HYPERLAB METHODS

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Blown glass apparatus and supplies for chemical laboratories
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The chemical analysis of wine aims at furthering and improving knowledge on the composition of the grapes, the must and the wine, studying the evolutionary kinetics during the transformation process in order to suggest to winemakers the most appropriate techniques for ensuring that the innate quality of the raw materials may best develop in the quality of the final product.

Preliminary Instructions

In some reaction kits it is possible to find the BLANK reagent; this product is almost never called for in the performing of the Hyperlab methods applied to robotic instrumentation. BLANK reagent is found in certain reaction kits because they can also be used with manual or automatic flow cell instruments where the determination of the sample blank must necessarily be carried out separate from that of the reagent blank.

Once the daily analysis cycle has been completed, the bottles containing the reagents must be capped and placed in a refrigerator (4° to 8°C) together with the reagent holder. Leaving the bottles uncapped on the cooling plate in operation brings about a greater adsorption of oxygen by the reagents, which changes their titre, and consequently may compromise the results of subsequent analyses.

For correct use of Hyperlab, the water supply tank must be filled with double-distilled or distilled water, adding 1 ml of *System Solution* for each liter of water used. For example, when using 15 l of water, add 15 ml of system solution. The washing tank, provided exclusively for Hyperlab Plus, must instead be filled with a 0.1 N sodium hydroxide (NaOH) solution using double-distilled or distilled water. For example, to make 2 l of this solution, weigh 4 g of pure NaOH (drops or flakes) and dissolve it in 2 l of water.

For Hyperlab Plus and Basic it is recommended that the cuvettes cleaning procedure be carried out once a week from the *Maintenance Procedures* menu using the alkaline solution in position 1 (1 part bleach and 3 parts water) and the acid solution in position 2 (hydrochloric acid 0.1 N). When performing the methods that are most “dirtying,” such as polyphenols, catechins, sulfur dioxide, chlorides and calcium, the cleaning procedure should be carried out once per day. For Hyperlab Smart instead it is recommended that the sampling needle be washed monthly.

The kit components are stable until the expiration date indicated on the package. They must be stored away from light and oxygen at 4° to 8°C (in the refrigerator).

Whenever possible, it is recommended that analyses be performed with kits and Auto methods that have a ratio of 4:1 between reagent 1 and reagent 2 instead of 40:1 as for traditional methods, and there is no blank reagent. These methods are more efficient and less expensive as regards managing reagents.

CONTENTS

The Hyperlab methods for the Plus, Basic and Smart models and the characteristics of the reagent kits for each individual analysis method are described below according to the following scheme:

METHOD CODE – METHOD NAME – REAGENT KIT CODE NO. – REAGENT KIT VOLUME
The description of each individual method includes an introductory part that gives the following information about the analyte: origin, oenological function, expected concentration and legal terms (if applicable). This is followed by a table regarding the use of the reagents found in the reaction kit and instructions for preparing the reagents when necessary.
The reaction principles and the characteristics of the method are then indicated.
Lastly, if there is the reference *Notes*, recommendations and tips are provided for optimizing the analysis.

ACETAU – ACETIC ACID AUTO – SQPE068205 – 125 ml

Acetic acid is an indicator of the health of the grapes and the quality of the wines. Its concentration generally must not exceed 0.6 g/l for red wines and 0.4 g/l for white wines, after which it is possible to perceive acetic notes nasally and/or retronasally. The maximum limits established by Italian law are: 1.08 g/l for white and rosé wines and 1.2 g/l for red wines.
Acetic acid is virtually absent in must, especially in the case of healthy grapes, and its content increases as a result of metabolic activity by microorganisms, such as alcoholic and malolactic fermentation. Values that are worrying, i.e. greater than or equal to 0.6 g/l, suggest an important microbiological alteration in progress or at least excessive aeration, resulting in oxidation of the product.
As for the unwelcome hint of vinegar, rather than the acid itself, the very volatile molecule involved is that of ethyl acetate, the ester that is formed chemically or enzymatically between the acetic acid and ethanol.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
ACET AUTO R1A (ACA1A)	1 x 50	large
ACET AUTO R1B (ACA1B)	1 x 50	large
ACET AUTO R2 (ACAR2)	1 x 25	small

Method principle and characteristics

Acetyl-CoA-synthetase converts acetic acid into acetyl-CoA, which reacts with oxaloacetate in the presence of citrate synthase to form citrate. The oxaloacetate necessary for the reaction is produced through the activity of the enzyme malate dehydrogenase on malate and on NAD, therefore NADH is generated. The increase in absorbance due to the formation of NADH, measured at 340 nm, is proportional to the amount of acetic acid in the sample.
Linearity with total V / sample V = 75: up to 1.2 g/l
Precision: CV% <1.1 in white wine; CV% <1.5 in red wine
Calibration and control: multiparameter standard SQPE053234 acetic acid 1g/l; multilevel standard SQPE081638 acetic acid 1 g/l (level 1), 0.8 g/l (level 2), 0.6 g/l (level 3), 0.4 g/l (level 4), 0.2 g/l (level 5).

ACETIC – ACETIC ACID – SQPE059575 – 5 x 20 ml

Acetic acid is an indicator of the health of the grapes and the quality of the wines. Its concentration generally must not exceed 0.6 g/l for red wines and 0.4 g/l for white wines, after which it is possible to perceive acetic notes nasally and/or retronasally. The maximum limits established by Italian law are: 1.08 g/l for white and rosé wines and 1.2 g/l for red wines.
Acetic acid is virtually absent in must, especially in the case of healthy grapes, and its content increases as a result of metabolic activity by microorganisms, such as alcoholic and malolactic fermentation. Values that are worrying, i.e. greater than or equal to 0.6 g/l, suggest an important microbiological alteration in progress or at least excessive aeration, resulting in oxidation of the product.
As for the unwelcome hint of vinegar, rather than the acid itself, the very volatile molecule involved is that of ethyl acetate, the ester that is formed chemically or enzymatically between the acetic acid and ethanol.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
ACET R1A (ACE1A)	5 x 10	small
ACET R1B (ACE1B)	5 x 10	small
ACET R2 (ACER2)	1 x 2.5	cup
ACET BL	2 x 50	do not use

Method principle and characteristics

Acetyl-CoA-synthetase converts acetic acid into acetyl-CoA, which reacts with oxaloacetate in the presence of citrate synthase to form citrate. The oxaloacetate necessary for the reaction is produced through the activity of the enzyme malate dehydrogenase on malate and on NAD, therefore NADH is generated. The increase in absorbance due to the formation of NADH, measured at 340 nm, is proportional to the amount of acetic acid present in the sample.
Linearity with total V / sample V = 75: up to 1.2 g/l
Precision: CV% <1.5 in white wine; CV% <175 in red wine
Calibration and control: multiparameter standard SQPE053234 acetic acid 1g/l; multilevel standard SQPE081638 acetic acid 1 g/l (level 1), 0.8 g/l (level 2), 0.6 g/l (level 3), 0.4 g/l (level 4), 0.2 g/l (level 5).

ALDEID – ACETALDEHYDE - SQPE059576 – 5 x 20 ml

Acetaldehyde is mainly produced by yeasts during alcoholic fermentation and is the precursor of ethyl alcohol. It can also accumulate from the metabolic activity of other microorganisms, and above all from the chemical oxidation of ethanol during the storage of wines (especially in wooden barrels), such that it is considered to be a marker of the state of oxidation of wines. Acetaldehyde, or ethanal, characterizes the color of wines, as it participates in the formation of the "ethyl bridge" (chemical bond) between tannins and anthocyanins and tends to bond with sulfur dioxide, causing a decrease in the level of free sulfur dioxide. Thus its determination is functional to the adding of sulfur dioxide (64 mg of SO2 bonds with 44 mg of acetaldehyde), which prevents the oxidation of ethanol, first to ethanal and then to acetic acid. The concentration of acetaldehyde in wine varies from 10 to 100 mg/l; this substance is characterized by a marked oxidized green apple aroma.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
ALD R1A (ALD1A)	5 x 20	small
ALD R1B (ALD1B)	1 x 2.5	cup
ALD R2 (ALDR2)	1 x 2.5	cup
ALD BL	2 x 50	do not use

Homogenize the ALD R1B suspension by shaking manually before use.

Method principle and characteristics

Acetaldehyde is quantitatively oxidized by NAD to acetic acid in the presence of the enzyme aldehyde dehydrogenase (ALDH).
The amount of NAD reduced to NADH during the reaction, measured at 340 nm, is directly proportional to the amount of acetaldehyde present in the sample.
Linearity with total V / sample V = 20: up to 150 mg/l
Precision: CV% < 1.5 on decolorized wine
Calibration and control: standard SQPE053229 acetaldehyde 100 mg/l.

Notes: With the repetition of the sample/standard over time, it is advisable to renew the sample, given the high volatility of the acetaldehyde. In addition, to improve the accuracy of the data, red wines can be decolorized, especially if the total polyphenols are over 2500 mg/l, using activated carbon or PVPP 1% weight/volume.

ANTOCI – ANTHOCYANINS - SQPE054971 – 40 x 50 ml

Anthocyanidins are the red pigments of the plant kingdom; they accumulate in the exocarp of the grape, and after maceration they are found in wine in a variable concentration of up to 600-800 mg/l in wines obtained from varieties rich in anthocyanins. The determining of anthocyanidins is important for the characterization of both wine color and flavor, as they play a role in polymerization along with tannins, causing a decrease in astringency. They also tend to react with SO2 (decolorization) and are influenced by temperature, pH and oxidation conditions during aging.
Anthocyanins show maximum absorbance at 520 nm, and, in the absence of a reference standard, the calculation factor used in the method was derived from experimental correlation tests with HPLC analytical methods.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
ANTO R1 (ANTOC)	4 X 50	large

Method principle and characteristics

Anthocyanins are ionized in an acidic environment by a controlled ionic strength buffer, which also contains solubilizers for eliminating any turbid protein formations, and measured at 520 nm. The anthocyanin fraction involved in polymerization with other polyphenolic substances is not detected.
Linearity with total V / sample V = 10: up to 600 mg/l
Precision: CV% < 1.5 in rosé and red wine
Result unit of measurement: mg/l

Notes: Must that is excessively turbid needs to be centrifuged or filtered before the analysis.

RAN – READILY ASSIMILABLE NITROGEN

This is a compound method and is derived from the sum of the concentration of amino nitrogen and the concentration of ammoniacal nitrogen. The characterization of the reagents and of these two methods is indicated below. In Hyperlab code terms: NH2 + NH3 = RAN (mg/l).

ASCORB – ASCORBIC ACID - SQPE072166 – 115 ml

An organic acid widely found in nature that acts as a remarkable antioxidant directly against dissolved oxygen. It thus preserves the colors and aromas of wines. It is generally added to must and/or during pre-bottling (1 to 10 g/l). It must always be protected from sulfur dioxide, as its oxidation leads to the formation of dehydroascorbic acid and hydrogen peroxide. The latter is a strong oxidizing agent.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
L-ASC R1 (ASCR1)	2 X 50	large
L-ASC R2 (ASCR2)	1 x 10	small
L-ASC R3 (ASCR3)	1 x 5	cup

These 3 reagents are the same used in the ASCR and ASCB coded methods that start automatically launching the ASCORB method.

Method principle and characteristics

The concentration of ascorbic acid in the sample is provided by the difference between the total quantity of reducing agents in the sample (ASCR) and a blank sample in which the ascorbic acid (ASCB) is removed. Therefore ASCR - ASCB = ASCORB, or the concentration of ascorbic acid. These determinations are performed automatically by launching exclusively the ASCORB method.
At an enzymatic level, all the reducing agents present in the sample reduce MTT (Thiazolyl Blue Terazolium Bromide) in the presence of PMS (Phenazine methylsulfate) to form formazan. The removal of ascorbic acid, upon which the calculation is based, is instead carried out by ascorbate oxidase. The difference in absorbance obtained, measured at 578 nm, is proportional to the amount of L-ascorbic acid present in the sample.
Linearity with total V / sample V = 50: up to 300 mg/l
Precision: CV% < 0.87 in white wine

Calibration and control: standard preparation at 250 mg/l, dissolve 125 mg/l of ascorbic in a 500 ml flask and then bring to volume.

Notes: Add 0.5 ml of propionaldehyde with a concentration of 10 g/l in 2 ml of sample to limit interference from SO2. Wait 5 minutes before starting the analysis. The result obtained must be multiplied by 1.25 (0.5 ml + 2 ml = 2.5 ml; 2.5 x 0.5 = 1.25). Do not leave the sample to be analyzed too long on the sample holder, given the high reactivity of ascorbic acid.

Ca2+ – CALCIUM – SQPE054972 – 2 X 100 ml

Calcium is in grapes, but it can also be present in wine due to treatments with calcium carbonate, with bentonite and with fossil flour, or it can come from storage in reinforced concrete containers.
Calcium in ionic form tends to precipitate during alcoholic fermentation because the progressive increase in ethanol decreases its solubility. It is important to determine its concentration, which in the finished wine should not exceed 80 mg/l, since over time higher concentrations could cause the precipitation of neutral calcium tartrate, a salt which originates from very slow formation kinetics.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
CA R1 (CALCR)	2 x 100	small
CA BL (CALCB)	2 x 100	small

Method principle and characteristics

In a sub-acidic environment, calcium reacts with Arsenazo III, a highly selective chromogen for this element, producing a colored complex of intensity proportional to the amount of calcium present in the sample. Optical density is measured at 650 nm.
Linearity with total V / sample V = 60: up to 150 mg/l
Precision: CV% < 2 in white wine, CV% < 2.5 in red wine
Calibration and control: standard SQPE053231 calcium 100 mg/l

Notes: Centrifuge or filter excessively turbid must before the analysis. Given the high sensitivity of this method, it is best to avoid any calcium contamination that may derive from: water that is not perfectly distilled and/or demineralized or from glassware with calcium residues. Furthermore, before performing the analysis, it would be best to carry out the "cuvette cleaning" procedure with two bottles containing 0.1N hydrochloric acid, in addition to performing this method on its own.

CATEC – CATECHINS – SQPE054972 – 5 x 20 ml

Catechins are located in different organs of the grapevines, especially in the grape seeds and in the epidermis of the grape exocarp. The term catechins indicates the monomeric (flavan-3-ol) and polymeric units represented by the isomers of catechin and epicatechin. These compounds take part in the formation of color in wines, act as antioxidants, can impart bitterness and astringency to wines and can interact with other macromolecules such as complex polyphenols and mannoproteins. Their concentration is strongly influenced by genetic, environmental, cultural and technological factors (5 - 100 mg/l in the musts) and is measured mainly for white wines, given their oxidizability starting from the pressing.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
CATE R1 (CATRM)	5 x 15	small (3 parts)
CATE R2 (CATRM)	5 x 5	small (1 part)
CATE BL	2 x 50	do not use

Mix three parts of R1 with one part of R2 to obtain a single CATRM working reagent (small bottle). It is possible to add one bottle of R2 to one of R1 (1:3), making sure to store any remaining amount at 2-8°C away from light.

Method principle and characteristics

In a strongly acidic environment and in a non-aqueous solvent, catechins react with cinnamaldehyde to form a chromophore with maximum absorption at 644 nm. Consequently, the measurement is done at 650 nm.
Linearity with total V / sample V = 6: up to 50 mg/l
Precision: CV% < 1.5 in white wine
Calibration and control: standard SQPE082087 catechin 500 mg/l (dilute 10 times to obtain the standard at 100 mg/l). Once diluted it will keep for one week in the refrigerator.

CITRC – CITRIC ACID – SQPE0076313 – 5 x 10 ml

A tri-carboxylic organic acid present in grapes at concentrations varying between 0.3 - 0.6 g/l. It does not play a primary role in oenological science and its level generally remains constant in wine. It can be metabolized by both lactic and acetic bacteria, with the resulting production of acetic acid. At present it is possible to add citric acid to wine, following the current laws in force (maximum 1 g/l), given its complexing action on ferric iron, preventing any ferric precipitations following aeration. In addition, with its acidifying action (0.93 g/l of citric acid increases the titratable acidity by 1%) this acid improves color and brings freshness to the product.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
CITR R1A (CIT1A)	5 x 10	small
CITR R1B (CIT1B)	1 x 1.25	cup
CITR R2 (CITR2)	5 x 0.25	cup
CITR BL	1 x 50	do not use

The R2 reagent (lyophilized) must be reconstituted by pouring 0.5 ml of distilled water into the cup. It remains stable for about 2 days. Shake the R1B reagent well before use.

Method principle and characteristics

In the presence of the enzyme citrate lyase, citric acid is converted to oxaloacetate, which is then reduced to malate by malate dehydrogenase, in the presence of NADH. Due to the possible decomposition of the oxaloacetate in reaction conditions, any pyruvate formed is converted to lactate by lactate dehydrogenase, in the presence of NADH.
The decrease in absorbance due to the consumption of NADH, measured at 340 nm, is proportional to the amount of citric acid present in the sample.
Linearity with total V / sample V = 75: up to 0.8 g/l
Precision: CV% < 1.3 in wine
Calibration and control: multiparameter standard SQPE053234 citric acid 1g/l; multilevel standard SQPE081638 citric acid 0.8 g/l (level 1), 0.6 g/l (level 2), 0.4 g/l (level 3), 0.3 g/l (level 4), 0.1 g/l (level 5).

Notes: In the case of strongly colored red wine (total polyphenols > 2500 mg / l) it is advisable to decolorize the sample with activated carbon or 1% PVPP weight/volume.

Cl- – CHLORIDES – SQPE055024 – 2 x 100 ml

The legal limit for chlorides in wines is 1 g/l of sodium chloride. Generally the salt concentrations found in wines are below the legal limit; however, the products coming from certain terroirs can have higher concentrations. At an organoleptic level, sapidity is a highly sought-after characteristic in the wine industry, especially in recent decades.
Some oenological adjuvants, such as liquid gelatins, contain sodium chloride.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
CL R1 (CLORR)	2 x 100	small
CL BL (CLORB)	2 x 100	small

Store the reagents at room temperature (15-20°C) and away from light.

Method principle and characteristics

Chloride ions react with mercury thiocyanate, developing a proportional amount of thiocyanate ions. The thiocyanate ions react with the iron (III) ions present in solution to form a red-colored complex characterized by an absorbance peak at 480 nm. The optical density is thus measured at 492 nm.
Linearity with total V / sample V = 60: up to 1.5 g/l
Precision: CV% < 1.5 in must and wine
Calibration and control: standard SQPE056370 chlorides 1 g/l NaCl

COLORE – COLOR – SQPE054875 – 4 x 100 ml

The color of wines is one of the main parameters for sensory evaluation. It also reflects some characteristics of the product such as the grape variety, the vintage, and the evolutionary state. In oenology it is common practice to observe absorbance values at 420 nm, 520 nm and 620 nm, since the sum 420 nm + 520 nm gives the coloring intensity of the product, and further adding the absorbance value at 620 nm one obtains the modified coloring intensity, while the 420 nm /520 nm ratio provides the hue of the product. These data are helpful and can be used in the objective evaluation of the product both over time and for new vintages. For example, for red wines, a correct aging process with controlled O2, in which the color stabilization takes place (polymerization between tannins and anthocyanins by means of an ethyl bridge), involves an increase in the contribution given by the wavelength at 620 nm; whereas in the case of unwanted oxidation processes, the contribution given by the wavelength at 420 nm increases.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
COL R1 (COL1)	4 x 100	large

Method principle and characteristics

The sample in question undergoes various programmed dilutions by means of a buffer stabilized at a known pH and with high ionic strength, thus avoiding the alteration of the ratio between the substances that contribute to the formation of the color. Following this, a mathematical algorithm is used to compare the results to the sample as is, therefore without considering the dilutions.

Notes: to observe the complete results, click on the green bar of the test of the sample being examined without dilution type C (Completed), which gives: the color intensity IC, the modified color intensity ICM, the hue TON and the pie chart with the different % contributions of the three absorbance values that contribute to the color formation.
Linearity up to 40 OD
Precision: CV% < 1.5 in red wine

Cu2+ – COPPER SELF BLANK – SQPE075544 – 50 ml

Grapes contain tens of mg/l of copper, but when ripe this concentration increases due to copper-based treatments applied to the vines. Copper can also be found in wine when bronze or brass equipment is used in the cellar. The amount of copper generally decreases after alcoholic fermentation, as it is incorporated in the lees. Excess copper can lead to unwanted cloudiness (copper casse) and catalyze the chemical oxidation process of the polyphenols, which leads to the formation of free radicals (strongly oxidizing).
In addition, monitoring of the copper concentration is considered necessary in the pre-bottling stage, since it is subject to a legal limit of 1 mg/l.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
CU R1	1 x 21	small
CU R2	1 x 4	cup
CU BL	1 x 25	use as R1

The reagent kit must be stored outside the refrigerator at room temperature (15±25°C). If it is at a lower temperature, the R1 and/or BL reagent must be warmed, for example in a thermostated bath at a temperature of about 30°C.

Method principle and characteristics

The 3,5-Di-Br-PAESA chromogen reacts with copper 2+ in the presence of sodium dodecyl sulfate to form a blue-violet complex, the absorbance of which is measured at 578 nm. The amount of chromophore formed is proportional to the amount of copper present in the sample. The specificity of the reaction is ensured by a specific environment, especially as regards the pH value.
Linearity with total V / sample V = 3: up to 1.2 mg/l
Precision: CV% < 2 in wine
Calibration and control: standard SQPE056386 copper 2 mg/l.

D-LATT –D-LACTIC ACID – SQPE059194 – 5 x 20 ml

Significant quantities of lactic acid (L-levorotatory 0.5 - 2.5 g / l) can be found in wine, since it represents the main product of the malolactic fermentation carried out by lactic acid bacteria.
However, lactic acid can be produced by the metabolism of sugars (fermentation) both by yeasts (L-lactic acid) and by bacteria (D-lactic acid). Thus the presence of D-lactic acid (D+ dextrorotatory) indicates an unpleasant bacterial development starting from the residual sugars which always goes along with the production of acetic acid. This fault is called mannitic fermentation.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
D-LAT R1 (DLAT1)	5 x 20	small
D-LAT R2 (DLAT2)	1 x 2.5	cup
D-LAT BL	2 x 50	do not use

Method principle and characteristics

D-lactate is converted into pyruvate by the enzyme D-lactate dehydrogenase in the presence of NAD. Given that the reaction equilibrium is clearly in favor of the D-lactate, the pyruvate that is formed is progressively removed from the reaction environment by the activity of the enzyme D-glutamate pyruvate transaminase, which transforms it into D-alanine.
The NADH that is formed from the oxidation of D-lactate is in a stoichiometric concentration with the analyte D-lactic acid. Thus the absorbance is measured at a wavelength of 340 nm.
Linearity with total V / sample V = 75: up to 1 g/l
Precision: CV% < 1.3 in wine
Calibration and control: multiparameter standard SQPE053234 D-lactic acid 3 g/l; multilevel standard SQPE081638 D-lactic acid 3.0 g/l (level 1), 2.4 g/l (level 2), 1.8 g/l (level 3), 1.2 g/l (level 4), 0.6 g/l (level 5).

D-MAL – D-MALIC ACID – SQPE067017 – 5 x 25 ml

The D(+) dextrorotatory isomer of malic acid plays a marginal role in oenology, because grapevines tend to accumulate mainly L-malic acid, the substrate of malolactic fermentation. D-malic acid may be present in traces; significant quantities can arise from food adulteration. The addition of L-malic acid for correcting the acidity of wines is currently allowed by law.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
D-MAL R1A (DML1A)	5 x 20	small
D-MAL R1B (DML1B)	1 x 25	small
D-MAL R2 (DMLR2)	1 x 2.5	cup
D-MAL BL	2 x 62.5	do not use

Shake R2 well before using.

Method principle and characteristics

The enzyme D-malate dehydrogenase catalyzes the selective oxidation of D-malic acid in the presence of NAD. The isomer L-malic acid, naturally present in solution, does not undergo reaction. The increase in absorbance due to the formation of NADH, measured at 340 nm, is proportional to the amount of D-malic acid in the sample.
Linearity with total V / sample V = 15: up to 1 g/l
Precision: CV% < 1.3 in wine
Calibration and control: multiparameter standard SQPE053234 D-malic acid 5 g/l; multilevel standard SQPE081638 D-malic acid 5.0 g/l (level 1), 4.0 g/l (level 2), 3.0 g/l (level 3), 2.0 g/l (level 4), 1.0 g/l (level 5).

DO420 – OD 420 nm – SQPE054875 – 4 x 100 ml

The color of wine is one of the main parameters for sensory evaluation. It also reflects some characteristics of the product such as the grape variety, the vintage, and the evolutionary state. In oenology it is common practice to observe absorbance values at 420 nm, 520 nm and 620 nm, since the sum 420 nm + 520 nm gives the coloring intensity of the product, and further adding the absorbance value at 620 nm one obtains the modified coloring intensity, while the 420 nm /520 nm ratio provides the hue of the product. The direct reading on the sample of the optical density at 420 nm can be useful in the case of unwanted oxidation processes in which, at the chromatic level, the contribution given by the wavelength at 420 nm increases.
This method does not provide for the use of reagents.

Method principle and characteristics

The sample being examined is read spectrophotometrically at 420 nm.
Linearity up to 3 OD

Notes: direct reading method suitable for white and rosé wines.

DO520 – DO 520 nm – SQPE054875 – 4 x 100 ml

The color of wine is one of the main parameters for sensory evaluation. It also reflects some characteristics of the product such as the grape variety, the vintage, and the evolutionary state. In oenology it is common practice to observe absorbance values at 420 nm, 520 nm and 620 nm, since the sum 420 nm + 520 nm gives the coloring intensity of the product, and further adding the absorbance value at 620 nm one obtains the modified coloring intensity, while the 420 nm /520 nm ratio provides the hue of the product. The direct reading on the sample of the optical density at 520 nm can be useful for evaluating the red component of the color of the wines, which tends to change according to the aging techniques used.

Method principle and characteristics

The sample being examined is read spectrophotometrically at 520 nm.
Linearity up to 3OD

Notes: direct reading method suitable for white and rosé wines.

ETANOL – ETHANOL – SQPE078690 – 50 ml

Ethyl alcohol is the main product of alcoholic fermentation, and is found in wines in concentrations between 11% vol. and 16% vol. It can be monitored during alcoholic fermentation, especially during the first stages, in order to identify exactly what additions of concentrated must might be necessary to obtain the desired alcohol content.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
ETAN R1 (ETAN1)	1 x 25	large
ETAN R2 (ETAN2)	1 x 25	small

Method principle and characteristics

Ethanol is oxidized to acetaldehyde by alcohol dehydrogenase in the presence of NAD, resulting in the formation of NADH. The NADH that is formed is in stoichiometric concentration with ethanol, therefore the increase in NADH is measured spectrophotometrically at 340 nm.
Linearity with total V / sample V = 100: up to 2% vol.
Precision: CV% < 1.3 in wine

F – FRUCTOSE – SQPE063151 – 5 x 20 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.
Fructose is one of the simple sugars of which there is the most in the grape. It differs from glucose in that it has a ketonic function (less reactive) and a greater sweetening power as regards the sensory analysis. It is metabolized together with glucose by yeasts during alcoholic fermentation. A few g/l of fructose in wine can represent the energy source for any bacterial development; in stabilized and finished wines it gives sweetness and roundness. In winemaking it is more appropriate to discriminate simple sugars individually, and the enzymatic method is identified as official for these determinations.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GL+FR R1 (G+FR1)	5 x 20	small
GL+FR R2 (G+FR)	1 x 2.5	cup
GL+FR R3 (G+FR3)	1 x 2.5	cup
GL+FR BL	2 x 50	do not use

Method principle and characteristics

In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer. The latter is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of fructose present in the sample.
Linearity with total V / sample V = 100: up to 2 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE063151 glucose-fructose-sucrose 20 g/l, with fructose 5 g/l, glucose 5 g/l and sucrose 10 g/l.

Notes: Method indicated for medium-low fructose concentrations. Given that in the presence of the enzyme hexokinase the glucose in the sample is also converted into glucose-6-phosphate, this method is reliable exclusively for samples in which fructose prevails over glucose, which must be decidedly less (fructose/glucose > 2).
Furthermore, the fructose concentration is obtained from the difference between the determination of glucose-fructose minus glucose.

Fe2+ - BIVALENT IRON – SQPE062468 – 5 x 20 ml

Wine contains trace amounts of iron as regards the mg/l. It is naturally present in grapes, but is often found in wine due to contamination coming from the vineyard or from the cellar, such as from utensils or untreated cement tanks.
The monitoring of iron, especially bivalent (2+), is absolutely essential for understanding and countering those oxidation phenomena in wine that are mainly “chemical.” This oxidizing process begins with the oxidation of polyphenols, especially ortho-diphenols, with the consequential production of quinones. O2 does not react directly with phenolic compounds until this limitation is overcome by the progressive addition of a single electron, which can be supplied by reduced transition metal ions, and by bivalent iron in particular. This sequential electron transfer leads to the formation of free radicals with extremely strong oxidizing power, which begin to oxidize the wine’s organic molecules according to their concentration, starting with ethanol.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT

FE R1 (FERR1)	5 x 20	small
FE R2	1 X 20	do not use
FE R3 (FERR3)	1 x 2.5	cup
FE BL	2 x 50	do not use

Method principle and characteristics

The Fe (II) ions react with the Ferene-S to give a blue colored complex, whose absorbance at 578 nm is directly proportional to the concentration of iron in the sample. Thiourea is also added to eliminate copper interference.
Linearity with total V / sample V = 14: up to 40 mg/l
Precision: CV% < 1.5 in white wine, CV% < 2 in red wine
Calibration and control: standard SQPE053230 iron 20 mg/l.

FeION – IONIC IRON – SQPE062468 – 5 x 20 ml

Wine contains trace amounts of iron as regards the mg/l. It is naturally present in grapes, but is often found in wine due to contamination coming from the vineyard or from the cellar, such as from utensils or untreated cement tanks.
Bivalent iron (II) is a fundamental cofactor of the chemical oxidation of wines, while ionic iron (III), which can derive from the oxidation of bivalent iron as a function of the system's redox potential, can give rise to precipitation, such as: blue casse, which leads to the formation of insoluble compounds between polyphenols and excess iron; white casse, the combination of Fe (III), proteins, calcium and potassium ions, which together with the colloids form poorly soluble aggregates. Furthermore, insoluble crystals can form from combinations of phosphoric acid and excess iron (phosphate-ferric casse).
To counter these phenomena, in addition to the use of antioxidants and protective colloids, it is possible to make use of the chelating action of citric acid against iron (III).

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
FE R1 (FERR1)	5 x 20	small
FE R2 (FERR2)	1 X 20	small
FE R3 (FERR3)	1 x 2.5	cup
FE BL	2 x 50	do not use

Method principle and characteristics

The iron is ionized in an acid medium and then reduced by ascorbic acid to Fe (II). The Fe (II) ions react with Ferene-S to give a blue colored complex, whose absorbance at 578 nm is directly proportional to the concentration of iron in the sample. Thiourea is also added to eliminate copper interference.
Linearity with total V / sample V = 14: up to 40 mg/l
Precision: CV% < 1.5 in white wine, CV% < 2 in red wine
Calibration and control: multiparameter standard SQPE053230 iron 20 mg/l.

G – GLUCOSE AUTO – SQPE079100 – 50 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.
Glucose is the simple sugar most present in the grape and is the elective substrate of the energy metabolism of yeasts and many other microorganisms. It differs from fructose in that it has an aldehyde function (more reactive) and a lower sweetening power as regards the sensory analysis. It also represents the aglycone for many volatile aromatic compounds and for anthocyanins. Some g/l of glucose in wine can represent the energy source for the possible development of bacteria; in stabilized and finished wines it gives sweetness and roundness. Therefore in winemaking it is more appropriate to discriminate simple sugars individually, and the enzymatic method is identified as official for these determinations.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GL AUTO R1 (GLAU1)	1 x 40	large
GL AUTO R2 (GLAU2)	1 x 10	small

Method principle and characteristics

The glucose is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of glucose present in the sample.
Linearity with total V / sample V = 100: up to 8 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE063151 glucose-fructose-sucrose 20 g/l with glucose 5 g/l, fructose 5 g/l and sucrose 10 g/l.

Notes: Method indicated for medium-low glucose concentrations.

G+F – GLUCOSE + FRUCTOSE – SQPE063019 – 5 x 20 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.
Glucose and fructose are the fermentable hexoses in the must (on average 200 g/l), both of which are metabolized by yeast during alcoholic fermentation. The residual sugars, on the order of a few g/l, give sweetness to the product but at the same time they constitute the energy source for any unwanted bacterial development. In general, alcoholic fermentation is considered completed when the fermentable sugar content is less than 2g/l.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GL+FR R1 (G+FR1)	5 x 20	small
GL+FR R2 (G+FR2)	1 x 2.5	cup
GL+FR R3 (G+FR3)	1 x 2.5	cup
GL FR BL	2 x 50	do not use

Method principle and characteristics

In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer. The latter, together with the glucose present in the sample, is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of glucose/fructose present in the sample.
Linearity with total V / sample V = 100: up to 8 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE053233 glucose-fructose 20 g/l.

Notes: Method indicated for medium-low glucose-fructose concentrations.

GF-7 – GLUCOSE FRUCTOSE AUTO 0-7 g/l – SQPE068207 – 125 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.
Glucose and fructose are the fermentable hexoses in the must (on average 200 g/l), both of which are metabolized by yeast during alcoholic fermentation. The residual sugars, on the order of a few g/l, give sweetness to the product but at

the same time they constitute the energy source for any unwanted bacterial development. In general, alcoholic fermentation is considered completed when the fermentable sugar content is less than 2g/l.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GF AUTO R1 (GFAU1)	1 x 40	large
GF AUTO R2 (GFAU2)	1 x 10	small

Method principle and characteristics

In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer. The latter, together with the glucose present in the sample, is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of glucose/fructose present in the sample.
Linearity with total V / sample V = 100: up to 8 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE053233 glucose-fructose 20 g/l.

Notes: Method indicated for low glucose-fructose concentrations.

GF-50 – GLUCOSE FRUCTOSE AUTO 7-50 g/l– SQPE068207 – 125 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.
Fermentable sugars such as glucose and fructose can be found in wine in high concentrations, for example when fermentation is stopped, either involuntarily or voluntarily as in the production of dessert wines. These wines are often subject to sugar concentrations imposed by the production regulations.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GL AUTO R1 (GFAU1)	1 x 40	large
GL AUTO R2 (GFAU2)	1 x 10	small

Method principle and characteristics

In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer. The latter, together with the glucose present in the sample, is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of glucose/fructose present in the sample.
Linearity with total V / sample V =75: up to 50 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE053233 glucose-fructose 20 g/l; multiparameter standard SQPE067024 glucose/fructose 50 g/l.

Notes: Method indicated for medium glucose-fructose concentrations.

GF-300 – GLUCOSE FRUCTOSE AUTO 50-300 g/l – SQPE068207 – 125 ml

The determination of sugars to identify the technological maturity of the grapes according to the type of wine to be made begins after the veraison stage. For example, the grapes for making sparkling wine are harvested a month earlier than the grapes for making a long-aging red wine, because they must have a lower alcohol content must.
The main sugars that are found in grape must and are used by yeasts during alcoholic fermentation are glucose and fructose, on average 200 g/l in a ratio with slightly more fructose. Before alcoholic fermentation, or rather during the

early stages, the sugar concentration can be corrected by adding concentrated must, in order to modulate the desired alcohol content.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GF AUTO R1 (GFAU1)	2 x 50	large
GF AUTO R2 (GFAU2)	1 x 25	small

Method principle and characteristics

In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer. The latter, together with the glucose present in the sample, is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of glucose/fructose present in the sample.
Linearity with total V / sample V =50: up to 300 g/l
Precision: CV% < 1.5 in white must; CV% < 2 in red must
Calibration and control: multiparameter standard SQPE067024 with six solutions with different glucose-fructose concentrations: 50, 100, 150, 200, 250 and 300 g/l.

GFDRY – GLUCOSE FRUCTOSE 0-7 g/l – SQPE053688 – 5 x 20 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.
Glucose and fructose are the fermentable hexoses in the must (on average 200 g/l), both of which are metabolized by yeast during alcoholic fermentation. The residual sugars, on the order of a few g/l, give sweetness to the product but at the same time they constitute the energy source for any unwanted bacterial development. In general, alcoholic fermentation is considered completed when the fermentable sugar content is less than 2g/l.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GL-FR R1 (GFR1)	5 x 20	small
GL-FR R2 (GFR2)	1 x 2.5	cup
GL-FR BL	2 x 50	do not use

Method principle and characteristics

In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer. The latter, together with the glucose present in the sample, is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of glucose/fructose present in the sample.
Linearity with total V / sample V =100: up to 8 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE053233 glucose-fructose 20 g/l.

Notes: Method indicated for low glucose-fructose concentrations.

GFS – GLUCOSE FRUCTOSE SUCROSE – SQPE068207– 125 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.

Glucose, fructose and sucrose are the fermentable sugars in the must (on average 200 g/l), and they are metabolized by yeast during alcoholic fermentation. The residual sugars, on the order of a few g/l, give sweetness to the product but at the same time they constitute the energy source for any unwanted bacterial development. In general, alcoholic fermentation is considered completed when the fermentable sugar content is less than 2g/l. This method also quantifies any traces of sucrose that may be found in wine.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GF AUTO R1 (GFAU1)	1 x 40	large
GF AUTO R2 (GFAU2)	1 x 10	small
SACC	10 ml	small

The reagent SACC is sold individually with its own packaging, identified by the code SQPE063020 – SUCROSE – 10ml.

Method principle and characteristics

Sucrose is hydrolyzed by the enzyme invertase into D-glucose and D-fructose. In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer, which is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of sugars present in the sample.
Linearity with total V / sample V =50: up to 8 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE063151 glucose-fructose-sucrose 20 g/l with sucrose 10 g/l, glucose 5g/l and fructose 5 g/l.

GLICER – GLYCERIN – SQPE060138 – 2 x 100 ml

Glycerol is one of the most important compounds of wine quantitatively (5 - 8 g/l). It gives sensations of sweetness and roundness, contrasting the bitter and astringent sensations of wine. It is also one of the factors that contributes to the formation of “legs” when the wine is swirled in the glass.
Modest glycerol levels can be found in musts obtained from dried or rotten grapes. Glycerol is mainly produced by yeasts during the early stages of alcoholic fermentation: the NADH that is formed by the oxidation of glyceraldehyde-3-phosphate cannot release the H+ to acetaldehyde, which has not yet formed, therefore it is oxidized on dioxyacetone phosphate, which is reduced to glycerophosphate and then hydrolyzed to glycerin (glycerol-pyruvic fermentation). This metabolic strategy allows the maintaining of an adequate NAD/NADH ratio and therefore the continuation of glycolysis.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GLIC R1 (GLICR)	2 x 100	large
GLIC BL	2 x100	optional

Method principle and characteristics

Glycerol takes part in a series of enzymatic reactions that produce H2O2 by means of glycerol kinase and glycerol phosphate oxidase. Hydrogen peroxide reacts with p-chlorophenol and 4-aminoantipyrine in the presence of peroxidase, forming a red-colored quinoneimine compound. The color intensity, measured at 520 nm, is proportional to the amount of glycerol present in the sample.
Linearity with total V / sample V =30: up to 12.5 g/l
Precision: CV% < 1.5 in white wine; CV% < 2 in red wine
Calibration and control: standard SQPE053232 glycerol 10 g/l.

GLICUV – UV GLYCEROL – SQPE078691 – 50 ml

Glycerol is one of the most important compounds of wine quantitatively (5 - 8 g/l). It gives sensations of sweetness and roundness, contrasting the bitter and astringent sensations of wine. It is also one of the factors that contributes to the formation of "legs" when the wine is swirled in the glass.
Modest glycerol levels can be found in musts obtained from dried or rotten grapes. Glycerol is mainly produced by yeasts during the early stages of alcoholic fermentation: the NADH that is formed by the oxidation of glyceraldehyde-3-phosphate cannot release the H+ to acetaldehyde, which has not yet formed, therefore it is oxidized on dioxyacetone phosphate, which is reduced to glycerophosphate and then hydrolyzed to glycerin (glycerol-pyruvic fermentation). This metabolic strategy allows the maintaining of an adequate NAD/NADH ratio and therefore the continuation of glycolysis.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
GLIC R1A (GUV1A)	1 x 20	small
GLIC R1B (GUV1B)	1 x 20	small
GLIC R2 (GUV2)	1 x10	small

Method principle and characteristics

In the presence of ATP, glycerol is converted into glycerol-3-phosphate by the enzyme glycerol kinase, with the resulting formation of ADP. The ADP that is formed reacts with phosphoenolpyruvate in the presence of pyruvate kinase to form ATP and pyruvate. In the presence of the enzyme LDH the pyruvate is reduced to lactate, oxidizing NADH. The decrease in absorbance measured at 340 nm, resulting from the oxidation of NADH, is proportional to the amount of glycerol present in the sample.
Linearity with total V / sample V =50: up to 6 g/l
Precision: CV% < 1.5 in white wine; CV% < 2 in red wine
Calibration and control: standard SQPE053232 glycerol 10 g/l.

GLUCON – GLUCONIC ACID – SQPE076314 – 1 x 50 ml

The concentration of gluconic acid is one of the fundamental parameters to be checked in winemaking during pressing, especially in the case of grapes affected by rotting. Given that gluconic acid is mainly produced by *Botrytis cinerea* starting from the oxidation of glucose, it is an indicator of the health and quality of the grapes. In severe cases the content of this acid can reach 2 - 3 g/l, while the optimal concentration found in musts obtained from perfectly healthy grapes is lower than 0.1 g/l.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
D-GLUC R1 (GLUC1)	1 x 40	large
D-GLUC R2 (GLUC2)	1 x 10	small
D-GLUC BL	1 x 50	do not use

Method principle and characteristics

In the presence of the enzyme gluconate kinase, D-gluconic acid is phosphorylated to D-gluconate-6-phosphate, with the consequential conversion of ATP into ADP. D-gluconate-6-phosphate, in the presence of NADP which is reduced to NADPH, is decarboxylated by the enzyme 6-phosphogluconate dehydrogenase and becomes ribulose 5-phosphate. The increase in absorbance due to the formation of NADPH, measured at 340 nm, is proportional to the amount of D-gluconic acid present in the sample.
Linearity with total V / sample V =50: up to 3 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE053234 gluconic acid 1 g/l.

K+ - POTASSIUM – SQPE056387 – 1 x 100 ml

Wine can be defined as a potassium beverage given its higher concentration of this ion, 400 - 1500 mg/l depending on the degree of tartaric stability, compared to other beverages.
The main role in winemaking of potassium, which is derived from the grapes and strongly influenced by the environment and the soil, regards salification with tartaric acid, which can cause unwanted crystalline precipitates. The

degree of tartaric stability of the wine, as well as the potassium bitartrate content, depends on other factors such as: temperature, alcohol content, presence of protective colloids, and pH.
The significant decrease in the concentration of potassium, after stabilizing treatments, is therefore an indication of tartaric stability.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
K R1 (POTAR)	1 x 100	large
K BL (POTAB)	1 x 100	large

Do not shake the reagents before use; if this happens, wait for them to settle.

Method principle and characteristics

The potassium ion reacts with sodium tetraphenylborate, forming a colloidal solution. Other components eliminate interference and stabilize the suspension. Consequently, it generates a certain turbidity that is proportional to the potassium concentration, which is read spectrophotometrically at 500 nm.
Linearity with total V / sample V =150: up to 1500 mg/l
Precision: CV% < 10 in wine
Calibration and control: standard SQPE056388 potassium 1500 mg/l.

Notes: as for all turbidimetric methods, it is recommended that multipoint calibrations be performed and the accuracy checked with standards at different concentrations. Given that the products of reactions are dirtying, the cuvette cleaning procedure should be carried out at the end of the analysis. Since this is the determination of the potassium ion, do not interfere by using potassium hydroxide (use NaOH) as a basic agent for cleaning.

LLATAU – L-LACTIC ACID AUTO – SQPE074739 – 125 ml

L-lactic acid plays a fundamental role in the production of red wines. Significant quantities of lactic acid are found in wine (1.5 - 2.5 g/l) following the malolactic fermentation carried out by the lactic acid bacteria of the genus *Oenococcus*. The conversion of malic acid into lactic acid involves raising the pH, decreasing astringency and increasing the complexity of the aromas. Generally 0.64 g of lactic acid plus CO2 are obtained from 1 g of malic acid. The starting of malolactic fermentation, and therefore the bacterial development, is strongly influenced by environmental factors, such as: pH, temperature, alcohol content, and content of energy and nitrogenous sources.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
L-LAT AUTO R1 (LATA1)	1 x 40	large
L-LAT AUTO R2 (LATA2)	1 x 10	small

Method principle and characteristics

L-lactate is converted into pyruvate by the enzyme L-lactate dehydrogenase in the presence of NAD. Given that the reaction equilibrium is clearly in favor of L-lactate, the pyruvate that is formed is progressively removed from the reaction environment by the activity of the enzyme D-glutamate pyruvate transaminase, which transforms it into D-alanine.
The NADH that is formed from the oxidation of D-lactate is in a stoichiometric concentration with the analyte L-lactic acid. Thus the absorbance is measured at a wavelength of 340 nm.
Linearity with total V / sample V =100: up to 3 g/l
Precision: CV% < 1.3 in wine
Calibration and control: multiparameter standard SQPE053234 L-lactic acid 3 g/l; multilevel standard SQPE081638 L-lactic acid 3.0 g/l (level 1), 2.4 g/l (level 2), 1.8 g/l (level 3), 1.2 g/l (level 4), 0.6 g/l (level 5).

L-LATT – L-LACTIC ACID – SQPE059112 – 5 x 20 ml

L-lactic acid plays a fundamental role in the production of red wines. Significant quantities of lactic acid are found in wine (1.5 - 2.5 g/l) following the malolactic fermentation carried out by the lactic acid bacteria of the genus

Oenococcus. The conversion of malic acid into lactic acid involves raising the pH, decreasing astringency and increasing the complexity of the aromas. Generally 0.64 g of lactic acid plus CO2 are obtained from 1 g of malic acid. The starting of malolactic fermentation, and therefore the bacterial development, is strongly influenced by environmental factors, such as: pH, temperature, alcohol content, and content of energy and nitrogenous sources.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
L-LAT R1 (LLAT1)	5 x 20	small
L-LAT R2 (LLAT2)	1 x 2.5	cup
L-LAT BL	2 x 50	do not use

Method principle and characteristics

L-lactate is converted into pyruvate by the enzyme L-lactate dehydrogenase in the presence of NAD. Given that the reaction equilibrium is clearly in favor of L-lactate, the pyruvate that is formed is progressively removed from the reaction environment by the activity of the enzyme D-glutamate pyruvate transaminase, which transforms it into D-alanine.

The NADH that is formed from the oxidation of D-lactate is in a stoichiometric concentration with the analyte L-lactic acid. Thus the absorbance is measured at a wavelength of 340 nm.

Linearity with total V / sample V =150: up to 3 g/l

Precision: CV% < 1.3 in wine

Calibration and control: multiparameter standard SQPE053234 L-lactic acid 3 g/l; multilevel standard SQPE081638 L-lactic acid 3.0 g/l (level 1), 2.4 g/l (level 2), 1.8 g/l (level 3), 1.2 g/l (level 4), 0.6 g/l (level 5).

MALAUT – L-MALIC ACID – SQPE068206 – 125 ml

D-malic acid is the acid second most present quantitatively in grapes (1.5 - 4.5 g/l). Its concentration decreases as the fruit ripens, especially in the hottest years, as it is metabolically breathed by the plant, being an intermediate of the Krebs cycle. This acid contributes to the formation of the total acidity of the must, and thus it influences the technological maturity. It is also the substrate of the malolactic enzyme of the lactic acid bacteria of oenological interest that transform it into lactic acid, with a consequential increase in pH, decrease in acidity and increase in product softness.

Generally 0.64 g of lactic acid plus CO2 are obtained from 1 g of malic acid. The starting of malolactic fermentation, and therefore the bacterial development, is strongly influenced by environmental factors, such as: pH, temperature, alcohol content, and content of energy and nitrogenous sources.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
L-MAL AUTO R1 (MALA1)	2 x 50	large
L-MAL AUTO R2 (MALA2)	1 x 25	small

Method principle and characteristics

L-malic acid is oxidized to oxaloacetate by the enzyme L-malate dehydrogenase in the presence of NAD, and therefore with the formation of NADH. Given that the reaction equilibrium is shifted towards L-malate, there is the presence of the enzyme glutamate-oxaloacetate transaminase, which subtracts the oxaloacetate from the reaction environment. The NADH that is formed is in a stoichiometric concentration with L-malic acid, and this increase in NADH is measured at 340 nm.

Linearity with total V / sample V =100: up to 5 g/l

Precision: CV% < 1 in white wine CV% < 1.5 in red wine

Calibration and control: multiparameter standard SQPE053234 L-malic acid 5 g/l; multilevel standard SQPE081638 L-malic acid 5.0 g/l (level 1), 4.0 g/l (level 2), 3.0 g/l (level 3), 2.0 g/l (level 4), 1.0 g/l (level 5).

MALICO – L-MALIC ACID - SQPE053689 – 5 x 20 ml

D-malic acid is the acid second most present quantitatively in grapes (1.5 - 4.5 g/l). Its concentration decreases as the fruit ripens, especially in the hottest years, as it is metabolically breathed by the plant, being an intermediate of the

Krebs cycle. This acid contributes to the formation of the total acidity of the must, and thus it influences the technological maturity. It is also the substrate of the malolactic enzyme of the lactic acid bacteria of oenological interest that transform it into lactic acid, with a consequential increase in pH, decrease in acidity and increase in product softness.
Generally 0.64 g of lactic acid plus CO2 are obtained from 1 g of malic acid. The starting of malolactic fermentation, and therefore the bacterial development, is strongly influenced by environmental factors, such as: pH, temperature, alcohol content, and content of energy and nitrogenous sources.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
L-MAL R1 (MAL1)	5 x 20	small
L-MAL R2 (MAL2)	1 x 2.5	cup
L-MAL BL	2 x 50	do not use

Method principle and characteristics

L-malic acid is oxidized to oxaloacetate by the enzyme L-malate dehydrogenase in the presence of NAD, and therefore with the formation of NADH. Given that the reaction equilibrium is shifted towards L-malate, there is the presence of the enzyme glutamate-oxaloacetate transaminase, which subtracts the oxaloacetate from the reaction environment. The NADH that is formed is in a stoichiometric concentration with L-malic acid, and this increase in NADH is measured at 340 nm.
Linearity with total V / sample V =100: up to 5 g/l
Precision: CV% < 1 in white wine CV% < 1.5 in red wine
Calibration and control: multiparameter standard SQPE053234 L-malic acid 5 g/l; multilevel standard SQPE081638 L-malic acid 5.0 g/l (level 1), 4.0 g/l (level 2), 3.0 g/l (level 3), 2.0 g/l (level 4), 1.0 g/l (level 5).

Mg2+ – MAGNESIUM – SQPE056389 – 2 x 100 ml

Magnesium is one of the cations most abundant in living cells, acting as a cofactor for over 300 enzymes and representing a growth factor for microorganisms. Therefore it is also functional to the metabolic activity of yeasts, especially during alcoholic fermentation, as it stimulates the activity of the enzyme pyruvate decarboxylase during glycolysis. The amount of magnesium required is 50 - 100 mg/l, and there may be magnesium concentrations of up to 160 mg/l in the must. However, the concentration may be significantly lower due to certain amino acids and chelating agents that can complex magnesium, making it unavailable to microorganisms.
Magnesium can be added to the must by adding magnesium sulfate.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
MAGN R1 (MGR)	1 x 100	small
MAGN R2 (MGR)	1 x 100	small
MAGN BL (MGB)	2 x 200	small

Mix reagents R1 and R2 in equal parts (1:1) to make up the MGR reagent. Remains stable for 2 days if stored at 4°C.

Method principle and characteristics

In an alkaline environment, calmagite binds with the magnesium present in the sample, forming a chromophore complex proportional to the quantity of analyte. Specific chelating agents eliminate interference due to the presence of other samples. Spectrophotometric measurement is done at 520 nm.
Linearity with total V / sample V =100: up to 100 mg/l
Precision: CV% < 1.5 in white wine; CV% < 2 in red wine
Calibration and control: standard SQPE056390 magnesium 24.3 mg/l.

NH2 – AMINO NITROGEN - SQPE054974 – 2 X 60 ml

Sources of nitrogen are a fundamental factor for the multiplication and development of yeasts, the protagonists of alcoholic fermentation, and of the bacteria that are the protagonists of malolactic fermentation. These sources are present in the must, but are often added, for example in the hottest years, to obtain a level of RAN (Readily Assimilable

Nitrogen) of least 200 mg/l. The α-amino nitrogen is identified in the amino acid fraction of the must (the yeasts do not metabolize proline), which is metabolized and catabolized by the yeasts at differing intensities according to the fermentation process. These microorganisms generally use the ammoniacal fraction in the early stages of fermentation, while in the later stages they mostly use the amino acid fraction. The latter is integrated with the must/wine in the form of mixed activators and/or yeast derivatives. The nitrogen content also influences the organoleptic characteristics of wines, because it modulates the production of superior alcohols (precursors of esters with fruity notes) and of sulfur molecules that can give the wine reduced notes.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
NH2 R1	2 X 50	large
NH2 R2	1 x 25	small
NH2 BL	2 x 60	do not use

Method principle and characteristics

In a basic environment with controlled pH, the amino group of free α-amino acids (NH2) reacts with ortho-phthalaldehyde in the presence of N-acetylcysteine, forming chromophoric derivatives (isoindolic) in stoichiometric quantities with the free amino groups, which show an absorption band from 335 to 345 nm. The optical density read at 340 nm is proportional to the quantity of α-amino nitrogen present in the sample under examination.
Linearity with total V / sample V =75: up to 200 mg/l
Precision: CV% < 1.5 in white wine, CV% < 2 in red wine
Calibration and control: standard SQPE055107 α-amino nitrogen 160 mg/l

NH3 – AMMONIACAL NITROGEN - SQPE054975 -2 X 50 ml

Sources of nitrogen are a fundamental factor for the multiplication and development of yeasts, the protagonists of alcoholic fermentation, and of the bacteria that are the protagonists of malolactic fermentation. These sources are present in the must, but are often added, for example in the hottest years, to obtain a level of RAN (Readily Assimilable Nitrogen) of least 200 mg/l. Ammoniacal nitrogen is normally metabolized by the yeast at differing intensities according to the fermentation and composition of the must. These microorganisms generally use the ammoniacal fraction in the early stages of fermentation, while in the later stages they mostly use the amino acid fraction. The former can be added to the must /wine in the form of ammonium salts, considering that 30 g/Hl provide 63 mg/l of readily assimilable nitrogen.
The nitrogen content also influences the organoleptic characteristics of wines, because it modulates the production of superior alcohols (precursors of esters with fruity notes) and of sulfur molecules that can give the wine reduced notes.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
NH3 R1	2 x 40	large
NH3 R2	1 x 20	small
NHH3 BL	2 x 50	do not use

Method principle and characteristics

Ammoniacal nitrogen reacts with α-ketoglutarate in the presence of NADH under the catalytic action of the enzyme glutamate dehydrogenase, producing NAD and L-glutamate.
The photometric measurement at 340 nm of the quantity of NADH oxidized to NAD indicates the quantity of ammoniacal nitrogen present in the sample.
Linearity with total V / sample V =38: up to 100 mg/l
Precision: CV% < 1.5 in white wine, CV% < 2 in red wine
Calibration and control: standard SQPE054993 ammoniacal nitrogen 50 mg/l.

PIRUV – PYRUVIC ACID – SQPE056391 – 5 x 20 ml

Pyruvate is found mainly in the wine, as it is produced by the energy metabolism of yeast during glycerol-pyruvic fermentation where, starting from glucose, for each molecule of glycerin that is formed, a molecule of pyruvic acid accumulates. The concentration can vary from tens to hundreds of mg/l.

This acid, which is highly reactive chemically and enzymatically, is the precursor of various substances that can be at the origin of olfactory faults in wines: vinegary, mousey, burnt.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
PIRU R1 (PIRR1)	5 x 20	small
PIRU R2 (PIRR2)	1 x 2.5	cup
PIRU BL	2 x 50	do not use

Method principle and characteristics

Pyruvic acid is reduced to D-lactic acid in the presence of the enzyme lactate dehydrogenase, with the consequential oxidation of NADH to NAD. The amount of NAD formed is proportional to the amount of pyruvic acid, and the decrease in absorbance, due to the disappearance of NADH, is measured at 340 nm.
Linearity with total V / sample V =25: up to 0.6 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE056392 pyruvic acid 0.5 g/l.

Notes: to improve the performance of the analysis, it is recommended that structured red wines be decolorized with total polyphenols > 2500 mg/l.

POLIB – WHITE WINE POLYPHENOLS – SQPE054970 – 3 x 100 ml

Polyphenols accumulate mainly in the grape peel and seeds during the development of the berry and during the ripening of the fruit, therefore they are found in the must and especially in the wines undergoing maceration. The following classes of compounds fall within the category of polyphenols of oenological interest: hydrolysable tannins, condensed tannins, anthocyanins, flavonols and hydroxycinnamic acids. The concentration of total polyphenols depends on the grape variety, the environment, and technological factors.
These compounds determine a large part of the color and flavor quality of the wine, as they are at the basis of the astringency, the bitterness, the longevity and the evolutionary potential of the product. They are analyzed starting from the ripening of the grapes up to the bottling of the wine, given the numerous changes to which these compounds are subject.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
POLI3 R1 (POLI1)	2 x 100	large
POLI3 R2 (POLI2)	1 x 100	small

Method principle and characteristics

The potassium ion reacts with sodium tetraphenylborate, forming a colloidal solution. Other components eliminate interference and stabilize the suspension. Consequently, it generates a certain turbidity that is proportional to the potassium concentration, which is read spectrophotometrically at 500 nm.
Linearity with total V / sample V =30: up to 600 mg/l
Precision: CV% < 10 in wine
Calibration and control: standard SQPE054995 polyphenols 3 g/l (anhydrous gallic acid).

Notes: This method is suitable for white wines with a polyphenols concentration of up to 600 mg/l. To improve the accuracy of the method, it is advisable: to verify that the Blank value is less than 1; to perform the analysis of the polyphenols alone (not together with other methods); to clean the needle before the analysis.

POLIT – TOTAL POLYPHENOLS – SQPE054970 – 3 x 100 ml

Polyphenols accumulate mainly in the grape peel and seeds during the development of the berry and during the ripening of the fruit, therefore they are found in the must and especially in the wines undergoing maceration. The following classes of compounds fall within the category of polyphenols of oenological interest: hydrolysable tannins, condensed tannins, anthocyanins, flavonols and hydroxycinnamic acids. The concentration of total polyphenols depends on the grape variety, the environment, and technological factors.

These compounds determine a large part of the color and flavor quality of the wine, especially in red wines, as they are at the basis of the astringency, the bitterness, the longevity and the evolutionary potential of the product. They are analyzed starting from the ripening of the grapes up to the bottling of the wine, given the numerous changes to which these compounds are subject.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
POLI3 R1 (POLI1)	2 x 100	large
POLI3 R2 (POLI2)	1 x 100	small

Method principle and characteristics

In a strongly basic environment, the Folin-Ciocalteu reagent oxidizes the hydroxyl groups of the polyphenols present in the sample, so that the products of reduction formed are proportional to the analyte. These blue-colored products are detected spectrophotometrically at 700 nm.
Linearity with total V / sample V =101: up to 3000 mg/l
Precision: CV% < 1.5 in red wine
Calibration and control: standard SQPE054995 polyphenols 5 g/l.

Notes: This method is suitable for red wines with a polyphenols concentration between 600 and 3000 mg/l. To improve the accuracy of the method, it is advisable: to verify that the Blank value is less than 1; to perform the analysis of the polyphenols alone (not together with other methods); to clean the needle before the analysis.

PROT – PROTEIN – SQPE076312 – 1 x 50 ml

The protein concentration in wine can vary from a few mg/l up to about 100 mg/l. These substances come from the grapes, and their content increases with the plant's response to stress in the case of disease. The oenological interest in proteins regards white wines, given the ability of proteins to impart turbidity to the product. In fact, the denaturation of proteins, such as that caused by heating, followed by their interaction with other compounds such as tannins, leads to the formation of invisible insoluble particles that aggregate and cause the wine to become cloudy. Generally, and when necessary, protein stability is achieved by means of physical treatments (ultrafiltration), the addition of tannins, proteolytic enzymes, polysaccharides, hyperoxygenation of the must or aging on the lees. Bentonite remains the most common additive for removing proteins.
To assess the degree of protein stability technicians interpret the NTU values individually, whereas considering the protein concentration, the following stability thresholds can be considered, in reference to the type of product: below 25 mg/l, stable; 25 to 50 mg/l, at risk; and above 50 mg/l, unstable.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
PROT R1 (PROT1)	1 x 12.5	large
PROT R2 (PROT2)	1 x 50	small
PROT BL	1 x 25	do not use
STANDARD	1 x 10	Use as sample

As part of each analytical cycle, perform the analysis of two standards with a concentration of 50 mg/l (0.1 ml standard + 0.9 ml PROT BL) and 25 mg/l (1 ml standard at 50 mg/l + 1 ml of PROT BL), and then realign the results. Mix carefully to obtain a homogeneous solution, given the high viscosity of the standard.
The R1 reagent must be diluted 1:10 with distilled water (1 ml R1 + 9 ml H2O), while the R2 reagent must be shaken gently before use.

Method principle and characteristics

The sample is given a pretreatment with a basic solution to favor the solubilization of the proteins, which then react with the Brilliant Blue G chromogen. The protein-chromogen complex generates a decrease in absorbance at 465 nm and an increase in absorbance at 595 nm. Therefore the 595 nm/465 nm ratio is a function of the concentration of proteins present in the sample.
Linearity with total V / sample V =5.2: up to 100 mg/l

Precision: CV% < 5 in white wine
Calibration and control: standard proteins 1000 mg/l supplied with the Work Kit.

Notes: For samples containing more than 100 mg/l of protein, dilute the sample 1:4 with the BL reagent and multiply the result by 5. To increase the accuracy of the result, it is recommended that each analysis be performed three times and the result be considered as the average of the three values obtained.

S – SUCROSE - SQPE063020 – 10 ml

In the wine industry, the sugar concentration is determined to establish the technological maturity of the grapes, to follow the fermentation process, to evaluate the effect of adding concentrated musts, and to determine the residual sugars. In addition, the dosage of sugar for secondary fermentation is a common practice in the production of sparkling wines.
Sucrose may be present in wine in traces, but it must be monitored in the sparkling wine process and in the production of sweet wines.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
reagent R1 (GFAU1)	5 x 20	small
reagent R1B (GFAU2)	1 x 2.5	small
SACC	10	small

Reagents R1 and R1B are sold as GLUCOSE FRUCTOSE AUTO with the code SQPE068207.

Method principle and characteristics

Sucrose is hydrolyzed by the enzyme invertase into D-glucose and D-fructose. In the presence of phosphoglucose isomerase, D-fructose is converted into the D-glucose isomer, which is transformed into glucose-6-phosphate by the enzyme hexokinase, and is then oxidized to gluconate-6-phosphate by the enzyme glucose-6-phosphate dehydrogenase while reducing the NADP to NADPH. Thus the increase in absorbance, measured at 340 nm, due to the NADPH is proportional to the amount of glucose present in the sample.
Linearity with total V / sample V =100: up to 6 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE063151 glucose-fructose-sucrose 20 g/l with sucrose 10 g/l, fructose 5g/l and glucose 5g/l.

Notes: This method is suitable for samples containing low concentrations of glucose and sucrose.

SO2L – FREE SULFUR DIOXIDE – SQPE056384 – 2 x 100 ml

The concentration of SO2 found in wines depends on the ability of the yeast to produce SO2 during alcoholic fermentation, at most 10 mg/l, and to a greater extent on the quantity of SO2 that is added during the various stages of vinification. This additive is regulated by legal limits: 200 mg/l of total SO2 for white wines and 150 mg/l of total SO2 for red wines.
Sulfur dioxide is found in wine in different forms: molecular, antiseptic and antioxidasic action, free, antioxidant and antimicrobial action, combined, bound with acetaldehyde, with anthocyanins and/or with ketone acids. The different fractions depend on the composition of the wine, the temperature, the pH and the winemaking techniques.
Free SO2 is monitored especially during aging to maintain values between 30 and 40 mg/l, also according to how malolactic fermentation is carried out.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
SO2L R1 (SO2LR)	2 x 80	small
SO2L R2 (SO2LB)	1 x 40	small
SO2L R3	1 x 5	0.25 parts in SO2LB

Both reagents must be prepared as follows: SO2LR = 8 parts of R1 + 2 parts of R2 + 0.25 parts of distilled H2O; SO2LB = 8 parts of R1 + 2 parts of R2 + 0.25 parts of R3. The two reagents remain effective for 24 hours from the time of preparation.

Method principle and characteristics

The free sulfur dioxide in the sample reacts selectively with a chromogenic hydrochloride agent, developing a violet colored compound characterized by an absorption peak at 578 nm. Therefore the quantity of colored compound that is formed is proportional to the free SO2 present in the sample.
Linearity with total V / sample V =74: up to 100 mg/l
Precision: CV% < 2 in white wine; CV% < 3 in red wine
Preparation of a standard of 80 mg/l of free sulfur dioxide: weigh 1.23g of sodium metabisulfite and put it in a 100ml volumetric flask. Add 40ml of distilled water, and shake until completely dissolved. Bring to volume with distilled water. Take 1 ml from this solution, put it in another 100 ml volumetric flask, and bring to volume with distilled water.

SO2T –TOTAL SULFUR DIOXIDE – SQPE060413 – 5 x 20 ml

The concentration of SO2 found in wines depends on the ability of the yeast to produce SO2 during alcoholic fermentation, at most 10 mg/l, and to a greater extent on the quantity of SO2 that is added during the various stages of vinification. This additive is regulated by legal limits: 200 mg/l of total SO2 for white wines and 150 mg/l of total SO2 for red wines.
Sulfur dioxide is found in wine in different forms: molecular, antiseptic and antioxidasic action, free, antioxidant and antimicrobial action, combined, bound with acetaldehyde, with anthocyanins and/or with ketone acids. The different fractions depend on the composition of the wine, the temperature, the pH and the winemaking techniques.
The total SO2 is the sum of the different fractions present in the wine and is considered in reference to the legal limits. If a high total SO2 value is required to obtain effective free values, this indicates that a large part of the sulfur dioxide has combined with specific compounds of the wine.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
SO2T R1 (SO2T1)	2 x 80	small
SO2T R2 (SO2T2)	1 x 40	small
SO2T BL	1 x 5	do not use

Method principle and characteristics

The total sulfur dioxide in the sample reacts selectively with a chromogenic agent having a disulfide bridge, giving rise to a yellow colored compound characterized by an absorption peak at 420 nm. The intensity of the color formed is proportional to the amount of total sulfur dioxide present in the sample.
Linearity with total V / sample V =50: up to 300 mg/l
Precision: CV% < 1 in white wine; CV% < 2 in red wine
Preparation of a standard of 160 mg/l of free sulfur dioxide: weigh 1.23g of sodium metabisulfite and put it in a 100ml volumetric flask. Add 40ml of distilled water, and shake until completely dissolved. Bring to volume with distilled water. Take 1 ml from this solution, put it in a 50 ml volumetric flask, and bring to volume with distilled water.

TARTAR – TARTARIC ACID – SQPE070208 – 179 ml

Grapes are one of the few fruits with high concentrations of tartaric acid (5 - 15 g/l); together with malic and citric acid, tartaric acid contributes predominantly to the formation of titratable acidity. Tartaric acid tends to decrease less as the fruit matures, while in wine it decreases as a result of precipitation together with potassium (potassium bitartrate). This aspect can decrease the hardness of wines, especially reds.
If this acid is attacked by aerobic lactic bacteria, it brings about the formation of lactic and acetic acid, resulting in turbidity and unpleasant flavor. Good cellar cleanliness, anaerobiosis and the use of SO2 are generally sufficient to avoid this phenomenon.

COMPONENTS SUPPLIED (ID abbrev.)	*V(ml)*	BOTTLE-REAGENT
TART R1 (TART1)	1 x 136	large

TART R2 (TART2)	1 x 34	small
TART BL	2 x 85	do not use

Method principle and characteristics

Tartaric acid reacts with vanadic acid in an acid medium to develop a colored complex, the concentration of which is directly proportional to the amount of tartaric acid in the sample. The absorbance is measured at 492 nm.
Linearity with total V / sample V =22: up to 6 g/l
Precision: CV% < 1 in white wine; CV% < 1.5 in red wine
Calibration and control: standard SQPE054994 tartaric acid 5 g/l.

Notes: to improve the performance of the analysis, it is recommended that structured red wines be decolorized with total polyphenols > 2500 mg/l. In addition, the result found should be added to +0.14 (HM).