BOT 443 - Ecological Methods

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Course Syllabus

- Instructor : **Dr. Abdullah Ahmed AlGhmadi**
- **What it the course about?**
- This course covers initial concepts of ecological methods including : design and execution botanical experiments simulating ecological factors, recording, analysis, and processing ecological experimental data using computer models.

Practical Schedule:

- Week 1 : Introduction to Ecological Methods
- Week 2 : Experimental Design and Experiment Preparation
- Week 3 : Standard Solution Preparation
- Week 4 : Climatic Factor Instrumentation and Measurement
- Week 5 : Chlorophyll Analysis and Plant Sampling
- Week 6 : Digestion and Element Extraction
- Week 7 : Phosphorus analysis using spectrophotometer
- Week 8 : Calcium Analysis using Flame Photometer
- Week 9 : Sodium and Manganese Analysis using Atomic Absorption Spectrophotometer
- Week 10 : Mid Term Exam
- Week 11 : Iron and Magnesium Analysis using Atomic Absorption Spectrophotometer
- Week 12 : Statistical analysis of the collected data using SPSS, Excel, and XLStat
- Week 13 : Preparation of Final Report
- Week 14 : Submission of Final Report
- Week 15 : Final Exam

Evaluation:

- Weekly Practical Notes/Report 10 points
- Attendance 10 points
- Homework 10 points
- Individual Written Final Report 20 points
- Mid Term Exam 10 points
- Final Exam 40 points

Week 1 – Introduction to Ecological Methods

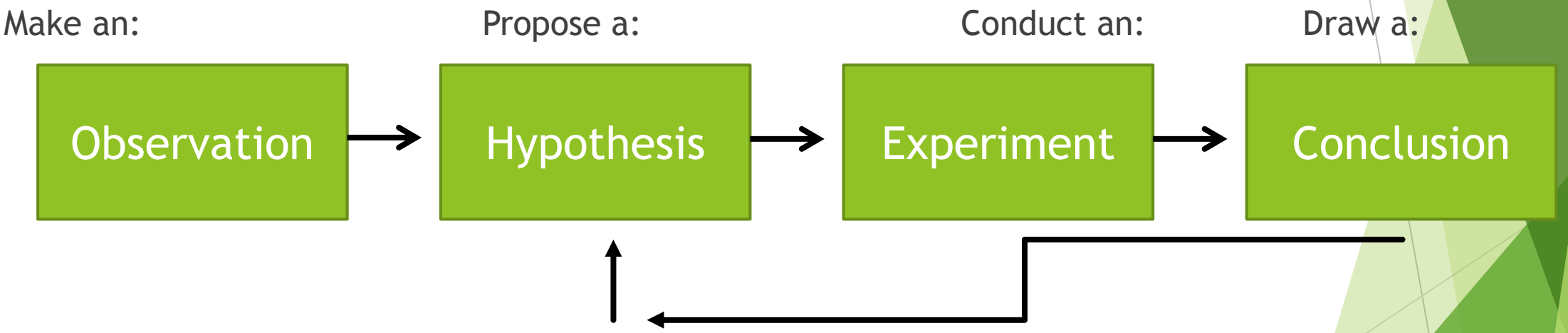
- Ecology is the scientific study of interactions among organisms and between organisms and their physical environment.
- Method is a particular procedure for accomplishing or approaching something, especially a systematic or established one.
- Ecological method is a particular procedure to study the interaction of organisms with other organism and their environment.

- Because ecologists work with living systems possessing numerous variables, the scientific techniques used by physicists, chemists, mathematicians, and engineers require modification for use in ecology. Moreover, the techniques are not as easily applied in ecology, nor are the results as precise as those obtained in other sciences.

- Controlled environmental chambers enable experimenters to maintain plants under known conditions of light, temperature, humidity, and day length so that the effects of each variable (or combination of variables) on the organism can be studied.
- The use of laboratory microcosms—aquatic and soil micro-ecosystems, consisting of biotic and nonbiotic material from natural ecosystems, held under conditions similar to those found in the field—are useful in determining rates of nutrient cycling, ecosystem development, and other functional aspects of ecosystems. Microcosms enable the ecologist to duplicate experiments and to perform experimental manipulation on them.

Scientific Method

- a set of techniques to ask and answer scientific questions



Week 2 – Experimental Design

Methods of Designing the Ecological Experiment

- Design experiments to mimic Ecological condition

Experimental design is planning to simplify the information and analysis for a good analysis and practical to reach the correct scientific conclusion.

There are some important point to be recognize to selecting experimental design:

1. Number of factor (temperature, humidity and soil texture, etc) in the experiment.

If is only one factor experiment, It called Simple experiment.

If it is more than one factor in the same time, It called Factorial experiment.

2. Type of experimental units (plants varieties) which you are going to apply your factorial study, is it homogenous or should be classified to overcome cause of different.

Simple Experiments

- The importance methods used for designing ecological experiments
- Simple experiments:
- In this experiment we need to make distribution to different levels.
- Examples: salinity with different concentration
- 1. Completely Randomized design.
- This design used when the experimental units are highly similar, and the distribution of treatment/factor using complete random distribution for the experimental unit as followed.
- Supposed that we have 5 concentration treatments. Can be coded as T1 (Control/0 % salt), T2 (5% Salt), T3 (10% salt), T4 (15%Salt), and T5 (20% Salt). For each treatments there are four replicates so the experimental design for this treatment can be distributes as followed

Completely randomized design

T3	T4	T1	T5	T5
T1	T2	T2	T4	T3
T3	T4	T3	T1	T5
T2	T1	T5	T2	T4

Randomized blocks design

Variety A	T3	T4	T2	T1
Variety B	T2	T1	T4	T3
Variety C	T1	T4	T3	T2
Variety D	T3	T1	T2	T4
Variety E	T4	T3	T2	T1

Latin Square Design

T3	T1	T2	T4
T4	T3	T1	T2
T1	T2	T4	T3
T2	T4	T3	T1

Factorial Design

Factor A (temperature)	Factor B (salinity)	Treatment Combination	Treatment Code
A1 (20 C)	B1 (0%)	A1 B1	T1
	B2 (5%)	A1 B2	T2
	B3 (10%)	A1 B3	T3
	B4 (15%)	A1 B4	T4
A2 (30 C)	B1 (0%)	A2 B1	T5
	B2 (5%)	A2 B2	T6
	B3 (10%)	A2 B3	T7
	B4 (15%)	A2 B4	T8

Plan an experiment

- Effect of ….. On the growth of …..

Week 2

- Methods Used for preparation of solution
- Prepare :
- Hoagland Solution
- Salt Solution

Week 3

- Preparing Standard Solution for element analysis
- Phosphorus analysis solutions for spectrophotometer
- Calcium for Flame photometer
- Sodium, Manganese, Zinc, and Magnesium for Atomic Absorption Spectrophotometer

Week 4

- Measurement of climatic Factor
- Weather Station and data logger

Week 5

- Harvesting and Sampling of experiment plant
- Note all plant morphological data
- Chlorophyll analysis

Sample Collection

- **Sample Collection**
- Samples are collected from designated area with stainless steel tools and placed in clean glass jars or acetone-washed aluminum foil. Jar lids are lined with acetone-washed aluminum foil. Jars are cleaned with acetone and dried. Samples are chilled/freezed with crushed ice during transfer from field to analytical laboratory. GPS locations and photographs of the sampling places are collected. (NEIC, 1985)
- Subsampling of local use pesticides are collected from the local field and markets. Glass pipets and acetone cleaned Widemouth glass jars are used to collect and store the pesticides samples.
- ***Water Sampling***
- Clean sampling glass jars are dipped to collect 1 liter of surface water. Replicates are collected for quality control analysis. Water samples are kept on ice and protected from direct sunlight. (NEIC, 1985)
- ***Soil Sampling***
- 500 grams of surface soil and under the root soil are collected with stainless steel scoop and stored in clean glass jar with Aluminum foil lined lid. Replicates are collected for quality control analysis. Samples are kept refrigerated in the analytical lab for further analysis. (NEIC, 1985)
- ***Sediment Sampling***
- Disposable core tubes are used to collect 500 grams of sediment and stored in clean glass jar with Aluminum foil lined lid. Replicates are collected for quality control analysis. Samples are chilled by crushed ice during transport to laboratory for further analysis. (NEIC, 1985)
- ***Plants Sampling***
- 500 grams of affected vegetation (water plants and surrounded land plants) are collected, clipper and small shovel are used to help collecting the plants leaves, shoot and roots. Unaffected plants are collected as clean control. Samples are stored in clean aluminum foil and chilled during transport to analytical lab. Plant samples are frozen with liquid nitrogen and stored in the freezer. (NEIC, 1985)

Chlorophyll

- The chlorophyll (Chl) and carotenoid (Car) contents of leaves were measured by the method described earlier (Lichtenthaler and Welburn 1983; Porra 2002). The pigments of 0.1 g of fresh leaf were extracted by acetone 80% in mortar and pestle. Extracts were filtered by filter paper and the absorbance of samples was measured at 663, 646 and 470 nm by UV-visible spectrophotometer (Spectronic 21 – Milton Roy Company). Chlorophyll a, b, carotenoid and total chlorophyll contents were calculated with an equation.
- $Chla = 12.21A663 - 2.81A646$
- $Chlb = 20.13A646 - 5.03A663$
- $Car = \frac{1000A470 - 3.27Chla - 104Chlb}{229}$
- $Total\ Chl = 17.76A646 + 7.34\ A663$

Week 6

- Digestion of sample materials

Sample digestion for extraction

- **Ca, K, Mg, Mn, and Na content**

- The samples are digested according to standard digestion procedures for plant materials (Price 1974). 200 mg of finely ground seedling samples were placed in 100 ml TECAM digestion flask in which 0.5 ml of sulfuric acid, 1 ml of perchloric acid and 5 ml of nitric acid were added. These flasks were heated at 110ºC and temperature was slowly increased to 330ºC. After it cool, the solution was transferred to 50 ml calibrated flask and made up to volume with double distilled water. Standard elements were prepared and Ca, K, Mg, Mn, and Na content were measured by Perkin Elmer AAS-300 Atomic Absorption Spectrophotometer. (Katz and Jennis 1983).

Week 7

- Phosphorus analysis using spectrofotometer

Week 8

- Mid Exam

Week 9

- Calcium Analysis using Flame Photometer

Week 10

- Analysis of Sodium, and Manganese using AAS

Week 11

- Second Mid Exam

Week 12

- Analysis of Iron and Magnesium using AAS

Week 13

- Statistical analysis of the collected data

Week 14

- Submit the Final Report
- Final Exam