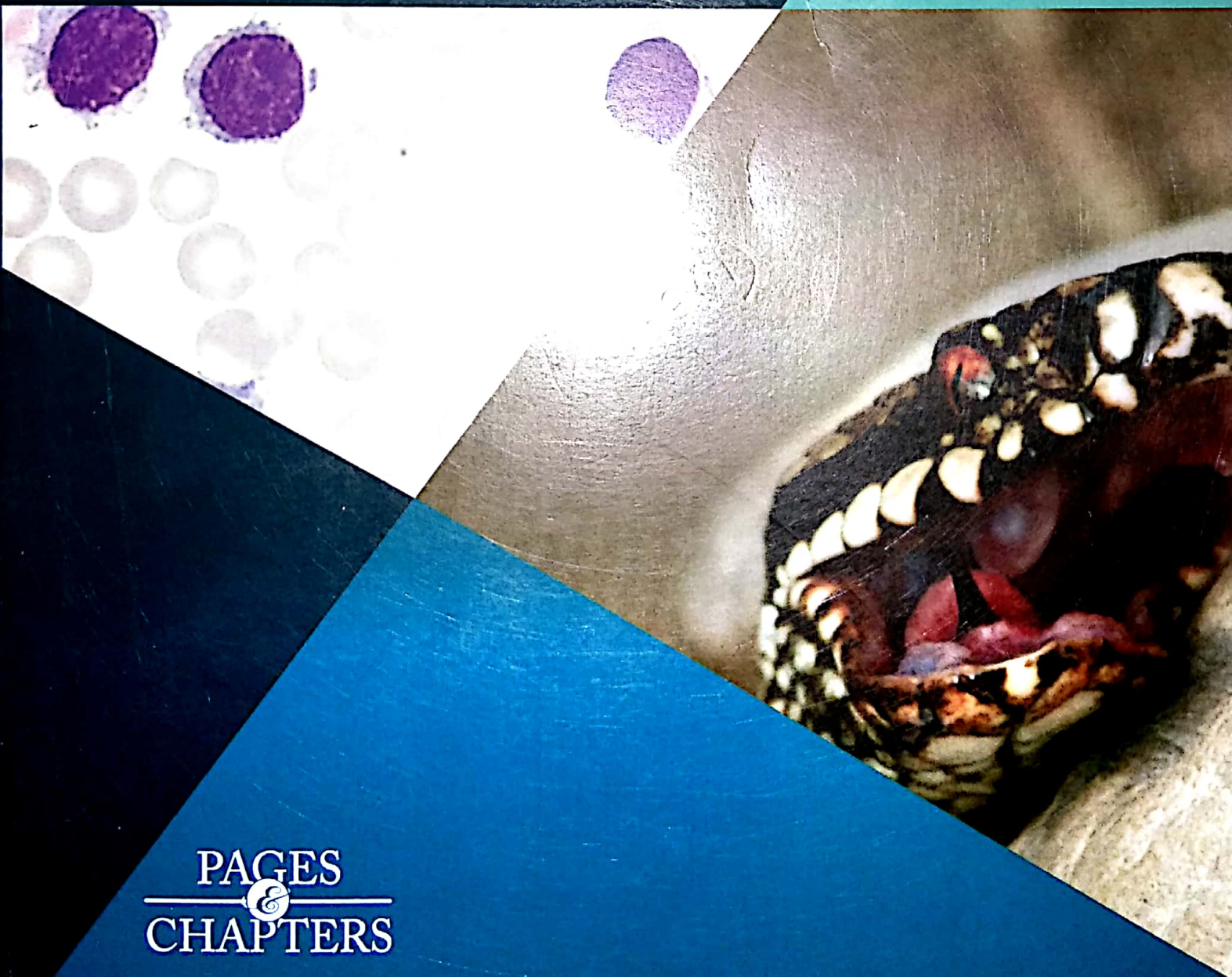


CONTEMPORARY LABORATORY AND FIELD EXPERIMENTS IN ZOOLOGY

DEPARTMENT OF ZOOLOGY

ACHARYA PRAFULLA CHANDRA ROY GOVERNMENT COLLEGE
SILIGURI

PAGES
&
CHAPTERS



CONTEMPORARY LABORATORY AND FIELD EXPERIMENTS IN ZOOLOGY

**PROCEEDINGS OF A HIGHER EDUCATION DEPARTMENT,
GOVERNMENT OF WEST BENGAL SPONSORED
NATIONAL LEVEL WORKSHOP**

HELD AT

**THE DEPARTMENT OF ZOOLOGY,
ACHARYA PRAFULLA CHANDRA ROY GOVERNMENT COLLEGE,
SILIGURI, DARJEELING DISTRICT, WEST BENGAL**

FROM

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Content

Contemporary Approach for the Courses in Zoological Science(<i>Key Note Address</i>)	7
Determination of LC_{50} or LD_{50} For A Particular Toxicant	10
NIMAI CH. SAHA	
Contemporary Taxonomy : Concept & Approach	14
D. RAYCHAUDHURI	
Arthropod Pest Diversity and Estimation of their Populations in Tea Plantations	21
ANANDA MUKHOPADHYAY	
Methodology in Field Biology	23
S K MUKHOPADHYAY, U SINGHA ROY & S ADHIKARI	
Separation of Lymphocytes From Different Mammalian Tissues	36
TAPAS KUMAR CHAUDHURI	
Specimen Preparation for Transmission Electron Microscope Observation.....	42
T C NAG	
Histochemical Localization of Glycogen by Periodic Acid Schiff (PAS) Staining Method (McManus 1946).....	45
T C NAG	
Identification and Staining of Barr Body	47
DR. SUBIR C. DASGUPTA	
Protocol for Collection, Preservation and Identification of Rumen Ciliates From Indian Goat (<i>Capra hircus</i> LINN.).....	50
DR. RUPENDU RAY	
How to make a People's Biodiversity Register	60
SILANJAN BHATTACHARYA	
Nesting and Mate Choice Decisions of Baya Weaverbirds (<i>Ploceus Philippinus</i>): An Example of Sexual Selection of Behavioural Traits.....	62
N GHORAI	
Single Nucleotide Polymorphism as Molecular Marker; A Bio-Informatic Approach	67
AMLAN GHOSH	
Bioinformatics As A Discipline – Perceptions and Promises	75
SAYAK GANGULI	
Study of Amphibian Sperm under Light & Electron Microscope.....	86
CHANDRANATH CHATTERJEE	
Protocol for Venom Collection from Non-Front-Fanged Caenophidian Snakes	89
SAIKAT SARKAR	
Measurement of Copper in Biological System	94
SURAJIT MAJUMDER	
<i>In Vivo</i> Study of The Effect of Genotoxicity Through Chromosome Aberrations of Bone Marrow Cells and Testicular Cells as Well as Micronuclei from Mouse Treated With Carbon Tetra Chloride (CCl_4)	96
SUJIT KR. BHOWAL	
Estimation of Cholesterol by $FeCl_3$ reagent	103
BIPLAB KUMAR BEHERA	

Methodology in Field Biology

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What is Field Biology ?

Modern biology may be divided into two categories, viz., Organismic Biology and Cell Biology. While Field Biology operates at the level of organism, population, community, ecosystem or landscape. Nature is the main laboratory of the field biologists.

How to plan a field study?

1. What is the specific question?
2. What results are necessary to answer the questions?
3. What data are needed to complete these results?
4. What protocol is required to obtain these data?
5. Can the data be collected in the time available?
6. Modify the planning in response to time available
7. Create data sheets
8. Start and encounter reality

Important Field Trip Activities

1. Density & distribution of different animals in their habitat
2. Behaviour of animals in their habitat
3. Activity patterns at the community level
4. Habitat preferences and niche space
5. Dynamic nature of the physico-chemical & biological world
6. Use of different tools and techniques for wildlife census/sampling

ACTIVITY-I

Biodiversity Estimation : Quadrat Analysis

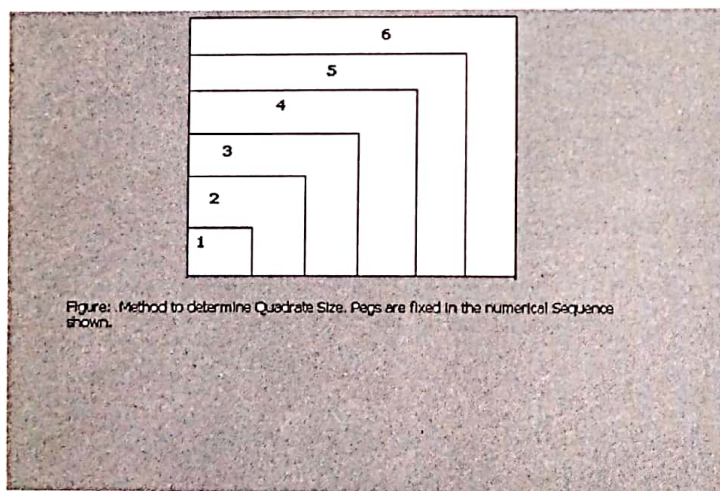
Census vs Sampling :

- Counting of animals or plants in whole of the population, if possible, such complete count is referred as true Census.
- However, in most cases such complete count is impossible and we study only part of the population through samples.
- For reliability of the estimate, one must ensure 1. suitable shape & size of sampling unit and sampling frame 2. samples are random 3. samples are representative 4. sufficient replication & 5. avoidance of bias

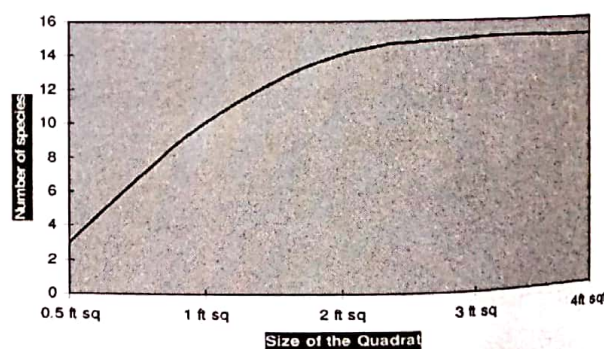
Quadrat Sampling

Determination of Quadrat size :—

The quadrat method is used to sample plants and sedentary animals in a community. The size of the quadrat must be of such dimension that all the species, which occur in that community, are fully presented. The optimum quadrat size is reached when 1% increase in quadrat size produce no more than 0.5% increase in the number of species present. A species area curve can be plotted to describe the importance of the determination of effective quadrat size (EQS).



Graph showing the determination of Effective Quadrat Size (3 ft sq is the effective quadrat size, which was later suitably converted to sq m area)



Calculations

$$\text{Densities (m}^2\text{)} = \frac{\text{Total number of individuals of the species}}{\text{Total number of Quadrat study} \times \text{Area of each quadrat}}$$

$$\text{Abundance (m}^2\text{)} = \frac{\text{Total number of individuals of the species}}{\text{Total number of Quadrat in which the species has occurred}}$$

$$\text{Frequency \%} = \frac{\text{Total number of Quadrat in which the species occurred}}{\text{Total number of Quadrat studied}} \times 100$$

Pit-fall Trap : Used to sample ground surface active organisms

Light Trap : Used to sample flying insects

Diversity Indices

INDEX OF DOMINANCE : $C = \sum (n_i/N)^2$

RICHNESS INDEX: $D = (S - 1)/\ln N$

SHANON-WEINER INDEX: $H' = -\sum (n_i/N) \ln (n_i/N)$

EVENESS INDEX: $E = H' / \ln S$

SIMILARITY INDEX: $s = 2C/(A+B)$

Niche Breadth (Niche Width or Niche Size)

Levins's (1968) Measures of Niche Breadth: $B = 1/\sum P_j^2$

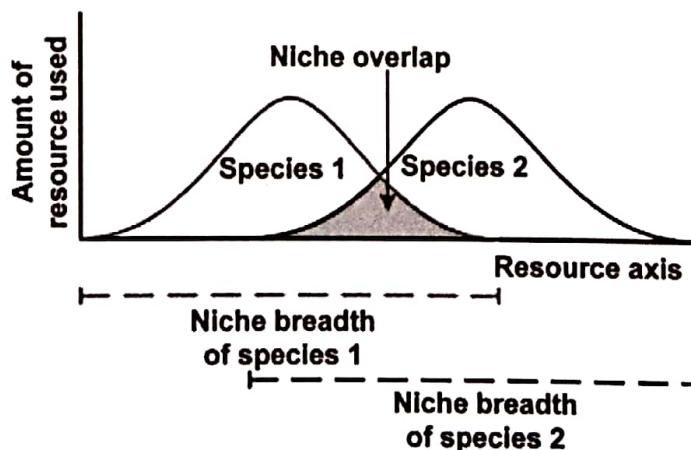
Where, P_j = number of individuals found in or using resource state "j"

It is often useful to standardize niche breadth to express it on a scale from 0.0–1.0. This can be done following measures for standardized niche breadth as suggested by Hurlbert (1978): $B_A = (B-1)/(n-1)$;

Where, B_A = Levins's standardized niche breadth;

B = Levins's measure of niche breadth;

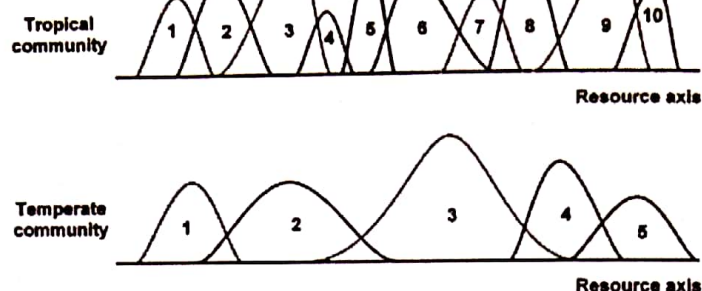
n = number of possible resource states



Niche Overlap

MacArthur and Levins's measure (1967): $M_{jk} = \sum P_{ij} P_{ik} / \sum P_{ij}^2$ where, M_{jk} = MacArthur and Levins's niche overlap of species "k" on species "j" and P_{ij} = proportion that resource "i" is of the total resources used by species "j" and P_{ik} = proportion that resource "i" is of the total resources used by species "k"; n = total number of resource states

(c) Much niche overlap example



ACTIVITY-II

Distance Methods: A Plotless Spatial Mapping

Plotless sampling, applied to a single species of plant or animals, and is effective in estimating the density alongside the spatial distribution pattern. In these methods distance is measured using a range finder by two general approaches :

- Select random points and measure the distance from the point to the nearest organism
- Select random organisms and measure the distance to their nearest neighbours

Calculations :

For point-to-organism distance: $D'_1 = n / \pi \Sigma (x_i^2)$

Where,

D'_1 = Population density for point-to-organism data;

n = Sample size;

x_i = Distance from random point i to nearest organism.

For organism-to-nearest neighbour distances :

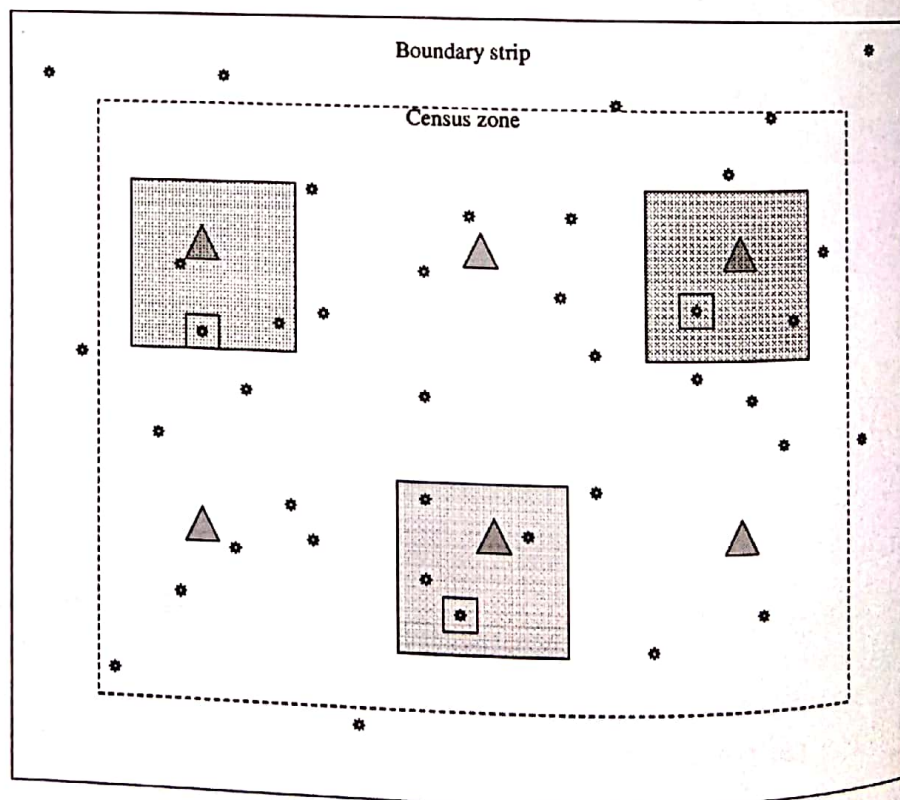
$D' = n / \pi \Sigma (r_i^2)$

Where,

D' = Population density for organism-to-neighbour data;

n = Sample size;

r_i = Distance from random organism i to nearest neighbour.



Semi-systematic sampling scheme recommended by Blyth and Ripley (1980) for distance sampling of large areas in which complete enumeration is not possible. In the figure the desired sample size is chosen to be 3 (n), so 6 (2n) points triangles are laid out in a systematic manner within the study area (boundary in dashed line). Three points are randomly selected for point-to-organism measurements, and the three point remaining shaded areas are used for setting out small plots for a random selection of individuals for nearest-neighbour distances. The three trees selected at random for nearest-neighbour measurements are shown as boxed squares.

ACTIVITY-III

Density Estimation Using Line Transects Method

Transects, are long thin sample units converted by a single traversed usually down the centre of the transect. In vehicle or trek counts, the road forms a convenient centre line and the counting transect is a strip of land on either side of the road as in the figure below.

Sample counts give the density data ("D"), often referred to as Estimate of Density () is calculated as number per unit area :

$$D' = \text{Number of animal seen} / 2 * \text{Transect width} * \text{Transect length}$$

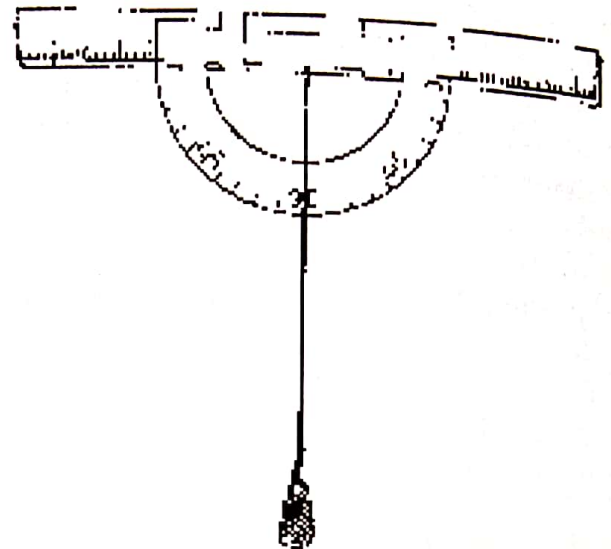
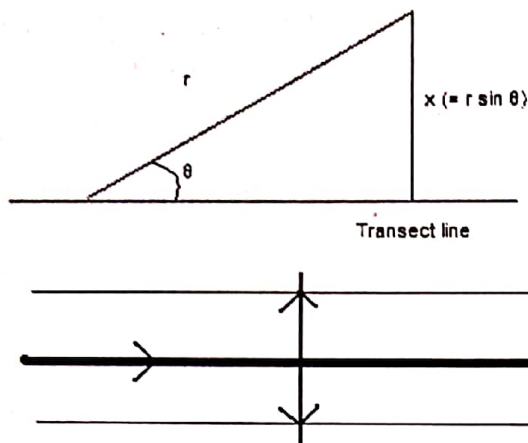
Procedure

- A transect line of the fixed length is to be demarcated. The transect time has to be a straight line.
- The line should be placed randomly for the animal to be counted.
- The perpendicular distance to each detected object of transect from the transect line (x) is calculated.
- A number of lines arranged as regular grid are taken into consideration for the final estimation.
- The sighting distance "r" is recorded from the observer.
- Sighting angler "θ" which is angle of the object from transect line is recorded for the calculation of x.

Material used

100 meters of transect lines marked at each 10m distance can be demarcated by using a 'Casio' digital Pedometer. A 'range finder' records sighting distance. The sighting angle is measured by using a simple device where a protractor tied with a thick thread along the 90° marking line is used to measure the angle of slopes.

Relationship between sighting distance r ,
Sighting angle θ , & perpendicular distance x ,
From the transect line

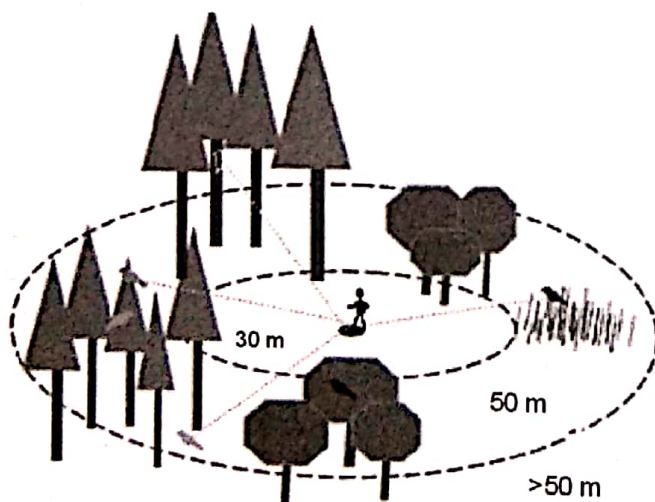


ACTIVITY-IV

Density Estimation By Point Transect Method

A fixed radius circular-plot Point Transect method is ideal to estimate the density of actively flying avian species at the selected habitat patches. At each point three kinds of data within a specified time period (10 min) may be recorded :

- the number of individuals of each species detected within a 30-m radius surrounding the observer
- the abundance of individuals of each species detected beyond the 30-m radius but still within the habitats of interest
- the identity of individuals carefully detected while the observer walked between count points to have a complete species list for the concerned habitat patch within the study duration.



Birds that are originally detected outside the 30-m radius boundary but that later moved

$$\text{Density} = [(n_1 + n_2) / (pr^2m)] \ln[(n_1 + n_2) / n_2]$$

Where,

n_1 = number within the circle,

n_2 = number outside the circle

r = radius of the circular point,

m = number of replicates of counts

to within 30 m of the observer may be considered as occurring within the fixed-radius circle. As it was assumed that all birds within 30 m are detected, the census would facilitate comparisons among vegetatively different habitats. Birds are to be identified on the spot. All birds (or flocks) seen (perched or flying under the canopy) or calls heard should be recorded.

ACTIVITY-V

Relative Density of Birds Estimated From Calls

Many bird species, especially the males, use calls to indicate territory ownership or to advertise for a mate. Production of calls varies seasonally, being most frequent at the start of the breeding season, and varies diurnally. If you tour in the spring, the breeding season of a variety of vocal/song birds, you are surely to note vocalization in the study locations. Night birds f calls may be noted between 19.00 and 22.00 hrs and day birds f calls between 6.00 and 8.00 and between 16.00 and 18.00 hrs. Five individual recordings may be averaged. Recorded numbers are standardized per hour basis and from the call frequencies the relative densities of the avian species are calculated to have an idea of the variety and the relative density of bird species active at the study areas.

Call of a few Forest Nocturnal Birds :

Collard Scops Owl	Mellow interrogative wut; chattering ack, ack, ack ...
Oriental Scops owl	uk kook-krook
Brown Hawk Owl	hoowup...hoowup...hoowup...hoowup
Jungle Owlet	KAO KAO KAO kao-kuk (kao-kuk kaokuk kaokukkaokukkaokuk)
Spotted Owlet	Harsh screeches and chuckles
Tawny Fish Owl	Cat-like mewling; whoo-hoo
Brown Fish Owl	Very low bwoum; boou
Eurasian Eagle Owl	Deep resonant wu-hoo
Grey Nightjar	Engine-like chuck-chuck-chuck...at 3-4/sec
Indian Nightjar	chuck chuck (chuck) chk-r-r-rrrrr
Savanna Nightjar	sweesh or chweez

ACTIVITY-VI

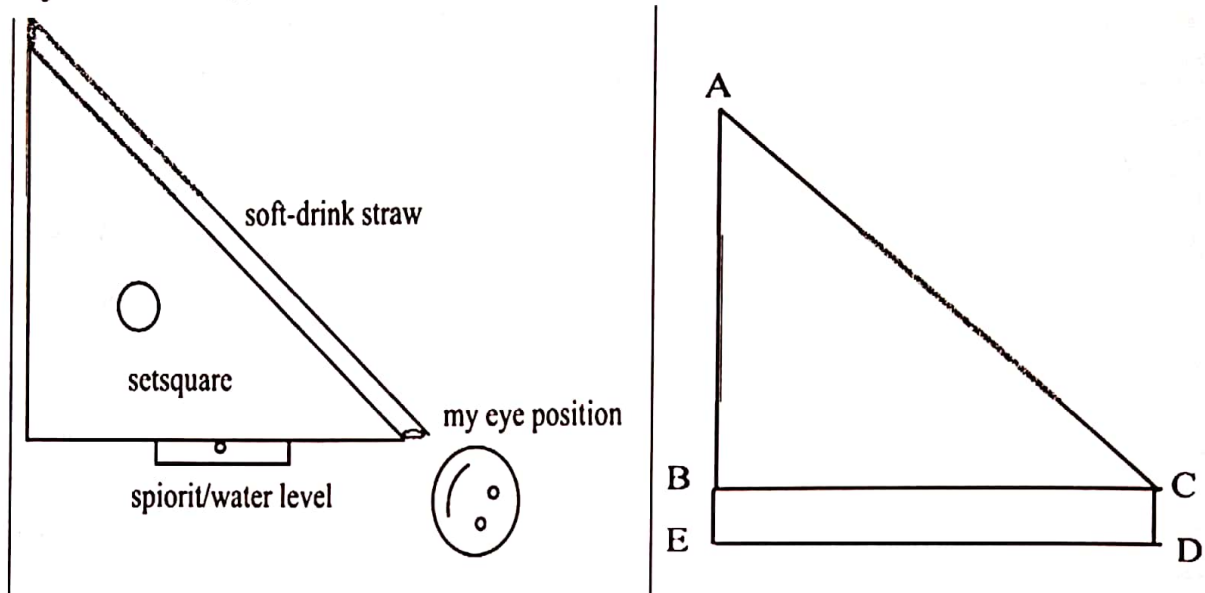
Estimation of Water Birds Using Binocular

To estimate waterbird populations, we can conduct random hanci frame and binocular-frame counts (Gopal 1995) of the birds in selected distance-ranges, 25m, 50m, 100m and 150m. Areas of both hand-frame and binocular-frame should be worked out and standardized by the average of three measurements. To work out the area coverage in binocular/hand frame, the ground cover on land at the pre-set distances should be worked out using the same binocular of the hand frame of the observer. Several random counts covering the water surface may be averaged at different time intervals to get the representative data of a particular month.

ACTIVITY-VII

Determination of Log Volume of Forest Trees

Make your own Hypsometer



If AB is the height of the tree & CD is the height of the observer, Then, $CD = BE$; as $\angle ACB = 45^\circ$ & $\tan 45^\circ = 1$, therefore, $AB/BC = 1$. As $DE = BC =$ distance of observer from the tree, then, $AB = BC$. So, $AB + BE = BC + CD$, Or, $AE = BC + CD = DE + CD =$ length of the tree (L)

Determination of Log Volume

In India volume of logs are usually calculated by Quarter Girth Formula (also known as Hoppus Rule). In the field we applied this technique in the field:

$$\text{Volume of the log} = (g/4)^2 \times L$$

When 'g' is the Girth of the log at chest height and L is the height of the tree or log.
Volume (V) = $\pi r^2 L$ and girth (g) = $2\pi r$

$$\text{Then, } r = g/2\pi$$

$$V = \pi (g/2\pi)^2 \times L$$

$$V = (g^2/4\pi) \times L$$

π is actually equal to 3.14, but for easy calculation it has been roughly taken to be equal to 4

$$\text{Therefore, } V = (g^2/4 \times 4) \times L = (g/4)^2 \times L$$

ACTIVITY-VIII

Wildlife Density As Estimated By Indirect Evidences Like Spoor/Pug Marks, Scratch Marks, Scat Counts Etc

Indirect evidences of Animal activities are a reliable indicator of animal presence and are used for estimating abundance of wildlife. Currently two indirect count methods are popular to estimate relative woodland wildlife activities:

- the '*clearance plot*' method, which is based on the number of pellet groups deposited within an area that was initially cleared of all pellets;
- and the '*standing crop*' method, in which all pellet groups within some previously defined area are counted.
- An alternative approach, is the use of '*belt transects methods*' using a pedometer to mark each evidence in up and down journeys along a defined path length to avoid double counting. Recorded numbers for such activities may be represented per square kilometer basis or as relative density (%).

A Field Guide to Animal Signs (Jayson, E. A. and Esa, P. S. *A Field Guide to Animal Signs*, KFRI Handbook No.4, KFRI, Peechi, Kerala, 1997) is very helpful. Confirmation of identification should be done with the assistance of forest watcher and later confirmed by the forest officials.

ACTIVITY-IX

Dynamic Nature of Ecosystem: Limnological Studies***Estimation of Physico-Chemical Factors***

Recording Physical & Chemical Factors of Water Samples:

Record air and water temperature (digital thermometer), pH, IDS (digital multifactor testing meter), illuminance (digital illuminometer) and chemical factors such DO, Free CO₂, Acidity, Alkalinity, Hardness, Phosphate, Nitrate, Silicate etc using Emerck (Germany) Merckoquant and Aquamerck field-testing kits. Observations may be made at 06 hrs and 18 hrs at sampling site. Therefore, changes in two sets of data for physico-chemical factors over a period of 12 hours may be compared to comment on dynamic nature of water

Estimation of Zooplankton Density: Qualitative sampling:

Pond littoral surface water is strained by plankton net (No. 25 silk bolting cloth) in such a way that the margin of the plankton net moves just below the water surface. The net should towed through the surface water for 1 minute in a uniform towing speed. The towing time may be increased or decreased on the basis density of Zooplankton. The net should also be towed through the aquatic macrophytes.

Quantitative sampling

For quantitative estimation Zooplankton density samples are collected by straining a known volume of pond littoral water (usually 20 liters) through a plankton net. Extreme care must be taken to avoid stirring of the bottom sediment.

Fixation & Preservation of Zooplankton

For both Qualitative & Quantitative Zooplankton samples are usually narcotized and fixed with 5 drops of 2% Formalin in 25 ml zooplankton concentrate however, after a period of 1 hour 10 more drops are to be added to fix the larger zooplankters like cladocera, copepod, arthropod. Formal Alcohol (5% Formalin 25%, Glycerin 5% & 70% Alcohol 70%) may be used as a preservative. Samples must be preserved in known volume (say 25 ml/50 ml) of preservative.

Determination of Lindeman's Efficiency

Lindeman's Efficiency or trophic level energy intake efficiency is the ratio of energy flow at the first trophic level. Steps are as follows :

Determine Gross Primary Productivity using light-dark bottle technique of Gaardar and Gran (1927) with not more than three hours of incubation period. GPP for a span of 12 hours while total respiration (R) for 24 hours $L - 1 = NPP$; $1 - B = R$; $NPP \text{ h}^{-1} \times 12 + R \text{ h}^{-1} \times 24 = GPP \text{ in mg O}_2 \text{ L}^{-1} \text{ d}^{-1}$; therefore :

$[GPP \text{ in mg O}_2 \text{ L}^{-1} \text{ d}^{-1} \times \text{Average depth in m} \times 3.5]/PQ 1.125 = GPP \text{ expressed as Kcal m}^{-2} \text{ d}^{-1}$; note that $1 \text{ mg O}_2 = 3.34 \text{ cal}$; *vide* Timothy R, Parsons M, Takahashi B & Margrave B. Biological Oceanographic Processes, Pergamon Press, 1984

Measure incident radiant energy using a illuminometer during incubation period for at least three times and average lux value should be converted to energy value; note that $1 \text{ lux} = 4104 \times 10^{-5} \text{ Kcal m}^{-2} \text{ d}^{-1}$; *vide* Wetzel R G, Limnology : River & lake Ecosystems, Academic Press, 2001

Lindeman's Efficiency or trophic level energy intake efficiency = P_G/L , where, P_G = gross photosynthetic energy fixation by producers and L = energy in incident light. Generally P_G/L is 1 - 5% (Odum, 1971) or 1 -2% (Tailing & Lamoalle, 1998).

ACTIVITY-X

Behaviour Patterns & Construction of Ethogram

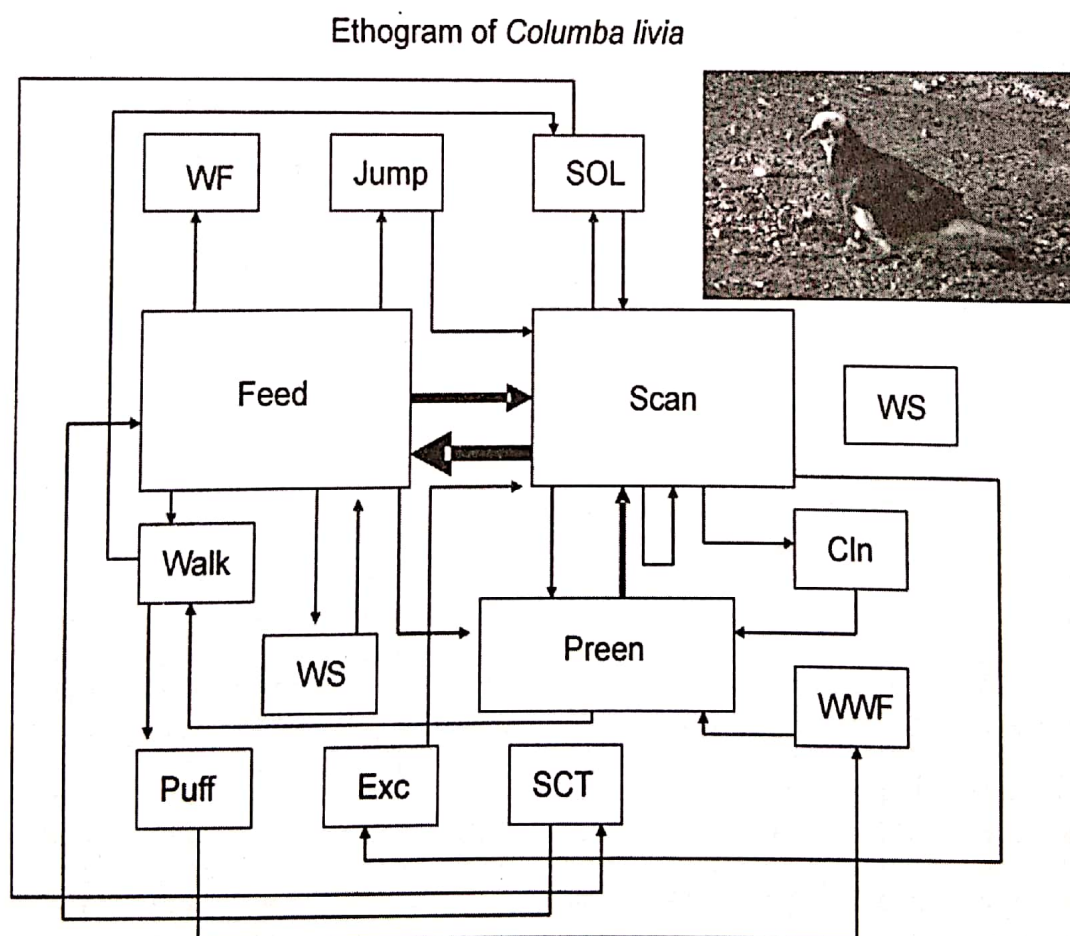
Sampling Methods

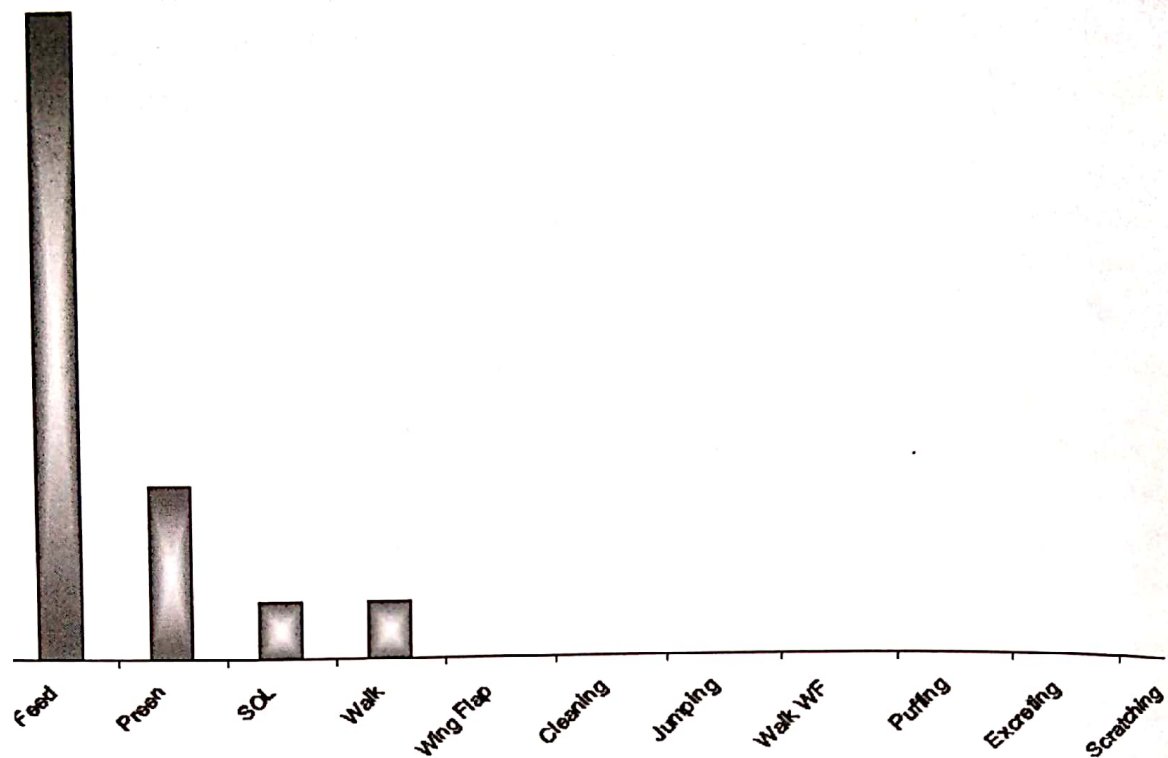
A variety of sampling methods can be used to obtain a a valid picture of the behavior in question. In most instances, behaviors can either be classified as short events or states lasting an appreciable time:

1. *Ad Libitum* Sampling : records as much information as possible at liberty. It is informal, non-systematic, and often used in field notes
2. Focal Animal Sampling: occurrences of specified actions of one individual are recorded during a predetermined sample period (e.g., one hour) recording the amount of time the focal animal is in view ("time in"). This method can provide unbiased data relevant to a wide variety of questions, particularly if animals remain in the field of view.
3. All Occurrence Sampling: The observer focuses on a particular behavior rather than a particular individual. This is a useful method for providing the rate of occurrence of a behavior (# occurrences per unit time) or for studying the synchrony of behaviors within a group.
4. Scan Sampling: An animal's activities are recorded at pre-selected moments (e.g., every 30 seconds). If the behaviors of all members of a group are surveyed within a short period of time, we call it scan (or instantaneous) sampling.

Ethogram Construction:

In its simplest form, an ethogram is a quantitative description of an animal's normal behavior. Constructing a useful ethogram demands time spent, serious watching, careful noting, and making sense of the observed behaviours. Leading to an annotated





Frequency of different activities observed in pigeon during the study

catalogue of behavioral patterns grouped in a coherent fashion that describes what a given species does in a given environment.

To practice construct an ethogram of *Columba livid* or any animal of your choice by noting all occurrence pattern for fixed durations (say for 10 minutes).

In common rock pigeon, altogether 13 different behaviour may be noted and are abbreviated as follows:

Matrix	Feed	Wing Flap	Scan Scan	Preen	Clean	Jump	SOL	Walk	Walk WF	Puff	Excrete	Scratch	Walk
Feed	0.00	2.56	20.51	0.00	0.00	0.00	0.0 0	0.00	0.00	0.00	0.00	2.56	2.56
Wing Flap	2.56	0.00	0.00	0.00	0.00	0.00	0.0 0	0.00	0.00	0.00	0.00	0.00	0.00
Scan	12.82	0.00	2.56	7.69	0.00	2.56	2.5 6	0.00	0.00	0.00	2.56	0.00	0.00
Preen	2.56	0.00	2.56	0.00	2.56	0.00	0.0 0	0.00	2.56	0.00	0.00	0.00	0.00
Clean	0.00	0.00	2.56	0.00	0.00	0.00	0.0 0	0.00	0.00	0.00	0.00	0.00	0.00
Jump	2.56	0.00	0.00	0.00	0.00	0.00	0.0 0	0.00	0.00	0.00	0.00	0.00	0.00
SOL	0.00	0.00	2.56	0.00	0.00	0.00	0.0 0	2.56	0.00	0.00	0.00	0.00	0.00
Walk	2.56	0.00	0.00	2.56	0.00	0.00	0.0 0	0.00	0.00	0.00	0.00	0.00	0.00
Walk WF	0.00	0.00	0.00	0.00	0.00	0.00	0.0 0	0.00	0.00	2.56	0.00	0.00	0.00
Puffing	0.00	0.00	0.00	0.00	0.00	0.00	0.0 0	2.56	0.00	0.00	0.00	0.00	0.00
Excrete	0.00	0.00	2.56	0.00	0.00	0.00	0.0 0	0.00	0.00	0.00	0.00	0.00	0.00
Scratching	0.00	0.00	0.00	0.00	0.00	0.00	2.5 6	0.00	0.00	0.00	0.00	0.00	0.00
Walk Scan	2.56	0.00	0.00	0.00	0.00	0.00	0.0 0	0.00	0.00	0.00	0.00	0.00	0.00

Feeding (Feed); Scanning (Scan); Preening (Preen); Standing on one leg (SOL); Walking (Walk); Wing flapping (Wing Flap); Cleaning; Jumping; Walking with wing flapping (Walk WF); Puffing; Excreting; Scratching and Walking and Scanning (Walk Scan).

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