ADPI Analytical Method #007
Determination of Titratable Acidity

#007a: Phenolphthalein Indicator Method
#007b: Autotitrator Method

1.0 Purpose

This Analytical Method defines the standard operating procedure for determination of titratable acidity in milk and dairy products.

2.0 Scope

This SOP is applicable to determination of titratable acidity in fluid milk and in other dairy products.

3.0 Definitions

3.1 **Titratable acidity** is the lactic acid stoichiometric equivalent to the quantity of standardized sodium hydroxide reagent required to titrate the sample preparation to endpoint, determined either by a color change from colorless to pink as visualized using phenolphthalein indicator solution or by instrument measurement of the equivalent pH of 8.3.

3.2 **Titration** is the process of controlled, incremental volumetric addition of a reagent (the **titrant**) to a sample preparation, allowing that titrant to react with components of the sample until a predefined chemical endpoint is achieved.

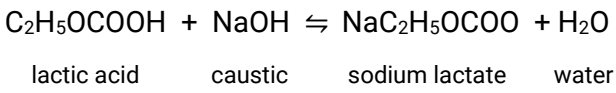
3.3 Titration is usually accomplished either manually using a volumetric apparatus called a **burette** (or sometimes “buret”) where the titrant is added manually, and the added quantity is read visually from a finely graduated scale incorporated into the burette; or automatically via a specialized lab instrument called an **autotitrator** where the addition process and the determination of the added titrant quantity are automated. Manual titration is most often accompanied by use of an indicator substance, while autotitration is most often performed to a point which can be measured by a probe such as a pH electrode or via a potentiometric electrode such as a dual pin platinum probe.

3.4 An **indicator** is a substance added to a titration which enables the visualization of the end of the chemical reaction in question. Typical indicator substances change from one color to another, or from a colorless form to a colored one. In the case of this method, phenolphthalein is the

indicator substance, and it changes from colorless at acidic pH levels to bright pink at an alkaline pH of 8.3 and higher.

3.5 The **endpoint** of a titration, sometimes called the equivalence point or the inflection point, is reached when the quantity of titrant added to the sample preparation becomes equivalent to the corresponding reactant in the sample. In this case, the endpoint is that moment when the alkaline titrant has reacted equivalently with the acidic components of the sample and the pH of the reacted solution rises to pH 8.3, where the phenolphthalein indicator chances color from colorless (acidic pH) to its first persistent, characteristic pink color.

3.6 **Stoichiometry** is the known quantitative relationship between reagents in a specific chemical reaction. In this method, the key stoichiometric relationship is the quantitative reaction between acid in the sample (presumed to be specifically lactic acid) and the base of known concentration (specifically sodium hydroxide). Each lactic acid molecule is neutralized by one molecule of sodium hydroxide as the titrant is added, forming sodium lactate and water in the process. When all lactic acid has reacted, any further hydroxide ions which are added then begin to accumulate in excess in the solution, causing an immediate rise in pH which corresponds to the endpoint of the chemical reaction.



4.0 Principle

The combination of the known concentration of the titrant, the measured volume of titrant addition, and the stoichiometry of the chemical reaction of the titration, allow for accurate determination of the quantity of the corresponding sample constituent that reacts with the chosen titrant. In the case of this method, which depends on simple acid/base titration, the titrant is 0.1000 *N* sodium hydroxide solution, which reacts stoichiometrically with acidic constituents of the sample preparation. For purposes of this method, those acidic constituents are presumed to be lactic acid, and the results of the test are reported as the quantity of lactic acid, on a percentage basis.

Because the visualization of a colorimetric endpoint can be subjective (and may even be entirely unachievable on a consistent basis, whether due to limitations in acuity of the specific analyst and/or due to coloration of the sample being tested), the reliance on non-visual means (e.g., pH measurement with an instrument probe) for endpoint determination has become commonplace and may contribute to more reliable, consistent, repeatable results.

5.0 Reagents and Materials

Adhere to the following requirements carefully for consistent and accurate results.

5.1 Laboratory balance, with capacity of approximately 500 grams and with sensitivity of ± 0.1 grams or better;

5.2 Spatula, or equivalent, suitable for handling solid samples;

5.3 Volumetric flask, 100.0 mL, Class A, calibrated to contain, or equivalent apparatus, suitable for diluting dry dairy samples before testing;

5.4 Mixer, or equivalent, suitable for dispersing and rehydrating reconstituted dry dairy samples (optional);

5.5 Distilled water, or higher purity, preferably boiled to drive off dissolved carbon dioxide which can interfere with the accuracy of the titration, allow to cool to ambient temperature before use;

5.6 Graduated cylinder, or equivalent, suitable for adding the 36 g / 36 mL of water as described below;

5.7 Casserole, Erlenmeyer flask, or equivalent, suitable for conducting the titration steps;

5.8 Burette, Class A, 25 mL, calibrated to deliver, graduated in 0.1 mL increments (for method #007a; auto-zeroing type is acceptable), or equivalent autotitrator may be substituted (method #007b);

5.9 Phenolphthalein indicator solution, ethanolic, 1% m/v (method #007a);

5.10 Pipette, or equivalent apparatus, suitable for adding 0.5 mL of the phenolphthalein indicator solution (method #007a);

5.11 Autotitrator probe, pH, potentiometric, dual-pin, gold, or equivalent, capable of accurately and consistently sensing the pH 8.3 endpoint of the titration (method #007b);

5.12 Sodium hydroxide (NaOH) solution, 0.1000 *N*, certified to a known and reported normality.

6.0 Personal Safety Precautions

In all cases, the practitioner's company's internal policies and procedures regarding personal safety supersede the following ADPI recommendations:

6.1 Milk (dairy) is globally classified as an allergen and should be properly handled with personal safety needs in mind.

6.2 Read and understand all precautions for safe handling and disposal shown in the Safety Data Sheets (SDS) for all the reagents required by this method, including use of any prescribed Personal Protective Equipment (PPE).

6.3 Dairy ingredients are foods and as such are exempt from U.S. requirements regarding Safety Data Sheets (SDS), where ingredient-specific safe handling instructions would be provided. Despite this exemption, many dairy ingredients are manufactured and marketed in powder form, and powders should be recognized as potential physical irritants, such as to the eyes, nose, and if inhaled.

6.4 Some testing apparatus described above may be susceptible to breakage, therefore be aware of associated personal risks. Inspect apparatus before use and replace any items which are compromised.

6.5 Exercise care when boiling water. Use appropriate PPE.

6.6 The ethanolic phenolphthalein indicator solution is extremely flammable. Utilize caution when handling.

7.0 General Considerations

General procedural considerations:

7.1 This method describes gravimetric sample preparation for all types of dairy samples to be evaluated, rather than volumetric approaches, e.g., the utilization of the classical Babcock- or Gerber-type pipettes for handling of milk and other fluid samples. Such volumetric apparatus, when maintained in good condition and used in careful compliance with established best practices, are suitable substitutes for the gravimetric approach described below and may be freely substituted.

7.2 Samples should be homogeneous, representative, and equilibrated to ambient temperature before handling, in a manner which will not compromise their suitability for testing.

7.3 Follow Good Laboratory Practices (GLPs) wherever applicable.

8.0 Initial Sample Preparation

8.1 Fluid sample materials require no prior reconstitution or dilution before testing; proceed to section 9.0 for these. Examples include:

a. Milk;
b. Fluid whey;
c. Melted ice creams, sherbets, ices;
d. Dairy mixes;
e. Yogurts.

8.2 To prepare dry samples, follow the appropriate scheme as shown in the following table to reconstitute, diluting the required weight of powder to the specified volume with water:

Sample type	required weights and volumes	
	sample weight (g)	QS to a final volume of (mL)
Dry whey	6.5	100.0
Nonfat dry milk Skim milk powder Malted milk Dry buttermilk Buttermilk product Whey protein concentrate Whey protein isolate Milk protein concentrate Milk protein isolate	10.0	100.0
Dry whole milk Whole milk powder	13.0	100.0

8.3 Mechanical mixing such as with an electric mixer may be useful to help disperse and re-hydrate dairy powders; exercise caution not to cause excessive foaming;

8.4 Allow the reconstituted material to stand for approximately 1 hour; mix gently and then proceed to section 9.0.

9.0 Titration

9.1 Accurately weigh 18.0 g of the liquid sample or the reconstituted preparation into a suitable titration vessel (e.g., casserole or Erlenmeyer flask);

9.2 Add about 36 g of water to the titration vessel;

9.3 Add 0.5 mL of phenolphthalein indicator solution (method #007a only);

9.4 Titrate with 0.1 *N* sodium hydroxide (NaOH) titrant solution until the first persistent (30 seconds) faint pink color is achieved (method #007a) or until pH 8.3 is reached (method #007b);

9.5 Record the volume of titrant required to reach the endpoint;

9.6 Proceed to section 10.0 for calculation of results.

10.0 Results Calculations

The key stoichiometric equivalence for this titration is that 1.0 mL of 0.1 *N* NaOH corresponds to 0.009008 g of lactic acid. The volume of titrant required to reach the endpoint, corrected to 0.1 N using the actual normality of the titrant, is used to calculate the equivalent amount of lactic acid (i.e., the titratable acidity) of the sample; and then this quantity of lactic acid is divided by the actual sample weight to express the results as a percentage.

Calculations are shown in simplified form, with factors for stoichiometric equivalence and conversion to a percentage basis worked into the formulas. Consult SMEDP for the long-form details if necessary.

10.1 Calculate the titratable acidity for undiluted samples on the as-is basis as follows:

$$\textbf{titratable acidity (\% as lactic acid)} = \frac{(\text{NaOH titrant, mL}) \times (\text{normality of NaOH}) \times (9.008)}{(\text{sample weight, grams})}$$

10.2 Calculate the titratable acidity for reconstituted samples on the reconstituted basis as follows:

$$\textbf{titratable acidity (\% as lactic acid)} = \frac{(\text{NaOH titrant, mL}) \times (\text{normality of NaOH}) \times (9.008)}{(\text{reconstituted sample weight, grams})}$$

10.3 Calculate the titratable acidity for dry powder samples on their original solids basis, using the reconstituted basis result from 10.2 above. Dividing the total weight of the reconstituted preparation by the weight of powder yields the dilution factor which, when multiplied by the result on the reconstituted basis, gives the titratable acidity of the powder on its original as-is basis:

$$\textbf{titratable acidity (\% as lactic acid)} = \frac{(\text{result from 10.2 above})(\text{weight of powder plus water, grams})}{(\text{weight of powder, grams})}$$

11.0 External References

Standard Methods for the Examination of Dairy Products ("SMEDP"), 17th edition, Ch. 15 – Chemical and Physical Methods, sections 15.020 – Acidity Tests.

12.0 ADPI Document Linkages

Analytical Method #001: *Sampling Dry Powders*

13.0 Revision History

Version	Effective Date	Notes
1.0	???	First officially approved version of this Standard Operating Procedure.
2.0	09/08/2023	Migrated this analytical method to the new modernized Standard Operating Procedure format as established by the ADPI Vice President of Technical Services.