

Review

# A review of the analytical methods to determine organic acids in grape juices and wines

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## Abstract

The determination of low molecular weight organic acids in grape juices and wines is important because of their influence in organoleptic properties (flavour, colour and aroma) and in the stability and microbiologic control of these beverages. The analysis of these acids allows to check the process of maturation of grapes and to control the evolution of the acidity of wines during the several stages of their elaboration process (alcoholic fermentation, malolactic fermentation, aging process, etc).

In this work, a comprehensive review of the analytical methods developed for the determination of short-chain organic acids in grape juices and wines has been shown. This review included non-enzymatic spectrophotometric, enzymatic, chromatographic and electrophoretic methods. Details of the chromatographic and electrophoretic conditions of these methods have been collected in tables.

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## Contents

1. Introduction . . . . .	1176
2. Spectrophotometric methods . . . . .	1176
3. Enzymatic methods . . . . .	1176
4. Chromatographic methods . . . . .	1177
4.1. Thin layer chromatography (TLC) . . . . .	1177
4.2. Gas chromatography . . . . .	1177
4.3. Liquid chromatography . . . . .	1177
4.3.1. High performance liquid chromatography . . . . .	1177
4.3.2. Ion chromatography . . . . .	1181
5. Electrophoretic methods . . . . .	1181
References . . . . .	1186

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## 1. Introduction

Low molecular weight organic acids are an important group of compounds in grape juices and wines since their influence in some respects like organoleptic properties (flavour, colour, and aroma) or in the stability and microbiologic control of these beverages. These acids come directly from the grape and/or from the processes which are subjected, as alcoholic fermentation, malolactic fermentation, oxidation of the ethanol, etc. (Peynaud, 1999).

In grape juice, tartaric and malic acids are the predominant organic acids and succinic and citric acids are present in minor proportion. In the case of wine, a common differentiation is made between acids which come directly from the grape (tartaric, malic and citric acids) and those that are originated, fundamentally, in the fermentation process (succinic, lactic and acetic acids). Also, there are small amounts of other acids like galacturonic, glucuronic, citramalic, dimethylglyceric, pyruvic, ketoglutaric, etc. (Berlitz & Grosch, 1992; Peynaud, 1999).

The evolution of tartaric and malic acids in grapes are useful for checking their processes of maturation (Lamikanra, Inyang, & Leong, 1995). In the case of wines, the analysis of organic acids allows to control the evolution of the acidity during the different steps of the winemaking process (alcoholic fermentation, malolactic fermentation, aging process, etc). The level of tartaric acid is a parameter of critical control in the stabilization of wine. These organic acids also have great importance in the detection of wine alterations and/or illnesses, because they suppose a modification of acids content. For example, some wine alterations are related to increase in the levels of acetic and lactic acids (acetic or lactic sharpness, respectively).

For all these reasons, several methods have been developed for identifying and quantifying these organic acids in grape juices and wines, so much individually (non-enzymatic spectrophotometric and enzymatic methods) or as a group of them simultaneously (chromatographic and electrophoretic methods).

The aim of this work has been to carry out a revision of methods for the determination of the more thoroughly organic acids in grape juices and wines and to make easy the approach for solving one specific problem.

## 2. Spectrophotometric methods

Spectrophotometric methods are based on the reaction of the organic acid with a certain substance, giving rise to the formation of a compound or coloured complex that is measured at a certain wavelength. In order to avoid interferences, organic acids are isolated by precipitation, ionic exchange resins, etc.

Rebelein (1961) determined tartaric, malic and lactic acids with a spectrophotometric method. These organic acids were separated by means of a strongly basic ion exchange resin. The portions of the eluent underwent to several treatments in order to form the coloured compounds that were measured at several wavelengths (490 nm for tartaric acid, 420 nm for malic acid and 530 or 570 nm in the case of lactic acid).

Other methods for the determination of tartaric acid have been developed having the grounds of the described method by Rebelein (1961). Basically, these methods differ in the way of clarifying the samples in order to avoid the interferences owed to the colour of wine and, also, in the automation of the process (Vereda, García de, Torres, Rivero, & Cano, 1998).

These methods are tedious and there are no data about the determination of important acids as succinic, citric or acetic acids.

## 3. Enzymatic methods

Enzymatic methods have been used, mainly, for the quantification of malic, lactic and citric acids in grape juices and wines. However, it would be possible the determination of other acids like tartaric, acetic, L-ascorbic, dehydroascorbic/L-ascorbic, formic, D-gluconic/D-glucono- $\delta$ -lactone, D-isocitric, oxalic and succinic acids (Bergmeyer, 1985; Boehringer Mannheim GmbH, 1995). In the case of wines, the method for the determination of citric acid is a Final Action method in the AOAC (Association of Official Analytical Chemists, 2000).

These methods are based on the measurement of the increase or decrease in absorbance of the coenzymes NADH (nicotinamide-adenine dinucleotide, reduced form) or NADPH (nicotinamide-adenine dinucleotide phosphate, reduced form) which absorb in the distance wavelength region (Boehringer Mannheim GmbH, 1995). The measurement was usually made with a spectrophotometer at 340 nm.

The main advantage of these methods is the high specificity because the determination of L- and D-isomers of some acids is possible and easy when enzymatic methods were used. Nevertheless, only one organic acid is determined in each assay, so this is the main disadvantage of these methods together time-consuming.

A way of these methods automatization, in order to reduce their cost and analysis time, consists on the use of flow injection analysis (FIA). Puchades, Herrero, Maquieira, and Atienza (1991) determined L-malic and L-lactic acids simultaneously in wine samples by FIA with tubular open reactors with enzymes immobilized in them. Lima and Rangel (1992) also determined these two acids in wine by FIA with spectrophotometric detection. In this case, the flow injection system had

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