COURSE GUIDE

SLM 505
SOIL MICROBIOLOGY AND BIOCHEMISTRY

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INTRODUCTION

SLM 505 a two credit course is a comprehensive course for the study of soil microbiology and biochemistry, designed for use by advanced undergraduate students who wish to have an overview of the subject to those who need to have a comprehensive knowledge of the field of soil microbiology and biochemistry.

The course has 25 units divided into five broad subject areas, starting with background and continuing through the soil biota, interactions between organisms, microbial transformations, biochemistry (enzymatic activities) and concluding with the impact of human processes through pesticide applications. These topics therefore will attempt to improve the understanding required to provide a foundation for the inter-disciplinary approaches that will continue to provide exciting new ideas in the field of agriculture.

You are encouraged to devote, at least two hours studying each of the 25 units. You are also advised to attempt the self-assessment exercises as they are important for better understanding of the units. You are advised also to pay attention to the Tutor – Marked Assignments (TMA) (details provided in a separate file).

There will be tutorial classes. Details of location of the tutorials and time will be made known to you; this is a great opportunity for you to have direct contact with your course coordinator. Areas not understood in the course of study will be properly explained.

WHAT YOU WILL LEARN IN THIS COURSE

The overall aim of this course is for you to have a comprehensive knowledge of the field of soil microbiology and biochemistry. An understanding of this course is important to further understanding agricultural and environmental impacts on life. During the course therefore, you will learn about the importance of microorganisms in the soil environment and you will be furnished with knowledge to solve issues related to the functions of other biological systems on earth – biochemistry (the chief support or engine room of soil microbial functioning).

COURSE AIMS

The aim of the course is to give you a better and comprehensive understanding of the microorganisms living in the soil and their activities that are of agricultural and environmental significance. The aim of the course will be achieved by:

- Introducing you to the origin and contributions of soil microbiology to agriculture.
- Introducing you to the organisms in the soil.
- Helping you to understand the importance of soil organisms.
- Giving an insight into the microbiology of the rhizosphere.
- Exposing you to the interrelationships of organisms in the soil.
- Helping you to understand transformations mediated by microorganisms.
- Helping you to understand biochemical functioning in the soil environment.
- Introducing you to application of pesticides and their importance in agricultural ecosystems.
- Helping you to understand the effects of pesticides on soil microflora.

COURSE OBJECTIVES

To achieve the aims mentioned above, the course sets overall objectives. In addition, each unit has its specific objectives. The unit objectives are given at the beginning of the unit; you should read them before you start working through the unit. You may also want to refer to them during your study of the unit so as to check on your progress. You should always look at the unit objectives after completing a unit. By this, you can be sure that you have done what was required of you by the unit.

Below are the broader objectives of the course, as a whole. By meeting these objectives, you should have achieved the aims of the course as a whole. On completion of the course, you should successfully be able to:

- define soil microbiology and soil biochemistry
- explain the importance of soil microbiology and biochemistry in terms of their contributions to agriculture
- explain the major types of soil organisms in the soil habitat and their beneficial roles in agriculture
- explain the factors affecting soil organisms
- explain microbiology of the rhizosphere concepts
- discuss the effect of plants on microorganisms
- describe the types of microbial soil interactions
- explain transformations mediated by microorganisms such as organic matter, nitrogen, phosphorus, sulphur and iron
- define and classify enzymes
- state the enzymes important in agricultural ecosystem and why?
- discuss the functions of enzymes
- discuss the factors affecting enzyme activities
- define basic concepts in enzyme reactions

- explain enzyme kinetics and the transformations of Michaelis-Menten equation
- explain enzyme inhibition and various types of inhibition
- define pesticide and state the types of pesticides
- explain the merits and demerits of pesticides
- discuss microbial degradations of pesticides
- discuss the effects of pesticides on soil microorganisms.

WORKING THROUGH THIS COURSE

To complete this course you are required to read the study units, as well as other related materials. Each unit contains self-assessment exercises, and at certain points in the course, you are required to submit assignments for assessment purposes. At the end of the course, you are going to sit for a final examination. The course should take you about 18/ 21 weeks in total to complete. Below you will find listed all the components of the course, what you have to do and how you should allocate your time to studying the course.

COURSE MATERIAL

The first five units center on the fundamentals and brief history of soil microbiology and biochemistry and the organisms that live in the soil; **Module 1**.The next five units that address the microbiology of the rhizosphere and the interactions among microorganism and between microorganisms and plants;**Module 2**has units 11 – 15 highlights the microbial transformation of cycles in soil and factors affecting them – **Module 3** and**Module 4** comprising of units 16 – 18 deals with enzymes and their reactions in soil; while **Module 5**having units 19 – 22 deals with the effect of pesticides, especially on soil microorganisms.

The major components of the course include the following:
1. Course Guide
2. Study Units
3. Textbooks and References
4. Assignment File
5. Presentation Schedule

STUDY UNITS

There are 25 study units in this course as follows:

Module 1 Fundamentals

Unit 1 Introduction and Brief History of Soil Microbiology

Unit 2 Introduction to Soil Biochemistry
Unit 3 TheSoil Habitat
Unit 4 Microbial Composition of Soil (Soil Organisms)
Unit 5 Physiology of Microbial Growth

Module 2 Microbiology of the Rhizosphere and Microbial Interactions

Unit 1 RhizosphereSoil
Unit 2 Soil Microbial Interactions
Unit 3 Non-Symbiotic VsSymbiotic Associations
Unit 4 Rhizobium Association
Unit 5 MycorrhizalAssociation

Module 3 Microbial Transformations of Cycles in Soil

Unit 1 Soil Organic Matter Transformation
Unit 2 Nitrogen Transformation
Unit 3 Phosphorus Transformation
Unit 4 SulphurTransformation
Unit 5 Micronutrient Iron (Fe) Transformation

Module 4 Soil Enzymes

Unit 1 What are Enzymes?
Unit 2 Enzymes Classification and Nomenclature
Unit 3 Soil Enzymes Importance in Agriculture
Unit 4 Kinetics of Enzyme Reactions in Soil
Unit 5 Enzyme Inhibition

Module 5 Pesticides in Soil

Unit 1 Nature of Pesticides
Unit 2 Behavior of Pesticides in Soil
Unit 3 Effect of Pesticides on Soil Organism
Unit 4 Biodegradation of Pesticides
Unit 5 Pesticide Persistence in Soil

Text Books and References

Experience in teaching this subject has shown that you often stumble over many new terms that are introduced during the course of study. To help you overcome this obstacle, a comprehensive description or explanation of soil microbiology terms is given under each unit or where appropriate.

You should note that bolded terms in each unit are defined in the text. Recommended textbooks and references are listed below:

1) Alexander, M. (1991). *Introduction to Soil Microbiology*. (2nd ed.). Wiley, New York.
2) Alexander, M. (1994). *Biodegradation and Bioremediation*. Wiley, New York.
3) Atlas, R.M. &Bartha, R. (1981). *Microbial Ecology: Fundamentals and Applications*.Reading, Massachusetts:Addison-Wesley Publishing Company.
4) Bala, A. (2012). *Lead Farmer Training*.In:*Biological Nitrogen Fixation and Grain Legume Enterprise*.Adapted by AbdullahiBala for the Training ofTrainers and Lead Famers within theN2Africa West Africa Outreach Project..
5) Brady, N.C.&Weil, R.R. (2002). *The Nature and Properties of Soils*.(13th ed.).Prentice Hall, Upper Saddle River.
6) Eldor, A.P. (Ed.). (2007). *Soil Microbiology, Ecology and Biochemistry*. (3rd ed.). Canada:Academic Press Publications..
7) Hassan, G.D. (2010). *Soil Microbiology and Biochemistry*. New India Publishing Agency.
8) Lynch, J.M. &Hobbie, J..E. (1988). *Microorganisms inAction: Concepts andApplication*.In:*Microbial Ecology*. Blackwell, Oxford.
9) Maier, R. M; Pepper, I. L&Gerba, C. P. (2009). *Environmental Microbiology*.San Diego, California:Academic Press.. 589pp.
10) Nannipieri, P., Kandeler, E. &Ruggiero, P. (2002). *Enzyme Activities and Microbiological and Biochemical Processes in Soil*. In: R.S. Burns andR.P. Dick (Eds.). "*Enzymes inThe Environment Activity, Ecology And Applications*. New York:Dekkers.
11) Nelson, D.L. &Cox, M.M. (2002). *Lehninger Principles of Biochemistry*. (3rd ed.). New York: New Delhi Worth Publishers.
12) Palmer, T. (2001). *Enzymes: Biochemistry, Biotechnology and Chemical Chemistry*. (3rd ed.). England: Horwood Publishing, Chichester.
13) Price, N.C. &Stevens, L. (1999). *Fundamentals ofEmzymology*. (3rd ed.). Oxford:Oxford University Press.
14) Soil Science Society ofAmerica (1997). Glossary of Soil Science Terms. SSSA, Madison, WI. 138pp. Available Online At Www.Soils.Org/Sssagloss/.
15) Sylvia, D.M., Hartel, P.G., Fuhrmann, J.J. &Zuberer, D.A. (2005). *Principles and Applications of Soil Microbiology*. (2nd ed.).New Jersey: Prentice Hall, Upper Saddle River.
16) Wilson, K. &Walker, J. (Eds.). (2000). *Principles and Techniques of Practical Biochemistry*. (5th ed.). Cambridge University Press.

ASSIGNMENT FILE

In this file, you will find the details of the work you must submit to your tutor for marking. The marks you obtain will form part of your total score for this course.

ASSESSMENT

There are two aspects to the assessment of this course. The first being the Tutor-Marked Assignments (TMA) and the second the final written examination. You are advised to attempt the exercises as this will help you apply the information, knowledge and techniques learnt during the course.

The assignments must be submitted to your tutor for formal assessment in accordance with the deadlines stated in the presentation schedule and assignment file. The work you submit to your tutor for assessment will count for 30% of your total course mark. At the end of the course, you will sit for a final examination; this examination will count for 70% of your total course mark.

TUTOR – MARKED ASSIGNMENT

You are advised to submit your assignments as required. Each assignment counts for 6% of your marks for the course. You should be able to complete your assignment from the information obtained from the study units and other recommended texts. However, it is advisable that you research more and study other references as this will give you a broader view point and many provide a deeper understanding of the subject.

When you complete each assignment, send it to your tutor. Make sure that each assignment reaches your tutor on or before the deadline given in the presentation schedule and assignment file. If for any reason, you cannot complete your work on time, contact your tutor to discuss the possibility of an extension. Extensions will not be granted after the due date, except for exceptional circumstances.

FINAL EXAMINATION AND GRADING

The final examination for this course will take three hours and have a value of 70% of the total course grade. The examination will consist of questions which reflect the types of self-assessment exercises and tutor-marked assignments you have previously encountered. All areas of the course will be assessed. Take time to revise the entire course before the examination. The examination covers all parts of the study units.

PRESENTATION SCHEDULE

Your course materials will give you important dates for attending tutorials and the timely completion and submission of your tutor-marked assignments. Remember that you are required to submit all assignments by the due date. Please guard against lagging behind in your work.

COURSE MARKING SCHEME

The following table shows how the marking scheme is divided.

Table 1: Course Marking Scheme

Assignment	**Marks**
Assignments 1 – 5	Five assignments count for 6% each = 30% of course marks
Final examination	70% of overall course marks
Total	**100% of course marks**

COURSE OVERVIEW

This table brings together the units, the number of weeks you should take to complete them and the assignment that follows.

Table 2: Course schedule

Units	Title of Work	Weeks activity	Assessment (end of each module)
	Course Guide		
	Module 1: Fundamentals		
1	Introduction and Brief History of Soil Microbiology	1	Assignment 1
2	Introduction to Soil Biochemistry	1	
3	The Soil Habitat	1	
4	Microbial Composition of Soil (Soil Organisms)	1	
5	Physiology of Microbial Growth	1	

Units	Title of Work	Weeks activity	Assessment (end of each module)
	Module 2: Microbiology of the Rhizosphereand Microbial Interactions		
1	RhizosphereSoil		
2	Soil Microbial Interactions	1	
3	Non-Symbiotic VsSymbiotic	1	Assignment 2

	Associations		
4	Rhizobium Association	1	
5	MycorrhizalAssociation	1	

Units	Title of Work	Weeks activity	Assessment (end of each module)
	Module 3: Microbial Transformations of Cycles in Soil		
1	Soil Organic Matter Transformation	1	Assignment 3
2	Nitrogen Transformation	1	
3	Phosphorus Transformation	1	
4	SulphurTransformation	1	
5	Micronutrient Iron (Fe) Transformation	1	

Units	Title of Work	Weeks activity	Assessment (end of each module)
	Module 4: Soil Enzymes		
1	What are Enzymes?	1	Assignment 4
2	Enzymes Classification and Nomenclature	1	
3	Soil Enzymes Importance in Agriculture	1	
4	Kinetics of Enzyme Reactions in Soil	1	
5	Enzyme Inhibition	1	

Units	Title of Work	Weeks activity	Assessment (end of each module)
	Module 5: Pesticides in Soil		
1	Nature of Pesticides	1	Assignment 5
2	Behavior of Pesticides in Soil	1	
3	Effect of Pesticides on Soil Organism	1	
4	Biodegradation of Pesticides	1	
5	Pesticide Persistence in Soil	1	
	Revision	1	
	Examination	25	

HOW TO GET THE MOST FROM THIS COURSE?

In distance learning, the study units replace the conventional university lecturer. This is one of the advantages of distance learning; you can read and work through specially planned study materials at your own pace, time and place that suits you best.

Each of the study units follows a common format. The first item is an introduction to the subject matter of the unit and how a particular unit is integrated with other units. Next is a set of learning objectives. These objectives let you know what you should be able to do by the time you have completed the unit. You should use these objectives to guide your study. When you have finished the unit, you must go back and check whether you have achieved the objectives. If you make this a habit you will improve significantly your chances of passing the course.

FACILITATORS/TUTORS AND TUTORIALS

There are 20 hours of tutorials provided in support of this course. As soon as you are allocated a tutorial group, you will be notified of the dates, times and location of tutorials, together with the name and phone number of your tutor.

Your tutor will mark and comment on your assignments; he/she will keep a close watch on your progress and on any difficulties you may encounter and provide assistance to you during the course. You must mail your tutor-marked assignments to your tutor well before the date due (at least two working days are required). They will be marked by your tutor and returned to you as soon as possible.

Do not hesitate to contact your tutor by telephone, e-mail or via the discussion board if you need help. The following might be circumstances in which you would find help necessary. Contact your tutor if:

- You do not understand any part of the study unit
- You have difficulty with the assignments/exercises
- You have a question or problem with your tutor's comments on any assignment or with the grading of an assignment

Try your best to attend tutorials as this is the only chance to have face to face contact with your tutor and to ask questions. You can ask questions on any problem encountered during the course of your study. To gain maximum benefit from the tutorials, make a list of questions before hand and participate actively in the discussions.

SUMMARY

Soil microorganisms and the biochemical reactions they carryout in the soil play major role in nutrient release and nutrient use efficiency. When you consider the health of the soil or the quality of the soil using biochemical compounds released that enhance or improve microbial and biochemical activities in soil, the importance of soil microbiology and biochemistry will be understood by you and productivity of soils will be improved leading to enhanced crop yield on a sustainable basis which is the ultimate goal of the small scale farmer. The important questions that require answer shows that many of the new ideas in agriculture will come through the study of the physiology and ecology of soil organisms, including processes they carry out such as nutrient transformations and biogeochemical cycles.

This course (SLM 505), therefore has topics on the physiology to biochemistry of soil organisms as well as on ecology in an attempt to improve the understanding needed to have a foundation for the interdisciplinary approaches that will continue to provide exciting new concepts in the field of agriculture.

We wish you the best and hope that you will find the course both interesting and useful. All the best.

MAIN COURSE

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MODULE 1 FUNDAMENTALS

UNIT 1 INTRODUCTION AND BRIEF HISTORY OF SOIL MICROBIOLOGY

CONTENTS

1.0 INTRODUCTION

Soil microbiology may be broadly defined as the study of microorganisms and their processes in soil. The interaction of organisms in the soil with each other and their environment is called soil ecology while soil biochemistry involves microbial processes, soil enzyme activities, formation and turnover of soil organic matter.

In this unit you will be looking at the origin of soil microbiology and the contributions of some of the early founders of soil microbiology. Also the importance of soil microbiology in agricultural ecosystems will be studied.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- define soil microbiology
- state some important names in soil microbiology and their contributions

- discuss the importance of soil microbiology in agriculture.

3.0 MAIN CONTENT

3.1 Brief History of the Origin of Soil Microbiology

The origin of soil microbiology is linked with the development of microbiology as a science and cannot be understood alone. The first documented fact of soil and soil biota, originates in the East by the Chinese. They classified soils and regarded earthworms as "angels of the earth" while the Romans saw earthworms as "intestines of the earth". They classified soil based on colour (organic matter content) and saw the importance of land forms, erosion, vegetation, land use and human health implications. Fungi were the 1st microorganisms to be classified because of their fermentation reactions. Later Eastern and Roman scholars recognised the importance of legumes and crop residues in enhancing soil quality.

3.2 Some Important Names in Soil Microbiology and Their Contributions

(a) Waksman, S.A.:
He wrote the book "Principles of soil microbiology" and encouraged research in soil microbiology. He studied the Actinomycetes – Streptomyces and discussed Streptomycin.

(b) Louis Pasteur:
He was responsible for the process of pasteurisation (process of heating liquids to partially sterilise them). He also researched on microbial fermentation leading to the delineation of anaerobic metabolism.

(c) Robert Kock:
He pioneered microbial culture technique and showed that microorganisms were disease – causing agents in his postulate called Koch's postulates. Kock's postulates are as follows:

a) A specific microorganism can always be found with a given disease.
b) The microorganism can be isolated and grown in a pure culture in the laboratory.
c) The pure culture will produce the disease when inoculated into a susceptible host.
d) The microorganism can be recovered from the infected host and grown again in a pure culture

(d) Sergei Winogradsky

He is called "Father of soil microbiology" because of his contributions to nitrification, anaerobic (N_2) nitrogen fixation, sulphur oxidation and microbial autotrophy. His studies on nitrification and sulphur oxidation were well noted, leading to the concept of microbial autotrophy where inorganic substrates are used as source of energy for growth by microorganisms. He came up with the column for studying sulphur cycle and investigated microbial growth on carbondioxide (CO_2) and inorganic ions in a process called chemoautotrophy. He also investigated microbial oxidation of ferrous iron (Fe^{2+}) which is the reduced form of Fe^{3+}, an essential component of rust.

(e) Beijerinck, Martins

Cultured the 1st symbiotic nitrogen-fixing bacteria that grew in association with legumes and the 1st non-symbiotic aerobic nitrogen-fixing bacteria as free living soil organism. These were Rhizobium and Azotobacter respectively. The non-symbiotic nitrogen fixing bacterium-Beijerinckia is named after him. He also extracted the first virus from plants (Tobacco Mosaic Virus – TMV) and developed enrichment techniques and was credited with saying "Everything is every-where, the environment selects".

(f) Lipman, Jacob

Was reported to have found the Department of Soil Chemistry and Bacteriology at New Jersey Agricultural Experiment Station in 1901. He is called the "Father of Soil Microbiology in the United States". He was interested in the effects of soil organisms on soil fertility and plant growth. He wrote the book called "Bacteria in Relation to Country Life". This was the 1st attempt to popularise the Science of Soil Microbiology.

SELF - ASSESSMENT EXERCISE

List the important names in soil microbiology and their contributions to soil microbiology.

3.3 What Is Soil Microbiology?

Soil Microbiology is defined as the study of microorganisms that live in the soil; their metabolic activity, their roles in energy flow, and importance in nutrient cycling. The soil science society of America defines soil microbiology as "the branch of soil science concerned with soil inhabiting microorganisms, their functions and activities.

3.4 Importance of soil microbiology in agriculture

Soil Microbiology is said to be the centre of all life, right from the air we breathe to food and the water we drink. Understanding of the study of soil microbiology is important to further understanding agricultural and environmental impacts on life. In effect, it will furnish you with knowledge to solve issues related to the functions of other biological systems in the soil.

4.0 CONCLUSION

From the origin of soil microbiology and the contributions made by some of the important names in soil microbiology, microorganisms have had great impact on man. Some of the effects are beneficial while some are detrimental. On the beneficial side, microorganisms are important in nutrient cycling and sustainability of life. On the detrimental side, they may contribute to environmental problems (green-house gases).

Soil microbiologists in this 21st Century, have continued studies in those areas that have brought us to our present position and encourage us to step boldly into the future. Hence, soil microbiologists have concentrated on the opportunities created by these contributions for the betterment of society through their work. If the past shows sign of good work, then the future is very promising. May be the solution to the need for enhanced food production or reduced environmental problem will cause some microbiologists (perhaps yourself) into the levels of Nobel Prize winners lie Waksman Selman who was honoured for his work on antibiotics.

5.0 SUMMARY

Early studies in soil microbiology focused on the study of soil biota and cycling of nutrients and their availability to agricultural plant. The study of soil microbiology today, is more of an interdisciplinary study which involves agricultural production, aquatic and biogeochemical sciences, microbial biodiversity and climate change. The contributions of Winogradsky the “Father of soil microbiology” and Lipman the founder of American Soil Microbiology stands as a landmark in soil microbiology and has spread throughout the whole world. Their contributions have improved crop production and given room to sound use of environmental resources worldwide.

6.0 TUTOR-MARKED ASSIGNMENT

1) Define soil microbiology.
2) State some important names in soil microbiology and their contributions.
3) Discuss the importance of soil microbiology in agriculture

7.0 REFERENCES/FURTHER READING

Brady, N.C.& Weil, R.R. (2002).*The Nature and Properties of Soils*. (13thed.).Prentice Hall, Upper Saddle River, NJ.

Eldor, A.P. (Ed.).(2007). *Soil Microbiology, Ecology and Biochemistry*.(3rded.). Canada :Academic Press Publications.

Sylvia, D.M., Hartel, P.G., Fuhrmann, J.J. &Zuberer, D.A. (2005).*Principles and Applications of Soil Microbiology*. (2nded.).New Jersey :Prentice Hall, Upper Saddle River.

UNIT 2 INTRODUCTION TO SOIL BIOCHEMISTRY

CONTENTS

1.0 INTRODUCTION

Soil Biochemistry is one of the disciplines of soil science and closely related with soil microbiology (one of the major disciplines in soil science). It is the science that deals with the chemical nature and behaviour of biological living matter in the soil. For this reason, it is thus referred to as biological chemistry or chemical biology.

In this unit, we will discuss the importance of soil biochemistry in agriculture and its relation to soil microbiology.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- define and explain soil biochemistry.
- distinguish between soil biochemistry and soil microbiology
- discuss the role of soil biochemistry in agriculture.

3.0 MAIN CONTENT

3.1 What is Soil Biochemistry?

Soil Biochemistry is defined as a branch of soil science hidden under the major discipline of soil microbiology in the sense that it deals with the chemical, nature and behavior of living matter, the transformations carried out and the energy changes that occur during these biological transformations. Therefore, the study of soil biochemistry includes the constituents (substances) that make up the living organisms, substances produced through their transformation processes and their functions or chemical activities which involves energy changes. Thus, soil biochemistry is the backbone of soil microbial functioning.

3.2 Importance of Soil Biochemistry in Agriculture

Soil biochemistry is of great importance in agriculture in the sense that it is the backbone of soil microbial functioning without which microorganisms cannot function effectively in the soil. As earlier mentioned, it includes the chemical compounds that make up part of the organism living cell or produced by or in the organisms itself. Some of the biochemical compounds produced by soil microorganisms that have been observed to be of great importance in agriculture are:

- Enzymes - catalyse chemical reactions during organic matter degradation (decomposition) and transformations of cycles in soil.
- Protein synthesis.
- Chalates formation - molecules that bind metal ions to form complexes and cause nutrient mobilization.
- Toxins formation - like antibiotics beneficial to some soil organisms in terms of protection from predators or reduces competition among soil organisms.
- Production of growth regulating substances or hormones like indole acetic acid (IAA), gibberlins and other substances that enable the plant to tolerate or withstand salinity problems. From these biochemical compounds produced you can see that soil biochemistry helps us to:
- Evaluate the nutritional value of agricultural and livestock products.
- Biodegrade toxic substances into non-toxic substances (e.g. breakdown or removal of groundnut toxin called Aflatoxin).
- Serves as food preservative and for processing.
- Helps plants to become disease and drought resistant.
- Helps in the formulation of balanced diet.
- Helps in cell metabolism (the formation and breakdown of cells tissues).

3.3 Relationship between Soil Biochemistry and Soil Microbiology

Soil microorganisms and the biochemical compounds they produce play important role in nutrient transformations and use. This inform, improves soil quality and soil productivity. The biology of the soil helps to determine the characteristics of the soil, while microbial decomposition of soil organic matter enhances soil fertility, growth of plants and microorganisms, improves or sustains the carbon storage of soil and improves soil structure. From the above discussion and many more, you can see the relationship between soil microbiology and soil biochemistry.

SELF-ASSESSMENT EXERCISE

Why do soil microbiology and soil biochemistry matter?

4.0 CONCLUSION

From our discussion in this unit, you can see that soil biochemistry is a major component of soil microbiology without which soil microorganisms cannot adequately carryout their metabolic activities. The biochemical compounds produced by or in these soil organisms form or make up part of the living cell and are necessary in biochemical activities such as nutrient transformation and use resulting in soil productivity. Thus, soil biochemistry is the pillar of soil microbial functioning, characteristics and dynamics of OM and the transformations by enzymes and soil organisms.

5.0 SUMMARY

Soil biochemistry is an essential component of soil science that you cannot do without (that is, cannot be ignored). Soil microbiology cannot be fully achieved or complete with the biochemical aspect of it. This is because soil biochemical takes care of all the transformation, function and energy changes that occurred during these processes. This unit, therefore looked at the importance of soil biochemistry in agriculture and its relationship with soil microbiology.

6.0 TUTOR-MARKED ASSIGNMENT

1) Define and explain soil biochemistry.
2) Distinguish between soil biochemistry and soil microbiology.
3) Discuss the role of soil biochemistry in agriculture.

7.0 REFERENCES/FURTHER READING

Eldor, A.P. (Ed.). (2007). *Soil Microbiology, Ecology and Biochemistry.* (3rd ed.).Canada: Academic Press Publications.

Hassan, G.D. (2010). *Soil Microbiology and Biochemistry*.New India Publishing Agency.

Sylvia, D.M., Hartel, P.G., Fuhrmann, J.J.&Zuberer, D.A. (2005).*Principles and Applications of Soil Microbiology*. (2^{nd}ed.).,Prentice Hall, Upper Saddle River, New Jersey.

UNIT 3 THE SOIL HABITAT

CONTENTS

1.0 INTRODUCTION

Soil is defined as a natural medium that supports plant growth on the earth surface, that is, it is both biologically active and living. It creates a habitat for microbial growth which feed and supports other plant life. The microorganisms survive as micro-colonies or biofilms isolated on mineral or inorganic particles, organic matter and roots depending on nutrients movements either by mass flow of soil water or diffusion in the soil. In this unit, you will be looking at the soil as a medium for microbial growth and development.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- define soil habitat
- identify some important microbes in the soil habitat
- discuss the importance of soil microbes in the soil habitat.

3.0 MAIN CONTENT

3.1 What is soil?

The soil is a natural medium that supports plant and animal growth and development on the earth surface and it is biologically active and living. The microorganisms survive as micro-colonies or biofilms isolated on mineral (inorganic) particles, organic matter and root depending on nutrient movements either by mass flow or diffusion in the soil. It is a heterogenous medium of solid, liquid and gaseous phases that vary in its physical, chemical and biological properties.

3.2 Components of Soil

The major components of soil are air, water, mineral and organic matter and consists of three phases – solid, liquid and gas phases. The solid phase consists of mineral or inorganic and organic matter that make up the skeletal framework of soils (pore spaces). The pore spaces are occupied by air and water and can change anytime depending on weather conditions. They influence the behavior and productivity of soils. The three phases mix together and encourage simple and complex reactions within and between soil components for growth and development of plants.

3.3 Factors Affecting Soil Habitat

Due to the differences in the physical, chemical and biological properties, the soil environment/habitat is a competitive one. This competition exists mainly among the major microorganisms in the soil such as bacteria, actinomycetes and fungi as well as other soil animals and plant roots due to the stresses or unfavourable conditions in the soil environment. These unfavourable conditions are classified as biotic and abiotic stresses.

3.3.1 Biotic and Abiotic Stresses

Biotic stress includes competition from other microbes while abiotic stress includes physical and chemical properties of the soil habitat. Competition is as a result of lack of adequate nutrient or substrate, water, space or growth factors. As a result of competition, some microbes can secrete allelopathic substances (toxic or inhibitory) like antibiotics that suppress neighbouring organisms while others are predatory or parasitic on other microbes.

3.4 Functions of Soil in the Habitat

The soil plays several roles in the soil environment:

a) Support plant growth as a medium for plant roots and supply nutrient element necessary for growth and development (medium for plant growth).
b) Control the fate of water in the soil water system – water loss, use, contamination (pollution) and purification (regulates water supply).
c) Serves as a recycling medium of nature for raw materials
d) Serves as a medium for microbial existence (habitat for soil organisms).
e) It is an engineering medium for human environment serving as an essential building material (baked soil material). Also provides

the foundation for road, houses we live in and air-port for air planes (Engineering medium).

SELF-ASSESSMENT EXERCISE

i. List the important soil components and phases of soil habitat.
ii. What is the function of the soil habitat?

4.0 CONCLUSION

From this unit, it can be concluded that:

4) The soil is a medium for plant growth.
5) Competitive inhabitant for soil organisms.
6) Regulates soil water supply.
7) Recycles raw materials in the soilecosystem.
8) Essential building material (engineering medium) for human environment.

5.0 SUMMARY

The soil habitat is a natural medium that supports plant and animal life on the surface of the earth and is both active biologically and living. It is a medium of solid liquid and gas phases. The solid phase consists of inorganic and organic matter which makeup the skeletal framework. The pore spaces are filled by air and water and can change at any time depending on weather conditions. The solid, liquid and gas phases mix together and enhance simple and complex reactions within and between soil components for growth and development of plants.

6.0 TUTOR MARKED ASSIGNMENT

1) Define soil habitat.
2) Identify some important microbes in the soil habitat.
3) Discuss the importance of soil microbes in the soil habitat.

7.0 REFERENCES/FURTHER READING

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UNIT 4 MICROBIAL COMPOSITION OF SOIL

CONTENTS

1.0 INTRODUCTION

A great number and diversity of microbes live in the soil. The microbes exist as micro-colonies or biofilms isolated on mineral particles, organic matter and roots depending on the movement of nutrients in the soil. In this unit, you will be looking at the microbial composition of soil and the roles the soil organisms play in the ecosystem.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain what soil organisms are
- classify the soil organisms living in soil
- discuss the activities of soil organisms in the soil.

3.0 MAIN CONTENT

3.1 What are Soil Organisms?

Soil organisms are creatures that live most of their lives in the soil. They include plants, animals, fungi, bacteria, actinomycetes, viruses, protozoans, nematodes, algae and so on. They are mostly single cells except for fungi and some small animals. Many of the soil organisms are small and can only be seen with the aid of a magnifying glass (microscope). The smallest being the bacteria, followed by actinomycetes, fungi and then algae. The major groups found in the soil are shown in the table below:

Organism	Size (µm)	Numbers (g^{-1} soil)	Biomass (kg wet mass ha^{-1} soil)
Viruses	0.02 x 0.3	10^{10} - 10^{11}	300 – 3,000
Bacteria	0.5 x 1.5	10^{8} – 10^{9}	300 – 3,000
Actonomycetes	0.5 – 2.0[a]	10^{7} – 10^{8}	500 – 5,000
Fungi	8.0[a]	10^{5} – 10^{6}	10 – 1,500
Algae	5 x 13	10^{3} – 10^{6}	5 – 200
Protozoa	15 x 50	10^{3} – 10^{5}	1 – 100
Nematodes	1,000[b]	10^{1} – 10^{2}	10 – 1,000
Earthworms	100,000[b]	-	-

Adapted from Metting (1993)
- a- Diameter of hyphae
- b- Length

3.2 Classification of Soil Organisms

a) Soil organisms are classified based on size and ecological function.
b) Soil organisms are classified based on action or activities.
c) Classification based on growth characteristics.

3.2.1 Classification Based on Size and Ecological Function

Soil animals (fauna) range from macrofauna (earthworms, millipedes) through mesofauna (mites, springtails) to microfauna (nematodes, protozoans). The plants (macro and micro flora) include the roots of higher plants, as well as microscopic algae and diatoms. The macroflora are largely autotrophs such as vascular plants (feeder root, mosses) while microflora (root hairs, diatoms, yeast, molds, mildews, mushrooms, cyanobacteria). The macro being larger than 2mm in width, meso between 0.1 and 2mm, and micro less than 0.1mm.

3.2.2 Classification Based on Action or Activity

Activities of soil organisms (flora and fauna) are in what we call food chain or food web. This relationship show how soil organisms are engaged in plant residue degradation. As one organism eats another, nutrients and energy are passed from one trophic level to another higher trophic level. The first trophic level is that of the primary producers while the second trophic level is the primary consumers that eat the producers. The third trophic level is the predators which eat the primary consumers and finally the fourth trophic level are the predators that eat predators and so on.

3.2.3 Classification Based on Growth Characteristics

Soil organisms are classified on their growth characteristics, nutritional, ubiquitous and affinity for substrate (nutrient). According to Winogradsky, organisms that grow very fast (rapidly) when high-energy containing nutrients are readily available are called zymogenous while those that colonise the more resistant materials remaining after the zymogenous attack are known as autochthonous (slow acting) microflora.

Ohta and Hattori isolated bacteria called Oligotrophs – those that grow better at low concentration of nutrients while those that grow better at relatively high concentration of nutrients are called Copiotrophs. Griffin, classified fungi as primary fungi colonisers – those that have the ability to metabolise residual substrates of increasing chemical complexity and recalcitrance while those that colonise residues at the later stages of decomposition are called secondary fungal colonisers. They are slower growing and less competitive at growth compared to primary fungal colonisers. They also produce fewer and more durable spores than primary fungal colonisers.

3.3 r – k Theory of Colonisationand Succession

Andrews (1984) came up with the possibility of linking microbial classification based on growth properties with a theory called r – k Theory of colonisation and succession. This theory suggests that "a species needs to adopt a different strategy to colonise an environment in which it is initially present at low density (r – strategy) than to persist in an environment in which it is already present near to its carrying capacity (k – strategy)". The r – strategy place large percentage of their available energy into reproduction, occupy the niche quickly and are poorly adapted to environmental stress and periods of low nutrient availability. Hence, they undergo large changes over time. Examples in this group are bacteria that belong to the Zymogenous or copiotrophic traits.

The k-strategy growth and reproduction rate are not affected by limited resources available and hence have the ability to withstand periods of low nutrient availability and environmental stress. Examples in this group are fungi and actinomycetes that belong to the autochthonous or oligotrophic traits.

3.4 Functions of Soil Organisms

- Breakdown of organic materials and pollutants to water and CO_2 (carbon dioxide).

- Breakdown or decomposition of organic compounds to simple inorganic forms in a process called mineralisation.
- Oxidation of reduced mineral element forms example sulphur (S) to sulphate (SO_4^{2-}).
- Reduction of oxidised nutrient element forms e.g. NO_3^- (nitrate) to N_2 (dinitrogen gas)
- Stabilisation of soil humus forms soil aggregate particles and improve soil structure.
- Buffer the soil pH.
- Nutrient retention and increased water infiltration and water holding capacity of soil.
- Take part in nutrient cycling/transformation and some act as food source for other microbes.

SELF-ASSESSMENT EXERCISE

List the important soil microbes and their contributions to the soil habitat.

4.0 CONCLUSION

This unit identified and discussed the major soil organisms and their roles in the soil ecosystem. Soil organisms are classified based on their size and ecological function, activities and growth characteristics. They play important role in nutrient cycling/transformation, soil formation, breakdown of harmful organic materials to simple forms and in the improvement of soil fertility and crop productivity. It is therefore clear that the soil is a habitat for a wide diversity of organisms that are both beneficial and detrimental to the soil environment.

5.0 SUMMARY

From the soil as a habitat for microbial growth and development, and the functions or contributions of soil microbes in the soil habitat, microbes have made great impact on soil environment. Some are beneficial while some are detrimental. On their beneficial role soil organisms are essential in nutrient cycling/transformation and sustainability of both plant and animal life though some contribute to green- house gases due to their release of methane has which causes green-house effect.

6.0 TUTOR MARKED ASSIGNMENT

1) Explain what soil organisms are.
2) Classify the soil organisms living in soil.
3) Discuss the activities of soil organisms in the soil.

7.0 REFERENCES/FURTHER READING

Brady, N.C.& Weil, R.R. (2002).*The Nature and Properties of Soils.* (13th ed.*)*.,Prentice Hall, Upper Saddle River.

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UNIT 5 PHYSIOLOGY OF MICROBIAL GROWTH

CONTENTS

1.0 INTRODUCTION

Growth is defined as the continuous or progressive development of an organism. It is seen as the development of some specific organs or the entire organism. The growth of soil organism is essential for soil ecosystem function and changes in soil depending on nutrient availability and adequate physical and chemical conditions.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain what growth is
- discuss the growth characteristics of soil organisms using the growth curve
- derive the growth equation.

3.0 MAIN CONTENT

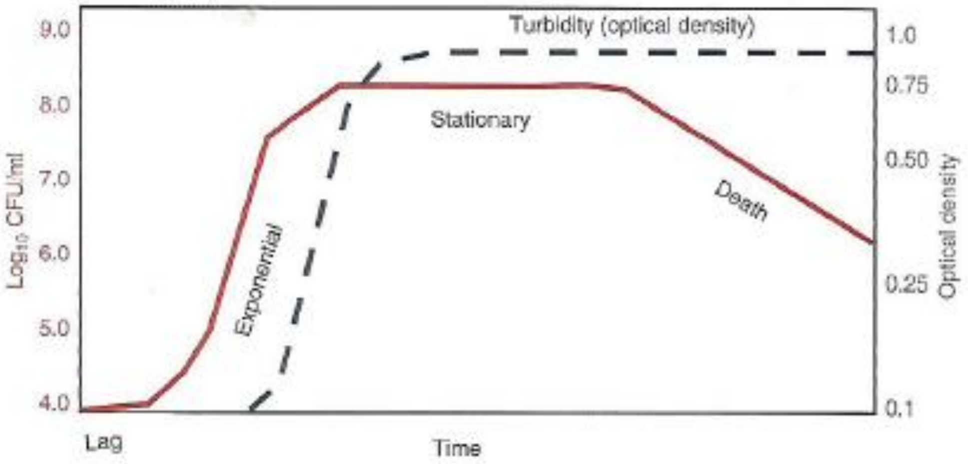
3.1 Growth Characteristics of Soil Organisms

Due to differences in nutrient availability in soil, different growth strategies are required. For instance, under dry surface soil conditions, biodegradable nutrients may accumulate due to lack of adequate water to support soil organism's action but when adequate or sufficient moisture is added, rapid organism growth which has been shown to take place. This transient period of nutrient availability can be differentiated with rhizosphere where constant nutrient supply is available.

Hence the environment provides the organism with the opportunity to express their growth characteristics and compete for nutrients with their unique characteristics.

3.2 Growth curve

Under favourable condition of sufficient nutrient supply, organism growth goes through various phases as shown below:



Adapted from Maier *et al.* (2009).

After the adaptive or lag phase, the organism enters a period of unlimited growth. When nutrient supply decreases, toxic metabolites accumulate and growth slows down. Then growth ceases and followed by death.

3.3 Growth Equation

At the unlimited growth phases, cell numbers increase exponentially or double per time interval and this is referred to as doubling time or generation time (abbreviated *td*). The number of cells per unit volume (Nt) after a growth period (t), while at initial population the number of cells per unit volume is No and number of doubling (n) during the time interval (t) is expressed as:

$$Nt = (No) \times 2^n$$

Where

$$n = \frac{t}{td}$$

Using linear form of equation

$$\text{In Nt} = \text{In No} + \frac{In\, 2\, x\, t}{td}$$

Using logarithm of base 10

$$Log_{10}Nt = Log_{10} No + \frac{Log_{10} 2 x t}{td}$$

Where

a) $\frac{Log_{10} 2}{td}$ = growth rate constant (N) = slope

$$N = \frac{In 2}{2.303 x td}$$

4.0 CONCLUSION

From the physiology of soil organisms, growth is the continuous development of an organism and could be either a specific organ or the entire organism depending on the availability of nutrients and the adequacy of physical and chemical conditions of the soil environment. The growth of soil organism goes through various phases namely lag, logarithmic, limited, and stationary and death phases.

5.0 SUMMARY

This unit identified and discussed the various growth stages of soil organisms depending on nutrient availability and the physio-chemical soil conditions. After the lag phase, the organism then enters a period of unlimited growth until nutrient supply decreases and toxic metabolites accumulate thereby slowing down growth rate and this may eventually cause growth to cease and finally death of organism.

6.0 TUTOR MARKED ASSIGNMENT

1) Explain what growth is.
2) Discuss the growth characteristics of soil organisms using the growth curve.
3) Derive the growth equation.

7.0 REFERENCES/FURTHER READING

Brady, N.C. &Weil, R.R. (2002).*The Nature and Properties of Soils*.(13th ed.), Prentice Hall, Upper Saddle River, NJ.

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MODULE 2 MICROBIOLOGY OF THE RHIZOSPHERE AND MICROBIAL INTERACTIONS

UNIT 1 RHIZOSPHERE SOIL

CONTENTS

1.0 INTRODUCTION

As earlier mentioned, the study of the properties and interaction of organisms in the soil with each other and their environment is called soil ecology. In the soil, physical and chemical properties of the soil affect microbial processes or activities. All these affect the overall makeup of the soil microbial community. These changes in the microbial community structure can determine the suitability of the soil for crop production. This unit will introduce you to the topic of soil ecology with special emphasis on the rhizosphere and the effects of plant and microorganisms on each other and the soil.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain what a rhizosphere is and differentiate it from spermosphere
- discuss the effect of plants on the microorganism in the ehizosphere
- discuss the effect of microorganisms in the rhizosphere.

3.0 MAIN CONTENT

3.1 What is Rhizosphere and Spermosphere?

The zone of soil under the influence of living roots is called the rhizosphere and usually extends more than 5mm from the root and more importantly, is the area of increased microbial activity because of the root exudation of substances that affect microbial activity. The area of increased microbial activity around the seed is called spermosphere. The term spermosphere arose because the seed is commonly referred to as the sperm hence; the term spermosphere is in line with the rhizosphere. The spermosphere can extend up to 10 mm from the seed but distances up to 20mm have been reported. Seed colonisation is the 1st step towards root colonisation in the soil. Therefore, spermosphere is the zone of soil under the influence of the seed.

The rhizosphere and spermosphere have different properties that change in space and time as the distance from the root or seed increases. These changes affect the relationship between the plant and the microbial community. The microorganisms that inhibit the root are known as endophytes.

The biological and chemical properties of the rhizosphere are different from those of the bulk soil. For instance, soil acidity may be 10 times higher (or lower) than the bulk soil. Roots affect nutrient supply by withholding or withdrawing dissolved nutrients or by solubilizing the nutrients. As a result of these, roots affect the mineral nutrition of soil microorganisms just as the microorganisms affect the nutrients available to the plant roots.

SELF-ASSESSMENT EXERCISE

i. The rhizosphere is the zone of increased microbial activity, why?
ii. Spermosphere is an important area of study, why?

3.2 Definitions of terms in the Rhizosphere

The rhizosphere represents a poorly defined zone of soil with a microbiological gradient in which the maximum effect of roots and microorganisms are in the soil nearest to the root. The terms outside and inner rhizosphere are terms used to define the zone of influence.

a) Inner rhizosphere (Rhizoplane)
This is the surface of a plant rot with any adhering soil particles, Also called Rhizoplane.

b) Outer Rhizosphere
This is the zone beyond the rhizoplane but still under the influence of plant root. Microscopic and plate count examinations have revealed presence of microbial activities around the root and root hairs.

c) R:S ratio
This is the ratio of the number of organisms per gram of the rhizosphere soil (R) to the number of organisms per gram of the controlled soil (S).

d) Rhizosphere effect (RE)
Rhizosphere effect is the ratio that expresses the extent to which the plant roots affect the number of microorganisms. It is usually greater for bacteria than for other microorganisms inhabiting the rhizosphere. Fungi and actinomycetes may be abundant but their active count is usually low.

SELF-ASSESSMENT EXERCISE

Distinguish between rhizosphere, rhizosplane and rhizosphere effect.

3.3 Factors affecting microbial activity in the Rhizosphere

1) Organic matter requirements
Plant dead roots and leaves and soil organic matter are energy sources for a majority of soil organisms especially the heterotrophs. Energy-rich organic substances including plant root exudates stimulate growth and activity of microbes. Bacteria respond mostly and rapidly to additions of freshly introduced or incorporated simple compounds such as starches and sugars while fungi and actinmycetes respond more to cellulose and the more resistant compounds. In addition, organic materials left on the soil surface are dominated by the microbial activity of fungi while bacteria play larger role when incorporated.

2) Oxygen requirements

While most microorganisms are aerobic, some are anaerobic and use substances like nitrate (NO_3^-), sulphide (S^{2-}) as their electron acceptors. Facultative bacteria use either aerobic or anaerobic forms of metabolism. These forms of metabolism are carried out at same time in different habitats within the soil.

3) Moisture and temperature

Optimum moisture ranges between -10 to -70 kPa and this is the best for aerobic microbes while anaerobic microbes prefer waterlogged conditions, where oxygen supply is limited. Microbial activity is best at temperatures between 20 – 40 ^oC. The warmer end of this range favoursactinomycetes while extreme temperatures kill bacteria and sometimes temporarily suppress their activity. The only excepts are cryophilic microbes that strive below 5 ^oC (biological zero) and thermophilic that strive beyond 40 ^oC.

4) Exchangeable calcium (Ca) and pH

Exchangeable calcium (Ca) and pH help to determine which type of organisms thrive in the soil. High Ca levels and near-neutral pH generally result in the largest and most diverse bacterial populations while low pH (acidic conditions) favour fungi. This is why fungi dominate forested soils.

3.4 Influence of microorganisms in the Rhizosphere

3.4.1 Beneficial effects

The microorganisms in the rhizosphere may have beneficial or detrimental influence on the development of plants. This is because, since microbes are so closely related to the plant root systems, any beneficial or toxic substance produced can cause an immediate and profound effect on the microorganism. Therefore, microbes are important to plant productivity and ecological functioning of soils.

1) Organic material decomposition

This is the most important contribution of microorganisms to higher plants in the rhizosphere. In this process, dead leaves, roots and other plant tissues are broken down (degraded) converting the organically held nutrients to inorganic (mineral) forms available for plant uptake. Soil microbes also assimilate wastes from animals and other organic materials. Some of the new compounds produced help to stabilize soil structure and others contribute to humus formation.

2) Breakdown of toxic compound

Many organic compounds toxic to plant and soil organisms which find their way into the soil (as metabolic by-products) are broken down into non-toxic compounds by those organisms that produce enzymes that use these toxins as food. Some of these toxins are xenobiotic (artificial) and may resist attack by microbial enzymes. Bacteria and fungi are important in maintaining a non -toxic soil environment. The detoxicifying activity of soil microbes is greatest on soil surface where microbial numbers are concentrated in response to greater availability of organic matter and oxygen.

3) Inorganic transformations

Inorganic transformation is of great importance to the functions of soil ecosystem. Nitrate (NO_3^-), sulphate (SO_4^{2-}) and to a lesser extent phosphate (PO_4^-) ions are present in soil due to microbial action. Bacteria and fungi takeup some of N, P and S in the organic materials they degrade. Excess of these nutrients are then excreted into the soil solution in organic forms. Also nematodes and protozoa that feed on them takeup these nutrients. Other essential nutrients like Fe and Mn (micronutrients) are determined largely by microbial action. In well-drained soil (aerobic) they are oxidized by autotrophic organisms to higher valency state making them insoluble. This keeps Fe and Mn mostly in insoluble and non-toxic forms even under fairly acid condition. If such oxidation did not occur, plant growth would be adversely affected due to the toxic quantities of these elements in solution.

4) Nitrogen fixation

The fixation of atmospheric nitrogen that cannot be used directly by higher plants is one of the most important microbial processes in soils. For instance, actinomycetes (*Frankis* sp.) fix major amounts of nitrogen in forest ecosystems, cyanobacteria in flooded rice paddies and wetlands and rhizobia in agricultural soils in association with legume root nodules. The greatest amount of N fixed by these organisms occurs in root nodules.

5) Rhizobacteria

The rhizosphere soil and rhizophane support large population of microorganisms. Bacteria that live in this zone are called rhizobacteria. Many of them are beneficial to higher plants called the plant growth promoting rhizobacteria. Because the root surface is almost covered with bacterial cells, little interaction between the soil and plant root take place and this cannot be done without some microbial influence. Many rhizobacteria enhance nutrient uptake or hormonal stimulation while some damage plants by invading the root cells and live as parasites. Other rhizobacteria inhibit root growth and function, thereby causing stunting, wilting, foliar discolouration, nutrient deficiency and even death of plants. Buildup of deleterious rhizobacteria cause yield declines especially under long-term monocultures, even in planting new trees in old orchanrds. Their main beneficial effect is in weed control, thus weed scientists hope to reduce weed seed germination and seedling growth, thereby reducing the use of herbicide.

6) Plant protection

Some soil organisms attack higher plants while others protect plant roots from parasites and pathogen invasion.

3.4.2 Detrimental Effects of Microorganisms in the Rhizosphere

1) Microorganisms also require some anions and cations for their own growth and development leading to nutrient immobilization. For instance, when absorption of soluble inorganic phosphate is affected, immobilization of Mn, Fe, Zn, K is stimulated.
2) Some microbes have injurious effect on higher plants as plant parasites or by producing certain toxic substances.

3.5 Influence of Plants on Soil Microorganism

The activities of plant roots greatly affect the soil chemical, physical and biological properties depending on the soil type and plant species. The effect of plants on organisms in the soil are as follow:

1) Increase soil aggregate stability

The plant root hairs increase root surface area available to absorb water and nutrient from soil solution, thereby encouraging soil aggregate stability. The root exudate supports large community of microorganisms which further help to stabilize soil aggregates.

2) Increase humus content of soil (SOM)

When the dead plant parts decompose, they increase soil humus both at the surface and subsurface layers of soil. Three types of organic compounds are released at the soil surface:

a) Low molecular-weight organic compounds secreted by root cells, e.g. organic acids, sugars, amino acids and phenolic compounds which exert growth stimulating or regulating effects on plants and soil microbes.
b) High molecular – weight mucilages secreted by root-cap cells and epidermal cells e.g. mucigel.
c) Cells from rootcap and epidermis (slough offs, root materials)

All these are called rhizodeposition.

3. Microorganisms are affected by plant root respiration which alters the pH or availability of some inorganic nutrients due to the release of CO_2. The pH of the rhizosphere is however more than the surrounding soil when roots are assimilating NH_4^+ (ammonium ion) but higher when NO_3^- (Nitrate) is being taken up.
4. Root penetration improves soil structure which favour microbial oxidation.
5. Increase in the population of denitrifying bacteria due to the presence of NO_3^- which enhance N_2 and NO (nitrous oxide) release by plant growth.
6. Presence of certain plants reduces abundance of nitrifying organisms.
7. Increase in the abundance of celluloytic microorganisms due to response to cellulose produced from slough-off root materials. These microbes degrade cellulose providing carbonaceous substrate for some microorganisms.

SELF-ASSESSMENT EXERCISE

What role does plant exudates in the rhizosphere?

4.0 CONCLUSION

The rhizosphere is the zone of soil under the influence of living roots usually extends more than 5mm from the root and spermosphere is the zone of soil under the influence of the seed. Soil microorganisms are affected by organic compounds released from plant roots or seed and these compounds alter the microbial community structure, increase microbial population, activity and interactions among microbes, plant and the soil. The microorganisms in the rhizosphere can exert beneficial or detrimental effects in plant, thereby affecting plant productivity.

5.0 SUMMARY

Rhizosphere which is the zone of soil under the influence of living roots is the area of increased microbial activity because of the root exudation substances, that affect microbial activity.

From the study in the unit, soil microorganisms play important role in plant growth and development and this could be positive or negative, likewise plants exert great effect on soil microorganisms due to their exudate excretion which alter the soil chemical and physical properties of the soil.

6.0 TUTOR-MARKED ASSIGNMENT

1) Explain what a rhizosphere is and differentiate it from spermosphere.
2) Discuss the effect of plants on the microorganism in the ehizosphere.
3) Discuss the effect of microorganisms in the rhizosphere.

7.0 REFERENCES/FURTHER READING

Brady, N.C. & Weil, R.R. (2002).*The Nature and Properties of Soils*. (13^th^ed.).Prentice Hall, Upper Saddle River.

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UNIT 2 SOIL MICROBIAL INTERACTIONS

CONTENTS

1.0 INTRODUCTION

Due to the limited nutrient supply and environmental problems in the soil, microorganisms compete within and among themselves for the limited nutrient supply, space, time and water supply in other to survive the adverse soil conditions. In other to mitigate these adverse soil problems, soil microorganisms interact with themselves and with other organisms in the soil. Many of these interactions occur through some biochemical signaling in the rhizosphere. These interactions are seen on the basis of whether one or both organisms gain benefit from the association. The study of these interactions among microorganisms and their environment is called soil ecology.

Biochemical Signaling
This is the information passed within and among microorganisms that are created and transmitted through the production of biochemical compounds by the microorganisms. These biochemical compounds are instruments used by the soil microorganisms to carry out their functions essential for plant growth, development and nutrition.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- distinguish the various types of interactions among soil microorganisms
- classify them into beneficial (positive) or detrimental (negative) interactions
- explain microbial succession.

3.0 MAIN CONTENT

3.1 Types of interactions

A number of possible interactions that may occur between two species are:

1) Commensalism
2) Neutralism
3) Symbiosis (mutualism)
4) Proto-cooperation (synergism)
5) Competition (Antagonism)
6) Ammensalism
7) Parasitism and predation

3.1.1 Positive Interactions

1) Commensalism

This is an interaction in which one population or microorganism benefits from the interaction and the second remains unaffected. This is the basis for nitrification in nitrogen cycle. For instance, the ammonia-oxidising bacteria (*Nitrosomonas*sp.) transform NH_3 into product (Nitrate) that is a substrate for the nitrite-oxidising bacteria (*Nitrobacter* sp.).

2) Neutralism

This is an interaction in which the two microorganisms (populations) behave entirely independent of each other.

3) Protocooperation (synergism)

In this ineraction, both populations of microorganisms benefit from the association but the interaction is not compulsory for their existence or performance in the environment. For instance, in anaerobic soils, the methane producing bacteria benefit from the acetate and hydrogen gas produced by the fermentative bacteria while the fermentative bacteria benefit by the lowering of the hydrogen gas and acetate concentrations by methane producing bacteria thereby allowing them to put together heat changing (thermodynamically) biochemical reactions to cell growth.

4) Symbiosis (mutualism)

This interaction occurs between specific organisms rather than populations. In this interaction both microorganisms benefit from the association and cannot survive (grow) without the other in the association.

This association can be found between:

a) Bradyrhizobium or Rhizobium with leguminous plants.
b) Actinomycetes (*Frankia* sp.) with plants (Actinorhizal).
c) Algae and fungi (Lichens).
d) Fungi with roots of higher plants (mycorrhizae).
e) Bacteria within protozoan cells.
f) Protozoan in underground termites (similar to bacteria in intestine of ruminants).
g) Some ants, beetles and termites grow some fungi (the insects provide the fungi with nutrients for growth such as dead plant debris (roots, leaves) and faeces while the insect feed on the fungal biomas).

3.1.2 Negative Interactions

1) Predation and Parasitism

This is an interaction in which there is direct attack of one organism.In another, in this interaction, one population has a negative effect on the size of another population by feeding on them and reducing their number.For instance, nematodes and protozoa feed on bacteria and fungi.

2) Competition (Antagonism)

This is an interaction in which there is a suppression of one organism as the two species struggle (compete) for limiting quantities of nutrients, water, oxygen and other needed requirements thereby having a negative effect on each other. Example: Heterotrophic microorganisms and ammonia-oxidising bacteria (*Nitrosomonas*) compete for ammonium ion (NH_4^+).

3) Ammensalism

This is an interaction in which one species is suppressed by toxins produced by the second species and the second species is not affected, thereby gaining a competitive advantage.

Examples:
a) Many fungi and bacteria produce antimicrobial growth substances that inhibit other organisms.
b) Sulphur-oxidisingmicroorganism causes soil acidification, which affect acid – sensitive organisms like streptomyces sp.

3.2 Microbial Succession

This is an important series of change in microbial populations in the soil environment, resulting in the arising of a climax community. Microbial succession takes place because when organisms grow, they alter the soil environment around them giving way for the development of other different microbial populations. The initial populations that respond to the addition of freshly introduced carbonaceous materials are called primary microflora. As theprimary microflora grow, they alter their environment and the carbonaceous materials thereby changing the products into microbial biomass and metabolic waste products. Then the secondary microflora takes over which feeds on the products of the primary microflora. This results in a progressive change in microbial populations as substrates are broken down under different soil environmental conditions such as pH, oxygen and Eh (redox potential).

4.0 CONCLUSION

Nutritional limitations and environmental conditions of the soil as well as time and space are the causes of microbial interactions in the soil, in order for soil microorganisms to mitigate (solve) these adverse soil problems. These interactions mostly occur through biochemical signaling. The interactions that have been reported to occur are commensalism, neutralism, photocooperation, symbiosis, competition, ammensalism, predation and parasitism. The 1st four types of interactions are positive interactions while the rest others are negative interactions. However, soil physical and chemical properties as well as carbonaceous materials introduced into the soil put together affect soil microbial metabolic activity and determine microbial community structure.

5.0 SUMMARY

From the discussion in this unit, the types of interactions within and among soil microorganisms can be summarized as follows in the table below:

Type of interaction	Effect of interaction within and among soil microorganism	
	1st population	2nd population
Commensalism	+	0
Synergism (photocooperation)	+	+ or 0
Mutualism (symbiosis)	+	+
Competition (Antagonism)	-	-
Predation and Parasitism	-	+
Ammensalism	-	+ or 0

Note
+ means positive effect, - means negative effect, 0 means no effect (Reproduced from Sylvia *et al*, 2007).

6.0 TUTOR-MARKED ASSIGNMENT

1) Distinguish the various types of interactions among soil microorganisms
2) Classify them into beneficial (positive) or detrimental (negative) interactions.
3) Explain microbial succession.

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UNIT 3 NON-SYMBIOTIC VS SYMBIOTIC ASSOCIATION

CONTENTS

1.0 INTRODUCTION

Nitrogen is an essential element for all living organisms not just soil microorganisms. It is a major component of protein (amino acids). Nitrogen in the atmosphere is transformed into the plant available forms in a process called biological nitrogen fixation (BNF). This process is carried out by the bacteria – Rhibobium and Bradyrhizobium in a mutualistic association with legumes forming root nodules.

Biological nitrogen fixation occurs as non (free living) symbiotic fixation and symbiotic nitrogen fixation. Free-living (not plant associated) and symbiotic plant associated – root nodule symbiosis).

2.0 OBJECTIVES

At the end of this unit, you shouldbe able to:

- discuss BNF and the types of BNF
- understand cross inoculation group
- discuss or explain the process of nodule formation and effectiveness
- explain the factors affecting symbiotic nitrogen fixation.

3.0 MAIN CONTENT

3.1 Biological Nitrogen Fixation (BNF) and Types

Biological nitrogen fixation is the process by which atmospheric nitrogen is converted into plant available forms. This process is brought about by either the free living bacteria (blue-green or cyanobacteria) – non symbiotic fixation or by Rhizobium or Bradyrhizobium in symbiotic fixation with higher plants.

3.2 Non-Symbiotic Nitrogen Fixation

An example of this non-symbiotic nitrogen fixation is the association or interaction between algae (green or blue-green algae) and fungi (lichens). In this interaction the algae are known as the phycobiont while the fungi are known as the mycobiont (Ascomycetes and Basidomycetes). The algae (phycobiont) benefits from the protection provided by the fungi which envelopes the algae and protects it from environmental stress. The fungus (mycobiont) gains by making use of the carbon dioxide (CO_2) fixed by the algae.

The free living heterotrophic nitrogen fixing bacteria are divided into three groups namely:

1) Obligatory aerobes

Examples are: *Azotobactervirinlandi, A. biejerinkia*) and *Biejerinkia* spp., *Pseudomonas* spp., *Norcaclia* sp., found mainly in tropical acidic soils.

2) Facultative anaerobic bacteria

Examples are the *Bacillus* sp. (*Bacillus polymyxia, B. mercerous, B. circulars*), and those belonging to the Enterobacterium family (*Klebsiella, pneumonia, Enterobacter cloacae, Escherischiatrictly anaerobic bacteria*).

3) Strictly anaerobic bacteria

Example: *Clostridium pasterClostridiumpasterianium, Desulphuruibro, methano bacterium chlorobium*and *chromatium*.

Azotobactersp are the most active in this type of symbiosis. They can fix about 5 – 20 µg of N and 1g of sugar; hence do not contribute much to the soil. The blue-green algae also called cyanobacteria fix N under waterlogged conditions (e.g. sugarcane plantations) and fix about 15 – 20 kg N.

Some of the factors affecting the non-symbiotic nitrogen fixers are:

4) C:N ratio
5) Oxygen content
6) Molybdenum content
7) pH
8) Ca^{2+}
9) Excess Fe and Al
10) Temperature level

3.3 Mechanism (Process) of Nitrogen Fixation

The mechanism through which BNF proceeds and the enzyme responsible for the reaction is shown in the overall reaction below:

nitrogenase

$N_2(g) + 8e^- + 8H^+ + 16Mg\ ATP \longrightarrow 2NH_3 + 16\ MgADP + 16\ Pi + H2$

enzyme

Where,

N = Nitrogen, NH_3 = Ammonia, H^+ = Hydrogen ion,

ATP = Adenosine Triphosphate,

ADP = Adenosine Diphosphate, Pi = inorganic phosphorus

From the reaction equation one molecule of nitrogen gas is reduced to two molecules of ammonia using energy to carry out the reduction process. It is estimated that 12 moles of Mg ATP are required for one mole of N_2 (gas) to be reduced to 2 moles of NH_3 (ammonia). The enzyme responsible for carrying out this reaction is called nitrogenase and consists of two proteins namely:

11) Iron-protein (Fe-protein) (Azoferrodoxin or Azofer) also known as dinitrogenasereductase.
12) Molybdenum – Fe protein (Molybdo-ferredoxin/Azofermo called component 1) also known as dinitrogenase

These nitrogenase proteins (Fe-protein and Mo-Fe protein) are Fe-sulphur components and the Fe protein is the smaller of the two proteins. The Fe protein is inactivated by oxygen. The molecular weight of Fe protein ranges between 50,000 – 70,000 while Mo –Fe protein is inactivated by O_2 but not as quickly as Fe protein. They need Mg^{2+} ion to be active and also inhibited by ADP. From the mechanism of nitrogenase enzyme; it is seen that:

- The Fe protein accepts electrons from ferredoxin and binds to two Mg ATP and then transfers electrons to Mo Fe protein.

- Fe protein and Mo - Fe protein then forms a complex, the electron is transferred and 2 Mg ATP are hydrolysed to 2 Mg ADP + Pi (in organic phosphate).
- Fe protein and Mo - Fe protein dissociated and the process is repeated.
- When nitrogenase has gotten electrons enough, it binds one N_2 molecule, reduces it to form NH_4^+.
- The Mo-Fe protein than accepts more electrons from Fe protein to repeat the cycle.

3.4 Symbiotic Nitrogen Fixation

Symbiotic nitrogen fixation is a mutualistic (symbiotic) interaction between two organisms rather than between populations. Both organisms benefit from the association and most times,no organism can grow in the absence of the other. In this association, the microorganism is the prokaryotic organisms (Rhizobium, Bradyrhizobium, Klebsiella, Nostoc or Frankia) and the plant a eukaryotic usually photosynthetic (leguminuous or non-leguminous plant, water fern or liverwort). Plants are the primary source of reduced carbon for the soil microbial community, and the plant growth is also linked by the availability of N and P. Therefore, plants solve this environmental adverse conditions or problems by forming associations with various soil microorganisms. Two examples of agricultural importance in association are:

13) Fungi and roots of higher plants
14) Rhizobium with legumenuous plants

The amount of nitrogen fixed in legumes vary and could go up as high as 600 kg N fixed per hectare (600 kg Nha^{-1}).

3.4.1 Cross Inoculation Groups

Various legumes prefer certain rhizobia strains thus rhizobia are grouped based on their nodulation but not on effectiveness. Some legumes (e.g. soybean) are promiscuous and can form nodules with more than one type of rhizobia. Therefore, cross-inoculation grouping is used to understand rhizobium grouping better. Cross – inoculation group therefore, refers to a collection of leguminous species that can form or develop nodules when exposed to bacteria gotten from the nodules of any member of that plant group. About 20 cross inoculation groups have been established but only about eight (8) of them have achieved prominence. Some of the selected cross-inoculation groups and their legume host are given in the table below:

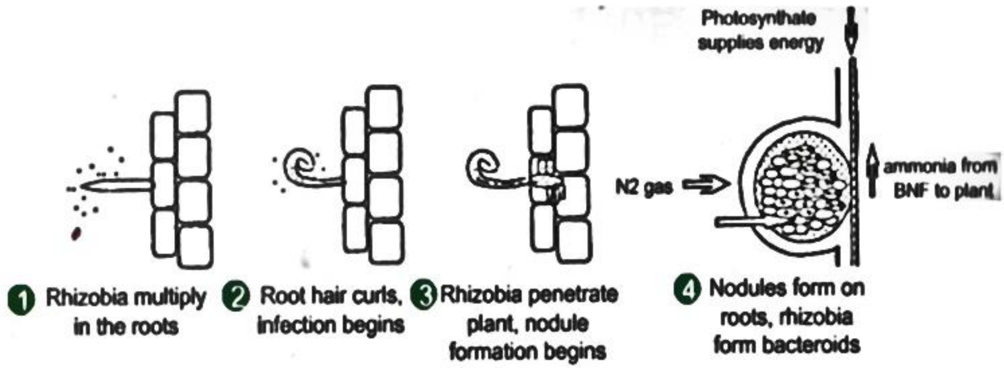
Cross inoculation group	**Rhizobium sp.**	**Host genera**	**Legumes included**
1) Alfalfa gp	*R. melilotus*	Medicago Melilotus Trigonella	Alfalfa,Sweet clover, Fenugreek (masala)
Clover gp	*R. trifoli*	Trifolium	Clovers
Pea gp	*R. leguminosarum*	PisumVicia Latyrus Lens	Pea,VetchSweet pea lentil
Bean gp	*R. phaseoli*	Phaseolus	Beans
Lupine gp	*R. Lupini*	Lupinusornithopus	Lupines,Serradella
Soybean gp	*R. japonicum*	Glycine vigna	Soybean
Cowpea gp	Cowpea Bradyrhizobium	Vigna,LespedezaCrotolaria, ArachisPhaseolus, Pueraria	Cowpea Lespedeza crotalaria Peanut Lima bean Kudzu
Fast lotus and Leucaena gp	R. Isolates	Lotus,Leucaena	

Based on Alexander, M. (1977).

3.5 Nodule Formation and Effectiveness

3.5.1 Nodule Formation

In the Rhizobium-legume symbiosis, the nodule bacteria infect the host plant (legume) by entering the host through the root hairs. This first step involves the release of biochemical compounds that produce signals in the rhizosphere that stimulate the nodule bacteria. These biochemical compounds cause increase in the population of the rhizobium in the rhizosphere. The biochemical compounds excreted by the legume host roots stimulates only the bacteria of the specific cross inoculation group. In the root hairs, infection threads are formed resulting in the curling of the root hairs (evidence of infection). The infection thread provides enterance for the rhizobia into the plant tissue resulting in the development of nodules. Rapid multiplication of the plant cell occurs. The bacteria are released into the cytoplasm where they multiply for a short period. The host then builds a layer of cell in which an extensive vascular system is formed containing the rhizobia and the cutile area. The rhizobia after multiplication converts into non-motile, non-growing cells called bacteriods. Bacteriods vary in size and shape. These bacteriods are essential for fixation to take place. Alongside the bacteriods, legume haemoglobin which is important in the fixation of N is developed and this gives the effective nodule its pink colour. The legume haemoglobin changes to give greenish colour when ageing. The size, shape and location of nodule also vary but to a limited extent the bacteria strains. Legume plants with tap root (soybean, cowpea) have larger but fewer nodules while those with branched root (groundnut) have smaller but more nodules. The diagrams below show the structure of a nodule and bacteriods.



Adapted from Bala (2012)

3.5.2 Nodule Effectiveness

Nodule effectiveness is the ability of the symbiotic system when formed to take up (assimilate) molecular N. Different legumes combine with Rhizobia in their symbiotic association to produce nodules and fix N and their ability to nodulate and fix N varies. Some combinations (associations) produce long lived nodules which fix N at high rate. These are the highly effective associations. On the otherhand, when no detectable N is fixed by the nodule, it is termed ineffective and may become parasitic by reducing the yield of the nodulating plant.

SELF-ASSESSMENT EXERCISE

Differentiate between nodule effectiveness and ineffectiveness.

3.6 Factors Affecting Symbiotic Nitrogen Fixation

Several factors have been observed to influence nodulation and symbiotic N_2 fixation. These factors may affect negatively the rhizobium population and survival rate, including the legume host plant. These factors include:

15) High temperature – kills rhizobia and reduces nodulation.
16) Acidity – Limit rhizobial growth and persistence.Acidity reduces nodulation and this is common in acid soils probably because few rhizobia persist and acidity affects rhizobia attachment to their host. Fast growing rhizobia are more sensitive than the slow-growing Bradyrhizobia though, few low pH tolerant strains exist.
17) Drought – kills rhizobia and reduces BNF (Biological nitrogen fixation).
18) Waterlogging – *Rhizobium* do well in waterlogged soils (anaerobes) but legumes growth and development are affected due to deficiency (lack) of oxygen and accumulation of toxic minerals.
19) Salinity and alkalinity – legume are more sensitive to salinity and alkalinity than rhizobia. Salinity reduces nodulation while alkalinity limits the availability of micronutrient in the soil, thereby affecting plant growth and N_2 fixation.
20) Mineral nutrients – Mineral nutrient deficiency affects specific functions in nodulation and N_2 fixation. Mineral nutrient deficiency will cause yellowing (chlorosis). Ca, P and micronutrient (Mo, Co, B and Fe) reduces nodulation and N_2 fixation. Mo causes nodule ineffectiveness; B and Zn reduce nodule number and size; Co delay nodulation; Cu reduce BNF and Fe reduce nodule initiation and development.
21) Extent of nodulation and rhizobium effectiveness.
22) Adequate carbohydrate (food) supply.

4.0 CONCLUSION

Biological nitrogen fixation results from the infection of host roots by rhizobia as a result of nodule development, function and senescence. The process of nitrogen fixation is brought about by either free-living Bacteria (non-symbiotic fixation) or by *Rhizobium* or *Bradyrhizobium* (symbiotic fixation) with higher plants. Nitrogenase enzyme is the enzyme responsible for carrying out the process of nitrogen fixation and has two components namely, the Fe protein and the Mo-Fe protein. The rhizobia are grouped based on their cross inoculation group (nodulation) but not on effectiveness. The nodules formed could be either effective (pink in colour) or ineffective (greenish in colour). Effective nodules are those that have the ability to fix dinitrogen while the ineffective nodules are those that form nodules but do not fix dinitrogen in soil. Several physical and chemical factors such as temperature, acidity, drought, waterlogging, mineral nutrients, salinity and alkalinity, carbohydrate supply and nodulation and rhizobia effectiveness was reported to affect symbiotic nitrogen fixation.

5.0 SUMMARY

This unit dealt with the ability of legumes to fix dinitrogen in association with rhizobium. It also discussed that symbiotic nitrogen fixation could be carried out by either free-living Bacteria in non-symbiotic association or by rhizobium in the symbiotic association. The process of nitrogen fixation was discussed including the cross inoculation groups of rhizobia. Nodule effectiveness and ineffectiveness was introduced noting the importance of nitrogenase enzyme and legume haemoglobin in nitrogen fixation.

6.0 TUTOR-MARKED ASSIGNMENT

1) Discuss BNF and the types of BNF.
2) Explain cross inoculation group.
3) Discuss or explain the process of nodule formation and Effectiveness.
4) Explain the factors affecting symbiotic nitrogen fixation.

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UNIT 4 RHIZOBIUM-LEGUME ASSOCIATION

CONTENT

1.0 INTRODUCTION

In the soil habitat, there are soil organisms which interact with leguminous plants to form symbiotic associations, also called mutualistic interactions. These soil organisms are called Rhizobia. Leguminous plants form mutualistic symbiosis with root nodule bacteria mostly of the genera *Bradyrhizobium* and *Rhizobium*. In this unit, we will discuss the importance of Rhizobium bacteria in the soil, its life cycle and give brief explanation of its association on legume plant roots.

2.0 OBJECTIVES

At the end of this unit, you shouldbe able to:

- discuss the life cycle of *Rhizobium*
- discuss its role in soil and on plant roots.

3.0 MAIN CONTENT

3.1 The Rhizobium Bacteria

Legumes have a long history of use in agriculture especially in association with the root nodule bacteria (*Rhizobium*). Not all legumes are nodulated by the bacteria rhizobia. They live in the soil along with other soil microorganisms without their host plant (legume). While in soil, they are saprophytes normally referred to as native rhizobia. They range from zero to thousands per gram of soil depending on the soil conditions.

3.2 Life-Cycle of Rhizobia

As previously mentioned, rhizobia live in the soil alone with other soil microorganisms (without the legume host) as saprophytes. This stage is called the saprophytic phase. They persist in the soil due to the presence of decaying root nodules from previous symbiosis or in the presence of non-host plants. Then when in association with the legume host, the infective phase occurs after several steps in the rhizobia life cycle. This phase is affected by environmental conditions such as soil salinity and low pH. Then the symbiotic phase takes place after the infective phase. This phase is the development and root nodule function stage in the life cycle of the rhizobia. It is also influenced by harsh environmental conditions that affect the host plant. In the soil, the native rhizobia population is high and there is always competition when new inoculant of rhizobia is introduced.

3.3 *Rhizobium* and Legume Roots

The association between the Rhizobium and legume roots is called symbiotic or mutualistic interaction. Both the Rhizobium and the plant (legume) root benefit from the association and occasionally the partners cannot grow in the absence of the association. The plants are the source of reduced carbon for the rhizobia. The plant growth is often affected or limited by the availability of N and P. Therefore, in order to solve this problem, plants form association with various soil organisms such as rhizobia. The association is mostly with the genera *Bradyrhizobium*and *Rhizobium*. The rhizobia invade the legume through the root hairs or by passage between the epidermal or cortical cells. As a result of some organic bio-chemical compounds released, signals are passed between the rhizobia and the legume and root nodule are formed. The rhizobia fix atmospheric dinitrogen (N_2) gas into ammonia, which the legume plant supplies the rhizobia (bacteria) with reduced carbon as energy source for metabolic processes which includes nitrogen fixation. Because of this interaction, legume plants can grow in extremely mineral nitrogen deficient soil condition.

4.0 CONCLUSION

The rhizobium and *Bradyrhizobium* interact with the legume plant in a symbiobic (mutualistic) association. The rhizobia are special soil microorganisms that are beneficial to the legumes. They can live in the soil alone (Saprophytes) or in the legume root nodules. The rhizobia life cycle involves three phases namely saprophytic, infective and symbiotic phases. In the symbiotic phase, atmospheric dinitrogen (N_2) gas is fixed by the rhizobia in the legume root nodule which the plant assimilates into amino acid, while the plant supplies the rhizobia with reduced carbon as energy source. Because of this association, the legume plant can grow in extreme nitrogen deficient soil.

5.0 SUMMARY

From the discussion in this unit, it can be seen that rhizobia are special bacteria having three stages or phases in it life cycle. It can exist in the soil alone as saprophytes or in association with legumes in root nodules where atmospheric dinitrogen gas is fixed in a symbiotic association. They are specific in their association with legumes. In the soil, the native rhizobia population is high and there is always competition when new species or inoculants of rhizobia are introduced.

6.0 TUTOR-MARKED ASSIGNMENT

1) Discuss the life cycle of *Rhizobium.*
2) Discuss its role in soil and on plant roots

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7.0 REFERENCES/FURTHER READING

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UNIT 5 MYCORRHIZA ASSOCIATIONS

CONTENTS

1.0 INTRODUCTION

Another group of rhizosphere microorganisms which form symbiosis with roots of higher plants is mycorrhiza fungi. A mycorrhiza forms when the right fungus invades a plant root in a process that is similar to the infection by pathogenic fungi. In this association, the fungus and the plant co-exist so that both parties benefit from the relationship. In this unit, you will see the benefits of mycorrhiza symbioses, types of mycorrhiza fungi, the mechanisms of nutrient uptake and the factors affecting mycorrhiza fungi.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- discuss mycorrhiza association
- explain their benefit in agroecosystem
- identify mechanisms of nutrient uptake especially P.
- discuss the factors affecting mycorrhiza fungi.

3.0 MAIN CONTENT

3.1 What is Mycorrhizae?

The term mycorrhizae mean “fungus root”. It is an association where certain fungi invade plant roots and form an extensive network of hyphae that penetrate either between or into the cortical cells and into the soil environment. This association is called mycorrhiza (that fungi and the root of higher plant).

3.2 MycorrhizaAssociation

In this association, the fungi act as an extension of the plant root system helping the plant root to assimilate (take up) nutrients especially phosphorus (P) that is slightly mobile in soil. Due to the low solubility of P, the availability of P in the soil solution is limited. As a result of this problem the fungi helps the plant in the uptake of nutrients from soil solution by penetrating the soil in search of P and other nutrients through the process of active transport mechanism. The fungal hyphae then release the nutrients and the plant roots take them up. Thus the fungus, acts as a sink for nutrients (or reservoir) for further plant use. The mechanism of uptake and release of nutrients especially is not well known. The plant host supplies the fungus with reduced carbon compounds such as the 6-C sugars (hexose). Then the fungus benefits the plant in several ways such as:

a) The fungus increases the uptake of mineral nutrients and water from the soil.
b) The fungus efficiently absorbs mineral nutrients that are in low concentrations compared to non-mycorrhiza roots.
c) The fungal hyphae release hydrolytic enzymes that release N and P from organic compounds. As a result of this mycorrhiza plants are more competitive and better able to tolerate environmental stresses than non- mycorrhiza plants.

3.3 Types of Mycorrhiza Fungi

Mycorrhiza fungi are regularly found in most soils and form extensive network of hyphae that connect different plant species. It has been observed and reported that on larger root systems, different fungi can infect the same root system. Mycorrhiza fungi are classified based on their structure and function namely:

a) Arbuscularmycorrhiza fungi (Endo-mycorrhiza)
b) Ecto-mycorrhiza
c) Ericaceous mycorrhiza (Ericord type)
d) Orchideceousmycorrhiza
e) Ectendomycorrhiza

The first two types are considered the most important especially in agroecosystems and will be discussed in full in this unit.

a) ArbuscularMycorrhizae (AM)

The most important in the group of endo-mycorrhiza association also called endo-mycorrhiza or vesicular-arbuscularmycorrhiza. It is preferably called Arbuscularmycorrhizae because not all form vesicles but all have arbuscles. In this type of mycorrhiza, the fungus initially grows between cortical cells, but soon penetrates the host cortical root cell wall and grows within the cell. As the fungus grows, the plant cell membrane invaginates and envelops the fungus, creating new structures where materials of high molecular complexity are deposited. This structure allows for efficient transfer of nutrients between the plant root and the fungi. These structures are small and highly branched and called arbuscules. They serve to transfer mineral nutrients from the fungi to the host plant and sugar from the host plant to the fungus. Other structures produced are residues, auxillary cells and asexual spores. Vesicles are thin-wall, lipid filled structures which form in the intercellular spaces and serve as storage organs and reproductive propagules for the fungus. Auxillary cells are reported to form in soils and their function is not known. Asexual spores (reproductive) are formed either in the root or soil. The spores are asexual spores. Examples of crop plants that form AM association are corn, cotton, wheat, potatoes, soybean, sugarcane, cassava, rice, apples, grapes, citrus, cocoa, coffee, rubber while cabbage, mustard, canola, broccoli, sugar-beet spinach do not form AM association.

b) Ecto-mycorrhiza

They are associated with forest trees and shrubs like pine, birch, oak, hemlock, spruce and fir found mostly in temperate or semiarid regions. The fungi are mostly basidiomycetes. They grow between root cortical cells producing net – like structure called Hartig net. They produce large quantities of hyphae on the root and in the soil. Many of them produce mushrooms on forest floor. These hyphalsheath (mantle) cover the surface of absorbing root, thereby increasing the effective absorptive surface area of the roots. The hyphae also absorp and translocate inorganic nutrients and water. They also release nutrients from layers of litter. The hyphae penetrate the roots and develop in the tree space around the cells of the cortex but do not penetrate the cortex cell wall. They form stubby white rootlets with a characteristic Y-shape (Hartig net structure).

c) Ericaceous mycorrhizae

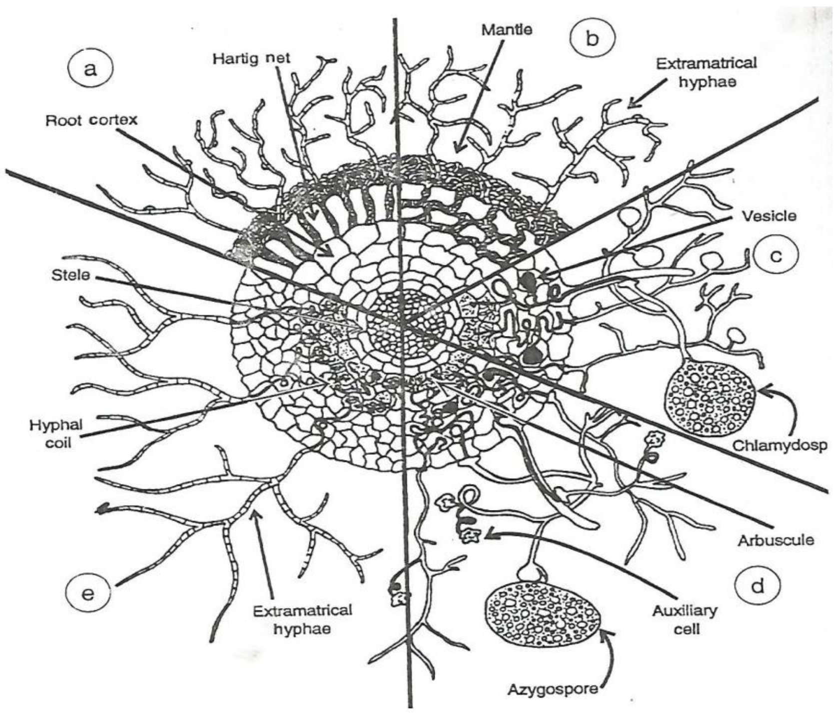
These are found on plants in the order Ericales (Ericaceous shrubs). They dominate temperate soils that are high in organic matter (high organic heathland soils). The ascomycetous fungi are among this group. They grow within (intracellular) cortical cells (endomycorrhizal habit) but with few or little extension into the soil. No arbuscules are formed. The fungus produces extracellular enzymes that mineralize organic matter (OM), thereby making the plants to have access to the nutrients tied up in the OM.

d) Orchidaceous mycorrhizae

Found in the family Orchidaceae (orchids). Orchids have small seeds with little nutrient reserve. The basidiomycetes are among this group. The plant getscolonised after germination. The fungus supplies carbon and vitamins to the developing embryo. The fungus grows into plant cell, investigating the cell membrane forming hyphal coils within the cell. The coils are active for a few days and then lose turgor (strength) and degenerate, and the developing orchid absorbs the nutrient contents. In the mature orchid, mycorrhizae enhance nutrient uptake and translocation also.

e) Ectendomycorrhizae

This is a type of mixed mycorrhiza infection. It forms a typical ectomycorrhizal structure, except that the mantle is thin or lacking and hyphae in the Harting net may penetrate root cortical cells. As the seedling matures, ecto-mycorrhiza is replaced by ectendomycorrhiza. The fungi initially in this group are the designated “E-strain” and later shown to be ascomycetes. In this type of mixed mycorrhiza infection, the host can support more than one type of mycorrhiza association. Examples of this type of mycorrhiza infection is found on Eucalyptus tree (have both AM and Ectomycorrhizal association on the same plant/tree).



Adapted from Brady and Weil (2002)

3.4 Benefits of mycorrhiza fungi in ecosystems

The benefits of mycorrhiza fungi in the ecosystem are seen in its functions and these are:

a) Increase effective absorptive root surface area. This is the root parameter that controls uptake.
b) Increase nutrient uptake and transfer of immobile nutrients.
c) Mycorrhizae impact drought tolerance (improve water uptake)
d) Protect plant from uptake of excessive salt, heavy metal and Al in saline, acid or contaminated soils.
e) Protect plant from pathogen association in the rhizosphere such as parasitic nematodes, soil borne diseases by releasing antibiotics, thereby altering root epidermis.
f) They (mycorrhizae) help in determining plant community structure and plant succession.
g) They improve or help stabilise soil aggregate
h) Their presence enhance nodulation and N_2 fixation by legumes.

3.5 Mechanism of nutrient uptake (especially P)

The fungi produce large quantities of hyphae on the root of plant and in the soil. These hyphal mantle or sheath covers the surface of the absorbing root, thereby increasing the effective absorptive surface area of the absorbing roots. The mycorrhiza roots enhance the uptake of mineral nutrients and water because the external mycelia explore greater volume of soil. The fungus then absorbs mineral nutrients at low concentrations more efficiently by producing various hydrolytic enzymes that release N and P from organic compounds (OM) that are unavailable to the plant.

SELF-ASSESSMENT EXERCISE

What is mycorrhizae? and enumerate the types and functions of mycorrhiza fungi in the soil environment.

3.6 Factors that Affect Mycorrhiza Fungi

a) Conventional agronomic practices affect adversely the diversity and abundance of AM fungi e.g. soil tillage destroys the hyphal networks and mono-cropping under long periods of fallow.
b) Application of large quantities of Phosphorus fertilisers inhibit growth and activity of vegetative mycelium
c) When surface soil is stockpiled during mining and construction activities, it loses its mycorrhiza inoculum potential.
d) Fumigant pesticides adversely affect AM activity in soil
e) Low light (photosynthate limiting) affect growth of mycorrhizae

f) Nutrient status of soil
g) Inoculum potential of the fungi.

4.0 CONCLUSION

From the discussion in this unit, mycorrhiza association is a symbiotic interaction between a fungi and root of higher plant. Extensive network of hyphae is produced that penetrate between or into the cortical cells and soil. The plant supplies the fungus with reduced carbon while the fungus benefits the plant by supplying the plant with nutrients and water especially in deficient or infertile soil thereby improving plant growth and reproduction. Hence, plants that have mycorrhiza association are more competitive and able to withstand harsh environmental condition than non-mycorrhiza plants. Mycorrhiza fungi are classified based on their structure and function. Endo-mycorrhiza (AM) and ecto-mycorrhiza are two important fungi in agroecosystem. The AM fungi form highly branched structures called arbuscules which serve to transfer mineral nutrients from fungus to the plant and sugars from plant to the fungus. Other structures include vesicles (storage organs), auxillary cells and asexual spores. Ectomycorrhizal fungi are mostly basidiomycetes forming Hartig net. Several factors affect mycorrhiza fungi such as host plant diversity and abundance, fertilizer application (especially phosphate fertilizer), mining and construction activities, pesticides and low light and the nutrient status of the soil. They improve soil structure, nutrient availability and water uptake. They also give protection to the plant against diseases and pathogen interaction in the rhizosphere.

5.0 SUMMARY

From the discussion in this unit, a summary table is given below indicating host plant, fungi and important functions of the mycorrhizal associations

Mycorrhizal type	Host involved	Fungi involved	Characteristic structures	Characteristic functions
Arbuscular	Bryophytes Pteridophytes some gymnosperms many angiosperms	Zygomycetes spp., Glomeromycota	Arbuscules vesicles Auxillary cells	Nutrient uptakes soil aggregation
Ectomycorrhizae	Mostly gymnosperms, some angiosperms Restricted to woody plants	Basidiomycetes some ascomycetes Few zygomycetes	Hartig net Mantle Rhizomorphs	Nutrient uptake, mineralization of OM, soil aggregation
Ericaceous	Ericales	Ascomycetes,	Same with	Mineralization of

		Basidiomycetes	either Hyphae in cell or Mantle and net	OM, transfer between plants
Orchidaceous	Orchidaceae	Basidomycetes	Hyphal coils	Supply carbon and vitamins to embryo
Ectendomycorrhizae or mixed infection	Mostly gymnosperms	Ascomycetes	Hartig net, with some cell penetrating, Thin mantle	Nutrient uptake, mineralization of OM

Adapted from Sylvia *et al*. (2007).

6.0 TUTOR-MARKED ASSIGNMENT

1) Discuss mycorrhiza association.
2) Explain their benefit in agroecosystem.
3) Identify mechanisms of nutrient uptake especially P.
4) Discuss the factors affecting mycorrhiza fungi.

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MODULE 3 MICROBIAL TRANSFORMATION OF CYCLES IN SOIL

UNIT 1 SOIL ORGANIC MATTER TRANSFORMATION

CONTENTS

1.0 INTRODUCTION

The vast array of organic matter that is produced on earth during photosynthetic processes does not keep accumulating, rather it is consumed and degraded, and a delicate global balance of carbon is maintained, CO_2 is removed from the atmosphere during photosynthesis and released during respiration. The balance is as a result of the biologically driven, characteristic cycling of carbon, nitrogen, phosphorus, sulphur and iron which are cycled in predictable and definable ways between biotic forms and abiotic forms.

Understanding microbial transformation of cycles in soil allows scientist to understand and predict the development of microbial communities and activities in the environment. As decomposition of plant residues proceeds, microbes slowly breakdown complex compounds into simpler compounds. The soil microbes then metabolise the resulting simpler compounds.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- define soil organic matter
- explain the various components of soil organic matter
- discuss the importance of soil organic matter
- explain the role of microbes in organic matter transformation
- identify how soil organic matter can be maintained.

3.0 MAIN CONTENT

3.1 Definition of Soil Organic Matter

Soil organic matter consists of a wide range of organic substances, including living organisms, remains of organisms that once occupied the soil, and organic compounds produced by current and past metabolism in the soil. The remains of plants, animals and microorganisms are continuously broken down in the soil and new substance issynthesised by other microorganisms.

3.2 Components of Soil Organic Matter

Classification of soil organic matter components is separable by chemical and physical criteria. They include:

- Living organisms (Biomass).
- Identifiable dead tissue (Detrites).
- Non-living, non-tissue.

Humus

Humic substancesNon humic substances

Humic acid Fulvic acid

3.3 Importance of Soil Organic Matter

Soil organic matter is one of the best indications of good soil health (nutrition and structure). There are two major roles or importance of soil organic matter namely

- Improvement of physical properties of soil.
- Enhancement of microbial activitiesPhysical benefits.
- Reduces evaporation loss.

- Stubble mulching.
- Surface mulches control soil erosion.

3.4 Microbial Processes in Organic Matter Transformation

As decomposition of plant residues proceeds, microbes slowly breakdown complex compounds into simpler compounds whilethe soil microbes then metabolise the resulting simpler compounds.

Diverse populations of microorganisms are found in most productive soils and total about several hundred million per gram. Thus, to maintain organic matter, water and air are the critical factors that can limit functions of microorganisms.

Microbial processes include:

- Organic matter decomposition
- Conversion of elements to plant available forms
- Nitrogen fixation.

Organic matter decomposition
A good rate of decomposition is required and the conditions that favour rate of organic matter decomposition by microbes include:

- Adequate soil moisture
- Well aerated soil
- Adequate temperature (optimum)
- C:N ratio
- Quantity and quality of plant residue
- Size of plant materials.

Conversion of elements to plant available forms (e.g. nitrogen transformation) changes in soil N and microbial activities are important in farm management, For example,

- Ammonification – This is the process where organic nitrogen compounds yield ammonia when they decompose or transform.
- Nitrification – The oxidation of ammonia to nitrate by action of nitrifying bacteria *Nitrosonomas* bacteria oxidises ammonia to nitrite while *Nitrobacter* bacteria oxidises nitrite to nitrate.
- Denitrification – This is the biological reduction of nitrate to gaseous nitrogen. This is the most important way of loss of available N in soils.

Nitrogen fixation
This is the process of conversion of atmospheric N into plant available form by the action of microorganisms. Inoculation and growth of

leguminous plants has been major practical use of this, of recent the discovery that N may be fixed by certain bacteria living in close association with the roots of a number of tropical grasses has attracted attention. Examples of N-fixing microorganisms are: *Azotobacter*, *Clostridium*, Blue-green algae and *Rhizobium*.

3.5 Maintenance of Soil Organic Matter

The need to maintain organic matter in soil is of immense importance for sustainable agricultural production. The maintenance or improvement of organic matter includes:

- Crop or plant residues incorporation
- Use of green manures, mulching (cover crop)
- Farmyard or animal manure incorporation
- Compost application
- Use of urban and industrial wastes
- Rotation/Intercropping with legumes
- Fertiliser application and living
- Adoption of reduced tillage (minimum tillage).

4.0 CONCLUSION

In the unit, we have discussed the definition, components, importance, and maintenance of soil organic matter as well as microbial processes in organic matter transformation. Following these discussions, it can be concluded that organic matter produced on earth does not keep accumulating;ratherit is consumed and degraded by microbes for it to be available for sustainable production as well as their importance component and maintenance.

5.0 SUMMARY

In this unit we have learnt that:

- Soil organic matter consists of both living and dead remains of plants and organisms as well as the substances they produce.
- Soil organic matter aids in improving soil structure, texture and water holding capacity and how it can be maintained using some materials.
- As decomposition of plant residues proceeds, microbes slowly breakdown complex components into simpler compounds. And soil microbes then metabolise the resulting simpler compounds.
- Understanding microbial transformation of cycles in soil allows scientists to understand and predict the development of microbial communities and activities in the environment.

6.0 TUTOR-MARKED ASSIGNMENT

1) Define soil organic matter.
2) Explain the various components of soil organic matter.
3) Discuss the importance of soil organic matter.
4) Explain the role of microbes in organic matter transformation.
5) Identify how soil organic matter can be maintained.

7.0 REFERENCES/FURTHER READING

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UNIT 2 NITROGEN TRANSFORMATION

CONTENTS

1.0 INTRODUCTION

More money and effort have been, and are being spent on the management of nitrogen than any other mineral element. And for good reason: The world's ecosystems are probably influenced more by deficiencies or excesses of nitrogen than by those of any other essential element. Were it not for the biological fixation of nitrogen from the atmosphere by certain micro-organisms, and for the recycling back to the soil of much of the nitrogen taken up in natural ecosystems, deficiencies of nitrogen would be even more widespread.

Nitrogen is an essential component of protein. Because of its nutritional importance and relative scarcity, protein is highly sought after by most animals, humans included. Supplying sufficient nitrogen often presents a major expense in agricultural production.

The processes of the nitrogen cycle transform nitrogen from one form to another. Many of those processes are carried out by microbes, either in their effort to harvest energy or to accumulate nitrogen in a form needed for their growth.

2.0 Objectives

At the end of this unit, you should be able to:

- understand the nitrogen cycle
- discuss its reservoirs
- discuss the importance of nitrogen
- explain the various transformations (interconnection and control.

3.0 MAIN CONTENT

3.1 Nitrogen cycle

There is great interest in the nitrogen cycle because nitrogen is the mineral nutrient most in demand by micro-organisms and plants. It is the fourth most common element found in cells, making up approximately 12% of cell dry weight. For example, nitrogen can exist in numerous oxidation states, from -3 I ammonium (NH_4^+) to +5 in nitrate (NO_3^-). The nitrogen cycle is the process by which nitrogen is converted between its various chemical forms. This transformation can be carried out through both biological and physical processes. Important processes in the nitrogen cycle include fixation, ammonification, nitrification and denitrification. The nitrogen cycle is dominated by microbial action.

3.2 Nitrogen Reservoirs

Nitrogen in the form of the inert gas, dinitrogen (N_2), has accumulated in earth's atmosphere since the planet was formed. Nitrogen gas is continually released into the atmosphere from volcanic and hydrothermal eruptions, and is one of the major global resources of nitrogen. A second major reservoir is the nitrogen that is found in earth's crust as bound, non-exchangeable ammonium. Neither of these reservoirs is actively cycled; the nitrogen in earth's crust is unavailable and the N_2 in the atmosphere must be fixed before it is available for biological use. Because nitrogen fixation is energy – intensive process and is carried out by a limited number of micro-organisms, it is a relatively slow process. Smaller reservoirs of nitrogen include the organic nitrogen found in living biomass and indeed organic matter and soluble inorganic nitrogen salts.

3.3 Importance

Nitrogen is an integral component of many essential plant compounds. It is a major part of all amino acids, which are the building blocks of all proteins – including the enzymes, which control virtually all biological processes. Nitrogen is also essential for carbohydrate use within plants. A good supply of nitrogen stimulates root growth and development, as well as the uptake of other nutrients. It can stimulate plant productivity as well as increasing the plumpness of cereal grains, the protein content of both seeds and foliage and the succulence of such crops as lettuce and radishes.

3.4 Transformations

3.4.1 Nitrogen Fixation

Ultimately, all fixed forms of nitrogen, NH_4^+, NO_3^-, and organic N, come from atmospheric N_2. Nitrogen fixation is an energy – intensive process and until recently was performed only by selected bacteria and cyanobacteria. As fertilizers are expensive, management alternatives to fertilizer addition have become attractive. These include rotation between nitrogen-fixing crops such as soybeans and non-fixing crops such as corn. Nitrogen is fixed into ammonia (NH_3) by over 100 different free-living bacteria, both aerobic and anaerobic as well as some Actinomycetes and cyanobacteria. For example, *Azotobacter* (aerobic), *Beijerinckia* (aerobic), *Azospirillium* (facultative), and *Clostridium* (anaerobic) can all fix N_2. Because fixed nitrogen is required by all biological organisms, nitrogen fixing organisms occur in most environmental niches Symbiotic nitrogen fixing bacteria such as Rhizobium usually live in the root nodules of legumes (such as peas, alfalfa and locust trees).

3.4.2 Ammonification

The release of ammonium from organic nitrogen compounds by broad groups of heterotrophic organism is called ammonification

$$\text{Organic N} \longrightarrow NH_4^+$$

3.4.3 Nitrification

The conversion of ammonia to nitrate is performed primarily by soil living bacteria and other nitrifying bacteria. In the primary stage of nitrification, the oxidation of ammonium (NH_4^+) is performed by bacteria such as the *Nitrosomonas* species, which converts ammonia to nitrites (NO_2^-). Other bacterial species such as *Nitrobacter*, are responsible for the oxidation of the nitrites into nitrates (NO_3^-). It is important for the ammonia to be converted to nitrates or nitrites because ammonia gas is toxic to plants.

3.4.4 Denitrification

Denitrification is the reduction of nitrates back into the largely inert nitrogen gas (N_2), completing the nitrogen cycle. This process is performed by bacterial species such as pseudomonas and clostridium in anaerobic conditions. They use the nitrate as an electron acceptor in the place of oxygen during respiration these facultative anaerobic bacteria can also live in aerobic conditions. Denitrification happens in anaerobic conditions e.g. waterlogged soils. The denitrifying bacteria use nitrates in the soil to carryout respiration and consequently produce nitrogen gas, which is inert and unavailable to plants.

3.4.5 Immobilisation

This is the reverse of mineralisation. All living things require N, therefore microorganisms in the soil compete with crops for N. Immobilisation refers to the process in which nitrate and ammonium are taken up by soil organisms and therefore become unavailable to crops. Incorporation of materials with a high carbon to nitrogen ratio (e.g. saw dust) will increase biological activity and cause a greater demand for N and thus result in N immobilisation.

3.4.6 Volatilisation

It is the loss of N through the conversion of ammonium to ammonia gas, which is released to the atmosphere. The volatilisation losses increase at higher soil pH and conditions that favor evaporation (e.g. hot and windy).

3.4.7 Mineralisation

It is the process by which microbes decompose organic N from manure, organic matter and crop residues to ammonium. Because it is a biological process, rates of mineralisation vary with soil temperature, moisture and the amount of oxygen in the soil (aeration).

3.4.8 Leaching

It is the pathway of N loss of a high concern to water quality. Soil particle do not retain nitrate very well because both are negatively charged. As a result, nitrate easily moves with water in the soil. The rate of leaching depends on soil drainage, rainfall, amount of nitrate present in the soil and crop uptake.

3.4.9 Crop uptake

This is the prime goal of N management on farms. The greatest efficiency occurs when adequate N is applied at a time when the crop is actively taking it up.

4.0 CONCLUSION

In this unit, we have discussed the nitrogen cycle, nitrogen reservoirs, importance and the various transformations how they are interconnected and controlled. Following these discussions, it can be concluded that nitrogen is essential for agricultural production, the processes of the nitrogen cycle transform nitrogen from one form to another and these processes are carried out by microbes.

5.0 SUMMARY

In this unit we have learnt that:

- Nitrogen in an essential nutrient that is in demand by microorganisms and plants.
- Nitrogen is gotten from different reservoirs.
- All the transformation processes in nitrogen are interconnected and controlled.

6.0 TUTOR-MARKED ASSIGNMENT

1) Explain the nitrogen cycle.
2) Discuss its reservoirs.
3) Discuss the importance of nitrogen.
4) Explain the various transformations (interconnection and control.

7.0 REFERENCES/FURTHER READING

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UNIT 3 PHOSPHORUS TRANSFORMATION

CONTENTS

1.0 INTRODUCTION

Soil microorganisms have a profound effect on the transformations involved in a large number of biogeochemical cycles other than C and N, such as the macronutrients phosphorus (P) and sulphur (S), and various micronutrients and environmental pollutants.

Phosphorus is found in soil, plats and in microorganisms in a number of organic and inorganic compounds. It is second only to nitrogen as mineral nutrient required by plants and microorganisms, its major physiological role being in certain essential steps in the accumulation and release of energy during cellular metabolism.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain the phosphorus cycle
- discuss its reservoirs
- explain the importance of phosphorus
- discuss the various microbial transformations.

3.0 MAIN CONTENT

3.1 Phosphorus Cycle

Phosphorus cycle is similar to several other mineral nutrient cycles in that P exist in soils and minerals, living organisms and water. Although P is widely distributed in nature, P is not found by itself in elemental form. The phosphorus cycle is the process by which phosphorus is converted between its chemical forms. Important processes in the phosphorus cycle include mineralisation, immobilisation, oxidation and reduction and solubilisation.

3.2 Phosphorus Reservoirs

The largest global reservoir of P is the ocean sediment pool which represents a small but steady sink for terrestrial P released by the weathering of minerals or released by the biota. Phosphorus occurs mainly in inorganic phosphates and in organic phosphates derivatives. Phosphorus in organic molecules constitutes 30 – 50% of the total P in most soils, ranging from as low as low as 5% to as high as 95%.

3.3 Importance

Phosphorus is an element essential to life. It plays both structural and functional roles in virtually all organisms and is found in cell components such as phospholipids, nucleic acids, and DNA as well as promoting root growth and stimulatestillering and hastens maturity which isimportant in cell division and development of new tissues.

3.4 Transformations

3.4.1 Mineralisation

Organically bound P is not directly available to organisms because it cannot be absorbed into cells in this form. For cellular uptake to occur, P must first be released from the organic molecule through the action of phosphate.

3.4.2 Immobilisation

Soil microorganisms can cause fixation or immobilisation of P, either by promoting the formation of inorganic precipitates or by assimilation into organic cell constituents or intracellular polyphosphate granules. The extent of immobilisation of P as affected by the C:P ratio of the organic materials being decomposed and the amount of available P in solution.

3.4.3 Oxidation and Reduction

A number of soil bacteria and fungi have been shown to be capable of oxidising reduced phosphorus compounds (e.g. phosphite) either aerobically or anaerobically. The biochemical pathway for such a microbially medicated reaction has been characterised molecularly and generally, providing some evidence for a previously under-appreciated microbial redox cycle for P.

3.4.4 Solubilisation

The low solubility of P in soils makes it one of the major nutrients limiting plant growth. Frequent applications of soluble forms of P are needed, more than really necessary, because only a fraction is used by plants while the rest rapidly forms insoluble complexes. Traditional P fertilizer production is based on chemical processing of insoluble mineral phosphate ore, which is expensive and environmentally undesirable. In areas where commercially produced P fertilizer is to costly, the microbial solubilisation of phosphate rock is seen as a viable alternative.

4.0 CONCLUSION

In this unit, we have discussed the phosphorus cycle, phosphorus reservoirs, importance and the various transformations how they are interconnected and controlled. Following these discussions, it can be concluded that phosphorus is essential for agricultural production, the processes of the phosphorus cycle transform phosphorus from one form to another and these processes are carried out by microbes.

5.0 SUMMARY

In this unit we have learnt that:

- Phosphorus is an essential nutrient most in need by microorganisms and plants.
- Phosphorus is found in soil, plant and in microorganisms in a number of organic and inorganic compounds.
- Important processes in the phosphorus cycle include mineralisation, immobilization, oxidation and reduction and solubilisation.

6.0 TUTOR-MARKED ASSIGNMENT

1) Explain the phosphorus cycle.

2) Discuss its reservoirs.
3) Explain the importance of phosphorus.
4) Discuss the various microbial transformations.

7.0 REFERENCES/FURTHER READING

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UNIT 4 SULPHUR TRANSFORMATION

CONTENTS

1.0 INTRODUCTION

Soil microorganisms have a profound effect on the transformations involved in a large number of biogeochemical cycles other than C and N, such as the macronutrients phosphorus (P) and sulphur (S), and various micronutrients and environmental pollutants.

Sulphur is a mineral nutrient required by plants and microorganisms, its major physiological role being in certain essential steps in acids stabilisation of protein structure, metabolic activities of vitamins, biotin, thiamine and coenzymes A.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- discuss the sulphur cycle
- explain its reservoirs
- explain the importance of sulphur
- discuss the various microbial transformation.

3.0 MAIN CONTENT

3.1 SulphurCycle

The terrestrial sulphur cycle involves significant interactions between the pedosphere, the hydrosphere the biosphere, and the atmosphere. The S cycle is similar to the soil N cycle and unlike P, these elements undergo chemical and microbial mediated transformations leading to volatilization. Not only is the biosphere a repository for highly mobile forms of S, but several key reactions of the cycle are accelerated by, and sometimes completely controlled by microbiological activity.

3.2 SulphurReservoir

The largest global reservoir of S is the lithosphere. The atmospheric content of S represents a relatively small pool, but one that has increased significantly in recent times due to the burning of fossil fuels.

3.3 Importance

Sulphur is an element essential to life. It serves as a building block of proteins and vitamins, key ingredient in chlorophyll formation, synthesis of oils, increases crop yield and improves quality and the activation of enzymes.

3.4 Transformations

3.4.1 Mineralisation

Carbon-bonded S is mineralised through various pathways:

- Direct aerobic mineralisation during the oxidation of C as an energy source
- Anaerobic mineralisation of organic matter (desulfurisation)
- Incomplete oxidation of organic S into inorganic S compounds
- Biological oxidation of H_2S to sulphate via element S and sulphite.

3.4.2 Immobilisation

Inorganic S is usually assimilated into organic compounds as SO_4^{2-} by plants and most microorganisms.

3.4.3 Oxidation

In the presence of available election acceptors, reduced forms of S are oxidized by both chemical and microbial pathways: A wide variety of organisms are capable of oxidising S in a wide variety of environments. e.g. green and purple sulphur bacteria, Bacteria, Fungi, *Arthrobacter*, *Bacillus* and *Pseudomonas*, etc.

3.4.4 Reduction

Reduction of oxidized forms of S, particularly SO_4^{2-}, by micro-organisms occurs in two different ways. I the first, S is incorporated into cellular constituents such as the S in amino acids. This process is referred to as assimilatory sulphate reduction, or immobilization as described above. In the other, the reduction leads to the following of sulphide (e.g. H_2S) as the end product.

This is referred to as dissimilatory, or respiratory, sulfate reduction. This process is mediated by anaerobic, oligotrophic organisms that use low-molecular weight organic compounds or H_2 as electron donors and the oxidised S compounds as terminal electron acceptors in a process similar to denitrification.

4.0 CONCLUSION

In this unit, we have discussed the sulphur cycle, phosphorus reservoirs, importance and the various microbial transformations. Following these, it can be safely concluded that sulphur is an essential element to life and agricultural productivity, the processes of the sulphur cycle transform sulphur from one form to another and these processes are carried out by microbes.

5.0 SUMMARY

In this unit we have learnt that:

- Sulphur is an essential element to life
- It largest reservoir is the lithosphere
- Important processes in the sulphur cycle include mineralisation, immobilisation, oxidation and reduction.

6.0 TUTOR-MARKED ASSIGNMENT

1) Discuss the sulphur cycle.
2) Explain its reservoirs.

3) Explain the importance of sulphur.
4) Discuss the various microbial transformation.

7.0 REFERENCES/FURTHER READING

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UNIT 5 IRON TRANSFORMATION

CONTENTS

1.0 INTRODUCTION

Despite the fact that it is only a minor nutrient for the growth of most of the microscopic life of the soil, iron is an element which readily undergoes transformation through the activity of the microflora. Iron is always abundant in terrestrial habitats, and it is one of the major constituents of the earth's crust. Microorganisms are implicated in the transformations of iron in a number of distinctly different ways, and the form of the element may be affected through a variety of biological means.

2.0 OBJECTIVES

At the end of the unit, you should be able to:

- identify iron cycling in soil
- discuss its reservoirs
- explain the microbial transformations.

3.0 MAIN CONTENT

3.1 Iron

3.2 Iron Reservoir

Iron is the fourth most abundant element in earth's crust. Iron generally exists in three oxidation states: 0, +2 and +3 corresponding to metallic iron (Fe^{o}), ferrous iron (Fe^{2+}) and ferric iron (Fe^{3+}). In the environment, iron is actively cycled between the +2 and +3 forms. Under aerobic conditions iron is usually found in its most oxidised form (Fe^{3+}) which has low aqueous solubility. Under reducing or anaerobic conditions Fe^{3+} is reduced to the ferrous form, Fe^{2+}, which has higher solubility. Iron is an essential but minor element for biological organisms, making up approximately 0.2%of the dry weight of a bacterial cell.

3.3 Importance

Iron is an essential component of many enzymes, necessary for the synthesis and maintenance of chlorophyll in plants, and plays an essential role in nucleic acid metabolism.

3.4 Transformation

3.4.1 Oxidation and Reduction

Biologically mediated, reduction-oxidation reactions are often associated with energy production by the organisms. Reduced inorganic compounds are oxidized by chemoautotrophs to generate electrons used in ATP production, while oxidized compounds are reduced as an alternative terminal electron acceptor. Micro-organisms are involved in the oxidation of Fe, but not all Fe oxidation is microbial mediated. Ferrous iron (Fe^{2+}) is rapidly oxidized chemically in aerated solution at pH 5. It is therefore difficult to demonstrate conclusively the microbial role of Fe oxidation under mental or alkaline pH conditions. The best evidence for microbial mediated Fe oxidation is from acidophilic bacteria operating at pH 5.

4.0 CONCLUSION

In this unit, we have discussed iron and its reservoir, importance and its microbial transformation. Following these, it can be concluded that iron though a minor nutrient but essential. The process of its transformation from one form to another is carried out by micro-organisms.

5.0 SUMMARY

In this unit we have learnt that:

- Iron is a minor nutrient but essential
- Iron is always abundant in terrestrial habitats
- Important process in iron cycle is oxidation and reduction

6.0 TUTOR-MARKED ASSIGNMENT

1) Identify iron cycling in soil.
2) Discuss its reservoirs.
3) Explain the microbial transformations.

7.0 REFERENCES/FURTHER READING

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MODULE 4

UNIT 1 WHAT ARE ENZYMES?

CONTENTS

1.0 INTRODUCTION

Enzymes are biological catalyst that helps to speed up the rate of reactions without themselves being affected in the reaction or without a permanent change. Most reactions in biological systems take place with the aid of enzymes. The need to study enzymes therefore becomes important considering the fact that these enzymes are involved in almost all biological reactions. The study of these reactions in most proper and convenient way will not be possible if it is not incorporated with the study of enzymes, hence the term enzymology.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- describe soil enzymes
- explain the properties of enzymes
- discuss the roles of soil enzyme activities

3.0 MAIN CONTENT

3.1 The definition of enzymes

Enzymes are biological catalysts that help speed up the rate of a reaction without undergoing any permanent change or alteration. They are specialised proteins that combine with a specific substrate and act to catalyse a biochemical reaction (specific activators). That is, they are specific for the types of chemical reactions they participate in.

The rate of reaction is the amount in moles or grams of reacting particles transformed or products formed per unit time.

3.2 Soil Enzymes

Soil is a living system where all biochemical activities proceed through enzymatic processes. Sources of enzymes in soil are primarily the microbial biomass, though they can also come from plant and animal residues.

Enzymes accumulated in soils are present as free enzymes such as exo-enzymes (extra-cellulars release from living cells); endo-enzymes (intra-cellar released from disintegrating cells; and enzymes bound to cell constituents (enzymes present in disintegrating cells, in cell fragments, and in viable but non proliferating cells)

Free enzymes in soils are adsorbed on organic or mineral constituents or complexed with humic substances or both. The amount of free enzymes in the soil solution is small compared to that in the adsorbed state. Most of the enzymes added to soil by decaying microbial tissues and plant and animal residues are degraded by soil protease and what remains is incorporated with humus.

Therefore, soil as a system of humus and minerals contain both immobilised enzymes and occluded microbial cells. Each of the organic and mineral fractions in soil has a special effect on enzyme activity.

3.3 Properties of Enzymes

1) The catalytic activity of most enzymes requires the activity of some components called cofactors.Cofactors are organic or inorganic components require for enzyme activity. A cofactor can be inorganic (metal ions like Fe^{2+}, Mg^{2+}, Mn^{+}) or organic like cytochromes. When cofactors are covalently bound to an enzyme, they are called Coenzymes. It is therefore, important to note that the catalytic of an enzyme is inefficient without cofactors. The protein component of an enzyme called Apo-protein.Apo-protein complexes with cofactors to form an active enzyme able of catalytic activity are called Holoenzyme (Aproprotein + cofactor).

2) Most enzymes are proteins contrary to the former opinion about enzymes that all enzymes are proteins. It has been observed that some nucleic acids (RNA molecules) have catalytic activity.

3) Enzymes are specific. They are specific in both choice of substrate and the reaction they catalyse. e.g. (1) proteolytic enzymes act on proteins and never on lipids. (2) Trypsin is specific for clearing the carboxyl bond (-Co-) of Arginine and lysine on a polypeptide chain.

Also, specificity of enzymes can either be group specificityor stereo-specificity. In group specified, the enzyme is specific for group of substrates. E.g. Amino acid oxidase – act as an oxidase enzyme in any amino acid. In stereo-specificity, the enzyme is specific for a particular stereo-isomer e.g. Arginine will only act on L-Arginine and not D – Arginine.

4) $E + S \;\; ES \rightleftharpoons E + P \rightleftharpoons$

Enzyme active site

Enzymes (E) have the capacity to form ES (enzyme substrate complex) during catalytic activity. The association of the E and S in the ES complex during catalytic activity is through the region or portion of the enzyme called the Active Site (AS). The AS of an enzyme is the region that binds the S and contributes the residues that are involved in catalytic activities (breakage or formation of bonds). These residues are called catalytic groups.

5) Enzymes have high turnover number (TON) compared to conventional catalyst.
TON= Number of moles of product per minute or per second produced by 1 mole of an active enzyme in a condition whereby the enzyme is saturated with the substrate.

6) Enzymes (Es) catalyse reactions by lowering the activation energy (Ea) of that particular reaction

7) Enzymes catalyse reactions without changing the equilibrium constant of the reaction.

e.g. $A + B \underset{K_2}{\overset{K_1}{\rightleftharpoons}} C + D$

$$\text{Keg} = \frac{[C][D]}{[A][B]} = keg\frac{K_2}{K_1}$$

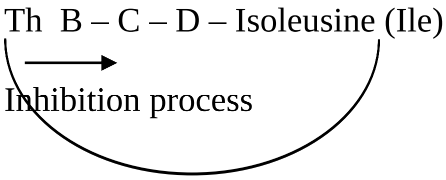
$V_1 = K_1[A][B]$, $V_1 = 20\ sec^{-1}$
$V_2 = K_2[C][D]$, $V_2 = 40\ sec_{-1}$

$\therefore$ keg = 2

Some enzyme reactions can be regulated. The regulation of enzyme activity can be in various forms:

a) Covalent modification:Some enzymes are regulated by covalent groups or addition of certain group or modification of enzymes with a group. E.g.the enzyme that breaks down glycogen. This enzyme is converted to an active form by addition of a PO^{4-} (phosphate) group.

b) Feedback inhibition: This mode of enzyme regulation is typical for reactions that involve steps, e.g. formation of isoleucine from Threomine – the reaction has about four different steps namely:



Reaction of Threominedeaminase byIsoleusine

1. Catalysis by threominedeaminase
2. Inhibition by high concentrations of product Isoleucine thereby stopping the chain reaction
3. Conversion of inactive forms of enzymes (Zymogens) to active forms
4. Cleavage of certain bond or groups in enzymes e.g. (2) Typsinogen is the inactive form of enzyme trypsin (active form). Trypsin formed by cleavage of certain peptide bonds.

3.4 Role of Soil Enzyme Activities

1) Soil enzyme activities are indicators of soil quality because they play important role in soil biology and respond rapidly to changes in soil management.
2) Soil enzyme activities respond to agronomic practices like fertilizer, amendments, vegetation cover, pesticides and soil conversation practices.
3) They are important components of soil humus and take part in biochemical fertility of the soil.

4) Because all biochemical transformations in soil are dependent on or related to the presence of enzymes, therefore soil enzyme activities gives an insight into the biochemical processes in soil.
5) Soil enzyme activities are substrate specific and are related to specific reactions.

SELF-ASSESSMENT EXERCISE

i) Differentiate between cofactors, prosthetic group and coenzymes.
ii) Differentiate between Apo-protein and holoenzyme.
iii) Differentiate between covalent modification and feedback inhibition.

4.0 CONCLUSION

From this unit, it can be concluded that:

- Enzymes are biologically specialised proteins and combine with specific substrate to catalyse all biochemical reactions without undergoing permanent alteration.
- Their catalytic activity requires the activity of some components of the enzymes.
- The primary source of soil enzymes is microbial biomass and is free living soil as either endo- or exo-enzymes.
- They respond rapidly to changes in soil management hence are used as indicators of soil quality.

5.0 SUMMARY

From the discussions in this unit, it can be summarized that soil enzymes are biologically specialized protein that are specific in their reactions and their activities requires the activity of some components of the enzyme. They are of great importance in soil quality assessment due to their rapid response to changes in soil management. Hence, the need to select indicators such as soil enzymes which show adverse effects on yields become serious

6.0 TUTOR-MARKED ASSIGNMENT

1) Describe soil enzymes.
2) Explain the properties of enzymes.
3) Discuss the roles of soil enzyme activities.

7.0 REFERENCES/FURTHER READING

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UNIT 2 ENZYME CLASSIFICATION AND NOMENCLATURE

CONTENTS

1.0 INTRODUCTION

Enzyme nomenclature and classification came to be as a result of the realization that different enzymes have similar names and the same enzymes discovered by different scientists were having different needs. In 1995, the IUPAC and IUB came together to set up an enzyme commission (EC) which is responsible for:

- Development of a standard way of enzyme classification
- Development of a standard way of enzyme nomenclature
- Development of a standard unit of enzyme activity
- Development of standard methods of enzyme assay

In 1961, the EC submitted its first report. Based on the report, enzymes where classified into six (6) different classes.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- write the trivial and systematic names of enzymes
- give the figurative representation of enzymes
- explain or differentiate between the various classes of enzymes

3.0 MAIN CONTENT

3.1 Classes of Enzymes

3.1.1 Class 1 – Oxidoreductases

This group of enzymes catalyses oxidation-reduction reactions. In most reactions involving oxidoreductase NADH, NADPH, NAD^+ and $NADP^+$ serve as donors and acceptors respectively. In most cases NADH and NAD+ serve the same purpose. These enzymes are further subdivided into six or more different sub-classes such as:

1) Dehydrogenases

These enzymes catalyse dehydrogenation reactions, e.g. Alcohol dehydrogenase

$$\text{Ethanol} \xrightleftharpoons[\text{alcoholdehydrogenase}]{NAD^+} \text{aldehyde} + NADH + H^+$$

2) Oxidases

These are enzymes that catalyse addition of oxygen into a substrate with the resultant formation of hydrogen perioxide (H_2O_2). In some oxidase enzyme reactions, water molecules are formed instead of H_2O_2.
e.g. cytochromeoxidase

$$\text{Glucose} \xrightleftharpoons[\text{Glucose oxidase}]{} \text{gluconolactone} + H_2O_2$$

3) Oxygenases

These enzymes catalyse the addition of molecular oxygen (O_2) into a substrate
e.g. catechol oxidase

$$\text{Catechol} \xrightarrow[\text{Catechol oxidase}]{O_2} \text{Cis-muconica}$$

4) Hydroxylases

These enzymes catalyse addition of O_2 into a substrate with resultant formation of a molecule of water (H_2O).

$$\text{e.g. progesterone} + \xrightarrow{NADP^+ + H^+ + O_2} \text{deoxycorticosterone}$$

hydroxylase

5) **Peroxidases**

These enzymes use H_2O_2 as anoxidant

$$H_2O_2 + NADH + H^+ \rightleftharpoons 2H_2O + NAD^+$$

6) **Catalyses**

In these enzymes H_2O_2 serve as an electron donor and electron acceptor and helps in regulating the H_2O_2 produced in living cells.

e.g.

$$H_2O_2 + H_2O \rightleftharpoons 2H_2O + O_2$$

catalase

3.1.2 Class 2 – Transferases

These enzymes catalyse the transfer of groups and/or subgroups from a donor to an acceptor. The subclasses include:

1) Kinases (enzymes transferring phosphate groups)
2) Transaminases (enzymes transferring NH_2 group)
3) Glycosyltransferases (enzymes transferring glycosyl groups)

Other transferases are involved in the transfer of one methyl group (1-carbon transfer), ethyl group among others.

3.1.3 Class 3 – Hydrolases

These enzymes catalyse the hydrolytic cleavage of a group from the molecule. The bonds undergoing the cleavage include C-N, C-C, C-S and C-O bonds. The subclasses include peptidases, esterases, phosphatases, β-glucosidasesulphatases among others.

3.1.4 Class 4 – Lyases

These enzymes catalyse the removal of groups or subgroups with the introduction of a double bond. The subclasses include decarboxylases, citratelyases, deaminase.

3.1.5 Class 5 – Isomerase

These are groups of enzymes that catalyseisomerisation reaction which includes cis-trans isomeriosm, keto isomerism, epimerism among others. Isomerases can act as mutases (enzymes that catalyse the intramolecular arrangement of groups) or as Racemase (enzymes that catalyse the conversion of L-stereoismer to a D-stereoisomer. Also Epimerase is another subclass which catalyseepimerisation reactions.

3.1.6 Class 6 – Ligases

These are group of enzymes that catalyse the addition of two groups at the expense of ATP – Adenosinetriphosphate (ATP is hydrolysed during the reaction) or another form of triphosphate. Ligases can also be called synthetases.

3.2 Enzyme Nomenclature

In the nomenclature of enzymes, two important parameters are to be noted. First, the systematic name and secondary the figurative representation or classification of the enzyme. It is important to note that apart from trivial names of enzymes, systematic names also exist. In the figurative representation/classification, four figures are involved. The first (1st) figure denotes the class of enzyme and the last figure denotes the number of the enzyme in the list of enzymes belonging to the group. The 2nd and3rdfigures depends on the class of enzymes and may differ with respect to classes of enzymes.

a) Oxidoreductases

In naming this class of enzymes, the 2nd digit denotes the group involved in oxidation-reduction (donor) and the 3rd digit denotes the acceptor molecule. The table below shows the figurative representation of oxidoreductase class of enzymes.

	2nd digit (donor, group involved in oxidation – reduction)			3rd digit (acceptor molecule)
1	Alcohol	Primary (1^{o} and secondary 2^{o})	1	NAD^+, $NADP^+$
2	Aldehyde		2	Cytochrome, Fe^{3+}
3	C_2H_5 (-C-C-)		3	O_2 (oxygen)
4	1^{o} amine			
5	2^{o} amine			
6	NADH			

e.g. Alcohol dehydrogenase EC 1.1.1.1

b) Transferases

	2nd digit (group transferred)		3rd digit (chemical nature of the group transferred)
1	One carbon atom	1	Methyl group
2	Aldehyde or ketone	2	Hydroxymethyl group
3	Acyl	3	Acids (carboxyl)

4	Glycosidic (carbohydrate)	
5	Alkyl group (except methyl ($-CH_3$ group)	
6	Nitrogenous group	
7	Phosphate(phosphorus containing group)	
8	Sulphate group	

e.g. Glucokinase EC 2.7.1.2

c) Hydrolases

The trivial names for enzymes under this class are mostly in the form of addition of "ases" at the end of the substrate undergoing hydrolytic action e.g. in glycosidase, the glycosidic bond is cleaved hydrolytically. In the systematic name for hydrolase enzymes, the word hydrolase is written after the substrate being hydrolysed. The 2nd and 3rd digits denotes type of bond being hydrolysed and specific nature of the chemical groups hydrolysed respectively as shown below:

	2nd digit (bond type hydrolysed)		3rd digit (chemical nature of the group hydrolysed)
1	Esther	1	Acid $C-O^-$
2	Glycosidic	2	Thiolesther $C-S^-$
3	Ether	3	Phosphate monoesther O^- -P=O
4	Peptide	4	Phosphate diester O-P-P=O
5	C-N		
6	Acid anhydride		
7	C-C		
8	Acid halide P-N		

e.g. Acid phosphate EC 3.1.3.2

d) Lyases

The systematic name involves the addition of "Lyase' after the substrate undergoing the enzyme reaction. The trivalnames involves decarboxylase indicating elimination of CO_2 (carbondioxide). Example of a typical trival name is Oxaloacetate decarboxylase. The enzyme decarboxylates oxaloacetates. The systematic name should be oxaloacetate carboxylase (EC 4.1.1.3).

	2nd digit (type of bond lysed)		3rd digit (chemical nature of the group lysed)
1	C-C	1	Carboxylic acids
2	C-O	2	Aldehydes

3	C-N	3	Keto acids
4	C-S		

e.g. Oxolocarboxyacetate EC 4.1.1.3

e) Isomerases

Here, the name of the substrate undergoing isomerisation reaction is written first followed by the type of isomerism. E.g. Glucose-6-phosphate-1-epimerase (G-6-P-1 epimerase) EC 5.1.3.1

	2nd digit (type of isomerisation)		3rd digit (nature of the group isomerisation)
1	Isomerism at a chiral centre (e.g. Racemase, epimerism)	1	Amino acids
2	Cis-trans isomerism	2	Hydroxyacids
3	Intramolecular oxidation/reduction	3	Carbohydrates
4	Intramolecular group transfer		

f) Ligases

In the nomenclature of ligase enzyme the systematic name is written as two groups undergoing ligation

e.g. A + B A – B ⟶ 1st group =:
A:B ligase 2nd group = ligase

The trivial name of ligase enzyme is synthetase. All synthetase reactions utilize energy from ATP. E.g. (1) Acetyl CoA synthetase (Trivial name) Acetate: COASH ligase (systematic name)
EC 6.2.1.1

	2nd digit (type of bond formed)		3rd digit (nature of chemical bond formed)
1	C-O	1	Amide bond –C-NH_2
2	C-S	2	Peptide bond –C-N-
3	C-N (mostly considered in 3rd digit)	3	
4	C-C		
5	O-P		

e.g. (2) Glutamine synthetase (Trival name) L-glutamate:ammonia ligase (systematic name)
EC 6.3.1.2

SELF-ASSESSMENT EXERCISE

i. Write the trivial and systematic names of two enzymes from each class of enzyme and give the figurative representation of each.
ii. Write noting their 4th digits/figures.

4.0 CONCLUSION

From the unit, it can be concluded that enzymes are classified into six classes and each class has subclasses, using the standard developed by enzyme commission (EC). The systematic name and figurative representation or classification is the two important parameters in enzyme nomenclature. Four figures or digits are involved in the figurative representation.

The 1st being the class of enzyme while the last denotes the number of the enzyme in the list of enzymes in that group. The 2nd and the 3rd digits figures depends on the class of enzyme and differ due to their classes.

5.0 SUMMARY

Having realised that different enzymes have similar names and different needs, it became necessary to set up an enzyme commission that developed the standard for enzyme classification and nomenclature putting into consideration enzyme activities and enzyme assays. Enzymes were classified into six classes as shown below:

	Class	**Catalytic function**
1	Oxidoreductases	Oxidation – reduction reaction
2	Transferases	Group transfer reaction
3	Hydrolases	Hydrolysis reactions (i.e. hydrolytic cleavage reactions)
4	Lyases	Reactions involving removal of a group (leaving a double bond) or addition of a group to a double bond
5	Isomerases	Reactions involving isomerisations
6	Ligases	Reactions joining together two molecules coupled with the utilisation of high energy bonds

6.0 TUTOR-MARKED ASSIGNMENT

1) Write the trivial and systematic names of enzymes.
2) Give the figurative representation of enzymes.
3) Explain or differentiate between the various classes of enzymes.

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UNIT 3 SOIL ENZYMES IMPORTANT IN AGRICULTURE

CONTENTS

1.0 INTRODUCTION

The last unit discussed the six classes of enzymes and their catalytic functions. In this unit, you will learn of some enzymes that are of great importance in agriculture. Enzyme activities are substrate specific and specific in reactions, therefore one enzyme activity value alone cannot give an overall picture of the soil status. Hence, measurement of various enzymes will be more useful to evaluate biochemical activity and related processes. The enzymes selected are mainly responsible for biochemical processes in carbon, nitrogen and phosphorus cycles in soil.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain the importance soil enzymes in agriculture
- discuss the reactions they catalyse

- determine the enzymes reactions in soil.

3.0 MAIN CONTENT

3.1 Soil Enzymes Important in Agriculture

3.1.1 Dehydrogenase (EC 1.1.1.48)

Dehydrogenases are often measured because they are found only in living systems, and are important to enzyme systems of all microorganisms (that is, they are intracellular enzymes). They are responsible for the oxidation – reduction reaction of soil organic matter by transferring electrons or hydrogen from substrate to acceptors. Their activity gives an indication of a good measure of microbial oxidative activity. Therefore, dehydrogenases are used in evaluating metabolic activity of soil microorganisms.

3.1.2 Phosphatase (EC 3.1.3.2)

There are two types of phosphatase enzymes namely acid phosphatase and alkaline phosphatase. These enzymes are under the class of hydrolase enzymes. They are good indicators of soil facility. They affect the P-acquisation and P-use efficiency of plants and catalyse the hydrolysis of P-ester bonds ad anhydrides of H_3PO_4 from the organic matter resulting in the release of inorganic phosphorus. They hydrolyse the P-ester bond of the P-nitrophenyl phosphate and then P-nitrophenol is released. They are the key enzymes in P-cycling in soil and release phosphorus (P) from both cellular and extracellular organic compounds at different stress levels. Their activity is higher where higher quantity of organic C is found. Management practices in mixed cultures which include P stress in the rhizosphere may affect the secretion of these enzymes. Understanding the activity of the enzyme is important for predicting their interactions as their activities may in turn regulate nutrient uptake and plant growth in soil.

3.1.3 Beta-Glucosidase(EC 3.2.1.21)

Beta (β)-glucosidase formerly known as gentiobiase or cellobiasecatalyses the hydrolysis of β-glucosides such as B-D-glucopyranosides and gives an idea about the decomposition of organic matter. This enzyme is related with carbon cycle (C-cycle). This enzyme converts cellulose to glucose which is an important energy source for soil microorganisms. Cellulose is the most important organic compound and its mineralisation and degradation in soil is a major process in the carbon cycle. β-glucosidase activity therefore, is considered an indicator for biomass turnover. This enzyme is more dominant in soils than Alpha (α)-glucosidase.

3.1.4 Protease (ec 3.4)

This enzyme catalyses the hydrolysis of proteins (Proteolysis) which is an important step in the organic nitrogen cycle (ammonification and nitrification) of soils. It helps in the maintenance of soil fertility and catalyse substrates with peptides. They reflect the proteolytic potential of a soil and thus indicate the protein degradation capacity.

3.1.5 Urease (EC 3.5.1.5)

Urease (urea amidohydrolase) is the enzyme the catalyses the hydrolysis of urea to CO_2 (carbon dioxide) and ammonia (NH_3)

$$NH_2CONH_2 + H_2O \longrightarrow CO_2 + 2NH_3$$

Urease acts on the C-N bonds other than the peptide bonds and two C-N bond are broken in hydrolysis of urea. It is the first enzyme protein to be crystallised. It is very widely distributed in nature and has been found in microorganisms, plants and animals.

3.2 Enzyme Assay

When investigating enzyme reactions in soil, you must develop an assay for the enzyme. Enzyme assay is the addition of a known amount of soil to a solution containing a known concentration of substrate species and determining the rate at which the substrate is converted to a product under carefully controlled conditions of temperature, pH and ionic strength.

1) **Dehydrogenase assay**
   This method involves the release of tetrazoliumformazan when soil is treated with tris-HCl buffer (pH 7.5) and INT solution added (iodonitrotetrazolium chloride – INT) and kept at 40 ^oC for one hour (1 hr). Then methanol is added and sample mixed and left in the dark for some minutes. The INTF released is extracted by filtration and determined spectrophotometrically.

2) **Phosphatase assay**

This method involves the colorimetric estimation of the P-nitrophenol released by phosphatase activity when soil is incubated with buffered (pH 6.5 for acid phosphate activity and pH 11 for alkaline phosphate activity) sodium P-nitrophenyl phosphate solution and toluene at 37 ^oC for one hour. The calcium chloride ($CaCl_2$) and NaOH (sodium hydroxide) are added and the soil suspension filtered.

3) β-glucosidase assay

The principles of this method are similar to that of phosphatase. In this case P-nitrophenol is released by β-glucosidase when soil is inculcated with buffered (pH 6.0) PNG solution (P-nitrophenyl- β-D-glucoside and toluene at 37 ^oC for one hour. The P-nitrophenol released is extracted by filtration after-addition of $CaCl_2$ and THAM buffer (pH 12) and determined colorimetrically.

4) Protease assay

This method involves the release of Tyrosine when soil is treated with casein denatured by heat and dissolved in Tris-HCl buffer (pH 8) and incubated at 51 ^oC for one hour 30 minutes. Then Tris-HCl and acetic acid are added. The amino acid formed is extracted by filtration and determined colorimetrically.

5) Urease assay

The method designed for assay of urease under optimum conditions are based on determination of the $NH4^+$ (ammonium ion) released when soil is incubated with THAM buffer (Tris(hydroxymethyl) amino methane), urea solution and toluene at 37 ^oC for two hours. The NH_4^+ released is determined by treatment of the incubated soil sample with KCl (potassium chloride) containing Ag_2SO_4 (silver sulphate) and steam distillation with MgO for few minutes.

All the enzyme activities measured are expressed as micromole of product released per hour per gram of soil used.

3.3 Factors Affecting Enzyme Assay

Soil treatments and methods of preparation have marked effect on the results obtained in enzyme assays. Air-drying, storage conditions, storage temperature and grinding affect enzyme assays. Preparation of homogenous soil sample is essential because enzymes have been shown to be unevenly distributed in the soil fractions. Shaking the soil substrate mixture affects enzyme activities and should be put into consideration when studying kinetics of enzyme reactions in soil.

4.0 CONCLUSION

From this unit, soil enzymes are potential indicators of soil quality because of their role in soil biology, biochemical fertility of soil and rapid response to changes in soil management. In conclusion, because all biochemical transformations in soil are dependent on, or related to the presence of enzymes, studying soil enzyme activities will give you an insight into the biochemical processes in the soil.

5.0 SUMMARY

The enzymes and the reactions they catalyse are summarized in the table below:

Enzyme	**Reaction**
Dehydrogenase	Oxidation of organic compound using hydrogen as acceptor INT + H^+ $\longrightarrow$ INTF + HCl
Acid phosphatase	Orthophosphoric monoester + H_2O $\longrightarrow$ An alcohol + orthophosphate
B-glucosidase	Hydrolysis of terminal, non-reducing β-D-glucose residues with release of β-D-glucose
Protease	Hydrolysis of proteins to petides and amino acids
Urease	Hydrolysis of urea to CO_2 and NH_4^+

6.0 TUTOR–MARKED ASSIGNMENT

1) Explain the importance soil enzymes in agriculture.
2) Discuss the reactions they catalyse.
3) Determine the enzymes reactions in soil.

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UNIT 4 KINETICS OF ENZYME REACTIONS IN SOIL

CONTENTS

1.0 INTRODUCTION

Two most important properties of enzymes are their specificity and catalytic efficiency. This involves the velocities of chemical reactions of the enzymes (chemical kinetics). The reaction of any enzyme is measured by the rate of the chemical reaction being catalysed by the enzyme. In this unit, you will learn about Michaelis-Menten theory of enzyme reactions in soil and other transformations of the equation derived from this theory. Also the factors affecting the rates of enzyme reactions will be discussed.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain the Michaelis-Menten theory of enzyme reactions
- derive the Michaelis-Menten equation and other transformations of the equation
- discuss the factors affecting the rate of enzyme reactions.

3.0 MAIN CONTENT

3.1 Theory of Enzyme Reactions in Soil

Enzymes are proteins that are specific in their reactions and catalytic efficiency. The enzyme reactions called chemical kinetics because it involves velocities of chemical reactions is measured by the rate of the chemical, reaction being catalyzed by the enzyme. Equation describing the kinetics of enzyme reactions in heterogenous system like soil is given below:

K1K3
E + S ⇌ K2 ES → E + Product
Enzyme substrate Enzyme-substrate complex

This is a rate equation of enzyme with a single substrate where K_1, K_2, K_3 are velocity constants or rate constants of the three processes. Generally, from the above equation, the complex of enzyme and substrate is unstable and proceeds through a number of steps of rearrangement to form the product plus the original enzyme. This theory of enzyme action was proposed by Michaelis and Menten (1913).

3.1 Derivation of one Substrate Enzyme Catalyzed Reaction (Michaelis-MentenEquation)

E + S ⇌ ES…… (1)
K +1
ESK-1 E + P ..….. (2)

Assumption 1 by Michaelis-Menten

- Formation of ES complex
- Breakdown of ES complex

Assumption 2

- Reaction must be reversible
- Rate constant of forward and backward reactions have positive and negative subscripts respectively

The initial velocity Vo = Rate of breakdown of ES complex
V_o = K + 2 [ES] …… (3), cannot be determined directly
Rate of formation of ES = $\frac{d[ES]}{dt}$

$\frac{d[ES]}{dt}$ = K + 1 [(ET) – (ES)][S] …. (4)

Note: $E_T - E_S = E_1$

$\frac{d[ES]}{dt}$ = K – 1 [ES] + K +2 [ES] …… (5) Reverse reaction

At steady state, rate of formation of ES = Rate of formation of ES

$\therefore$ K + 1([ET] – [ES]) [S] = K – 1 [ES] + K + 2 [ES] …… (6)

Rearranging equation (6), you have

$\frac{K-1+K+2}{K+1}$ = Km (Michaelis constant) …… (7)

$\therefore$ [ES] = $\frac{[ET][S]}{Km+[S]}$ ……. (8)

From equation (8) Vo = K + 2 [ES]

[ES] = $\frac{Vo}{K+2}$

Substitute foe ES in equation (3), its value from equation 8 will be

Vo = $\frac{K+2[ET][S]}{Km+[S]}$ ……….. (9)

K + 2 [ET] = V_{max}………… (10)

Therefore V_o = $\frac{V\max.[S]}{Km+[S]}$ …….. Michaelis-Menten equation of ES complex

3.2 Transformation of MichaelisMentenEquation

From the equation $\frac{V\max.[S]}{Km+[S]}$, three other linear transformations are obtained

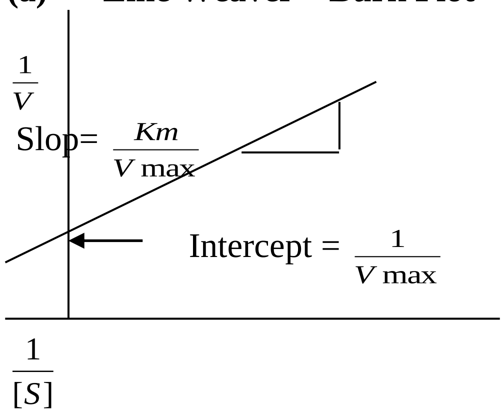
a) $\frac{1}{V}=\frac{1}{V\max}+\frac{Km}{V\max}\frac{1}{[s]}$ - This is called Linear Weaver Burk Transformation

b) $\frac{[S]}{V}=\frac{Km}{V\max}+\frac{1}{V\max}.[S]$ - Hane-Wolf Transformation

c) $V = V\max - Km \bullet \frac{V}{[S]}$ - Eadie-Hofstee Transformation

Plots of the above transformations give straight lines. Once the K_m and $V_{max}$are known for an enzyme reaction, the reaction velocity (V) can be calculated for any substrate concentration. K_m is important in estimating the substrate concentration that will give V_{max}.

(a) Line Weaver – Burk Plot

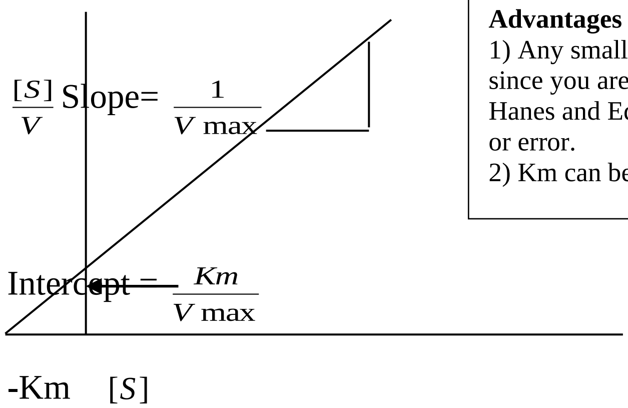


Disadvantages of LWB plot

When plotting V against [S], the graph appears to be a rectangular hyperbolic curve. You cannot know when you are approaching Vmax, hence you cannot adequately calculate Km.

1) Because you are taking reciprocals $(\frac{1}{V} and \frac{1}{[S]}$ any small error will be magnified.
2) There is clustering of the points when 1/V is plotted against 1/[S]

(b) Hane-Wolf Plot

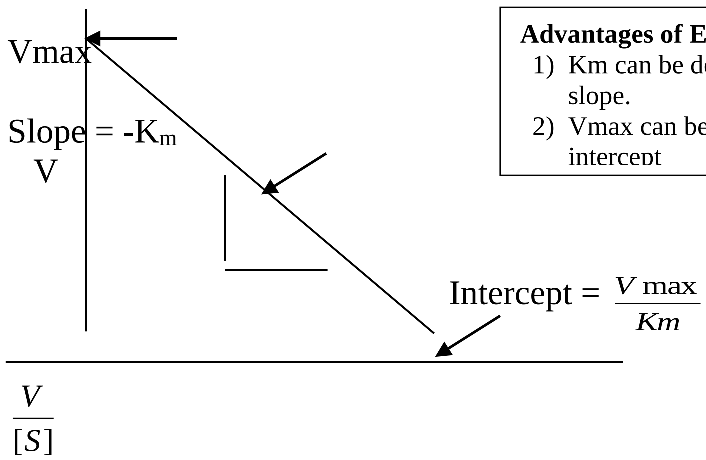


Advantages of Hane and Eddie

1) Any small error in LWB will be magnified since you are plotting 1/V against 1/[S], hence Hanes and Eddie plots will correct the mistakes or error.

2) Km can be calculated from the errors in LWB.

(c) Eddie-Hofstee Plot



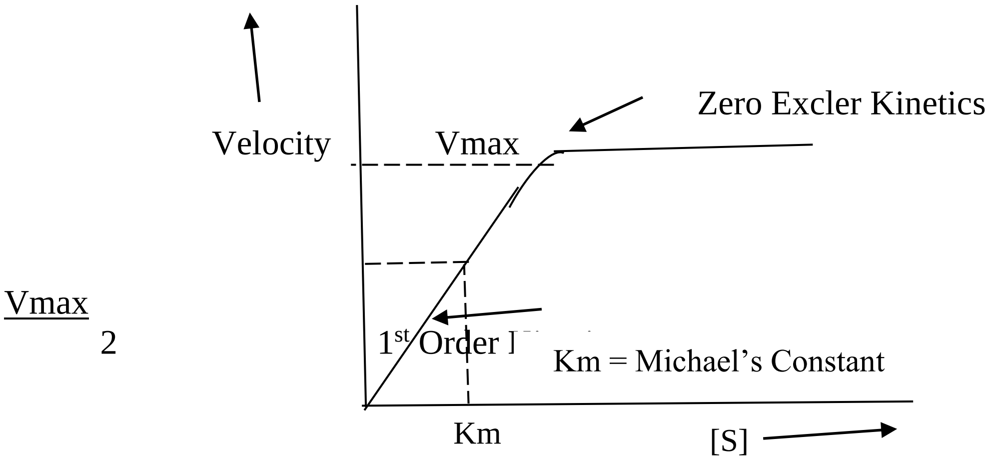
Advantages of Eddie Plot

1) Km can be determined directly from slope.
2) Vmax can be determined directly from intercept

3.3 Factors Affecting Rates of Enzyme Reactions

3.3.1 Concentration of Enzyme and Substrate

Most enzyme catalyzed reactions are either zero order reaction (reaction where the rate is constant and independent of substrate concentration) or first-order reaction (reaction where the rate at any time is proportional to the existing substrate concentration). Irrespective of the order of reaction, the rate of reaction is proportional to the enzyme concentration.



The graph shows when the enzyme concentration is constant and the substrate concentration vary over a wide range; the velocity of enzyme catalyzed reaction is shown.

1) At low substrate concentration, the velocity of reaction follows a first order reaction equation.
2) At higher substrate concentrations, a maximum velocity is obtained independent of further amounts of substrate and this follows a zero order reaction equation.
3) The magnitude of the Vmax (maximum velocity) is proportional to the concentration of the enzyme. That is, the higher the enzyme concentration, the higher the Vmax.

3.2.2 Temperature

The activity of any chemical reaction increases with temperature, doubling for every 10 ^{o}C increase. The rate of enzyme reaction increases until it reaches a maximum beyond which the rate reaction begins to decrease due to enzyme inactivation. Enzyme catalyzed reactions are less sensitive to temperature changes compared to uncatalysed reaction. Enzyme catalyzed reaction rate increases by a factor of < 2. The dependence of temperature on the rate constant is described by Arrhenius equation.

$$K = A \exp\left(-\frac{Ea}{RT}\right)$$

Where
A = Pre exponential factor
Ea = Activation energy
R = Gas constant
T = Temperature (°K)
The log form of the equation is written as

$$\text{Log } K = \left(\frac{-Ea}{2.303RT}\right) + \log A$$

Log A and Ea are intercept and slope respectively of the graph of logK on $\frac{1}{T}$

3.2.3 pH

Enzymes being proteinaceous, show marked changes in ionisation due to pH fluctuations, each enzyme has a pH value at which the rate of reaction is optimal and below or above this optimum the rate is lower. Therefore, the catalytic action of enzymes operates within a pH range. Enzymes are more stable close to their optimum pH and are irreversibly denatured at extremes in acidity or alkalinity. The use of buffer is essential to maintain optimum pH for the reaction duration.

3.2.4 Cofactors, Activators, Inhibitors and Ionic environment

Many enzymes are not catalytically active except in combination with a non-protein (cofactor) component. These non-protein components are heat stable, dialysable substances of low molecular weight, cofactors are activators, coenzymes and prosthetic groups.

Prosthetic groups are substances that are bound firmly to the enzyme. Coenzymes are organic substances that are freely dissociable from enzymes. Some enzymes are activated by inorganic ions are not changed during the catalysed process but are required before the enzyme can carry out the reaction. These are called activators.

Inhibitors are compounds that slow down the rate of an enzyme catalysed reaction. There are many ways they can act:

1) Inhibitors usually form complexes with various enzyme forms and thus lowers the amount of enzyme available for participation in a normal sequence.
2) They can also inhibit by complexing the substrate

3) The product of a reaction can also inhibit by recombining with the enzyme form that results from the dissociation of the product.
Thereby raises the steady state concentration of the enzyme form that yields the products when it dissociates.

A dead end inhibitor is one which neither acts as substrate nor product but forms complexes with one or more enzyme forms.
Salts (Ionic environment) also affect or show clear effects on enzyme activity. Changes in electrolyte concentration cause changes in the activity coefficients of the reactants and the activated complex. The degree of ionization of acidic and basic groups in the protein molecule may be changed by the presence of salts. All these effects may either accelerate or slow down the catalytic activity of the enzyme.

3.4 Factors Affecting Enzyme Activities in Soil

Among the many factors previously mentioned that affect enzyme activities, these factors listed below are included:

- Cropping history
- Soil amendments
- Environmental factors such as seasonal variations, rainfall
- Water logging (this affects Redox potential (Eh) and pH thereby affecting enzyme activity)
- Vegetation
- Agricultural chemicals (fertilizers, pesticides)
- Industrial pollutants.

SELF-ASSESSMENT EXERCISE

i. Why do all enzyme – catalyzed reactions show a pH optimum?
ii. State the advantages and disadvantages of any two of the transformations of Michaelis equation.
iii. Using suitable diagram, distinguish between Hane-Wolf, Eddie Hofstee and Lineweaver-Burk plots.

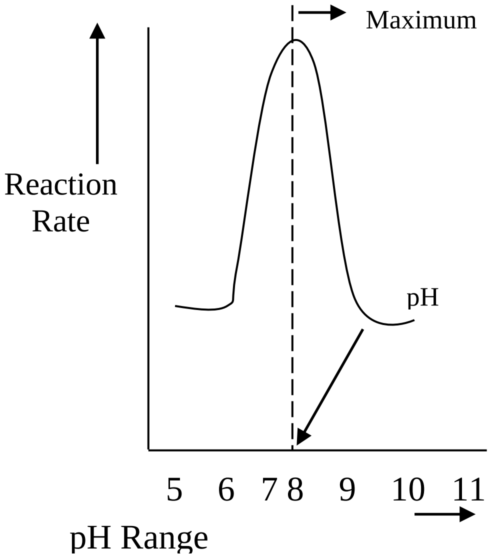
4.0 CONCLUSION

From the study enzymes are specialized proteins that combine with specific substrate and specific activators. They are specific for the types of reactions they carry out or participate in various physical and chemical factors have been identified that affect enzyme reactions in soil. These factors are concentrations of enzyme and substrate, temperature, pH, cofactors, ionic strength of the environment, cropping history, rainfall, vegetation type, agrochemicals and industrial pollutants. All these factors depend on soil type, dose of chemical applied and the conditions of the study area.

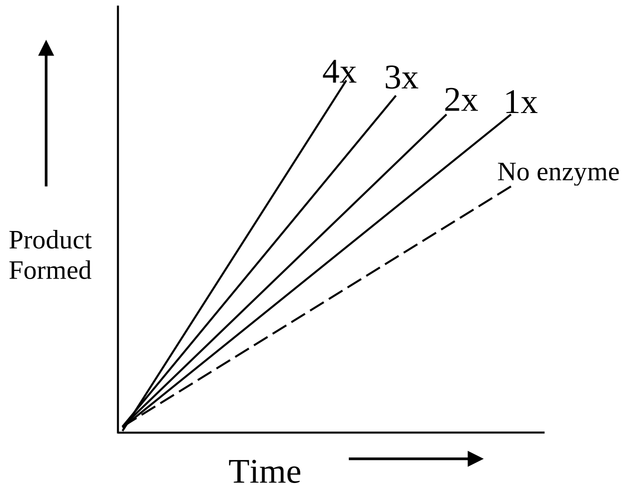
5.0 SUMMARY

Graphical representation of factors affecting enzyme activity.

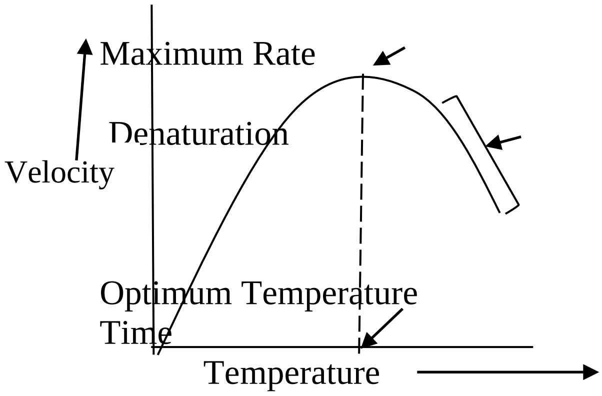
1)pH effects on enzyme activity



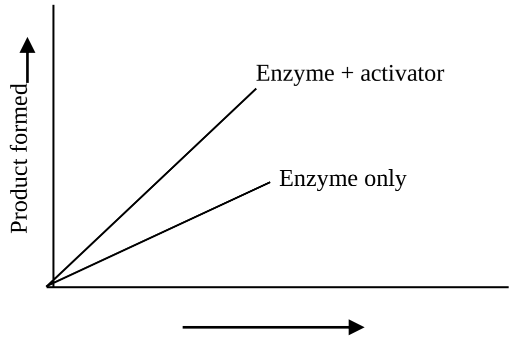
4) Effect of enzyme concentration



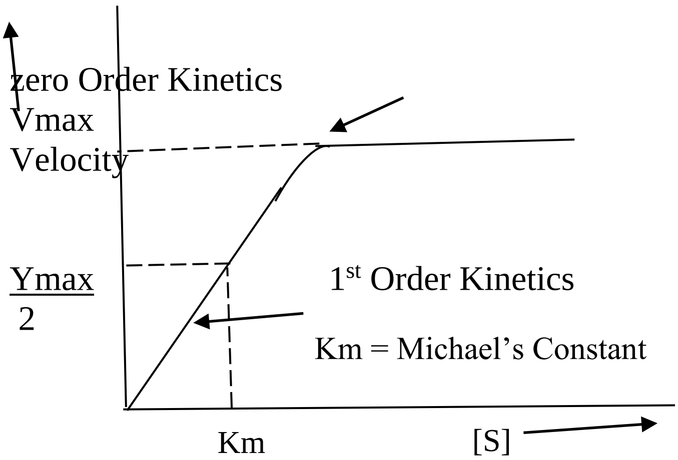
2) Temperature effect



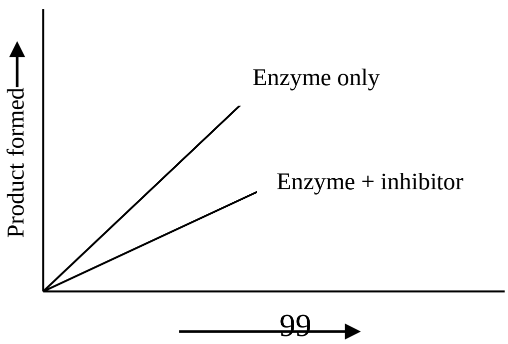
5) Effect of activator



3) Effect of substrate concentration



6) Effect of inhibition



Time

6.0 TUTOR –MARKED ASSIGNMENT

1) Explain the Michaelis-Menten theory of enzyme reactions.
2) Derive the Michaelis-Menten equation and other transformations of the equation.
3) Discuss the factors affecting the rate of enzyme reactions.

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UNIT 5 ENZYME INHIBITION

CONTENTS

1.0 INTRODUCTION

Soils receive a variety of organic and inorganic chemicals and some of these compounds like fertilisers, pesticides, municipal and industrial wastes are added as part of soil and crop management. Other compounds such as salts of trace elements are added to soils as impurities in lime and fertilisers and as industrial pollutants. These compounds inhibit and slow down the rate of enzyme catalysed reactions.

2.0 OBJECTIVES

At the end of this unit, you should be able:

- explain an inhibitor and define enzyme inhibition
- discuss the various types of enzyme inhibition
- differentiate between the various types of enzyme inhibition using suitable diagrams.

3.0 MAIN CONTENT

3.1 Definition and Functions of Inhibitors

3.1.1 Definition

An inhibitor is a compound that slows down the rate of an enzyme catalysed reaction. The process is called inhibition. Enzyme inhibitors are available in:

3.1.2 Functions of Enzyme Inhibition

a) Mechanism and pathway of enzyme catalysis
b) Substrate specificity of enzymes
c) The nature of final groups at active sites
d) Helping to determine the groups that can participate in maintaining the active conformation or structure of the enzymes.

3.2 Types of Enzyme Inhibition

Enzyme inhibition has been classified into two namely reversible and irreversible inhibitions.

3.2.1 Reversible Enzyme Inhibition

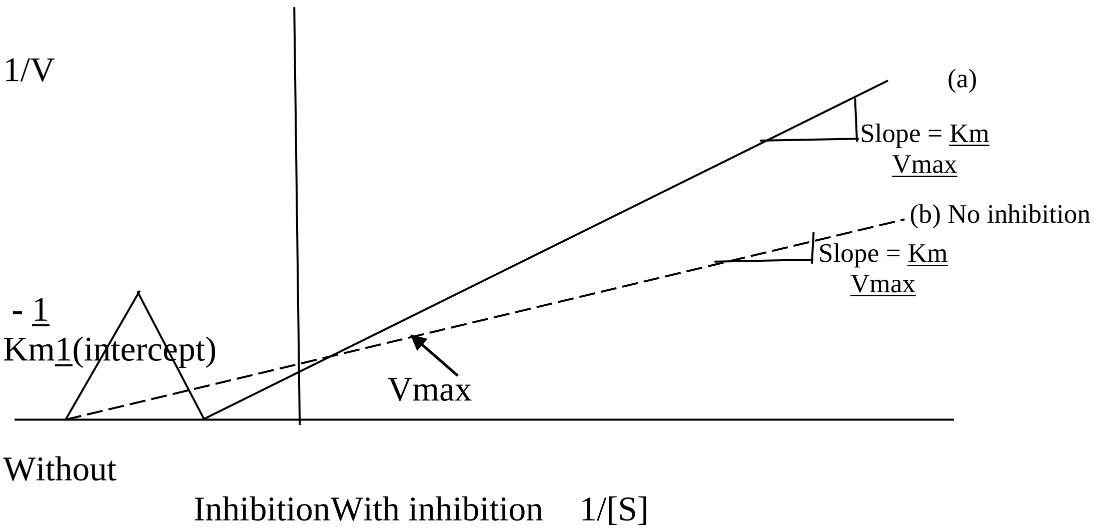
There are three major types of reversible enzyme inhibition. They are:
a) Competitive (binds with only enzyme)
b) Uncompetitive (inhibitor binds with ESC)
c) Noncompetitive or mixed inhibition (inhibitor binds with either enzyme or ESC).

The reversible inhibition pattern can be distinguished by the effect of the reaction kinetics on the enzyme. This is analysed based on Michaelis-Menten equation. For valid kinetic analysis the inhibitor must react rapidly and reversibly with the enzyme or with the enzyme-substrate complex (ESC).

3.2.1.1CompetitiveInhibition

The inhibitor can combine with free enzyme in such a way that it competes with the normal substrate for binding at the active sites. The competitive inhibitor reacts reversibly to form enzyme-inhibitor complex (EI) similar to ESC. The inhibitor molecule is not chemically changed by the enzyme. The presence of competitive inhibitor increases the apparent Km of substrate to achieve its maximum velocity.

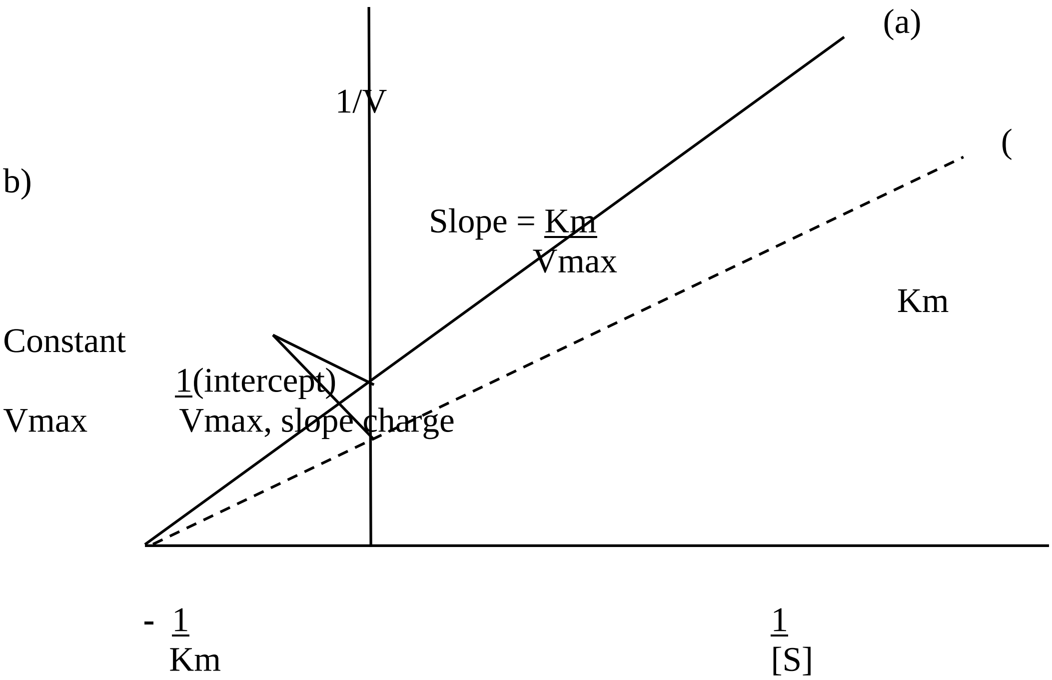
Conversely, competitive inhibitor does not affect the rate of breakdown of enzyme-substrate complex (ESC).



LWB Plot of Michaelis-MentenEquation Showing Competitive Inhibition (A) in The Presence of Inhibitor.

3.2.1.2 UncompetitiveInhibition

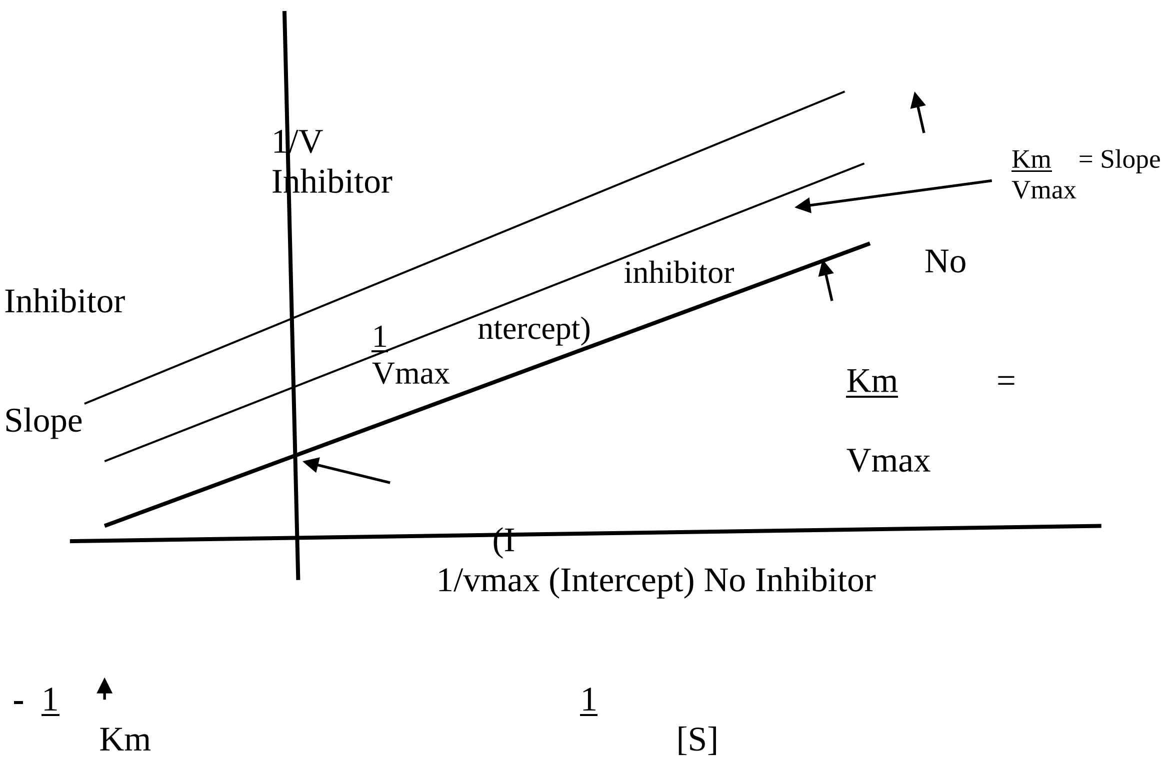
The inhibitor does not combine with the free enzyme or affect its reaction in normal substrate but combined with ESC to give an enzyme-substrate inhibitor complex (ESI) which cannot undergo reaction to give a product. From the graph below, this relationship suggests that degree of inhibition may increase with increase in substrate concentration. This type of inhibition can be recognised in the double reciprocal plot.



The slope of the plot remains constant as the concentration of the inhibitor increases. Uncompetitive inhibition is rare in one substrate catalyzed reaction but is common in two substrate reaction.

3.2.1.3 Non-competitive or Mixed Inhibition

It can interfere with the free enzyme or with the ESC (that is, a noncompetitive inhibitor) interfering with the action of growth. This inhibitor binds to a site on the enzyme other than the active site with the sole effect of deforming the enzyme. So it does not form the enzyme substrate complex (ESC) or if formed cannot decompose at the normal rate to form the product. These effects are not reversed by increasing substrate concentration. The inhibition reaction results in two inactive forms of EI complex and ESI complex. Some enzymes that require metal ions for activity are inhibited non competitively by agents capable of binding metal ion.



LWB-plots of Michaelis-Menten equation showing Non-competitive or mixed inhibition (a) in the presence of inhibitor and (b) in the absence of inhibitor.

3.3 Irreversible Inhibition

Some enzymes undergo irreversible inhibition when they are treated with agents that can covalently and permanently modify their final groups which is needed for catalysis. As a result, the enzyme molecule becomes inactive. Irreversible inhibition cannot follow Michaelis-Menten principles which assume reversible formalism. Irreversible inhibitors are very valuable in mapping of active sites.

4.0 CONCLUSION

From this unit, you have learnt that an inhibitor slows down the rate of an enzyme catalyzed reaction by complexing with the enzyme or substrate or the product recombines with the enzyme. Two types of enzyme inhibition were identified namely reversible and irreversible inhibition and the reversible inhibition consisted of three major types, competitive, uncompetitive and Non-competitive or mixed inhibitions.

5.0 SUMMARY

The table below summarises the effect of the inhibitors on the Lineweaver-Burk (LWB) plot

Inhibition	**Slope**	**Intercept**
No inhibition	$\frac{Km}{V\max}$	$\frac{1}{V\max}$
Competitive	$\frac{Km}{V\max}\left(1+\frac{[1]}{KI}\right)$	$\frac{1}{V\max}$
Uncompetitive	$\frac{Km}{V\max}$	$\frac{1}{V\max}\left(1+\frac{[1]}{KI}\right)$
Mixed or Noncompetitive	$\frac{Km}{V\max}\left(1+\frac{[1]}{KI}\right)$	$\frac{1}{V\max}\left(1+\frac{[1]}{KI}\right)$

7.0TUTOR-MARKED ASSIGNMENT

1. Explain an inhibitor and define enzyme inhibition.
2. Discuss the various types of enzyme inhibition.
3. Differentiate between the various types of enzyme inhibition using suitable diagrams.

7.0 REFERENCES/FURTHER READING

Eldor, A.P. (Ed.). (2007). Soil Microbiology, Ecology and Biochemistry. (3rd ed.). Canada:Academic Press Publications

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MODULE 5 PESTICIDES IN SOIL

UNIT 1 NATURE OF PESTICIDES

CONTENTS

1.0 INTRODUCTION

A lot of synthetic organic compounds are manufactured yearly such as plastics, plasticisers, lubricants, refrigerants, fuels, solvents, preservatives and pesticides. Some of these organic compounds or chemicals are extremely toxic to microbes and humans while some are inactive and harmless. These synthetic organic chemicals are found everywhere in our environment (soil, groundwater, plants and even our bodies). They reach our environmentaccidentally through leakage and spills, planned spraying as is done on our agricultural fields, or by other treatments. Pesticides are therefore synthetic organic chemicals designed to kill pests (any organism that the pesticide user see to be damaging). Hence, in this unit, you will be looking at the nature of pesticides, their benefits and costs, behavior in soil, effects of soil organisms and their persistence.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain the nature of pesticides
- discuss the benefits and costs of pesticides
- differentiate between the various types of pesticides.

3.0 MAIN CONTENT

3.1 What are Pesticides?

Pesticides have become the most widespread organic pollutants associated with soils. Pesticides are synthetic organic chemicals designed to kill or inhibit pests. A pest is any organism that the pesticide users see to be damaging. When pesticides are applied, those that find their way into the soil may inhibit or kill soil organisms resulting in the imbalance of the soil community. Others may be transported from the soil to air, water or vegetation (plants) where they may be contacted, inhaled or ingested by some organisms. Therefore, it is important to control the release of these organic chemicals and know their fate (behavior/mode of action) and effects in the soil. Some may enter as contaminants from industrial organic wastes applied or spilled on soils as components of machine parts thrown away, lubricants or as agrochemicals (pesticides) applied to agricultural fields.

For those meant to be applied on plants as pesticides, most of it reaches the soil because they have missed the insect or plant leaf that was the target (purpose) of the application. Hence, they are the most widely causes of organic pollutants connected with soils.

3.2 Classification of Pesticides

Pesticides are classified into two:
1) Based on their target (pest) organisms
2) Mode of action

3.2.1 Classification Based on Target Organism

Pesticides under this group are namely:
a) Insecticides – insect killers

b) Fungicides – fungi killers
c) Herbicides – weed killers
d) Rodenticides or Alkalicides – mice or rodent killers
e) Nematicides – nematode killers
f) Molluscides – snails or slugs/Mollucs killers
g) Bactericides – bacteria killers.

The first three are used in large quantities and are therefore more likely to contaminate the soil. Most pesticides contain aromatic rings of some kind.

3.2.2 Classification Based on Mode of Action

Pesticides are further divided into two main groups based on their mode of action namely:
a) Contact pesticides
b) Systemic pesticides.

3.2.2.1 Contact Pesticides

These pesticides do not penetrate into the tissue and therefore are not translocated within the vascular system or tissue or transport system of the target plant. They are susceptible to weather effects and cannot stop infection once infection has begun or started to spread through plant tissues. They are only protectants, example: cypermethrin designed to prevent insect attack on cowpea during flowering and podding stages of growth and fungicide designed to prevent seed rot, example: Benomyl (Benlate).

3.2.2.2 Systemic Pesticides

These pesticides effectively penetrate the cuticle and move through the plant vascular system. Example: Benomy (benlate) is a fungicide chemotherapeutant. It not only protects from fungal attack but cures and inhibits any established infection.

Systemic pesticides have three qualities namely:
a) Those that are not absorbed – protectants
b) Those absorbed but not translocated – chemotherapeutants
c) Those absorbed and translocated which are the main systemic pesticides.

3.3 Types of Pesticides

3.3.1 Insecticides

There are three general groups here namely:

a) The chlorinated hydrocarbons
Example: DDT
DDT was the most extensively used insecticide in this group until banned due to its low biodegradability and persistence as well as toxicity to birds and fish.

b) The organophosphates
They are generally biodegradable and less likely to build up in soils and water but they are extremely toxic to humans, hence great care must be used in handling and applying them. Example: Parathion.

c) The Carbamates
They are the least dangerous because they are readily/rapidly biodegradable and have relatively low mammalian toxicity but highly toxic to honeybees and other beneficial insects and earthworms. Example: Carbaryl.

3.3.2 Fungicides

Fungicides are mainly used to control diseases of fruits and vegetable crops and also used as seed coating (dressing) to protect against seed rot (Benlate (Benomyl)). Some are used to protect harvested fruits and vegetables from decay andto prevent wood decay and to protect clothings from mildew. Examples: Thiocarbamates and Triazoles are currently in use.

3.3.3 Herbicides

The amount of herbicides used is much compared to insecticides and fungicides combined. Most herbicides are biodegradable and relatively low in mammalian toxicity but some are quite toxic to fish and other wildlife.

Also, they can have adverse effects on beneficial aquatic vegetation that provide food and habitat for fish and shell fish.
Examples:

a) The Triazines – Atrazine used for the control of weed in corn.
b) Substituted ureas – Linuron
c) Some carbamates
d) The relatively new sulphonylureas – Nicosulphuron. They are potent (very effective) at very low rates
e) Dinitroanilines – Trifluralin
f) Acetanilides – Alachlor (This is quite mobile in the environment).

3.3.4 Nematicides

Some of them are known to contaminate soils and water draining from treated soils. Examples: Some carbamates used as nematicides are quite soluble in water, not adsorbed by soil (clay particles) and hence leached downwards and into the ground water.

3.3.5 Molluscides

They are used for the killing of slugs and snails.

3.3.6 Rodenticides/Alkalicides

They are used for the killing of mice or rodents such as rats. Quite a number of rat killers are found in the market.

3.4 Benefits and cost of Pesticides

3.4.1 Benefits of Pesticides

Pesticides have provided many benefits to the society such as:

1) Control of mosquitoes and other vectors that cause human diseases such as yellow fever and malaria.
2) Protect crops and livestocks against insect pests and diseases.
3) Control weeds by the use of herbicides without which conservation tillage (especially no-tillage/zero tillage) would be much difficult to adopt.
4) Much of the progress made in controlling soil erosion would not have come about without herbicide.
5) They reduce food spoilage from farms or fields (that is, they are used for storage of food and grains).
6) Biological nitrogen fixation process is enhanced (stimulated) by some pesticide use by reducing protozoa activity and other competitors or predators of nitrogen fixing bacteria.

3.4.2 Cost of Pesticides

As the benefits to society are great, so are the costs. Pesticides should not be seen as something you cannot do without (indispensible), because some farmers produce profitable yields without the use of pesticides. Even with the use of pesticides, insects, diseases and weeds still cause crop loss. Therefore, the use of large amount/quantities of pesticides cause:

1) Imbalance in microbial community: Though they are designed to kill living things, many are potentially toxic to organisms other than the pests for which they are intended.
2) Detrimental to beneficial insects (butterflies, bees, etc) and certain soil organisms (Rhizobium, nitrifying bacteria).
3) Fungicides especially fumigants have adverse effect on soil fungi and actinomycetes thereby slowing down the humus formation in soil.
4) They cause health hazards to humans and animals. Those that do not quickly breakdown may be biologically magnified (i.e. increased) as they move up the food chain, for instance, earthworms ingest contaminated soil and the chemicals tend to concentrate in their bodies. When birds and fish eat them, the pesticides build up to lethal levels.
5) The near extinction of certain birds of prey is as a result of the devastating environmental pollution consequences of pesticide use.
6) Studies have shown that human hormone balance (endocrine) may be disrupted by small traces of some pesticides found in water, food and air.

SELF-ASSESSMENT EXERCISE

1. Explain the nature of pesticides.
2. Discuss the benefits and costs of pesticides.
3. Differentiate between the various types of pesticides.

4.0 CONCLUSION

From our study or discussions in this unit, pesticides are synthetic organic compounds and classified into about seven groups based on their target organisms and into two major groups based on their mode of action. They are beneficial to the society and soil organisms and at the same time have detrimental effects; hence care must be taken to control the release of pesticides.

5.0 SUMMARY

In this unit we have learnt that pesticides are classified in broad terms as insecticide, fungicides, herbicides, nematicides, molluscides, rodenticides (alkalicides) and bactericides used to control or inhibit insect pests and diseases. In terms of mode of action, they are classified as contact and systemic pesticides.

They have provided many benefits but also have adverse effects on both humans and microorganisms, hence you must exercise caution in the use of pesticides.

6.0 TUTOR-MARKED ASSIGNMENT

1. Explain the nature of pesticides.
2. Discuss the benefits and costs of pesticides.
3. Differentiate between the various types of pesticides.

7.0 REFERENCES/FURTHER READING

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UNIT 2 BEHAVIOR OF PESTICIDES IN SOIL

CONTENTS

1.0 INTRODUCTION

Once pesticides reach the soil, they move in one or more ways from the soil to the air, water or vegetation (plants) where they may be contacted, inhaled or ingested by some organisms. Therefore, it is important to control their release and know their fate (behavior or mode of action) and effects in soil.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- identify the various ways in which pesticides move in soil
- discuss or explain these various modes of action in soil.

3.0 MAIN CONTENT

3.1 Fate of Pesticides in Soil

Once pesticides reach the soil, they move in one or more ways such as:

1) They may vaporise into the atmosphere without chemical charge.
2) They may be adsorbed by soil particles like silicate clays.

3) They may move downwards through the soil in liquid or solution form and be lost from soil by leaching into underground water.
4) They may undergo chemical reactions within or on the soil surface.
5) They may be broken down by soil microorganism.
6) They may wash into streams and rivers in surface runoff and leaching.
7) They may be taken up by plants or soil animals (organisms) and move up the food chain.

Note that the specific fate of these pesticides is determined at least in part by their chemical structures, which are highly variable.

3.1.1 Volatility

Pesticides vary in their volatility and susceptibility to atmospheric loss. Example, fumigants like methyl bromide (now banned) was selected because of its high vapour pressure that allows it to penetrate the soil pores to reach the target organisms.

But this encourages rapid loss to the atmosphere after treatment, unless the soil is covered or sealed. A few herbicides (Trifluralin) and fungicides (Pentachloro nitrobenzene -PCN B) are volatile and make vaporization their primary means of loss from soil.

3.1.2 Adsorption

This depends on the characteristics (structure) of the compound and the soil type to which they are added. For instance, soils high in organic matter and soils with high surface area clays tend to be the strongest adsorbents for some compounds while soils with oxide coatings on soil particles strongly adsorb others. The presence of functional groups like OH, NH_2, NHR, $CONH_2$, COOR and $^+NR_3$ in the chemical structure encourages adsorption especially on the soil humus. Hydrogen bonding and protonation (addition of H^+ to a group such as an $–NH_2$ (amino) group) promotes some of the adsorption. Large organic molecules with many charged sites are more strongly adsorbed. Herbicides like Diaquat and Paraquat with positively charged groups are strongly adsorbed by silicate clays. Adsorption by clays of some pesticides is pH dependent with maximum adsorption at low pH, thus encouraging protonation.

3.1.3 Leaching and Runoff

The rate of leaching from soil depends on their solubility in water and their potential for adsorption. Example: Chloroform and Phenoxyacetic acid are more water soluble than DDT (1,1 – Trichloro 2,2 bis 4-

chlorophenyl ethane) and Pentachloro benzene (PCB) which are soluble in oil but not in water. High water solubility favours leaching losses while strongly adsorbed molecules are not likely to move down the profile. Hence, conditions that favour adsorption discourage leaching. The greatest leaching occurs in highly permeable sandy soils that are low in organic matter. High rainfall periods at the time of application promote leaching and runoff losses. Herbicides are more mobile than fungicides and/or insecticides and therefore more likely to find their way to groundwater supplies and streams.

3.1.4 Contamination of Groundwater

Pesticides reach the groundwater through accidental spillage and from normal agricultural use. Many people depend on ground water (well water) for their drinking supply; therefore leaching of pesticides is of great concern because the amount of pesticides discovered in drinking water was found to be high.

3.1.5 Chemical Reactions

Some pesticides undergo chemical modification independent of soil organisms once they get in contact with soil. Examples are:

1) Fe-cyanide decompose within hours or days on exposure to bright sunlight
2) DDT, Diquat and Triazines photo-decompose slowly in sunlight.
3) Triazine herbicides (example: Atrazine) and organophosphate insecticides (example: Malathion) undergo hydrolysis and subsequent degradation. Therefore, degradation independent of soil organisms does occur.

3.1.6 Plant bsorption

Pesticides are commonly absorbed by higher plants especially systemic insecticides and most herbicides because they are taken up to perform their intended function. The absorbed chemical may remain inside the plant or be broken down (degraded). Some of the products of degradation may be harmless or toxic than the original chemical absorbed. Those that remain on the plant parts whether as fresh fruits and vegetables or as processed food became of concern to the society.Hence, the strict regulation of the use of pesticide residues in food to ensure human safety by regulatory agencies.

3.1.7 Microbial Metabolism

This is the most important method by which pesticides are removed from soils (biochemically degraded ≡ biodegradation). Polar groups like –OH, COO^- and $–NH_2$ provide points of attack for the organisms.

Insecticides such as DDT and other chlorinated hydrocarbons like Aldrin, Dieldrin and Heptachlor are degraded slowly and persist in soil for long (about 20 years or more). Organophosphates (Parathion) are degraded rapidly by a number of organisms. Herbicides like 2, 4-D, Phenylureas, Aliphatic acids and Carbamates are readily attacked by a host of organisms but Triazines are slowly degraded mainly by chemical action.

Most fungicides depend on microbial degradation (decomposition) though their rate of breakdown is slow causing residue problems.

SELF-ASSESSMENT EXERCISE

Enumerate the fate of pesticides in soil and discuss microbial metabolism of pesticides.

4.0 CONCLUSION

From our discussion in this unit, pesticides once they reach the soil, behave in seven different ways namely volatilization, adsorption, leaching and runoff, groundwater contamination, chemical reactions, plant absorption and microbial metabolism, where they may be contacted, inhaled or ingested by some organisms. Therefore, in conclusion, it is important to control their release having known their fate (behavior/mode of action) in soil as they are the most widespread synthetic organic chemicals associated with soil.

5.0 SUMMARY

In summary, most of the pesticide applied in agricultural soils finds their way through or into the soil in one or several ways namely through:

- Volatilisation
- Adsorption
- Leaching and runoff
- Contamination of groundwater
- Chemical reactions
- Plant absorption
- Microbial metabolism

Therefore, knowing their mode of action will help to control their release in order to minimise environmental pollution, health hazards on both humans and soil organisms. Therefore, the knowledge of pesticides behavior will be of great importance in agricultural production.

6.0 TUTOR-MARKED ASSIGNMENT

1) Identify the various ways in which pesticides move in soil.
2) Discuss or explain these various modes of action in soil.

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UNIT 3 EFFECT OF PESTICIDES ON SOIL ORGANISM

CONTENTS

1.0 INTRODUCTION

Most of the pesticides meant to be applied on plants missed the insect or plant leaf that was the target of application and reach the soil. The soil is the habitat for diversity of organisms. These pesticides vary in their effects on soil ecology (soil microorganisms). Their effects could be detrimental or beneficial.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- discuss how pesticides adversely affect various soil organisms
- explain the effects of pesticides on the beneficial soil microorganism.

3.0 MAIN CONTENT

3.1 Effect of Pesticides on Soil Organisms

3.1.1 Detrimental Effects of Pesticides on Soil Organisms

Pesticides vary widely in their effects on soil ecology (soil organisms) and this in turn affects plant growth. Some of the major effects of pesticides on soil organisms are:

- Imbalance in the soil ecology especially on soil microorganisms (microflora)

- Detrimental (adverse) effects on the fertility of the soil and crop productivity
- Adverse effect on nitrification and nitrogen fixation by affecting the bacteria responsible for these processes, e.g. *Rhizobium*, *Azotobacter*, *Nitrosomonas*, *Nitrobacter*. Insecticides and fungicides affect both processes more than most herbicides.Thus, the soil Nitrogen balance is altered.
- Symbiotic (mutualistic) associations in soil are adversely affected such as mycorrhiza association and root nodulation in legumes
- Rhizosphere microorganisms are altered due to the changes in the soil physical to chemical properties.
- Fungicides especially fumigants affect adversely the fungi and actinomycetes activities thereby slowing down the humus formation in soil.

The negative effects of most pesticides on the beneficial soil microorganisms are temporary and after a few days or weeks, organism numbers recover. Hence, care must be taken to apply them when alternative means of pest management are not available.

3.1.2 Effects of Pesticides on Beneficial Soil Organism

1) The bacterial levels in the soil are not so badly affected by pesticides but the organisms (bacteria) responsible for nitrification and nitrogen fixation are badly affected sometimes. For instance, insecticides and fungicides affect both processes more than most herbicides though some of the later (herbicides) can reduce the number of organisms (population) carrying out these two reactions.
2) Studies have shown that some pesticides enhance biological nitrogen fixation (BNF) by reducing the activity of protozoa and other competitors or predators of nitrogen fixing bacteria.
3) Ammonification is enhanced (stimulated) as a result of the above (2 above).

SELF-ASSESSMENT EXERCISE

In what ways do pesticides affect soil organisms?

4.0 CONCLUSION

From our discussions in this study, it can be concluded that pesticides have detrimental effects on most soil organisms, especially on the beneficial soil organism responsible for nitrification and nitrogen fixation. They also affect or slow down soil formation by adversely affecting fungi and actinomycetes activities. Studies have also shown that pesticides enhance BNF by reducing the activities of the competitors and predators (protozoa) of nitrogen fixing bacteria resulting in increased ammonification process.

5.0 SUMMARY

From our study, we have learnt that pesticides have effect on the soil microbial ecology. Their effects were both beneficial and detrimental to soil organisms and to the soil itself by adversely affecting fungi and actinomycetes activities in soil formation.

6.0 TUTOR-MARKED ASSIGNMENT

1) Discuss how pesticides adversely affect various soil organisms.
2) Explain the effects of pesticides on the beneficial soil microorganism.

7.0 REFERENCES/FURTHER READING

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UNIT 4 BIODEGRADATION OF PESTICIDES

CONTENTS

1.0 INTRODUCTION

Pesticides applied on plant leaves in agricultural fields directly or indirectly reach the soil. The ability and effectiveness of the soil to degrade the toxic organic compounds to nontoxic compounds depends on the physical, chemical and biological properties of the soil. The biological properties (soil microorganisms) play the most important role in pesticide degradation. This process of pesticide degradation by soil organisms is called “Biodegradation”. It is important to note that all pesticides are not biodegradable as shown in Unit 2 – fate of pesticides in soil. Those that resist biodegradation are called “recalcitrant”.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain biodegradation
- discuss the various reactions of biodegradation

3.0 MAIN CONTENT

3.1 What is Biodegradation?

Pesticides in soil are turned into nontoxic compounds by soil microbes. Because the process takes place in soil, it is affected by several soil factors such as organic matter content, moisture, temperature, pH including microbes (microorganisms). These soil factors are essential in the degradation of pesticides applied into soil. Pesticides degrade faster under tropical (warmer) climate compared to temperate (cooler) climate. Studies show that most organic pesticides degrade within 3 – 6 months under warm conditions. Soil organisms such as bacteria, fungi and actinomycetes play most of the role in pesticide degradation in soil.

3.1.1 Conditions Favourable for Biodegradation

The following conditions have been observed or reported for proper and good biodegradation of pesticides in soil:

1) The soil must be favourable for microbial and plant growth as well as for enzyme activity.
2) The soil organism carrying out the degradation process must have the needed catabolic activity in order to lower the concentration of the contaminant.
3) The contaminant must be biologically available (i.e. must have life in it).
4) The disadvantages of bioremediation must be less than the process of removal of contaminant (biodegradation).

3.2 Other Reactions of Biodegradation

Other reactions that take place during biodegradation are:

1) Detoxification:
   This is the process whereby the toxic compounds in a pesticide are converted to nontoxic compounds. This reaction takes place without the help of microorganisms.
2) Degradation:
   This is the breakdown of toxic complex compound into simple products through the process of mineralisation. Degradation and mineralisation go together.
3) Conjugation:
   Here the microorganism makes the substrate more complex or combines the pesticide with the cell metabolites.
4) Activation:
   This is the conversion of nontoxic substance a toxic compound
5) Change in toxicity spectrum:
   Here the pesticide is metabolized into products inhibitory to other organisms.

SELF-ASSESSMENT EXERCISE

What is biodegradation and recalcitrant?

4.0 CONCLUSION

From our discussion in this unit, soil microorganisms play the most important role in biodegradation, though not all pesticides are biodegradable. Biodegradation of pesticides are affected by soil factors hence the soil must be favourable for microbial and plant growth as well as enzymatic activity.

The microorganisms must have catabolic activity to lower the concentration of the contaminant. Other reactions apart from degradation by microorganisms that take place are detoxification, conjugation, activation and change in toxicity spectrum. In conclusion, the ability of soil to properly degrade pesticides depends on the physical, chemical and biological properties of soil.

5.0 SUMMARY

In this unit, you have learnt that the ability of the soil to degrade pesticides properly depends on the soil factors including soil microorganisms. Most pesticide degradation are carried out by soil organisms, though there are exceptions. The soil condition must be favourable for proper and good degradation to take place. Other reactions reported to take place during biodegradation were discussed such as detoxification, conjugation, activation and change in toxicity spectrum.

6.0 TUTOR-MARKED ASSIGNMENT

1) Explain biodegradation.
2) Discuss the various reactions of biodegradation.

7.0 REFERENCES/FURTHER READING

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UNIT 5 PESTICIDE PERSISTENCE IN SOIL

CONTENTS

1.0 INTRODUCTION

The persistence of pesticides in the soil is a summation of all the reactions, movements, and degradations affecting the pesticides. Majority of pesticides degrade rapidly to prevent build-up in soils with normal annual (yearly) applications. Those that resist biodegradation cause environmental damage mostly. Continuous use of the same pesticide on the same land (especially on agricultural lands) can increase the rate of microbial (microorganisms) breakdown of that pesticides. This is because there will be a population build-up of those microorganisms with the enzymes needed to breakdown the compound. This is an advantage but the breakdown sometimes may become rapid to reduce pesticide effectiveness.

2.0 OBJECTIVES

At the end of this unit, you should be able to:

- explain what pesticide persistence means
- enumerate the effects of persistence of pesticides in soil.

3.0 MAIN CONTENT

3.1 What Do You Mean By Pesticide Persistence?

When we talk of pesticide persistence, we mean how long it takes the pesticide in question to breakdown to its nontoxic compounds that will no longer be effective on the target (i.e. the purpose for which it is applied) organism.

Persistence of pesticide therefore, is the summation of all the reactions, movements and degradations affecting the pesticide(s). Some pesticides degrade rapidly while others take long periods (even years) depending on the chemical structure (especially the functional group) and properties of the organic compounds in the pesticide.

3.2 Effects of Pesticides Persistence in Soil

When a pesticide persists longer than expected in soil, it could result (lead) to:

1) Chronic health hazards: When pesticides are taken up (assimilated) by plants, they move up the food chain and accumulate to toxic levels as root tubers and leaves. When eaten by humans or livestocks, it would lead to chronic health problems.
2) Toxicity to microorganisms in soil: Accumulation of the organic compounds in a pesticide to a level that becomes toxic to microorganisms thereby inhibiting microbial activity in soil or resulting in the death of the microbes.
3) Contamination of ground water, streams, and rivers: Some pesticides reach the streams, rivers and underground water (well waters) through accidental spillage or pesticides applied to land ecosystems. The amount of pesticides so far discovered in drinking water was found to be high and this has been of great concern health wise. Some of the effects observed is the discoloration of teeth and food poisoning (when the water is used for cooking).
4) Environmental pollution: It could lead to environmental pollution especially those that resist biodegradation (recalcitrants), that are mostly chemically broken down by chemical reactions and volatilization processes.

There are clear differences in their persistence and this varies from a few days to years depending on the chemical structure and properties of the organic compound in the pesticide.

For instance,

1) Organophosphate insecticides may last only a few days in soils.
2) 2, 4-D persist for only 2 – 4 weeks
3) PCBs, DDT, Aldrin, Chlordane, Mercurial fungicides and Chlorinated hydrocarbons persist for 3 – 20 years or longer. DDT and Mercurial fungicides (used for the control of insect pests on agricultural fields) persists longer than expected. DDT (was the most extensively used insecticide) and Mercurial fungicides were banned due to their low biodegradability and persistence as well as toxicity, leading to food contamination and adverse health

hazards. Hence they are banned to use in agriculture and public health departments.

4.0 CONCLUSION

In this unit and from our discussions organophosphate pesticides degrade rapidly compared to all the other pesticides such as the chlorinated hydrocarbons. They are found or observed to degrade within few days to weeks while chlorinated hydrocarbons persist up to 3 – 20 years and even longer.

The persistence of pesticides has resulted in many health, hazards, environmental pollutions and agricultural problems (due to their effects on the beneficial microorganisms). Therefore, care must be taken in the use or application of pesticides as how long a pesticide persists in soil is important in pest management and environmental pollution.

5.0 SUMMARY

From the study of persistence of pesticides in soil, a range of persistence of a number of pesticides is given below:

	Pesticide	Persistence
1)	Chlorinated hydrocarbon insecticides (DDT, chlordame, Aldrin, Dieldrin)	3 – 20 years
2)	PCBs	2 – 10 years
3)	Triazine herbicides (Atrazine, Simazine)	1 – 2 years
4)	Benzoic acid herbicides (Amiben and Dicamba)	2 – 12 months
5)	Urea herbicides (Monuron, Diuron)	2 – 10 months
6)	Vinyl chloride	1 – 5 months
7)	Phenoxy herbicides (2, 4-D, 2,4,5-T)	1 – 5 months
8)	Organophosphate insecticides (malathion, diazinon)	1 – 3 months (1– 12 weeks)
9)	Carbamate insecticides	1 – 2 months (1-8 weeks)
10)	Carbamate herbicides (Barban, (IPC)	2 – 8 weeks

Adapted from

6.0 TUTOR-MARKED ASSIGNMENT

1) Explain what pesticide persistence means.
2) Enumerate and discuss the effects of persistence of pesticides in soil.

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