **		22.00%	22.00%	22.00%	22.00%	22.00%	22.00%
HORRATR		0.91	1.93	1.05	1.02	0.96	0.87
Limit of repeatability, r (2.77 X sr)	mg/l	0.008	0.020	0.007	0.018	0.030	0.009

Limit of reproducibility, R (2.77 X sR)	mg/l	0.017	0.114	0.019	0.031	0.061	0.024
Rel. limit of repeatability		26.09%	20.32%	22.28%	36.00%	28.38%	19.64%
Rel. limit of reproducibility		55.67%	117.45%	64.05%	62.44%	58.45%	52.84%
No. of laboratories after elimination of outliers		9	9	8	8	9	10
No. of measurement values without outliers		18	18	15	16	18	20

Table 4: Diethyl phthalate (DEP)**[2]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006
No. of laboratories that submitted compliant results		12	11	11	11	10	12
Mean	mg/l	0.048	0.065	0.030	0.039	0.021	0.059
Median	mg/l	0.044	0.076	0.029	0.041	0.023	0.061
Assigned value	mg/l	0.057	0.092	0.031	0.056	0.030	0.089
Rel. dev. assign. value		-22.8%	-17.4%	-6.5%	-26.8%	-23.3%	-31.5%
Repeatability s.d.	mg/l	0.006	0.010	0.005	0.004	0.003	0.002

Reproducibility s.d.	mg/l	0.026	0.026	0.015	0.017	0.008	0.019
Rel. repeatability s.d.		10.49%	11.32%	15.28%	7.00%	11.41%	2.53%
Rel. reproducibility s.d.		45.36%	28.49%	47.95%	29.71%	25.74%	20.98%
Modified Horwitz s.d. **		22.00%	22.00%	22.00%	22.00%	22.00%	22.00%
HORRATR		2.06	1.30	2.18	1.35	1.17	0.95
Limit of repeatability, r (2.77 X sr)	mg/l	0.017	0.029	0.013	0.011	0.009	0.006
Limit of reproducibility, R (2.77 X sR)	mg/l	0.072	0.073	0.041	0.046	0.021	0.052
Rel. limit of repeatability		29.05%	31.35%	42.32%	19.40%	31.60%	7.01%
Rel. limit of reproducibility		125.66%	78.91%	132.81%	82.29%	71.30%	58.12%
No. of laboratories after elimination of outliers		11	10	11	9	10	11
No. of measurement values without outliers		21	20	21	17	20	22

Table 5: Diisobutyl phthalate (DIBP)**[3]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006

No. of laboratories that submitted compliant results		11	10	11	10	10	11
Mean	mg/l	0.049	0.087	0.076	0.119	0.054	0.046
Median	mg/l	0.049	0.085	0.076	0.123	0.055	0.045
Assigned value	mg/l	0.035	0.076	0.058	0.107	0.061	0.045
Rel. dev. assign. value		40.0%	11.8%	31.0%	15.0%	-9.8%	0.0%
Repeatability s.d.	mg/l	0.003	0.006	0.007	0.009	0.002	0.004
Reproducibility s.d.	mg/l	0.011	0.019	0.014	0.023	0.012	0.013
Rel. repeatability s.d.		7.43%	7.71%	11.55%	8.81%	4.04%	9.54%
Rel. reproducibility s.d.		32.18%	25.23%	24.48%	21.95%	19.98%	28.37%
Modified Horwitz s.d. **		22.00%	22.00%	22.00%	22.00%	22.00%	22.00%
HORRATR		1.46	1.15	1.11	1.00	0.91	1.29
Limit of repeatability, r (2.77 X sr)	mg/l	0.007	0.016	0.019	0.026	0.007	0.012
Limit of reproducibility, R (2.77 X sR)	mg/l	0.031	0.053	0.039	0.065	0.034	0.035

Rel. limit of repeatability		20.58%	21.35%	31.98%	24.42%	11.19%	26.44%
Rel. limit of reproducibility		89.15%	69.88%	67.80%	60.81%	55.35%	78.58%
No. of laboratories after elimination of outliers		11	10	11	10	10	11
No. of measurement values without outliers		21	20	21	20	20	22

Table 6: Dibutyl phthalate (DBP)**[4]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006
No. of laboratories that submitted compliant results		12	11	12	11	11	12
Mean	mg/l	0.103	0.264	0.078	0.728	0.090	0.178
Median	mg/l	0.103	0.266	0.074	0.666	0.089	0.174
Assigned value	mg/l	0.107	0.281	0.057	1.039	0.032	0.153
Rel. dev. assign. value		-3.7%	-5.3%	29.8%	-35.9%		
Repeatability s.d.	mg/l	0.009	0.014	0.011	0.033	0.004	0.012
Reproducibility s.d.	mg/l	0.022	0.048	0.021	0.314	0.018	0.022
Rel. repeatability s.d.		8.24%	5.03%	19.11%	3.21%	13.79%	7.87%
Rel. reproducibility s.d.		20.73%	17.01%	36.78%	30.25%	57.05%	14.66%

Modified Horwitz s.d. **		22.00%	19.36%	22.00%	15.91%	22.00%	21.22%
HORRATR		0.94	0.88	1.67	1.90	2.59	0.69
Limit of repeatability, r (2.77 X sr)	mg/l	0.024	0.039	0.030	0.092	0.012	0.033
Limit of reproducibility, R (2.77 X sR)	mg/l	0.061	0.132	0.058	0.871	0.051	0.062
Rel. limit of repeatability		22.81%	13.92%	52.94%	8.89%	38.21%	21.80%
Rel. limit of reproducibility		57.43%	47.12%	101.88%	83.79%	158.03%	40.60%
No. of laboratories after elimination of outliers		12	11	12	10	11	11
No. of measurement values without outliers		23	22	23	20	22	22

Table 7: Benzyl butyl phthalate (BBP)**[5]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006
No. of laboratories that submitted compliant results		11	10	11	10	10	11
Mean	mg/l	0.049	0.026	0.033	0.074	0.075	0.050
Median	mg/l	0.050	0.027	0.034	0.075	0.078	0.051
Assigned value	mg/l	0.057	0.029	0.037	0.088	0.087	0.053
Rel. dev. assign. value		-12.3%	-6.9%	-8.1%	-14.8%	-10.3%	-3.8%

Repeatability s.d.	mg/l	0.002	0.001	0.003	0.004	0.003	0.003
Reproducibility s.d.	mg/l	0.008	0.004	0.005	0.011	0.015	0.007
Rel. repeatability s.d.		4.30%	4.96%	8.08%	5.10%	3.31%	4.78%
Rel. reproducibility s.d.		13.71%	13.82%	13.93%	12.72%	17.00%	14.00%
Modified Horwitz s.d. **		22.00%	22.00%	22.00%	22.00%	22.00%	22.00%
HORRATR		0.62	0.63	0.63	0.58	0.77	0.64
Limit of repeatability, r (2.77 X sr)	mg/l	0.007	0.004	0.008	0.012	0.008	0.007
Limit of reproducibility, R (2.77 X sR)	mg/l	0.022	0.011	0.014	0.031	0.041	0.021
Rel. limit of repeatability		11.90%	13.75%	22.38%	14.14%	9.16%	13.23%
Rel. limit of reproducibility		37.98%	38.27%	38.58%	35.23%	47.09%	38.77%
No. of laboratories after elimination of outliers		9	8	10	9	9	10
No. of measurement values without outliers		17	15	19	18	18	20

Table 8: Dicyclohexyl phthalate (DCHP)**[6]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006

No. of laboratories that submitted compliant results		9	8	9	8	8	9
Mean	mg/l	0.079	0.042	0.030	0.088	0.046	0.031
Median	mg/l	0.076	0.044	0.033	0.091	0.050	0.033
Assigned value	mg/l	0.084	0.048	0.038	0.105	0.057	0.036
Rel. dev. assign. value		-9.5%	-8.3%	-13.2%	-13.3%	-12.3%	-8.3%
Repeatability s.d.	mg/l	0.005	0.006	0.003	0.005	0.002	0.001
Reproducibility s.d.	mg/l	0.024	0.008	0.005	0.011	0.011	0.006
Rel. repeatability s.d.		5.60%	13.13%	6.75%	4.84%	3.25%	3.67%
Rel. reproducibility s.d.		28.46%	16.05%	12.93%	10.20%	18.83%	16.37%
Modified Horwitz s.d. **		22.00%	22.00%	22.00%	22.00%	22.00%	22.00%
HORRATR		1.29	0.73	0.59	0.46	0.86	0.74
Limit of repeatability, r (2.77 X sr)	mg/l	0.013	0.017	0.007	0.014	0.005	0.004
Limit of reproducibility, R (2.77 X sR)	mg/l	0.066	0.021	0.014	0.030	0.030	0.016
Rel. limit of repeatability		15.53%	36.37%	18.69%	13.40%	9.00%	10.18%
Rel. limit of reproducibility		78.83%	44.46%	35.82%	28.24%	52.15%	45.35%

No. of laboratories after elimination of outliers		9	7	8	7	7	8
No. of measurement values without outliers		18	14	15	14	14	16

Table 9: Bis (2-ethylhexyl) phthalate (DEHP)[7] – Results of data evaluation

		S001	S002	S003	S004	S005	S006
No. of laboratories that submitted compliant results		12	11	12	11	11	12
Mean	mg/l	0.101	0.028	0.602	0.150	0.741	1.032
Median	mg/l	0.099	0.026	0.654	0.180	0.709	1.115
Assigned value	mg/l	0.217	0.046	1.049	0.328	1.569	2.013
Rel. dev. assign. value		-54.4%	-43.5%	-37.7%	-45.1%	-54.8%	-44.6%
Repeatability s.d.	mg/l	0.017	0.005	0.206	0.016	0.122	0.266
Reproducibility s.d.	mg/l	0.019	0.011	0.238	0.063	0.465	0.563
Rel. repeatability s.d.		7.72%	11.54%	19.66%	4.82%	7.78%	13.20%
Rel. reproducibility s.d.		8.92%	24.15%	22.70%	19.11%	29.61%	27.96%
Modified Horwitz s.d. **		20.13%	22.00%	15.88%	18.92%	14.95%	14.40%
HORRATR		0.44	1.10	1.43	1.01	1.98	1.94
Limit of repeatability, r (2.77 X sr)	mg/l	0.046	0.015	0.571	0.044	0.338	0.736
Limit of reproducibility, R (2.77 X sR)	mg/l	0.054	0.031	0.660	0.174	1.287	1.559

Rel. limit of repeatability		21.39%	31.98%	54.45%	13.36%	21.54%	36.55%
Rel. limit of reproducibility		24.70%	66.91%	62.87%	52.93%	82.03%	77.46%
No. of laboratories after elimination of outliers		10	10	12	9	11	12
No. of measurement values without outliers		20	20	23	18	22	24

Table 10: Di-n-octyl phthalate (DNOP)**[8]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006
No. of laboratories that submitted compliant results		11	10	11	10	9	10
Mean	mg/l	0.031	0.015	0.051	0.073	0.016	0.026
Median	mg/l	0.035	0.015	0.049	0.061	0.019	0.028
Assigned value	mg/l	0.086	0.031	0.059	0.114	0.036	0.054
Rel. dev. assign. value		-59.3%	-51.6%	-16.9%	-46.5%	-47.2%	-48.1%
Repeatability s.d.	mg/l	0.007	0.003	0.021	0.005	0.004	0.005
Reproducibility s.d.	mg/l	0.010	0.003	0.023	0.038	0.008	0.011
Rel. repeatability s.d.		7.84%	9.25%	36.33%	4.51%	11.18%	9.23%
Rel. reproducibility s.d.		11.50%	9.33%	38.90%	33.40%	23.32%	20.10%

Modified Horwitz s.d. **		22.00%	22.00%	22.00%	22.00%	22.00%	22.00%
HORRATR		0.52	0.42	1.77	1.52	1.06	0.91
Limit of repeatability, r (2.77 X sr)	mg/l	0.019	0.008	0.059	0.014	0.011	0.014
Limit of reproducibility, R (2.77 X sR)	mg/l	0.027	0.008	0.064	0.105	0.023	0.030
Rel. limit of repeatability		21.73%	25.61%	100.62%	12.50%	30.97%	25.56%
Rel. limit of reproducibility		31.85%	25.85%	107.76%	92.52%	64.60%	55.66%
No. of laboratories after elimination of outliers		9	8	10	9	7	8
No. of measurement values without outliers		18	15	18	16	14	16

Table 11: Diisononyl phthalate (DINP)**[9]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006
No. of laboratories that submitted compliant results		9	8	10	8	8	9
Mean	mg/l	0.027	0.108	1.820	0.059	0.115	0.064
Median	mg/l	0.028	0.116	1.497	0.058	0.136	0.051
Assigned value	mg/l	0.054	0.242	3.134	0.104	0.271	0.057

Rel. dev. assign. value		-48.1%	-52.1%	-52.2%	-44.2%	-49.8%	-10.5%
Repeatability s.d.	mg/l	0.004	0.019	0.520	0.005	0.010	0.003
Reproducibility s.d.	mg/l	0.006	0.027	1.067	0.019	0.072	0.040
Rel. repeatability s.d.		8.14%	7.84%	16.60%	5.17%	3.83%	5.51%
Rel. reproducibility s.d.		10.27%	11.18%	34.06%	18.41%	26.60%	70.59%
Modified Horwitz s.d. **		20.00%	20.00%	20.00%	20.00%	20.00%	20.00%
HORRATR		0.51	0.56	1.70	0.92	1.33	3.53
Limit of repeatability, r (2.77 X sr)	mg/l	0.012	0.053	1.441	0.015	0.029	0.009
Limit of reproducibility, R (2.77 X sR)	mg/l	0.015	0.075	2.957	0.053	0.200	0.111
Rel. limit of repeatability		22.55%	21.71%	45.99%	14.32%	10.61%	15.27%
Rel. limit of reproducibility		28.44%	30.98%	94.35%	50.99%	73.69%	195.53%
No. of laboratories after elimination of outliers		5	6	9	7	6	6
No. of measurement values without outliers		10	11	17	13	12	12

Table 12: Diisodecyl phthalate (DIDP)**[10]** – Results of data evaluation

		S001	S002	S003	S004	S005	S006

No. of laboratories that submitted compliant results		8	7	8	7	7	8
Mean	mg/l	0.096	0.103	0.677	0.152	0.186	1.828
Median	mg/l	0.102	0.107	0.540	0.152	0.181	1.660
Assigned value	mg/l	0.275	0.186	0.200	0.281	0.427	3.070
Rel. dev. assign. value		-62.9%	-42.5%	170.0%	-45.9%	-57.6%	-45.9%
Repeatability s.d.	mg/l	0.009	0.018	0.477	0.048	0.027	0.202
Reproducibility s.d.	mg/l	0.025	0.018	0.505	0.058	0.109	1.676
Rel. repeatability s.d.		3.42%	9.61%	238.49%	17.11%	6.27%	6.57%
Rel. reproducibility s.d.		9.11%	9.61%	252.34%	20.51%	25.43%	54.59%
Modified Horwitz s.d. **		20.00%	20.00%	20.38%	20.00%	20.00%	20.00%
HORRATR		0.46	0.48	12.38	1.03	1.27	2.73
Limit of repeatability, r (2.77 X sr)	mg/l	0.026	0.050	1.321	0.133	0.074	0.559
Limit of reproducibility, R (2.77 X sR)	mg/l	0.069	0.050	1.398	0.160	0.301	4.642
Rel. limit of repeatability		9.46%	26.62%	660.61%	47.40%	17.37%	18.21%
Rel. limit of reproducibility		25.25%	26.62%	698.98%	56.82%	70.44%	151.21%

No. of laboratories after elimination of outliers		7	5	7	7	7	7
No. of measurement values without outliers		14	10	13	14	14	14

References

- Report on the Method Performance Study of a Method to Determine Phthalates in Wine Determination of Ten Phthalates in Wine by Gas Chromatography Mass Spectrometry (GC-MS), Wenzl Thomas, Karasek Lubomir, Giri Anupam. Publications Office of the European Union 2015 doi :10.2787/666948 (online) https://publications.europa.eu/en/publication-detail/-/publication/b3ebef67-f1db-4fb2-97ce-bfc301c8ce68/language-en

[1] Type IV method
[2] Type II method
[3] Type II method
[4] Type II method
[5] Type II method
[6] Type II method
[7] Type IV method
[8] Type IV method
[9] Type IV method
[10] Type IV method