RESOLUTION OENO 8/2000

MEASUREMENT OF ALCOHOLIC STRENGTH BY VOLUME OF WINE BY ELECTRONIC DENSIMETRY USING FREQUENCY OSCILLATOR

THE GENERAL ASSEMBLY,

HAVING CONSIDERED Article 5 of the October 13, 1954 international convention for the unification of means of analysis and appreciation of wine,

WITH THE PROPOSAL of the Sub-commission of the means of analysis and appreciation of wine

DECIDES to introduce in Annex A of the Compendium of International Methods of Analysis the following "**Measurement of alcoholic strength by volume of wine by electronic densimetry using a frequency oscillator**".

1. MEASUREMENT METHOD

1.1. Strength and introduction

The alcoholic strength by volume of wine must be measured before being commercialised mainly in order to conform to labelling rules.

The alcoholic strength by volume is equal to the number of litres of ethanol contained in 100 litres of wine; these volumes are both measured at 20°C. The symbol is “ % vol.”.

1.2. Precautionary safety measures

Respect the safety guidelines for the usage of distillation apparatuses, the manipulation of hydro-alcoholic and cleaning solutions.

1.3. Object and field of application

The method of measurement described is electronic densimetry using a frequency oscillator.

In reference to the provision of the rules in the existing law, the trial temperature is stopped at 20 °C.

1.4. Principle and definitions

The principle of the method consists firstly of distilling the wine volume by volume. The distillation procedure is described in the Compendium. This distillation enables

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the elimination of non-volatile substances. The ethanol counter parts in addition to ethanol and the ethanol counter parts involved in esters are included in the alcoholic strength since they are present in the distillate

The distillate density of the distillate is measured. The density of a liquid at a given temperature is equal to the ratio of its density to its volume.

$p = m / V$, for a wine, it is expressed as g/ml

For hydro-alcoholic solutions such as distillates, given the known temperature, the graphs correspond to the alcoholic strength by volume (OIV, 1990). This alcoholic strength corresponds to that of wine (distillation of volume to volume).

In the present method the distillate density is measured by electronic densimetry using a frequency oscillator. The principle consists of measuring the period of oscillation of a tube containing the sample undergoing an electromagnetic stimulation. The density is thus calculated and is linked to the period of oscillation by the following formula:

$$p = T^2 \times \left(\frac{C}{4\pi^2 V}\right) - \left(\frac{M}{V}\right) \quad (1)$$

- p = density of sample
- T = period of induced vibration
- M = mass of empty tube
- C = spring constant
- V = volume of vibrating sample

This relation is in the form of, $p = A\,T^2 - B$ (2), There is a linear relationship between density and the period squared. The A and B constants specific to each oscillator are estimated by measuring the period of fluids of the known density.

1.5. Reagents and products

1.5.1. Reference fluids

Two reference fluids are used to adjust the densimetry. The densities of reference fluids must encompass the densities of the distillates to be measured. A spread between the densities between reference fluids above 0.01000 g/ml is recommended.

The density must be known with an uncertainty under +/- 0.00005 g/ml, for a temperature of 20.,00 +/- 0.05 °C.

The measuring of alcoholic strength by volume of wine by electronic densimetry of reference fluids:

- dry air (unpolluted),
- double distilled water or of an equivalent analytical purity,
- hydro alcoholic solution of density determined by pycometry (reference method),
- solutions connected to national standards of viscosity under 2 mm^2/s.

1.5.2. Cleaning and drying products

- detergents, acids ...,
- organic solvents: ethanol 96% Vol., pure acetone ...

1.6. Apparatus

1.6.1. Electronic densimetry by frequency oscillator

Electronic densimetry contains the following elements:

- a measuring cell containing a measurement tube and a temperature controlled enclosure,
- a system for setting up an oscillation tube and measurement of the period of oscillation,
- a timer,
- a digital display and possibly a calculator.

The densimetry on a perfectly stable support isolated from all vibrations.

1.6.2. Temperature control of measuring cell

The measurement tube is located in the temperature-controlled enclosure. Temperature stability must be better than +/- 0.02 °C.

It is necessary to control the temperature of the measuring cell when the densimetry makes this possible, because this strongly influences .the indication results. Density of this hydro alcoholic solution with an alcoholic strength by volume of 10% Vol., and is at 0.98471 g/ml at 20°C and at 0.98447 g/ml at 21°C or a spread of 0.00024 g/ml.

The trial temperature is stopped at 20°C. The temperature is taken at the cell level and done with a resolution thermometer 0.01°C and connected to national standards. This must enable a temperature measurement with an uncertainty of under +/- 0.07°C.

1.6.3. Calibration of the apparatus

The apparatus must be calibrated before using for the first time, then every six months or is the verification is not satisfactory. The objective is to use two reference fluids to calculate the constants A and B (cf. (2)). To carry out the calibration refer to the user's manual of the apparatus. In principle, this calibration is carried out with dry air (take into account the atmospheric pressure) and very pure water (double distilled and/or very high micro filtered resistance, for example > 18 M □

1.6.4. Calibration verification

In order to verify the calibration we measure the density of the reference fluids.

- Every day, a density check of the air is carried out. A difference between the theoretical density and the observed density of more than 0.00008 g/ml may indicate that the tube is clogged. In that case, it must be cleaned. After cleaning, verify the air density again. If the verification is not conclusive adjust the apparatus.

- Check the density of water, if the difference between the theoretical density and the density observed is greater than 0.00008 g/ml, adjust the apparatus.
- If the verification of cell temperature is difficult, it is possible to directly check hydro alcoholic density of the alcoholic strength by volume compared to the distillates analysed.

1.6.5. Check

When the difference between the theoretical density of the reference solution (known with an uncertainty of +/- 0.00005 g/ml) and the measurement is above 0.00008 g/ml the temperature of the cell must be taken.

1.7. Sampling and preparation of samples

(Cf. Compendium if International methods of wine and musts 1990, page 59, Obtaining distillate)

1.8. Operating procedure

After obtaining a distillate, (OIV, 1990) we measure the density or the alcoholic strength by volume by densimetry.

The operator must ensure the stability and the temperature of the measuring cell. The distillate in the densimetry cell must not contain air bubble and must be homogeneous. If there is an available lighting system, turn off quickly after checking because the heat generated by the lamp can influence the measuring temperature.

If the apparatus only provides the period, density can be calculated by the A and B constants (cf. A.4 c). If the apparatus does not provide the alcoholic strength by volume directly, we can obtain the alcoholic strength by volume using the (OIV, 1990) tables if we know the density.

1.9. Expression of results

The alcoholic strength by volume is obtained from the distillate. This is expressed as “ % vol. ”.

If the temperature conditions are not respected, a correction must be made to express the temperature at 20°C. The result is quoted to two decimal places

1.10. Comments

The volume introduced into the cell must be sufficient enough to avoid possible contamination caused from the previous sample. It is thus necessary to carry out two testing. If this does not provide results included in the repeatability limits, a third testing may be necessary. In general, results from the last two testing are homogeneous and we then eliminate the first factor.

1.11. Reliability

For alcoholic strength by volume samples between 4 to 18% Vol.

Repeatability (r) = 0.067 (% vol.),

Reproducibility (R) = 0.0454 + 0.0105 x alcoholic strength by volume.

2. INTERLABORATORY Tests. RELIABILITY AND ACCURACY ON ADDITIONS

2.1. Samples

The samples used for this joint study are described in Table 1.

Table 1: Samples for joint study

Num	Nature	Approx alcoholic strength by volume (% vol.)
C0	Cider (filtered through membrane to remove CO_2)	5
V0	Filtered wine	10
V1	Filtered wine then doped	11
V2	Filtered wine then doped	12
V3	Filtered wine then doped	13
P0	Liqueur wine	16

All samples are homogenised before filling the bottles to be sent to the participants. For wine, 40 litres of wine are homogenised before sending and carrying out the additions

For the additions, pour absolute ethanol into a 5 litre volumetric flask and then fill up to the line with filtered wine. This operation is repeated two times. The volumes of ethanol are respectively 50, 100 and 150 ml for the V1, V2 and V3 samples.

2.2. Participating laboratories

The participating laboratories in the joint study are outlined in Table 2.

Laboratoire	C.P.	Ville	Personne à contacter
ALKO Group LTD	FIN-00101	Helsinki	Monsieur Lehtonen
Bénédictine	76400	Fécamp	Madame Pillon
Casanis	18881	Gemenos	Madame Cozon
CIVC	51200	Epernay	Monsieur Tusseau
Cointreau	49181	St Barthélémy d'Anjou	Madame Guerin
Courvoisier	16200	Jarnac	Monsieur Lavergne
Hennessy	16100	Cognac	Monsieur Calvo
IDAC	44120	Vertou	Madame Mars
Laboratoire Gendrot	33000	Bordeaux	Madame Gubbiotti
Martell	16100	Cognac	Monsieur Barboteau
Ricard	94320	Thiais	Monsieur Boulanger
SOEC Martin Vialatte	51319	Epernay	Madame Bertemes

In order not to introduce a methodological angle, the *Station Viticole du Bureau National Interprofessionnel du Cognac,* the joint study organiser, will not be taken into account.

2.3. Analyses

The C0 and P0 products are distilled two times, the V0, V1, V2 and V3 products three times. Three alcoholic strength by volume tests were done for each distillate. The results were carried over to the results table.

2.4. Results

The second testing (out of the three carried out) is kept of the accuracy study (Table 3).

Table 3: Results (second testing per distillate) (% vol.)

Laboratory	C0	V0	V1	V2	V3	P0
	6,020	9,500	10,390	11,290	12,100	17,080
1	5,970	9,470	10,380	11,260	12,150	17,080

		9,450	10,340	11,260	12,150	
2	6,040	9,500	10,990	11,270	12,210	17,050
	6,040	9,500	10,390	11,280	12,210	17,050
		9,510	10,400	11,290	12,200	
3	5,960	9,460	10,350	11,280	12,170	17,190
	5,910	9,460	10,360	11,280	12,150	17,200
		9,450	10,340	11,260	12,170	
4	6,020	9,470	10,310	11,250	12,160	16,940
	6,020	9,450	10,350	11,250	12,120	17,070
		9,450	10,330	11,210	12,130	
5	5,950	9,350	10,250	11,300	12,050	17,000
	5,950	9,430	10,250	11,300	12,050	17,000
		9,430	10,250	11,300	12,050	
6	6,016	9,513	10,370	11,275	12,222	17,120
	6,031	9,513	10,336	11,266	12,222	17,194
		9,505	10,386	11,275	12,220	
7	5,730	9,350	10,230	11,440	12,080	17,010
	5,730	9,430	10,220	11,090	12,030	16,920
		9,460	10,220	11,080	11,930	
	5,990	9,400	10,340	11,160	12,110	17,080

8	6,000	9,440	10,320	11,150	12,090	17,110
		9,440	10,360	11,210	12,090	
9	6,031	9,508	10,428	11,289	12,180	17,089
	6,019	9,478	10,406	11,293	12,215	17,084
		9,509	10,411	11,297	12,215	
10	6,030	9,500	10,380	11,250	12,150	17,130
	6,020	9,510	10,380	11,250	12,150	17,100
		9,510	10,380	11,250	12,160	
11	6,020	9,480	10,400	11,260	12,150	17,040
	6,000	9,470	10,390	11,260	12,140	17,000
		9,490	10,370	11,240	12,160	

2.5. Repeatability and reproducibility calculations

Repeatability and reproducibility calculations are carried out in compliance with the standard NF X 06-041, September 1983, ISO 5725.Table 4 presents the standard deviation per cell (laboratory x sample).

Table 4: Dispersion table (standard deviation in % vol.)

Laboratory	C0	V0	V1	V2	V3	P0
1	0,0354	0,0252	0,0265	0,0173	0,0289	0,0000
2	0,0000	0,0058	0,3436	0,0100	0,0058	0,0000
3	0,0354	0,0058	0,0100	0,0115	0,0115	0,0071
4	0,0000	0,0115	0,0200	0,0231	0,0208	0,0919

5	0,0000	0,0462	0,0000	0,0000	0,0000	0,0000
6	0,0106	0,0046	0,0255	0,0052	0,0012	0,0523
7	0,0000	0,0569	0,0058	0,2050	0,0764	0,0636
8	0,0071	0,0231	0,0200	0,0321	0,0115	0,0212
9	0,0085	0,0176	0,0115	0,0040	0,0202	0,0035
10	0,0071	0,0058	0,0000	0,0000	0,0058	0,0212
11	0,0141	0,0100	0,0153	0,0115	0,0100	0,0283

Three cells presented strong dispersions (Probability with Cochran test under 1%). These cells are represented in grey (Table 4).

For laboratory 7 and the V3 product, the standard deviation of 0.0764 is maintained despite the Cochran test because it is on the same high level as that observed at the same laboratory on the V0 product.

An examination of figures for each distillate leads us to eliminate (Table 3):

- laboratory 2, product V1, value 10.990,
- laboratory 7, product V2, value 11.440.

After eliminating these two values, the cell averages are calculated (laboratory x sample) (Table 5).

Table 5: Table of averages (averages in % vol.)

Laboratory	C0	V0	V1	V2	V3	P0
1	5,9950	9,4733	10,3700	11,2700	12,1333	17,0800
2	6,0400	9,5033	10,3950	11,2800	12,2067	17,0500
3	5,9350	9,4567	10,3500	11,2733	12,1633	17,1950

4	6,0200	9,4567	10,3300	11,2367	12,1367	17,0050
5	5,9500	9,4033	10,2500	11,3000	12,0500	17,0000
6	6,0235	9,5103	10,3640	11,2720	12,2213	17,1570
7	5,7300	9,4133	10,2233	11,0850	12,0133	16,9650
8	5,9950	9,4267	10,3400	11,1733	12,0967	17,0950
9	6,0250	9,4983	10,4150	11,2930	12,2033	17,0865
10	6,0250	9,5067	10,3800	11,2500	12,1533	17,1150
11	6,0100	9,4800	10,3867	11,2533	12,1500	17,0200

The figures given by laboratory 7 are generally low (Table 5). In the case of cider the average for this laboratory is very far from the figures of the other laboratories (Associated probability to the Dixon test under 1 %). The results of this laboratory for this product are eliminated.

Table 6 presents the calculated repeatability and reproducibility.

Table 6: Calculation of repeatability and reproducibility

Sample	P	n	TAV	S2r	S2L	r	R
C0	10	20	6,002	0,000298	0,001033	0,049	0,103
V0	11	33	9,466	0,000654	0,001255	0,072	0,124
V1	11	32	10,344	0,000255	0,003485	0,045	0,173
V2	11	32	11,249	0,000219	0,003113	0,042	0,163
V3	11	33	12,139	0,000722	0,003955	0,076	0,194
P0	11	22	17,070	0,001545	0,004154	0,111	0,214

Key:
p : number of laboratories retained
n : number of values retained
TAV: average alcoholic strength by volume (% vol.)
S2r: repeatability variation (% vol.)2
S2L : interlaboratory variation (% vol.)2
r : repeatability (% vol.)
R : reproducibility (% vol.)

Reproducibility increases with the samples' alcoholic strength by volume (Figure 1). The increase in repeatability according to alcoholic strength by volume is less noticeable and global repeatability is calculated according to the average repeatability variation. As such, for the alcoholic strength by volume samples between 4 and 18% vol.,

Repeatability (r) = 0.067 (% vol.),

Reproducibility (R) = 0.0454 + 0.0105 x alcoholic strength by volume.

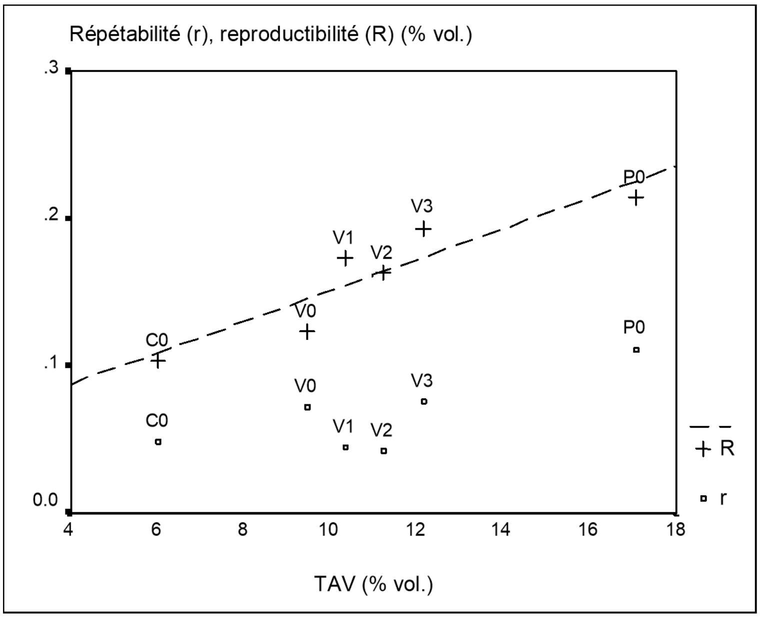

Figure 1: Repeatability and reproducibility according to alcoholic strength by volume

2.6. Accuracy on additions carried out on wine

The regression line of alcoholic strength after the addition according to the volume of ethanol supplied, for a volume of 0 ml, an estimation of the initial alcoholic strength of product (Figure 2). This regression is carried out with average values for each laboratory (Table 5).

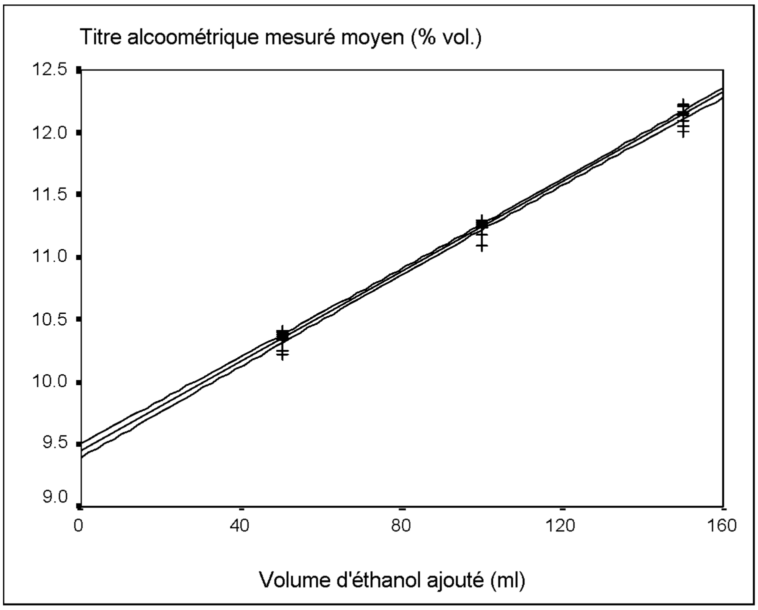

Figure: Regression of measures alcoholic strength by volume of added ethanol

Measurements carried out on initial products are not included in this estimation. This estimation is made up of the average of measurements taken on this product before additions; the intervals of relative confidence on these two estimations are calculated (Table 7).

Table 7: Additions on products

BI	Average measurements	BS	BI	estimation with measurements on products + additions	BS
9,440	9,466	9,492	9,392	9,450	9,508

Key:

BI : lower bound of confidence interval at 95%

BS : upper bound of confidence interval at 95%

The two confidence intervals cover a large overlapping spreading centre. Thanks to the measurements on doped samples, the alcoholic strength by volume of the initial product can be found.

2.7. Conclusion of interlaboratory trials

The repeatability and the reproducibilty indications by interlaboratory trials provide the following equations, for alcoholic strength by volume products between 4 to 18% vol.:

Repeatability (r) = 0.067 (% vol.),

Reproducibility (R) = 0.454 + 0.0105 x alcoholic strength by volume (% vol.).

The Horwitz indicators, Hor and HoR are weak (Table 8). These indicators provide good details of the method compared to the level of analyte measured.

Table 8: Table summary of method reliability

Samples	C0	V0	V1	V2	V3	P0
n	20	33	32	32	33	22
p	10	11	11	11	11	11
Alcoholic strength by volume	6,0019	9,4662	10,3443	11,2492	12,1389	17,0699
r	0,0489	0,0724	0,0452	0,0419	0,0760	0,1113
sr	0,0173	0,0256	0,0160	0,0148	0,0269	0,0393
RSDr	0,2878	0,2702	0,1543	0,1316	0,2214	0,2303
RSDrH	2,0159	1,8822	1,8573	1,8340	1,8131	1,7224
Hor	0,1428	0,1436	0,0831	0,0718	0,1221	0,1337
R	0,1033	0,1237	0,1731	0,1634	0,1935	0,2136
sR	0,0365	0,0437	0,0612	0,0577	0,0684	0,0755
RSDR	0,6080	0,4616	0,5912	0,5131	0,5634	0,4423

RSDRH	3,0543	2,8519	2,8141	2,7788	2,7471	2,6097
HoR	0,1991	0,1619	0,2101	0,1847	0,2051	0,1695

Key:
n : *number of values retained*
p : *number of laboratories retained*
Alcoholic strength by volume: average rate (% vol.)
r : *repeatability (% vol.)*
sr : Standard deviation of repeatability (% vol.)
RSDr : *Repeatability coefficient of variation (sr x 100 / TAV) (%)*
RSDrH : *Horwitz repeatability coefficient of variation (.0.66 x RSDRH) (%)*
Hor : *Horrat repeatability value (RSDr/RSDrH)*
R : *Reproducibility (% vol.)*
sR: Reproducibility standard deviation (% vol.)
RSDR : *Reproducibility coefficient of variation (sR x 100 / TAV) (%)*
RSDRH: Horwitz reproducibility coefficient of variation (2(1-0,5log(TAV))) (%)
HoR:Horrat reproducibility value (RSDR/RSDRH)

Interlaboratory trials’ measurements carried out on wine with additions enable us to find the value obtained before the addition. We respectively find 9.45 and 9.47% vol.

Bibliography

1. OIV, 1990. Recueil des méthodes internationales d’analyse des vins et des moûts, (Compendium of international methods of analysis of wine and musts) Office International de la Vigne et du Vin ; Paris.