DEPARTMENT OF PLANT SCIENCE

Central University of Kerala

Ph.D. Plant Science
Syllabus
March, 2017

No.		Course title	Credits
1	Course I BPS 7111	Research Methodology in Biological Sciences	6
2	Course II BPS 7112	Frontiers and Techniques in Plant biology	6
3	Course III	Specific paper related to scholars area of research	6

Programme outcome: Students should be acquainted with the theory and protocols of research methods in biology. They should also learn the latest developments in the research field.

Programme specific outcome: Course III deals with different topics based on students research area. This will enable students to equip with recent developments in their area of research.

BPS 7111: Research Methodology in Biological Sciences Credit: 6

This course teaches fundamental skills for the design and application of research in the biological sciences, including experimental design and data analysis. Through this course the student is introduced to the development of scientific hypotheses and models and covers the design, analysis and interpretation of experiments and sampling programs. Additionally, skills for writing and assessing scientific papers and reports, including the scientific peer review process, oral presentations and critical reviews are covered

Unit 1: Scientific Conduct and Ethics

- Brief Introduction to Ethics, Scientific Conduct and Misconduct.
- Misconduct and Why It Occurs.
- Fabrication and Other Forms of Misconduct Affecting the Truth Claims of Scientific Findings.
- Authorship Issues, The Investigation and Punishment of Scientific Misconduct – few cases will be discussed.

Unit 2: Design and Analysis of Experiments.

- Introduction, Identifying a Scientific Problem, Literature, Methods and Techniques Research Conditions, Data Types, Techniques, Repeatability, Reproducibility and Reliability Validity, Effect Measure and Choice of Statistical Test, Experimental Protocol, Experimental Design: Blocking One-way (RCBD)Two-way (Latin Square)Split Plot Statistical analysis :Probability Theories, Properties of Normal Distributions, Poisson Distribution, Binomial Distribution, One Sample Test - Two Sample Tests/ Chi-Square Test, t-Test,one way ANOVA, two way ANAOVA,ANACOVA, Regression analysis.
- Computer based tools and software's for data analysis, SAS, MAT-LAB

Unit 3: Scientific Communications

- Introduction, Preparing a Scientific Paper and Poster, review, book, Guide to grant applications, grant writing, Preparing and Delivering a scientific presentation

References:

1. *Asking Questions in Biology.***A Guide to Hypothesis Testing, Experimental Design and Presentation in Practical Work and Research Projects.**Gilbert, G., Mcgregor, P. and C. Barnard (2011).4th ed., Pearson Education Ltd.
2. *Experimental Design and Data Analysis for Biologists***.** Quinn, G.P. & M. J. Keough (2003).

Cambridge University Press.

3. *Biometry : The principles and practice of statistics in biological research.* Sokal, R.R. and F. J. Rohlf (1995).3rd ed., W.H. Freeman.
4. *Introductory Statistics for Biology Students*. Watt, T.A. (1993/1997). Chapman & Hall.

BPS 7112: Frontiers and Techniques in Plant Biology Credit: 6

This course provides an intensive overview of topics in plant genetics. It emphasizes recent results from model organisms including Arabidopsis, Rice, Maize and Tomato as well as a variety of other plants and provides an introduction to current methods used in basic and applied plant biology.

UNIT I – Biochemistry of medicinal plants

- Chemistry of medicinal molecules.
- Synthetic pathways, accumulation of the major medicinal molecules.
- Extraction, purification of active ingredients, large scale extraction, characterization and quantification methods.
- Pharmacology of medicinal plants.
- Biotechnological manipulations of medicinal plants.
- Secondary metabolite production in cell cultures- Case studies

UNIT II - Electrophoresis:

- General principles – apparatus, methods and applications
- Moving boundary, electrophoresis and zone electrophoresis, Paper electrophoresis, thin layer column chromatography
- starch gel, agarose gel and polyacrylamide gel electrophoresis
- SDS, Non-SDS, DIS electrophoresis Isoelectric focusing, Istotachophoresis and immuno electrophoresis.
- Separation of nucleic acids, Southern blotting – DNA sequencer, DNA synthesizer.

UNIT III - Centrifugation**:**

- Basic principles of sedimentation
- Types of centrifuges and their uses –Preparative and Analytical centrifuge
- Sedimentation equilibrium method, Sedimentation velocity method
- Density Gradient Centrifugation
- Isokinetic and isopycinic centrifugation
- Differential centrifugation. Ultra Centrifugation

UNIT IV – Plant Genetics and Genomics

- Gene isolation methods
- Construction of gene libraries
- Molecular markers (RAPD, SSR, AFLP, CAPS, SNPsetc) and their analysis
- Case study of SSR markers (linkage map, QTL analysis etc)
- SNP identification and analysis
- Generation and screening of mutants
- Transposon mediated mutagenesis
- High throughput sequencing techniques

UNIT V - Intellectual property Rights

Forms of protection – copy right, trade mark, patent. Plant Breeders rights

References:

1. Plant Biotechnology: The genetic manipulation of plants 2nded. By Adrian Slater, Nigel Scott, Mark Fowler, Oxford university Press, UK**ISBN-10:**0199282617
2. Principles of Plant Genetics and Breeding 2nd Ed. By George Acquaah Wiley- Blackwell Publishers, NJ ISBN: 978-0-470-66476-6
3. Plant Functional Genomics:Methods and ProtocolsEditors: Grotewold, Erich (Ed.) Springer Publishers, ISBN: 978-1-58829-145-5
4. Plant genome editing by novel tools: TALEN and other sequence specific nucleases (2015),ThorbenSprink, JaninaMetje, Frank Hartung, Current opinion in Plant biology,32: pp 47-53 (review article)
5. Patent Law Essentials: A Concise Guide by Alan L. Durham (1999) ISBN-13: 978-1567202427
6. Wadehra, B.L. 2000. Law relating to patents, trademarks, copyright designs and geographical indications.Universal Law Publishing.

BPS 7113: Plant tissue culture and phytochemistry

I. PLANT TISSUE CULTURE

Unit 1. The Botanical basis for plant tissue culture

- Regeneration from tissues- induction of regeneration, shoot and root regeneration.
- Culture methods, media and various additives.
- Factors affecting seed germination.
- Isolation of individual seedlings and its development in to mature plantlets.

Unit 2. Somatic embryogenesis

- Initiation of somatic embryogenesis, growth and maturation in medium.
- Induction of somatic embryos, various stages of somatic embryo development.
- Production of embryogenic suspension cultures – initiation, explant, media, proliferation and selective subculture.
- Somaclonal variation and its application in plant improvement.

Unit 4. Haploid plant production

- Anther culture, importance and limitations, stage of anther to culture, media, plant growth regulators and factors affecting anther culture, androgenic embryos, embryo development, haploid testing.
- Microspore culture, techniques, and its importance over anther culture.
- Gynogenesis, importance of unfertilized ovary, ovule or inflorescence culture, techniques and various steps involved media, conditions and hormones that control gynogenic response. Advantages over anther culture and limitations.
- Doubling of chromosome numbers in haploids and importance of haploids.

Unit 5. Triploid plant production

- History and importance, conventional method of triploid production and its difficulties
- Stage of endosperm at the time of culture, mature and immature endosperm culture, role of embryo, factors affecting endosperm proliferation.
- Shoot regeneration from endosperm, organogenesis and embryogenesis. Ploidy determination, Rooting and field transfer of plantlets.

Unit 6. Protoplast isolation, culture, fusion and regeneration

- Isolation: Enzyme treatment, protoplast purification, visualization of cell walls,

determination of protoplast viability, counting protoplasts

- Culture: Media, Culture procedures – embedding in agarose, liquid over agar or agarose, hanging drop culture
- Protoplast fusion: chemical fusion, electrical fusion, selection of heterokaryons and somatic hybrids, Manual selection, complementation, Flow cytometry, genetic consequences of protoplast fusion, cybridization, characterization of somatic hybrid plants

Unit 7. Germplasm storage

- Short term storage – cold storage, desiccation, modification of gaseous environment
- Long term storage -cryopreservation -choice of plant material and preculture, pregrowth, chemical cryoprotection, vitrification, cryoprotective dehydration, encapsulation and dehydration, freezing and storage, thawing and recovery.

II. PHYTOCHEMISTRY

Unit 8. Plant Biochemistry

8.1 The nature of molecules

- Covalent bond, hydrogen bond, ionic bond, *van der Waals* force of attraction and hydrophobic interaction.
- Water as an ideal biological solvent, Concept of pH and Buffers.

8.2 The chemical building blocks

- Carbohydrates: monosaccharides, oligosaccharides, polysaccharides
- Proteins : Amino acids - structure and classification, Structure of proteins - primary, secondary, tertiary and quaternary , globular and fibrous proteins
- Lipids :Simple lipids- fatty acids; saturated , unsaturated and triacyl glycerols, Compound lipids -phospholipids, glycolipids, and sphingolipids
- Nucleic acids: ribonucleic acid and deoxyribonucleic acid, sugar, nitrogenous bases and phosphate group

8.3 Photosynthesis

- Light reactions and dark reactions

8.4 Respiration

- Glycolysis, TCA cycle and oxidative phosphorylation

8.5 Plant Growth regulators

- Auxin and cell elongation and growth
- Gibberlin and stem elongation
- Cytokinin and cell division

- Abscisic acid and water balance of the plant
- Ethylene and senescence

Unit 9.Phytochemical techniques

- Chromatography – general principle, Coloumn chromatography. Paper chromatography, Thin Layer chromatography, Partition chromatography, Gel permeation chromatography, Affinity chromatography, Gas chromatography, HPLC, Ion exchange chromatography.
- Spectrophotometry – GCMS, U.V., NMR.
- Electrophoresis and autoradiography

References

1. Bhojwani, S.S. and Razdan, M. K (1996) Plant tissue culture: Theory and Practice, a revised edition, Elsevier Science B.V, Amsterdam.
2. Dixion, R.A. and Gonzales, R.A (Ediors) (2004) Plant Cell culture – A practical Approach 2nd edn., Oxford University Press, Newyork.
3. Ravishankar, G.A, and Venkataraman, L.V (1997) Recent advances in biotechnological applications of plant tissue and cell culture, Oxford and IBH Publishing company private Limited, New Delhi.
4. Srivastava, P.S. (Editor) (1998) Plant Tissue culture and Molecular Biology – Applications and Prospects, Narosa Publishing House, New Delhi.
5. Kyte, L., and Kleyn ,J. (1996) Plants from test tubes – An introduction to Micropropagation, 3rd edn., Timber Press, Portland, Oregon.
6. Toonen, M.A.J., and de Vries, S.C. (1996) Initiation of somatic embryos from single cells. In: Wang, T.L., Cuming, A. (Editors) Embryogenesis the generation of a plant. Pp. 173-189, Bios Scientific Publishers Limited, Oxford, UK.
7. Gurumani, N. (2007) Research methodology for Biological Sciences, MJP Publishers, Chennai.
8. Taiz, L. and Zeiger, E. (2002) Plant Physiology, 3rd edn., Sinauer Associates, Sunderland.
9. Heldt, H.W. (2005) Plant Biochemiostry, 3rd edn., Elsevier Academic Press, California, USA.
10. Harbone, J.B. (2005) Phytochemical methods : a guide to modern techniques of plant analysis, 3rd edn., Springer international edition.

Course 3	**BPS 7141: Plant Cell Wall- Biochemistry of the cell wall molecules and principles of wall architecture**
AIM	This course aims to make the learners understand the structure, complexity, characterization and modification of plant cell wall polymers in perspective to biofuel production
Objectives	➢ To understand the structure of important plant cell wall polymers ➢ To know how wall polymers are synthesized in plant cell and its complexity and extraction ➢ To gain knowledge on different biochemical and analytical methods to characterize plant cell wall and how the genetic make-up could be modified for biorefinery applications.
Learning outcome	➢ This course will help the students to understand the importance of plant cell wall polymers in biofuel yield. The knowledge gained would be applicable in designing novel pretreatment methods as well as re-engineering feedstocks for fuel production.
Units	**Theory**
1	Derivation of cell walls and wall architecture- Cellulose, Hemicelluloses, Pectic polysaccharides, mixed linked glucans, proteins and glycoproteins, Lignin, suberin, cutin, waxes.
2	Biosynthesis of cell wall polymers-General mechanism of polymer assembly. Glycosyl transferases and polysaccharide synthases, regulation of polysaccharide synthesis.
3	Wall polymers: Extraction and fractionation- Polysaccharides; Wall phenolics, hydroxycinnamates; Feruloylation and Coumarylation Lignin-carbohydrate complexes
4	Modification/ engineering of plant cell wall for better fuel production: Hemicellulose and Lignin engineering
5	Cell wall profiling**:** by different biochemical and analytical methods- Cellulose and hemicellulose estimation, compositional analysis of hemicellulose by HPLC/HPAEC, estimation of hydroxycinnamates and lignin, polymer analysis by FTIR and NMR, OLIMP, PACE, FE-SEM, Confocal microscopy

Course 4	**BPS 7142: Roadmap for biomass to bioenergy and bioactive components**
AIM	This course aims to make the students understand how plant biomass can be converted to biofuels and value added products.
Objectives	➢ To understand the current International and national status of biofuel production ➢ To understand the structural complexity of cell wall polymers and their conversion to biofuel by efficient pretreatment methods ➢ To discuss on the fermentation parameters for high fuel yield
Learning outcome	➢ This course is having high significance in current scenario of research in the field of alternative energy resources. The learners will gain knowledge on how plant biomass can be completely utilized for biofuel production cost effectively and could be helpful for them to secure career in the biorefinery sector.
Units	**Theory**
1	Bioenergy current status: National and international; Recent advances in second generation biofuel production and its advantages; future perspectives
2	Plant cell walls: Renewable energy resource of biofuel and chemical; Feedstocks- Important bioenergy crops, agri-residues as potential Feedstocks; Value added products from lignocelluloses; Recalcitrance of cell wall
3	Cell wall degradation- Biomass pretreatment-different pretreatment methods- Physical, Chemical, Biological, Recent advances in cost effective pretreatment methods; Microbial source for cell wall degrading enzymes: Cellulolytic, Xylanolytic and Ligninolytic microbes and their identification
4	Saccharification and fermentation: Estimation of the saccharification efficiency of the pretreated biomass; Factors affecting saccharification, Simultaneous saccharification and Fermentation.
5	Environmental and economic aspects: Environmental impacts of biofuel production; Value-added processing of biofuel residues and co-products; Policies and regulations on biofuel production: biofuel polices, underlying drivers and technical standardisation

References:

1. Goldstein WE. 2016. The Science of Ethanol:CRC Press;
2. Fry SC. 2001. The Growing Plant Cell Wall: Chemical and Metabolic Analysis The Blackburn Press
3. Hayashi T. 2006. The Science and Lore of the Plant Cell Wall: Biosynthesis, Structure and Function Brown Walker Press
4. Linskens HF and Jackson, JF. 2011. Plant Cell Wall Analysis. Springer; Softcover reprint of the original 1st ed. 1996 edition

5. Ahluwalia VK 2018. Renewable Energy in India; Impacts and Responses for the Built Environment. Booh Shores, ,second edition
6. Singh RS and Pandey A. 2017. Biofuels Production and future Prospectives. Edgard gnansounou, crc press
7. Shoukat S. 2011. Progress In Biomass and Bioenergy Production:vol 7,IntechOpen
8. Khanna M and Zilberman D. 2017. Handbook of Bioenergy Economics and Policy: Springer
9. Marco Aurelio Dos Santos Bernardes. 2011. Biofuel production:;Recent Developments and Prospects:vol 8,IntechOpen
10. Lima MAP, Policastro Natalense AP. 2012. Bioethanol:Intech,
11. Albersheim P, Darvill A, Roberts K, Sederoff R and Staehelin A. 2010. Plant Cell Walls. Garland Science; 1 edition
12. Li Y and S.K. Khanal SK. 2016. Bioenergy: Principles and Applications. ISBN 9781118568316 (paper) / 9781118568378 (epub). Wiley Blackwell
13. Vairavan K, Thukkaiyannan P, Paramathma M Venkatachalam P, Sampathrajan A. 2007. Biofuel Crops: Cultivation and Management (Jatropha, Sweet Sorghum and Sugarbeet) Published by Agrobios

CORE COURSE II : BPS 7121 – Algal Biotechnology

S. No	Topics
1.	Brief taxonomic descriptions and identification of micro and macro algae of fresh water and marine habitats. General principles of Culturing Algae in Laboratory and growth measurement. Isolation and Culture of Algae of different forms (single cell, colonial, filamentous and thallus forms). Chemical composition of Culture media for fresh water and marine algae (Botryococcus braunii, Chlorella, Dunaliella tertiolecta, Gracilaria, Kappaphycus, Sargassum).
2.	**Algae cultivation:** Photobioreactor: various types for Photobioreactors and their components. Cultivation of micro algae in various types of pond systems. Environmental conditions, Open pond, Closed loop system.
3.	**Algae Biofuel Product and Processes:** Harvesting and Oil Extraction: Principles and methods-Chemical processes, Bio-diesel, Thermochemical processes, Biochemical processes, Bio-ethanol, Bio-butanol, Bio-methanol, Bio-hydrogen.
4.	**Seaweeds farming** – Objectives – Site selection, Installation of test plants, Kinds of test planting, Introduction of test plants. Preparation of the farm site and other culture activities –construction of farm – Line method, Rope & Raft methods, Net method – Floating bamboo method – Mangrove stakes and nets-method. Management – Seed selection and preparation, Tying of seedings, Planting, Harvesting, Pre-harvest activities, Harvesting procedures, Drying. Maintenance of the farm. Marking of seaweeds.
5.	Generalized uses of seaweeds, Human food, Seaweed Baths, Cosmetics, Seaweed as agricultural fertilizers, Liquid Seaweed Extracts, Seaweed industrial gums: Alginates, Agars, Carrageenans, other polysaccharides and their Medicinal Uses. Microalgae for high-value chemicals from algae: β-carotene, astaxanthin, docosahexaenoic acid, eicosahexaenoic acid, phycobilin pigments and algal extracts for cosmetics. Microalgae for cosmaceuticals, nutraceuticals and functional foods. Microalgae in liquid waste managements, Biological waste treatemnt, Algae-bacterial system.

6.	**Practical:** Isolation and Culturing of fresh water and marine Algal forms in Laboratory and growth measurement using suitable medium. Cultivation of microalgae using photobioreactor and in pilot pond systems. Algal biomass harvesting: Algae oil extraction by mechnical and chemical methods (Solvents/soxlet extraction), Pretreatment- Saccharification (Thermochemical processes). Ethanol fermentation.

Suggested Readings:

1. H Stein (1973) Handbook of Phycological methods. Culture methods and growth measurements, Cambridge University Press.
2. Christopher S. Laban and Michael James Wynne (1981) The Biology of seaweeds, University of California Press.
3. David Sieg (2011) Making algae biodiesel at Home.
4. Gavin C. Torn, Jr. (1988) Manual on seaweed culture – FAO Manual.
5. Klaus Lüning (1990) Seaweeds: their environment, biogeography and ecophysiology, Wiley-IEEE
6. Clinton J. Dawes (1998) Marine Botany, 2nd ed, John Wiley & Sons, Inc.
7. Relevant information in the reviews and research articles.

CORE COURSE III : BPS 7123 – Microalgae genetic engineering

UNIT 1. Importance of microalgae:

Brief introduction to ecological, commercial and industrial applications of microalgae.

UNIT 2. *Haematococcus pluvialis*:

A detailed study on habit, habitat, cell structure and life cycle of the microalgae *Haematococcus pluvialis.*

Unit 3. Astaxanthin:

Structure, biosynthetic pathway, genes involved in biosynthesis, commercial uses of Astaxanthin, commercial production value and biomedical potential of Astaxanthin.grid15

Unit 4. Transformation techniques in microalgae:

Brief notes on

1. Agrobacterium mediated transformation
2. Particle bombardment

3. Electroporation
4. Viruses
5. Agitation with glass beads
6. Biolistic delivery
7. Agitation with silicon carbide whiskers
8. Cell wall-deficient strains

Unit 5. Selection of transgenic microalgae:

1. Selection of nuclear transformants based on auxotrophic growth
2. Selection of nuclear transgenic algae based on herbicide resistance

Unit 6. Gene promoters in microalgae.

1. Cauliflower Mosaic Virus (CaMV35S)
2. RBCS2
3. HSP70A

References

1. A.V.S.S. Sambamurty, (2005) Text book of algae, I K International pvt. Ltd.
2. Kathiresan S, Chandrashekar A, Ravishankar A, Sarada R. Agrobacterium-mediated transformation in the green alga Haematococcus pluvialis (Chlorophyceae volvocales). J Phycol 2009;45:642–9.
3. Diaz-Santos,E., delaVega,M., Vila,M., Vigara,J. ,and Leon,R. (2013). Efficiency of different heterologous promoters in the unicellular microalga Chlamydomonas reinhardtii. Biotechnol.Prog. 29,319–328.
4. Ying-Fang Niu, Tan Huang, Wei-Dong Yang, Jie-Sheng Liu and Hong-Ye Li (2016) Genetic Engineering of Microalgae. OMICS Group e book.

5. Astaxanthin: A Review of the Literature *The scientific evidence on the uses, contraindications, and interactions of this carotenoid :* Natural Medicine Journal.

Core III: BPS 7115 **Algae metabolites and Drug discovery**	
Objectives	This course is designed to teach, • , extraction, separation, identification and bio-evaluation of phytochemicals.
Learning outcomes	Upon successful completion of this course, students are ➢ Able to know how to cultivate the algae ➢ have the knowledge on the extraction, isolation, purification and characterization of bioactive compounds of commercial importance. ➢ will have the competence to initiate start-ups or job opportunity in phytochemical and pharmaceutical industries.
Skill components	Practical's are the skill components of the course.
S.No	
1.	**Algae resources and cultures : Microalgae & macroalgae.**
2.	**Methods of Extraction:** Solvent Extraction methods-Maceration,Decoction, Reflux extraction, Soxhlet extraction, ultrasonic and microwave-assisted extraction. **Methods of Separation, Isolation and concentration:** Separation by solvent method-polarity gradient separation; precipitation methods, salting out, dialysis**;** Separation byChromatography Ion-exchange, gel-filtration, HPLC and HPTLC; Concentration by evaporation- Lyophilization and flash evaporation.
3.	**Identification of phytochemicals by chromatographic techniques:** Phenolic Compounds, Terpenoids, Organic Acids, Lipids and Related Compounds, Nitrogen Compounds, Sugars and their Derivatives, Macromolecules like nucleic acids; Proteins; Polysaccharides by HPLC and HPTLC.
4.	**Biosynthesis and characterization of nanoparticles:** Silver nanoparticle synthesis using plant extracts like polysaccharides, Phenolic Compounds and Terpenoids.

5.	**Identification and characterization of phytochemicals by various analytical and spectroscopic methods:** UV/Visible, Fluorescent, FTIR and XRD and FE-SEM.
6.	**Bio-evaluation:** Anti-oxidants, Anti-viral, Anti-bacterial, anti-fungal and anti-cancer by cell line assay.
7.	**Practicals:** • Introducing basic protocols in cell culture-Bacteria, Fungi, Algae and Callus. • Analysis of monosaccharide components in poly/oligosaccharides by HPLC • Soxhlet extraction of plant metabolites • Extraction and partial purification of crude enzyme samples • Gel filtration chromatography for separation of oligosaccharides • Concentration of the plant/algal extracts by lyophilisation and flash evaporation. • Silver nanoparticle synthesis using plant/algal polysaccharides • Identification of phenolic compounds by HPTLC • Characterization of plant/algal polysaccharides by FTIR • Isolation and characterization of plant/algal polysaccharides using XRD • Analyzing antioxidant/anticancer/antiviral properties of plant/algal polysaccharides

References:

1. Arunkumar,K., Rathinam Raja, V. B. Sameer Kumar, Ashna Joseph, T. Shilpa and Isabel S. Carvalho. 2020. Antioxidant and cytotoxic activities of sulfatedpolysaccharides from five different edible seaweeds, *Journal of Food Measurement and Characterization*,51.
2. Hahn-Deinstrop E. Applied Thin Layer Chromatography:Best practice and avoidance of Mistakes. Wiley-VCH, Weinheim, Germany. 2000.
3. Hancock WS. High Performance Liquid Chromatography in Biotechnology. Wiley-Interscience, New Jersey, USA. 1990.
4. Harborne JB. Phytochemical Methods: A guide to modern techniques of plant analysis. 2nd Edition. Chapman and Hall publishers: 3, Springer. Germany.1998
5. Jim Clark (Chemguide.co.uk); Introducing Chromatography: Thin Layer Chromatography; Jun 6, 2019
6. Katz ED. High Performance Liquid Chromatography:Journal of Pharmacognosy and Phytochemistry Principle and Methods in Biotechnology (Separation science Series). John wiley& sons, New Jersey, USA.1995
7. Mark F. Vitha Spectroscopy: Principles and Instrumentation ISBN: 978-1-119-43664-5
8. Roseline, T.A., Murugan, M., Sudhakar, M.P., Arunkumar, K.2019. Nanopesticidal potential of silver nanocomposites synthesized from the aqueous extracts of red seaweeds, *Environmental Technology and*

Innovation, 13 , pp. 82-93.