A nine-point pH titration method to determine low-concentration VFA in municipal wastewater

Hainan Ai, Daijun Zhang, Peili Lu and Qiang He

ABSTRACT

Characterization of volatile fatty acid (VFA) in wastewater is significant for understanding the wastewater nature and the wastewater treatment process optimization based on the usage of Activated Sludge Models (ASMs). In this study, a nine-point pH titration method was developed for the determination of low-concentration VFA in municipal wastewater. The method was evaluated using synthetic wastewater containing VFA with the concentration of 10–50 mg/l and the possible interfering buffer systems of carbonate, phosphate and ammonium similar to those in real municipal wastewater. In addition, the further evaluation was conducted through the assay of real wastewater using chromatography as reference. The results showed that the recovery of VFA in the synthetic wastewater was 92%–102 and the coefficient of variance (CV) of reduplicate measurements 1.68%–4.72%. The changing content of the buffering substances had little effect on the accuracy of the method. Moreover, the titration method was agreed with chromatography in the determination of VFA in real municipal wastewater with $R^2 = 0.9987$ and CV = 1.3–1.7. The nine-point pH titration method is capable of satisfied determination of low-concentration VFA in municipal wastewater.

Key words | determination, municipal wastewater, titration, volatile fatty acid (VFA)

INTRODUCTION

Wastewater contains a variety of organic components with different physical and chemical properties, among which the volatile fatty acid (VFA) is considered as the only form that can be directly utilized by the micro-organisms in a biological wastewater treatment system. The other biodegradable organics need to be transformed to VFA for their degradation. According to this recognition, VFA is incorporated into Activated Sludge Model No. 2 and No. 2d (ASM2, ASM2d) as an important COD fraction (Henze et al. 1995, 2000). Especially, for enhanced biological phosphate removal, VFA is the only carbon resource for phosphate-accumulating organisms (PAOs) to synthesize PHA (Oehmen et al. 2007). As a result, the determination of VFA in wastewater becomes necessary for the usage of ASMs, as well as the understanding of wastewater composition (Lu et al. 2010).

So far, several methods such as titration, chromatography, colorimetry and distillation have been proposed for the measurement of VFA in wastewater. The chromatography, colorimetry and distillation method are mainly used to determine high-concentration VFA, such as the VFA in industrial wastewater or anaerobic fermentation broth (Feitkenhauer et al. 2002; Lahav & Loewenthal 2000). As for the measurement of the low concentration VFA, which is typical in municipal wastewater, the pH titration method based on buffer theory has been proven feasible and to be

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the most cost effective analysis (DiLallo & Albertson 1961; Vanrolleghem & Lee 2003). The five-point pH titration method was proposed by Moosbrugger et al. (1993a, 1993b). However, there was a larger deviation between the tested results and the real values, probably due to either a residual liquid junction potential or poor pH meter calibration. The correction of the test pH was proposed to overcome this issue. Later, Lahav et al. (2002) took insight of the five-point method and concluded that the error in pH observations was random rather than systematic and it was therefore difficult to ascribe a “systematic pH error” to the liquid junction effects. Based on the principle of the five-point pH titration method, Lahav et al. (2002) proposed the eight-point pH titration method. However, the principle of eight-point pH titration method was debatable due to its subjectivity. Moreover, this method was proposed aiming at determining high concentration VFA in the anaerobic reactor, while its capacity in determining low concentration VFA in municipal wastewater still needs to be investigated.

The five-point and eight-point pH titration methods (Moosbrugger et al. 1993b; Lahav et al. 2002) are based on the total alkalinity of weak acids and bases in wastewater and the algebraic calculation procedures were used to solve VFA concentration. As a result of the presence of various buffer subsystems in wastewater and taking no consideration about the contribution of other buffer subsystems than carbonate and VFA acids to the determinative error of the total alkalinity, the eight-point pH titration methods had a poor accuracy in the measurement of the low concentration VFA (Lahav et al. 2002). In this study, with taking consideration about the contribution of all buffer subsystems to the determinative error of the total alkalinity and by using the least-square optimizing calculation procedures, a total alkalinity based nine-point titration method was proposed for determining low concentration VFA in municipal wastewater, in which three pH titration observations were used to calculate total alkalinity and the others were used to determine VFA. The method was evaluated using synthetic wastewater containing low-concentration VFA and real municipal wastewater.

**MATERIALS AND METHODS**

**Expression of the total alkalinity**

Besides VFA, municipal wastewater also contains other weak acid/base buffer systems, such as carbonate, ammonium, sulfide and phosphate. These buffer systems display different buffer intensities and buffer ranges. At a specific pH value, the theoretical total alkalinity ($M^{TH}_{tot}$) in wastewater is the sum of the mass of acceptable protons (eq 1):

$$M^{TH}_{tot} = C_T F_C (pH_x) + A_T F_A (pH_x) + N_T F_N (pH_x)$$

$$+ P_T F_P (pH_x) + S_T F_S (pH_x) + [OH^-]_{pH_x}$$

$$- [H^+]_{pH_x}$$

(1)

where $M^{TH}_{tot}$, the total alkalinity of wastewater at some pH (calculated by CaCO3), mg/L; $C_T$, the carbonate concentration in wastewater, mg/L; $A_T$, VFA concentration in wastewater, mol/L; $N_T$, the ammonia concentration in wastewater, mol/L; $P_T$, the phosphate concentration in wastewater, mol/L; $S_T$, the sulfide concentration in wastewater, mol/L. $F_C (pH_x)$, $F_A (pH_x)$, $F_N (pH_x)$, $F_P (pH_x)$ and $F_S (pH_x)$ are functions which relate to the ionization constants of all levels of various substances at a certain pH; $[OH^-]_{pH_x}$ and $[H^+]_{pH_x}$ refer to the amounts of OH and H⁺ which are produced by ionization of water or other strong acidi and alkaline substances in wastewater at a certain pH.

When the wastewater sample titrated using strong acids, the total alkalinity at each titration endpoint can be established according to Equation (1):

$$M^{TH}_{tot} - V_x C_a = C_T F_C (pH_x) + A_T F_A (pH_x) + N_T F_N (pH_x)$$

$$+ P_T F_P (pH_x) + S_T F_S (pH_x) + [OH^-]_{pH_x}$$

$$- [H^+]_{pH_x}$$

(2)

where $V_x$, the volume of the added strong acid, mL; $C_a$, the concentration of the added strong acid, mol/L.

Considering the possible error $M^{ER}_G$ during the testing process of the total alkalinity, the relation among the theoretical total alkalinity, the tested total alkalinity ($M^{TH}_{tot}$), and the test error was shown as Equation (3):

$$M^{TH}_{tot} - V_x C_a = C_T F_C (pH_x) + A_T F_A (pH_x) + N_T F_N (pH_x)$$

$$+ P_T F_P (pH_x) + S_T F_S (pH_x) + [OH^-]_{pH_x}$$

$$- [H^+]_{pH_x} \pm M^{ER}_G$$

(3)

There are 6 unknown variables in Equation (3), whereas the contents of ammonium, total phosphorus, and sulfide in wastewater could be determined with the standard methods. Only $C_T$, $A_T$ and $M^{ER}_G$ remain unknown once the initial total alkalinity of wastewater is known. If $M^{ER}_G$ was smaller than
the error limit of the total alkalinity ($M_{\text{G}}^{\text{FR}} / M_{\text{G}}^{\text{TS}} \leq 5\%$ in this study), $C_T$, $A_T$ and $M_{\text{G}}^{\text{FR}}$ in wastewater could be determined.

### Determination of the total alkalinity ($M_{\text{TS}}^{\text{tot}}$)

Gran titration method (Gran 1952) is recognized as one of the most precise methods to determine the total alkalinity. Only $H_2PO_4^-$ accepts protons when the strong acid solution is added in the pH range of 2.4–2.7:

$$M_{\text{TS}}^{\text{tot}} = V_e \cdot C_a - V_s \cdot C_a = \{[H_2PO_4^-]_e - [H^+]_e\} \times (V_x + V_s)$$  \hspace{1cm} (4)

where $V_e$ the volume of the strong acid required to add when neutralizing the total alkalinity of the solution, mL; $V_s$, the volume of the added strong acid, mL; $V_{TS}$, the original volume of the solution, mL; $C_a$, the concentration of the added strong acid, mol/L.

Expressing $H_2PO_4^-$ with $P_T$ and its equilibrium constant, Equation (5) was produced:

$$C_a(V_e - V_s) = (V_s + V_x) \times \left\{ \frac{K_{p1} K_{p2}(H^+)^2}{(H^+)^2 + K_{p1} K_{p2}(H^+) + K_{p1} K_{p2} K_{p3}} \times \frac{V_x}{V_s + V_x} - 10^{-pH_{x}} \right\}$$  \hspace{1cm} (5)

where $K_{p1}^t$, $K_{p2}$ and $K_{p3}$, the ionization constants of $HPO_4^{2-}$, $H_2PO_4^-$ and $PO_4^{3-}$, greatly affected by temperature and pH value; ($H^+$), the activity of $H^+$. When there was the high concentration ion in the water sample, the ionic activity was smaller than the ion concentration and their difference could be expressed with activity coefficient. These three constants were corrected. In accordance with the regulations of IUPAC (International Union of Pure and Applied Chemistry), the following equation could be used for the mixed acidity constant:

$$K' = \frac{(H^+)[B]}{[HB]}$$  \hspace{1cm} (6)

where $(i) = n_i$; $n_i$, the activity coefficient which could be calculated by Davies equation:

$$\log n_i = -AZ_i^2 \left[ \frac{\mu^{1/2}}{1 + \mu^{1/2}} - 0.2 \mu \right]$$  \hspace{1cm} (7)

where $A = 1.82 \times 10^{8}(T)^{-3/2}$; $\mu$, the ionic strength of the solution; $\epsilon$, the dielectric constant; $Z_i$, the electric charge number of ion $i$. While the ionic strength in the solution had the following relation with the total dissolved solid (TDS):

$$\mu = 2.5 \times 10^{-5} \times TDS(\text{mg.L}^{-1})$$  \hspace{1cm} (8)

$$TDS(\text{mg.L}^{-1}) = 6.7 \times EC(\text{mS.m}^{-1})$$  \hspace{1cm} (9)

where EC referred to the conductivity of the solution which could be determined with the conductivity meter.

After correcting the equilibrium constant with the ionic strength, the correction with the temperature could be finished with Van’t Hoff equation.

$$\frac{d \ln K}{dT} = \frac{\Delta H^0}{RT^2}$$  \hspace{1cm} (10)

Provided $\Delta H^0$ did not change with the temperature within the limited temperature range, and the product of integrating the above equation was as follows:

$$\ln \frac{K_1}{K_2} = \frac{\Delta H^0}{R} \left( \frac{1}{T'_2} - \frac{1}{T'_1} \right)$$  \hspace{1cm} (11)

where $K$, the equilibrium constant; $T$, the thermodynamic temperature; $\Delta^0$, the standard enthalpy of formation; and $\Delta^0$ of all substances could be found out in American Handbook of Chemistry and Physics (David 2006).

All right variables of Equation (5) were known, defined as $F_X$ The slope $(V_e)$ of linear relation between $F_X$ and $V_X$ was obtained, further calculating the initial total alkalinity $M_{\text{Tot}}^{\text{TS}}$ with Equation (12):

$$M_{\text{Tot}}^{\text{TS}} = V_e C_a$$  \hspace{1cm} (12)

### Selection of pH titration points

As the carbonate buffer system would affect the measurement of VFA in wastewater, the pH titration points were selected so that the carbonate buffer system and the VFA buffer system could fully reflect the buffer intensity at the selected pH points. The first and second titration points selected were symmetric to $pK_a$ of $\text{HCO}_3^-$, while the third and fourth titration points were symmetric to $pK_a(\text{Ac}^-/\text{HAc})$ (it was suggested to select the symmetric titration points within 0.05 pH of $pK$ value). The $pK_a$ value of $\text{HCO}_3^-$ and the $pK_a$ value of acetic acid are 6.3 and 4.75 (25), respectively. Consequently, $\text{pH} = 6.85$, $\text{pH} = 6.35$, $\text{pH} = 5.85$, $\text{pH} = 5.25$, $\text{pH} = 4.75$. 

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pH = 4.75 and pH = 4.25 were firstly selected as calculated titration points, plus three titration points used by Gran titration method, there were totally 9 pH titration points.

The main instruments used during the titration process included Leici ZDJ-4 A Automatic potentiometric titrimeter and DDS–11 A Digital Conductivity Meter produced by Shanghai Accuracy Instrument Co., Ltd. During the titration process, NaOH was used firstly to adjust pH to over 6.85 if the initial pH value of the water sample was below 6.85, and then the titration of the preset pH points was performed one by one and the titration results were recorded. The concentration of PO\textsubscript{4}\textsuperscript{3-} and ammonium in the solution was determined with the standard method (APHA 1998). Each sample was parallel-titrated for 4 times.

RESULTS AND DISCUSSION

Firstly, the synthesized wastewater containing acetate, carbonate, ammonium, and phosphate was used to evaluate the accuracy and reproducibility of the method based on the recovery and the CV of several repeated measurements. The components and their concentrations of the synthesized wastewater were close to real municipal wastewater. The results were shown in Table 1 (the influence of sulfide on this method was not taken into consideration in experiments).

Secondly, the proposed method was used to measure VFA in real municipal wastewater from a full-scale wastewater treatment plant and then compare and find out the correlation between it and the ion chromatography as reference. The sample was firstly flocculated with 0.06 mol/L ZnSO\textsubscript{4} solution and filtrated with 0.45\textmu m membrane, and then it was divided into two parts, one for titration and the other for ion chromatograph. The used ion chromatograph was DX-120 ion chromatograph produced by Dionex Corporation (USA), 0.4 mmol/L perfluorobutyric acid-acetonitrile (v/v: 97:3) as eluent, flow rate 1.0 ml/min, 5 mmol/L tetrabutylammonium hydroxide as regeneration liquid, sample injection volume 50\textmu L/time. The results were shown in Figure 1.

The results in Table 1 showed that the presence of carbonate alkalinity slightly affected VFA concentration measured by the nine-point pH titration method. Such influence was not significant when VFA concentration was considerable to the carbonate alkalinity concentration, while became significant with the increase of the carbonate concentration, namely the increase of C\textsubscript{T}/A\textsubscript{T}, mainly because this system was a relatively but not absolutely close system. Therefore, the volatilization of CO\textsubscript{2} still occurred, and the volatilized volume of CO\textsubscript{2} increased during the titration.

From Table 2, it could be seen that the presence of the ammonium buffer system had little influence on the test results by the nine-point pH titration method, and the standard recovery reached over 93%, CV was controlled below 4.43% and gradually decreased with the increase of the acetic acid concentration, until to the minimum value of 2.28%. This was because with the increase of A\textsubscript{T}/N\textsubscript{T} within the titration range, the dominated buffering effect of the acetic acid system in the titration process also increased, which made the influence of the ammonium buffer system gradually decrease.

The phosphate in municipal wastewater was lower than the ammonium content. From Table 3, it could be seen that the phosphate alkalinity had a slight influence on the titration results, the standard recovery reached over 94%, and the titration error and CV gradually decreased with the increase of the acetic acid concentration in the solution. The phosphate affected VFA determination little because the relevant equilibrium constant of the phosphate system (i.e., pK\textsubscript{p2} 7.2) is much closer to pK\textsubscript{a} than to pK\textsubscript{C1}.

As shown in Table 4, under the co-presence of several buffer systems, the measured results of VFA concentration

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Co-presence of acetic acid and carbonate alkalinity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water sample</td>
<td>HAc (mg/L)</td>
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<tr>
<td>---------</td>
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</tr>
<tr>
<td>20</td>
<td>19.17</td>
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<tr>
<td>40</td>
<td>18.84</td>
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<td>20</td>
<td>18.61</td>
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<td>18.59</td>
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<tr>
<td>100</td>
<td>18.49</td>
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</table>
with the proposed method always kept a high accuracy. The carbonate buffer system was the major buffer system in municipal wastewater and could produce a small amount of CO$_2$ due to accepting protons, thus it was the buffer system with significant influence on the titration method, which was demonstrated in the five-point pH titration method (Moosbrugger et al. 1993a; 1993b) and also in the eight-point titration method (Lahav et al. 2002). The test results of VFA by the proposed method maintained a high accuracy at all the time, the standard recovery was 94.2%–101.12%, and the relative error was lowest to 1.12%. However, the concentrations of ammonium and phosphate in municipal wastewater are low; these two substances have little influence on the test results of VFA by the proposed method.

It could be seen from Figure 1 that, after parallel-testing three water samples for 5 times, the results with the nine-point pH titration method kept stable and CV was low (within 2%), which indicated that the proposed method had

| Table 2 | Co-presence of acetic acid and ammonium alkalinity

<table>
<thead>
<tr>
<th>Water sample (alkalinity)</th>
<th>HAc (mg/L)</th>
<th>NH$_4^+$–N (mg/L)</th>
<th>Measurement mean (mg/L)</th>
<th>Standard recovery (%)</th>
<th>CV (%)</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>9.49</td>
<td>94.90</td>
<td>4.43</td>
<td>9.49 ± 0.88</td>
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<td></td>
</tr>
<tr>
<td>30</td>
<td>29.29</td>
<td>97.63</td>
<td>3.54</td>
<td>29.29 ± 1.50</td>
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<td></td>
</tr>
<tr>
<td>50</td>
<td>50.53</td>
<td>101.06</td>
<td>2.05</td>
<td>50.53 ± 1.26</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>9.34</td>
<td>93.40</td>
<td>4.20</td>
<td>9.34 ± 0.67</td>
<td></td>
<td></td>
</tr>
<tr>
<td>30</td>
<td>28.65</td>
<td>95.50</td>
<td>3.36</td>
<td>28.65 ± 1.28</td>
<td></td>
<td></td>
</tr>
<tr>
<td>50</td>
<td>49.22</td>
<td>98.44</td>
<td>2.28</td>
<td>49.22 ± 1.09</td>
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| Table 3 | Co-presence of acetic acid and phosphate alkalinity

<table>
<thead>
<tr>
<th>Water sample (alkalinity)</th>
<th>HAc (mg/L)</th>
<th>PO$_4^{3-}$–P (mg/L)</th>
<th>Measurement mean (mg/L)</th>
<th>Standard recovery (%)</th>
<th>CV (%)</th>
<th>95% CI</th>
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<tbody>
<tr>
<td>10</td>
<td>9.55</td>
<td>95.50</td>
<td>4.34</td>
<td>9.55 ± 0.76</td>
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<tr>
<td>30</td>
<td>29.17</td>
<td>97.23</td>
<td>3.36</td>
<td>29.17 ± 1.33</td>
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<tr>
<td>50</td>
<td>49.63</td>
<td>99.26</td>
<td>2.07</td>
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<tr>
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<td>9.41</td>
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<td>28.95</td>
<td>96.50</td>
<td>3.08</td>
<td>28.95 ± 1.43</td>
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<tr>
<td>50</td>
<td>49.12</td>
<td>98.24</td>
<td>1.76</td>
<td>49.12 ± 0.86</td>
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| Table 4 | Co-presence of acetate, carbonate, ammonium, and phosphate

<table>
<thead>
<tr>
<th>Composition of the water sample</th>
<th>NaAc (mg/L)</th>
<th>Carbonate (mg/L)</th>
<th>NH$_4^+$–N (mg/L)</th>
<th>PO$_4^{3-}$–P (mg/L)</th>
<th>A$_T$ measurement mean (mg/L)</th>
<th>Standard recovery (%)</th>
<th>Relative error (%)</th>
<th>CV (%)</th>
</tr>
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<tbody>
<tr>
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<td>20</td>
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<td>9.55</td>
<td>95.50</td>
<td>-4.5</td>
<td>4.9</td>
<td>4.34</td>
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<tr>
<td>20</td>
<td>40</td>
<td>20</td>
<td>3</td>
<td>18.84</td>
<td>94.20</td>
<td>-5.8</td>
<td>4.2</td>
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</tr>
<tr>
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<td>3</td>
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<tr>
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<td>2.2</td>
<td>2.7</td>
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</tr>
<tr>
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<td>10</td>
<td>50.56</td>
<td>101.12</td>
<td>1.12</td>
<td>2.4</td>
<td>2.46</td>
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</table>
a very good reproducibility when testing VFA in real wastewater. Besides, the findings showed that the results with the nine-point pH titration method were greater than those with ion chromatography, with a difference of about 9.35%–10.61%; meanwhile there was a good linear relation between the results measured with two methods, with a correlation coefficient of 0.9987. The titration method titrated the total VFA concentration in the water sample, namely the sum of the contents of all C1-C6 volatile fatty acid, while it was reported in the relevant literatures (Elisabeth & Paul 1998) that the VFA in municipal wastewater mainly included acetic acid and propionic acid, the both accounted for approximately 90% of the total VFA, of which the former accounted for 80%. There was very few of formate acid, butanoic acid, pentanoic acid, and hexanoic acid in municipal wastewater, which could not be measured in the ion chromatogram due to the sensitivity of the ion chromatography column. Therefore, theoretically, the measurement results with the nine-point titration method should be slightly greater than those with ion chromatography, and the difference between them also should be about 10%, as shown in Figure 1.

Compared with the five-point and eight-point methods (Moosbrugger et al. 1993b; Lahav et al. 2002), the nine-point method proposed in this paper had the following advantages: The nine-point method fully considered the effects of various buffer subsystems throughout the titration process and used the total alkalinity in wastewater as baseline at the beginning, with a reasonable titration system. The nine-point method adopted the least-squares fitting for solution, while the five-point and eight-point methods used algebra for solution (Moosbrugger et al. 1993b; Lahav et al. 2002), thus the calculation method of the nine-point method was more reasonable and accurate. The eight-point method was mainly applied for the titration of high concentration VFA (≥50 mg/L) (Lahav et al. 2002), while the nine-point method could precisely measure the low concentration VFA in municipal wastewater.

CONCLUSIONS

In this paper, a new nine-point pH titration measurement method is proposed based on the basic definition and expression of the total alkalinity in wastewater, which focuses on the solution of the issue that the test results by the traditional titration methods are affected by various buffer systems in wastewater. The results from the evaluation of the proposed method with the synthetic and real wastewater samples show that the method has a good advantage in the aspects of accuracy, precision, and reproducibility.

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