DIGESTION OF FAT IN MILK
Investigating the effect of temperature on lipase activity

Phenolphthalein is an indicator that is pink in alkaline solution of about pH 10. When the pH drops below pH 8.3, phenolphthalein turns colourless.

5% lipase solution
0.05M sodium carbonate solution

Approximately 4 minutes for colour change

Digestion of fat in milk

Effect of fat content on lipase activity

Compare fat content of different types of milk
DIFFERENT TYPES OF MILK AND THEIR FAT CONTENT

1. **100% Fresh Milk**
   - 3.5%

2. **1 1/2 fat Fresh Milk**
   - 1.8%

3. **1.5% Low Fat Slimilk**
   - 1.5%

4. **0.3% Fat Skimmed**
   - 0.3%
### Concept of Evidence Related to the Design of Investigation

<table>
<thead>
<tr>
<th></th>
<th>Comparison of fat content in different types of milk</th>
<th>Effect of fat content on lipase activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Independent variable (categoric vs continuous)</td>
<td>Types of milk (categoric)</td>
<td>Fat content of milk (continuous)</td>
</tr>
<tr>
<td>Dependent variable</td>
<td>Fat content measured as the time taken for colour change of phenolphthalein / final pH of the reacting mixture</td>
<td>Rate of reaction expressed as time taken for colour change / time taken for reaching stable pH</td>
</tr>
<tr>
<td>Controlling variables</td>
<td>...</td>
<td>...</td>
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<tr>
<td>Assumptions</td>
<td>...</td>
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</tbody>
</table>
PREDICTION OF THE RESULTS

Time taken for colour change in ascending order should be:

Fresh milk (3.5%), ½ fat fresh milk (1.8%), slimmed milk (1.5%), skimmed milk (0.3%)

Because …
IN TRIAL RUNS,

We used:

- 5% lipase
- 0.05M sodium carbonate solution

Results:

<table>
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<tr>
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<th>Fresh milk (3.5%)</th>
<th>½ Fat fresh milk (1.8%)</th>
<th>Slimmed milk (1.5%)</th>
<th>Skimmed milk (0.3%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time (s)</td>
<td>474</td>
<td>598</td>
<td>402</td>
<td>1206</td>
</tr>
</tbody>
</table>

Time taken for colour change in ascending order:

Slimmed milk (1.5%), fresh milk (3.5%), ½ fat fresh milk (1.8%), skimmed milk (0.3%)
A: Fresh milk
B: ½ Fat fresh milk
C: Skimmed milk
D: water

Lipase added simultaneously

Photo taken when the first tube turned from pink to white, i.e. tube C
CONCEPT OF EVIDENCE RELATED TO MEASUREMENT

Any replicates? (concept of accuracy and repeatability / or reliability)

- Repeated and same results are obtained
- Rule out errors due to manipulations
Validity of the design:

Is there any problem with the design?

- Can the experimental design yield data that can answer the problem?
- Can the measurement strategy collect data that are accurate and believable / reliable?
Is the measurement valid? (concept of ‘choice of instrument’)

- Set-point system, colour change (at pH 8.4) taken as the end point, what are the limitations of such measurement?
- How can we improve the measurement?
REPEATED USING pH SENSOR AND DATA LOGGER

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<th>Slimmed milk (1.5%)</th>
<th>Skimmed milk (0.3%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial pH</td>
<td>9.8</td>
<td>9.8</td>
<td>9.7</td>
<td>10.0</td>
</tr>
<tr>
<td>pH at end point</td>
<td>8.4</td>
<td>8.4</td>
<td>8.4</td>
<td>8.4</td>
</tr>
<tr>
<td>Time taken to reach end point (s)</td>
<td>400</td>
<td>579</td>
<td>410</td>
<td>1747</td>
</tr>
<tr>
<td>Final pH</td>
<td>6.7</td>
<td>7.4</td>
<td>7.4</td>
<td>8.4</td>
</tr>
<tr>
<td>Time taken for reaching stable pH (s)</td>
<td>1731</td>
<td>1073</td>
<td>1608</td>
<td>2238</td>
</tr>
</tbody>
</table>
With reference to the results, which set of data is most valid for comparing the relative amount of fat in different types of milk? Explain why.

Do the results obtained by pH sensor and data logger support the anomalous results of the method of using phenolphthalein to show the end point?

Why slimmed milk used a shorter time to reach end point for phenolphthalein method but a longer time to reach stable pH than ½ fat milk?
Will lipase work in lower pH? Will the change in pH of the mixture affect lipase activity?

Compare the ingredients of different types of milk, suggest a hypothesis to account for the anomalous results. How can you test this hypothesis?
What can be deduced from the ingredient labels?
EXPERIMENT TO SHOW THE FAT DROPLETS IN MILK
Hence, suggest why slimmed milk and skimmed milk are regarded as dairy products while the other two can be named fresh milk.
Other possible investigations:

- Dilute the fresh milk to the fat content of $\frac{1}{2}$ fat fresh milk and/or slim milk and compare the results
- Add bile salts to the mixture
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Lower cognitive demand:

- design an experiment to compare the relative fat content of fresh milk and ½ fat fresh milk (0.05M sodium carbonate)
  - No anomalous result
  - Simple data handling
  - Depend on which method of measurement (phenolphthalein or data logger), may have discussion on the concept of ‘choice of instrument’

If higher concentration of sodium carbonate is used, can include slimmed milk and skimmed milk
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Medium cognitive demand:

design an experiment to compare fat content of different types of milk using phenolphthalein method

- Ask students to predict results using their knowledge
- The anomalous results create cognitive conflict (discussion about replicas, validity and reliability)
  - If focusing on the measurement method, can lead students to the design of experiment with improvement on measurement method, compare the results obtained from the two methods
  - If focusing on application of knowledge, ask students to study the ingredient labels and propose a hypothesis to explain the anomalous results and action plan / another experimental design to test the hypothesis
- Or it can be a series of investigations to cover all
Higher cognitive demand:

provide them with materials for both methods, let them have some hands-on experience and come up with their own design to compare the fat content of different types of milk

- Justification of their choice of instrument / method
- Anomalous results
- Study of food label, apply knowledge to suggest an hypothesis for the anomalous results
- Further investigations to test their hypothesis
Instead of carrying out SBA assessment, the series of investigations can be used as a teaching package for concept of evidence.