

Assessing wine quality with IC

Organic acid analysis using suppressed conductivity detection

Summary

Nature and concentration of organic acids are important parameters in enology. They affect organoleptic properties (color, flavor, and aroma), wine stability, and help to track alteration processes and the wine's authenticity [1]. Tartaric acid and malic acid represent the greatest fraction of organic acids in wine, coming from the grapes themselves. Free tartaric acid decreases during wine storage when it binds to other components and precipitates, and malic acid can metabolize to lactic acid. Other organic acids are formed as products during alcoholic fermentation [1] influencing flavor. Acetic acid for example causes an undesirable vinegar taste. Overall, monitoring of organic acids is crucial to improve flavor and quality, and to fulfill universal standardized criteria such as the International Code of Oenological Practices [2]. Analytically, organic acids can be properly determined with ion chromatography (IC) and suppressed conductivity detection. As a multicomponent method, inorganic acids can also be resolved which are also valuable tracers for wine quality and taste. This Application Note presents two IC methods for wine quality analysis: a fast isocratic screening method of major organic acids and anions including sulfite, and a complex monitoring method with a binary gradient to separate 15 organic acids. Inline Ultrafiltration was used for economical sample treatment.

Configuration



2.940.1440 - 940 Professional IC Vario ONE/SeS/HPG

The 940 Professional IC Vario ONE/SeS/HPG is the intelligent IC instrument with sequential suppression and binary high-pressure gradient. It can be extended with the 942 Extension Modules to up to a quaternary gradient system. An 800 Dosino can be used for the regeneration of the suppressor. The instrument can be used with any separation and detection methods. Typical areas of application: Gradient applications for anion or cation determinations with sequential suppression;



2.850.9010 - IC Conductivity Detector

Compact and intelligent high performance conductivity detector for intelligent IC instruments. Outstanding temperature stability, the complete signal processing within the protected detector block and the latest generation of DSP – Digital Signal Processing – guarantee the highest precision of the measurement. No change of measuring ranges (not even automatic ones) is required, due to the dynamic working range.



6.1006.630 - Metrosep A Supp 7 - 250/4.0

Disinfection byproducts from water treatment are suspected not only of being health hazards but even of being carcinogenic. Oxyhalides have therefore become the subject of many investigations and standards (e.g., EPA 300.1 Part B, EPA 317.0, EPA 326.0). Of primary concern is bromate, which forms from bromide during the ozonization of drinking water. The Metrosep A Supp 7 - 250/4.0 is a high-performance separation column for the parallel determination of standard anions, oxohalides and dichloroacetic acid. With this column, these ions are determined with certainty and precision down to the lower $\mu\text{g/L}$ range. The high detection sensitivity is achieved through the use of the $5\ \mu\text{m}$ polyvinyl alcohol polymer, with which extremely high plate numbers and thus outstanding separation and detection properties are achieved. In addition, the separation can be adapted to the specific requirements of the application by modifying the temperature.



2.858.0020 - 858 Professional Sample Processor – Pump

The 858 Professional Sample Processor – Pump processes samples from 500 µL to 500 mL. The sample transfer takes place either with the installed bidirectional two-channel peristaltic pump or with an 800 Dosino.



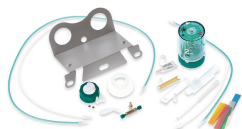
2.800.0010 - 800 Dosino

Drive with write/read hardware for intelligent Dosing Units. With fixed cable (length 150 cm).



6.2842.000 - MSM-HC Rotor A

Suppressor rotor for all IC instruments with MSM-HC (Metrohm Suppressor Module with high capacity)



6.5330.190 - IC equipment: Dosino regeneration

Accessory set for the assembly of a Dosino for automatic regeneration of the Metrohm Suppressor Module (MSM).

Sample and sample preparation

Samples of red and white wine were diluted (10–50-fold) in ultrapure water (UPW). To minimize oxidation, vials were capped with polyester lids. Using Inline Ultrafiltration, samples were automatically filtered through a 0.22 µm (regenerated cellulose) membrane prior to injection.

Experimental

Since all organic acids easily ionize, their conjugate bases can be analyzed by ion chromatography with suppressed conductivity detection.

For **fast screening analysis** the chromatographic separation was performed on A Metrosep A Supp 10 column with isocratic elution (**Figure 1**). Within less than 20 minutes the organic acids acetate, malate, tartrate, oxalate and the anions chloride, phosphate, sulfite, and sulfate are separated. Sulfite was calibrated separately to avoid a potential sulfate contamination. It was stabilized with 2-propanol (2% in working standard solutions). Although sulfite can be determined within in this multi-component run, for dedicated sulfite analysis please refer to the described methods [3].

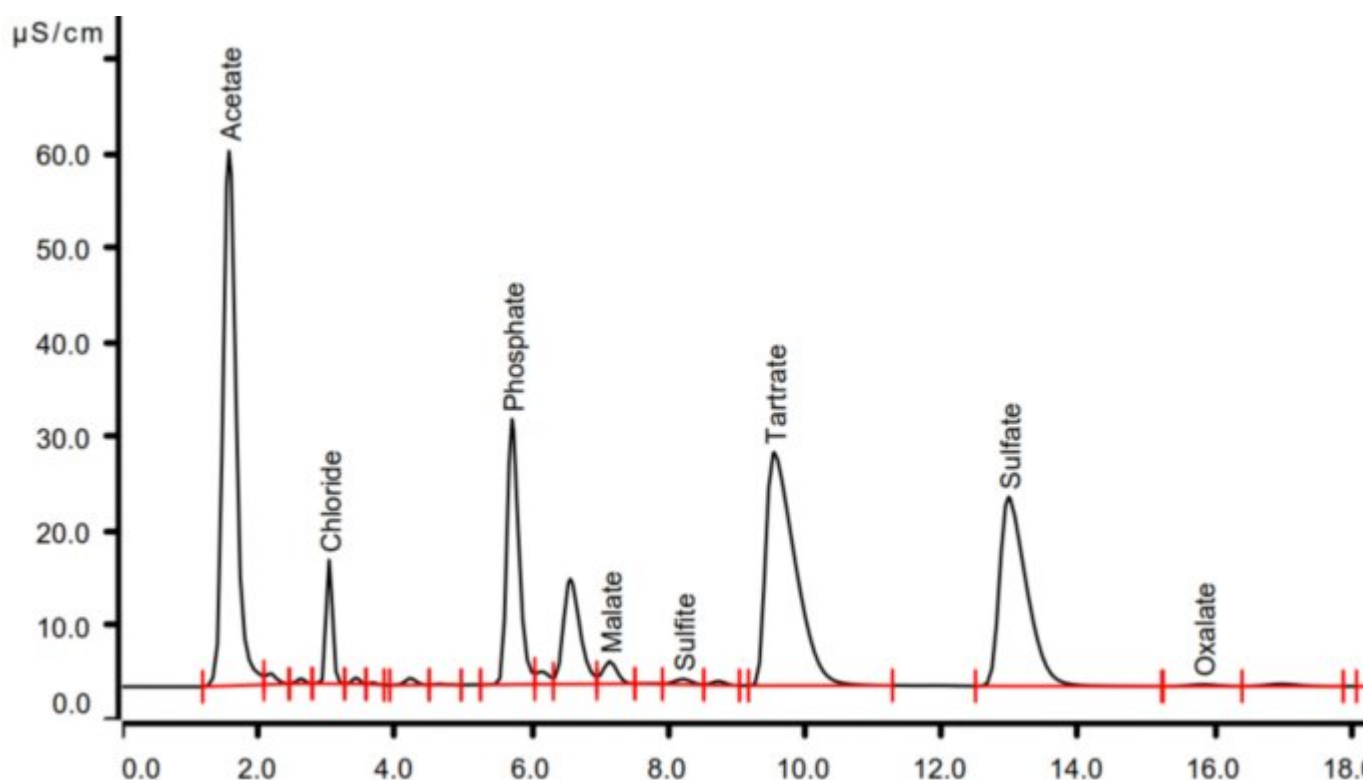


Figure 1. Fast screening analysis of major organic acids (acetate (not quantified), malate (105 mg/L), tartrate (1534 mg/L) and oxalate (<10 mg/L)) and major anions (chloride (22 mg/L), phosphate (818 mg/L), sulfite (29 mg/L), and sulfate (367 mg/L)) in a white wine sample (injection volume 20 μL). Isocratic elution was performed on a Metrosep A Supp 10 - 100/4.0 column using a carbonate eluent. (5.0 mmol/L Na_2CO_3 + 5.0 mmol/L NaHCO_3 + 5 mol/L HClO_4 , flow rate 1 mL/min, column temperature 35 $^\circ\text{C}$). Suppressed conductivity detection enables detection with a low background for detection in the lower mg/L range.

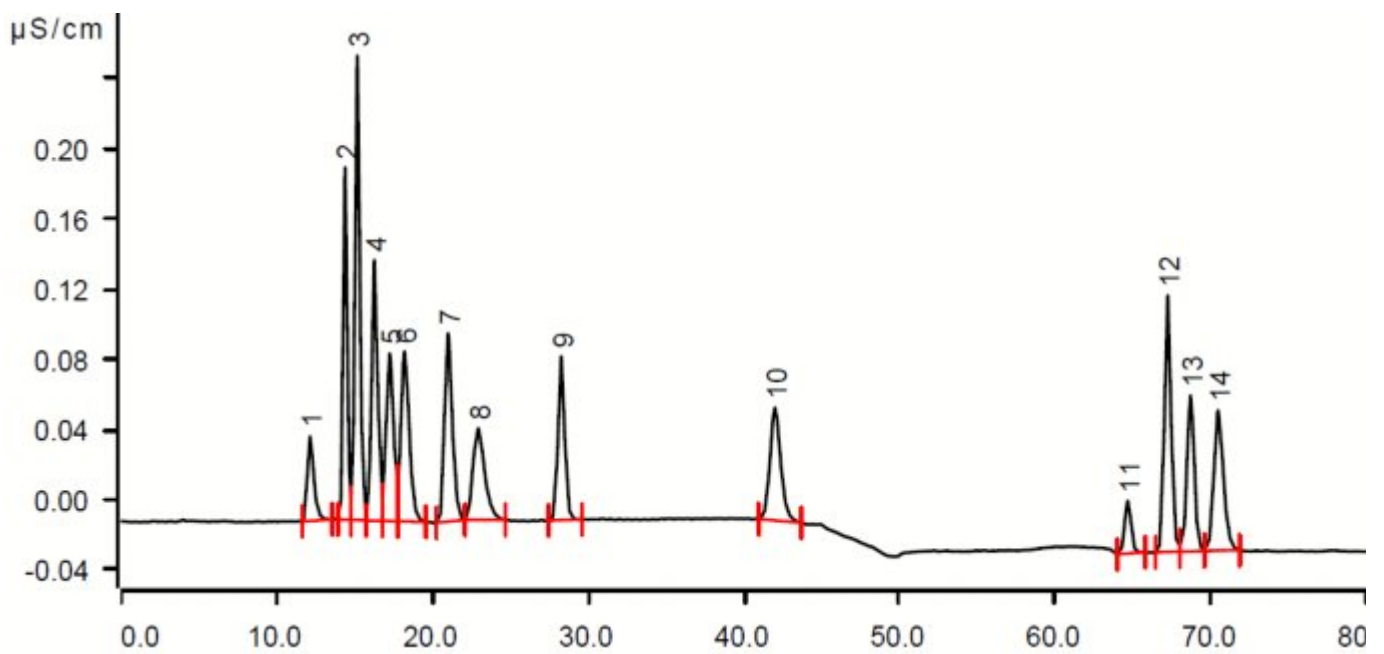


Figure 2. The figure shows the suppressed conductivity signal for the complex organic acid monitoring of gluconate (1), lactate (2), acetate (3), propionate (4), iso-butyrate (5), butyrate (6), methacrylate (7), valerate (8), methyl sulfate (9), dichloroacetate (10), malonate (11), malate (12), glutarate (13), adipate (14), and phthalate (15) in a 1 mg/L mixed standard (injection volume 20 L). Separation was on a Metrosep A Supp 7 - 250/4.0 column with a binary gradient (eluent A: ultrapure water, eluent B: 6.4 mmol/L Na₂CO₃ + 2.0 mmol/L NaHCO₃, flow rate 0.7 mL/min, column temperature 45 °C).

A comprehensive view of the organic acid composition for **complex monitoring** can be obtained by separation with a Metrosep A Supp 7 column using a binary gradient (**Figure 2**). With the carbonate-UPW gradient the following 15 organic acids could be resolved: gluconate, lactate, acetate, propionate, isobutyrate, butyrate, methacrylate, valerate, methyl sulfate, dichloroacetate, malonate, malate, glutarate, adipate, and phthalate.

The experimental setup is shown in **Figure 3**.

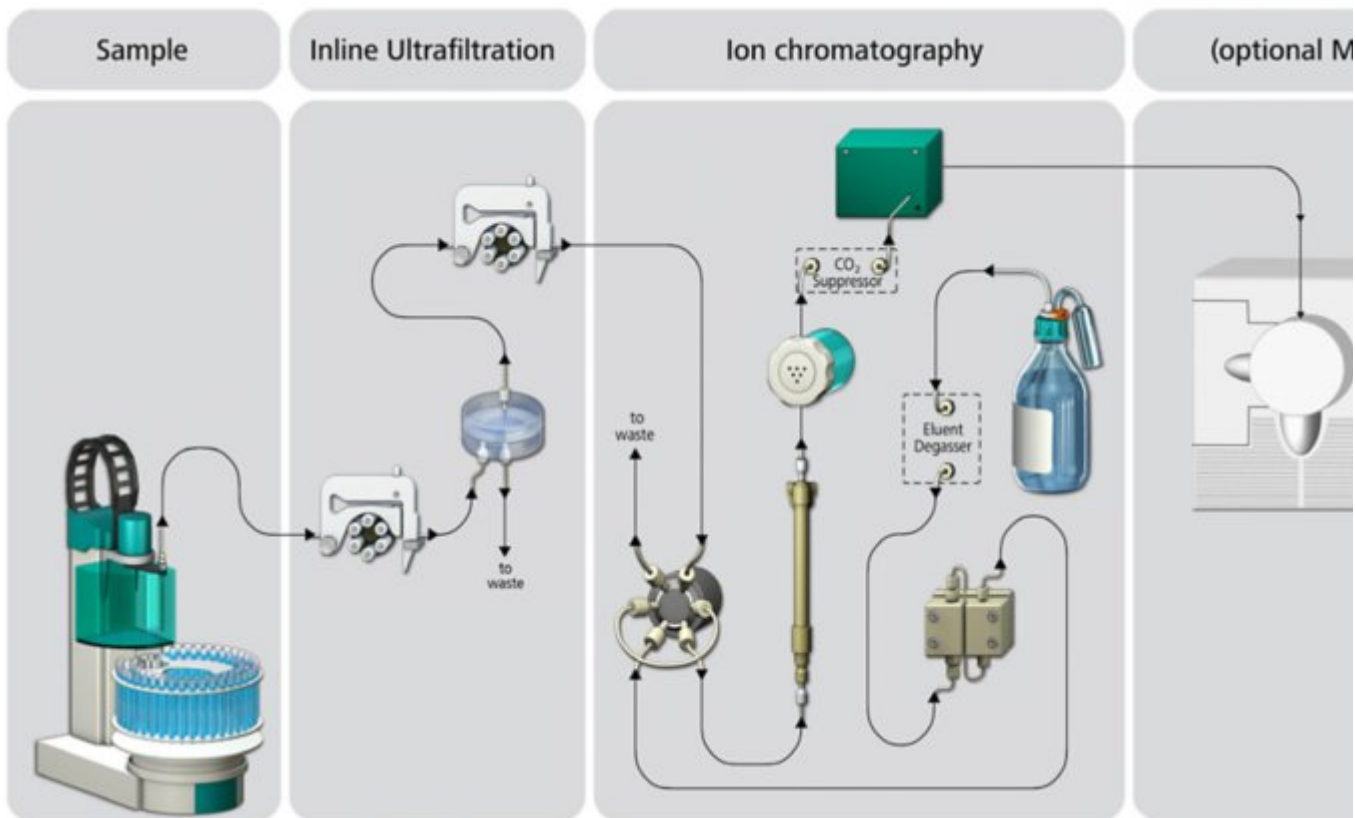


Figure 3. Schematic flow path for fast screening analysis of organic acids and anions with ion chromatography and suppressed conductivity detection. As sample preparation step, Inline Ultrafiltration is used to optimize the overall analysis in terms of laboratory time and laboratory expenses. After introduction of the sample (858 Professional Sample Processor) the sample passes the Ultrafiltration cell. The samples are filtered with a 0.2 m regenerated cellulose membrane. Up to 100 samples depending on the matrix can be analyzed before changing the membrane and with less than 0.1% carryover speeding up this unavoidable process in routine analysis. After injection and separation with a high capacity anion column sequential suppression removes cations and carbonate resulting in a very low background signal in the conductivity detector. For the complex organic acid monitoring a second high pressure pump and a mixing capillary need to be added to the system. Connection of the outlet of the conductivity detector to a mass spectrometer can be a valuable addition for peak confirmation and even better detection limits.

Results

The **fast screening analysis** of organic acids and anions took less than 20 minutes. Tartrate was the major organic acid in both samples and phosphate and sulfate the dominant anions, with slightly lower contents in white wine for tartrate and sulfate (**Table 1**). Triplicate injections showed a relative standard deviation of less than 2% for both the white wine and the red wine (**Table 1**).

Table 1. Organic acids and anions quantified in a red and white wine sample. Sample dilution was performed in UPW with a dilution factor of 10 (50 for tartrate). Samples were analyzed with the fast screening analysis resolving major organic acids and anions in wine samples.

Analyte	Red wine (mg/L) (RSD)	White wine (mg/L) (RSD)
Chloride	60 (0.03%)	22 (0.04%)
Phosphate	771 (0.2%)	818 (0.1%)
Malate	92 (0.1%)	105 (0.2%)
Sulfite	27 (2%)	29 (0.4%)
Tartrate	1756 (0.1%)	1534 (0.6%)
Sulfate	553 (0.01%)	367 (0.01%)
Oxalate	< 10	< 10

A gradient elution improved peak resolution for a complex monitoring analysis of 15 organic acids. Suppressed conductivity detection enabled a sensitive detection in a working range of 0.1 to 5 mg/L.

Both methods show excellent performance in the lower mg/L range. Detection of the suppressed conductivity signal omits interferences from UV-active components seen with UV-detection. Sample preparation with **Inline Ultrafiltration** makes this unavoidable (generally manual) step both time and cost effective while also guaranteeing column protection.

Conclusion

Ionic composition profiles in wines are readily quantified with IC and conductivity detection. Ion exchange chromatography allows the simultaneous determination of inorganic anions and

organic acids in one run, in contrast to ion-exclusion which only separates organic acids. With the **fast multi-component screening analysis** sample throughput in laboratories can be maximized. Sample preparation can be facilitated with Inline Ultrafiltration, protecting the column and enhancing instrument performance. Further increase of the economic potential can be achieved by combination with Metrohm Inline Dilution including the possibility of automatic calibration. The manual error-prone dilution step of samples and standards is omitted while laboratory time is saved, and accuracy and precision improved.

The **complex organic acid monitoring** with suppressed conductivity detection benefits from higher sensitivity compared to UV-detection methods and reduced interferences from UV-active sugars and phenols in such wine samples.

If peak identity needs confirmation, or very low detection limits are required, the IC setup can be combined with a sensitive mass specific detector (**Figure 3**).

References

1. Waterhouse et al. (2016), John Wiley & Sons, UK, ISBN 1118627806
2. International Organization of Vine and Wine (OIV) (2021), OIV, France, ISBN 978-2-85038-030-3
3. Metrohm, WP-065 Simplified sulfite determination in foods and beverages using ion chromatography

Metrohm AG

*Ionenstrasse
9100 Herisau*

<mailto:info@metrohm.com>