

Effect of mineral deficiency on plant growth



Aim

To investigate the effect of mineral deficiencies towards the growth of Lemna sp

Introduction

Plants need water, carbon dioxide and sunlight to synthesise carbohydrates during photosynthesis. To synthesise nutrients and other organic substances, plants need additional elements. Mineral elements are one of the essential chemical elements required by plants in order to achieve optimal growth and development. Mineral elements are mostly obtained in the form of inorganic ions from the soil through their roots. Most of the time, the mineral ions in soil water are present only in low concentration, often lower than that in cytoplasm of root cells. So, in order to obtain mineral ions into the cells, they are taken up selectively against the concentration gradient by active transport, using energy from the respiration of plants. The uptake process occurs with the help of the specific protein pumps in the plasma membrane of the root cells.

This usually results in accumulation of mineral ions in the root cells. The mineral ions are then carried in the apoplast or symplast pathway until they reach the endodermis containing impermeable Casparian strip. They then enter the cytoplasm of cells either by diffusion or active transport and reach the xylem of the plant to be transported in the water that moves up continuously in the transpiration stream. Once the mineral ions reach the tissues where they are needed, they move out of the xylem into the cells either by diffusion or active transport, depending on the permeability of cell membranes and relative concentrations of ions inside and outside the cells.

Generally, mineral elements required by plants can be divided into two categories, macronutrients and micronutrients. Macronutrients can be broken into two more groups: primary and secondary nutrients. The primary nutrients are nitrogen (N), phosphorus (P), and potassium (K). These major nutrients usually are lacking from the soil first because plants use large amounts for their growth and survival. The secondary nutrients are calcium (Ca), magnesium (Mg), and sulfur (S). There are usually enough of these nutrients in the soil so fertilization is not always needed. Also, large amounts of Calcium and Magnesium are added when lime is applied to acidic soils.

Sulfur is usually found in sufficient amounts from to slow decomposition of soil organic matter, an important reason for not throwing out grass clippings and leaves. Nitrogen is a part of all living cells and is a necessary part of all proteins, enzymes and metabolic processes involved in the synthesis and transfer of energy. Nitrogen is a part of chlorophyll, the green pigment of the plant that is responsible for photosynthesis. Helps plants with rapid growth, increasing seed and fruit production and improving the quality of leaf and forage crops. Nitrogen often comes from fertilizer application and from the air (legumes get their N from the atmosphere, water or rainfall contributes very little nitrogen).

Like nitrogen, phosphorus (P) is an essential part of the process of photosynthesis. Involved in the formation of all oils, sugars, starches, etc. Helps with the transformation of solar energy into chemical energy; proper plant maturation; withstanding stress. Effects rapid growth. Encourages blooming and root growth. Phosphorus often comes from fertilizer, bone meal, and superphosphate. Potassium is absorbed by plants in larger

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amounts than any other mineral element except nitrogen and, in some cases, calcium. Helps in the building of protein, photosynthesis, fruit quality and reduction of diseases. Potassium is supplied to plants by soil minerals, organic materials, and fertilizer. Calcium, an essential part of plant cell wall structure, provides for normal transport and retention of other elements as well as strength in the plant. It is also thought to counteract the effect of alkali salts and organic acids within a plant. Sources of calcium are dolomitic lime, gypsum, and superphosphate. Magnesium is part of the chlorophyll in all green plants and essential for photosynthesis. It also helps activate many plant enzymes needed for growth. Soil minerals, organic material, fertilizers, and dolomitic limestone are sources of magnesium for plants. Sulfur is essential plant food for production of protein. Promotes activity and development of enzymes and vitamins. Helps in chlorophyll formation. Improves root growth and seed production. Helps with vigorous plant growth and resistance to cold. Sulfur may be supplied to the soil from rainwater. It is also added in some fertilizers as an impurity, especially the lower grade fertilizers.

Lemna is a genus of free-floating aquatic plants from the duckweed family. These rapidly-growing plants have found uses as a model system for studies in community ecology, basic plant biology, in ecotoxicology, in production of biopharmaceuticals, and as a source of animal feeds for agriculture and aquaculture.

The duckweeds have been classified as a separate family, the Lemnaceae, but some researchers (the AGP II) consider the duckweeds members of the Araceae. Lemna species grow as simple free-floating thalli on or just beneath

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the water surface. Most are small, not exceeding 5 mm in length, except *Lemna trisulca* which is elongated and has a branched structure. *Lemna* thalli have a single root, which distinguishes them from related genera *Spirodela* and *Landoltia*.

The plants grow mainly by vegetative reproduction: two daughter plants bud off from the adult plant. This form of growth allows very rapid colonisation of new water. Duckweeds are flowering plants, and nearly all of them are known to reproduce sexually, flowering and producing seed under appropriate conditions. Certain duckweeds (e. g. *L. gibba*) are long day plants, while others (e. g. *L. minor*) are short day plants. When *Lemna* invades a waterway, it can be removed mechanically, by the addition of herbivorous fish (e. g. grass carp) or treated with a herbicide. The rapid growth of duckweeds finds application in bioremediation of polluted waters and as test organisms for environmental studies. It is also being used as an expression system for economical production of complex biopharmaceuticals. Duckweed meal (dried duckweed) is a good cattle feed. It contains 25-45% proteins (depending on the growth conditions), 4.4% fat, and 8-10% fibre, measured by dry weight.

Lemna has been transformed by molecular biologists to express proteins of pharmaceutical interest. Expression constructs were engineered to cause *Lemna* to secrete the transformed proteins into the growth medium at high yield. Since the *Lemna* is grown on a simple medium, this substantially reduces the burden of protein purification in preparing such proteins for medical use, promising substantial reductions in manufacturing costs. In addition, the host *Lemna* can be engineered to cause secretion of proteins

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with human patterns of glycosylation, an improvement over conventional plant gene-expression systems. Several such products are being developed, including monoclonal antibodies.

Problem Statement: How do the deficiencies of minerals affect the growth of Lemna sp ?

Hypothesis: Lemna plants grow healthily in complete culture solution. When there is deficiency of certain minerals, Lemna plants will show the symptoms of deficiency of that particular minerals.

Variables

Manipulated: Type of mineral deficiencies

Responding: The growth of Lemna sp

Fixed: Intensity of sunlight, type, size and number of Lemna plants used, concentration of carbon dioxide and volume of solution

Apparatus

- Petri dishes
- Petri dishes cover
- forceps
- measuring cylinder
- droppers.

Materials

A range of solutions including solutions with

all solutions present

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lacking nitrogen, NO_3^-

lacking phosphate, PO_4^{3-}

lacking potassium, K^+

lacking magnesium, Mg^{2+}

lacking calcium, Ca^{2+}

lacking zinc, Zn^{2+}

lacking iron, Fe^{2+}

Lemna

Stickers

Tissue paper

Procedures

One petri dish and its cover is washed using water and dried using tissue paper.

Four pairs of Lemna sp are picked out using a forceps and placed inside the clean petri dish. This step is done with extra care as to minimise the damage done to the Lemna sp.

Step 1 and 2 are repeated 8 times to prepare 8 petri dishes , each containing 4 pair of Lemna sp.

The culture solutions are measured at 15ml using a measuring cylinder.

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Every petri dish is then filled with different culture solutions with different mineral deficiencies, NO_3^- , PO_4^{3-} , K^+ , Mg^{2+} , Ca^{2+} , Zn^{2+} , Fe^{2+} .

The last petri dish is filled with perfect culture solution in which all the minerals are present.

All the petri dishes are placed at a spot where light intensity is just sufficient for the Lemna sp to grow.

The Lemna sp are observed carefully for a full 10days of study.

The Lemna sp. are observed for the number of leaves , the colour of leaves and the number of plants with roots.

The observations or data are recorded into a table.

Results

Day 2

Number of live plants: 4

Number of green leaves: 10

Number of plants with roots: 1

Growth abnormalities: None

Day 4

Number of live plants: 6

Number of green leaves: 17

Number of plants with roots: 3

Growth abnormalities: None

Day 6

Number of live plants: 12

Number of green leaves: 28

Number of plants with roots: 6

Growth abnormalities: None

Day 8

Number of live plants: 15

Number of green leaves: 36

Number of plants with roots: 11

Growth abnormalities: None

Day 10

Number of live plants: 20

Number of green leaves: 57

Number of plants with roots: 13

Growth abnormalities: None

Table 1 Data for normal culture solution (Control experiment)

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Number of live plants:

Number of green leaves:

Number of plants with roots:

Growth abnormalities:

Day 2

Number of live plants: 4

Number of green leaves: 8

Number of plants with roots: 21

Growth abnormalities: None

Day 4

Number of live plants: 4

Number of green leaves: 11

Number of plants with roots: 2

Growth abnormalities: Leaves turn pale green

Day 6

Number of live plants: 6

Number of green leaves: 17

Number of plants with roots: 4

Growth abnormalities:

- Growth of root stunted
- Leaves turn yellowish

Day 8

Number of live plants: 7

Number of green leaves: 22

Number of plants with roots: 5

Growth abnormalities:

- Growth stunted
- Leaves turn paler and yellowish

Day 10

Number of live plants: 8

Number of green leaves: 27

Number of plants with roots: 6

Growth abnormalities:

- Growth of root stunted
- Leaves turn very pale green or yellowish

Table 2 Data for culture solution lacking nitrate ions

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Day 2

Number of live plants: 4

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Number of green leaves: 8

Number of plants with roots: 0

Growth abnormalities: None

Day 4

Number of live plants: 6

Number of green leaves: 13

Number of plants with roots: 1

Growth abnormalities: None

Day 6

Number of live plants: 7

Number of green leaves: 16

Number of plants with roots: 2

Growth abnormalities: Leaves turn pale green

Day 8

Number of live plants: 9

Number of green leaves: 20

Number of plants with roots: 4

Growth abnormalities:

- Leaves turn pale green and yellowish
- Some leaves appear to be twisted

Day 10

Number of live plants: 11

Number of green leaves: 24

Number of plants with roots: 6

Growth abnormalities:

- Most leaves turn yellowish
- Some leaves bleached

Table 3 Data for culture solution lacking of sulphate ions

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Day 2

Number of live plants: 4

Number of green leaves: 8

Number of plants with roots: 0

Growth abnormalities: Edges of leaves turn yellow

Day 4

Number of live plants: 5

Number of green leaves: 10

Number of plants with roots: 1

Growth abnormalities: Leaves turn yellow

Day 6

Number of live plants: 7

Number of green leaves: 15

Number of plants with roots: 2

Growth abnormalities: Some leaves curl and crinkle

Day 8

Number of live plants: 9

Number of green leaves: 19

Number of plants with roots: 3

Growth abnormalities: Leaves turn yellow

Day 10

Number of live plants: 10

Number of green leaves: 22

Number of plants with roots: 5

Growth abnormalities:

- Leaves turn yellow
- Some leaves decompose

Table 4 Data for culture solution lacking of potassium ions

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Day 2

Number of live plants: 4

Number of green leaves: 8

Number of plants with roots: 0

Growth abnormalities: None

Day 4

Number of live plants: 5

Number of green leaves: 10

Number of plants with roots: 0

Growth abnormalities: Leaves turn pale green

Day 6

Number of live plants: 7

Number of green leaves: 19

Number of plants with roots: 1

Growth abnormalities: Leaves turn pale green or yellowish

Day 8

Number of live plants: 8

Number of green leaves: 22

Number of plants with roots: 2

Growth abnormalities: Leaves turn white and yellowish

Day 10

Number of live plants: 9

Number of green leaves: 25

Number of plants with roots: 4

Growth abnormalities: Almost all leaves yellow or bleached

Table 5 Data for culture solution lacking of magnesium ions

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Day 2

Number of live plants: 2

Number of green leaves: 4

Number of plants with roots: 0

Growth abnormalities:

- All plants die
- Areas between leaf veins turn yellow

Day 4

Number of live plants: 0

Number of green leaves: 0

Number of plants with roots: 0

Growth abnormalities: Leaves turn white

Day 6

Number of live plants: 0

Number of green leaves: 0

Number of plants with roots: 0

Growth abnormalities: Leaves turn white

Day 8

Number of live plants: 0

Number of green leaves: 0

Number of plants with roots: 0

Growth abnormalities: Leaves turn white and disintegrate

Day 10

Number of live plants: 0

Number of green leaves: 0

Number of plants with roots: 0

Growth abnormalities: Leaves disintegrate

Table 6 Data for culture solution lacking of calcium ions

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Day 2

Number of live plants: 4

Number of green leaves: 9

Number of plants with roots: 0

Growth abnormalities: None

Day 4

Number of live plants: 6

Number of green leaves: 17

Number of plants with roots: 2

Growth abnormalities: Leaves turn yellowish

Day 6

Number of live plants: 9

Number of green leaves: 23

Number of plants with roots: 6

Growth abnormalities: Some leaves turn pale green

Day 8

Number of live plants: 13

Number of green leaves: 30

Number of plants with roots: 11

Growth abnormalities: Some leaves turn dark green with red or purple spots

Day 10

Number of live plants: 15

Number of green leaves: 34

Number of plants with roots: 12

Growth abnormalities:

Stunted growth

Roots grow poorly

Table 7 Data for culture solution lacking of phosphate ions

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Day 2

Number of live plants: 4

Number of green leaves: 8

Number of plants with roots: 1

Growth abnormalities: None

Day 4

Number of live plants: 7

Number of green leaves: 16

Number of plants with roots: 4

Growth abnormalities: Leaves turn pale green

Day 6

Number of live plants: 11

Number of green leaves: 29

Number of plants with roots: 7

Growth abnormalities: Leaves turn pale green or yellow

Day 8

Number of live plants: 14

Number of green leaves: 32

Number of plants with roots: 10

Growth abnormalities: Some leaves completely bleached

Day 10

Number of live plants: 18

Number of green leaves: 41

Number of plants with roots: 14

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Growth abnormalities: Leaves turn pale green or yellow or completely bleached

Table 8 Data for culture solution lacking of iron ions

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Discussion

In this experiment, we are determining the effect of minerals deficiencies on the growth of Lemna sp. The manipulated variable used is the type of minerals deficiencies. The culture solutions used in this experiment have different minerals deficiencies which are Calcium, Magnesium, Sulfate, Phosphate, Nitrate, Iron, and Potassium ions. There is a solution which does not have any minerals deficiencies. It is a perfect culture solution. This culture solution is used as a control in this experiment to compare the effect of different minerals deficiencies with the perfect culture solution. The conditions of Lemna sp. in other culturesolutions with minerals deficiencies are compared with the Lemna sp. in the control solution. The responding variable is growth of the Lemna sp. where we count the number of leaves and observe the colour of leaves at the end of experiment. Lemna sp. is used because they are easy to find, observe and count the number of leaves. The number and colour of the leaves are recorded into a table. The fixed variable used is the amount of sunlight and air obtained. The petri dishes which are involved in the experiment are placed on a spot where sunlight can reach them. This situation is fixed so that it will not affect the result which is the number and colour of the leaves. Besides, volume of culture solution is also one of the fixed variable. This is an important fixed variable because different volume of culture solution will affect the rate of growth of Lemna

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sp. moreover, different volume of culture solution will also affect the number of leaves at the end of experiment. Thus, constant or fixed volume of each culture solution is needed so that it will give a valid result for the data. Last but not least, type of plant used is also another fixed variable. Different plants have different growth rate. Therefore, Lemna sp. is used because it is easy to find and observe. Every petri dishes need to be synchronised together by using Lemna sp.

Interpretation of Data

The analysis of data can be done by tabulating the data from Table 1. Table 1 show the observations obtained on the growth of Lemna plants after ten days of investigation. Based on the results obtained, it can be seen that the absence of different mineral elements has different effects on growth of Lemna plants. Lemna plants grow into healthy plants in complete culture solution. Their growth increases rapidly as seen from the table with number of live plants increases from 4 to 20, number of green leaves increases from 10 to 57 and number of plants with roots increases from 1 to 13. There is no growth abnormality. This is because normal culture solution provides them with all the necessary mineral. ions at appropriate concentrations for optimal growth.

In culture solution deficient of nitrogen ions, chlorosis takes place as the leaves turn pale, due to lack of chlorophyll as it plays a role as a major component of chlorophyll. Nitrate ion is needed for the formation of amino acid, enzymes and plant hormones. Lacking of nitrate ion causes no amino acids, enzymes and plant hormones produced at all. Photosynthetic enzymes and hormones which is essential for the metabolism of plant cell cannot be

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made. Therefore, the growth is stunted with only minor increase of number of live plants from 4 to 8, and increase in number of green leaves until the end of experiment with a difference of 5 and a notable decrease in number of plants with roots from 21 to 6.

Sulphate ions deficiency affects the production of chlorophyll leading to an overall chlorosis of the leaves with general yellowing of leaves observed. This is observed with a steady increase in number of green leaves, with difference of 4 at the end of experiment, indicating that more leaves are turning yellow. Some leaves appear to be twisted and brittle. Stunted root growth is also observed as towards the end of the experiment, the root growth is constant with difference of 2 plants with roots observed every 2 days.

Potassium ions deficiency shows first in marginal chlorosis, that is yellowing at the edges of the leaves as observed on Day 2. It is also observed that the number of live plants and green leaves increases with a decreasing rate with the leaves curl and being crinkly. Some parts of the plants decompose as seen on Day 10 as premature death occurs because lack of potassium affects various activities of plants such as protein synthesis, carbohydrate metabolism, enzymatic activities and others. Potassium ion is needed for the active transport in the guard cells. Guard cells actively pumps in potassium ion so that the stoma will open as water flow by osmosis from low solute concentration region to high solute concentration region. Lacking of potassium ion causes the stoma to close. This eventually affect the rate of photosynthesis. Sugars cannot be made and the metabolism of plant cells is

affected. Therefore, the growth rate is affected and the number of leaves at the end of experiment is reduced.

As magnesium is an essential part of the chlorophyll molecule, plants lacking of magnesium show chlorosis in which all the leaves turn yellow and eventually die. In the absence of magnesium, the synthesis of the chlorophyll is inhibited. This is shown with the small and decreasing difference in number of live plants and green leaves towards the end of the experiment, illustrating that number of leaves turning yellow or white increases, because of chlorosis.

In culture solution deficient of calcium ions, areas between leaf veins become yellow are observed on second day. The leaves have distorted shape. Growth of the leaves is stunted and the growing points die back too as lack of calcium affects plant cell growth and enzymatic reactions. This is shown with no more living Lemna plants from Day 4 onwards as all the leaves turn white and disintegrate. This happens because calcium ion is needed for the formation of cell wall during cell division. Lacking of calcium ion will cause no cell division occur as calcium pectate cannot be formed. Permeability of the cell is also affected. Lacking of calcium causes the cell sap and the cytosol diffuse out of the cell. This will cause the death of cells.

Phosphorus deficiency results the leaves turning to dark green color with red or purple spots on them surfacing on the day 8 of the experiment. They grow slowly and their roots grow poorly as compared to others with only difference of 1 plant with root between Day 8 and 10. This happens because phosphorus is necessary in almost all aspects of growth and metabolism in

plants. In culture solution deficient of iron, there is chlorosis (yellowing of leaves) at the base of the leaves, leading to some completely bleached leaves.

Deficiency in iron leads to decrease in chlorophyll molecules, causing chlorosis. However, in this experiment, the number of lives plants, green leaves and plants with roots generally increases instead of decreases. Ferum ion is needed for the formation of chlorophyll. Lacking of ferum ion causes no synthesis of chlorophyll. No production of chlorophyll will cause the colour of leaves to change as the colour of chlorophyll is green.

Source of errors and ways to overcome these error:

Errors are present when the experiment is being conducted, leading to discrepancy and inaccuracy in results obtained.

No measurement is exact. All types of measurement will have some degree of error or uncertainty. Generally, errors can be divided into systematic errors and random errors. Systematic errors are cumulative errors that can be corrected, if known. Random errors are errors arise from unknown and unpredictable variations in condition while carrying out the experiment.

Random errors may be due to human limitations, lack of sensitivity, natural environment and use of wrong technique of measurement. Random errors are present in this experiment. Thus, it is best to be minimized by repeating the experiment a couple of times.

Parallax error is one example of random errors. It is an error in reading an instrument when the observer's eyes are not in a line perpendicular to the plane of the scale of measuring instrument. For this experiment, the culture

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solution placed into each petri dish may not be exactly 15cm³. This happens when the position of the eyes is not directly perpendicular to the meniscus of the culture solution in the 10cm³ measuring cylinder. Varying volume of culture solution in different petri dishes may slightly affect the growth of the Lemna plants as the higher the volume of culture solution, the higher the amount of mineral nutrients present, the higher the rate of growth of Lemna plants. Thus, to improve the accuracy of data, parallax error should be avoided while taking any measurement in the experiment.

Furthermore, human errors like being too harsh when handling the Lemna plants is also one of the sources. This may do damage to the plants and greatly affects the survival rate and the growth of the plant. The observers may also wrongly count the number of leaves leading to unreliable results being produced.

Limitations:

The impurities found in the culture solution will affect the validity of the result. This occurs when the culture solution is being prepared. The impurities contained inside the water will enter the culture solution and hence contaminate the solution. This varies the amount of minerals found within each solution and this may affect the growth of Lemna sp. In addition, there is also limitation from the visual method of diagnosis used in obtaining results of the experiment. This is because symptoms of certain mineral deficiencies may be suppressed by other factors besides lack of certain mineral elements. For example, the weather conditions such as light illumination. Light illumination to which the Lemna plants are exposed to may vary when a few petri dishes are placed too close together or on top of

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another, blocking each other, affecting the amount of light received by the Lemna plants in the petri dishes. All these may lead to wrong results and conclusions being obtained. Thus, weather conditions should be ensured being one of the constant variables in this investigation too. Besides, the disease-causing microorganisms will cause the death of the Lemna sp. indirectly. This will also affect the number of leaves at the end of experiment obtained and the observers may be ignorant for this fact and attribute the death of the Lemna plants to the deficiencies of certain minerals.

Intraspecific competition is also present among the Lemna plants. They may compete for all the known necessities, like water, air sunlight as well as minerals. Those that failed to compete maybe wiped out and hence lead to death. This point also points out that the death in number of Lemna plants may not be completely due to the deficiencies of minerals in plants.

Human limitation is one of the limitation and random errors in this experiment. The results of the experiment may have been affected due to the limitation in the observer's observation skills when observing the growth of plants. Although the deficiency symptoms shown by Lemna plants for each mineral elements investigated are quite visually distinct, mistakes can still occur especially when they are viewed by different observers. For example, different observers may misidentify and count different number of Lemna leaves in a petri dish. Besides that, observer may also mixed up between different deficiency symptoms shown by Lemna plants especially those complicated ones, leading to inaccuracy of results. To minimize such error, the observations on the Lemna plants should be carried