

Organic Acid Analysis Using Capillary Ion Analysis



Capillary Electrophoresis
Application Brief EBI 11/91

Breakthrough in Organic Acid Analysis.

CIA™, or Capillary Ion Analysis, brings distinct and productivity-boosting benefits to the analysis of beverages.

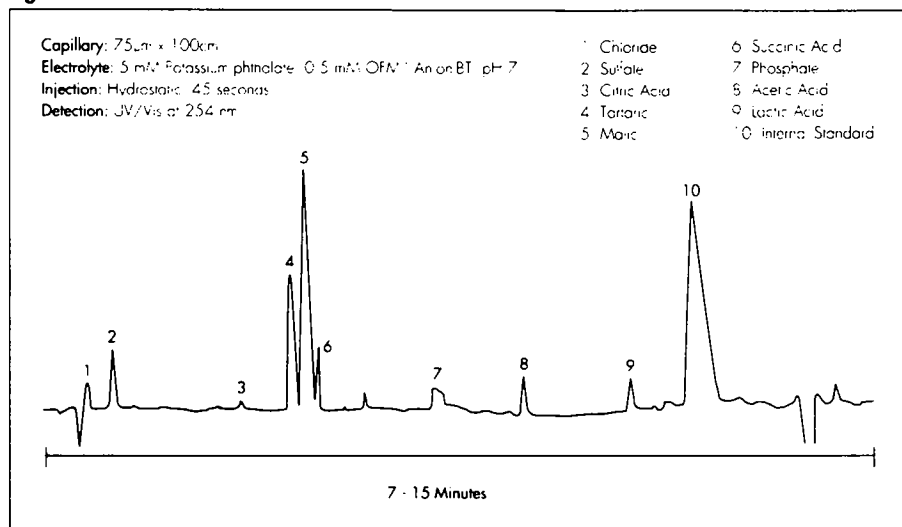
Enzyme analysis and HPLC have traditionally been used to determine authenticity and organoleptic qualities as well as to monitor fermentation and stability. But these methods suffer from drawbacks, such as long sample preparation, lack of specificity, and interfering compounds. Some analyses for existing methods are also either expensive or aren't rugged enough for routine use. CIA can accomplish all these tests and more in less time, for less money, and with more reliability.

Simple, Fast Analysis.

To test the technique, a Chablis wine sample was analyzed on the Waters Quanta® 4000 Capillary Electrophoresis System. Sample preparation was minimal – the sample was simply diluted in a 1/50 ratio in water and an internal standard was added. Other sample preparation procedures are eliminated by the automatic purge feature of the Quanta 4000 system.

The sample was injected via hydrostatic mode and a 75µm x 100cm capillary was used. The electrolyte components were potassium phthalate and OFM™ Anion-BT. The latter reagent is part of Waters proprietary NICE-Pak™* chemistry kit. Detection

Figure 1.



This 17 minute CIA separation resolved ten compounds at a cost of 50 cents.

was at 254 nm. All data was collected on Waters 820 Chromatography Workstation.

As you can see in Figure 1, the Quanta 4000 system and the proprietary chemistries resolved the organic acids and inorganic ions in 17 minutes. This includes a two minute automatic purge. Due to unique selectivities and the autopurge before each run, there are no sugar or phenolic interferences, therefore making results easy to interpret.

Reliable Results.

To test the quantitative value of this analysis, ten aliquots of the wine were analyzed. Two components of interest, malic and tartaric acids, proved to have a reproducibility of better than 2% RSD over the repetitive runs.

100% Recovery.

In order to ensure the accuracy of our methods, the recovery of the malic and tartaric acids was tested. The acids were mixed with a grape juice sample at different levels. As shown in Figure 2, overall recovery was 100% for both.

* Patent pending.

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Exceptional Linearity.

After vigorous testing this method proved to be linear over a broad range of concentrations. The lactic acid component of the sample illustrates this point. To determine linearity, the area and height response was compared between 10 and 167 $\mu\text{g/ml}$. As shown in Figure 3, the area response demonstrated a r^2 value of 0.999. The 10 to 167 range reflects the levels of these acids in an actual, diluted sample.

Cost Efficient.

Due to minimal reagent costs, simple sample preparation, and inexpensive capillaries, this method is extremely cost efficient on a cost-per-test basis. As compared to HPLC (which costs about \$2.70 per test) or enzyme analysis (which costs about \$12 per test), CIA averages a mere 50¢ per test.

Replaces Conventional Methods.

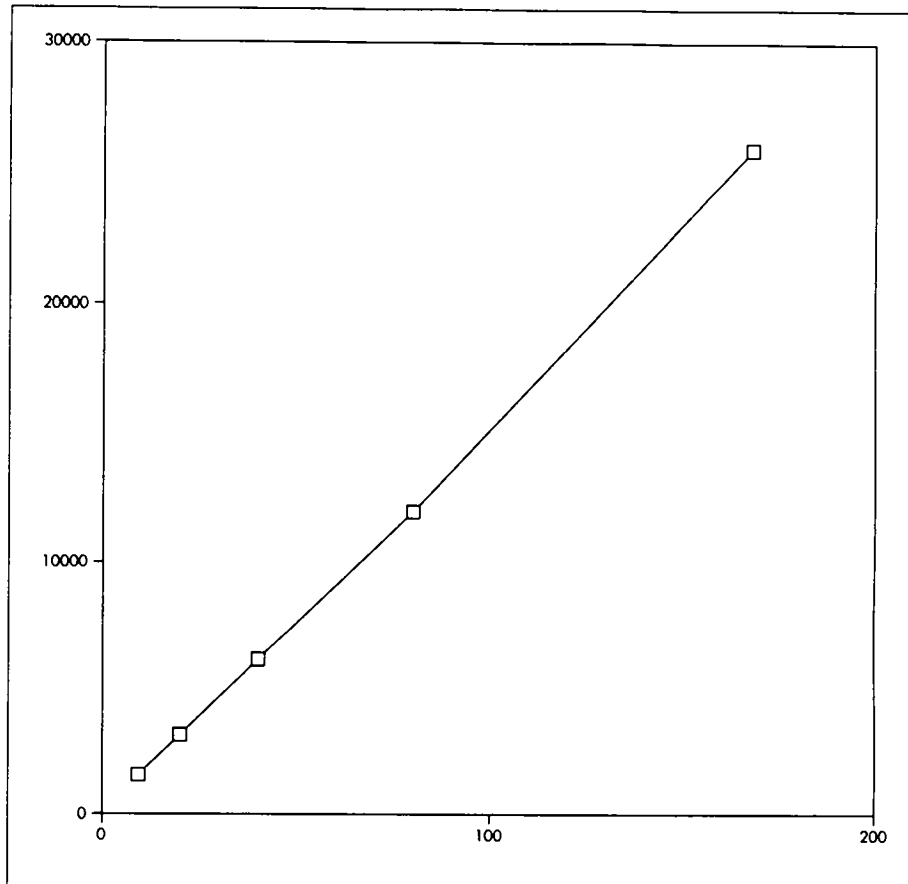
Capillary Ion Analysis significantly reduces the time and cost of beverage organic acid analysis. It is quantitative, linear, reproducible, and offers good recoveries. It can be used for a wide variety of samples, including coffee, apple, and citrus, as well as fermented beverages, with little or no sample preparation.

Figure 2: Recovery of the Organic Acid Methods on the Quanta 4000.

Analyte	$\mu\text{g/ml}$	$\mu\text{g/ml}$	% Recovery
Tartaric	0	4330	
Tartaric	1160	5525	103
Tartaric	2319	6703	102
Malic	0	3867	
Malic	1289	5153	100
Malic	2577	6283	94

To ensure the accuracy of the methods, the recovery of two components of interest was tested in dark grape juice. Average recovery was 100%.

Figure 3.



The linearity of the method was tested using the lactic acid component of the wine sample. As shown, the test on the Quanta 4000 system resulted in a 0.999 linearity.

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