SPREP HANDBOOK OF FIELD METHODS FOR STUDYING COASTAL ECOSYSTEMS: MANGROVES

w p. 5

Mangrove ecosystems

Mangrove ecosystems are defined by the presence of woody shrubs and trees growing below the high tide level, and generally extending down to the mid-tide level. They may comprise a variety of plant species and support a diverse and varied fauna. Above tide level the fauna is essentially terrestrial, while below tide level it is coastal and marine. Much of the flora and fauna is highly adapted to the mangrove environment and does not occur elsewhere but some species migrate or wander into adj.cent ecosystems. A number of the coastal fish and crustaceans may migrate between the mangroves and offshore zone at some stage during their life cycle. A simplified mangrove food web is shown in figure 1.

The mangrove flora and fauna varies regionally, and there is generally a decline in the variety and number of species eastwards across the Pacific and way from the equator. Within a region, the mangrove ecosystem varies with the substrate, wave action, freshwater run=off, rainfall, etc., and there is usually a pronounced zonation with different species becoming abundant at different tidal levels. This zonation is caused by both the physico-chemical characteristics of the environment and biological interactions with other species: the zonation may therefore change depending upon which other species are present.

MANGROVE FOOD WEB



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Definition of the Mangrove Area

Mangrove trees are mostly distinct in appearance, both from the ground and from the air, so the boundary of an area of mangroves is easily defined. On aerial photographs mangroves can usually be identified from the topography and their uniform tree canopy. If the mangrove trees are distinctly zoned then this is usually apparent on aerial photographs. It is recommended that aerial photographs of the region are obtained, at scales of 1:10,000 - 1:25,000, from which the mangroves may be mapped. Colour photography is preferable but not essential. If possible, an accurate large-scale base map should be used.

SPREP HANDBOOK OF FIELD METHODS FOR STUDYING PACIFIC COASTAL ECOSYSTEMS

Mangrove Trees

Mangrove trees have evolved in several plant families but many come from a single family; the Rhizophoraceae. These trees are able to live below high tide level, an environment in which their roots are periodically inundated with saline or brackish water. At times the substrate may be soaked with freshwater run-off or rain, creating low salinities, but after long dry spells the salt concentration at the inland edge of the mangroves may rise well above the seawater concentrations because of evapotranspiration Relatively few plants can live in these conditions and the mangroves show a variety of adaptations to coping with salt. The root systems are also adapted to growing in soft and anaerobic sediments: various species possess hollow root systems to facilitate gas exchange to roots, surface feeding roots, aerial roots, prop-roots, buttresses and spreading roots. The fruits and seeds of most species are well adapted to dispersal by the sea, and some species germinate before the fruit falls from the trees, thereby permitting very rapid establishment when a suitable substrate is encountered.

Several guides are available for the identification of mangrove trees in the pacific region.

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1997

nb. Genera and species not included in this guide may be encountered. For more details refer to the bibliography which follows. COMBRETACEAE

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24.42

Lumnitzera

L. littorea:

Mangrove tree, - 25 cm DBH, -20m high

Landward zone

Flowers red



Fruit green







1.

MELIACEAE

Xylocarpus

Mangrove tree, -75 cm DBH, -25 m High X. granatum:

Mid to Landward zone



Flower cream

΄.



1 cm

Seed angular brown





ι.

Fruit glossy green





RHIZOPHORACEAE

Ceriops

歴史

1000

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and a

C. <u>tagal</u>: Mangrove tree, ~15 cm, -15 m high. A Middle zone and along muddy channels.





SONNERATIACEAE

•,

Sonneratia

5. alba: Mangrove tree, -100 cm DBH, 30 m high

· · · ·

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Mid to Landward zone





VERBENACEAE

Avicennia

100

and the second

F

A. eucalyptifolia Mangrove tree

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J.A.

Leaf grey beneath, yellow vein above



A. marina:

As above but ovate leaves.

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S.P.R.E.P. HANDBOOK OF FIELD METHODS FOR STUDYING COASTAL ECOSYSTEMS

Mangrove Trees

A. Wood: Standing Crop

The standing crop of trees, measured as the volume ^ or weight of wood or useable timber, is useful for several purposes:

- i) It is a measurement of/quantity of plant material and this is a useful characteristic of the mangrove community. The standing crop of old stands of trees is a good indication of site productivity.
- ii) Total wood weight is a useful measurement for charcoal and firewood yield.
- iii) Stem or bole volume are useful measurements for timber yield.

The standing crop of trees is generally estimated in two stages;

1) The population of trees is determined in a study area, and for each tree one or more size indices are measured, e.g. D.B.H. (Diameter of the trunk at Breast Height = 137 cm Λ /Basal Area (cross-sectional area of trunks), and Height (either of the bole or the whole tree).

2) In addition to the size indices, the total or useable wood volume (or weight) is measured for a number of trees of different sizes; sufficient to establish a regression relationship or graphical relationship. The wood or timber volume of the trees in the population may then be estimated and a total volume or weight calculated.

Several alternative procedures are available for each of these two stages.

1) a. Quadrat Sampling

A number of quadrats (at least 30) are randomly or haphazardly located in the study area. Random quadrats are best located, ' using grid coordinates, while haphazard quadrats are best located at intervals along transect lines within each zone of 159 the mangroves. The quadrat size should normally include about 5-10 of the larger trees. A circular quadrat of 20m radius is commonly used by foresters and is relatively easy to define using an optical range finder, or a single rope or chain of 20m length. Obviously, a quadrat large enough to contain 5-10 large trees may contain many small trees, and rather than recording all the smaller trees it is better to use smaller quadrats for smaller trees. An appropriate scheme is to record:

e.g. All trees > 40 cm D.B.H. within a 20m radius. Trees > 10 cm D.B.H. < 40cm DBH within a 10m radius, Trees > 1.37m tall < 10 cm D.B.H. within a 5m radius, Seedlings < 1.37m tall within a 2.5m radius.

Such size class is analysed separately.

If optical rangefinders and optical trunk diameter gauges are available then all measurements can be taken from a central point (visibility permitting which greatly reduces the time required for each quadrat. There is, therefor more time available to record other quadrats and to increase the precision of the survey. Optical tangefinders are relatively cheap (e.g. 2-30m rangefin Cost 19835) by toptical dimeter gauges are expensive (e.g. Spiegel Relaskop ; Cost US\$550)

FORMAT FOR RECORDING OBSERVATIONS:

Quadrat Sampling:

Date: day, Month, Ye

Location:

Observer:

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•	Transect Jumber	Quadrat Number	Quadrat Size	Tree Number	Tree	Species	D. B. H.	Height	Notes
					-				
				0.0					
1									

b. Point-quarter samples

(see also section on sampling corals)

It is possible to use the distance (Di) from a randomly located point to the nearest tree as an estimate of tree density. This estimate is biased if trees occur in clusters, and an improvement is the point-quarter method which involves measuring the distance from a point (randomly or haphazardly located) to the nearest tree in each of four quadrants (e.g. compass quadrats; N.E. - S.E., S.E. - S.W., S.W. - N.W., N.W. - N.E.).

The average area per tree (Ai) is estimated as ,

 $\bar{A}i = ((D1 + D2 + D3 + D4)/4)^2,$

where D1 - 4 are the distances to the nearest tree in each quadrant. This estimate is not seriously biased in most forests but it will be biased if trees are highly clustered. Isolated trees are over-represented.

and clustered trees under-represented, so, if size is correlated with isolation then the method will give biased size-frequency information.

Generally it is desirable to sample different sizes of tree separately at each point, since tree-density alone (i.e. including seedlings - large trees) is of little value. Suitable size classes are similar to those for quadrat sampling;

e.g. < 1.37m tall 0 - 10 cm D.B.H. > 10 < 25cm D.B.H. > 25 cm D.B.H.

It is impractical to set a large D.B.H. for the lower limit of the largest D.B.H. class because large trees will be relatively sparse and long distances will be involved. In practice, it is desirable that individuals in every size class should be visible from the central point: no time is then wasted in searching for the trees.

If optical rangefinders and optical trunk diameter gauges are available then measurements may all be made from the central point and the sampling is very rapid, a large number of points may be visited (\gg 30) and results may be obtained with great precision. If optical equipment for remote measurement of distances and trunk diameters are not available, then each tree must be visited (and measured with a tape) and the method is much slower.

Since the method assumes that a tree can be found in each quadrant it does not work near the edge of the mangroves where one or more quadrants may contain no trees of suitable size. Strictly, it is not valid simply to calculate a mean distance from the other quadrants because this biases the result.

If a tree occurs on the border between two quadrants it should be allocated to one quadrant, i.e. rotate the quadrant slightly; a single tree should not be counted in two quadrants.

POINT OF ARTER SAMPLING:

Observer:	Date:	day,	month,	year
Loc tion:				

Innsect Number	Point Number	Tree Size	Iree arber	Distance (m)	Average distance	D.B.H. (cm)	Species	Notes
			1		1			
			2		1			
			3					
			1)			
		ĺ	2					

c. Basal Area

If the only parameter of interest is the total biomass or amount of timber, and not the number or size of trees or the relative abundance of different species, then the Bitterlich method may be used to estimate the total basal area of tree⁻ trunks per unit of ground area.

The technique works on the principle that forest basal area may be estimated from trunk diameters and distance of the trees from a point. for all the trees around a point. The trunk diameter forms an angle with the point and knowledge of this

angle is sufficient to estimate the tree's contribution to forest basal area (either the tree is small and nearby or large and distant). Instead of determining the angle for all the trees around a point, it is possible to estimate forest basal area simply by counting the number of trees around a point which exceed a particular angle, and multiplying this number by a conversion factor related to the angle. The angle is usually chosen to give a simple conversion factor to basal area in either metric (sq. m/ha) or imperial (square feet/acre) units. Although a range of angles could be used, it is usual to select an angle with a metric conversion factor of about 5, or an imperial conversion factor of about 20 or 40. Smaller conversion factors give more precise results per point but they are unsuitable if visibility is hindered by saplings, prop roots, etc. and for this reason they are not recommended in mangroves.

A variety of instruments are available for determining whether a tree exceeds the critical angle. The cheapest are 'angle gauges' which define the angle by sighting through a window at a fixed distance from the eye (e.g. 'Cruz-all', 'Panama' gauges: Cost US\$ 5-10). The most convenient are wedge prisms (Cost US\$16-30), which are small hand-held clear or tinted glass prisms. The image of the tree is displaced to the side by the prism; if the image overlaps with the trunk the tree is included in the count, if the image does not overlap the tree is excluded:





This technique is very rapid and, therefore, many samples may be taken (>>30) and great precision may be achieved. The limitation of the method is that it only provides the total basal area per unit of ground area. Total biomass must be obtained by calibrating the basal area records with known biomass records. Once a calibration has been made, however, the technique is a very rapid means to estimate biomass, especially for firewood or charcoal production in which bole size is not critical. For timber production the individual tree size of particular species is usually important.

An indication of total above ground wood volume may be obtained by assuming that, on average, tree shape is constant and that volume may be predicted from basal area and the height of the tree canopy:

V = K.H.BA
Wherm V - Tree volume, in m³ per ha.
K = Shape coefficient, 0.3 - 0.5.
H = Canopy height, of trees in m.
TA = Basal area, in m² per ha.

Date;

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Sq. m/Ha or Sq. ft/Ac

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Transect Number	Point Number	Count	Basal Area	Notes, e.g. Common species
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2) Estimation of biomass, weight, or volume of individual trees.

To estimate forest biomass it is necessary to establish relationships between D.B.H. or Basal Area and the biomass of individual trees. A wide range of tree sizes should be studied, including equal numbers -20 (about 10_A each) of small, medium, and large trees to provide a clear relationship. The relationship is generally non-linear but linearity may be acheived by taking the logarithms of both D.B.H. and biomass. Biomass is the weight of organic material in the trees and may be estimated by taking the oven-dry weight of the tissues (strictly, the inorganic content should be removed from the oven-dry weight, and this component is usually determined by burning a sample and determining the weight of ash remaining: the ash content is usually

1-5. of the oven-dry weight). Instead of oven-drying all the tissues in a tree, it is usual to dry small samples of wood, bark, leaves, etc. and to determine a wet weight, or volume, to oven-dry weight ratio. The ratio of weight : volume is the density of the surple.

Although biomass is the most appropriate measure for biological studies and for fuel yields, timber is usually measured by volume.

There are two common approaches to measuring tree blomass:

- 1) Maasure tree volume and use density determinations to estimate biomass,
- Measure tree weight directly and use wet weight: OMEN-dry weight ratios to estimate biomass.
 - i) a) <u>Tree volume</u>. The simplest means to measure the volume, of a large tree varies with the tree's growth form (shape) and whether the tree is standing or felled.

Wood volume is measured beneath the bark. Bark thickness may be found,

The tree is treated as though it were a series of simple geometric shapes, e.g. sections of cones, and the length (or height) and width of each section is measured. This is relatively simple, with a measuring tape, for fallen or felled trees but for standing trees it may be necessary to use optical instruments which measure trunk or branch. diameter and height above ground e.g. Spiegel Relaskop (Cost: US\$550).

The most accurate method is to measure the volume of all the branches, roots (if necessary), and trunk, but in practice it is easier to count the number of small branches or roots (e.g. \leq 5cm diameter) and to determine the volume or weight of a small sample only. Depending upon the shape of the tree it may be desirable to increase or decrease the size at which branches are counted and sub-sampled rather than measured individually. Counting should replace where individual measurement there are more than about 10 branches of a particular size. The sub-sample of branches should be taken from different parts of the tree, and may also be used for estimating the number or weight of leaves.

It the length of each trunk or branch section is relatively short (e.g. < 2 metres) then the mid-point diameter of the section gives a reasonable estimate of volume, i.e.

 $V = \pi r^2 h$, Where $V = Volume of section, cm^3$ $\pi = 3.142$ r = 5ection mid-point diameter/2, in cmh = length or beight of section, in cm

For longer sections and greater accuracy it is necessary to consider

the tapered nature of the section, ... i the 'mid-point' should be moved closer to the thick end of the section; e.g. at 43% of section length for a simple conical taper.

[entity. There its values r thads for determining the density
f = elet, and most require estimation of 'oth volute and weight.
'oth resculd be deter ined on the fresh sagie, not after it has
it and hrenk. V give pricedure are available:

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.) 's sum the diverse llength of the section - as discussed and - length volume.

(i) Ant opinge of the distinct it has regular rectangular sides.
 Which man be computed, and calculate volume.

i) The set the set of the spectrum. fill a container with stors to it to ver ally overflowing, gently submerge the spectrum of the store which overflows, measure summer storight (1 sm=1 co¹) is this water. This should

ii) <u>Measurement of the tree weight</u>. If trees may be felled and cut into small sections then it may be practical to weigh all the pieces of the tree. Several

B. Wood: Production

Standard forestry procedures are applicable to mangrove trees, but these are restricted by two factors;

1) Absence of well defined growth rings

The tropical climate may lack much seasonal variation, and the root environment is tidal. Variations in wood anatomy (which do occur) are not readily identified as annual rings. The use of increment borers to determine annual wood increment is unlikely to be useful except in very seasonal climates.

ii) Irregular trunk shepe

Many mangroves form buttresses or prop-roots which may extend considerable distances up the trunk. A clear, straight, circular bole may not be formed, or not until a height of several metres above the ground. The standard D.B.H. (Diameter at Breast Height (137 cm)), may not be useful: it is recommended that measurements are made above the buttresses and prop-roots.

Wood production is generally measured using the annual increment in bole diameter. Diameter may be related to wood volume (see section **an** standing crop) and annual diameter increments wolume or may, therefore, be expressed in terms of standing crop increment. In the absence of clearly defined annual growth rings, and a worker of the standard of the second of the second of the house of the second of the second of the second of the house of the second of the second of the second of the house of the second of the second of the second of the house of the increase in trunk diameter.

Annual trunk diameter increments vary with the species, tree size, and competition (especially shading). Isolated trees, free of competitors, may have diameter increments of 5-20 mm per year at diameters of 5-20 cm, declining to about 2-5 mm per year in trees exceeding 50 cm diameter. Under shaded competitive conditions, diameter increments rise to a level of about 1-4 mm per year when the trees reach the upper canopy level (usually with D.8.H.of 20-35 cm⁵).

Accurate measurement of growth requires techniques capable of measuring a diameterincrement le s than 0.5 cm per year or 0.5% per year. Either very accurate measuring techniques are required or the measurements must be taken over a period of several years. Since trunk growth varies from year to year, even the accurate techniques are of 11.tle value for periods of less than 2-3 years, and periods of about 3-3 years are preferable.

There are several methods commonly used for measuring trunk diameter increment:

a' Repaired girth observations

Select a portion of the trunk, 1-2 m above ground level, which is approximately circular in section and free of proproots or buttresses. Scrave off any 1 ose outer bark and epiphytes to form a clear band, 5-10 cm wide, around the trunk. Using a steel or fibreglass tape the the trunk circulference accurately to the nearest Q.5 mm, - core the value. Ising 0.9. Tree Marking paint or Boundary Marking a bind around the trunk where the circumference was measured, and plint a reference, on the truck ; it is usually simplest to paint the nut or vertically down the trunk (n.b. In some countries Forestry D.p. rtments use different coloured paints for different purposes, e.g.)range for research, Blue for logging, etc.). An aluminium •r ;lustic numbered tag may be nailed to the tree some distance above or below the band but there is often the risk that these may b removed or damaged by people and other animals. π subsequent occasions the trunk is remeasured around the painted hand, perhaps once each year.

b) Attached girth bands

A modification of the previous technique is to fix an aluminulum strip (1 cm wide) around the trunk; the two ends overlapping and held tightly in place by stainless steel spring joining the end of one band to the other band. The aluminium is scored with a vertical line in the zone of overlap, such that both ends of the strip are scored and coincide. As the tree grows the spring stretches

dotail:

and the distance between the scored lines on the ends of the bands increases and may be accurately measured with an optical or mechanical micrometer. This technique is potentially more precise and accurate than 'i' but any interference with the bands, e.g. by people, climbing animals, falling branches, atc, is liable to destroy the band and the record. Because of this it is always desirable to use method 'i' in addition to method 'ii'. Method ii is more time consuming to set up and much more expensive than method'i '(A stainless steel spring and aluminium band may cost US\$1-2), so it is often preferable to allocate the cost and time to sampling a larger population of trees with method i.

c) Anatomical markers

There are a variety of methods for accurately measuring diameter growth on one side of the trunk, but this may not be typical of the whole trunk.

These methods generally involve some damage to the cambium (e.g. temoving cores, injecting dyes, fixing pegs into the wood, etc.), may introduce diseases to the trunk, and may cause callous tissue formation: all of which are likely to produce atypical growth rates. Despite their precision, these methods are liable to be biased and to give *un*reliable estimates of diameter increment. Their primary value is for studying wood anatomy and seasonality of growth.

Selection of trees

To define the growth characteristics of a tree population it is customary to define a study area and to monitor all, or most, of the trees in this area, except for the smallest sizes. This is not a good procedur because the sample generally includes many small trees but few medium to large trees, so the growth rates are poorly known for medium large trees. It is desirable that an equal number of trees in each size class are selected, randomly, or haphazardly, from within a large plot. A sample of at least 60 trees, representing the full range of sizes, is desirable for each species

[7]

and for each set of invironmental conditions. It is important to realise that growth rates a hieved in (1 - p) = n or logged area will be quite different growth rates in an undisturbed forest are. Seedlings planted in an open area will become computitors as they grow larger and, therefore, their growth rates will decline to the levels in competitive hibitats.

Morrality estimation

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not In "thempti's st make wood production accurately it is sufficient s - 1. to know the initial tree repulation and growth rates: the yield is reduct by monthliny. Contality may be measured as the proportion of individuals in a size class which die during a relatively short interval. In a staribute of every individual tree, lot nly about (0.5 in the stand 15 - 100% of seedlings die each year, the sample size regulred to achieve stimute in whit I by increases, and very large sample sizes ir rounod who goo ortion, the reach Glor 100%. Depending upon the k t t i v 1 int linv, m les of several hundred trees in each The late hould be observed with year for a 2 - 3 (-10) year period In reason of estimat sol portality. Mortality is caused by intits of fact rs, including ompetition (natural thinnning), disease ropic counts such as lithcing, cyclones etc. Some causes of "" iffect is lited individuals, some affect slow growing stunted indian als, and some affect fast growing tall individuals. Larger in the study periods are required if catastrophic event it a major cluse of mortality.

Recruitment

The standing crop of trees is periodically increased by seedlings. The biomass increment is minimal but these seedlings counteract the . losses due to tree mortality. Seedling growth and survival are generally greatest where there is a gap in the tree canopy, such as that causedby tree death. Ecological studies may emphasize the early stages of seedling establishment, growth and survival but forestry studies generally ignore the early stages becaused of the high and variable mortality. Forestry studies genrally measure the recruitment to the smallest tree class, e.g. plants with a height of 137 - 200 cm. or a D.B.H. of 1-5 cm. In either situation, the methods are similar. Permanent plots, e.g. Im x 1m to 10m x 10m (depending on the seedling or sapling size at which recruitment is to be recorded), are set up and plants exceeding the recruitment size are tagged. Periodically the plot is revisited, the growth and survival of the tagged plants is recorded and the appearance of new plants exceeding the recruitment size are also recorded. For seedling studies the visits may be at 1-3 monthly intervals, for forestry studies the interval is usually 1 year and coincides with measurements of tree growth.

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C. Litter fall

The quantity of leaves, stipules, flowers and fruits falling from the mangrove trees and epiphytes are of interest for two main reasons;

- i) They indicate the primary productivity of the mangroves.
- 11) They are a major source of food for intertidal animals, fungiand bacteria and they may be carried seawards by the tide where they act as a source of food or nutrients for sea-grass, mud-flat, lagoon, coral reef and ocean ecosystems.

Mangrove leaves are used as fodder for domesticated grazing animals in some places. The leaves appear to be nutritious and digestable. Litter production figures indicate the potential yield of leaves, though grazing on live plants may cause shoot damage and decrease the yield.

Mangrove litter fall may represent as much as half the total plant production, and ranges from about 2-4 gm dry weight $m^{-2} day^{-1}$ in mangroves at about Mean Sea Level to 1-2 gm dry weight $m^{-2} day^{-1}$ at the landward edge of the mangroves, 1.5-2.0 m above Mean Sea Level, and on sites lacking an input of silt and nutrients.

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The principle behind litter fall trapping is simply to place open bags, of known area, beneath the trees and to record the quantity of leaves falling into them over known periods. In common with other sampling techniques, replication is desirable to indicate the variability which arises between different trees, species, zones in the mangroves, etc. A large number of small traps is preferable to a few large traps.

Traps may be constructed from a circle of stiff wire (e.g. fencing wire, or plastic sheathed copper cable), from which a fine mesh sheet or bag (e.g. plastic mosquito mesh for windows) is suspended. The fine mesh may be sown or stapled onto the wire frame and should form a bag at least 30radeep. A stone may be placed in the bag to prevent the wind from lifting the mesh and spilling the contents. The wire frame may be either i) stood on three stiff wire or wooden legs which are firmly buried in the substrate or, ii) suspended from three lines (e.g. fishing line) tied from the trees to the net and then from the net to roots on the ground. It is important that the net cannot be blown around by the wind. In both cases the net must be above the level of the highest tides.



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The optimum size of the trap will vary with the rate of litter fall and periods of trapping; larger traps being required where the amount of litter per trapping interval is low.

Suggested dimensions for the frame and bag are: Wire frame; 220 cm length of wire, forming a circle of 200 cm circumference and 0.32 sq. m. area (Cost: US\$0.50).

Fine mesh; 1 sq. m. of grey or green plastic mosquito window mesh Legs; 3 x 1.5-2m poles or stiff wire , or fishing line (8-15m; Cost: US\$0.10 -0.30). Staples (preferably copper to resist rusting).

(cost; US \$1.50)

Traps may be labelled by writing on the mesh with a permanent ink felt tip pen or by attaching a plastic or aluminium label (Garden or Horticulture labels, US\$0.03 - 0.15 each).

Since the trap is left in the mangroves it is advisable not only to seek permission to use the area but also to explain to people who live Mearby or use the area (especially children) that the trap should not be touched. It is advisable not to place traps where they can be seen by casual passers.

Traps should be located as though they are quadrats (see section). Important considerations are whether survey is intended to provide values for the total area, or for comparison of selected zones or species. When the trap is set up it is suggested that a couple of clearly marked leaves (e.g. with an ink cross, or a staple punched through the blade) are placed in the trap. if the trap is working efficiently these leaves will remain in the trap until it is collected but if wind, birds, people, etc. are removing leaves then this should be apparent and some modification to the procedure may be required.

Traps should be collected every 7-30 days; the more frequently the better if time and cost are not major considerations. Edible fruits and leaves are likely to be consumed or removed from traps by various herbivores and decomposers so long trapping intervals are likely to underestimate litter fall.

Most tree species have seasonal patterns of growth and this is reflected in various litter components. Stipules indicate new leaves expanding; flowering, and fruit fall are usually fairly synchronous and may be annual, biennial or less frequent, the second spectra are decisions - studen of at least one year and preferably for several years.

The information to be recorded from these traps may include:

- i) The number of leaves, scipules, fruits, etc. from each species.
- 1i) The fresh and dry weights of each component. A sensitive spring balance may be used for weighing fresh samples (e.g. 10 gm, 100 gm ran Cost US). If numbers are recorded, then the average fresh and dry weights of each component should be determined so that total weights may be calculated.

The energy and chemical composition of the various litter components may generally be determined using standard techniques but care should be taken to ensure that the high salt content of some tissues does not interfere with the methods.

Project:

Collector:

Location:

Location No. (if any):

Date litter lastcol	lected:	day,	Month,	Year
Today's date	:	day,	Month,	Year

tter Trap Number	Tree Species Overhead	Number & Stipules	/or weigh Flowers	t of Fruits	Leaves	Total weight	Notes
						178	

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SPREP HANDBOOK OF FIELD METHODS FOR STUDYING PACIFIC COASTAL ECOSYSTEMS

Mangrove Molluscs

The mangroves, adjacent mud-flats, and sea-grass ecosystems have a diverse and abundant mollusc fauna comprising mostly bivalves and 'gastropods. The bivalves are filter feeders, while the gastropods are mostly carnivores, though some gastropods graze on algae and micro-organisms growing on surfaces. Many of these species are used as food items and they may supply more food than coastal fish and crustacean fisheries.

These molluses are characterised by the outer calcareous shell and most field observations are made on the shell rather than the living tissues. The shells grow by addition of material to the lip. Adult bivalves are more or less sessile and live either partly buried in mud and sand or attached to the surface of mangrove roots, rocks or other shells, sometimes forming dense clusters. The fertilised eggs give rise to freeswimming larvae which may be carried in tidal water before they settle and develop a shell similar to the adults. Juveniles of some species may be capable of limited migration but few species have mobile adults.

adult gastropods may be highly mobile. They lay jelly-like masses of eggs; the larvae which emerge are free swimming and may be transported considerable distances before settling and developing a shell.

A. Standing Crop of Mangrove Molluscs

Adult molluscs may be sessile or mobile but they do not move rapidly relative to man and may therefore be sampled as though they are sessile.

The simplest sampling procedure is to use quadrats haphazardly located within each tidal or mangrove zone. The quadrats may be located along transect lines, either a series of transects running from the sea to land, which may subsequently be divided into zones, or transects within each zone. Random quadrat locations are required if great accuracy is needed.

The quadrats should be as small as possible, but this will vary with the species of interest. A quadrat of 20 cm to 50 cm diameter is usually adequate, and the survey may include 50 - 150 quadrat sites. The quadrat may be projected vertically to sample buried individuals or individuals on trees.

Buried molluscs should be removed from the substrate, washing the excavated material through a sieve (e.g. 2-3 mm mesh spacing), small molluscs are easily overlooked in hand-sorted samples of sediment.

It is generally useful to record the size of the individuals in each quadrat, measuring to the nearest 0.1 - imm with a micrometer (see next section).

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Table for recording observations of molluscs in quadrats

Observer:	Date:	Day,	Month,	Year,
Location:				

Quadrat size =

, depth of excavation =

сm

Transect	Quadrat	Species	Langth of challe m
			Lengen de Suerrs, mm
Number	Number		
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SPREP HANDBOOK OF FIELD METHODS FOR STUDYING COASTAL ECOSYSTEMS

MOLLUSC PRODUCTION

1. Size and biomass

There is generally a very close relationship between the size of a mollusc shell (e.g. length, width) and the biomass (oven dry weight) of the fleshy tissues. By selecting about 60 individuals from a wide range of sizes it is possible to draw a graph of biomass v.s. shell size. This graph is unlikely to be linear and a straight line may be achieved by plotting log₁₀ Biomass v.s. log₁₀ Shell size.

If possible a regression line should be calculated. Having established the relationship, at a particular site, it is then possible to use shell size as an indication of biomass.

It is necessary to measure lengths (using calipers or micrometers) to the nearest 0.1 - 1.0 mm and biomass to the nearest 0.01 - 0.1 gm;

greater precision is required for the smaller species. TABLE FOR RECORDING DATA:

Species:

Location of samples:

Observer:

Date:

Shell No.	Shell síze (mm) Length/Width/Height	Oven Dry Weight of Flesh (gm)	Log ₁₀ Size	Log ₁₀ Weight
1				
· 2				. 2
I			,	

2. Measuring Growth in Shell Size

Samples of about 200 molluscs (or more if the species is mobile), representing the whole size range, should be collected in each subdivision of the study area. It is advisable to divide the size range into about 6-10 size classes and to collect on equal number from each size class.

a. Increase in total shell size.

Shells are individually numbered using paint, or tags fixed to the shell. The shell is then accurately measured, in a clearly defined directi and remeasured periodically, e.g. every 1-2 months.

The individual marking of shells presents considerable practical difficulty and several methods may be tried. The techniques are best suited to large species. The mark should not be so obvious that it will affect predation.

i) Paint the number onto a portion of the shell, which has been scrubbed with a tooth brush or wire brush to remove epiphytes, etc, and allowed to dry before the paint is applied. Various paints may be applied, e.g. fast drying outdoor acrylic lacquers (e.g. for cars), but care should be taken while the paint dries and with marine paints because some are toxic to molluscs (never use antifouling paint!). Depending upon the species and its habitat the paint may remain visible for periods of a few weeks or months: the paint gradually wears off so this technique is not advisable for studies lasting for many months.

ii) Aluminium or plastic tags with the identity number scratched or
 cut into the surface may be glued onto the shell using quick-setting

resin glues. It is advisable to scrub the surface clean of epiphytes and to roughen smooth surfaces with a file or emery paper. <u>n.b.</u> Some resin glues soften if continually submerged. Resin glues may be toxic before they set so care is needed to avoid poisoning the mollusc.

Small tags probably do not greatly inconvenience the mollusc and may remain intact for several months.

111) Large gastropods, e.g. <u>Trochus</u>, may be tagged by drilling a small hole through the shell (near the lip) and attaching a numbered aluminium washer (7-12 mm diam.) using a pop-rivet (2.4 mm diameter). The gastropod secretes a smooth layer of shell over the inner head of the rivet, and the tag remains attached virtually indefinitely. (Ref: Heslinga, G.A. & Orak, O. (1984). A permanent tag for large marine gastropods. <u>Aquaculture</u> 36, 169-172).

A similar technique may be used on bivalves but great care must be taken to drill near the outer margin where the animal will not be damaged. IV) Size Measurements

Size measurements should be taken along the axis which shows the greatest growth (maximum length) with a vernier calliper accurate to the nearest 0.01 - 0.1 mm.



b. Increment in shell size.

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Instead of measuring the overall change in shell size it is possible to record the amount of shell growth over a period of time, e.g. a few weeks or months. There are several techniques available, and these do not require the identification of individual marked shells, though this may be desirable for other reasons. These techniques are suitable for small species, e.g. less than 2 cm long.

i) Staining of shells

1.1.1

A stain such as Alizarin Red is virtually non-toxic and stains shells red. Unattached Molluscs may be placed in an aerated 1 litre beaker or plastic bag of seawater to which about 0.5 gm (about half a level teaspoon) of the dye has been added. It is advisable to scrub the shells clear of any epiphytes, etc. with a wire brush on toothbrush. If the container is aerated (Aquarium pump) the molluscs may be left for 6 - 18 hours in a shaded place but if the container is not aerated then the water should be changed after about 6 hours.

Note carefully whether the shells are stained up to the edge, and also whether the operculum (in gastropods) has stained.

The molluscs are then returned to their natural habitat and later recaptured. Growth since the initial marking is visible as unstained shell and should be measured with a micrometer. An optical micrometer, e.g. microscope is ideal, (See also the notes on growth rings). The stain may remain visible for several months.

Molluscs which are attached to the substrate, e.g. cysters and mussels, may be stained by making up a paste or viscous solution, e.g. 186

agar, cornflour, glycerin, to which the dye is added, the solution being painted on the shell at low tide, but it is difficult to obtain adequate staining.

ii) Physical marks

Using a small triangular metal working file (e.g. Saw tooth file) with a width of 2-3 mm, cut a Unotch or series of notches into the outer rim of the shell where the new shell is laid down. It is preferable that the notch is of a known depth, e.g. 2 mm, and in this case it may help to paint a mark on the file (if it tapers) where it is this width and to file the shell until the notch is as deep as the file.



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are damaged. The shell continues to grow, infilling the notch and laying down a new layer of shell. The infilled notch should be visible and the amount of growth since the notch was cut should be apparent and measurable.

(11) Growth ring induction.

Certain chemicals, e.g. E.D.T.A. interfere with calcium metabolism and prevent the deposition of shell material. If a mollusc is placed in an aerated % solution of E.D.T.A. for hr., shell growth is virtually suspended and this is apparent as a distinct ring on the shell. The shell is then returned to its natural habitat for a period of weeks or months so that the growth increment may be determined.

c. Growth rings

Most nolluse shells, especially bivalves, form fairly distint patterns of growth rings in their shells. Gastropods may also form similar rings on the operculum. These rings, where the shell is thinner or thicker are generally caused by fluctuations in environmental conditions such as temperature, salinity, food availability, etc. The tidal cycle is one cause of such environmental fluctuations and there are usually cycles of about 12½ hours (tidal cycle), 24 hours (diumal -nocturnal), 14 days (springneap tides), and annual climatic cycles. Growth may fluctuate with one or more of these cycles inducing regular patterns of rings. Molluses probably live for 1-10 years, depending on the species, so the total number of rings on large specimens will give an indication of the likely frequencies of i ring formation.

A low power microscope (e.g. binocular microscope with 10X eyepiece and 4X objective) or 10X hand lens will assist in studying these rings. If the rings are not clear it may be helpful to cut a section through the shell; grind it smooth with fine emery paper (e.g. P-600) and examine the cut_surface. Staining with Alizarin Red or other stains may help to show the rings.

By combining a study of growth rings with a study of growth rates it may be possible to interpret the ring pattern and thereby determine the growth history of individuals. It is then possible to define growth rates with greater accuracy.

A useful graphical method for analyzing growth, when distinct growth rings are present, is to use a Ford-Walford plot. The increment in growth (distance) between each ring is measured and used to plot a graph of total length up to ring number 'Lt' against total length up to ring number 'Lt+1'. For most mollusc shells the points on the graph will fall about a straight line. The line may be fitted with a regression procedure or, less satisfactorily, visually. The ratio of Lt+1 : Lt will approach 1.00 as the shells reach their maximum size, and graphically this is the point at which a regression line through the data intercepts a line with a 45° slope.

Intercept Data & Lt + 1regression 45° slope (Lt+1 = Lt)

Lt 89

3. Study Area

To obtain realistic information on growth rates it is necessary to return marked individuals to their natural habitat. Studies in Laboratories may give growth rates much greater or less than the natural rates.

Generally it is simplest to place the marked individuals in one or more plots. The plots may be rectangular with posts or stones marking their corners. If the area is frequented by people it is desirable to avoid using obvious markers since these may be removed and the plot disturbed. On mud flats the location of a plot may be defined by lining up two different pairs of distant objects, e.g. channel markers, hills, houses, trees etc.

Burrowing bivalves generally remain within the plots but the more mobile gastropods may migrate considerable distances. To recapture the marked gastropods it is necessary to search the surrounding areas; which may also give a useful indication of migration. It may help to place suitable baits, e.g. dead fish, crabs, or bivalves as attractants 1-3 hours before the capture is to be made.

To ensure sufficient recaptures of the smaller species of gastropod it may be necessary to mark 500-1000 individuals. If the risk of human interference is low, fine wire mesh enclosures or cages may be used to contain the marked individuals but, as in a laboratory experiment, these may change the environment significantly such that the results are atypical. The longer the cages are used the more atypical the results may become since the populations of other species will also change, and detritus may be excluded or may tend to accumulate. To minimise these problems, the cages or enclosures should measure at least 5m x 5m. It is necessary

to determine the climbing ability, and behaviour of the species before deciding on the structure of the enclosure (i.e. mesh size, depth of burial of mesh, height of walls, need for a roof or floor etc.).

4. Results

Growth rates (of size or biomass) vary with the size of the individual and generally increase from the larval stage until the shell is a few mm long and then gradually decline as the shell becomes larger,





It is, therefore, desirable to plot a graph of growth rate with shell size and either,

i) fit a curve to the results; probably of the shape shown above,
 or, ii) divide the results, according to the initial size of the shells,
 into size classes and calculate the average growth in each size class.

A size:age relationship may be calculated, starting with a small size or size class and estimating monthly growth to yield the shell size one month later; the process being repeated until the largest size is reached. This will generally yield a graph of the form shown below

Size

A curve of this type may be described by the logistic equation. In areas which are heavily exploited the larger molluscs may be very rare or absent and in this case the larger sizes will be missing in the samples and the growth rate will not slow down as noticeably as the graphs shown above indicate.

The instantaneous rate of growth (q) may be estimated as follows:

q =
$$\frac{\log_e W t_2 - \log_e W t_1}{t_2 - t_1}$$

where Wt_1 and Wt_2 are the weights at time t_1 and t_2 . It is usual to use \log_e rather than \log_{10} for these calculations.

To estimate production by a population it is necessary to known the rate at which new larvae colonise the area, and the mortality of each size class. This information is not easy to obtain accurately, but it may be estimated using capture-mark-recapture methods (see separate discussion) with very large samples.

An approximation to the production of useable biomass may be made by assuming that the population has constant rates of natality (births) and mortality and, therefore, that the frequency: Size class structure is fixed. If the species reproduces continually and steadily throughout the year then the frequency: Size class relationship should show a steady stepwise decline in numbers with increasing size. It is necessary to adjust the results to meet this condition; this may be done by increasing the range of eac size class. Since very small individuals may be missed because of inadequate sampling methods it must be assumed that these are present in numbers equal to or greater than the next larger size class. The decline in number between

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each size class is then taken as an estimate of mortality, and production is then estimated by the growth in the size of individuals in each class that are either alive at the middle of the period (estimated as the average of the number alive at the beginning and the end of the period) or have . been removed for human exploitation.

If the species reproduces seasonally, then the population may comprise a series of year-classes: these should be apparent as a series of obvious and fairly regular peaks in the frequency: Size class relationship. It is necessary to confirm that these peaks really represent distinct age classes and that they are not the result of insufficient sampling (interface distinguish where and forgete interface are of a sampling every 2-4 months should show that the peaks in the frequency: size class graph advance until one year later they have reached the size occupied by the previous peak. If the sampling technique is reliable, and gives absolut abundance, it should be possible to measure the annual decline in abundance (= mortality + emigration). Alternatively, the decline in total numbers in successive year classes may be used as estimates of mortality.

Examination of growth rings may indicate age classes which are not apparent from size-class data.

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SPREP HANDBOOK OF METHODS FOR STUDYING PACIFIC COASTAL ECOSYSTEMS

MANGROVE CUSTACEANS

There are many diverse and abundant species of Crustaceans living in the mangroves and adjacent ecosystems, of which three groups have considerable economic significance as food items: crabs, lobsters, and prawns. The crustaceans are a diverse group but they share certain characteristic features, including a chitinous exoskeleton and a life cycle involving 20-30 moults. This repeated moulting creates a problem for field studies because it is difficult to tag individuals for growth or population studies. Overall growth in size takes place by rapid expansion of the new cxoskeleton after moulting of the old shell; the exoskeleton retains this size until it is shed.

Mating occurs after the female has recently moulted, and the fertilized eggs may be carried until they hatch as free-swimming larvae. Crustacean larvae form one of the main components of the zooplankton in the coastal ecosystem.



BURROWING MANGROVE CRABS (Brachyurans)

Many species of crabs live as scavengers or predators within the mangroves. Many genera and species are widespread on western Pacific islands. In a particular region, the abundance of different crab species varies with the position in the tidal zone, salinity and the substrate. In productive habitats, such as some estuaries, the populations of *mud-sifting* scavenging crabs may reach $40 - 80 \text{ m}^{-2}$ (10 - 20 gm biomass m⁻²). Larger species such as Scylla, may average 1-10 crabs ha⁻¹ and yield 2-20 kg fresh weight ha

On firm silty substrates the burrows are obvious but on coral rubble or rocky substrates the crabs use natural cavities and these are not so easily located. Crabs may burrow into soft silty sediments without creating a visible burrow. On coral or rocky substrates lacking large cavities the larger crabs may take refuge amongst the roots buttresses, and cavities in mangrove trees.

Most crabs will avoid people, taking refuge in burrows or other cavities, but will resume their activity if the person remains still are active for some time. Most species are active by day and a few Aby night.

Most species feed at low tide while others feed at high tide. Some species use burrows constructed by other species. The behavioural and burrow characteristics of a species largely determine the most suitable methods for studying their populations and production.

A. Standing crop of crab species

Adult Crab populations may be studied by observation of individuals above ground level, excavating individuals from below ground, and by trapping. Larvae may be released within the mangrove zone at high tide or in spawning grounds some distance from the mangroves. The larvae are planktonic for the first 2-3 weeks and move with the tides and ocean currents before settling in the mangroves as juvenile crabs.

a) Observation of individuals above ground

Depending upon the abundance of the crabs, quadrats of im x 1m to be 10m x 10m may defined and watched by a stationary observer for 10-20 min. Video cameras or binoculars may assist in making these observations. The number, approximate size and sex of the crabs' which emerge may be recorded. The number of burrows should also be recorded to indicate the proportion of burrows containing active crabs. Inactive crabs, such as those which have recently moulted, are likely to remain underground. Crab activity is generally greatest as the tide recedes, and sampling after spring tides has been recommended

5) Removal of crabs from burrows

Crab burrows vary in size and shape with the species and size of crab. Usually burrows descend steeply for 15 - 40 cm and then extend horizontally for 5-30(-300) cm. It is useful to learn to recognize signs that a burrow may be occupied: fresh tracks, excavated piles of silt, and the absence of obstruction such as leaves and other' debris. Burrows are often blocked as the tide rises.

Crabs may be removed from burrows either by digging up the whole burrow, or by probing with a hooked, flexible stick (e.g. mangrove sapling) and pulling out the crab. Small crabs may be dug out relatively easily and caught by agile collectors. Large crab species may form long burrows and they are both more difficult to catch and

to handle. A single burrow may, at least temporarily, contain more than one crab; sometimes copulating crabs. In some species male crabs are found in burrows much more frequently than females. Female crabs, especially of the larger species and landward species, may be absent from the mangroves, travelling to and from spawning grounds. Spawning migrations usually coincide with the lunar cycle and seem to be most frequent about the time of the new moon; there may also be seasonal variation. As discussed above, some crabs may burrow in soft silt and not form obvious burrows. Some species may live above grounc and use tree cavities as refuges but tree climbing apecies are absent from the Pecific region.

A particular species may produce burrows scattered throughout a zone of the mangroves, e.g. some fiddler crabs (<u>Uca</u>), or they may make burrows in particular sites, e.g. the Mangrove Crab <u>(Scylla</u>) often burrows from within a small pool. With experience the tracks of a crab species may be recognized and traced to the burrow.

Once the crab has been excavated a variety of observations may be made, including size (carapace width), sex, presence of eggs, maturity, moulting condition, etc.; the crab may then be takked and released for further study.

Once the ratio of crabs: burrows has been established with reasonable precision is may be sufficient simply to count burrows.

c) <u>Traps</u>

The larger, rarer and more mobile species of crab, especially predators, may be captured using baited traps. Trapping does not yield an estimate of absolute abundance but it does give an indication of abundance and provides individuals which may be marked and released for mark-recapture estimates of population size or removed from the population to determine the determine

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Avariaty of crab traps have been devised, and there are often - local varieties which may be well adapted to local species and conditions. In general, a crab trap comprises a rigid cage of wood, bamboo, wire, or plastic with one or more passages leading to near the centre of the cage. A crab may enter through the passage but since the internal opening is raised from the floor of the cage it is unlikely to find its way out. There is usually a door for removing the crabs, and there may be an escape hole large enough for crabs below the legal minimum size to escape. The trap is baited with food remains, especially fish, placed into a small cage secured inside the trap. 62

A trap measuring about 50 cm x 50 cm x 80 cm, with one entry passage at each end, may be made from a roll of stiff galvanized wire measuring 300 cm x 100 cm. (Cost US \$3,00).

Severel (5-10) traps should be placed in pools or mangrove channels, where they will be continuously submerged, and should be visited every 6-12 hours. The traps should be secured with weights or ropes, and hidden to avoid theft.

Observations on size, sex, eggs, maturity, moulting condition etc. may be made, and crabs may be marked or tagged for further studies. Captured crabs rarely represent a random sample from the population. Population estimates based upon mark-recapture procedures on trapped crabs may, therefore, be biased. Some crabs remain in the vicinity of a particular burrow while others move about in a more or less haphazard manner and mix with other crabs in the vicinity. If markrecapture procedures are used, it is recommended that a simple Lincoln index is used over a short study period (12 - 24 hours). To reduce the bias introduced by 'trap-happy' crabs which revisit traps immediately it is suggested that either these receptures are ignored, or that traps

Marking and tagging crabs

Since the exoskeleton (shell) of crabs is shed at each moult any marks or tags attached to the shell are lost. The period between moults varies . from a few days for small crabs to several months for large crabs. It is, therefore, possible to conduct short-term marking studies by simply painting reference numbers onto the (dried) shell of the crab. Quick or boys, drying acrylic enamel paints, e.g. for automobiles Λ may be adequate for this purpose.

Long-term marking is more difficult, especially if individual indentification is required. Tags attached to the body of the crab such that they are visible but do not interfere with moulting have been used.

<u>T-bar anchor tags</u> may be attached to the crab at the point of juncture between the abdominal plate and carapace. The small plastic tag is inserted with a tagging gun (e.g. Dennison Mark II gun with Floy 68-B T-bar anchor tags). The needle of the gun may either be pushed through a hole in the abdominal plate, or inserted along the underside of the carapace until it is free of the abdominal plate, before the trigger is squeezed. The tag must be narrow and not widen appreciably because this may cause the old shell to catch on the tag and, either keep the crab attached to its old shell, or, this may rip out the tag and possibly damage the crab. Tags may be colour coded or have a written message.

Since crabs are often used as food items the tagged crabs may be captured by fishermen and it is desirable that the tag should include a message stating that a reward is offered for the return of the crab and tag. It is, obviously, advisable to discuss the trapping scheme with everyone fishing in the area. This may substantially improve the rate of recaptures and account for the loss of tagged crabs from the population.

2.01

Sheet for recording observations on Crustaceans

Collector:	Date:	day,	month,	yea
Location:				

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-							-	
	Site or trap	Species	Carapace Size mm	Moult hard/soft	Sex M/F	Egga	Taga	Notes
No. of Concession, No. of Conces								
						4		

Crab growth and production

During their life cycle, of perhaps 2-4(-6) years duration, a crab may post-larval undergo 15-25 moults, initially every 6-14 days, eventually every 60-90 (-180) days. After moulting the crab must take in water to swell out the new exoskeleton (shell), and the growth (carapace) increment is usually 5-20% in small species and 10-25% in larger species:

Carapace Increment = 100. Post-moult Carapace Width - Pre-moult Carapace Width
Pre-moult Carapace Width

Moulting is most frequent at the time of spring-tides (full and new moons), perhaps because of the need for water of suitable (sea water) salinity. The new shell is soft to the touch and takes 10 - 18 days days to harden. During the post-moult period, while the shell is soft, the crabs may initially remain buried or in burrows. Female crabs generally mate immediately after moulting.

The growth pattern for a crab species require information on two factors:

i) the variation in Carapace Increment with size

ii) the duration of the moult cycle as the trab grows Several methods may be used, and it is advisable to repeat the procedures on several occasions, especially if the climate is seasonal.

a) Laboratory observation

Laboratory studies are liable to be biased but they provide a simple means to determine moult cycle duration and growth for at least the first moult cycle after removal from a natural habitat. Crabs are placed into individual beakers of fresh mud with shallow seawater, changed daily, and the crab is observed daily for signs of moulting (and egg production). Measurements of the pre-moult and post-moult carapace width are made. The period between moults may also be recorded. Crabs may survive under these conditions for several moult cycles but the length of later cycles tends to be very variable.

b) Tagged crabs

In natural situations it may be possible to monitor the growth of individual tagged crabs, recording Carapace Increment and estimating the duration of the moult cycle. For accurate estimates of moult cycle duration it is necessary to recapture the crabs frequently, e.g. every 1-2 weeks.

c) Estimation of moult cycle duration

The average duration of the moult cycle (Dm), for a particular size range of crabs, may be estimated from the ratio of the average duration of the soft-shelled post-moult period (e.g. 10-18 days) (Dp): the proportion of crabs in the post-moult period (Pp).

Dm = Dp/Pp, in days

This estimate may be used on field populations, provided that individuals at all stages of the moult-cycle are captured (including buried individuals).

The duration of the post-moult period may be studied in the laboratory, as described above. Each day the crabs which initially had soft-shells are re-examined. If moulting occurs at all times, the duration of the soft-shell period is twice the average duration of the soft-shell period in the laboratory. If moulting occurs at spring should be tides then this should be apparent when comparing. Crabs collected on different days during the spring tide to neap tide cycle.

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d) <u>Growth layers as an indication of moult cycle duration</u> (no This technique is tentative: more research is required)

The cuticle of arthropods is layered and it has been suggested that layers are formed regularly. In crabs these periods may be tidal or daily. These growth layers are visible in sections of the cuticle when viewed with a microscope and may be visible in untreated speciment

Specimens of cuticle may be examined as follows:

Choose a relatively flat surface of the exoskeleton and remove a piece, e.g. 1 cm². Fix the specimen in 5% formaldehyde - in which it may be stored - and then decalcify the specimen in 70% ethylalcohol acidified with few drops of hydrochloric acid. Infiltrate the specimen with paraffin wax and section vertically at about 10 µm thickness. The growth layers may be more readily seen if they are stained with aniline blue and viewed with phase contrast. The layers are generally about 10-15 µm apart.

Before the growth layers may be used as an index of age since the last moult, it is necessary to establish the actual frequency of formation. This may be done by examining crabs of known age since the last moult, or by inducing a distinct layer in the cuticle and then examining the crabs on subsequent occasions. A distinct layer might be produced by placing the crab in an aquarium overnight, with or without chemicals such as E.D.T.A. which disrupt calcium metabolism.

Section of cuticle :



cuticle layers formed before old shell shed

Cuticle Layers formed" after old shell shed

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Cohort analysis for growth and mortality

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Crabs may have a distinct spawning season, or at least a peak in the proportion spawning, and this may produce a cohort of juveniles which can be monitored by sampling at 1-3 month intervals. The shift in the size-frequency peak will indicate the mean growth rate of the cohort. Since growth rates vary amongst individuals the peak will become less obvious as the crabs grow and cohorts are unlikely to be detected amongst large crabs. The decline in the number of individuals in the cohort will indicate mortality.

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SPREP HANDBOOK OF FIELD METHODS FOR STUDYING PACIFIC COASTAL ECOSYSTEMS

THE MANGROVE LOBSTER

The mangrove lobster, <u>Thalassina</u> <u>anomala</u>, is widespread in the Indowest Pacific region, and is considered a food delicacy in some areas but as a pest in other regions because it's burrows may damage dykes. The lobster forms characteristic large conical mounds (20-60 cm high) at the entrance to its burrows, which may be abundant near the inland edge of mangrove swamps. The adult lobster lives and feeds within the burrows, which extend over several metres and descend to depths of 30-60 cm. The long burrows make capture of the lobsters difficult. Lobsters may be flushed out of the mound by removing the plug of mud at the entrance and, using a foot, 'pumping' water into a section of the burrow some distance from the mound: the lobster is caught as it flees. A variety of traps may be set at the entrance to the burrows.

It is thought that there is a single breeding season each year and the population may form recognizable cohorts. Female lobsters may migrate to the sea carrying their fertilized eggs, where the larvae hatch, but the larvae may simply be released into the tidal water within the mangrove zone.

Most of the techniques for studying mangrove crabs may be applied to the mud-lobster.

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PENAEID PRAWNS

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Penagid prawns are a major and valuable food resource in many parts of the western Pacific and south-east Asia. The prawns are either caught as juveniles in mangrove creeks and inshore waters or as adults in offshore waters at depths of 5-20 (-40) m. Many species occur in the west of the Pacific region, e.g. 20 species in Malaya, but the number of species declines eastwards. Local fishermen often lump several species under a single name. On the basis of prawn fisheries, adjacent to areas of mangroves, it is estimated that prawn yields (fresh weight) are 15-25 (-100)kg/Ha/yr.

The penaeid prawns are heterosexual and breed and spawn in curbid offshore water, generally at depths of 10-30m. The numerous (20000 -100000) eggs are released at night, soon after fertilization, and pass through about 12 planktonic larval stages within 12-18 days. The larvae which are carried into near-shore waters may enter brackish estuaries, sea-grass beds lagoons, and channels amongst mangroves where their development proceeds through about 18 post-larval moults until they become immature adults 6-/2monthslater. The adults then migrate to deeper offshore waters and breed: they may live a further 6-18 months. The migration and breeding may be triggered by reduced salinities associated with high rainfall during the wet season. In seasonal climates there may be a single breeding season but in some regions there may be two or more peak breeding periods. Some penaed prawns do not require brackish inshore water for juvenile development, and it is not certain whether other species require shallow, mangroves or only the brackish inshore water. The prawns are omnivorous, feeding on live and detrital materials, much of which is derived from sea grass or. mangrove food web**s**

Prawns are most active at night, and at high tide, burrowing into the soft sediments at other times and to escape predators. Adult

Capture of Prawns

Prawns may be captured by a variety of procedures, including trawl nets, hand nets, spears and by hand. Trawls are most suitable for offshore sampling at depths greater than 1-2 metres. For research purposes a trawl of about 2m width, 50 cm height, and 3-5 *i*m length (with 1cm mesh net in the forepart of the net and 0.5cm mesh net at the back), raised about 10 cm from the substrate by *skids* may be towed behind a small boat at about 3 knots (1.5 cm/sec). Larger nets are used by commercial prawn fishing boats. Trawling, as with other trapping procedures, is liable to miss a proportion of the smaller, more active and buried prawns. Trawling is most effective at night.

In shallow inshore waters, such as sea-grass beds and mangrove channels, prawns may be captured with hand nets used to scoop up prawns. In clear water, prawns may be captured at night using lamps (the eyes of the prawns acting as reflectors), the prawns being caught with scoop nets, spears or by hand. The efficiency of the technique relies upon the skill of the collector. Sampling may be conducted along known lengths of the mangrove channel or as a transect across more level areas.

Push-nets may be used in shallow water (20-60 cm deep) on clear firm sandy substrates. These are similar in principle to a trawl net but the net is pushed by a person walking behind and the net has two tails with room to walk batween them. This type of net is described by Ailey (op 81-82, 286-290 <u>in N.A. Holme and A.D. McIntyre</u>, I.8.P. Handbook 16).

Tagging Prawns

Like other crustaceans, the frequent moult's complicate the procedures for marking or tagging prawns. For short-term capture-recapture studies it is possible to put paint (acrylic enamel) or waterproof ink numbers or or to glue tags onto the carapace (e.g. epoxy resin g marks on the (dnied) carapace . Prawns are generally rather small, which makes many long-term taggging procedures *Jifficult* but the cuticle is generally transparent and this enables the use of internal tags which are not affected by moulting.

Several types of internal mark may be used, including: '

- a) injected colour ink spots which form a coded sequence.
- b) magnetic wire strips with a recorded number: but these may need sophisticated equipment to use and may require excision of the wire.
- c) colour coded flexible rods placed beneath transparent cuticle.

C This technique has been successfully used on cave dwelling crayfish measuring as little as 7.5 mm Carapace length. A piece of monofilament nylon fishing line 1.3-3.5 mm in length is painted with 3 colour bands (using acrylic enamel, e.g. for toys and model kits). The coded piece of line is placed into a hypodermic needle of slightly larger diameter. The needle is then inserted through the exoskeleton, e.g. just posterior to the sclerotized ridge of the fifth sternite in the crayfish, slightly to one side of the midline to avoid nerves, and pushed forward into the space between the cuticle and the abdominal muscles. The tag is then ejected, using a wire plunger, as the needle is slowly withdrawn. In prawns the tag may be placed beneath a translucent section of cuticle on the dorsal surface, avoiding the heart, dorsal artery and gut.

The procedure takes about 5-10 minutes. The tags and tagging operation do not appear to have any obvious effect on subsequent

growth, moulting, reproduction or survival of crayfish and the tags remain visible for at least 1-2 years. Colour codes may be read from head to tail and the number of individual codes (N) may be calculated as,

 $N = n(n-1)^{K-1}$

where n = number of colours used

k = number of colour bands used on rods.

Ref: Weingarther, D.L. (1982), <u>A field-tested internal tag for crayfish</u> (Decapoda, Astacidea)

Crustaceana, 43; 181-188. (Includes references to other tagging procedures)

In order to study moulting of tagged prawns it may be desirable to mark the cuticle with a painted mark or by clipping a piece from one of the uropods.

Standing crop of prawns

The standing crop may be estimated using a variety of procedures. Since the prawns tend to flee or burrow into the sediment when disturbed by people it is difficult to obtain absolute counts of prawn populations, Instead, it is necessary to use capture procedures.

- a) The simplest method is to use an index of abundance, such as the number caught/area trawled or number caught/area searched. Provided that the technique is unchanged this may provide reliable indices. of abundance on different occasions.
- b) In a small channel it may be possible to significantly reduce the prawn population with a hand or trawl net. If the netting procedure is repeated, in identical manner, several times then the total prawn population will decline and the number of prawns caught on each occasion will decline. A graph of the number of prawns caught on each occasion with the cumulative number that have been caught up to that time may show a linear decline, and a straight line through these points intercepts the axis of cumulative catch at the probable population size. A limitation of this method is that since the prawns tend to hide when disturbed, the decline in capture rate may reflect the decline in the active population rather than the decline in the actual population.
- c) Capture-mark-recapture procedures may provide estimates of abundance in enclosed channels or pools. Since the sampling may be undertaken at night, it may be desirable to use a flourescent paint for marking the prawns, e.g. visible with an Ultra Violet light source. Prawns; especially adults, may migrate considerable distances and, since this

Productivity of prawns

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Recruitment, growth and mortality information is required to estimate prawn productivity. If the prawns have distinct spawning seasons and produce an obvious cohort of juveniles then this may be monitored at 1-2 monthly intervals to indicate the growth in size and decline in number (mortality) of the cohort. Several studies have indicated that prawn populations do typically have one or more cohorts of juveniles each year, although some juveniles may be present at all times.

If cohort analysis is not possible, then long-term monthly monitoring of tagged prawns may be used but this is likely to require very large numbers of tagged individuals because prawns are mobile and juveniles may experience high mortalities (e.g. 50-90%).

Whichever method is used, it is advisable to sample regularly at the same phase of the lunar cycle and the same tide level, preferably a rising tide and a new moon.

A of t of growth rate with size may be used to derive the terms of the Von Bertalanffy equation.

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Pollution of Mangroves

Mangrove ecosystems are liable to pollution from both terrestrial and marine sources. Terrestrial pollutants include a great variety of materials which are accidentally or deliberately washed or dumped into the mangroves, e.g. sewage; storm-water; garbage; industrial wastes; organic fuels, solvents, pesticides, herbicides, and detergents; inorganic fertilizers, and heavy metal salts; and radioactive materials. Marine pollutants enter the mangroves with the tides, and the major types are organic fuels and detergents. Pollutants are too diverse for it to be possible to outline or even generalise about all their impacts on mangrove ecosystems. Some pollutants, especially fertilizers and sewage, increase tree growth but may adversely affect some animals. Pesticides and heavy metals may kill specific groups of animals. Organic fuels, solvents, and detergents may kill much of the fauna and, in extreme cases, the trees as well.

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The major concern about pollutants is their impact on human health, though other species may deserve equal consideration, and two groups of pollutants are considered:

- i) disease organisms, which are generally associated with sewage disposal into mangroves and nearby areas,
- ii) toxic chemicals, which are generally associated with industrial effluent, and agricultural pesticides from adjacent areas.

The methods for analysing pollutants require well equiped laboratories and skilled technicians. Suitable methods are provided in the most recent edition of "<u>Standard Methods for the examination of water and waste</u> water; American Public Health Association, Washington, U.S.A."

and "UNEP (1982 -) <u>Regional Seas;</u> Reference Mathods for Marine,

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It is recommended that standard methods are used so that results are comparable with other studies and health standards established elsewhere.

i) Disease

Pathogenic bacteria may be present in human faeces and these may enter the mangroves either by direct defeacation or via sewage outfalls. Faeces will decompose in the mangrove environment and the products are both used by mangrove organisms and dispersed by the tides. Some mangrove animals, especially filter feeding bivalves, may accumulate faecal bacteria to levels much higher than that in the water and sediments. The risk to humans is, therefore, both from contact with polluted water, e.g. while bathing, and from eating contaminated animals.

Pathogenic bacteria are usually relatively rare and difficult to detect but human faeces contain numerous less dangerous coliform bacteria which may be easily detected and used as indicators of faecal pollution. These analyses involve the culture of potentially pathogenic organisms and should not be undertaken unless the people involved have a proper understanding of the theory and practise of sterile techniques. It is suggested that the analyses are undertaken by trained medical or university technicians.

(1) · Toxic chemicals

Poisonous chemicals generally enter the mangroves accidentally, either by dumping or spillage and overflow from industrial.

processes, or run-off from agricultural land. The pollution may result from a chronic low level discharge or occasional cause overflow more intense events; rain-storms may of industrial water purification plants and may also introduce large quantities of agricultural chemicals. Depending upon their solubility and density relative to the coastal waters, the pollutants may dissolve and disperse or precipitate near the point of discharge. Many soluble pollutants, both heavy metals and organic compounds, will be absorbed onto fine clay particles and thereby accumulate in certain sediments. Some of thes pollutants will enter food chains and many accumulate at various levels, reaching concentrations of 10-300 ppm in living tissues; which are $10^3 - 10^6$ times higher than the concentration in the water. The use of shell fish from polluted waters may be a serious health risk. Studies of these forms of pollution may be initiated either because diseases and ill-health are apparent or because potentially dangerous chemicals are being used nearby or somewhere within the water catchment.

Materials for observing pollutants

Pollutant levels may be determined in four types of materials:

- i) Water
- ii) Sediments
- (ii) food animals, e.g. prawns, molluscs
- iv) Man

Each of these materials offers different advantages and disadvantages. Generally pollutant concentrations increase from water to sediments to animals, and it is technically easier to measure the higher concentrations in animals; especially if concentrations are low.

Since most pollutants enter the mangroves in solution or suspension in water, it is generally appropriate to measure the introduction of pollutants by the concentration in the discharge water or coastal water.

Animals may live and accumulate pollutants for several years, and though it is unlikely that there will be a simple age: pollutant-concentration relationship, they are particularly suitable for measuring infrequent polluting events or very low levels of pollution.

Food animals, such as the molluscs, may be readily sampled (see appropriate section of handbook) at various locations so these are suitable for detecting spatial patterns of pollutants.

Although the aim of many studies is to determine the levels of pollutants injested by people, it is generally difficult to obtain suitable samples (except from hair, nail clippings, or autopsies) and it is often difficult

to determine how much exposure the person has received.

Sediments may accumulate pollutants over many years, and in some situations it may be possible to obtain a stratigraphic record of pollutants. Surface sediments are, however, liable to erosion and redeposition and unless the history and rates of sediment accumulation are known it may be difficult to relate pollutant concentrations in the sediment to the quantity of pollutants entering the ecosystem.

Spatial and temporal sampling procedures

() The extent and severity of pollution may be determined by taking samples of water, sediment, or animals, at the source of pollution and at various distances out to sea and along the coast. The dispersal of pollutants will vary with local conditions and though concentration are likely to decline away from the source, the dispersal may be directional and there may be site; of accumulation. If they are available and widespread, selected species of bivalves may be justed as pollution indicators.

Monitoring of pollution is usually most efficient if the source of pollution, e.g. discharge pipe or creek, is sampled regularly and also during events such as storm-water discharge which may transport unusually large amounts of pollutant.
Analysis of water is more useful than analysis of sediment and animals because the latter represent long-term accumulation.

If there are several sources of pollution in the vicinity, then monitoring may be carried out at sites where people are likely to be at risk, e.g. bathing, or fishing sites, rather than at the sources.

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Seminar/Workshop on Utilization and Management of Inshore Marine Ecosystems of the tropical Pacific Islands, Suva, 24-30 Nov., 1979.

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SPREP HANDBOOK OF FIELD METHODS FOR STUDYING PACIFIC COASTAL ECOSYSTEMS

ECONOMIC EVALUATION OF MANGROVE RESOURCES AND EXPLOITATION INTENSITY

Planners and developers frequently want to place economic values on natural resources so that they can establish the probable costs and benefits of different patterns of land-use. This might seem to be a simple procedure: estimate the standing crop or annual production of the species and calculate the market value of this quantity of resources. In reality the situation is more complicated because both the quantity and yield of resources will vary with the level of exploitation. Natural biological resources are not the same as either mineral resources, which are non-renewable, or agricultural resources, which are carefully nurtured so as to sustain yields. Natural populations, as in the mangrove ecosystem, may be exploited at various levels of intensity, varying from time to time and place to place. Generally, but not always, the largest individuals of a species are of greatest value, and as exploitation increases these individuals are harvested. This does not necessarily reduce the yield from an area, there may be more smaller individuals available when the larger individuals have gone. There may, however, come a stage at which so much of the population is being harvested, including quite small individuals, that the total yield from an area declines: this is the situation known as over-exploitation. If a biological resource is being overexploited it would be better to reduce the intensity of harvesting since this would cause an increase in the total yield.

The economic value of a particular resources may be represented by the present yield but this may not be a good indication of the optimum Yield which could be sustained. There are many examples of commercial ¹

fisheries which have been overexploited - not only has the total catch declined below an optimum level, but the catch per unit of fishing effort (e.g. trawling time) has fallen even more severely causing the collapse of the economic fishery for several years.

To determine the economic value of mangrove resources it is necessary to know the optimum sustainable yields. This is not easy to determine. Since populations fluctuate from year to year as a result of a variety of natural processes, it is difficult to establish the sustainable yield empirically by varying the level of exploitation. Generally it is desirable to harvest a species at a stage near its maximum growth rate, but it is also necessary to ensure adequate reproduction, so it is wise to only harvest individuals larger than the minimum size for reproductive maturity.

If these rules are applied then the population is likely to persist and fishing yield may approximate the optimum sustainable yield.

Most coastal animals produce very large numbers of eggs and mobile larvae, and even though the population may be overexploited locally, the mobile larvae may recolonize these areas. If it is difficult to enforce a minimum size for harvesting, then it may be possible to create a series of reserves in which harvesting is rigorously controlled: these reserves then serve as centres for re-colonization of adjacent areas. If the level of exploitation is low, then there is no need for any external control of exploitation.

Decisions based upon survey results

If planning or economic decisions are to be made on the basis of a survey, then it is important that both the accuracy and precision of the survey are known. Accuracy can only be acheived and recognised by using suitable procedures which do not give biased results: accuracy or inaccuracy . cannot be recognized from the results unless they are obviously deviant, from accepted values. The methods, calculations, etc. should then be checked.

The precision of a survey results are usually given by the standard deviation (s), standard error (SE), the 95% confidence interval (C I) or 95% confidence limits (\bar{x} - CI to \bar{x} + CI). These statistics indicate the spread of the observations and the uncertainty of defining the true mean value of the resource. Although the mean (π) or total (T) quantity of resources may be the desired statistic for evaluating resources may be extended and it is often desirable to base resource evaluation on confidence limits rather than mean values. The confidence limit to be used for decision making depends upon who is taking the risk and the costs involved.

For example, in a mangrove timber survey it may be so desirable to base the decision on whether to-log an area of forest upon the lower 95% Confidence limit rather than the mean timber volume/ha. This means that there is a very low risk (about 1 in 40) of deciding to log the area when in reality it does not have as much timber as expected. If the mean were used, then in 50% of cases there could be less timber than expected. If logging costs are more or less constant per unit area then it may be essential that there is a minimum amount of timber per ha.

If the decision involves compensation payments for damage or destruction of resources then it seems reasonable that the risk of an incorrect decision should be borne by those liable to pay the compensation rather than the in 'victim'. In this case the compensation should be determined accordance with the upper 95% Confidence Interval of the resource abundance since this will minimise the risk that the compensation is inadequate because of a lack of precision in the survey.

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2. Resource Surveys

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2.A General objectives

As discussed in section 1.8, mangrove resources include not only the economically important species but also the other components of the ecosystem which they depend upon. Since virtually all the living and nonliving components of the ecosystem interact, there are a great number and diversity of 'resources'. It is, however, impractical to attempt to survey all these resources.

The first stage of a resource survey is, therefore, to decide what are the precise objectives of the survey. Once the objectives are clear it is possible to define which resources need to be surveyed, what type of information is required, and what methods are appropriate.

It is important to recognise that there are many objectives when conducting a survey; some are political or economic (at personal, corporate regional, national and international levels), while others are scientific or technological him luding the hardvesting of resources). Generally, a survey must satisfy several political, economic and scientific objectives and is constrained by these different objectives. Scientific objectives tend to need a great deal of information and require a great deal of expenditure whereas the agencies granting funds for surveys seek the maximum of generalisation for a minimum of funding. The compromise between what is considered to be scientifically desirable and what is economically possible dictates much about the scope and methodology used in a resource survey. While it may seem desirable to know as much as possible about resources, the aim of an efficient survey is to obtain $\gamma_1 33$

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2.8 Scientific objectives

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Biological resource surveys are generally concerned with:

either i) The resources of a region,

or ii) Variation in the natural distribution of resources,

or iii) The impact of external factors on resources, e.g.

exploitation, pollution, isolation, etc.

These resources are generally measured in terms of one or more of

the following:

i) Presence/absence of a species

ii) Standing Crop; the amount of the resource present at a particular

time, usually measured as biomass or the number of individuals. Indices of standing crop are often used.

Production; the amount of energy or blomass incorporated by

a species over a known time period, especially the amount of useable biomass.

Production is estimated by various techniques, generally involving combinations of observations on metabolic rate, energy or food intake, growth, and reproductive output. Laboratory or field estimates on a few individuals are generally combined with Standing Crop estimates to provide Production estimates for the population and ecosystem.

iv' Bustainable Yield; the amount of the resource, either numbers or biomass, that can be repeatedly removed from the ecosystem without causing a long-term decline in the abundance of the resource Maximum sustainable yield usually occurs at only low-moderate levels of exploitation.

Sustainable yield is most satisfactorily estimated by repeatedly harvesting the resource at various levels of intensity and determining the effect and yield on the Standing Crop . It is difficult to estimate sustainable yield in small areas if there is much migration from other areas. There are also a variety of mathematical models which may be used to predict sustainable yields on the basis of population and individual growth rates and standi crop parameters but care must be taken to ensure that the assumptions made in 3. Survey procedure: basic considerations and decisions

When conducting a biological resource survey it is necessary to consider and make decisions about δ major aspects of the survey <u>before</u>the survey commences. The decisions made in the planning stage will determine the outcome and value of the survey, and deserve a great deal of careful thought.

- i) is the objective descriptive, e.g. total resources, on analytical, e.g. natural variation and variation associated with external facto
- What is the boundary of the study area? Is the survey supposed to relate only to the study area or is it supposed to be repres tative of a larger area of mangroves? It usually cannot be assumed that a single study area is representative of the mangro of the region, and several sites may be required to ascertain their variability: how should these sites be chosen?
- III) Which organisms are of interest? As discussed above, most mangrove ecosystems contain a great diversity of organisms which behave in different ways such that a variety of methods are required to study them. It may be tempting to attempt to survey every species which turns up but the effort required to make this worthwhile is almost certainly prohibitive. One practical strategy is to seek different types of information about different species, e.g. record presence/absence for all species in the locality, and record standing crops, productivit) or sustainable yields from only a few economically important species or groups of species.

Species with similar habitats and behaviour may often be sampled together so that each survey technique may yield estimates of standing crop for a group of species e.g. tetrestri plants; burrowing filter feeders; burrowing mobile species; fish: and plankton. -25

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iv) What type of information is required? Is qualitative or quantitative information required? Is it enough to know that a species occurs in the mangroves, or feeds in the mangroves, or breeds in the mangroves, or is it necessary to estimate how many individuals occur, at what density, at what time(s) and capable of yielding what weight of resources or sustained yield?

Since the methods and results of the survey depend very much upon the answers to these questions they should be given very careful consideration. It is always tempting to seek easily obtainable information about the species without fully considering whether it is the required information. Some information may be obtained with unnecessary precision while other useful information is not collected. There is no guide with which to answer these questions except a clear view of the overall objectives of the survey.

- v) How accurate does the survey need to be? In this context accuracy means how close the survey results are to the real(or true) results. In biological surveys several types of accuracy may be involved:
- a) Taxonomy

In both qualitative and quantitative surveys the accurate identification of species may be very important or it may be of minor importance; e.g. while it may be important to identify the species and, perhaps, the variety of an economically important organism. e.g. fish, crabs, shellfish, it may not matter which phytoplankton species occur provided they are present in sufficient quantity.

Specimens should always be taken, preserved and identified by a competent authority. These specimens should then be lodged at a suitable museum or herbarium, preferably with duplicates lodged at a local institution.

b) Biased richods

In quantitative surveys the estimates of abundance may deviate considerably from the real values if the survey methods are biased. Biased survey methods mean that certain individuals are more, or less, likely to be sampled than should occur by chance. ¹A biased survey method may either overestimate or underestimate the resources, and it is important to realise that this bias cannot be detected from the survey results. Bias can only be avoided, or minimised, by applying suitable methods. For slow moving or sessile organisms this generally involves random quadrat sampling. For mobile species this required unbiased trapping and perhaps mark-recapture methods. These procedures are described in various textbooks, e.g. Southwood, 197%; Chapman, 1976

It is important to check that the methods are not biased and, if necessary, correct the bias or determine correction factors to eliminate bias. For example; if burrowing species are estimate from the number of burrows then it is essential that the ratio of burrows to live individuals is also determined; if fish are estimated using nets or traps then it is essential that the efficiency of the nets or traps for the different sizes and sexes in the population is determined.

c) Biased estimates

Bias may also occur during analysis of results. One type of bias is 'bias of the estimating procedures' in which certain calculations tend to give biased results e.g. the ratio of two estimated and unbiased values is biased. These types of bias are generally minimised by taking large numbers of samples. If a mathematical model of growth or distribution is assumed to apply but does not, then, any calculations based on the model will be biased. Bias of this type may be corrected if it is recognised.

d) Error - Bias may also occur through incorrect application of the method or incorrect calculations: what is commonly thought of as accidental error.

vi) How precisely should the surveys results be estimated?

a) <u>Precision</u> Precision is measurement of the certainty with which a result is known, and is generally measured by statistical indices such as the Confidence Interval. It is important to distinguish between precision and accuracy: a biased survey

method may produce very precise results but they will be inaccurate, and, conversely, an unbiased survey method will produc accurate results but these may not be very precisely known. In general, precision is achieved by making many independent observations (samples) and this may be expressed by the equation:

precision 🗠 (Number of Samples) 5

e.g. To double the precision of the estimate, the number of samples must be quadrupled. Very great precision requires a very large number of samples while moderate precision may be achieved with few samples.

Great precision is a costly aim in surveys and may be unwarranted if there is much doubt about the accuracy of the methods. If the methods are thought to be accurate, or at least repeatable, then a large number of samples is the only approach to obtaining precise answers. Generally it is desirable to take many small samples rather than a few larger samples since this contributes most to defining natural variation and maximising precision.

Minimising error by stratified sampling. In the introduction (b)(1.E) it was noted that mangroves are a transitional ecosystem and may be treated as a gradient or series of zones with differen environmental characteristics and different species composition. If the mangroves are treated as a single zone then there is a great deal of natural variability in the environment and species which may be sampled. This variability means that very large numbers of samples are required to obtain precise estimates and that only major differences between two sites, or the same site at different times, will be detected. A stratified sampling scheme is a useful technique for reducing variability: each of the zones is treated as a separate sampling area, and the results may later be combined to obtain total values. Stratified sampling may reduce the number of samples required, increase the precision achieved, and **also** improve understanding of the mangrove ecosystem.

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There are several strategies for allocating samples and sampling effort to each zone, the simplest are to allocate samples in proportion to the area of each zone, or in proportion to the population in each zone (if the total population size is wanted).

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If the survey is only concerned with one study area then the definition of zones is not critical; they could usefully be defined by altitude, distance across the mangroves zone, by dominant species, etc. If the study involves more than one site, then the equivalent zones can only be usefully compared if there are clear criteria for defining each zone in all areas



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4. Survey procedure: practical considerations

At this stage there should be:

- i) A general objective (2.A)
- ii) A scientific objective which satisfies 'i' (2.8)
- iii) The scientific objective should be clearly stated in terms of the basic considerations (3)

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iv) The survey procedure should now be implemented.

4.B Definition of Study Areas

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If the objectives of the survey are to provide accurate estimates of mangrove resources in the entire region then the entire area of mangroves is the study area. In general, survey results only apply (with known accuracy and precision) to the region from which the observations represent a random sample. The survey results may be valid outside the study area but this assumption may be invalid and extrapolation to other areas is essentially a guess. If the objectives are to monitor changes in the resources, or to examine variation within the region in relation to factors such as exploitation or pollution, then other strategies may be adopted for defining more restricted study areas.

A general principle for defining study areas is that for every source of variation to be considered there should be at least two study sites; one with the source of variation expressed (e.g. exploitation, pollution, or an environmental zone), and another 'control' site. Within each site it is necessary to take replicated samples so as to determine the natural variation within each site which can be compared with the variation between sites. This logical approach to the analysis of sources of variation is apparent in the statistical technique of Analysis of Variance (ANOVA), which is generally the most appropriate method for analysis of the results. For this approach to be efficient, each site should be surveyed with an equal number. (perhaps 3-10) of observations. It is recommended that a statistician is consulted about the design of the survey to ensure that it will yield the desired information (see also Green, [413])

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If some factor, such as levels of exploitation, is to be examined then the exploited and non-exploited sites should be similar in all other respects. It may be necessary to examine several pairs of matched sites to establish clearly that exploitation is associated with particular differences in the mangrove resources. If a process is to be monitored then both the sites must be monitored.

If the study is concerned with estimating resources in the whole region then the samples must be randomly located within the region, with or without stratification.

If the survey is intended to reveal differences between areas which differ is factors such as exploitation, pollution, substrate, aspect, to, then the study areas may be relatively small and the main requirements are:

 The whole of each study area should be properly classified according to the external factor.

ii) The study areas should be paired to control for other factors. $f_{a}d_{a}$ were g_{a} . The simplest design for these sites is rectangular plots with one axis along the gradient from land to sea; the width is not so easily defined but should exceed any scales of pattern in the ecosystem, e.g. it should exceed the diameter of the largest trees by a factor of at least 5x. Plot width of 50 - 100 m are probably acceptable in most circumstances.

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4.C Stratified sampling within Study Areas

The use of stratified sampling to reduce variability in survey results has been discussed above (3.vi.b) and this procedure is evidently and applicable to the mangroves, which show distinct biological environmental gradients from sea to land. Each species tends to vary in abundance along the gradient, and by dividing the gradient into zones the abundance within each zone may be estimated more precisely, and the overall estimate of abundance is also more precise. The patterns of abundance vary from species to species and there is not a single pattern of zones that can be applied to all species. Similarly, the number of distinct zones that could be defined varies from species to species.

Although the mangrove species are clearly varying in a zonal fashion, the optimal zonation is not readily defined by casual observation and is not fully apparent until the area has been surveyed. While zonation cannot, therefore, be determined before the survey, it is possible to record the position of the sample site along the gradient and to define zones subsequently. It is generally desirable to divide the gradient into a number of equal width zones <u>(e.g. subsequently. It-is-generally</u> desirable-to-divide. the gradient-into-a-number of-equal width zones (eg 2-10 zones) and to sample equally and randomly within each zone, such that overall the samples are fairly uniformly apread along the gradient and the position of each sample is known.

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4.D Sampling for Presence/Absence of Species

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presence/absence information may be collected in various ways; by systemmatically or randomly searching the area for species, or by using more thorough sampling techniques which give information for defined sample areas or volumes. Casual surveys of the area are likely to miss the rare, cryptic and more mobile species which avoid the observer, so even if the objective is no more than a species list it is desirable to use a sampling scheme such as that for standing crop.

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4.E Sampling Standing Crop

Appropriate sampling techniques vary with the mobility, behaviour and habitat of an organism. At one extreme are the rooted plants, epiphytic plants, and epiphytic animals which are mobile as juveniles but remain fixed to the substrate during the rest of their life cycles. Burrowing and crawling species are mobile throughout their life cycles but many species do not travel far once the adult stage is reached. At the other extreme are the fishes and plankton which are highly mobile (actively or passively) and must be considered as migrants.

There are two major approaches to sampling;

 immobile species are sampled by examining known areas (i.e. quadrats) or volumes of substrate (or water) and recording the presence and/or standing crop of species in each sample.

ii) Mobile species, which may be defined as those that move away from an observerbefore they can be accurately recorded, are sampled by trapping.

A mobile species may be estimated by various procedures:

- a) A total count within a reasonably large area; large enough to avoid the problem of animals moving away from the observer.
- b) The whole population in an area may be trapped, using meta or poisons.
- c) The decline in captures over a series of repeated trapping attempts may be used to plot a graph of 'Captures in latest trial' against 'Total Captures to date': the intercept on the axis of the Total Captures to date'should be close to ' the total population.

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d) Capture-mark-recapture techniques are potentially applicable to all mobile species but they make several assumptions about the behaviour of the species, mortality, natality, and migration. These techniques may be inappropriate for mammals and some other vertebrates but they seem to be successful for many/invertebrates.

There are many other sampling techniques but few are so widely applicable and most make more assumptions about the distribution or behaviour of the species.

Quadrat Sampling

i) Random locations. Within the study area, or each zone of the study area, quadrats should be located randomly (using random numbers to choose co-ordinates or the serial number of a location) so that there is an equal probability of each part of the area being sampled.

The location of guadrats in randomly chosen places is difficult. especially so emongst the tangle of mangrove trunks and roots. In practise it is much easier to locate quadrats in a haphazard fashion, not truly randomly, but in a way that is easy to locate quadrats and does not introduce bias by favouring certain areas. The simplest procedure is to locate quadrats along transect lines across the study area, either within tidal zones, or across the zones. The quadrats may be regularly or randomly spaced along the transacts and should not be unnecessarily grouped in one part of the study area.

i) As discussed in section 3vi, precision and also the robustness of many statistical methods depends on the number of samples, and generally it is desirable to take many small quadrats rather than a few large ones. The minimum size for a quadrat should be a size at which only a small proortion of individuals fall across the boundary of the quadrat; generally the quadrat area (or volume) should be at least 20 x the area (or volume) of the organism.

(Another guideline is that the mean number of individuals per quadrat should be about 5-10. The variance yielded by quadrat sampling varies . with the pattern of distribution of the organism and it may be desirable to compare several quadrat sizes, during a pilot study, to determine
least variance and which size is the most economical to sample. Although minimising variance may be worthwhile if populations of a simgle species are to be estimated with great accuracy, quadrat sampling is often used to sample several species, and in this case it is unlikely that they will have similar patterns of variance, so no single quadrat size is optimal for all species.

For the mangrove trees, quadrats of about 10m x 10m to 20m x 20m are desirable, while the burrowing species of crabs, bivalves, etc. require quadrats of about 15 cm x 15 cm $\frac{15}{500}$ cm x 504m. Number of quadrats

Generally, the number of samples required (N*) may be estimated as:

 $N^{\times} = 1/E^2$, where E is the acceptable error about the mean

expressed as the ratio of the acceptable standard error (SE*) to the mean (\tilde{x})

E = SE*/X

e.g. An acceptable error (E) of 0.10, (= 10%) yields;

$$N = 1/(0.10)^2 = 100$$

The confidence interval (CI) is readily derived from the acceptable error, or vice versa;

 $CI' = \pm d.SE^* = \pm d.E.x$, where d'is the unit standard deviation of a normal distribution.

If E = 0.1, $CI_{95\%} = \pm 1.96.0.1\bar{x} = \pm 0.2 \bar{x}$

A more accurate estimate of the required number of samples may be derived from

 $N^{*} = \left(\frac{d.S_1}{E.\overline{x}_1}\right)^2$

where \bar{x}_1 is the mean and S_1 the standard deviation based on a pilot survey of the area. 'd' is usually set at 1.96 to give 95% Confidence. in the estimate. 'E' is usually set between 0.1 - 0.2 depending on the needs of the survey.

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iv) <u>Stratification</u> The study area may be stratified into about 2-10 zones, both to ensure more or less uniform density of sampling and to reduce variability in the results. Zones may be defined using: environmental criteria, eg. height, biological criteria, e.g. dominant species, arbitrary criteria, e.g. distance from sea. Zones of equal width, recording the distance of each sample from the sea, provides the simplest approach. Quadrats may be allocated to zones according to several different procedures, a) In proportion to the area of each zone. $n > h = n > k \cdot \frac{Ah}{A}$, where nth = Required number of samples in zone h, Ah = Area of zone h, $A = \sum_{i=1}^{n} Ah = Total Area.$ b) In proportion to the population of each zone $n \times h = n \times \cdot \frac{Nh}{N}$, where Nh = Number individuals in zone, h, N = S Nh = Total Number individu

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- 26 -c) If the effort (cost) of obtaining each sample is similar in all zones, and the variance · differs in each zone, then $n \times h = n \cdot Nh \cdot Sh$ ZNh. Sh where n = total sample size Nh= Ah/a = Area of Zone (Ah)/Area of quadrat (a) Sh = Standard Deviation in Zone h. n.b. This provides an optimum allocation of effort, and requires a pilot study to determine Rh. This is known as Neyman allocation. If the effort of obtaining samples in different zones differ, then there are other allocation procedures but it seems unlikely that this will be important for most mangrive Surveys. -----750

- 27-Procedure for combining stratified samples to obtain results for whole study area: a) Standard Error for each zone; SEzch $SE_{\overline{x}h} = Sh (n_h)^{\nu_2}$ where, Sh = Standard Denation in zone h Nh = Number of quadrate in zone h. _____.. b) Total population for whole study area: $X = \Sigma Nh. \overline{z}_h$ where, Nh = Number quadrats in zoneh, = Ah/a Jch = Mean denisty in zoneh. c) Overall mean; X $\overline{X} = X / N_{, where } N = \overline{\Sigma} N h.$ d) Standard error of total population ; SER $\frac{5E_{\bar{x}}}{N} = \left(\frac{\Sigma Nh^2 SE_{\bar{x}}h^2}{N}\right)^{-1/2}$ e) Confidence Internal 95% = ±1.96 SEX

V) Information The information required from each quadrat may be : presence / absence, number of individuals, sizes of individuals , weights of individuals (fresh or oven-dry), total weight of individuals (fresh or oven-dry), energy content of individuals.

While some of these measurements are easily made in a mangrove swamp, others require laboratory equipment (e.g. ovens, scales). It is, therefore, convenient to take Simple size index measurements in the swamp (e.g. length) and relate these to calibration curves based on a sample of the population. A calibration curve generally requires 30-100 individuals, representing as wide a range of sizes as possible, upon which both parameters (e.g. length & weight) have been accurately measured. A graph and/or regression line enables the estimation of the desired quantity (e.g. weight) from the field measurements (e.g. length).

vi) Transformations

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Although many statistical procedures assume the observations have a normal distribution, this assumption is not critical for minor deriations eg. slight skewness. The distribution of many species is, however, highly skewed; some quadrats contain many individuals while many contain very few; In this case transformation of the data is desirable. A useful transformation i log(x;+1) where r: is the parent f

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- 29-The term (xi+) is used instead of (2ci) to avoid the problem which arises with zero records Once the statistical analysis is complete the summary statistics should be transformed back to their original form $log_{10}(2c'+1) = 10^{(2c'+1)}$ nb. a) Subtract 1.0 from the results to obtain (>c') instead of (x'+1). b) Confidence Internals, standard deviations, etc will not be symmetrical about the mean Calculate the confidence limits 5c-d SE to 5c+d.SE. before reverting to the original state of the data.

Mark-Recapture methods:

The mark-recapture methods are based on the principle that individuals may be captured, marked in some way, and released without causing them any significant damage or affecting their behaviour. After the lapse of enough time for the marked individuals to mix in with any other individuals, the animals are trapped again, making sure that the technique does not favour or previously marked individuals. The proportion of marked individuals amongst those captured should have $\begin{pmatrix} m_{12} \end{pmatrix}$ the same proportion as the number of marked individuals released to $\begin{pmatrix} N_{1} \end{pmatrix}$ the total population at the time of release: $\frac{m_{12}}{C_2} = \frac{m_1}{N_1}$, so, $N_1 = \frac{m_1C_2}{m_12}$

Various adaptations of this basic principle allow population changes to be monitored over a series of Mark-recapture periods.

b) The assumptions upon which these methods are based vary slightly with different marking schemes. The basic assumptions are that:
a) Marks are not lost
b) Marks do not cause a change in the survivorship on behaviour of individuals.
e.g. Marked individuals may have higher mortality. Marked individuals may avoid recapture.
If these biases occur, then it may be possible to estimate the bias and apply correction factors:
c) Marked animals mix randomly with the other individuals is the possible to estimate the bias and apply correction factors:

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- 31 -It is important that the assumptions are checked carefully. This may be possible through careful monitoring of individuals, perhaps in enclosures, and by comparing the results of different survey methods. Most trapping techniques favour a particular size, age, or sex of individual, so the results are really estimates of a sub-population, not the whole population Repeated sampling not only allows a better estimate of population size, it also allows estimates of Longerity, natality and immigration, mortality ---and emigration. It is generally not worthwhile to undertake a repeated (multiple-capture) sampling program merely to obtain an estimate of standing crop. A 素装 great deal of demographic information may be estimate if unique marks are used and recorded for each individual, together_with records of individual size, sex, et Trapping. Any harmiess trapping technique may be used; nets, cages, pursuit, etc, bearing in mind ____ the biases it may introduce into the sampling. The more intelligent the species, the more difficult it is, a not only to trap them but also the more difficult it is to avoid biased estimates of population size. Mammals, birds and some other vertebrates generally learn to avoid or enter traps and therefore may

(10). -32 -There is no simple rule to indicate how many traps are required or how these should be located; this will depend upon the species abundance distribution, the type of trap, and the precision which is required. Precision For a simple capture - mark - recapture survey the standard error may be estimated as $S = \hat{N}_{1} = \hat{N}_{1} \sqrt{(\hat{N}_{1} - m_{1})(\hat{N}_{1} - C_{2})} m_{1} C_{2} (\hat{N}_{1} - 1)$ A pilot I is needed to yield estimates of the standard ever and therefore the required number of marked and recoptived_ individuals. HS a general guideline, for a single capture-markrecapture survey the number of recaptures should be at least 7. It it is suspected that there are N_individuals in the study area and that an equal number of individuals may be captured and marked. $m_1 = C_2 = (7 \cdot N)^{1/2}$ -

- 33 -Since, by definition, mobile species more from one area to another it is difficult to express results on an area basis. This problem arises at two stages of the survey; the placing of trap, and the calculate of results a) If individuals search an area larger than the study area, then the focation of traps in the study wear is arbitrary; the only consideration being that the traps should be effective b) If individuals move about in groups then different trapping procedures may be required depending on the nije of the group, eg size or number of traps. c) Ingeneral, trapping of individuals from a group which reaches an area larger than the study area will give an estimate of the group size and this will refer to their reaching area, not the study area. d) It individuals are solitary and rearch an area larger than the study area, then the recaptive rate will be no low (ramating of - 2?) that reliable estimates of the population rije cannot be made e) If individuals, or groups, are territorial or occupy different parts of the study wear, then the traps should be located throughout the area 17 15 then deviable to record the locations at which individuals are released and recaptured to determine mobility in the population. Either a grid or random lacation of transman he used

34 The capture-mark-recapture methods are generally less precise than quadrat methods for estimating population size in a particular area. This is partly a consequence of the mobility of the organisms and difficulty of defining the area used by the population, and partly because the method is based upon a ratio in which more than one term is subject to variation nad

R. " Acknowledgements thank Mrs Rahesh Garib for typing I would like to 1 - 40 F this manuscript. í

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