BCH 445- Biochemistry of Nutrition [Practical]

Determination of Total Acidity of Food

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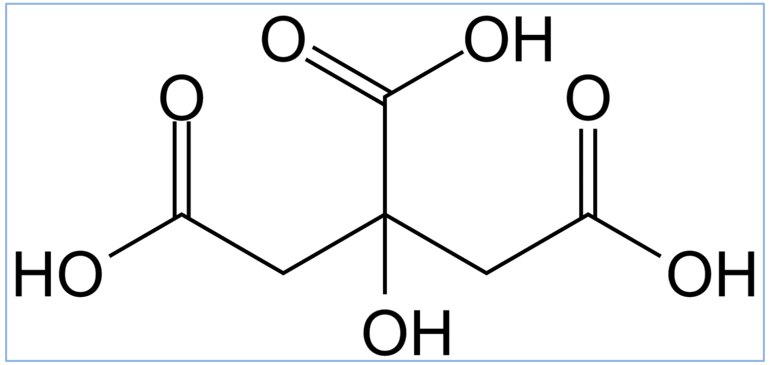
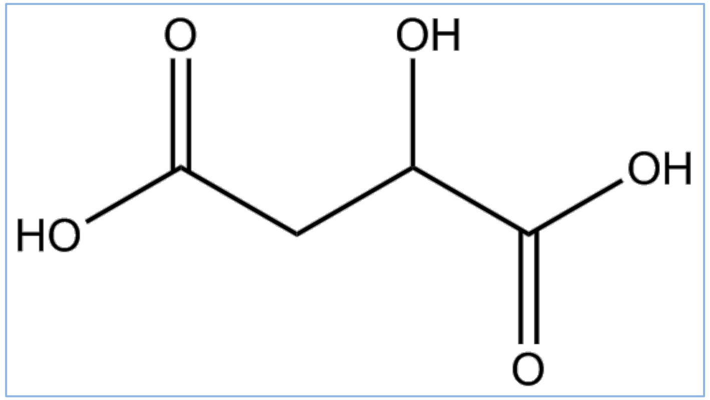
Food acidity

Food acids are usually **organic acids**, with citric, malic, lactic, tartaric, and acetic acids being the most common.

Organic acids are compounds that are characterized by their carboxyl group (–COOH).



Citric acid



Malic acid

However, inorganic acids such as phosphoric and carbonic (arising from carbon dioxide in a solution).

Acids often play an important and even predominant role in food acidulation.

The organic acids present in foods influence the:

1. Flavor (i.e., tartness).
2. Color (though their impact on anthocyanin and other pH-influenced pigments).
3. Prevent/retard the growth of microorganisms or inhibit the germination of spores.
4. Providing the proper environment for metal ion chelation, an important phenomenon in the minimization of lipid oxidation (e.g. citric acid, tartaric acid)

Organic acids may present:

- Naturally
- By Fermentation
- Added as part of a specific food formulation

(food acidulation)

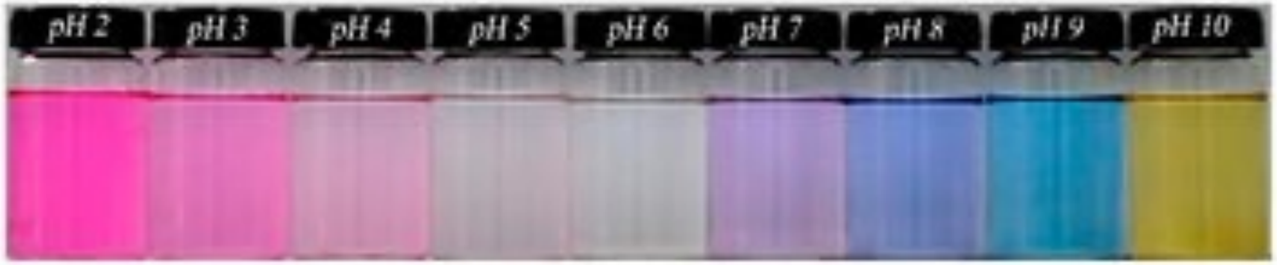


Figure 1. Color changes of anthocyanin in response to pH variation

The importance of determining food acidity

1- Determine the degree of maturity of fruits and vegetables

- The titratable acidity of fruits is used, along with sugar content, as an indicator of **maturity**, generally the higher the maturity, the lower the acid content. e.g. *in the ripening process*.

Such as tomatoes from green to mature stage, there is an increase in sugar content.

Maturity ↑ **Sugar content** ↑ **Acid content** ↓

2- To determine the freshness of foods

For example in milk, the more the lactic acid levels, means that milk is rotten.

The importance of determining food acidity

3- Acidity indicators reflect the quality of food

The amount of organic acids in food **directly** affects the food flavor, color, stability, and the level of quality.

4- Determination of acid on the microbial fermentation process

Such as, fermentation products in soy sauce, vinegar and other acids is an important indicator of **quality**.

Food acidity determination

There are two ways to express food acidity:

1- Titratable acidity

- Also called **total acidity**
- Simple estimate of the total acid content of food
- Better predictor of acid impact on flavor

2- Hydrogen concentration (pH)

- Depend on the strength of acid condition
- Quantify H^+ ions concentration

Acetic and citric acids are more sour than hydrochloric acid at the same pH.

Titratable acidity

- **Titratable acidity** is determined by neutralizing the acid present in a known quantity (weight or volume) of food sample using a **standard base.**
- The endpoint for titration is usually either a target pH or the color change of a pH-sensitive dye, typically phenolphthalein.

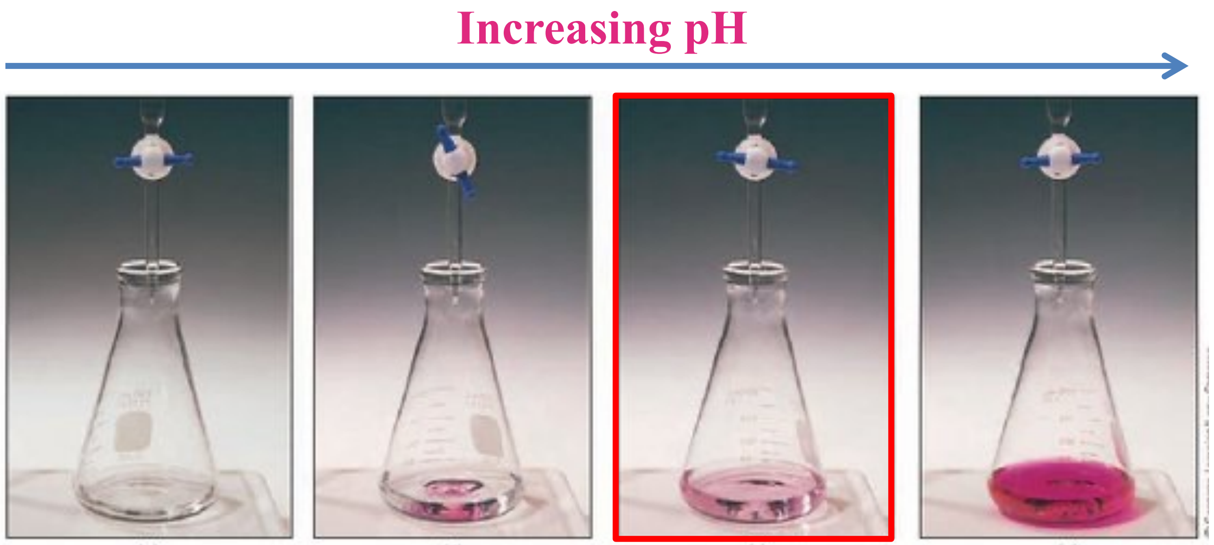


Figure 2. Color changes of phenolphthalein in response to pH variation

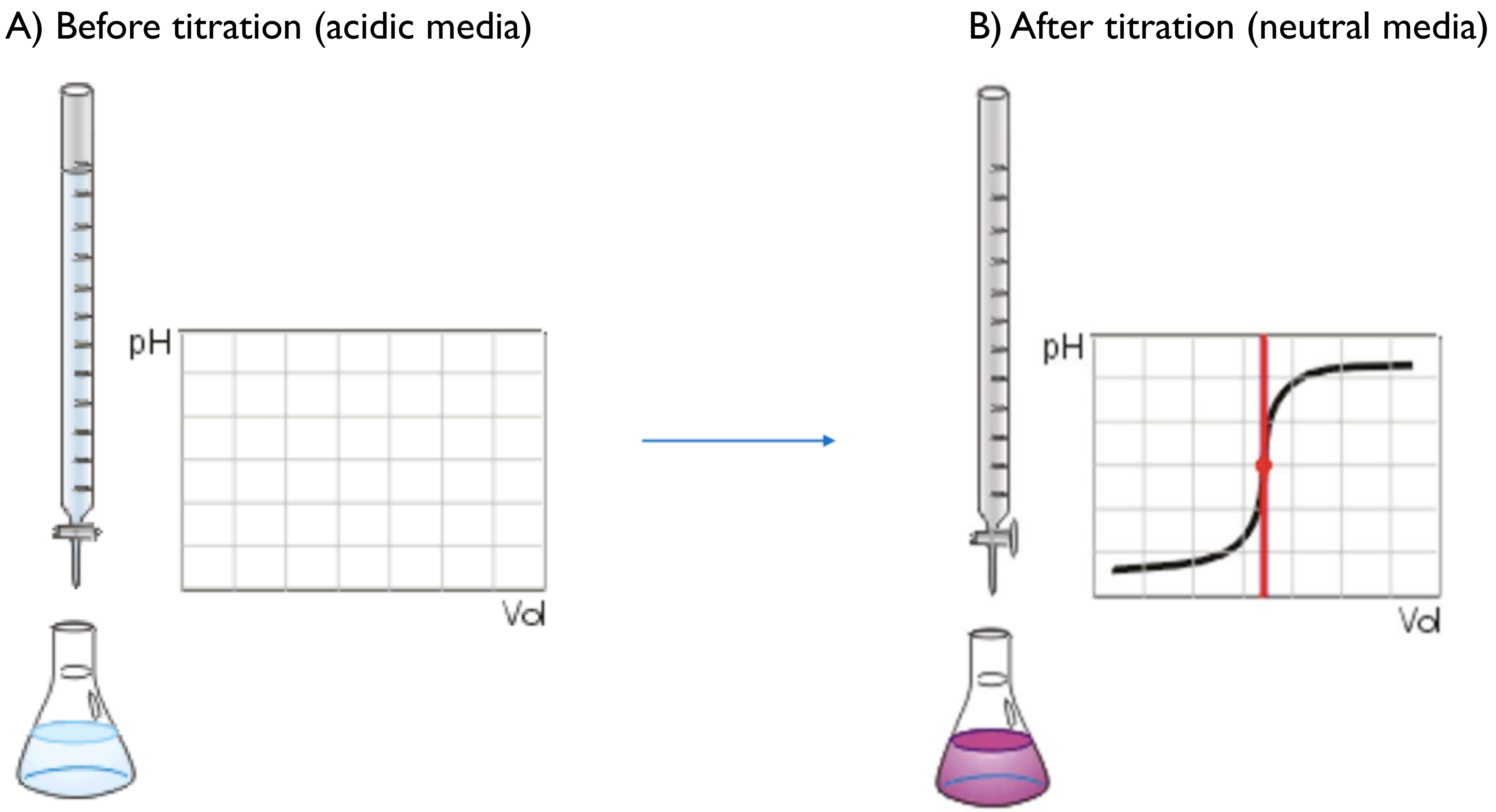


Figure 3. Schematic representation of the titration process

Practical Part

Objective:

- To determine total acidity of milk, juice, vinegar and oil acid value.

1-Determination of Milk Acidity (Titratable Acidity):

Measuring milk acidity is an important test used to determine milk quality.

The acidity of fresh milk is due to (natural acidity):

- Phosphates, casein and whey proteins, citrates and carbon dioxide dissolved during the process of milking

Developed acidity which is due to:

- **Lactic acid** produced by the action of bacteria on lactose in milk

Titratable Acidity:

- It is determine by titrating a known volume of milk with standard alkali to the point of an indicator, like phenolphthalein

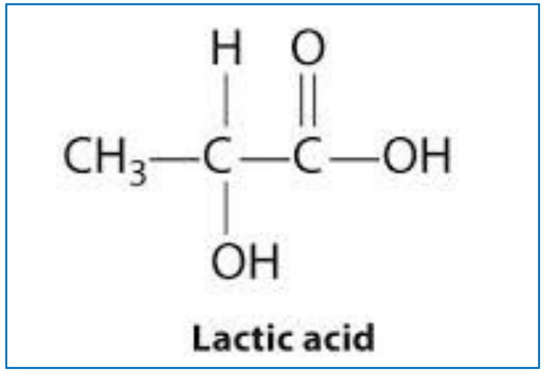
1-Determination of Milk Acidity (Titratable Acidity):

Normal range of milk acidity:

- **TA%**= 0.12%-0.16%, the average is 0.14%

- If it increased more than 0.16% indication of lactic acid by bacteria

➢ **Acidity is expressed as percentage of lactic acid** because **lactic acid** is the principal acid produced by fermentation



Method:

1. Fill the burette with 0.1 N NaOH solution
2. Mix the milk sample thoroughly by avoiding incorporation of air
3. Transfer 10 ml of milk to conical flask or beaker
4. Add equal quantity of distilled water
5. Add 3-4 drops of phenolphthalein indicator and stir
6. Rapidly titrate the contents with 0.1 N NaOH solution, continue to add alkali drop by the drop and stirring the content till first definite change to pink color which *remains constant for 10 to 15 seconds*
7. Note down the final burette reading

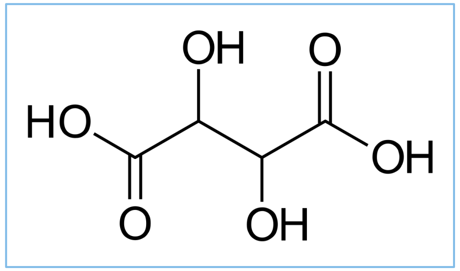
Calculations:

- **Lactic acid %=** $\frac{[0.1M\ NaOH\ X\ vol.\ of\ NaOH\ (in\ liter)]X\ 90.08)\ x\ 100}{Weight\ of\ the\ sample}$

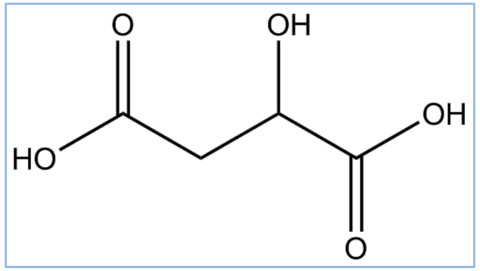
***90.08 g/ mol** is the molecular weight of Lactate

2- Determination of total acidity in juice:

- The acidity of natural fruit juices is the result mainly of their content of **organic acids**.
- **For example,** most fruits contain the tricarboxylic acid (citric acid) whereas grapes are rich in tartaric acid & peaches, apricots & plums in malic acids.
- Both tartaric & malic acids are **dicarboxylic acids**.



Tartaric acid



Malic acid

- The acidity of fruit juice may be determined by simple direct titration with 0.1M sodium hydroxide, using phenolphthalein as an indicator.

Method:

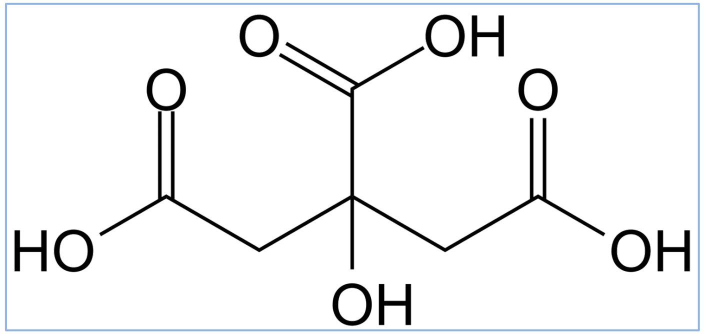
1. Transfer 10 ml of juice in a beaker
2. Add 25 ml of distilled water
3. Titrate with 0.1M NaOH, using 2 drops of phenolphthalein as an indicator

Calculations:

Calculate percent acidity of fruit juice (citric acid):

1- Wt. of citric acid= $\frac{0.1M\ NaOH \times vol.\ of\ NaOH\ (in\ liter) \times 192.43}{3}$

***192.43 g/mol** is the molecular weight of citric acid



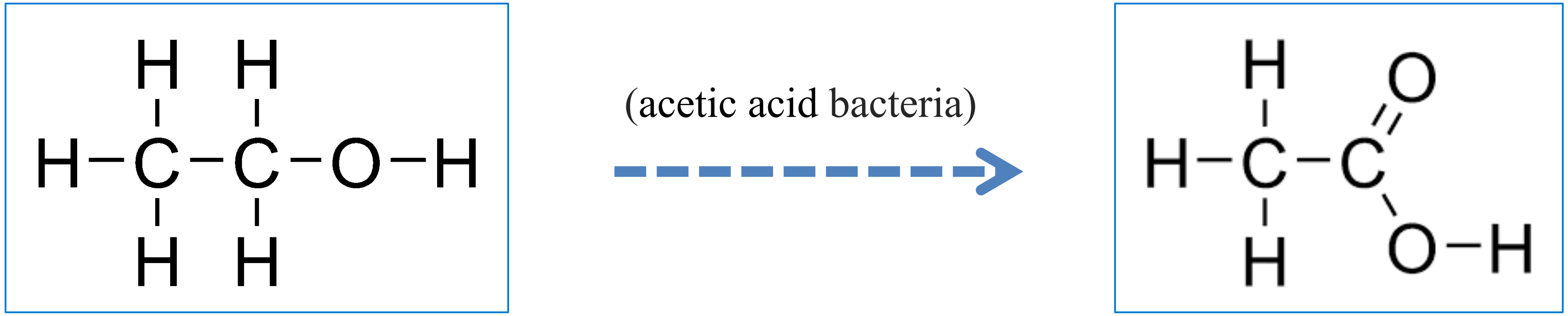
2-% of total acidity = (**wt. of acid** / wt. of sample) X 100

-Normal range for citric acid = **0.39 - 1.1 %**

*** Why the calculation of the weight of citric acid is divided by 3?**

3- Determination of total acidity in vinegar:

- The acidity of vinegar is derived by the fermentation of ethanol by acetic acid bacteria which produce **acetic acid (CH_3COOH)**, that is volatile.
- It may be determined by titration using **phenolphthalein** as an indicator .



Ethanol

Vinegar

Method:

Determination of total acidity:

1. Transfer 1 ml of vinegar.
2. Add 10 ml of distilled water.
3. Titrate with 0.1M NaOH, using 2 drops of phenolphthalein as an indicator.

Calculations:

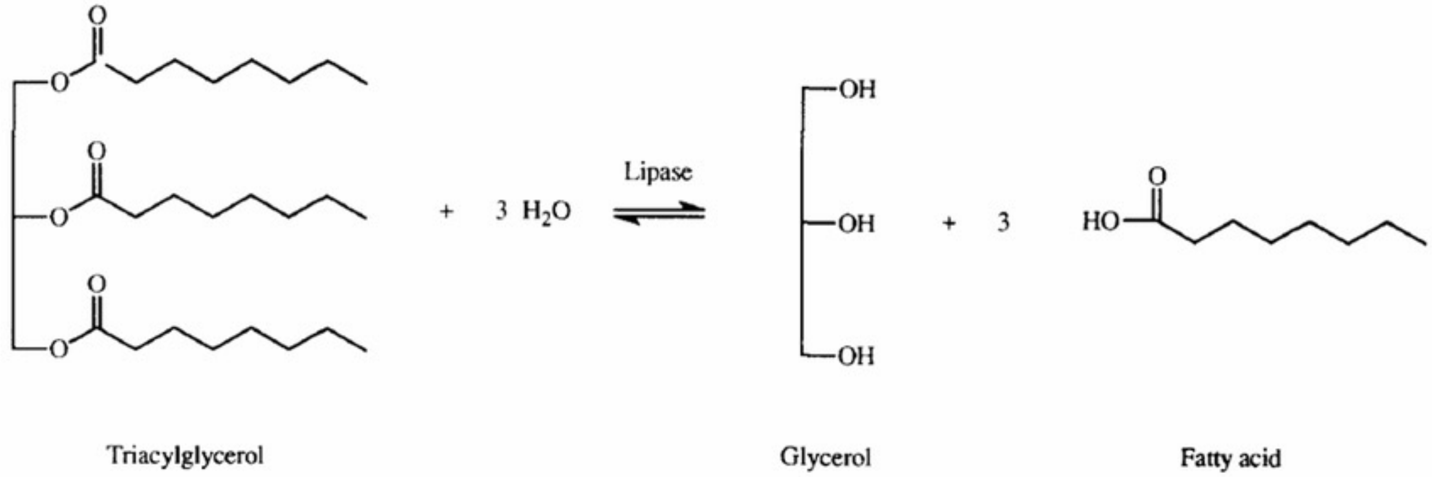
Calculate percent acidity as acetic acid (MW=60.05 g/mol)

1- **Wt. of acetic acid=** (0.1M NaOH X volume of NaOH in liter X **MW**)

2- **% of total acidity=** (wt. of acid / wt. of sample) X 100

4- Acid value:

- The acid value is defined as the number of milligrams of **base** required to neutralize the **free fatty acids** present in **one gram of fat.**
- As oil/fats rancidify, triacylglycerol are converted into fatty acids and glycerol, causing an increase in acid number.
- The amount of **free fatty acid** can be determined volumetrically by titrating the sample with sodium hydroxide .
- The value is also expressed as percent of free fatty acids calculated as **oleic acid.**



Principle

- The value is a measure of the amount of fatty acids which have been liberated by hydrolysis from the glycerides due to the action of **moisture, temperature and/or lipolytic enzyme lipase.**

- The acid value is determined by directly **titrating the oil/fat** in an alcoholic medium against standard potassium hydroxide/sodium hydroxide solution.

Method:

1. Mix the oil or melted fat thoroughly before weighting.
2. Weight accurately about 5 g of cooled oil sample in a 250 ml conical flask.
3. Add 50 ml of freshly neutralized hot ethanol.
4. Add one ml of phenolphthalein indicator solution.
5. Boil the mixture (in water bath) for about 5 minutes and titrate while hot against standard alkali solution shaking vigorously during the titration.

Calculations:

- Acid value = 39.997 X (V x N) / weight of sample

V = Volume in **ml** of standard potassium hydroxide or sodium hydroxide used.

N = Normality of the potassium hydroxide solution or sodium hydroxide solution.

W = Weight in g of the sample.

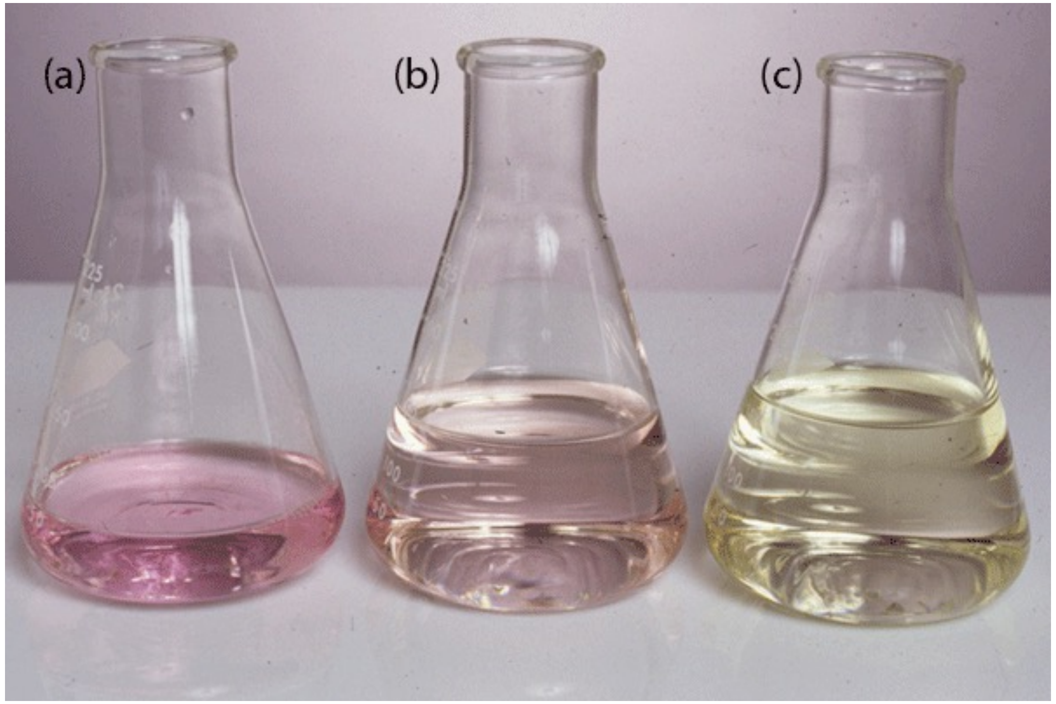
39.997 = The molecular weight of NaOH.

•**The maximum levels allowed for acid value of edible fats and oils is 0.6 mg NaOH/g**

Notes:

- Titratable acidity provide a **simple** estimate of acid in food.
- Routine titration **cannot** differentiate between individual acids.
- Therefore, titratable acidity is usually stated in terms of **predominant acid.**

Note the color at end point:



(a) Color in base

(b) Color at endpoint

(c) Color in acid

Discussion

Discuses the result you got for each sample and compare it to the normal range.

Homework

1. For each of the food products listed below, what acid should be used to express titratable acidity? Orange juice, yogurt, apple juice and grape juice.

2. **‘There is no direct relationship between pH and titratable acidity’** explain such statement

References

1. Nielsen S. Food Analysis. Springer Science & Business Media, 2014.
2. BCH 445- practical note

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