

# Investigating Nutrient Cycling in Context (IB Core)



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## Decomposition – Investigating Nutrient Cycling in Context

### Teachers' Notes

This unit has been set up by staff at the Eagle's Nest to enable students to take part in a long term investigation to study the relative decomposition rates of different types of leaves. Students dip into the investigation when they arrive and contribute to an ever-expanding data base – making up and burying 'decomposition bags' and examining the contents of bags buried by previous groups. The unit is a half day study.

The unit introduces students to the theory behind decomposition – the nitrogen and carbon cycles – and applies that theoretical knowledge to a real life situation. The factors that influence decomposition rate are measured, including soil moisture, pH, soil texture and temperature. The biotic community responsible for carrying out the initial stages of decomposition are sampled. The chi square test is applied to test the statistical significance of the difference in decomposition rate between different types of leaves left to decompose in different woodlands.

NB: Data from previous experiments are included on this CD-ROM in the 'secondary data' section.

### Key Syllabus Areas

#### Topic 5: Ecology and evolution

5.1 Communities and ecosystems.

5.1.14 State that saprotrophic bacteria and fungi (decomposers) recycle nutrients

### Reference List

Adds, J., Larkcom, E., Miller, R. (1997) *The Organism and the Environment*. 2nd Edition. Nelson.

Chenn, P. (1999) *Ecology – Advanced Biology Readers*. John Murray Publishers.

### Introduction

**Bacteria and fungi in the Nitrogen Cycle:** (The full cycle is summarised in Appendix 1.)

Organic nitrogen in dead tissues – be they plant or animal in origin – and in faeces and urine can be converted back into inorganic nitrogen by the action of saprotrophic bacteria and fungi found in abundance in most soils. These organisms are referred to as decomposers – they recycle organic nitrogen compounds into ammonium ions,  $\text{NH}_4^+$ . This recycling is a vital process that enables the continuation of organic life on the planet. Without it, all of the organic nitrogen would become bound up in dead material – the supply of nitrogen on earth and in its atmosphere is not infinite.

Plants are able to absorb ammonium ions, but most of the ammonium is further converted into nitrite  $\text{NO}_2^-$  ions and on to nitrate  $\text{NO}_3^-$  ions, by the nitrifying bacteria *Nitrosomonas* and *Nitrobacter*, respectively. Plants are able to absorb nitrate ions from the soil most readily. However, nitrate and nitrite ions are very soluble in water and do not bind to soil particles effectively – they can be easily leached from soil after heavy rain.

Nitrates can also be lost from the soil if the soil becomes saturated, creating anaerobic conditions. Denitrifying bacteria such as *Pseudomonas denitrificans* and *Thiobacillus denitrificans*, in the soil convert nitrates and nitrites into nitrogen gas, which is lost from the soil into the atmosphere.

Some bacteria can fix atmospheric oxygen, assisted by the enzyme nitrogenase. Some nitrogen fixers are free-living in the soil, such as *Azotobacter vinelandii*. One genus of nitrogen fixing bacteria, *Rhizobium*, forms a symbiotic relationship with leguminous plants – the plants develop swellings on their roots, called root nodules, which contain the bacteria. The bacteria provides the plant with ammonia, whilst taking carbohydrates and ATP from the plant.

### Bacteria and fungi in the Carbon Cycle: (The full cycle is summarised in Appendix 2.)

Carbon plays a central role in the structure and metabolism of living organisms. It is released into the environment as carbon dioxide by all respiring organisms. Carbon locked up in plant and animal tissues may become part of a food chain when the plant material is eaten by herbivores. The organic compounds assimilated by the animals may then be used as fuel in respiration. Alternatively carbon may be released when a plant sheds it's leaves, an animal defecates or excretes matter containing carbon, or after death.

This organic material is decomposed by bacteria and fungi in the soil. After breakdown by digestive enzymes, the now soluble organic molecules are absorbed into the bodies of these saprotrophic decomposers, and may be used as fuel for microbial respiration. Carbon is thus returned to the atmosphere.

Factors affecting the rate of decomposition are related to the soil and overall climate conditions, the presence of fauna to carry out the initial stages of decomposition (the detritivores), the presence of a suitable soil micro-organism community and the nature of the material being decomposed.

Ideally, the soil should be well aerated – the detritivore community require an open-texture soil through which it is easy to move. The nitrifying bacteria are aerobic bacteria – the conversion of ammonium to nitrite and later nitrate are oxidation reactions – and poorly aerated soil will see more ammonium being converted to atmospheric nitrogen by denitrifying bacteria.

The soil should be warm – low temperatures inhibit bacterial activity and growth of fungal hyphae. But temperatures above 35°C may inhibit the activity of some soil bacteria.

The soil should be reasonably moist, but not saturated. Dry conditions will inhibit the growth of bacteria and fungi, and detritivores do not tolerate very dry conditions.

The soil should have a neutral pH – very acidic or alkalis conditions will inhibit the activity of bacteria and the growth of fungi. Also, the large detritivores prefer neutral soil conditions – earthworms in particular are inhibited by acidic soils.

These factors collectively imply that the overall climate needs to be warm, humid, with a moderate rainfall and few frosts. The underlying bedrock should ideally produce a soil which is neither too acid or too alkalis for the detritivores and decomposer organisms.

### **Specific information**

To take part in this ongoing research, students make up and bury a series of decomposition bags as outlined in the method. These bags will exclude part or all of the detritivore community by being made up from material with an increasingly small mesh size. The bags will contain a measured amount of leaf material from one of three different tree species under investigation – beech or ash.

The bags will be buried at one of two locations – a conifer plantation or a beech wood. At each site, students will assess soil pH, texture, temperature and moisture. Students will collect in a batch of bags from a previous group (or one previously buried by Eagle's Nest staff) and determine the amount of decomposition that has taken place in each bag since burial. If time allows (you could make this into a full day unit), students will also sample the soil invertebrate community, to determine what detritivores are present, and construct a woodland food web.

Ash and beech leaves have been shown to have a differential rate of decomposition due to the different amount of waste products contained within the leaves. Ash leaves fall to the ground still green – containing much chlorophyll and few waste products. They also fall in the autumn, while the weather is still mild. Ash leaves tend to decompose rapidly due to their greater palatability to detritivores – increase the surface area of the leaves for the saprotrophic organisms. Beech leaves tend to remain on the tree until well into the winter months, and are dropped to the ground brown and dry, full of tannins and polyphenols. Woodland invertebrates do not favour beech leaves. Their toxin content, waxy cuticle and dryness make them unpalatable to most woodland invertebrates.

By excluding different groups of invertebrates (the macro, meso and micro-invertebrates), we can investigate the importance of detritivores in the decomposition process, and investigate the relative rates of decomposition of the leaf material as a result.

### **Aims**

- To determine the comparative rate of decomposition of the leaves of two tree species;
- To investigate the importance of soil factors in influencing decomposition rate;
- To study the detritivore community in two woodlands and determine their role in the decomposition process.

### **Objectives**

To bury decomposition bags containing equal volumes of beech or ash leaves in two locations – a beech wood and a conifer plantation;

To collect bags left by a previous group and determine the amount of leaf material which has been decomposed in each;

To measure and record soil temperature, pH, texture and moisture in two contrasting woodlands, and identify which soil factors are influential in the rate of decomposition;

To sample the detritivore community at the two locations and construct a woodland invertebrate food web.

### **Hypotheses**

The rate of decomposition of ash leaves will be greater than that of beech leaves. This is because ash leaves are shed from the trees still green – ie. containing chlorophyll, and few waste products – whilst the leaves of the beech are shed brown – ie. containing high levels of waste products such as tannins and polyphenols;

The rate of decomposition will be greater in the beech wood than the conifer wood, as the soil factors will be more favourable. The soil pH will be more neutral, as conifer needles acidify the soil by releasing organic humic acids when they decompose. The soil will be warmer and moister as the woodland is more open and sunlight and rain can get to the ground.

There will be a more diverse animal community in the beech wood compared to in the conifer wood. This is because the conifers are an introduced species and are unlikely to have a well-established associated soil fauna.

### **Equipment**

Soil pH kit and trowel

Safety blades

Soil thermometer

20x20mm templates

Soil texture chart

Bags of ash and beech leaves

Cotton fabric

Marker sticks

Hair nets or 10x10mm mesh

String

1x1mm mesh (fly netting or plain net curtain fabric works well)

Scissors

Tape measure

Cotton and needles or stapler

Pooters

Large white trays

Sweep nets

Plastic bags

Plastic funnels, beakers and table lamps for simple tulgren funnels

Identification sheets for invertebrates

### **Method**

Set up decomposition bags:

Each group makes up the decomposition bags for the following group. The class makes up 36 decomposition bags. This is enough for decomposition bags for three replicates of each treatment in the two woodlands.

Allocate responsibility for the bags to each group. In total, make up:

- 6 bags from cotton fabric @ containing 10 squares of beech leaf
- 6 bags from 1x1mm mesh @ containing 10 squares of beech leaf

- 6 bags from 10x10mm mesh @ containing 10 squares of beech leaf
- 6 bags from cotton fabric @ containing 10 squares of ash leaf
- 6 bags from 1x1mm mesh @ containing 10 squares of ash leaf
- 6 bags from 10x10mm mesh @ containing 10 squares of ash leaf

Cut the leaf squares carefully around the 20 x 20mm templates, using a safety blade. Tie the bags to the marker sticks and label the marker sticks 1 to 18, where:

- ✓ # 1 = 0.1 x 0.1mm fabric, ash leaves (replicate 1)
- ✓ # 2 = 1 x 1mm fabric, ash leaves (replicate 1)
- ✓ # 3 = 10 x 10mm fabric, ash leaves (replicate 1)
- ✓ # 4 = 0.1 x 0.1mm fabric, ash leaves (replicate 2)
- ✓ # 5 = 1 x 1mm fabric, ash leaves (replicate 2)
- ✓ # 6 = 10 x 10mm fabric, ash leaves (replicate 2)
- ✓ # 7 = 0.1 x 0.1mm fabric, ash leaves (replicate 3)
- ✓ # 8 = 1 x 1mm fabric, ash leaves (replicate 3)
- ✓ # 9 = 10 x 10mm fabric, ash leaves (replicate 3)
- ✓ # 10 = 0.1 x 0.1mm fabric, beech leaves (replicate 1)
- ✓ # 11 = 1 x 1mm fabric, beech leaves (replicate 1)
- ✓ # 12 = 10 x 10mm fabric, beech leaves (replicate 1)
- ✓ # 13 = 0.1 x 0.1mm fabric, beech leaves (replicate 2)
- ✓ # 14 = 1 x 1mm fabric, beech leaves (replicate 2)
- ✓ # 15 = 10 x 10mm fabric, beech leaves (replicate 2)
- ✓ # 16 = 0.1 x 0.1mm fabric, beech leaves (replicate 3)
- ✓ # 17 = 1 x 1mm fabric, beech leaves (replicate 3)
- ✓ # 18 = 10 x 10mm fabric, beech leaves (replicate 3)

Repeat this for the second set of bags. Go out and bury the decomposition bags in the two woodlands, one set of bags (1 to 18) in each woodland. Bury them in a 3 x 6 grid, 2m apart, and mark the position of the grid with a pole, for easy finding later.

- Collect the set of decomposition bags for you to assess.
- Measure and record the soil pH, temperature, soil texture and moisture at three sites within the grid where the bags were collected.
- Using pooters and sweep nets, carry out systematic sampling of the woodland floor. Empty invertebrates into a large white plastic tray for identification. Make a list of invertebrates found, and an approximate

count. It may be possible to rig up some simple tulgren funnels at the Centre – bring soil and leaf litter samples back to the Centre.

- Distribute the decomposition bags between the groups. Make sure that the contents of the bag are always known – it will correspond to the label on the marker stick – don't remove these until the last minute!
- (Results sheet 1) Each group should carefully open their decomposition bag one at a time and assess the amount of leaf square which has been decomposed by laying the disc onto a graph paper template and counting the number of grid squares that are missing from the leaf template.
- If asked to do so by your group leader, return the leaf material to the decomposition bags and reseal using needle and thread or a stapler. Reattach the marker stick using string.
- Set up a few simple tulgren funnels using funnels, table lamps and beakers. Place a soil and leaf litter sample onto a piece of 10 x 10cm mesh in the bottom of a funnel and prop in a beaker containing a little water. Shine the light over the funnel and leave over night (if possible). The invertebrates will be driven down the funnel, away from the light and heat from the lamp. They will collect in the beaker. Identify them using the ID sheet.

**Recording Sheet 1**

**Soil Factors**

Beech Wood	1	2	3	Average
Soil Factor				
Soil Temp. (oC)				
Soil Moisture (Collect sample for drying)				
Soil pH				
Soil texture				

conifer plantation	1	2	3	Average
Soil Factor				
Soil Temp. (oC)				
Soil Moisture (Collect sample for drying)				
Soil pH				
Soil texture				

### *Data Presentation and Analysis*

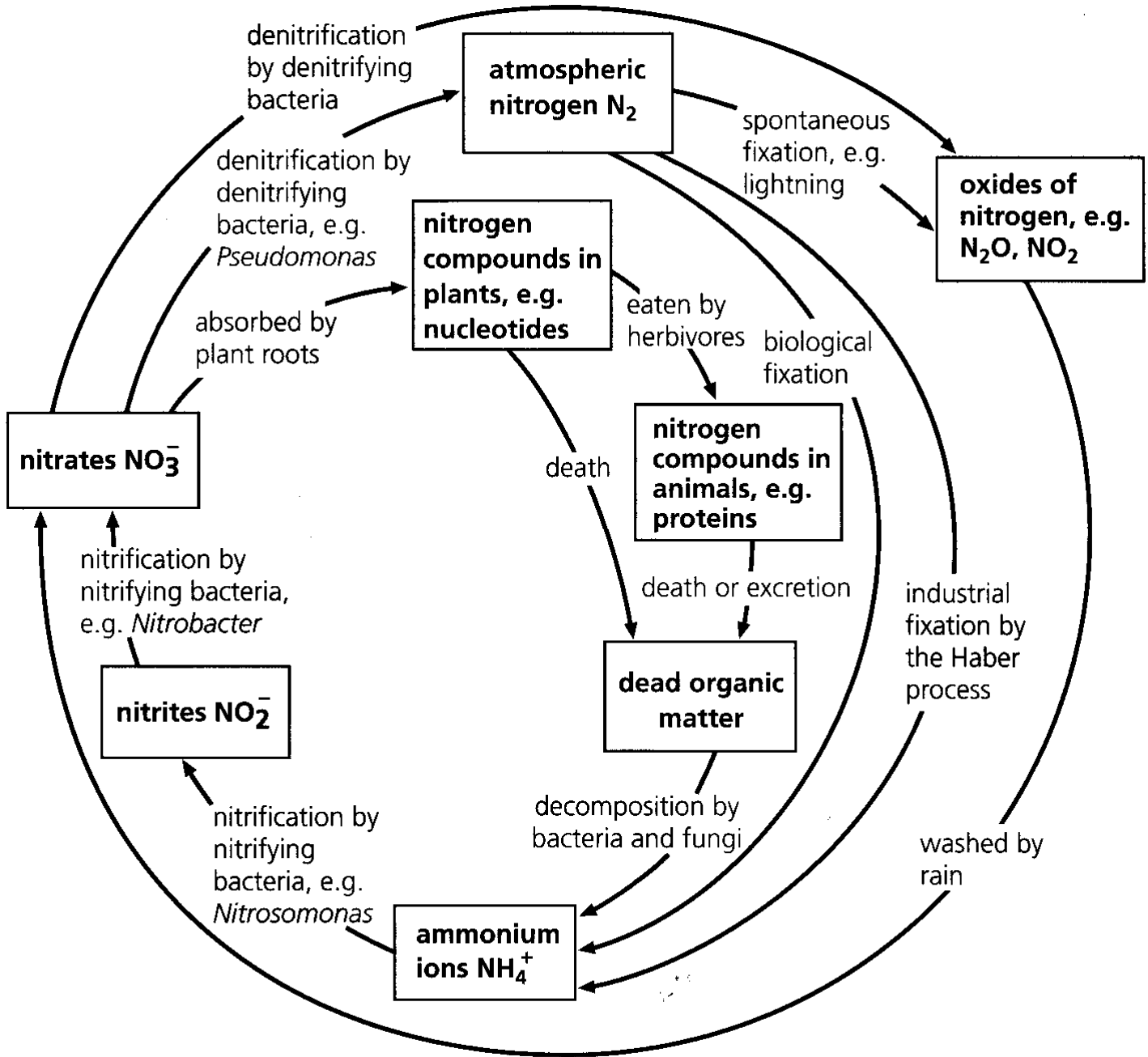
- Open each decomposition bag, carefully noting it's number. Lay each leaf square onto the acetate quadrat and count how many squares have been decomposed. Work out an average % decomposition for the ten squares. Enter this figure onto the computer spreadsheet.
- Carry out a chi square test to determine whether the decomposition rate between the two woodlands and two leaf types is significantly different.
- Using excel, present the soil data for the two sites as bar charts – one factor to each chart.
- Make a species list of soil invertebrates found and construct a woodland food web. Which invertebrates would have been excluded by which treatment? Can you categorise the detritivores into macro, meso and micro-invertebrates?

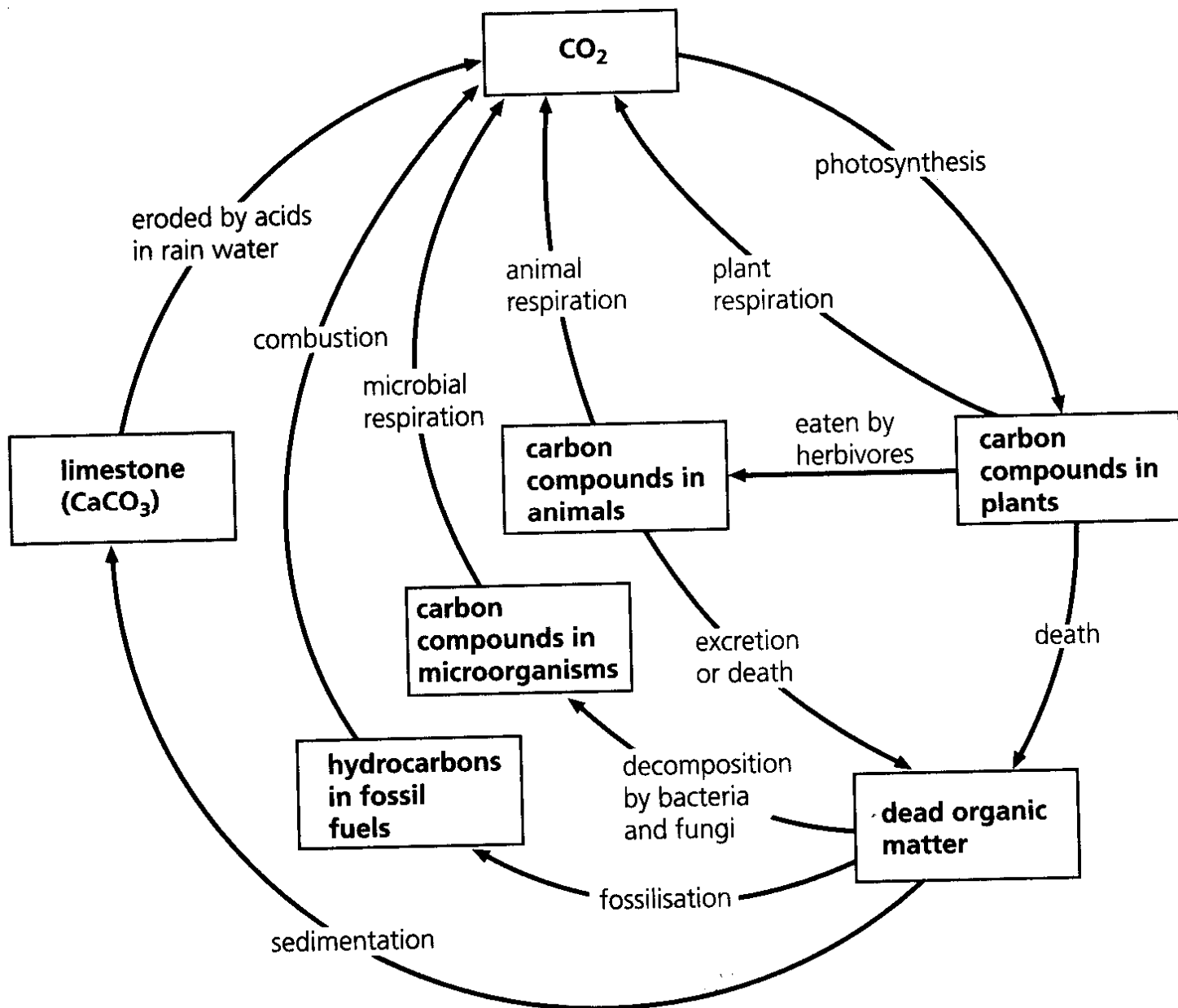
### *Discussion Points*

- Was there any possible source of error in the investigation that has been undertaken? Is there any possibility that there could have been:
  - Human error?
  - Equipment error?
  - Problems inherent with the techniques used?
- Was there a significant difference in the decomposition rates on the two leaf types? Why is this?
- Did the decomposition bags decompose differently in the two woodlands? What differences in the soil factors could account for these differences? What are the ideal soil conditions for decomposition?
- Which invertebrate detritivores were excluded by the different treatments? How did exclusion of the detritivores influence the rate of decomposition of the leaf squares?

Appendix 1 The Nitrogen Cycle

From Chenn (1999)





From Chenn (1999)