Determination of low citric acid concentrations in a mixture of weak acid/bases

Ori Lahav*, Emily Shlafman and Malka Cochva
Faculty of Civil and Environmental Engineering, Technion – Israel Institute of Technology, Haifa, 32000, Israel

Abstract

A titration approach was developed to measure low concentrations of citric acid ($C_6H_8O_7$) in a mixture of other weak acid/bases. Two methods were tested. The first and more practical method (a 4-point titration procedure) is applicable in conditions where volatile fatty acids (VFAs) are not normally present. The second method (a 5-point titration procedure) was developed for anaerobic environments where VFAs may be encountered. Generally, fairly accurate and repetitive results (precision >95%) were obtained for both situations although stability and accuracy were better in the absence of VFA. Both methods can be used for routine monitoring of biological reactors where citric acid is added as a carbon source and electron donor. Mg^{2+} and Ca^{2+} form complexes with citric acid and thus inhibit the use of the method. To overcome this, a sodium-saturated cationic ion exchanger was used to exchange these cations with Na^{+}. Following cation exchange, citric acid concentration was determined accurately using the method.

Keywords: citric acid measurement, acid titration, mixed weak-acid system, VFA

Introduction

Citric acid ($C_6H_8O_7$) is a natural component and common metabolite of plants and animals. It is the most versatile and widely used organic acid in foods, beverages, detergents and pharmaceuticals. Because of its functionality and environmental acceptability it is used in numerous industrial and research applications for chelation, buffering, pH adjustment, and also as a source of energy for controlled bacterial metabolism. The latter application is widely used in scientific research for the cultivation of both aerobic and anaerobic bacteria (Herzberg et al., 2003; Yoo et al., 2004). Often in these applications, process control depends on routine determination of the citric acid concentration.

Current determination of high concentrations of citric acid in the absence of "interfering" substances (other hydroxide-accepting species) is normally carried out by strong base titration (typically NaOH) to an endpoint pH of around 7.0 (*Standard Methods*, 1998). Low concentrations are currently determined by a spectrophotometric method based on a reaction with pyridine and acetic anhydrine (Hartford, 1962). Also, an enzymatic (Taraborelli and Upton, 1975) and HPLC methods (Guerrand, 1982) are in use. More recently, other methods based on ion chromatography (Saccani et al., 1995), gas chromatography (Chepurnoi and Bolbat, 1996), and fluorescence spectroscopy (Yedur and Berglung, 1996) have been proposed. All existing methods for determination of low citric acid concentrations lack simplicity, and some require relatively expensive equipment, and trained operators.

The work presented here was aimed at developing an accurate, simple, inexpensive, and rapid titration procedure for determination of relatively low citric acid concentrations (approximated range: 20 mg/ℓ to several hundred mg/ℓ) in a mixture of various weak acid species, primarily the carbonate system. The method was developed for two distinct situations. First, a mathematical algorithm and a matching titration procedure were developed for cases where the sole unknowns are the citric acid concentration and the total inorganic carbon concentration. This situation represents an aerobic environment in which volatile fatty acids (VFA) are normally not present, and the concentration of the other weak acid species (ammonia, phosphate, sulphide, etc.) is known. Second, the model is extended to solve a third unknown – the VFA concentration. The extended procedure can be used in the context of wastewater anaerobic reactors where VFA concentrations are normally encountered.

It is expected that the method will find use in bioreactor research where citrate is a popular carbon source for bacterial growth. The proposed method is not intended as a very accurate tool for citric acid determination, but rather as a fast and easy means of determining the concentration within an accuracy of ± 5 to 10 mg/ℓ. Such accuracy is in many cases sufficient in bioreactor operation practice.

In the following sections the equations comprising the mathematical algorithms for aerobic (VFA absent) and anaerobic (VFA present) conditions are described, and the corresponding titration procedures are developed and tested.

Model derivation

Apart from the carbonate system that is invariably present, other weak acid systems that affect the titration curve, and are thus considered in the "aerobic" model, are the phosphorus and the sulphide systems. In anaerobic environments, the volatile fatty acids (VFA) systems, that for practical purposes are simulated together by the acetic acid system (pK values of the acetic, propionic and butyric acids are all close to 4.75) are added. The ammonia system is not considered in the model because it has no buffering capacity at the proposed titration range.

A by-product of the model is the accurate determination of carbonate alkalinity, and also the VFA concentration in cases where it is present, the most important example being anaerobic biological reactors.

* To whom all correspondence should be addressed.
☎ +72 4 8292191; fax: +72 4 8295696; e-mail: agori@tx.technion.ac.il
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The basic model approach involves equating a mass balance relationship for alkalinity in terms of the volume of standard strong acid titrant added (Eq. (1)) to a mass balance of alkalinity in terms of the concentration of all proton-accepting species likely to be present in biological reactors (Eq. (2)). The approach follows the technique described by Moosbrugger et al. (1993), Lahav et al. (2002) and Lahav and Morgan (2004). Such a general approach includes the ammonia subsystem that is discarded later on. In cases where another weak acid system is present in significant concentrations, its proton accepting species concentration should be added to the right side of Eq. (2) in the same manner.

$$M_{\text{total alk (x)}} = V_e \cdot C_a - V_x \cdot C_a \quad (1)$$

where:

$M_{\text{total alk (x)}}$ = total mass of alkalinity after the addition of V_x mℓ of standard strong acid (mol)

V_e = the unknown volume of standard strong acid to be added to the alkalimetric end-point (ℓ)

V_x = the volume of standard strong acid added to a point x with pH equal to pH_x (ℓ)

C_a = concentration of standard strong acid titrant (mol/ℓ).

$$M_{\text{total alk (x)}} = \{[C_6H_7O_7^-]_x + 2[C_6H_6O_7^{2-}]_x + 3[C_6H_5O_7^{3-}]_x + 2[CO_3^{2-}]_x + [HCO_3^-]_x + [A^-]_x + [HS^-]_x + 2[S^{2-}]_x + [NH_3]_x + 3[PO_4^{3-}]_x + 2[HPO_4^{2-}]_x + [H_2PO_4^-]_x + [OH^-]_x - [H^+]_x\} \cdot (V_x + V_s) \quad (2)$$

where:

$[y]_x$ indicates molar concentration of species y after addition of x mℓ of standard acid (mol/ℓ)

$[A^-]$ = dissociated short-chain VFA species concentration (mol/ℓ)

V_s = volume of sample (ℓ)

Eq. (2) can be reformulated in terms of total weak acid species concentrations using equilibrium equations for the weak acid subsystems and mass balance equations for each of the weak acids as represented in Eqs. (3) to (7) below. For brevity reasons only the citric acid, VFA, and carbonate species are given. The other subsystems follow the same approach.

For the citric acid subsystem:

$$\{H^+\}_x \cdot [C_6H_7O_7^-]_x / [C_6H_8O_7]_x = K'_{CA1} \quad (3)$$

$$\{H^+\}_x \cdot [C_6H_6O_7^{2-}]_x / [C_6H_7O_7^-]_x = K'_{CA2} \quad (4)$$

$$\{H^+\}_x \cdot [C_6H_5O_7^{3-}]_x / [C_6H_6O_7^{2-}]_x = K'_{CA3} \quad (5)$$

$$CA_T \cdot V_s/(V_x + V_s) = [C_6H_8O_7]_x + [C_6H_7O_7^-]_x + [C_6H_6O_7^{2-}]_x + [C_6H_5O_7^{3-}]_x \quad (6)$$

For the carbonate subsystem:

$$\{H^+\}_x \cdot [HCO_3^-]_x / [H_2CO_3^*]_x = K'_{C1} \quad (7)$$

$$\{H^+\}_x \cdot [CO_3^{2-}]_x / [HCO_3^-]_x = K'_{C2} \quad (8)$$

$$C_T \cdot V_s/(V_x + V_s) = [H_2CO_3^*]_x + [HCO_3^-]_x + [CO_3^{2-}]_x \quad (9)$$

For the VFA subsystem (all the VFAs are considered to constitute a single weak acid system with an equilibrium constant K'_a because they all have pK values very close to each other):

$$\{H^+\}_x \cdot [A^-]_x / [HA]_x = K'_a \quad (10)$$

$$A_T \cdot V_s/(V_x + V_s) = [HA]_x + [A^-]_x \quad (11)$$

In Eqs. (3) to (11):

{ } denotes activity, [ ] denotes molarity, K' equals apparent equilibrium constant after adjustment for activity coefficients and CA_T, C_T, and A_T denote the total concentration of the respective weak acid subsystem (citric acid, carbonate, and VFA).

Solving for CA_T from Eqs. (3) to (6), for C_T from Eqs. (7) to (9) and for A_T from Eqs.(10) and (11) respectively gives the following equations for the representation of the species included in Eq. (2) as function of pH, V_x , and the total concentration of each weak acid subsystem:

$$[C_6H_7O_7^-]_x = CA_T \cdot V_s/(V_x + V_s) / \{1 + K'_{CA2} / \{H^+\}_x + \{H^+\}_x / K'_{CA1} + K'_{CA2} \cdot K'_{CA1} / \{H^+\}^2_x\} \quad (12)$$

$$[C_6H_6O_7^{2-}]_x = CA_T \cdot V_s/(V_x + V_s) / \{1 + K'_{CA3} / \{H^+\}_x + \{H^+\}_x / K'_{CA2} + \{H^+\}^2_x / (K'_{CA2} \cdot K'_{CA1})\} \quad (13)$$

$$[C_6H_5O_7^{3-}]_x = CA_T \cdot V_s/(V_x + V_s) / \{1 + \{H^+\}^2_x / (K'_{CA2} \cdot K'_{CA3}) + \{H^+\}_x / K'_{CA3} + \{H^+\}^3_x / (K'_{CA3} \cdot K'_{CA2} \cdot K'_{CA1})\} \quad (14)$$

$$[HCO_3^-]_x = C_T \cdot V_s/(V_x + V_s) / \{1 + K'_{C2} / \{H^+\}_x + \{H^+\}_x / K'_{C1}\} \quad (15)$$

$$[CO_3^{2-}]_x = C_T \cdot V_s/(V_x + V_s) \cdot K'_{C2} / \{\{H^+\}_x + K'_{C2} + (\{H^+\}_x)^2 / K'_{C1}\} \quad (16)$$

$$[A^-]_x = A_T \cdot V_s/(V_x + V_s) \cdot K'_a / \{\{H^+\}_x + K'_a\} \quad (17)$$

Similar equations can be developed for the phosphate, sulphide and ammonium proton accepting species. Substituting the equations for each of the species concentration into Eq. (2) gives an equation for total mass of alkalinity in terms of CA_T, A_T, C_T, P_T, N_T, S_T and pH_x:

$$M_{\text{total alk (x)}} = \{CA_T \cdot V_s/(V_s+V_x) \cdot F_{n1}(pH)_x + C_T \cdot V_s/(V_s+V_x) \cdot F_{n2}(pH)_x + A_T \cdot V_s/(V_s+V_x) \cdot F_{n3}(pH)_x + P_T \cdot V_s/(V_s+V_x) \cdot F_{n4}(pH)_x + S_T \cdot V_s/(V_s+V_x) \cdot F_{n5}(pH)_x + N_T \cdot V_s/(V_s+V_x) \cdot F_{n6}(pH)_x + 10^{-(14 - pHx)}/f_m - 10^{-pHx}/f_m\} \cdot (V_s + V_x) \quad (18)$$

where:

P_T, S_T and N_T represent the total phosphate, sulphide and ammonium concentrations (mol/ℓ),

f_m = monovalent activity coefficient, and

F_{n1} to F_{n6} are functions of pH_x and equilibrium constants for the citric acid, carbonate, VFA, phosphate, sulphide and ammonium subsystems (as given, for example, in Eqs. (12) to (17)).

Equating Eqs. (1) and (18) gives the desired equation linking the mass of alkalinity based on acid added to the mass of alkalinity based on species concentrations:

$$(V_e - V_x) \cdot C_a = \{CA_T \cdot V_s/(V_s+V_x) \cdot F_{n1}(pH)_x + C_T \cdot V_s/(V_s+V_x) \cdot F_{n2}(pH)_x + A_T \cdot V_s/(V_s+V_x) \cdot F_{n3}(pH)_x + P_T \cdot V_s/(V_s+V_x) \cdot F_{n4}(pH)_x + S_T \cdot V_s/(V_s+V_x) \cdot F_{n5}(pH)_x + N_T \cdot V_s/(V_s+V_x) \cdot F_{n6}(pH)_x + 10^{-(14 - pHx)}/f_m - 10^{-pHx}/f_m\} \cdot (V_s + V_x) \quad (19)$$

At each point in the titration (i.e. for each V_x and corresponding pH_x) Eq. (19) includes 4 unknowns: V_e, CA_T, A_T and C_T,

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assuming volatile fatty acids are present, and provided that the phosphate, sulphide and ammonium concentrations are measured, and temperature and TDS (or EC) are known. Thus, to solve for V_e, A_T and C_T only 4 titration data pairs (i.e. 4 values for corresponding V_x and pH_x pairs) are required. However, Moosbrugger et al. (1993) and Lahav et al. (2002) report that arbitrary selection of titration points invariably leads to poor prediction. They found that in order to attain accurate and stable readings titration points need to be located on both sides of the relevant pK values of the unknown weak acid system. First, such approach yields stable pH readings imperative for accurate model results, and second, when the equations composed from two titration points symmetrical about a pK value are subtracted from each other, the resulting equation is independent of weak acid subsystems that do not exert buffering capacity at the pH range between the two points, a fact that tends to simplify the calculations involved.

Algorithm for calculating citric acid and carbonate alkalinity concentrations in "aerobic" environments (volatile fatty acids not present)

In aerobic environments, where VFA concentrations are typically zero, Eq. (19) contains three unknowns: V_e - the volume of acid required to reach the alkalimetric end-point (ℓ), CA_T – the total citric acid subsystem concentration (mol/ℓ), and C_T – the total carbonate subsystem concentration (mol/ℓ). In accordance with the concept of titration to a pair of pH points roughly symmetrical about the relevant pK values, it was chosen here to titrate to four pH points (i.e. two titration pairs), the first pair symmetrical about pK_{CA2} (4.72 – the second pK of the citric acid subsystem), and the second symmetrical about pK_{C1} (pH = 6.35 – the first pK of the carbonate subsystem). The second titration pair is also roughly symmetrical about the third pK of the citric acid subsystem (pK_{CA3} = 6.33). In both cases titration is carried out to roughly 0.5 pH units on both sides of the relevant pK. In addition, the first pH point is recorded for subsequent calculation of the carbonate alkalinity value.

When inserted into Eq. (19), the data from the four points result in four equations. The pair of observations around pK_{CA2} (i.e. the third and the fourth points) is in a region where the buffer capacity of the citric acid subsystem dominates over that of the carbonate subsystem (for equal concentrations of the two subsystems), whereas for the 1st and 2nd titration points both subsystems are represented approximately evenly. As a result, subtracting the equation formed from the 4th data point from that derived from the 3rd, gives an equation in terms of CA_T and C_T in which the citric acid alkalinity term dominates (only two unknowns remain - V_e is eliminated). This technique also enables a relative separation between the subsystems and thus, an error in the first two pH observations (that may arise from either CO_2 loss and/or H_2S loss, or from inaccurate P_T or S_T input) would be somewhat "absorbed" by the carbonate subsystem, reducing the effect on the citric acid determination. Another advantage of this approach is that because only the difference in proton absorbing capacity between the two points is taken into account, weak acid subsystems that do not have buffer capacity at that pH region do not affect the calculation. Accordingly, for example, the ammonia weak acid system (pK = 9.25) does not need to be considered, and its term can be removed from Eq. (19).

Subsequently, the two new equations (resulting from the subtraction of the 1st point from the 2nd and of the 3rd point from the 4th) are solved to yield CA_T and C_T. For the output of the algorithm CA_T represents the total citric acid concentration (i.e. the sum of citric acid and acetate) while C_T is used in conjunction with the initial pH (pH_o) to calculate the carbonate alkalinity using Eq. (20):

$$\text{Carbonate alkalinity} = [HCO_3^-]_{(pHo)} + 2[CO_3^{2-}]_{(pHo)} = C_T / \{ 1 + K'_{C2} / (10^{-pHo}) + (10^{-pHo}) / K'_{C1}\} + 2C_T \cdot K'_{C2} / \{(10^{-pHo}) + K'_{C2} + ((10^{-pHo}))^2 / K'_{C1}\} \quad (20)$$

Algorithm for calculating citric acid and carbonate alkalinity concentration in "anaerobic" environments (volatile fatty acids present)

In the presence of VFA the analytical problem extends to three unknowns, i.e. CA_T, C_T and the total VFA concentration, represented by the symbol A_T.

Theoretically, titration to four points gives enough information to solve a set of three equations and three unknowns. However, it was empirically found that the most accurate and stable results are attained when a 5th titration point is added, i.e. titration to around pH 3.65. This titration point is located approximately 0.5 pH units above the lower pK of the citric acid system. The motivation for the addition of yet another titration point was to further differentiate between the citric and VFA weak acid subsystems, by titrating to a pH region where the buffer capacity of the citric acid subsystem is much higher than that of the VFA subsystem.

The calculation procedure in this case is as follows: three equations are formed, the first by subtracting the equation formed by the 1st titration point from the equation formed by the 2nd titration point, the second by subtracting the equation formed by the 3rd titration point from the equation formed by the 4th titration point, and the third by subtracting the equation formed by the 4th titration point from the equation formed by the 5th titration point. Because V_e is eliminated, the outcome of this procedure is a set of three equations with three unknowns. These are now solved to yield the values of C_T, CA_T, and A_T. The values of CA_T and A_T are used as the model output for the citric acid and VFA concentrations respectively, and C_T is used in conjunction with the initial pH to calculate the carbonate alkalinity using Eq. (20).

Materials and methods

Reagents and analytical measurements

Chemicals used for the laboratory-made samples were of analytical grade (AR). $NaHCO_3$ was used to generate the carbonate alkalinity; phosphate and sulphide concentrations were made using K_2HPO_4 and Na_2S respectively. Phosphate was measured using anionic Metrohm 761 ion chromatograph equipped with a Metrosep A Supp 3 anion separating column and suppressor using a carbonate/bicarbonate eluent. Sulphide was measured by the iodide method (*Standard Methods*, 1998). Ion exchanger used for calcium and magnesium removal was Amberlite™ IRC747 distributed by Rohmhaas, Inc.

Titration procedure (for both "aerobic" and "anaerobic" samples)

***Apparatus**:* good general-purpose pH meter and electrode, auto-titrator machine, magnetic stirrer with both slow and even stirring.

***Standards**:* prepare and calibrate standard HCl and NaOH 0.05 M (NaOH is needed only if the samples are at pH lower than 6.85) as set out in *Standard Methods* (1998).

TABLE 1
Measurements of laboratory-made solutions (no VFA present)

Results of method execution				Composition of laboratory made samples			
Relative error in total alkalinity measure-ment	Relative error in citric acid measure-ment	Measured Citric acid	No. of samples Tested	Phos-phate (K_2HPO_4)	Sulphide (Na_2S)	Carbo-nate alkalinity	Citric acid
(%)	(%)	(mg/ℓ as $C_6H_8O_7$)	-	(mg/ℓ as P)	(mg/ℓ as S)	(mg/ℓ as $CaCO_3$)	(mg/ℓ as $C_6H_8O_7$)
2.16	5.4	210.8 (7.8)	3	0	0	400	200
4.12	2.2	293.2 (4.3)	7	0	0	200	300
2.77	0.5	199.1 (12.2)	8	0	0	200	200
0.92	2.7	97.3 (6.5)	8	0	0	200	100
1.17	6.1	106.1 (6.5)	8	100	0	200	100
0.87	2.7	102.7 (7.5)	7	50	0	200	100
1.97	0.9	100.9 (12.4)	7	0	30	200	100
1.67	4.8	104.8 (10.4)	9	10	10	200	100
1.05	3.0	48.5 (10.2)	12	10	10	200	50
1.53	11.6	33.5 (2.6)	7	10	10	200	30
2.47	15.5	23.1 (4.4)	7	10	10	200	20

Procedure

1. Filter the sample to remove suspended material using a 0.45 µm filter.
2. Measure EC, phosphate, and sulphide concentrations of the filtrate. For routine measurement where phosphate and/or sulphide concentrations are known or very low this step can be skipped.
3. Dilute the raw sample to a C_T value of between 150 and 350 mg/ℓ as $CaCO_3$ (to minimise both CO_2 losses, and change in C_T during titration).
4. Recommended sample volume: 50 to 100 mℓ.
5. Record the temperature.
6. Stir slowly and allow pH to stabilise and then record initial pH. For both titration procedures, in case that the initial pH of the sample is lower than the initial titration point (i.e. pH = 6.85), the initial pH is recorded and a measured volume of strong base (NaOH, 0.05 M) is added to raise the pH to above 7. For the algorithm calculations, the volume of the strong base is added to all the V_x values. There is no need to record the pH attained after the addition of the strong base, and acid titration can begin immediately following its addition.
7. Continue slow stirring and start titrating, using the standard acid, down to pH values of approximately 6.85, 5.85, 5.25, 4.25, and 3.65 (the 5th pair only in case of VFA-containing samples). There is no need to bring the pH precisely to these values – a deviation of ± 0.1 pH units is acceptable, thus titration can proceed rapidly. Record each of these pH values and the corresponding V_x. Make sure the pH reading is stable before continuing between points.
8. Enter data into Excel program (The program is built as a simple MS Excel sheet and can be either programmed by the user based on the model as set out above, or requested from the authors free of charge). Data includes the initial pH, 4 (or 5) titration pairs (V_x, pH), total phosphate and sulphide concentrations of the undiluted sample, EC of the undiluted sample, temperature of the diluted sample, and volume of NaOH added (if applicable).
9. The program gives the following output: Citric acid concentration (in mol/ℓ and mg/ℓ as $C_6H_8O_7$), VFA concentration (in mol/ℓ and mg/ℓ as CH_3COOH), carbonate alkalinity (in eq/ℓ and mg/ℓ as $CaCO_3$), and total inorganic carbon concentration (C_T) (in mol/ℓ and mg/ℓ as $CaCO_3$).

Method evaluation

Samples not containing VFA (representing aerobic environments)

The assessment of the method was performed in two steps. First, laboratory-made samples containing known concentrations of all components (i.e. C_T, CA_T and other weak acid species) were tested using the method. In these measurements Ca^{2+} and Mg^{2+} ions, which form complexes with citric acid, and thus interfere with the titration procedure, were not added to the solution. Second, citric acid aliquots were added to tap water and the sample was measured to assess the extent to which the results reflected the expected concentrations, in a sample better resembling actual bioreactor waters. In order to overcome the complexes formed by citric acid with Mg^{2+} and Ca^{2+} the solution was first passed through a column filled with a Na^+ - saturated weak cation exchange characterised by high affinity toward divalent cations. Since sodium does not form complexes with citric acid it was hypothesised that the exchange of Na^+ for Ca^{2+} and Mg^{2+} will reduce the total soluble concentration of Mg^{2+} and Ca^{2+} to a degree that complexes, if formed, would have only a minor effect on the titration procedure.

Model evaluation with laboratory-made samples

Table 1 shows the average (+ standard deviation) concentrations of citric acid calculated by the method when laboratory-made samples comprising various concentrations of CA_T, C_T, P_T and S_T were titrated. The samples simulated a variety of citric acid concentrations in solutions containing a typical C_T value of 200 mg/ℓ as $CaCO_3$. Sulphide and phosphate concentrations that simulate the upper range of these species in biological reactors were added to the samples in order to assess their affect on the accuracy of the citric acid measurement. The term "total alkalinity" (right column in Table 1) reflects the sum of all proton-accepting

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TABLE 2 Measurements of laboratory-made solutions (VFA present)							
Average (and standard deviation) results from method execution					Composition of laboratory-made samples		
Relative error in VFA measurement	Measured VFA concentration	Relative error in citric acid measurement	Measured citric acid	No. of samples tested	Total alkalinity added	Acetic acid	Citric acid
(%)	(mg/ℓ as $C_2H_4O_2$)	(%)	(mg/ℓ as $C_6H_8O_7$)	-	(mg/ℓ as $CaCO_3$)	(mg/ℓ as $C_2H_4O_2$)	(mg/ℓ as $C_6H_8O_7$)
9.0	91.0±6.4	9.5	219.0±10.9	4	200	100	200
0.6	149.1±12.1	9.0	109.0±31.1	7	200	150	100
2.2	153.4±7.0	2.7	205.4±14.8	3	200	150	200
5.3	52.7±3.3	40.0	30.0±8.4	3	200	50	50

species in the sample, i.e. it includes the various forms of alkalinity of all weak acid subsystems present. The average accuracy in the determination of this value was added to Table 1 for the purpose of further evaluating the overall accuracy of the method. Because the total alkalinity of the original made-up samples was known, the value calculated back by the method gave a further assessment of the accuracy of the method. In contrast, the accuracy of the carbonate alkalinity value calculated by the method could not be directly assessed. However, its correctness could be implied from the accuracy of the values attained from the citric acid and total alkalinity concentrations.

Referring to the data listed in Table 1 the following can be concluded:

- The accuracy of the measurement of both the citric acid concentration and carbonate alkalinity (as inferred from the accuracy of the total alkalinity measurement) is high – typically an error of not more than a few mg/ℓ for both values. In percentage terms, the relative error is normally below 5% for both the alkalinity value and for citric acid concentrations of above 30 mg/ℓ as $C_6H_8O_7$. Lower citric acid concentrations resulted in higher relative errors (10.2% and 15.5% error for 30 and 20 mg/ℓ as $C_6H_8O_7$ respectively) but the actual error was small (3.5 and 3.1 mg/ℓ respectively). High repeatability in results was obtained for both citric acid and carbonate alkalinity values, with typical standard deviations not exceeding a few percents of the average value.
- In mixed weak acid systems, differentiation between the different weak acids by titration is less accurate when large differences exist between the concentrations of the different subsystems. As a result, it was expected that a less accurate result would be obtained for a low citric acid concentration mixed with a high total carbonate species concentration. This is explained by a masking effect, resulting from the fact that a small error (in %) in the determination of the high concentration species (C_T in this case) results in a large error in the determination of the low concentration species (the citric acid). However, down to citric acid concentration of 20 mg/ℓ (C_T to CA_T molar ratio of about 40 to 1), relatively good accuracy was obtained. Conversely, at a high molar ratio of 80 to 1 (citric acid = 10 mg/ℓ vs. C_T = 200 mg/ℓ as $CaCO_3$), the method gave meaningless results.

Samples containing VFA (anaerobic)

From the inception it was clear that this algorithm, aimed at solving a set of equations for three unknowns, although theoretically correct, would be much more prone to pH experimental errors than the simpler "aerobic" model that involves only two unknowns. This situation is exacerbated by the fact that the three weak acid systems involved share (for practical purposes) two pK values. The third (most basic) pK of the citric acid system (pK = 6.33) is practically identical to the first (acidic) pK of the carbonate system (pK = 6.35), and the second pK of the citric acid (pK = 4.72) practically overlaps with that of the VFA system (pK = 4.75). From a mathematical standpoint, this overlapping of the pK values resulted in a set of equations in which the contribution of each of the unknowns in each of the equations was not negligible. Consequently, small, unavoidable errors in observed pH values had the potential to cause large errors in results. This led to certain instability in the results obtained from this method.

The "anaerobic model" was assessed using laboratory-made solutions with no divalent ions present. Results are shown in Table 2. By and large, despite the initial concerns, fairly accurate results were attained for both citric acid and VFA concentrations, although accuracy was lower than in the absence of acetic acid, and also, from time to time, instable and erratic results were obtained. As mentioned, the lower accuracy and higher instability of this method was attributed to the sensitivity of the mathematical algorithm to small pH deviations. This sensitivity was even more apparent when low citric acid concentrations (50≥ mg/ℓ) were measured in the presence of a relatively high C_T. In these tests, a large relative error (40%) was encountered (although the absolute error was not extremely large - 20 mg/ℓ or 0.1 mM). With regard to the measurement of the VFA concentrations – the results attained in all experiments were accurate and repeatable, down to concentrations as low as 50 mg/ℓ.

Overcoming the inhibition caused by the formation of complexes of citric acid with Ca^{2+} and Mg^{2+}

Many water streams worldwide can be characterised as semi-hard or hard, i.e. containing a considerable amount of multivalent cations. In most natural waters calcium and magnesium ions are the dominant divalent ions present. As mentioned above, citric acid is an excellent complexing agent for Ca^{2+} and Mg^{2+}. Because complexation is not accounted for in the algorithm, meaningful interpretation of titration results in the presence of Ca^{2+} and Mg^{2+} is not possible. In order to overcome this problem the solution was passed trough a Na^+ saturated ion exchange

TABLE 3
Measurements of tap water + citric acid with and without (after ion exchange) divalent ions

Relative error in citric acid measurement	Citric acid concentration as measured by the method	No. of samples tested	Mg^{2+}	Ca^{2+}	Citric acid added	Sample description
(%)	(mg/ℓ as $C_6H_8O_7$)	-	(mg/ℓ as Mg^{2+})	(mg/ℓ as Ca^{2+})	(mg/ℓ as $C_6H_8O_7$)	
80.8	361.6±7.0	2	15	40	200	Tap water + citric acid
4.9	209.8±3.4	3	0.2	0	200	Tap water + citric acid after exchange with Na^+
1.1	101.1±8.7	7	0.02	0.2	100	Tap water + citric acid after exchange with Na^+
11.8	55.9±8.8	7	0.02	0.2	50	Tap water + citric acid after exchange with Na^+

resin. The goal was to substitute the two divalent cations with Na^+ that do not complex with citric acid, and thus allow for accurate execution of the method.

To demonstrate the feasibility of the concept, a sample of hard tap water (Ca^{2+} = 40 mg/ℓ; Mg^{2+} = 15 mg/ℓ) to which 200 mg/ℓ citric acid was added, was prepared. Table 3 (upper row) shows the citric acid concentration attained from applying the method to the raw sample containing Ca^{2+} and Mg^{2+}. Table 3 also shows (second to fourth row) the results obtained from the algorithm with tap water + citric acid, but after passing it through the cationic exchanger.

As shown, meaningless results were obtained from the method when the raw sample containing the divalent ions was titrated. In contrast, it is clear from the results that passing the sample through the ion exchanger not only reduced the calcium and magnesium concentrations significantly, but also enabled a correct and accurate execution of the method. It was therefore concluded that any sample containing Ca^{2+} and Mg^{2+} should be passed through a Na^+-saturated cation exchanger prior to applying the procedure.

Conclusions

A new multiple-point titration method was developed for measurement of low citric acid concentrations in a mixture of weak-acids. The method can be used for 2 distinct situations, in the absence (aerobic environments) or presence of VFAs (anaerobic environments).

Results indicate acceptable accuracy for both variants of the method, typically within ±5% for citric acid concentrations higher than 50 mg/ℓ, with relatively good repetition (average STDV in all "aerobic" samples = 4%). Both accuracy and precision were lower when the "anaerobic" method was applied to laboratory-made waters; however, an average prediction accuracy of over 90% was attained for samples with citric acid concentration of above 50 mg/ℓ.

Samples containing Ca^{2+} and Mg^{2+} concentrations should be passed through a Na^+ saturated cation exchanger prior to applying the method.

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