5th SEM B.Sc. ZOOLOGY
CALICUT UNIVERSITY

METHODOLOGY IN SCIENCE, BIOSTATISTICS AND BIOINFORMATICS

CODE: ZOL5B09T

2019 - ADMISSION

Prepared by
Jamshiya Jaithun C
Assistant professor
Department of Zoology

CPA College of Global of Studies, Puthanathani

FIFTH SEMESTER B.Sc. ZOOLOGY PROGRAMME
ZOOLOGY CORE COURSE- VIII [Theory]
METHODOLOGY IN SCIENCE, BIOSTATISTICS AND BIOINFORMATICS
Code: ZOL5B09T
[54 hours] [3 hours per week] [4 Credits]

COURSE OUTCOMES (COs)

COs Course Outcome Statements

1. CO1 Explain science, its importance, disciplines and the major steps in formulating a hypothesis, various hypothesis models, theory, law and importance of animal models, simulations and virtual testing (6 hrs)
2. CO2 Illustrate the principles and procedures in designing experiments and elaborate the requirements for carrying out experiments (4 hrs)
3. CO3 Describe the ethical concerns in practicing science (5 hrs)
4. CO4 Understand the Scope and role of statistics; methods and procedures of sampling; Construction of tables, charts and graphs (5 hrs)
5. CO5 Calculate central tendency and measures of dispersion and application of its knowledge on hypothesis testing as well as in problem solving (10 hrs)
6. CO6 Enumerate major biological databases and database search engines (8 hrs)
7. CO7 Perform DNA and protein sequence analysis, including sequence alignment and sequence similarity search using BLAST, FASTA, CLUSTAL W and CLUSTAL X (4 hrs)
8. CO8 Understand molecular phylogenetics and tools and methods for construction of phylogenetic trees (3 hrs)
9. CO9 Explain genome sequencing technologies, functional genomics, proteomic technologies and molecular docking and drug design (9 hrs)

Question paper pattern for external examination
[Module 1-3: Short answer 5x2=10 marks, Paragraph 2x5=10 marks, Essay 1x10= 10 marks
Module 4-5: Short answer 5x2=10 marks, Paragraph 2x5=10 marks, Essay 1x10=10 marks;
Module 6-9: Short answer 5x2=10 marks, Paragraph 4x5=20 marks, Essay 2x10 = 20 marks]

Section A: METHODOLOGY IN SCIENCE (15 hrs)

MODULE 1. Science, Scientific Studies and Methods (6 hrs)

Science and Scientific Studies ;Science as a human activity; scientific attitude; Empiricism; Science disciplines; Interdisciplinary approach. Scientific Methods ;Major steps: Observation, Defining the problem, Collection of information, Formulation of a hypothesis, Experimentation, Analysis of the results and Conclusion based on interpretation of the results.

Methods in scientific enquiry: Inductive and deductive reasoning.

Hypothesis: Formulation of a hypothesis, different thought processes in developing hypothesis (analogy, induction, deduction and intuition), hypothetico-deductive model, testing hypothesis, auxiliary hypothesis, adhoc hypothesis.

Theories and laws in science; peer review; importance of models, simulations and virtual testing (brief account).

MODULE 2. Experimentation (4 hrs)

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Types of experiments; design of an experiment: principles and procedures; necessity of units and dimensions; repeatability and replications; documentation of experiments; Planning of Experiments: design, selection of controls, observational and instrumental requirements; Test animals used in experiments.

MODULE 3. Ethics in Science and Animal Ethics (5 hrs)
Scientific information: Depositories of scientific information – primary, secondary and digital sources; Sharing of knowledge: transparency and honesty, Publications, Patents, Plagiarism.
Constitution of India Article 51A (g); Prevention of cruelty to animals Act of 1960 - Section 17.1(d), Committee for the purpose of control and supervision of experiments on animals (CPCSEA).

Section B: BIOSTATISTICS (15 Hrs)
MODULE 4. Introduction (5 hrs)
Definition; scope; role of statistics in life sciences; terminology and variables. Sample and Sampling: Sample size, sampling errors, methods of sampling.
Collection/documentation of data of the experiments. Classification of data; Presentation of data: Tabular, Graphical and Diagrammatic (histogram, frequency polygon and frequency curve; line diagram, bar diagram and pie diagram).

MODULE 5. Analysis and Interpretation of data (10 hrs)
Measures of central tendency: (raw data, discrete series data, continuous series data- problems are to be discussed)

a) Mean, b) Median and c) Mode.

Measures of Dispersion: (raw data, discrete series data, continuous series data – problems to be discussed)
a) Range, b) Mean deviation, c) Standard deviation, d) Standard error.

Hypothesis testing and Interpretation of results: (problems to be discussed)
a)'t' test, b) F- test - ANOVA
Significance of statistical tools in data interpretation; Statistics-based acceptance or rejection of hypothesis.

SECTION C: BIOINFORMATICS (24 hours)
MODULE 6. Introduction and Biological Databases (8 hrs)
Overview of bioinformatics, Scope and application of Bioinformatics.Major Databases in Bioinformatics: Biological databases, Features of a good database. Classification format of biological databases.
Primary databases: Nucleotide sequence databases – Mention EMBL, DDBJ,
Genbank; Protein sequence databases – Mention Swiss Prot, PIR, MIPS.
Structure databases: PDB, NDB.
Special databases – PROSITE, Pfam, CATH, OWL, PubMed.
Secondary databases: Mention PROSITE, PRINTS. Databases of patterns, motifs and profiles, EST databases, SNP databases.
Metabolite databases – Mention KEGG, EcoCyc.
Database Search Engines: Entrez at NCBI of USA, SRS at EBI of England, STAG at DDBJ of Japan. Data retrieval with Entrez and SRS.
Sequence submission to NCBI.

MODULE 7. Sequence Analysis (4 hrs)

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Web based and standalone tools for DNA and protein sequence analysis. Types of sequence alignment, methods of sequence alignment. Sequence similarity search – pair wise and multiple sequence alignments; BLAST, FASTA, CLUSTAL W,CLUSTAL X.

MODULE 8. Molecular Phylogenetics (3 hrs)

Basics of Phylogenetics; molecular evolution and molecular phylogenetics, cladistics and ontology. Gene Phylogeny versus species phylogeny. Phylogenetic tree construction methods and programmes. Forms of Tree representation.

MODULE 9. Genomics and Proteomics (9 hrs)

Genome sequencing technologies; Sanger capillary sequencing, Roche 454 (pyrosequencing), Illumina/Solexa, SOLiD System, Single molecule sequencing. Whole genome sequence assembly, annotation and analysis. Functional Genomics:Microarrays, SAGE, ESTs; Transcriptomics; Metabolomics. Metagenomics: Concept and applications.

Proteomics : Aims, strategies and challenges in proteomics. Brief account on proteomics technologies: 2D-electrophoresis, iso-electric focusing, LC/MS-MS, MALDI-TOF mass spectrometry, yeast 2-hybrid system. Protein-protein interactions: experimental and computational methods; structural proteomics. Deriving function from sequence databases.

Cheminformatics

Molecular docking and drug design; Structural Bioinformatics in drug discovery

MODULE 1

Science, Scientific Studies and Methods

SCIENCE

- Derived from *scientia* : meaning knowledge
- Knowledge gained through observation , experimentation , interpretation & generalization
- Anything observable in the universe
- Empirical
- Measurable
- Purpose:
  - Provides explanations for natural observations
  - Makes generalizations to predict relations of natural phenomena
- Systematic procedure : *scientific method*
- Pseudoscience: alternative science/fringe science : methodology/belief claimed to be legitimately scientific , though does not have standards of scientific status : eg: telepathy
- Junk science : scientific hypothesis which may be legitimate in themselves but are used to support an illegitimate claim
- **Major steps :**
  1. Observations
  2. Asking questions
  3. Formulation of hypothesis
  4. Testing of hypothesis
  5. Collection of data
  6. Interpretation of data
  7. Drawing conclusions
  8. Revision of hypothesis
  9. Formulation of theory / law

SCIENCE AS A HUMAN ACTIVITY

- Built-in-self-correction mechanism
- Involves collection of data , observable facts and convincing evidences , recording and analysis of data , drawing conclusions and inferences and formulation of theoretical explanations
- Ongoing human activity
- Collective and organized human activity
- Motivating forces : curiosity , desire , imagination , insight , skill , creativity ,traits and attributes
- Social perspective

SCIENTIFIC ATTITUDE/ TEMPER

- Rational & ideological outlook of mankind on issues and problems
- Free from bias , prejudices and pre-conceived notions
- Involves rational thinking , intellectual reasoning ,weighing of evidences
- Cardinal aspects:observation , analysis, verification , argument etc.
- Scientific attributes:
  1. Mental activism
  2. Curiosity

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3. Open-mindedness
4. Absence of dogmatism
5. Intellectual honesty
6. Rational thinking
7. Critical thinking
8. imaginative thinking
9. Selflessness
10. Objectivity
11. Humility
12. Aversion to superstitions
13. Systematic approach

EMPIRICISM

- Philosophical doctrine related to pursuit of knowledge through observation and experimentation
- Cardinal aspects: knowledge is founded on experience ; sense experience is the only source of knowledge ; knowledge can be acquired only through experience
- Sensory perception
- Fundamental part of scientific methods
- Science : methodologically empirical branch of knowledge
- Contradicts rationalism

SCIENCE DISCIPLINES

- 2 broad categories:
  1. Basic : facts and principles
     Natural science : physics
     Social science : psychology
  2. Applied : application : eg;animal husbandry
  3. Formal science : mathematics

Interdisciplinary approach

- Different disciplines of science are never mutually exclusive , but are interdepenedent and interrelated
- Multidisciplinary
- Biology=life science

METHODS IN SCIENTIFIC ENQUIRY: INDUCTIVE AND DEDUCTIVE REASONING

1. **Inductive**
2. **Deductive**
3. **Constructive** : variant of deductive method ; involves deductive construction of scientific theories in maths and logic
4. **Hypothetico-deductive** : certain generalizations are advanced as hypothesis and then subjected to verification by deduction and comparison with established facts

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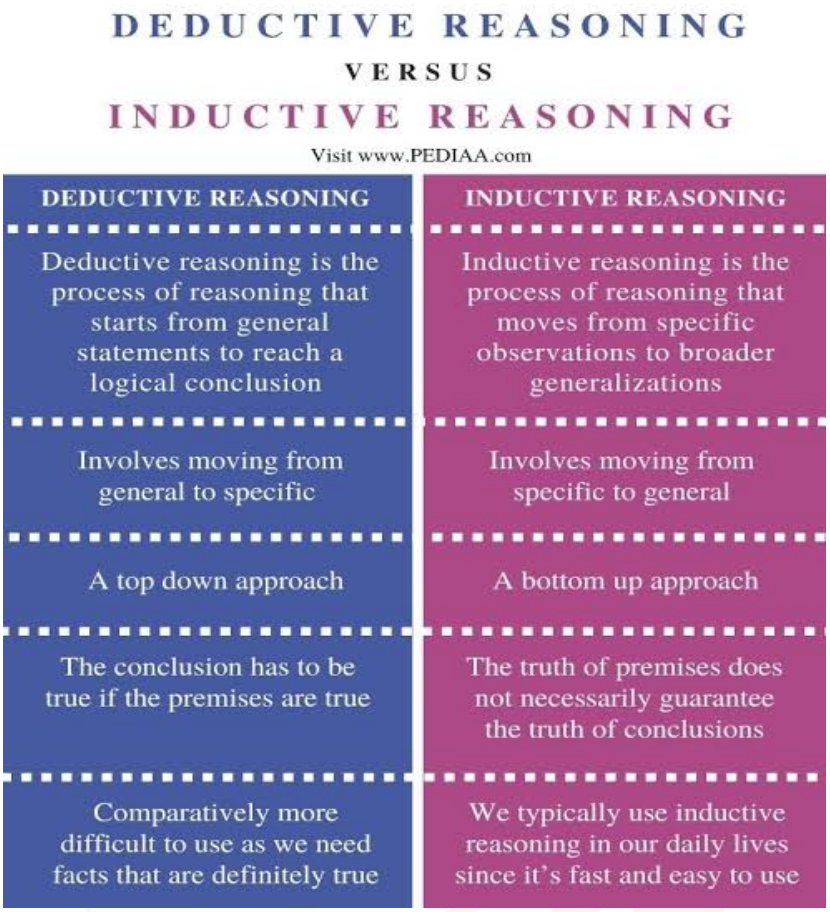

HYPOTHESIS

- Testable tentative based on previous knowledge & forms basis of reasoning
- Deduction based on a series of facts
- Assumptions made on evidence
- Initial point of investigation
- Offers adequate explanation of related facts
- Channels scientific inquiries in right direction
- Helps in selection of appropriate methods for testing
- Helps in formulation of laws,facts
- Helps in drawing conclusions

DIFFERENT THOUGHT PROCESSES IN DEVELOPING HYPOTHESIS

1. **Analogy** : similarity;previously established knowledge contributes to the formulation of a hypothesis
2. **Induction** : originates from observations of a specific case but reflects general pattern
3. **Deduction**: based on established knowledge
4. **Intuition :** ability for directly understanding the truth , without conscious reasoning/ study

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TYPES OF HYPOTHESIS

1. **Auxiliary hypothesis :** supplementary hypothesis used in scientific testing & verification ; considered to be true and necessary to support test implication
2. **Adhoc hypothesis :** auxiliary hypothesis , used solely for defending a particular test hypothesis whose validity/ acceptability is questioned by negative evidences

Theories and laws in science

- **Theories** :Organized set of reasoned ideas,intended to explain facts , phenomena & events
- Represent crux of one or more hypothesis
- **Law** : universally applicable / accepted theory with capabilities for making true predictions

Evidence & proof

- **Evidence:** facts & figures used to support , oppose , substantiate or invalidate a hypothesis, theory / law
- **Scientific proof :**logical deduction / process of reasoning applied to establish the truth of a proposition or an assertion ; based on accepted assumptions and proved inferences

Peer review

- Expert scrutiny of the results of a scientific investigation or ideas , hypothesis and theories formulated by an investigatory project
- Requires team of scholars
- Encourages to maintain required standard
- Helps in avoiding unwanted claims, irrelevant findings , factual errors etc

Models

- Representation of a real thing and serves as a very useful tool in scientific investigations
- Accurate / inaccurate representations of real thing
- Used to evolve theories when direct observation & study of actual phenomena is difficult
- Eg; hardware tools, software tools , mathematical models ; eg;atom model
- Eg; *E.coli*

Simulations

- Imitation of real thing, state of affairs or process
- Useful when testing in real condition is impossible
- Eg;study of origin of life
- Computer simulation in medical research

Virtual testing

- "not real"**;**But conceptual , possessing the salient features of the real
- Virtual reality technique : testing of hypothesis using computer methods

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MODULE 2.

EXPERIMENTATION

- Scientific experiments = to try out
- Method of investigating the casual relationships among variables
- Objectives :
  1. Commonly used for testing validity of hypothesis
  2. For supporting / negating theoretical assumptions
  3. For solving practical problems
  4. For gathering data about natural phenomena etc
- The experimental method involves the manipulation of variables to establish cause and effect relationships. The key features are controlled methods and the random allocation of participants into controlled and experimental groups.
- An experiment is an investigation in which a hypothesis is scientifically tested. In an experiment, an **independent variable** (the cause) is manipulated and the **dependent variable** (the effect) is measured; any extraneous variables are controlled.

TYPES OF EXPERIMENTS

1. Controlled Experiment

- A controlled experiment is simply an experiment in which all factors are held constant except for one: the independent variable.
- A common type of controlled experiment compares a control group against an experimental group. All variables are identical between the two groups except for the factor being tested.
- The advantage of a controlled experiment is that it is easier to eliminate uncertainty about the significance of the results**.**

2. Lab Experiment

- A laboratory experiment is an experiment conducted under highly controlled conditions (not necessarily a laboratory), where accurate measurements are possible.
- The researcher decides where the experiment will take place, at what time, with which participants, in what circumstances and using a standardized procedure.
- Participants are randomly allocated to each independent variable group.
- **Strength**:
  - It is easier to replicate (i.e. copy) a laboratory experiment. This is because a standardized procedure is used.
  - They allow for precise control of extraneous and independent variables. This allows a cause and effect relationship to be established.
- **Limitation**:
  - The artificiality of the setting may produce unnatural behavior that does not reflect real life, i.e. low ecological validity. This means it would not be possible to generalize the findings to a real life setting.
  - Limitation: Demand characteristics or experimenter effects may bias the results and become confounding variables.

3. Field Experiment

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- Field experiments are done in the everyday (i.e. real life) environment of the participants. The experimenter still manipulates the independent variable, but in a real-life setting (so cannot really control extraneous variables).
- **Strength:**
  - Behavior in a field experiment is more likely to reflect real life because of its natural setting, i.e. higher ecological validity than a lab experiment.
  - There is less likelihood of demand characteristics affecting the results, as participants may not know they are being studied. This occurs when the study is covert.
- **Limitation:**
- There is less control over extraneous variables that might bias the results. This makes it difficult for another researcher to replicate the study in exactly the same way.

4. Natural Experiment

- Natural experiments are conducted in the everyday (i.e. real life) environment of the participants, but here the experimenter has no control over the independent variable as it occurs naturally in real life.
- **Strength:**
  - Behavior in a natural experiment is more likely to reflect real life because of its natural setting, i.e. Very high ecological validity.
  - There is less likelihood of demand characteristics affecting the results, as participants may not know they are being studied.
  - Can be used in situations in which it would be ethically unacceptable to manipulate the independent variable, e.g. Researching stress.
- **Limitation**:
  - They may be more expensive and time consuming than lab experiments.
  - There is no control over extraneous variables that might bias the results. This makes it difficult for another researcher to replicate the study in exactly the same way.

5. Observational studies

- Similar to controlled experiments except that they lack equivalency between groups
- Common in medical field where ethical issues exists
- Results are much less convincing

DESIGN OF AN EXPERIMENT

- Represent design of all information – gathering and information –analysing exercises
- Includes ;
  1. Planning of various methods within a time-frame
  2. Fixing definite time period
  3. Planning definite experimental activities
  4. Selection of appropriate analytical methods etc

PRINCIPLES OF EXPERIMENTAL DESIGN

1. Comparison
2. Randomization
3. Replication
4. Blocking
5. Orthogonality

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6. Multi-factorial experiments
7. Confounding

PROCEDURE OF EXPERIMENTAL DESIGN

1. Selection of problem
2. Determination of variables to be studied
3. Determining the number of levels of independent variables
4. Determining the possible combinations
5. Determining the n umber of observations
6. Redesign
7. Considerations of ethical and legal requirements
8. Application of mathematical principles and models
9. Data collection
10. Data verification
11. Interpretation and deduction

NECESSITY OF UNITS AND DIMENSIONS

- Inevitable for generating and comparing quantitative data
- Measured physical quantities : dimensions
- Example : length, area ,temperature
- Dimension = value = unit
- SI Unit : international system of units
- 2 types : base (eg; length) & derived (eg;area)

DOCUMENTATION OF EXPERIMENTS

- Can be in the form of log book / notebook
- ***Scientific notebook*** : thorough record of the daily work of a research project ; permanent
- Science notebook : contains all details about experimental set up , data collected , explanation of the analysis procedures used & results
- Functions served by scientific notebook:
    1. Provides a complete coverage of the work for preparing scientific paper
    2. Helps others replicate/ repeat
    3. Furnishes valid evidences & supports in case of patent dispute
    4. Provides solid base for evaluation of the work if its result contradicts
- ***Scientific paper*** : detailed report of whole work in a standard format and published / presented so as to reach and shared by whole human society(IMRAD)

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MODULE 3. Ethics in Science and Animal Ethics

Depositories of scientific information – primary, secondary and digital sources

1. **Primary Source**

- Original materials that have not been filtered through interpretation or evaluation by a second party.
- Timing of publication cycle : Primary sources tend to come first in the publication cycle.
- Conference papers, dissertations, interviews, laboratory notebooks, patents, a study reported in a journal article, a survey reported in a journal article, and technical reports.
- Example : Article in scholarly journal reporting research and methodology.
- Eg; Ph.D. thesis

2. **Secondary Source**

- Sources that contain commentary on or a discussion about a primary source
- Secondary sources tend to come second in the publication cycle.
- Review articles, magazine articles, and books
- Example : articles analyzing and commenting on the results of original research; books doing the same"
- Eg; review papers, reference books , manuals

3. **Tertiary source**

- Consists of pri and sec source information which has been collected and distilled
- Present summaries of or an introduction to current state of research
- Example :year books, directories

4. **Digital sources**

- Scientific information in digital format
- Either in form of CD ROMS & as online sources
- Example : INFLIBNET

Sharing of knowledge: transparency and honesty

- Exchange of various kinds of knowledge among and between the different sections of people , friends , community / organizations
- Supported by knowledge management systems

Publications

- Process of placing on record the results of experimental investigations as part of scientific literature
- Scientific literature : includes publications which report the original empirical and theoretical work
- Primary: results of original work initially published in scientific journals
- Secondary literature: edited books , review journals
- Tertiary literature : encyclopedia
- **format of content :** introduction , materials & methods , observations & results , discussion & conclusion , references

Patent

- set of exclusive rights granted by a state to an inventor or to his assignee for a limited period of time in exchange for a public disclosure of an invention
- derived from *patere : to lay open*
- example : biopatents,machine patents

Plagiarism

- Criminal act by which a person willfully uses & represents the ideas , inventions , postulations , works etc of others as his own original contribution without their inform / permission

Constitution of India Article 51A (g)

- States that "*it should be the duty of every citizen of citizen of India to protect and improve the natural environment , including forests, lakes , rivers & wildlife and to have compassion for living creatures "*
- Provision for the management , protection & conservation of environment

Prevention of cruelty to animals Act of 1960 - Section 17.1(d)

- Envisages that all forms of cruelty to animals , including willful infliction of unnecessary pain & sufferings are punishable offences
- Also states that to ensure well being and protection of animals and to refrain from willfully inflicting unnecessary sufferings and misery on them , shall be mandatory for every person who is in charge of their care & protection

Committee for the purpose of control and supervision of experiments on animals (CPCSEA)

- Was instituted in 1964 under provisions of the Prevention of Cruelty to Animals Act 1960
- Main function : to see that laboratory animals are well maintained and the experiments conducted on them follow internationally prescribed ethical norms and regulations
- Make guidelines and rules for conducting experiments on animals

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