BRITISH ROUGH WINKLES: ASPECTS OF THEIR ANATOMY TAXONOMY AND ECOLOGY.

Thesis submitted in accordance with the requirements
of the University of Liverpool for the degree of
Doctor in Philosophy by Celia Jane Hannaford Ellis.

September 1980.

BRITISH ROUGH WINKLES: ASPECTS OF THEIR ANATOMY, TAXONOMY AND ECOLOGY.

By Celia Jane Hannaford Ellis.

Abstract: The characters used to identify rough winkles were described and the following assessments of their taxonomic usefulness were made: 1).Pallial oviduct. The structure is diagnostically different in rudis and arcana females. 2).Penis. The form is diagnostic in nigrolineata (Heller,1975a) but not in rudis and arcana and also possibly not in neglecta (cp. Heller,1975a). 3).Prostate. The appearance can aid in identifying rudis and arcana males, but is not diagnostic. 4).Ciliated field. The size difference between rudis and arcana is diagnostic and is used to identify males. 5).Sub-opercular pattern. This is useful in identifying neglecta only. 6).Radula. The radulae show cusp number decline and, in some species,changes in cusp shape with age. They are of little use in diagnosis. 7).Shell. L.neglecta and nigrolineata shells have diagnostic features (Heller,1975a),so these species can be identified on this basis. L.rudis and arcana shells are too similar to be used. By using combinations of these characters most animals can be diagnosed.

Various aspects of the biology and ecology of the four species were examined: 1).Sexual dimorphism was observed in all the species. Females are generally larger than males, usually maturing at larger shell heights and are sometimes more globose. In nigrolineata size dimorphism related to shell colour was observed; yellow-shelled animals generally grow larger and mature at larger sizes than white-shelled. 2).Ontogenetic changes in shell colour pattern of neglecta were observed: Young neglecta usually have banded shells, older animals' shells are usually tessellated.3).The reproductive seasonality of the four species was examined. L.neglecta,arcana and nigrolineata show marked periodicity, but rudis shows little. 4). The population size structure and composition changes of neglecta suggests that this species is an annual.5). The spatial and temporal zonation patterns of rudis and arcana in a community at Porth Swtan were examined. For much of the year the species seem truly sympatric; but arcana adults in breeding condition were found to migrate down to the top of the barnacle belt. 6). Spatial zonation differences were found between the species of the small-winkle community in the barnacle belt,though there were wide overlaps between species. Temporal effects may also be important: The breeding periodicity differences resulted in some staggering of juvenile production by the different species, so that the juveniles were of slightly different size ranges. This could be important in resource partitioning. Male neglecta were found to zone higher than females.

The intra-specific variation in rudis is particularly striking and two types commonly occur in Anglesey. One type is abundant in boulder habitats, the other in crevice habitats. Unusually at Porth Swtan both types (living in their respective habitats) can be found and appear to retain their identity in spite of their proximity. At another site a cline between the forms seemed to exist. The observations could support arguments for the existence of phenotypes,ecotypes or sibling species. Though some recognition of the different forms is desirable for ecologists, before any formal taxonomic recognition is given to any form of rudis (or any of the forms of other species), experiments, to discover if there is any genetic basis for the observed variation, should be undertaken.

CONTENTS

GENERAL DISCUSSION

GENERAL INTRODUCTION

In 1975 Heller resolved the British rough winkles of the Littorina saxatilis complex into four species, L.rudis, L.neglecta, L.nigrolineata and L.patula, distinguishing them on a combination of shell and anatomical characters. Though this classification was mainly welcomed by littorinologists, the distinction of patula from rudis has been questionned, since Heller's diagnosis of this species did not make it possible to separate a high percentage of specimens encountered into one or other species. This problem has been highlighted in Raffaelli's recent paper (1979). In this he concludes that the characters used by Heller to define patula are not sufficiently distinct from those of rudis to warrant its separation as a species, and that patula must be regarded as a synonym of rudis. Like Raffaelli's, my early attempts to distinguish patula and rudis also failed in the face of the apparently continuous variation in shell and penial characters. Most ovoviviparous individuals seemed to be intermediate between rudis and patula sensu Heller in at least some characters and could not be assigned to one or other species with any degree of surety. Thus, I, likewise came to regard patula as a synonym of rudis.

Concurrently a second taxonomic problem emerged. Females were found that exhibited pallial oviduct structure like that of nigrolineata, mariae and obtusata i.e. of oviparous design, which is quite distinct from ovoviviparous pallial oviduct structure. These females combined oviparous reproductive mode with shell characters that ranged from patula to rudis.

For a while it appeared that the two problems were unrelated, that patula was probably a variety of rudis and that a new, second oviparous rough winkle existed which (excepting Seshappa's observation (1947)) had evaded previous discovery. However it soon became apparent that the oviparous winkle was a widespread, abundant species and had influenced Heller's description of patula.

Once the character of the ciliated field had been noted and used to separate males, the penial characters of the males corresponding with the oviparous females could be identified. These proved strikingly like those noted for patula by Heller. Also re-examination of Heller's material at Liverpool revealed substantial numbers of the oviparous form and these must have been used in his diagnoses. Furthermore, oviparous specimens were sent to Dr. Heller which he returned labelled L.patula for those with asymmetrical shells and 'L.unknown' for the remainder. Thus it seems probable that he used primarily the shell and penial characters for distinguishing rudis and patula, and at such sites as Abraham's Bosom where rudis and arcana exhibit very different shell types, the penial differences would support such a division.

Despite the probability that patula was

described mainly from individuals of the oviparous species, it would have been inappropriate to re-describe patula as an oviparous species; the patula form of rudis undoubtedly exists, in as much as there are rudis individuals with the penis form and shell shape associated with patula by Heller (which, incidently he derived from Jeffreys' (1865) description and not the original description published in 1844 by Thorpe). Furthermore none of the other variety names in existence could be used as these all relate to shell types, and might have been secreted by individuals of either species. So I described L.arcana, in 1978, as a new species of which the main diagnostic characteristic is its oviparity, A fuller description appeared in 1979 (Hannaford Ellis 1978,1979).

Though the separation and description of arcana is of some importance and took a considerable amount of time, the work undertaken was not intended to be primarily taxonomic. The main objective was to examine the ecology of the then newly-described rough winkles generally.

Section 1 deals mainly with the identification of rough winkles to species and while especial attention is paid to the separation of rudis and arcana, it also covers the identification of juveniles which is usually omitted from species descriptions. Identification of juvenile and immature individuals was essential for the ecological work undertaken in the subsequent sections. Many of the characters which are useful for distinguishing the species are anatomical and the structure of the oviduct in particular is described in detail. Some new observations were made of this and also certain other structures, such as the ciliated field, sub-opercular patterns, prostate and radula, and these are described here.

Section 2 covers aspects of the autecology of the different rough winkle species, looking at sexual dimorphism, ontogenetic changes in the colour patterns of neglecta shells, reproductive seasonality and the changes in the structure of neglecta populations.

Section 3 looks at the synecology of rough winkles. Considerable overlaps in habitat are apparent especially rudis with arcana and neglecta with juveniles of other species and questions arise about the niche separation.

In both sections 2 and 3 some of the ecological differences between the look-alike species rudis and arcana are described which support the separation of these two species.

Section 4 is rather more speculative. It considers the variability observed in L.rudis, perhaps the most variable of the rough winkles and points to some of the many problems encountered with this species.

OUTLINE OF THE CURRENT CLASSIFICATION OF BRITISH WINKLES

The winkles of British shores are currently referred to one of eight species which between them exhibit three modes of reproduction.

Littorina littorea (L.) and *L. neritoides* (L.) both go through a planktotrophic stage in their life history. Eggs are released into the sea in capsules, from which young hatch as veliger larvae and are free-swimming until they metamorphose and settle.

Four species are oviparous, laying their eggs in egg masses in which they develop, hatching as tiny snails. These species are *L. obtusata* (L.), *L. mariae* Sacchi & Rastelli (1966), *L. nigrolineata* Gray (1839) redescribed by Heller (1975a) and Sacchi (1975), and *L. arcana* Hannaford Ellis (1978). *L. obtusata* and *L. mariae* were formerly not distinguished and were called either *L. littoralis* (L.) or *L. obtusata* (L.). The latter name, having page precedence in Linnaeus' *Systema Naturae*, is now used for the commoner species, (see Goodwin and Fish, 1977).

The other two species are ovoviviparous, retaining their eggs, while they develop into tiny snails, in a brood pouch. These are *L. neglecta* Bean (1844) redescribed by Heller (1975a) and *L. rudis* (Maton, 1797).

Four of these species, *arcana*, *rudis*, *neglecta* and *nigrolineata* were, prior to Heller's paper, classified as one species, *L. saxatilis* (Olivi, 1792). The common name given to *L. saxatilis* in Britain was the rough winkle. Consequently these four species are often referred to collectively as the rough winkles. However it should be realised that this is not a natural grouping within the British winkle species. The reproductive mode of *arcana* and *nigrolineata* groups them with the flat winkles, _mariae_ and _obtusata_, rather than the ovoviviparous species _rudis_ and _neglecta_.

SECTION 1: USE OF COMPARATIVE ANATOMY AND SHELL CHARACTERISTICS FOR IDENTIFICATION OF BRITISH ROUGH WINKLE SPECIES.

Identification and separation of winkle species has, until comparatively recently, relied primarily on shell characteristics. Many species, subspecies and varieties were named on the basis of shell characters and little regard was paid to the 'soft parts', the animals themselves, which were all too often thrown away.

It is, of course, very tempting to use principally the shell for specific identification. Shells can be easily examined and measured without necessarily killing the animal, and they are also easily preserved. However accessibility cannot be equated with reliability as a species marker. Speciation can occur without involving a distinctive change in shell characters and conversely separate species may show convergence. Furthermore the considerable variation that exists in shell characteristics within a species can obscure the division between species. For example, amongst the four species that were lumped together as saxatilis, two nigrolineata and neglecta in fact have diagnostic shell characteristics but the distinction was obscured by the wide variation in arcana and rudis shells. Also, ontogenetic changes can further complicate the task of identifying individuals to species e.g. in littorea which is commonly regarded as of uniform shell type, the uniformity is applicable only to adults; juveniles have shells which are totally dissimilar. Shells are none-the-less very useful markers provided the limits of their usefulness are known.

For the rough and flat winkles anatomical characters have been shown to be more reliable for distinguishing the species (Sacchi

& Rastelli, 1967, Sacchi, 1975, Heller, 1975a). These authors have used characters such as the pallial oviduct, penis, radula and pigmentation patterns. Of these, characters relating to the reproductive system have proved the most useful, being at once conservative within the species, and showing marked specific differences. The interspecific diversity in genital organ structure probably derives from the need of the winkles themselves to recognise conspecifics. Differences in structure may act as important isolating barriers to interspecific matings.

The following sections assesses the usefulness and limitations of several taxonomically important characteristics of anatomy and shell, particularly of rough winkles, attempting to distinguish characters as diagnostic, typical or indistinct. It is intended that in conjunction with Heller (1975a) the information given will provide a basis for identification of the vast majority of rough winkles of whatever size. Accordingly most detailed attention has been paid to features of <u>rudis</u> and <u>arcana</u> which were not distinguished by Heller. It should perhaps be pointed out that identification of species cannot always be made on a single character and combinations of characters have to be used for diagnoses.

Six anatomical features are considered, pallial oviduct, penis, prostate, radula, and two characters previously unused in littorinid taxonomy, the ciliated field and sub-opercular pigment pattern. Also, shell characteristics of both juveniles and adults of all British species are described and assessed.

The case for reliable identification of species throughout their age range and in all stages of their reproductive cycle cannot be overstressed. A great deal of work on winkles has been, and is being, wasted through lack of care in identification, or through inability to recognise juveniles or adults out of reproductive condition.

1.1 PALLIAL OVIDUCT CHARACTERISTICS

The pallial oviduct is that part of the oviduct lying within the mantle cavity. In Littorinids, besides its function in transporting the ova from the ovary to the outside world, it also serves as the site of fertilisation, secretes layers of nutritive and protective materials round the zygote, and in the ovoviviparous species provides the protected space in which the embryos develop.

The structure of the pallial oviduct varies with reproductive mode and, corresponding with the modes of reproduction seen in British Littorinids, there are three basic formats of pallial oviduct structure. Since reproductive mode is an important species characteristic, pallial oviduct structure can be a diagnostic character useful in identification of species.

The three reproductive modes are oviparity, ovoviviparity and production of planktotrophic larvae. Two species, littorea and neritoides, exhibit this last mode and since both species can be reliably recognised on the more easily examined shell characteristics the pallial oviduct is relatively unimportant for identifying them. In the case of the flat winkles, obtusata and mariae, which are not always easily distinguished from each other on their shell characteristics, the oviduct structure is of no assistance as both species are oviparous. However, oviduct structure is useful in separating rough winkle species, especially in separating rudis and arcana females from each other. Since rudis reproduces ovoviviparously and arcana oviparously, the pallial oviduct structure differs and these differences can be used for identification.

The following account describes and figures the structure of the oviducts, both immature and adult, of arcana and rudis and compares them. The oviduct structures of these species can be used as examples of oviparous and ovoviviparous oviduct formats and so the descriptions of the structure can be applied generally to the other species which reproduce similarly. Though both types of oviduct have previously been figured in the mature condition (rudis by Fretter & Graham 1962, Sacchi, 1975, Sneli & van Marion, 1979; nigrolineata by Sacchi, 1975) the appearance of the immature oviduct has not been described by previous authors. Since the appearance of the oviparous and ovoviviparous oviducts in the immature condition is as diagnostically different as in the mature, this can be used to separate immature rudis and arcana females. This is of particular importance in identifying arcana. The ability to distinguish immature, as well as mature, arcana from rudis is a prerequisite for any ecological study of these species since arcana has a markedly seasonal reproductive cycle (see section 2.4) and outside the reproductive period the majority of arcana on a beach will be in an immature condition.

1.1.1. Homology of Oviparous and Ovoviviparous Pallial Oviduct Structure.

The homology of these two oviduct structures has been pointed out by Sacchi (1975). The pallial oviduct of littorea and neritoides is by contrast not obviously homologous with these. The structural homology is clarified in the following descriptions and figures. Histological techniques applied to the oviducts have underlined the homology, similar tissues occurring in similar positions in the two systems (see below).

On an anatomical basis it seems probable that oviparous and ovoviviparous species are more closely related to each other than they are to the planktotrophic species littorea and neritoides.

1.1.2. Features Common to the Oviducts of Oviparous and Ovoviviparous Species.

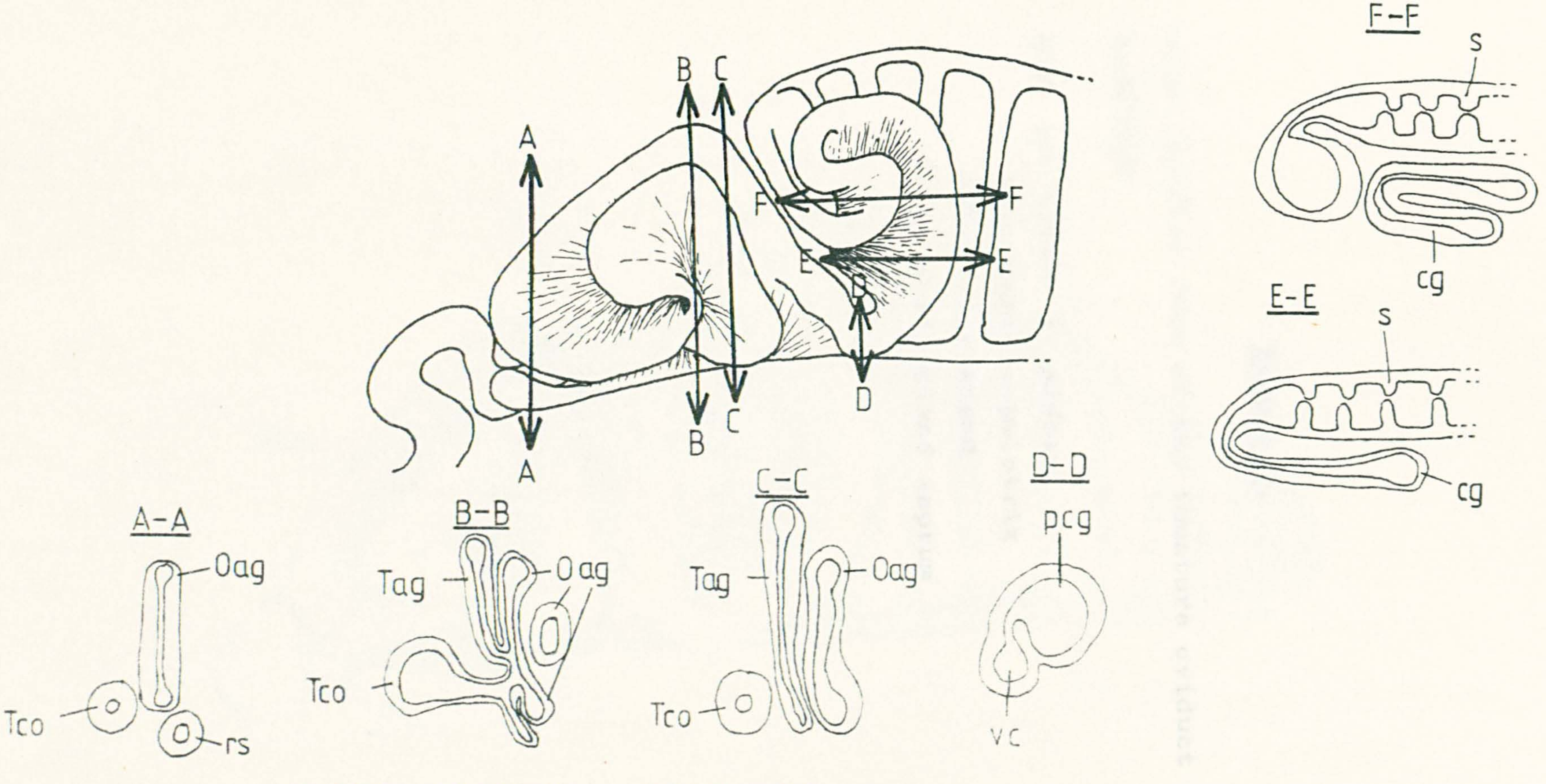
Oviduct anatomy has been studied by dissection and histological techniques in arcana, nigrolineata and rudis. Superficial examination of the oviduct of neglecta did not suggest any departure from the oviduct pattern of rudis.

Though essentially a tube, the walls of the oviduct are so elaborated and folded that the structure appears fairly complex. The anatomy is most easily understood by examining immature specimens, in which the lobing is relatively obvious (see Plate I, figs 2 & 6). The diagrammatic sections of fig. 1 should act as an aid to understanding how the duct is constructed. Where it enters the mantle cavity the oviduct is more or less tubular (the tubular coiled oviduct, Tco,) and in the mature animal its walls are swollen with glandular cells. The pattern of coiling is fairly uniform. The largest loop is not visible in right lateral view (the view observed when the animal is removed from the shell) as it is overlain by the lobed walls of the next section of the duct (fig. 3). Where the tubular coiled oviduct expands into the lobed second section (which is currently known as the albumen gland (see below) the receptaculum seminis (rs) opens and it is presumably in this area that fertilisation occurs.

Figure 1.

Diagrammatic sections through the proximal convolutions (based on an immature *L.arcana* oviduct). The positions of the sections are indicated in the central diagram.

Key: cg:- capsule gland
Oag:- opaque albumen gland
pcg:- passage to capsule gland
rs:- receptaculum seminis
s:- septum
Tag:- translucent albumen gland
Tco:- tubular coiled oviduct
vc:- ventral channel



DIAGRAMMATIC SECTIONS

Figure 2.

Right lateral view of the immature oviduct of L. arcana.

Key: see figure 1, also:-
Bc:- bursa copulatrix
Jg:- jelly gland
Jgs:- jelly gland septum

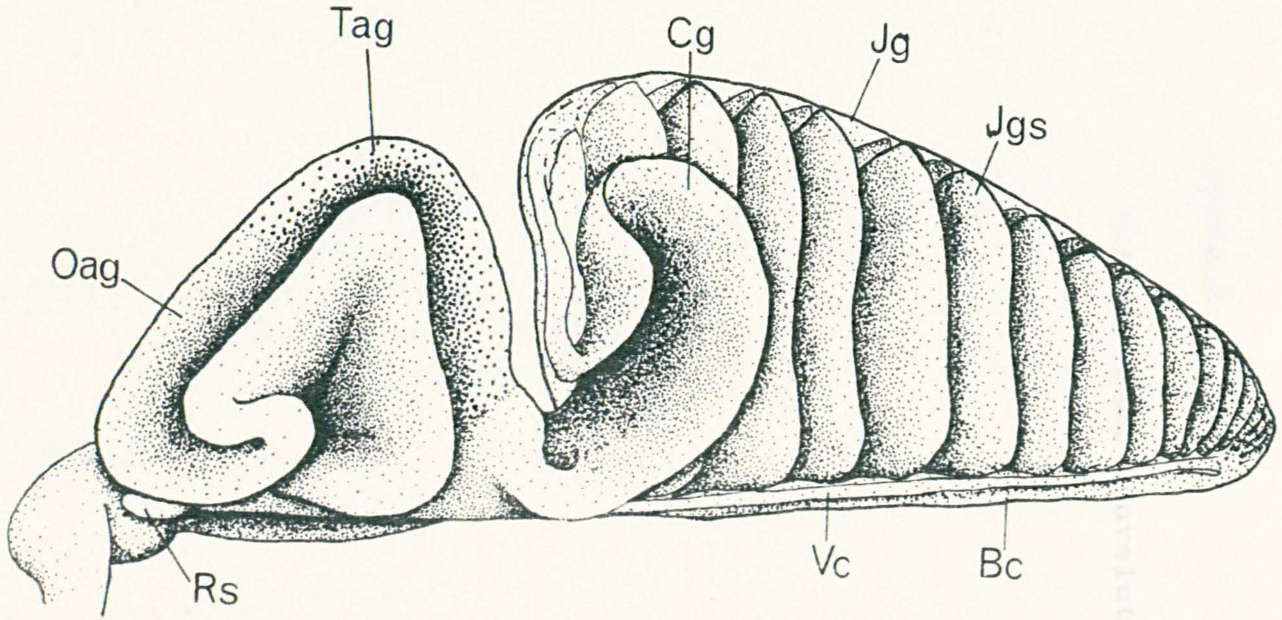
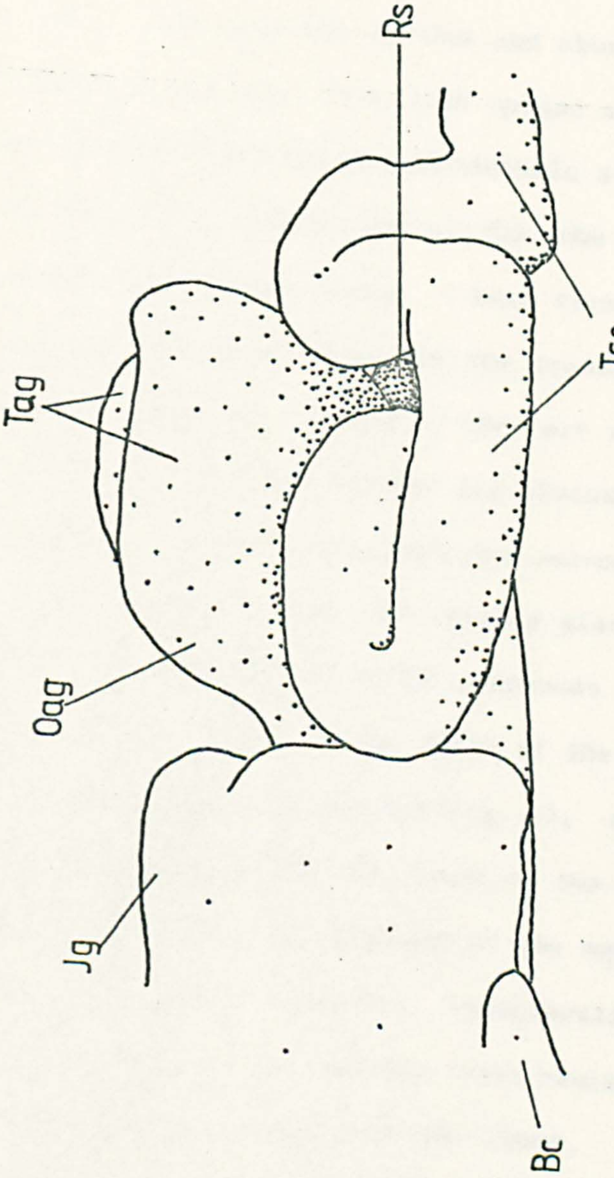

Tag
Cg
Jg
Jgs
Oag
Rs
Vc
Bc

Figure 3.

Left lateral view of the proximal convolutions of *L. arcana*.

Key: see figs. 1 & 2.


Rs
Tco
Tag
Oag
Jg
Bc

The next section of the oviduct, the so-called albumen gland (Linke, 1933; Sacchi, 1975) has an enormously expanded, flattened lobe which is folded once, into a characteristic shape. However this lobe is not one gland but two. This can be determined histologically and is frequently visually apparent as well. The proximal section and about half of the distal section of the lobe often look opaque and the tissue stains blue with modified Mallory-Heidenhain stain (Cason, 1950). The remaining, most distal part of the lobe appears translucent and does not take up the stain. I have termed these areas the opaque albumen gland (Oag) and the translucent albumen gland (Tag) (see fig. 2); possibly they are responsible for secreting respectively the fluid albumen and viscous albumen layers round the zygote. The albumen glands are succeeded by another laterally flattened lobe, the capsule gland. This probably secretes the egg membrane which surrounds the zygote and its albumen layers. Owing to the shape of the passage from the albumen gland, (see section D-D fig. 1), eggs coming down the oviduct are directed into the lumen of the capsule gland and thence enter the most distal part of the oviduct, the brood pouch or jelly gland, dorsally. Structurally the brood pouch and jelly gland are very similar, both having a complex of incomplete septa extending into the lumen.

The oviduct is also adapted for the reception and storage of sperm prior to fertilisation. The bursa copulatrix (Bc), a diverticulum just inside the opening of the oviduct, receives the sperm during copulation. Later the sperm travel along the ventral channel, a ciliated gutter in the ventral part of the distal oviduct. They pass ventrally into the albumen glands and thence to the receptaculum seminis. Thus the eggs and sperm are travelling in opposite directions along the oviduct and are kept apart through the complex folding of the duct walls (see fig. 1).

1.1.3. Adaptations of the basic oviduct plan in the representative Oviparous and Ovoviviparous species, L. arcana and L. rudis.

In arcana the proximal convolutions, (tubular, coiled oviduct, opaque and translucent albumen glands and the capsule gland) are large relative to the size of the oviduct, and the distal section (jelly gland) is comparatively short. In the mature condition the septa of the distal section swell and completely fill the lumen. This tissue is thought to secrete the jelly matrix of the egg mass. The bursa copulatrix is comparatively long, often about three-quarters of the length of the jelly gland and extending (usually) to below the capsule gland. When mature the oviduct appears as a large white mass (figs. 4 & 8, Plate 2) the structure of which is barely discernible.

In rudis the proximal convolutions are relatively small and the distal section relatively long. With maturity the septa of the distal section (the brood pouch) do not become markedly glandular as in arcana; instead, developing embryos fill the lumen.

Figure 4.

Right lateral view of the pallial oviduct of a mature *L. arcana* female.

Key: see figs.1 & 2, also:
Cf:- ciliated field
Cm:- columellar muscle

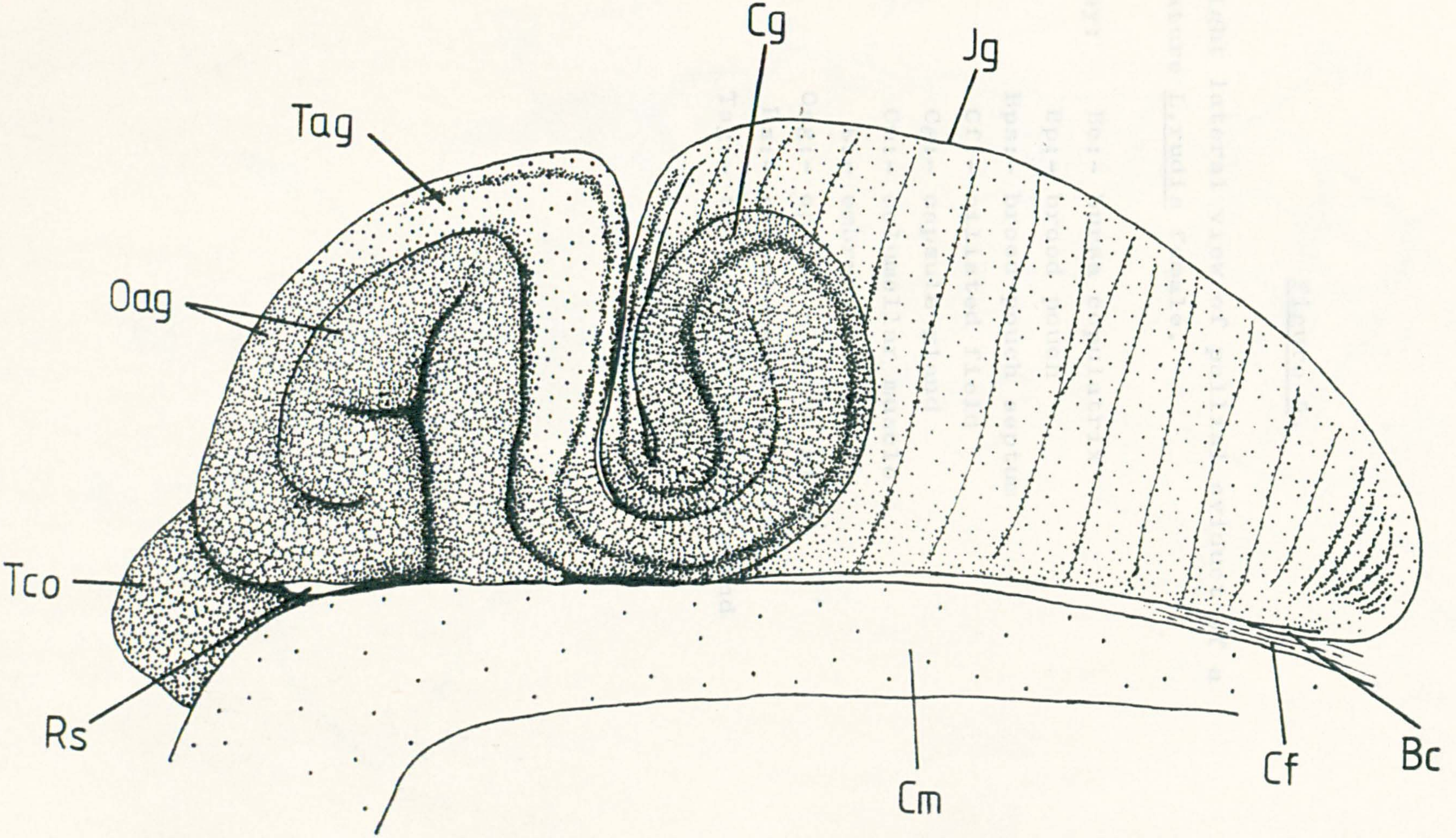

Tag
Cg
Jg
Oag
Tco
Rs
Cm
Cf
Bc

Figure 5.

Right lateral view of pallial oviduct of a mature *L.rudis* female.

Key:
Bc:- bursa copulatrix
Bp:- brood pouch
Bps:- brood pouch septum
Cf:- ciliated field
Cg:- capsule gland
Cm:- columellar muscle
E:- embryo
Oag:- opaque albumen gland
Rs:- receptaculum seminis
Tag:- translucent albumen gland

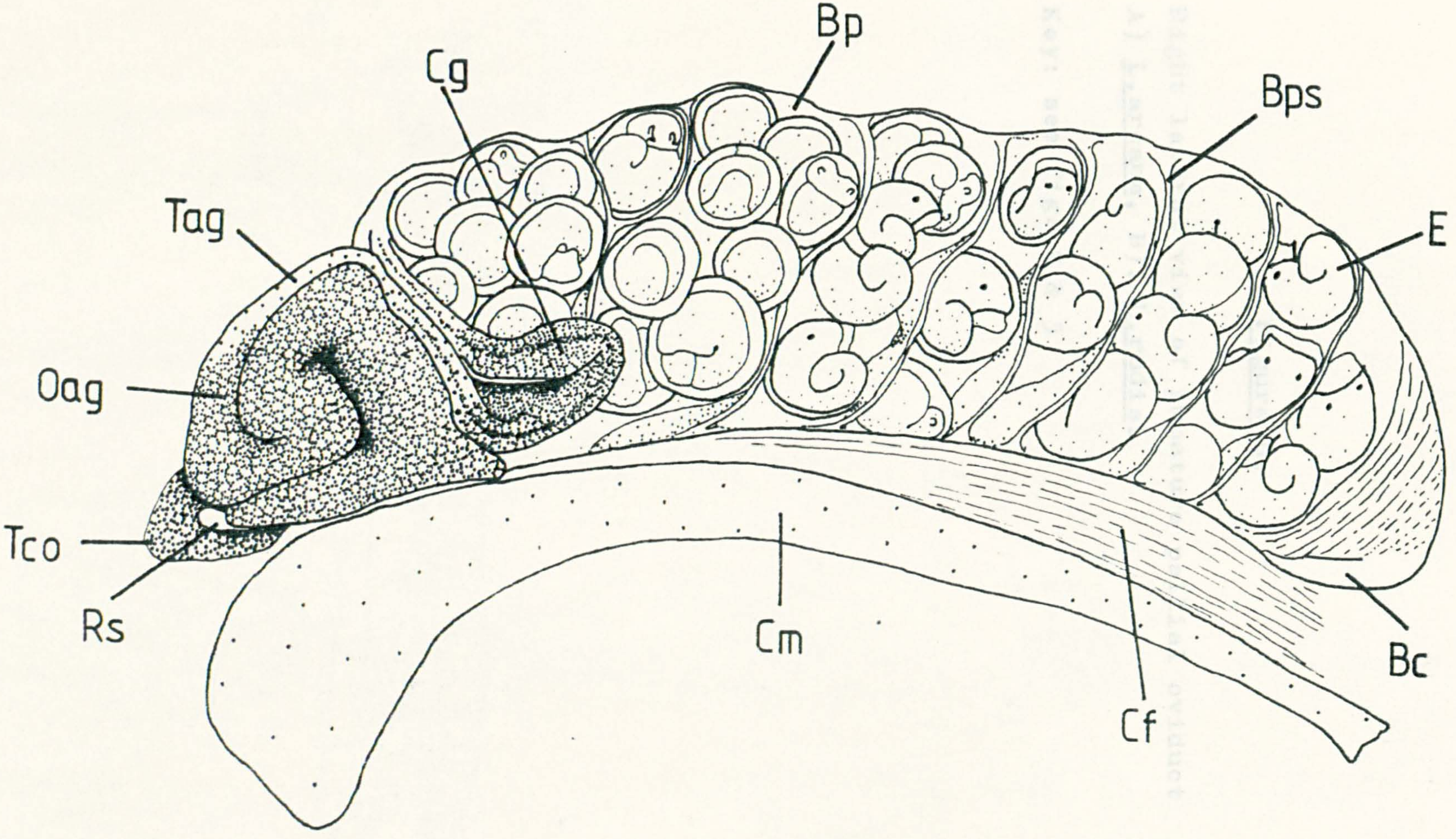
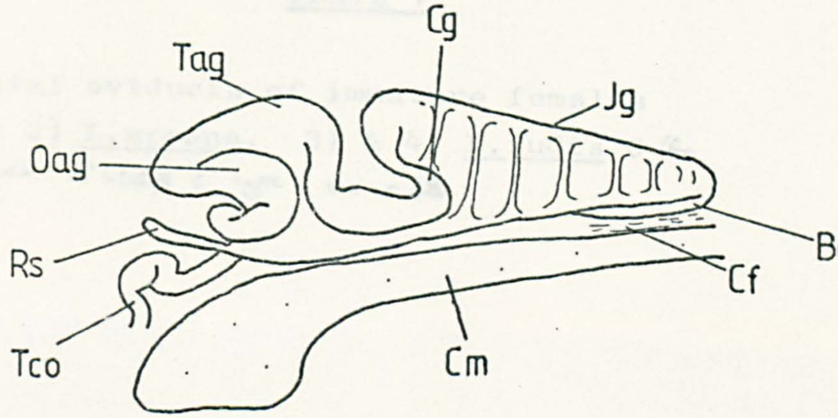

Bp
Cg
Bps
Tag
E
Oag
Tco
Rs
Cm
Bc
Cf

Figure 6.

Right lateral view of immature pallial oviduct A) *L. arcana*, B). *L. rudis*.

Key: see figs.2 & 5

A.



B.

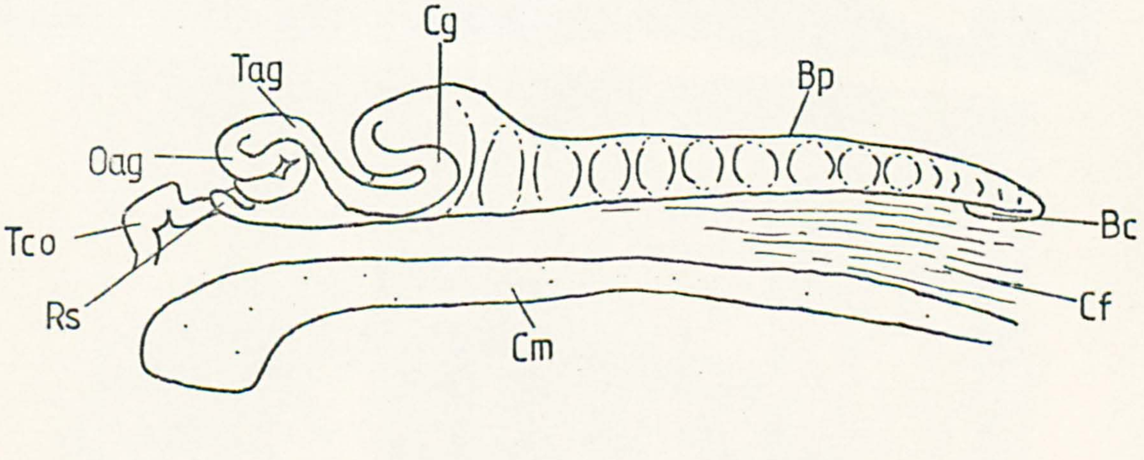


PLATE 1.

Pallial oviducts of immature females
1) & 2) L.arcana, 3) & 4) L.rudis C (for explanation of L.rudis 'C' type see p.48)

1

2

3

4

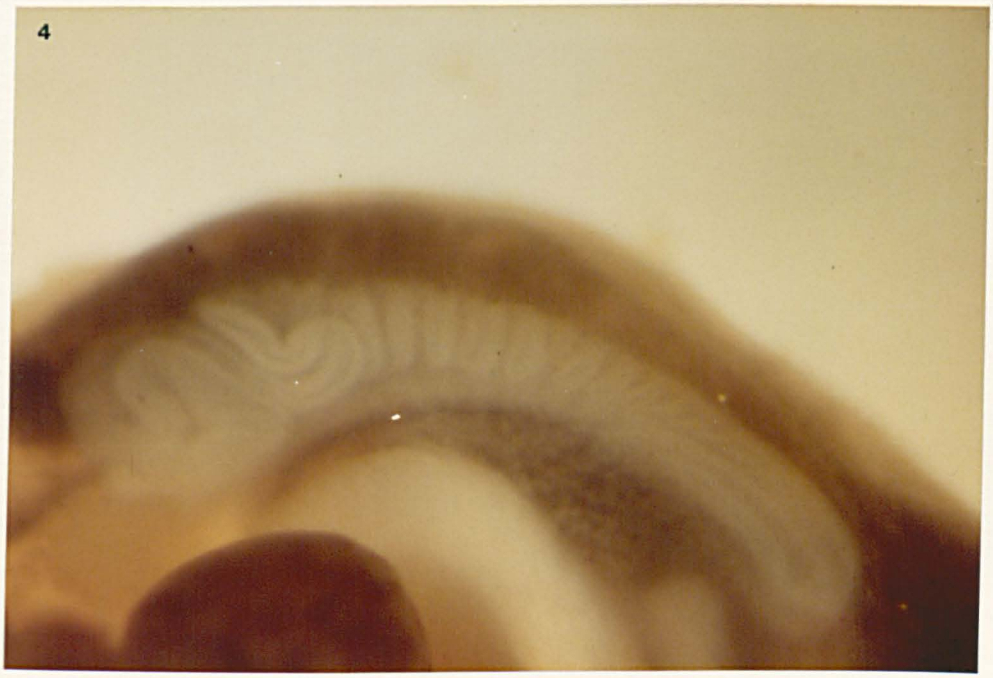


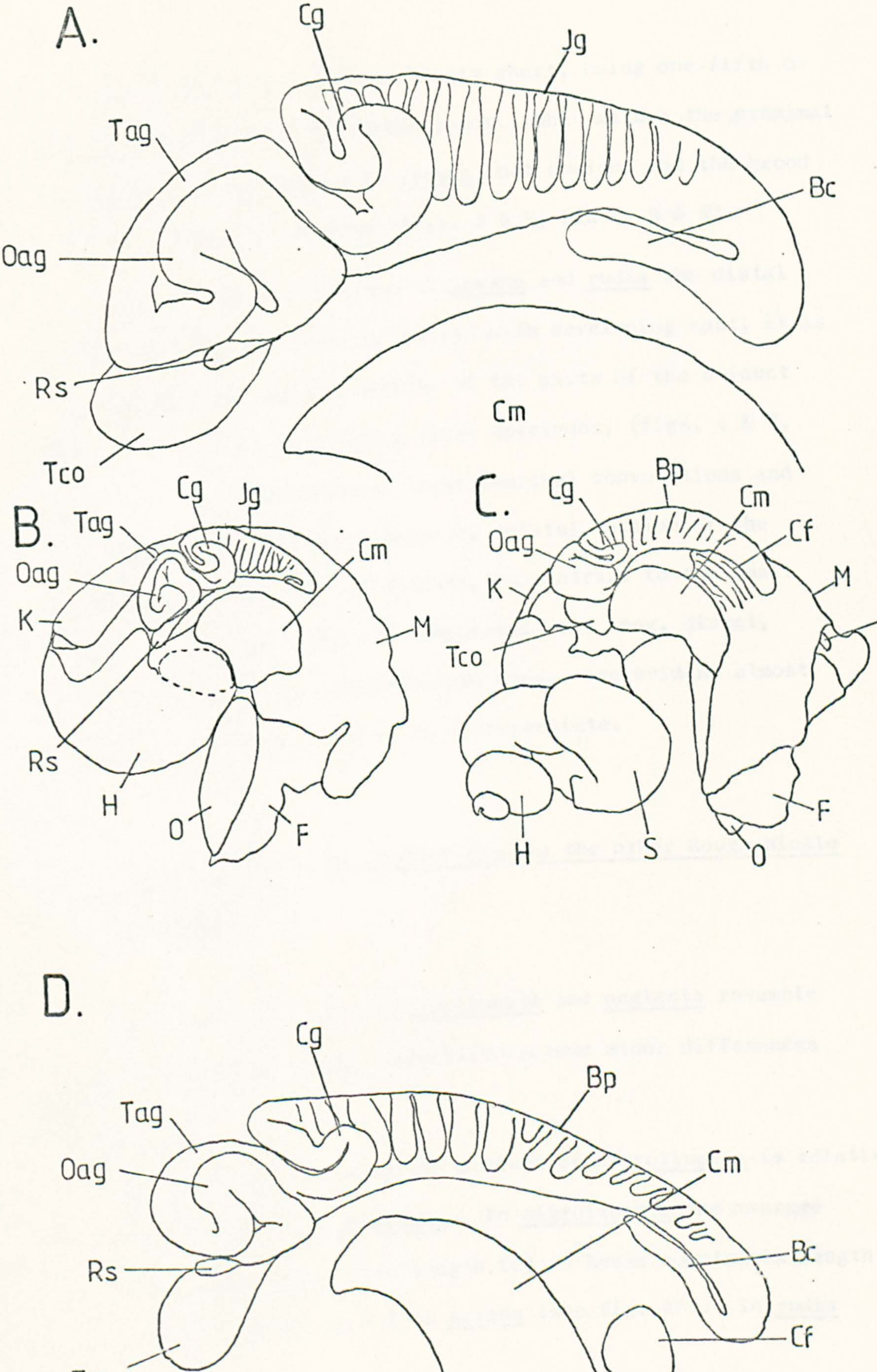
Figure 7.

Figures traced from Plate 1.

A). & B). L.arcana C). & D). L.rudis C

Key: see figs. 2 & 5, also:-

F:- foot
H:- hepatopancreas
K:- kidney
M:- mantle
O:- operculum
S:- stomach
Te:- tentacle
Tco:- tubular coiled oviduct


A.
Cg
Jg
Tag
Bc
Oag
Rs
Cm
Tco
B.
Cg
Jg
Tag
Oag
K
Cm
M
Rs
H
O
F
C.
Cg
Bp
Cm
Cf
Oag
K
M
Te
Tco
H
S
F
O
D.
Cg
Bp
Tag
Oag
Cm
Rs
Bc
Tco
Cf

The bursa copulatrix is comparatively short, being one-fifth or less, of the length of the brood pouch. When mature the proximal convolutions are, compared to arcana, not obvious and the brood pouch is filled with embryos (figs. 5 & 8, Plates 2 & 3).

Since in immature females of arcana and rudis the distal section is neither fleshy nor packed with developing spat, it is the difference in the proportions of the parts of the oviduct which is useful in identifying these specimens, (figs. 6 & 7, Plate 1). The comparatively large proximal convolutions and bursa copulatrix and short, septate, distal section of the oviparous format of pallial oviduct, in contrast to the small proximal convolutions and bursa copulatrix and long, distal, septate section of the ovoviviparous type, are evident almost as soon as the oviduct starts to differentiate.

1.1.4. Variations in the Oviduct seen in the other Rough Winkle Species.

Though the oviducts of nigrolineata and neglecta resemble those of arcana and rudis, respectively, some minor differences do occur.

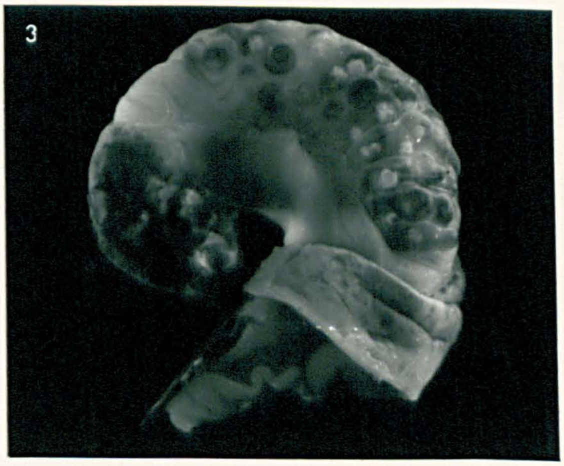
The bursa copulatrix of the oviduct of nigrolineata is relatively much shorter than that of arcana. In nigrolineata the average ratio, $^{c}/Bc$, of jelly gland length (c) to bursa copulatrix length (Bc) is 3.1, compared to 1.7 in arcana (see fig. 27). In rudis

PLATE 2.

Pallial oviducts of mature females
1) & 2) *L.arcana*, 3) & 4) *L. rudis* C


1


2


3


4

Figure 8.

Figures traced from Plate 2.
A). & B). L.arcana, C). & D). L.rudis C

Key: see figs. 2,5 & 7

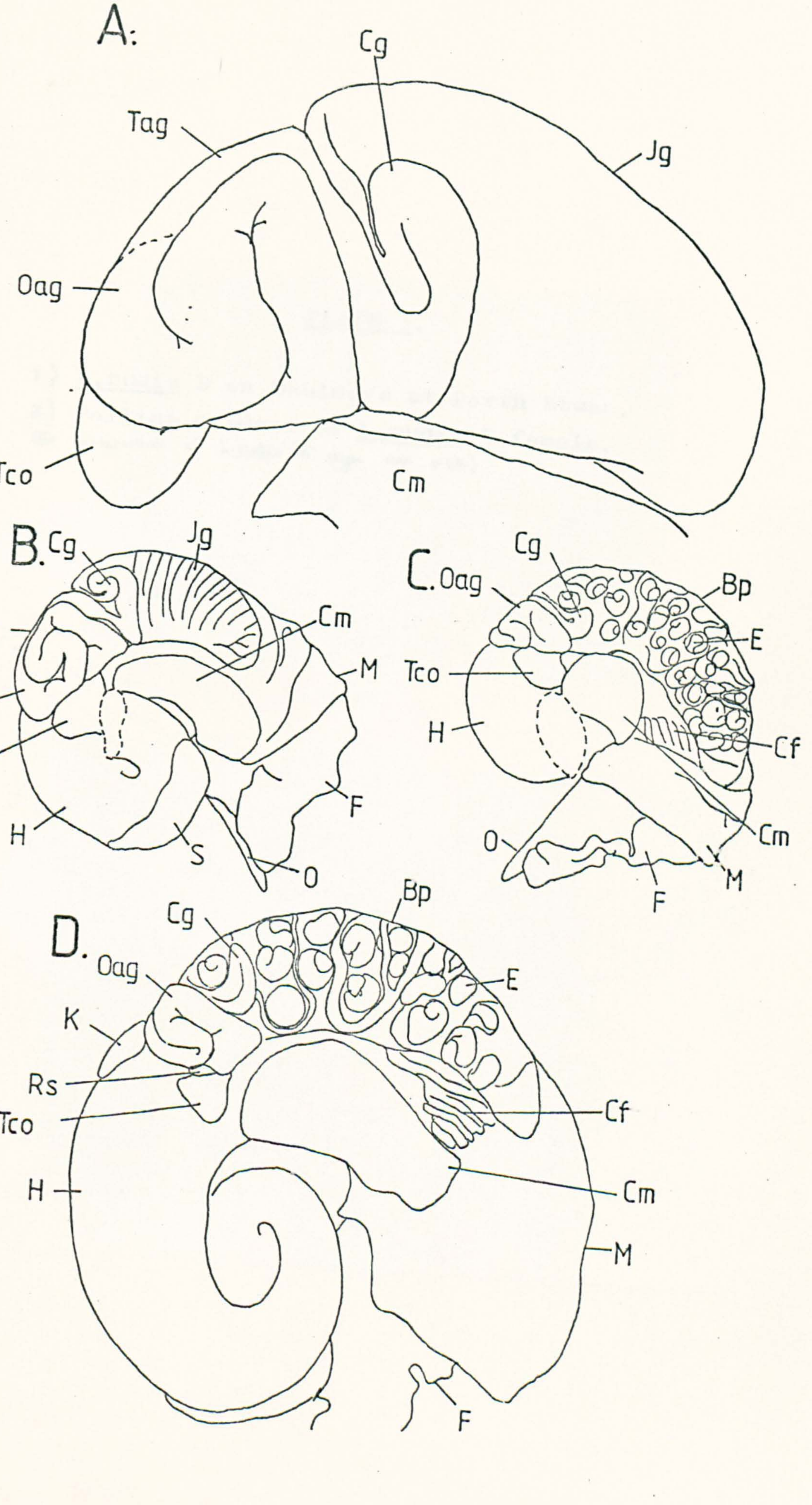

A:
Cg
Tag
Jg
Oag
Tco
Cm
B.
Cg
Jg
Tag
Cm
M
Oag
Tco
F
H
S
O
C.
Oag
Cg
Bp
E
Tco
H
Cf
Cm
O
M
F
D.
Bp
Cg
Oag
E
K
Rs
Cf
Tco
Cm
H
M
F

PLATE 3.

1) *L.rudis* B on boulders at Porth Stwan.
2) Pallial oviduct of *L.rudis* B female.
(for explanation of *L.rudis* 'B' type see p.48)


1


2

this ratio is 4.0, i.e. also much shorter than in *arcana*. This factor may be of significance in species recognition (see section 1.2.4). In other respects the pallial oviduct of *nigrolineata* and *arcana* seem very similar.

The oviduct of *neglecta* is likewise similar to that of *rudis*; however, there is probably a functional difference. In *neglecta* the eggs are almost certainly produced individually since embryos in the brood pouch are usually at different stages of development. Interestingly the embryos, unlike the parent, are not scaled-down versions of those of *rudis*. Indeed they are of an approximately equivalent size, so that the physically smaller brood pouch of *neglecta* rarely holds more than 10 embryos. Females of *rudis* often have 100 or more embryos in the brood pouch. Thus relative to the maternal size, the embryos of *neglecta* are very much larger than those of *rudis* and the number of offspring per mother are probably fewer.

1.1.5. Functional Significance of Pallial Oviduct Differences in Oviparous and Ovoviviparous Species.

Besides the difference in adaptation of the distal section as either brood pouch or jelly gland, in which the functional significance is readily apparent, there is also the previously unexplained difference in relative size of the proximal convolutions. These I think secrete the nutritive layers and protective egg membrane which surround the embryos of both oviparous and ovoviviparous species. One possible explanation is that the difference in size of the proximal convolutions is related to differences in the pattern of egg production.

Oviparous species may lay egg masses containing 100-300 eggs. Plate 4 shows an egg mass of arcana photographed shortly after it was laid. As this shows, in this egg mass (as in all egg masses I have seen) the eggs are very close in age. Assuming no inhibition of development occurs, it can be inferred from this that the pattern of egg production is such that all the eggs for an egg mass are produced in one batch.

It is interesting to note that, as this egg mass shows (the eggs are at the 1- 2- or 4-cell stage) eggs are laid at an early stage of development. It seems probable therefore that the time between fertilisation and laying of the eggs is short. Thus the nutritive and protective layers for all the hundred or more eggs in such an egg mass would have to be produced comparatively rapidly.

The pattern of egg production in rudis is rather different. Eggs appear to be released in small batches of about 20. Correspondingly the pattern of demand for outer egg layers is markedly dissimilar in these two representative species. Assuming that the gland cells of the proximal convolutions operate at similar rates in oviparous and ovoviviparous species, then for an oviparous species to coat quickly the large number of eggs produced for an egg mass, it would require a large number of gland cells ready to operate in unison. The gland cells in ovoviviparous species presumably have time to replenish their secretory products before the next batch of eggs is released into the oviduct. Fewer cells can therefore coat an equivalent number of eggs and so the proximal convolutions can be smaller.

PLATE 4.

1). Maternal L.arcana and recently laid egg mass.

2). Close-up of egg mass (scale in millimetres).
Note the pink coloration of the developing eggs.

1

2

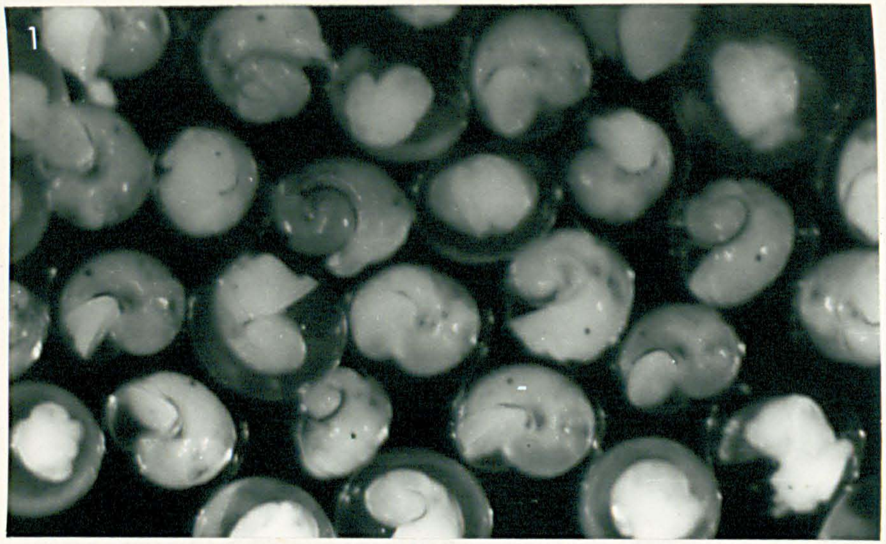
PLATE 5.

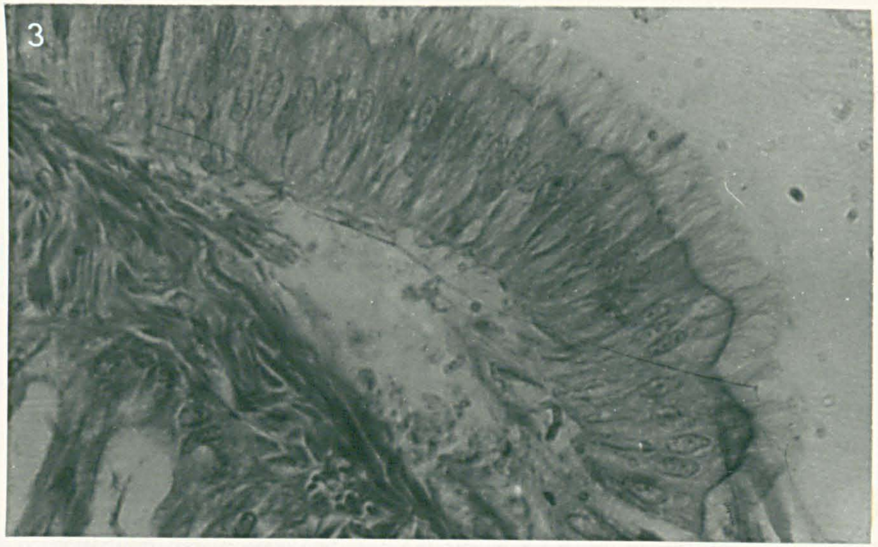
1). Embryos from the brood pouch of L.rudis.

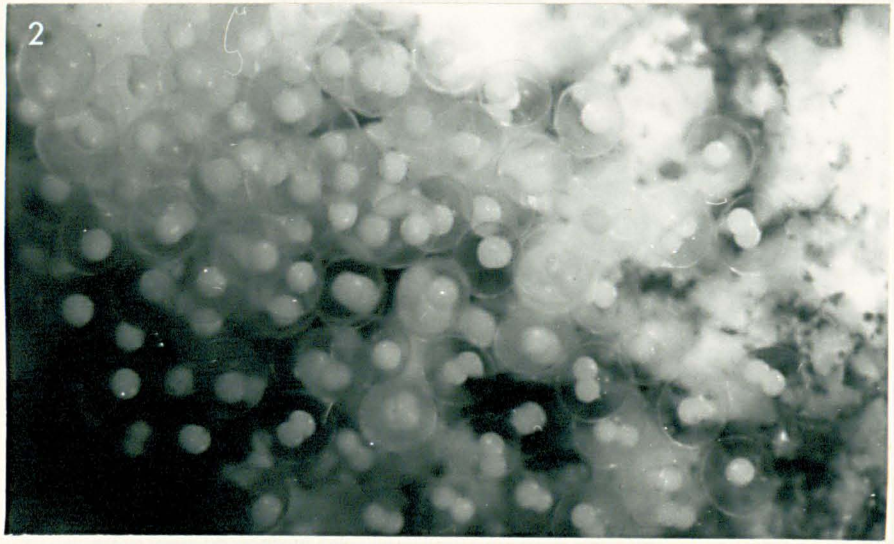
2). L.arcana egg mass, eggs at 1-,2- & 4-cell stages, (about two hours after being laid).

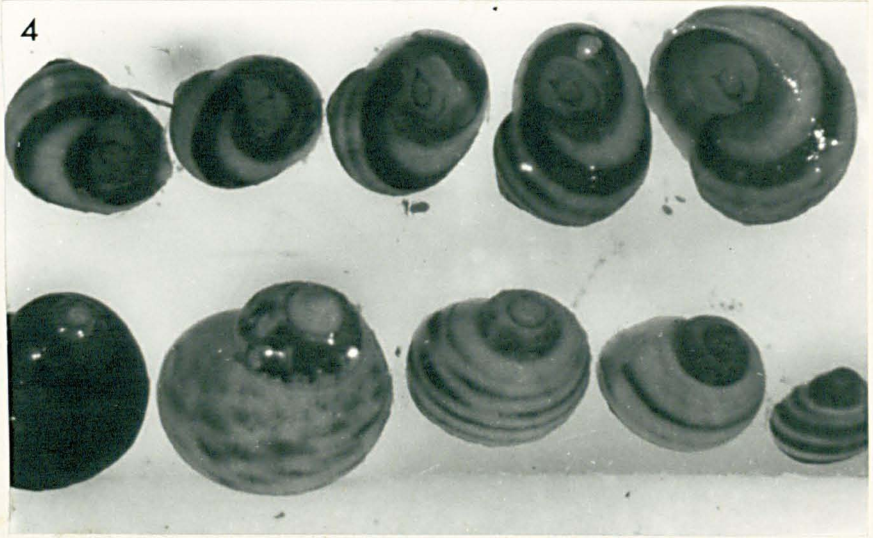
3). Ciliated field tissue.

4). L.neglecta, shell colour patterns and sub-opercular patterns.


1


3


2


4

1

2

1.2 PENIS CHARACTERISTICS

The number and arrangement of penial glands and the size and shape of the penis tip are the most useful features of the penial structure for taxonomic purposes. In the species obtusata and mariae these characteristics are diagnostic, obtusata having penes with tips considerably less than half the total length of the penis and more than 16 penial glands arranged in two or more rows, mariae having penes with tips approximately half the total length of the penis and 17 or fewer penial glands arranged in a single row.* Indeed it was the occurrence of these two distinct penis forms in what was considered one species, littoralis, that first drew the attention of Sacchi and Rastelli (1967) to the possibility that two species were present.

Heller (1975a) examined the penial forms of the rough winkles looking for equivalent diagnostic differences. He described differences in the four species he recognised, though as Raffaelli (1979) has pointed out the differences between rudis and patula were not diagnostic. Of the four species only nigrolineata has indisputably diagnostic penial characteristics. These have been adequately illustrated by Heller (1975a) and also Sacchi (1975) who independently redescribed the species in the same year. In this species the penis has a short tip (less than the width of one penial gland) and comparatively few penial glands (3-9 is the figure given by Heller, 7-12 by Sacchi, Naylor (1978) observed a range of 4-12 and at Porth Swtan I have seen 2-9).

The remaining three species which I recognise, rudis, arcana and neglecta I, likewise, have examined for possible diagnostic

---

*Goodwin & Fish (1977).

penial characteristics, especially rudis and arcana since diagnostic differences in penis form which might distinguish these species were of particular interest to me. Considerably more attention was paid to this problem than to the separation of neglecta and rudis on penial characters. In the future, however, this latter problem may be of equal importance as more is learnt about the possible speciation within rudis (see sections 1.2.3 and 4).

1.2.1. Comparison of Penial Characteristics in L. arcana and L. rudis.

In the following comparison of mature penial characteristics in arcana and rudis identification of species was based on the ciliated field character (see section 1.3). Assessment of maturity is discussed in section 2.1. Specimens from mixed populations at five sites were examined and various penial variables compared. Aspects of penial form which were considered important were a) the number and arrangement of penial glands b) relative size of the regions of the penis (tip, gland bearing region, basal region) and c) the size of the penis relative to the size of the animal.

The penial glands can be easily counted under a binocular microscope. They may be arranged in a regular single row, irregularly or in a double row (which is defined as four or more penial glands in a row parallel to the main row); very rarely triple rows were seen in arcana only. The data obtained from the five sites were lumped for each species and plotted in fig. 9 (see Appendix A, tables 1 &2 for data from individual sites).

Figure 9

Frequencies of patterns of arrangement of penial glands, and number of penial glands.

A) <u>L.rudis</u> C B) <u>L.arcana</u>

FREQUENCIES OF PATTERNS OF ARRANGEMENTS OF PENIAL GLANDS & NUMBER OF PENIAL GLANDS. A) L. rudis B) L. arcana

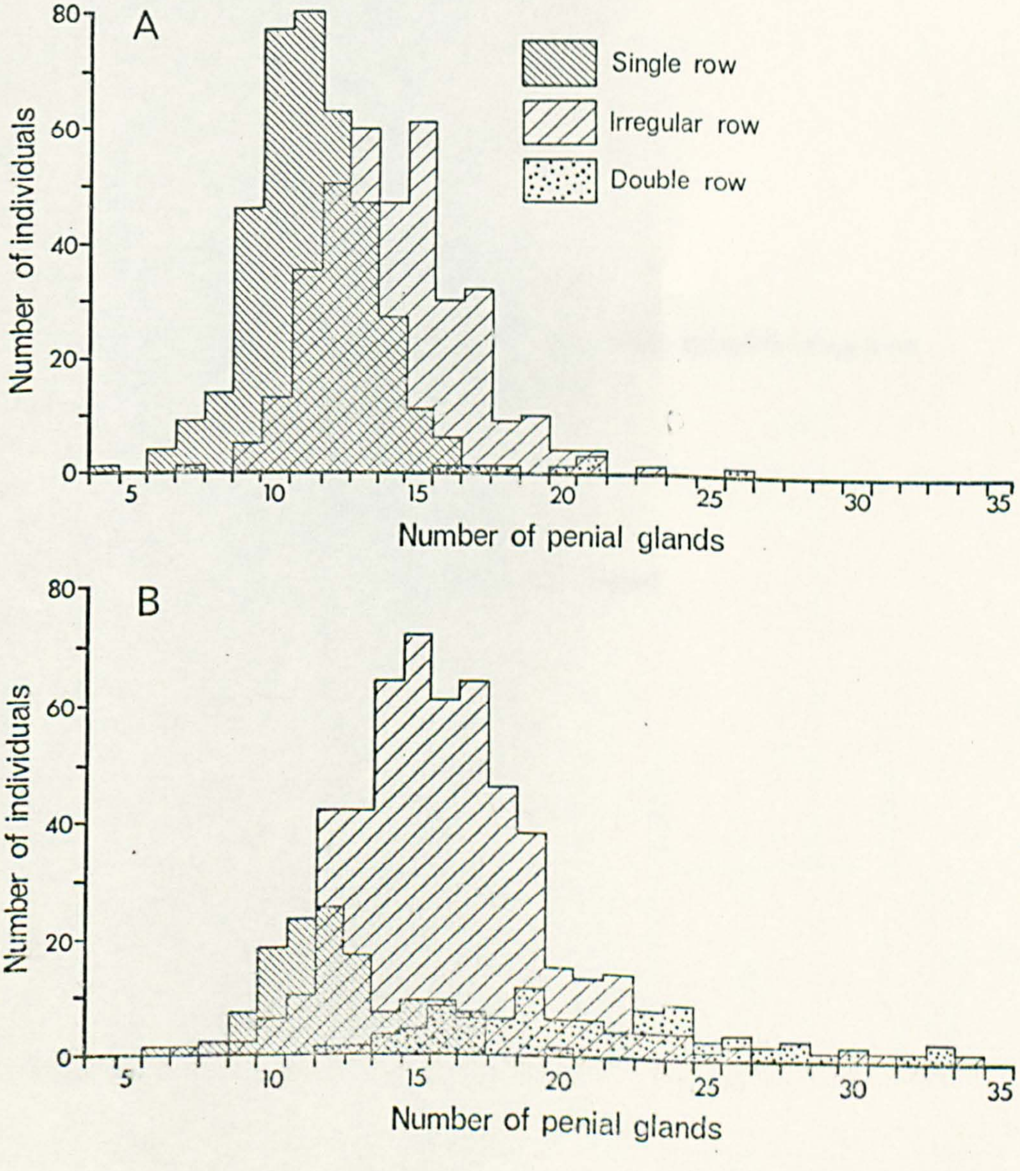
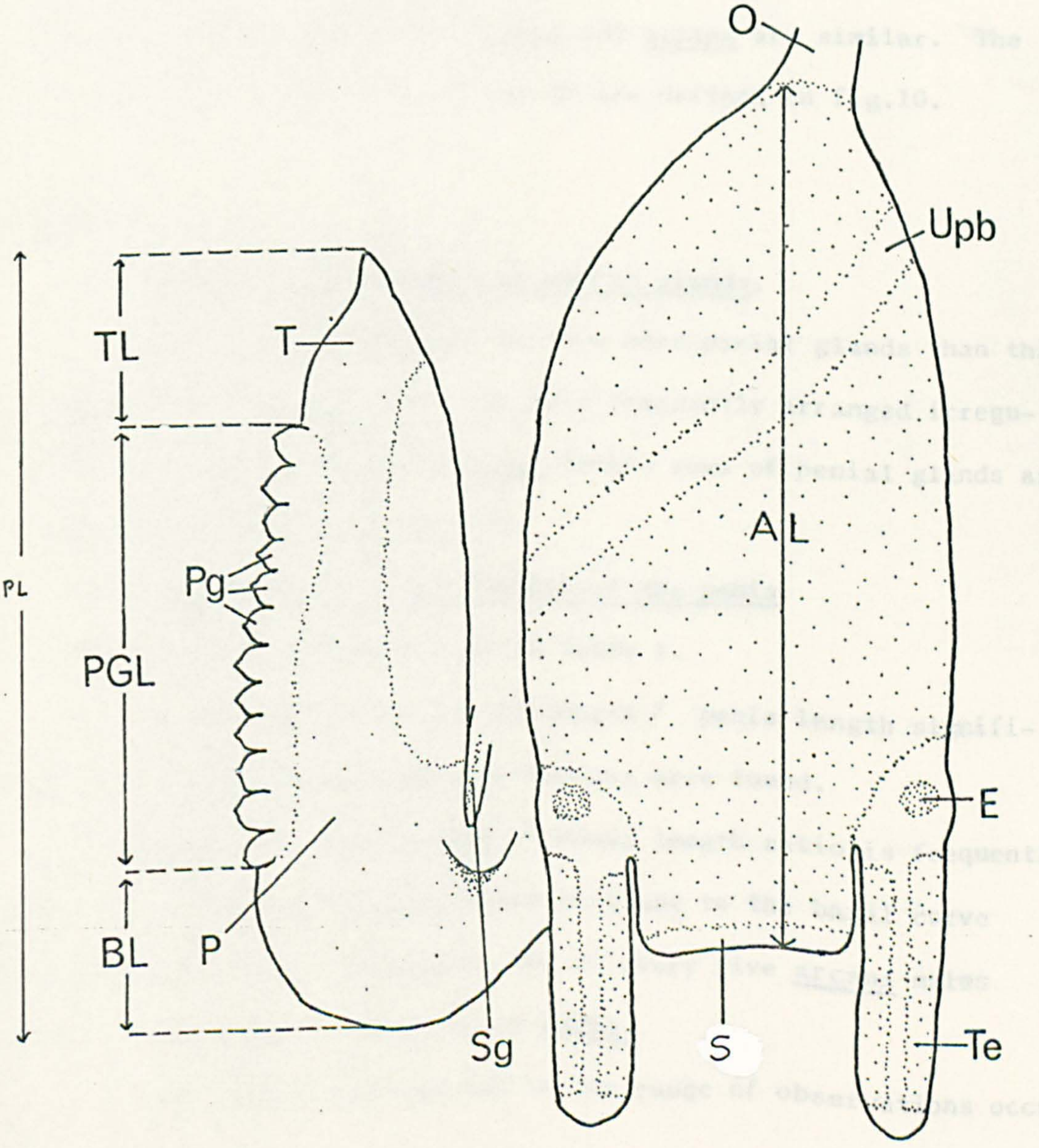


Figure 10.

Definition of measurements of penis.

Key: AL:- animal length
BL:- basal length
E:- eye
O:- oesophagus
P:- penis
S :- snout
Pg:- penial gland
PGL:- length of penial gland region
Sg:- sperm groove
T:- tip
Te:- tentacle
TL:- tip length
Upb:- unpigmented band


O
Upb
TL
T
PL
Pg
AL
PGL
E
P
BL
Sg
S
Te

The ratios of relative size of the regions, tip length / penis length (TL/PL), penial gland region length / penis length (PGL/PL) and basal region length / penis length (BL/PL), and also the size of the penis relative to the animal (PL/AL), for the two species at each of the five sites were compared using the Mann-Whitney U test (Siegel, 1956), against the null hypothesis that the ratios obtained from rudis and arcana are similar. The measurements, TL, PGL, BL, PL and AL are defined in fig.10.

Results

a) Number and arrangement of penial glands.

The penis of arcana tends to have more penial glands than that of rudis, and in arcana they are more frequently arranged irregularly or in double rows. In rudis double rows of penial glands are comparatively rare, (see fig. 9).

b) Relative size of the regions of the penis

The results are summarised in table 1.

In all the ratios except tip length / penis length significant differences between the two species were found.

In arcana the basal length / penis length ratio is frequently zero, i.e. the penial glands often continue to the basal curve (see fig. 11). This occurred in two of every five arcana males examined compared to one in 72 in rudis.

For each ratio some overlap in the range of observations occurred.

c) Size of the penis relative to the animal

At each site arcana males had significantly smaller penes

TABLE 1

Comparison of penial shape characteristics of L. arcana with L. rudis using Mann-Whitney U test.

		L. arcana			L. rudis				
Ratio	Site	n	median	range of observations	n	median	range of observations	z	two-tailed p
PL/AL	P.S.	86	.886	.650-1.275	100	1.120	.776-1.333	9.39	<.00006
	A.B.	61	.895	.648-1.143	50	1.066	.800-1.349	4.91	<.00006
	B.R.	68	.922	.712-1.156	72	1.091	.810-1.346	6.89	<.00006
	Y.W.	60	.821	.683-1.014	40	1.061	.840-1.324	7.79	<.00006
	D.	37	.867	.613-1.137	32	1.071	.877-1.500	5.58	<.00006
TL/PL	P.S.	84	.241	.155-.388	94	.235	.134-.365	2.72	.0066
	A.B.	61	.240	.172-.321	49	.250	.147-.352	.78	.4354
	B.R.	79	.222	.150-.381	73	.242	.159-.386	2.73	.0054
	Y.W.	60	.250	.143-.353	41	.244	.145-.347	.23	.8180
	D.	49	.241	.154-.362	34	.239	.169-.349	.84	.4010
PGL/PL	P.S.	89	.735	.529-.823	95	.606	.375-.780	9.38	<.00006
	A.B.	61	.716	.580-.815	50	.600	.448-.740	6.48	<.00006
	B.R.	79	.723	.455-.875	73	.603	.292-.726	8.46	<.00006
	Y.W.	60	.732	.600-.831	41	.577	.418-.744	7.83	<.00006
	D.	47	.729	.571-.821	35	.576	.241-.733	6.73	<.00006
BL/PL	P.S.	91	.000	.000-.2352	93	.1711	.000-.3750	10.31	<.00006
	A.B.	61	.0395	.000-.1695	49	.1408	.000-.3529	6.47	<.00006
	B.R.	79	.0333	.000-.2860	72	.1714	.000-.3750	7.34	<.00006
	Y.W.	60	.000	.000-.1600	41	.1710	.012-.3890	7.93	<.00006
	D.	48	.0385	.000-.1224	35	.1429	.0706-.4684	7.06	<.00006

Sites:- P.S. - Porth Swtan; A.B. - Abraham's Bosom; B.R. - Bell Rock;
Y.W. - Ynys Wellt; D. - Dunbar

Measurements:- A.L. - length of head mass; B.L. - base length;
P.G.L. - length of penial gland zone; P.L. - length of penis;
T.L. - tip length

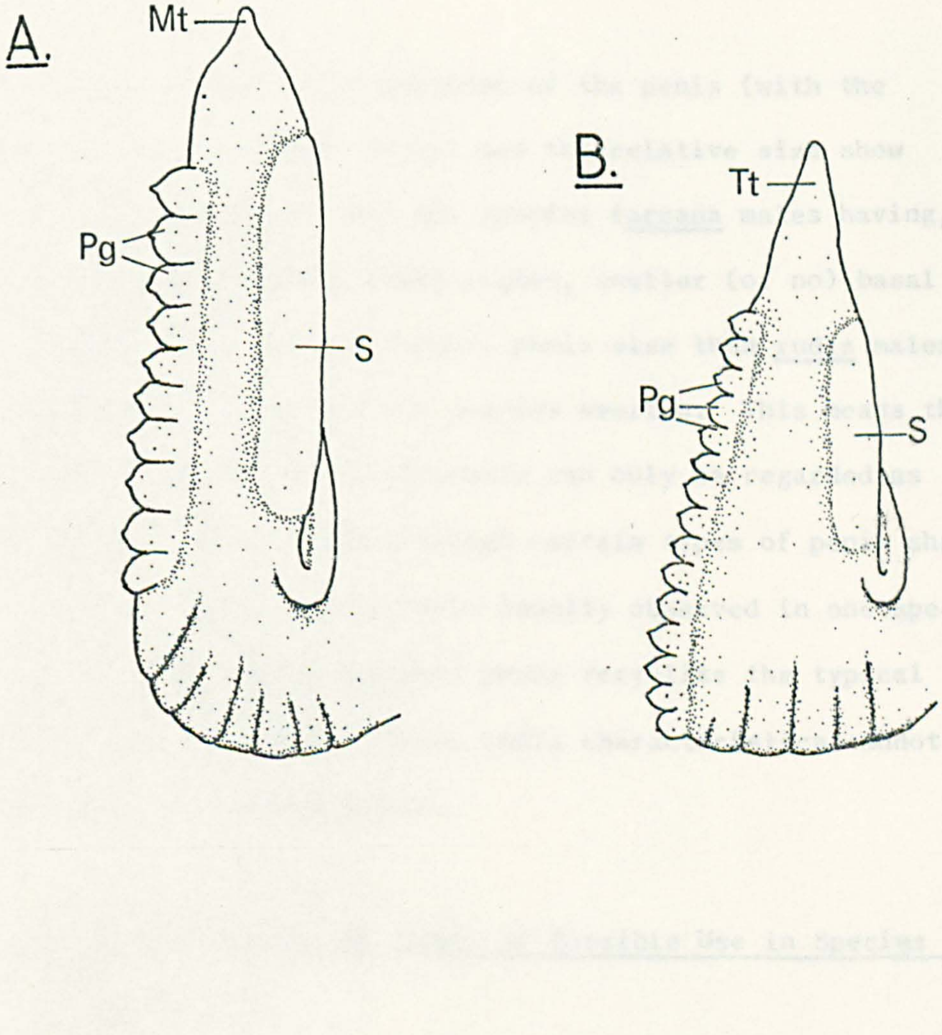
Figure 11.

Typical penis forms.

A). L.rudis

B). L.arcana

Key:-
Mt:- mucronate tip
pg:- penial glands
s:- swelling
tt:- tapering tip


A.
Mt
Pg
S
B.
Tt
Pg
S

relative to the size of the animal, compared to rudis.

Discussion of results and assessment of the usefulness of the penis in separating rudis and arcana

Though in general, penial gland number and arrangement differ in the two species, the differences are not diagnostic. Thus for example, if a male has a penis with 19 glands arranged in a double row it cannot be ascribed on this basis to one species or the other, though it is more likely to be an arcana male than a rudis.

Similarly though the proportions of the penis (with the exception of the tip length ratio) and the relative size show significant differences in the two species (arcana males having, on average, a larger penial gland region, smaller (or no) basal region and generally smaller overall penis size than rudis males) the ratios obtained from the two species overlap. This means that, as with penial glands, the differences can only be regarded as typical not diagnostic. Hence though certain types of penis shape (see figs 11& 12, Plate 7) are more usually observed in one species or the other, rudis males can have penes very like the typical arcana form and vice versa. These penis characteristics cannot therefore be used diagnostically.

1.2.2. Other Features of the Penis of Possible Use in Species Identification.

1.2.2.1.Tip shape

In rudis the tip of the penis is frequently of a mucronate

PLATE 7.

Penis and prostate appearance.

1) & 2). L.arcana

3) & 4). L.rudis C

1

2

3

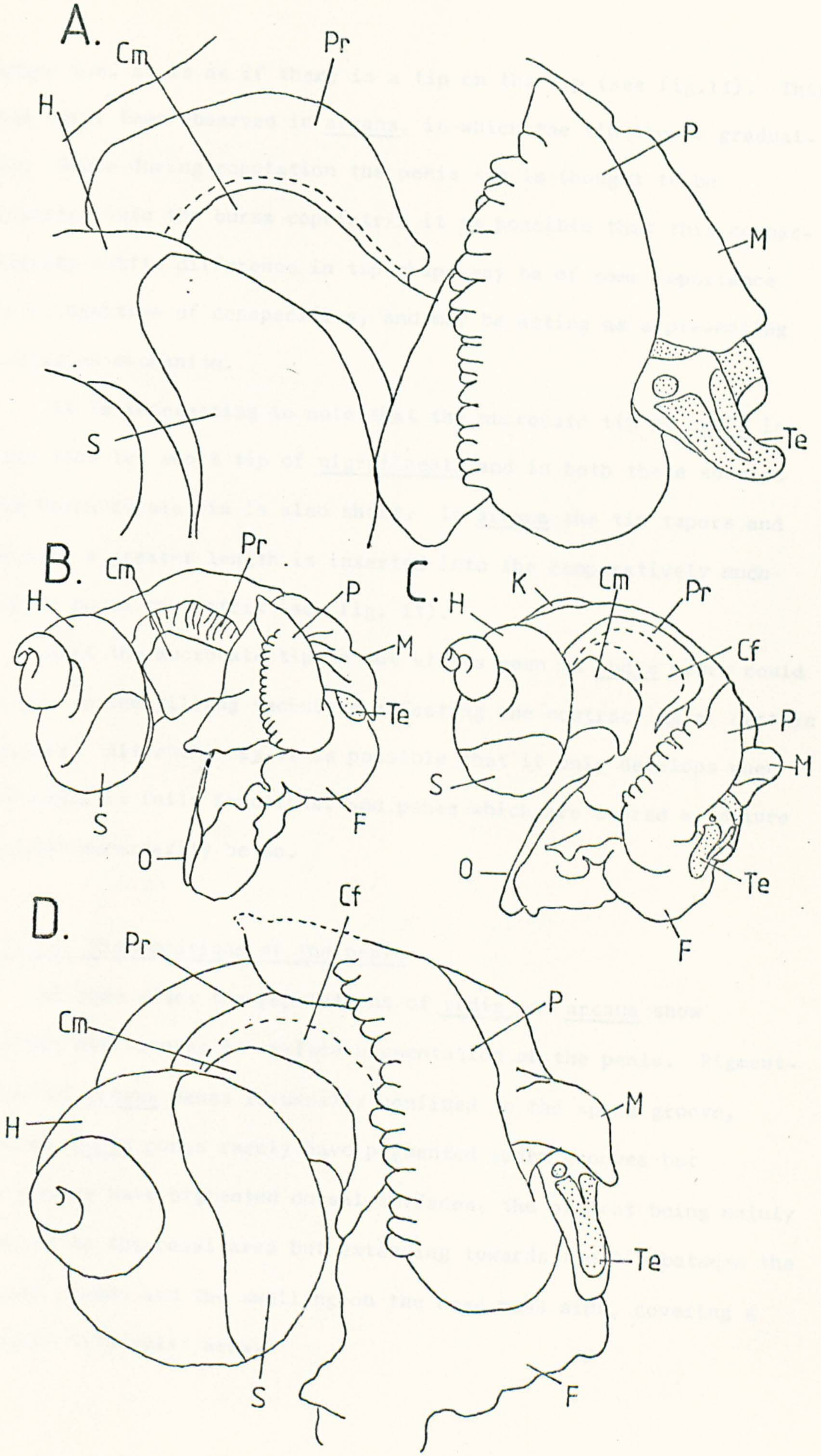
4

Figure 12.

Figures traced from Plate 7.
A). & B). L.arcana   C). & D). L.rudis C

Key: see figs. 4 & 10, also:-

F:- foot
H:- hepatopancreas
K:- kidney
M:- mantle
O:- operculum
Pr:- prostate
S:- stomach


A.
Cm
Pr
H
P
M
S
Te
B.
Pr
Cm
H
P
M
Te
S
F
O
C.
K
H
Cm
Pr
Cf
S
P
M
O
Te
F
D.
Cf
Pr
P
Cm
M
H
Te
S
F

shape i.e. it is as if there is a tip on the tip (see fig.11). This has never been observed in arcana, in which the tip tapers gradually. Since during copulation the penis tip is thought to be inserted into the bursa copulatrix it is possible that this comparatively subtle difference in tip shape may be of some importance in recognition of conspecifics, and may be acting as a pre-mating isolation mechanism.

It is interesting to note that the mucronate tip of rudis is very like the short tip of nigrolineata and in both these species the bursa copulatrix is also short. In arcana the tip tapers and perhaps a greater length is inserted into the comparatively much longer bursa copulatrix (see fig. 13).

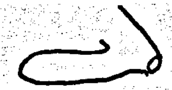
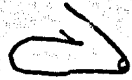
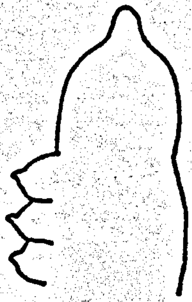
That the mucronate tip is not always seen in rudis penes could be due to the killing technique affecting the contracting of certain muscles. Alternatively it is possible that it only develops when the penis is fully functional and penes which are scored as mature may not necessarily be so.

1.2.2.2. Pigmentations of the penis

At some sites the populations of rudis and arcana show specific differences in surface pigmentation of the penis. Pigmentation of arcana penes is usually confined to the sperm groove, whereas rudis penes rarely have pigmented sperm grooves but frequently have pigmented dorsal surfaces, the pigment being mainly limited to the basal area but extending towards the tip between the penial glands and the swelling on the head mass side, covering a roughly triangular area.

Figure 13.

Suggested relationship between penis tip length and bursa copulatrix length.

Number of specimens measured	56	42	23
Range of $\frac{c}{Bc}$ ratio	2·49 - 7·0	1·33 - 2·29	2·34 - 4·15
Average $\frac{c}{Bc}$ ratio	4·0	1·7	3·1
Bursa copulatrix	 Short	 Long	 Short
Penis tip shape			
Species	*L. rudis*	*L. arcana*	*L. nigrolineata*

1.2.3. The Penis of L. neglecta

In some respects the penis of neglecta is very like that of rudis. It usually has a mucronate tip and the penial glands are similarly positioned. It differs in absolute size, in showing a more limited range of variation in penial gland number and arrangement and apparently relatively larger penial glands (Heller 1975). In neglecta the number of penial glands range from 2-6 and are always in a single row. However, this is not outside the range of variation seen in rudis, hence the only described diagnostic difference in penis separating rudis and neglecta is overall size. Heller (1975) indicates that the largest mature neglecta penis is considerably smaller than any mature penis of rudis.

This, however, only applies to the sites visited in Angelsey. At other sites even the size distinction breaks down. For example some specimens from Eddystone Rock, a very exposed site, were difficult to assign definitely to one species or the other. On the basis of present knowledge they could either be a population of small tessellated rudis or large neglecta without the distinctive wide band on the shell.

Furthermore a population, of what on shell characters were rudis, sampled at Esk Mouth (Scotland) an estuarine situation, which were inhabiting barnacle shells, mature animals no larger than neglecta were found. Peter van Marion (pers. comm.) has also found difficulty in assigning animals from some Norwegian sites to one species or the other.

As an addendum to the problem of the usefulness of the penis in distinguishing rudis and neglecta, it is interesting to note

Hughes' paper (1979). In this he describes a population of rudis from saltmarshes in South Africa, in which the penes all had 6 penial glands in one row. Not only is this a most unusual lack of variation but also places the population at least on this character between the two species.

1.3. PROSTATE CHARACTERISTICS

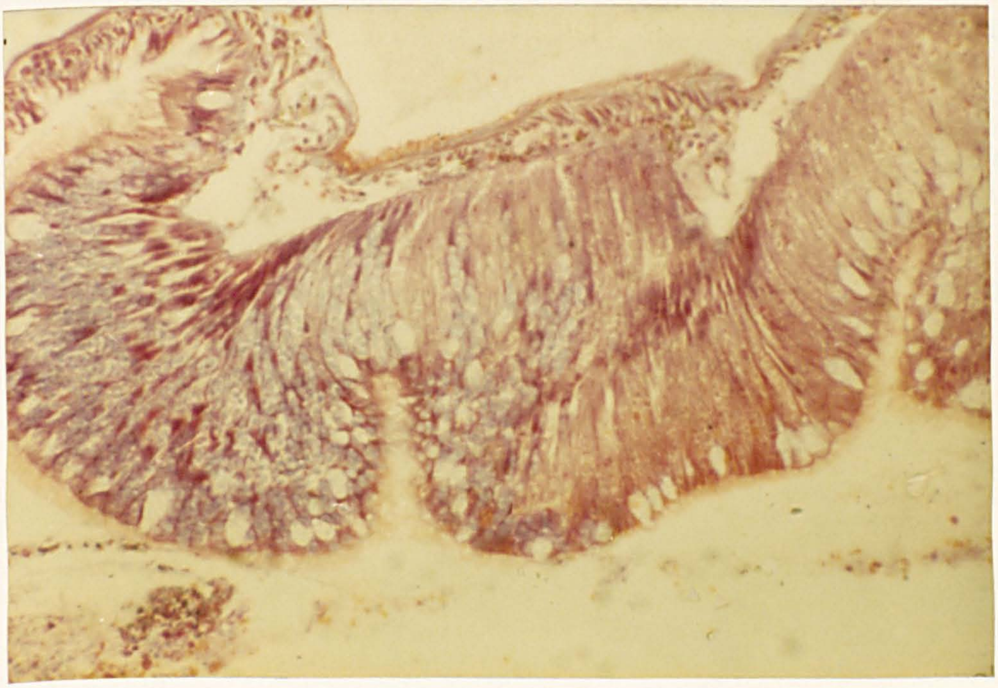
The appearance of the prostate can be a useful ancillary characteristic for distinguishing rudis and arcana males. Prostate appearance in other species was not examined.

In rudis the prostate often looks longer, stretching in an arc round the ciliated field, and is relatively smooth (Plate 7, fig. 12). In arcana the prostate usually appears puckered with transverse folds and comparatively short.

Though of no taxonomic importance, an interesting observation, apparently previously unnoted, was made. In many specimens it was possible to detect a tissue difference (fig. 12) between a small part of the proximal end and the rest of the prostate. This proximal section appears translucent pink in freshly killed animals and the distal (larger) section appears opaque pink. On histological examination the secretory cells of the proximal sections were found to produce a secretion which stained blue with modified Mallory-Heidenhain stain and was formed in round packages. The cells of the distal section produced irregularly shaped packages of an orange-staining secretory product (Plate 8). The precise chemical nature and the function of these secretions is unknown.

PLATE 8.

Oblique section through the prostate (L.arcana) showing the two types of glandular tissue, cells with round,blue-staining packages on the left and cells with irregular, orange-staining packages on the right.



1.4. CILIATED FIELD CHARACTERISTICS

In all rough winkle species there is a localised field of heavily ciliated epithelial tissue which lies between the genital tract and the columellar muscle. The position and extent of this ciliated field (cf. figs 4-8, 12, 14-17 & Plates 1-3,5 & 7) shows characteristic differences in arcana and rudis and has been of great importance in the diagnosis of arcana.

The ciliated field is only readily apparent in animals freshly killed (by briefly boiling them) when usually it is pink and stands out against the white or black of surrounding mantle tissues. In preservation it loses its colour and tracing its true extent can be difficult. In males, because the prostate is similarly pink, the ciliated field appears to be an extension of this gland.

1.4.1. Histology

Specimens of the ciliated field and some surrounding tissues were dissected from live rudis, arcana and nigrolineata, placed in Bouin's fixative, embedded, sectioned and stained with modified Mallory-Heidenhain stain (Cason 1950).

Examination of the sections showed that the ciliated field in all three species and both sexes appeared to be composed of densely ciliated epithelial cells with a few goblet cells (Plates 9 & 10 & figs 16 & 17).

This tissue was discernible in sections of arcana individuals in which the ciliated field was not seen during dissection under x 30 binocular microscope.

The ciliated field of neglecta was not studied histologically.

1.4.2. Function of the ciliated field

Tests made on the direction of beating of the cilia of rudis showed that small particles are carried towards the opening of the mantle cavity; thus their action appears to be to drive something out of the mantle cavity. There are a number of possible candidates for such expulsion e.g. water, particles, pheromones, faecal pellets, embryos, sperm etc. One fact suggests that the expulsion of particles may be its principle function. The field is largest and therefore presumably most important in rudis and this is the only rough winkle species regularly found in muddy habitats where particles are likely to enter the mantle cavity. L. arcana, in which the ciliated field is fairly small, has not been recorded from muddy areas.

1.4.3. Nature of the ciliated field in L. rudis and L. arcana and its use in specific identification.

1.4.3.1 Males

In rudis the ciliated field is comparatively extensive and lies adjacent to the distal end of the prostate between the prostate and the columellar muscle (fig. 14A and Plate 9). In arcana it is generally inconspicuous, usually forming a narrow band (less than the depth of the prostate) beside the ventral

Figure 14.

Ciliated field and prostate characteristics:
A) *L.rudis* B) *L.arcana*

Key: A:- anus
Cf:- ciliated field
Cm:- columellar muscle
Fp:- faecal pellet
Hm:- head mass
Pr:- prostate
R:- rectum
Upb:- unpigmented band

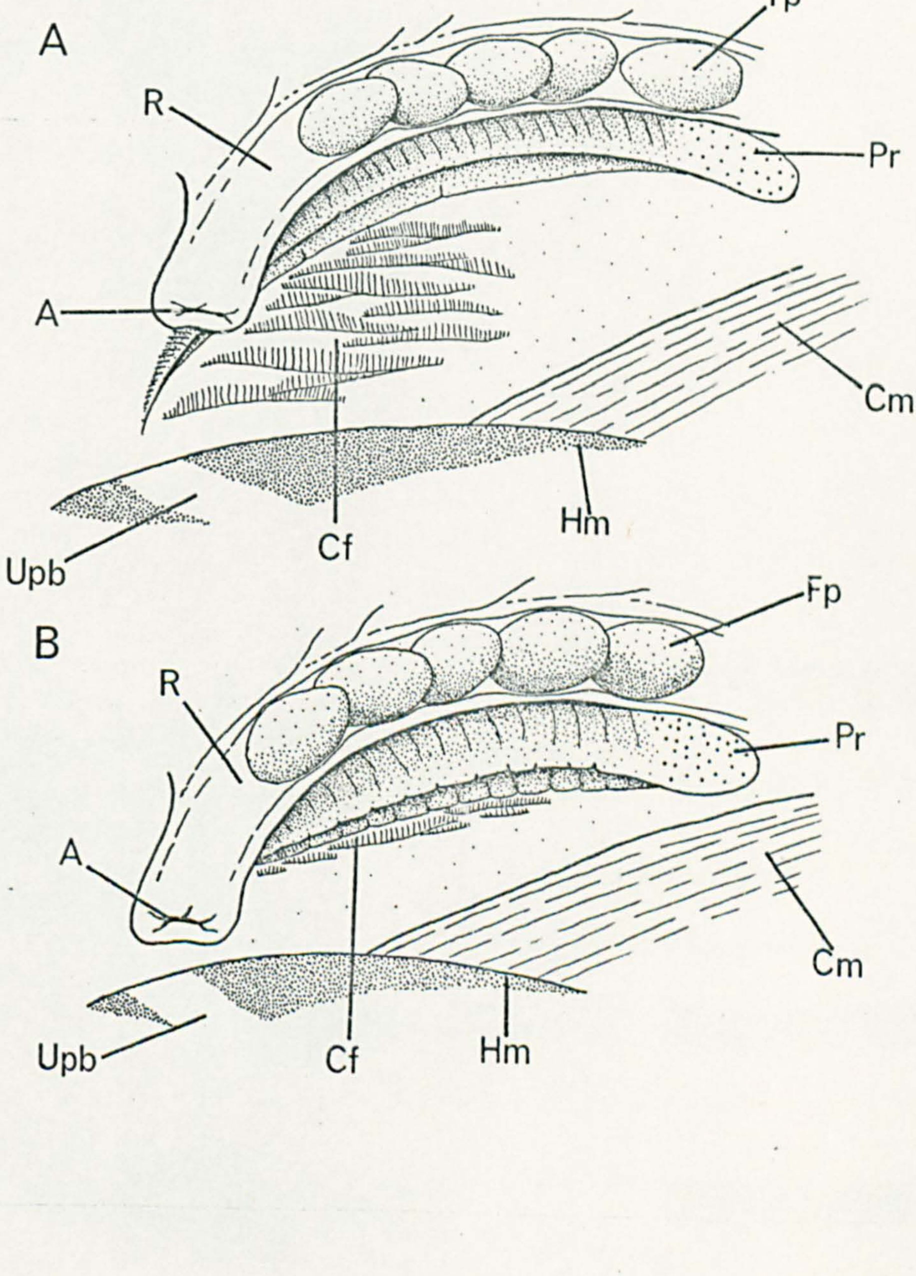
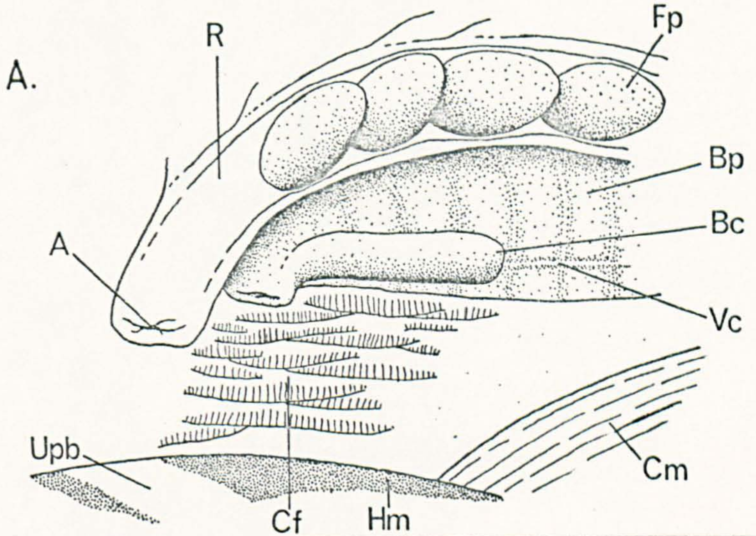

A
Fp
R
Pr
A
Cm
Cf
Hm
Upb
B
Fp
R
Pr
A
Cm
Upb
Cf
Hm

Figure 15.

Ciliated field position and extent in females
A). *L.rudis* B). *L.arcana*

Key: see figs. 1,2,5, & 14.


A.
R
Fp
Bp
A
Bc
Vc
Upb
Cm
Cf
Hm

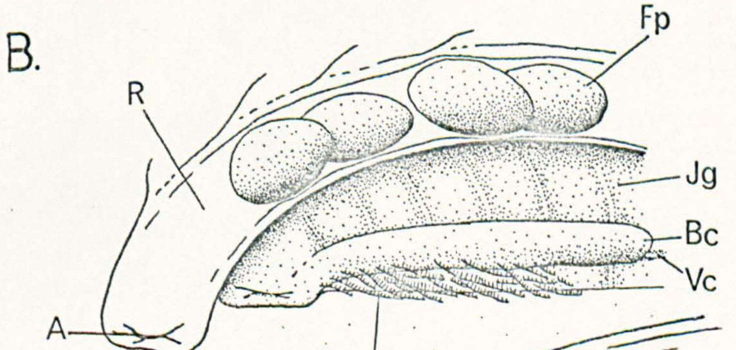
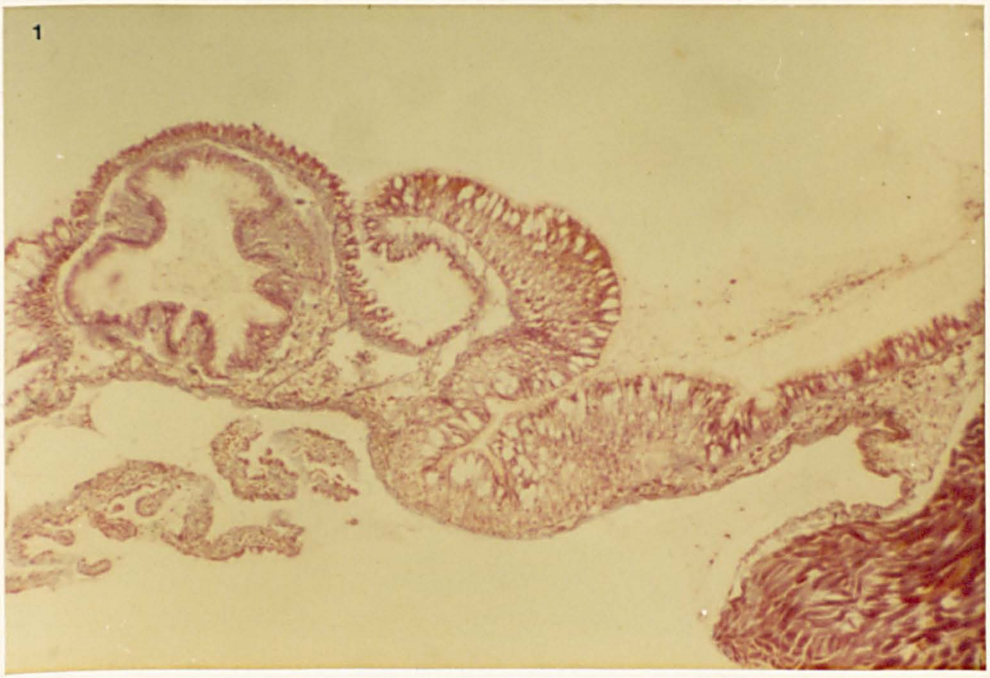

B.
Fp
R
Jg
Bc
Vc
A
Upb
Cf
Hm
Cm

PLATE 9.

Transverse sections through the rectum, prostate and ciliated field.

1). L.arcana 2). L.rudis

1



2

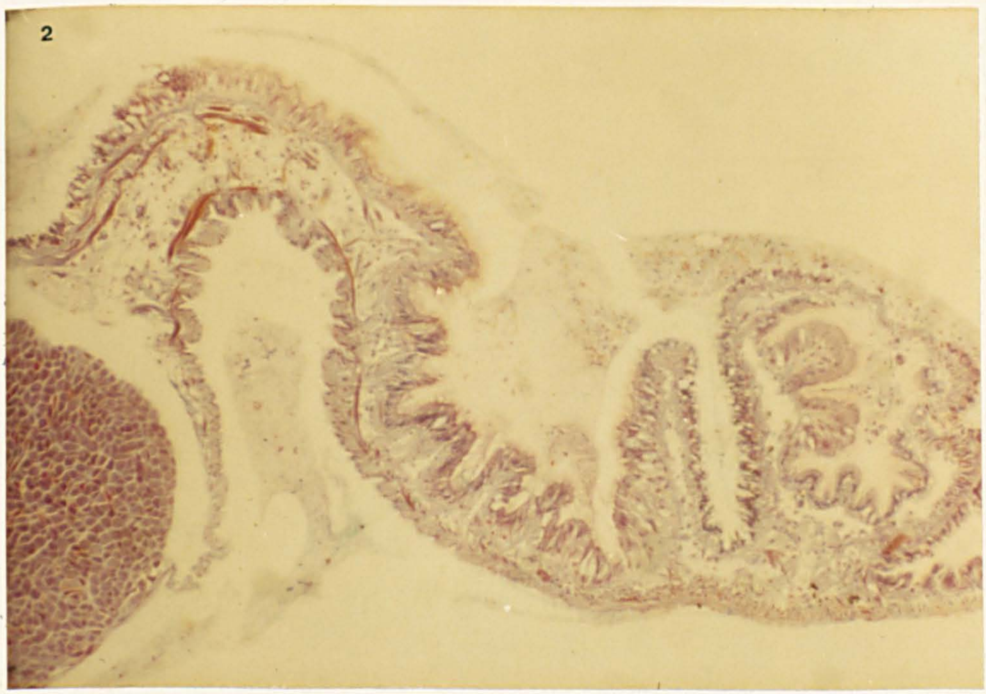
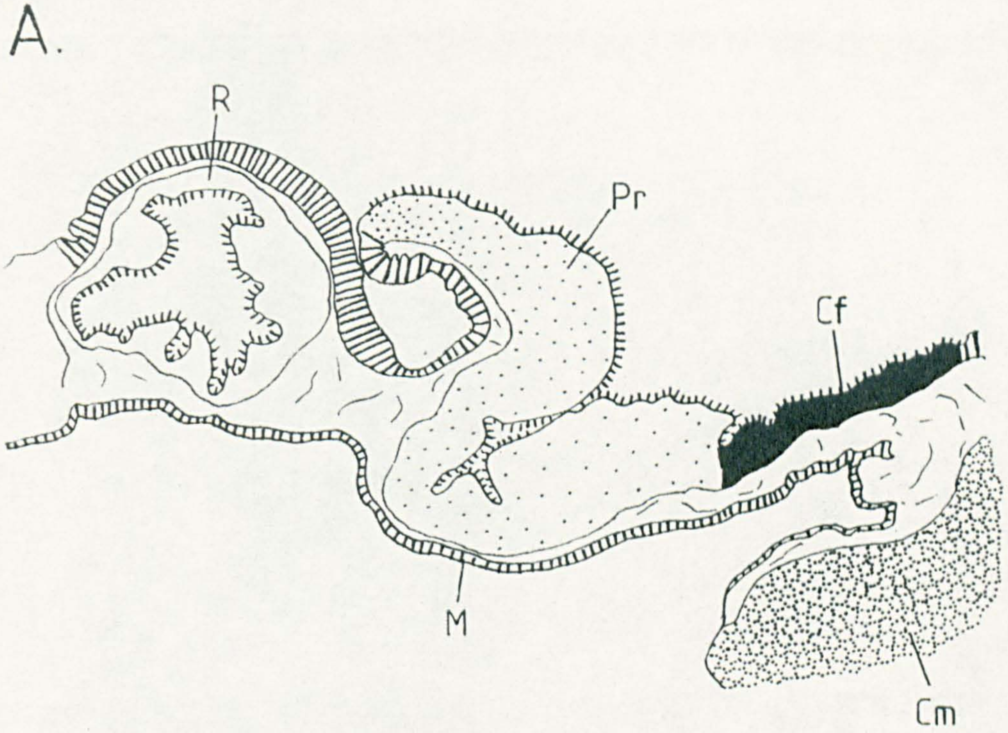


Figure 16.

Figures traced from Plate 9.

A). L.arcana B). L.rudis

Key:
Cf:- ciliated field
Cm:- columellar muscle
M:- mantle
Pr:- prostate
R:- rectum


A.
R
Pr
Cf
M
Cm

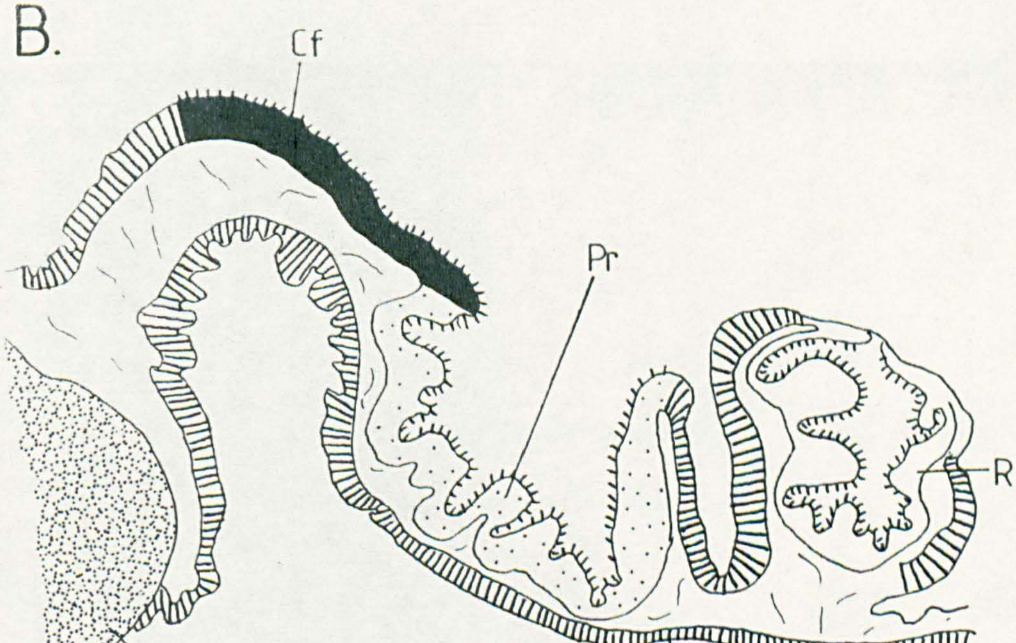
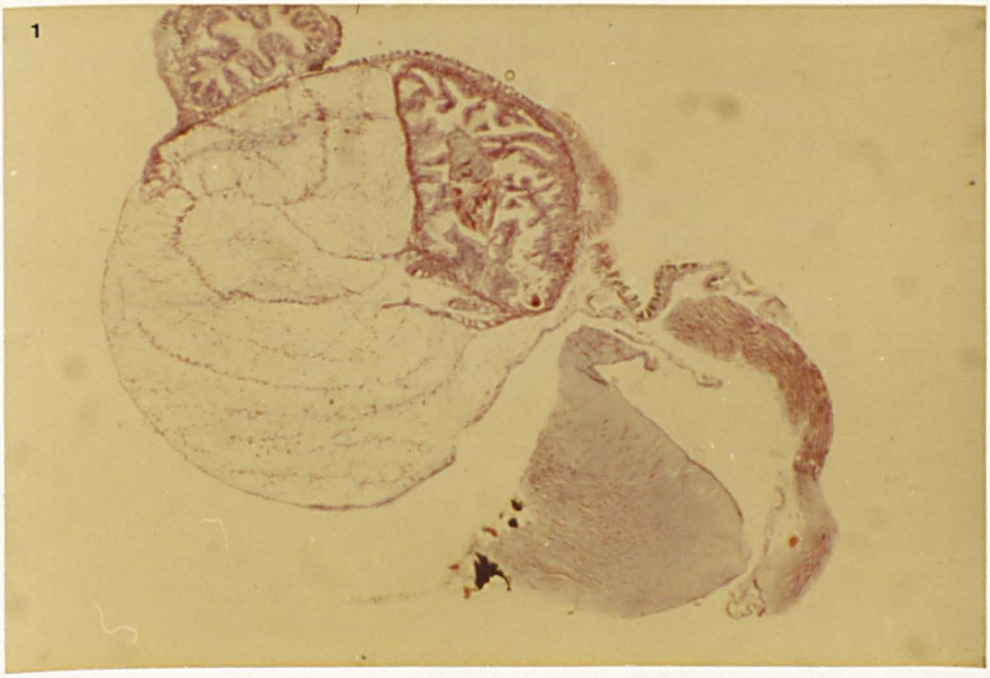

B.
Cf
Pr
R
Cm
M

PLATE 10.

Transverse sections through the oviduct and ciliated field.

1). L.arcana 2). L.rudis

1



2

Figure 17.

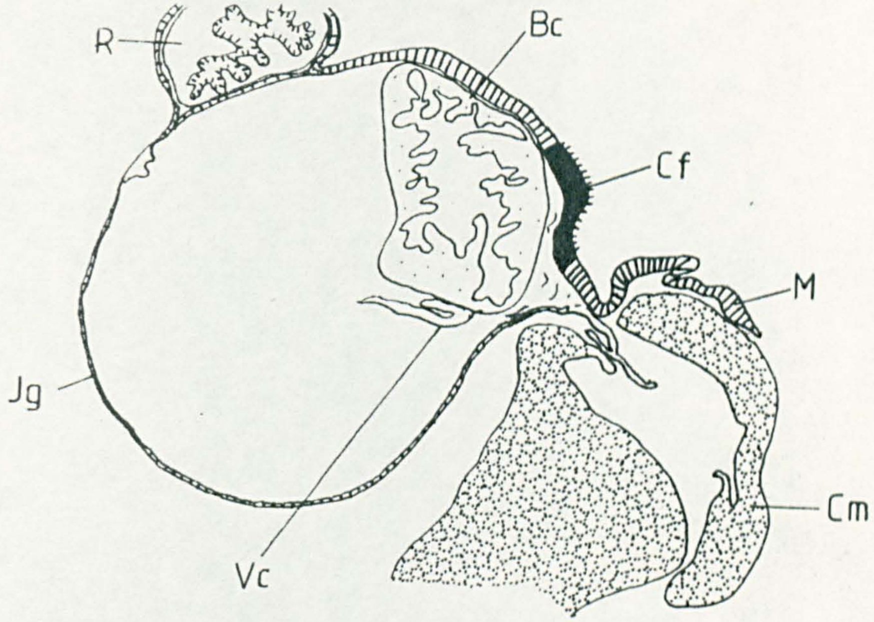
Figures traced from Plate 10.

A). *L.arcana* . B). *L.rudis*

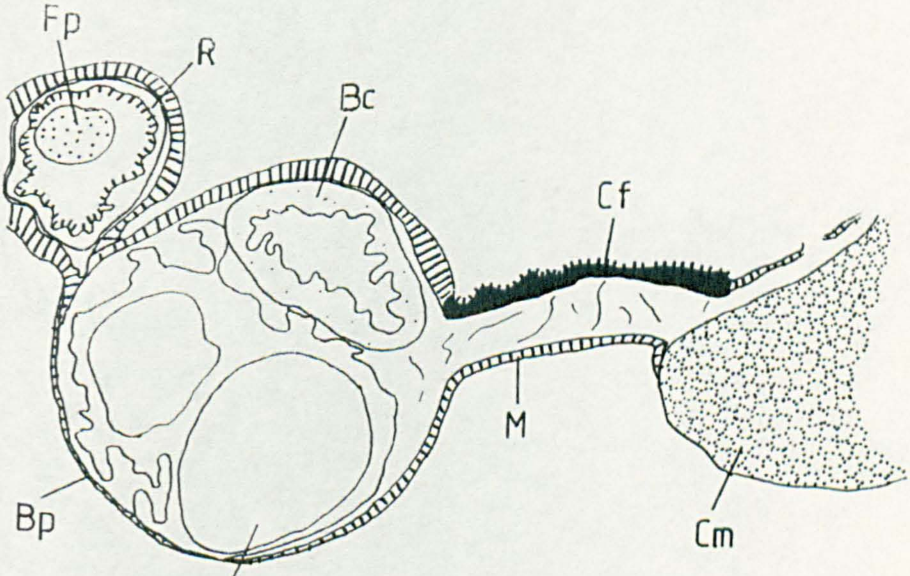
Key:

Key	Term
Bc	bursa copulatrix
Bp	brood pouch
Cf	ciliated field
Cm	columellar muscle
E	embryo
Fp	faecal pellet
Jg	jelly gland
M	mantle
R	rectum
Vc	ventral channel

A.



B.



edge of the prostate (fig. 14B and Plate 9). Because both ciliated field and prostate appear pink, the field can often be detected only histologically in arcana. The difference in extent of the ciliated field can frequently be seen through the mantle (Plate 7 and fig. 12).

The character of the ciliated field is independent of the presence of the penis and is apparent in males without penes (through shedding or parasitic castration) and in males without fully developed penes. It is crucial in identifying and separating males of these species since they can be confused on penial morphology. Males of the other related taxa can be distinguished on shell and penis characteristics (Heller 1975a).

1.4.3.2. Females

The ciliated field in female rudis, like that in the male is very extensive (figs 5-8 & 15A, Plates 1-3) and is frequently thrown into folds which may be seen through the mantle. In arcana the ciliated field is very small when compared to that of rudis. Often it includes a small part of the mantle covering the jelly gland (figs 6-8 & 15B, Plates 1-3). As in the males, the field is scarcely apparent externally and the pallial oviduct appears to lie adjacent to the columellar muscle throughout its length. In rudis, the large ciliated field separates the distal end of the pallial oviduct from the columellar muscle.

The extent of the field can, of course, vary from individual to individual and also from population to population. For example, in the arcana population at Porth Swtan, the ciliated field is barely discernible in most specimens, whereas in arcana from Dunbar it is readily apparent although small.

Female rudis and arcana can usually be easily distinguished on pallial oviduct structure, even when immature. However, the ciliated field characters can be useful in confirming or making specific diagnoses. Females of other related taxa can be easily distinguished by their shells.

1.5 SUB-OPERCULAR PATTERNS

The operculigerous disc of certain British littorinid species develops distinct black and grey markings. This was first observed during work for this thesis in neglecta, and in this species, as well as several others, the pattern of pigmentation is species-specific. There is no previous record of these sub-opercular patterns in the littorinids. However, it was subsequently found that Davis (1966) had recorded a similar feature in Hydrobia totteni. The pattern can be seen through the operculum, except in large specimens of the bigger species in which the operculum is very thick. Like tentacle pigmentation, the pattern increases in intensity with age.

Six species were examined in detail from several sites. Of these three had distinctive patterns, littorea, neritoides and neglecta. The patterns observed in rudis, arcana and nigrolineata were very similar and very variable and not distinct enough to be useful in distinguishing species.

The pattern can be broken down into three elements, an upper crescent, a lower line and a central ring or bar.

1.5.1. L. neritoides

Specimens examined from Filey Brigg, Porth Swtan and Abraham's Bosom all showed the following type of pattern (fig. 188). The upper crescent is typically darkly pigmented with a smaller inner grey crescent. The lower line is continuous with the upper crescent, and extends across the operculigerous disc and turns sharply to run parallel to the crescent for about $\frac{1}{3}$rd of its length. Short parallel lines frequently join the upper crescent to the reflexed part of the lower line, giving a ladder-like effect. The central element is typically a black bar often with a grey area above it.

A large area of the operculigerous disc is thus heavily pigmented and, when the winkle is withdrawn, increases the dark appearance of the aperture.

1.5.2. L. littorea

In this species only one element, the upper crescent, is present (fig. 18A) This is usually completely black. Specimens from three sites, Rhos-on-Sea, Filey Brigg and Porth Swtan were examined and all showed this pattern.

1.5.3. L. neglecta.

The upper crescent is usually only poorly marked in this species. However, the central element, an angled ring, is heavily marked and is joined by a dark line to the lower line which is similarly dark (fig. 18C). Specimens were examined from Filey Brigg, Porth Swtan and Porth Diana.

1.5.4. L. arcana, L. rudis and L. nigrolineata.

In these three species the upper crescent and central element (a ring) are the major components of the pattern. The lower line may or may not be present and all parts of the pattern are very variable in degree of pigmentation. The upper crescent is frequently split into zones of differing greys or black. The ring is also very variable and is not always complete (fig. 18D-F).

1.5.5. Use of the Sub-Opercular Pattern in Specific Diagnoses

The sub-opercular pattern is not particularly useful for specific identification, with one exception, *L. neglecta*. It cannot be used to distinguish *arcana*, *rudis* and *nigrolineata* from one another, and *littorea* and *neritoides* are easily identified by their shells. However, in the identification of *neglecta*, it is of some use. Though the distinctive tessellated or banded patterned shells of *neglecta* can be recognised as such, some *neglecta* have completely black or badly eroded shells and are difficult to separate from juveniles of *rudis* and *arcana*. The sub-opercular pattern can be useful in identifying these *neglecta*. Not only do the juveniles of other species either lack a sub-opercular pattern or it is very faint, but also the distinctive *neglecta* pattern is a good marker. In the size range below 5mm, only *neglecta* individuals have a well-developed pigment pattern.

Though the character of the sub-opercular pattern is only of limited use with British winkles, it is possible that such patterns may be of use in the identification of littorinids from other parts of the World.

Figure 18.

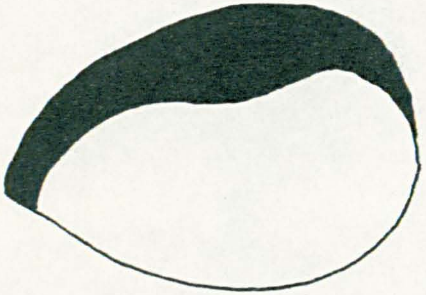
Sub-opercular patterns.

A). L.littorea

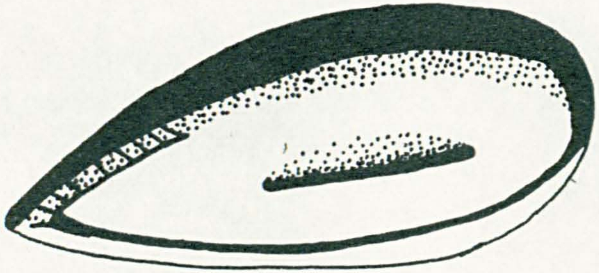
B). L.neritoides

C). L.neglecta

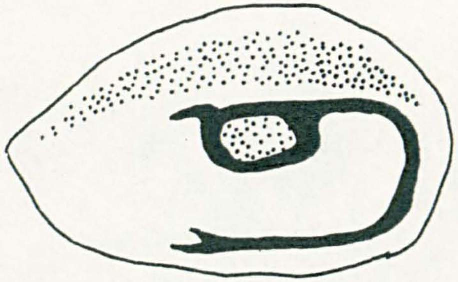
D) - F). Some of the patterns seen in L.rudis, L.arcana and L.nigrolineata.



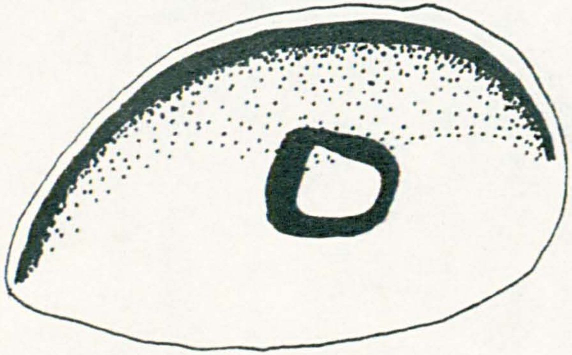
A. *L. littorea*



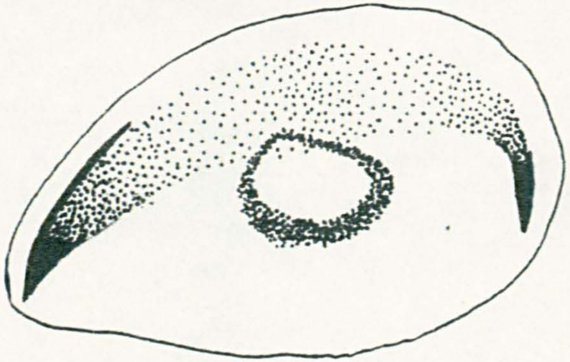
B. *L. neritoides*



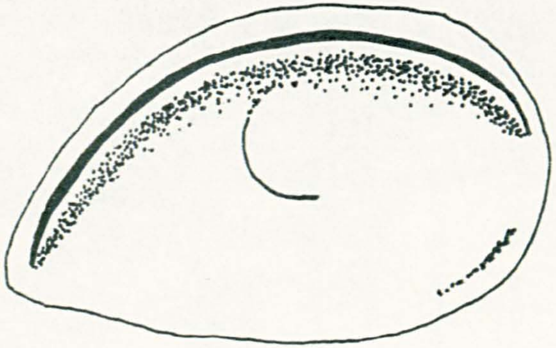
C. *L. neglecta*



D.



E.



F.

1.6. RADULA CHARACTERISTICS

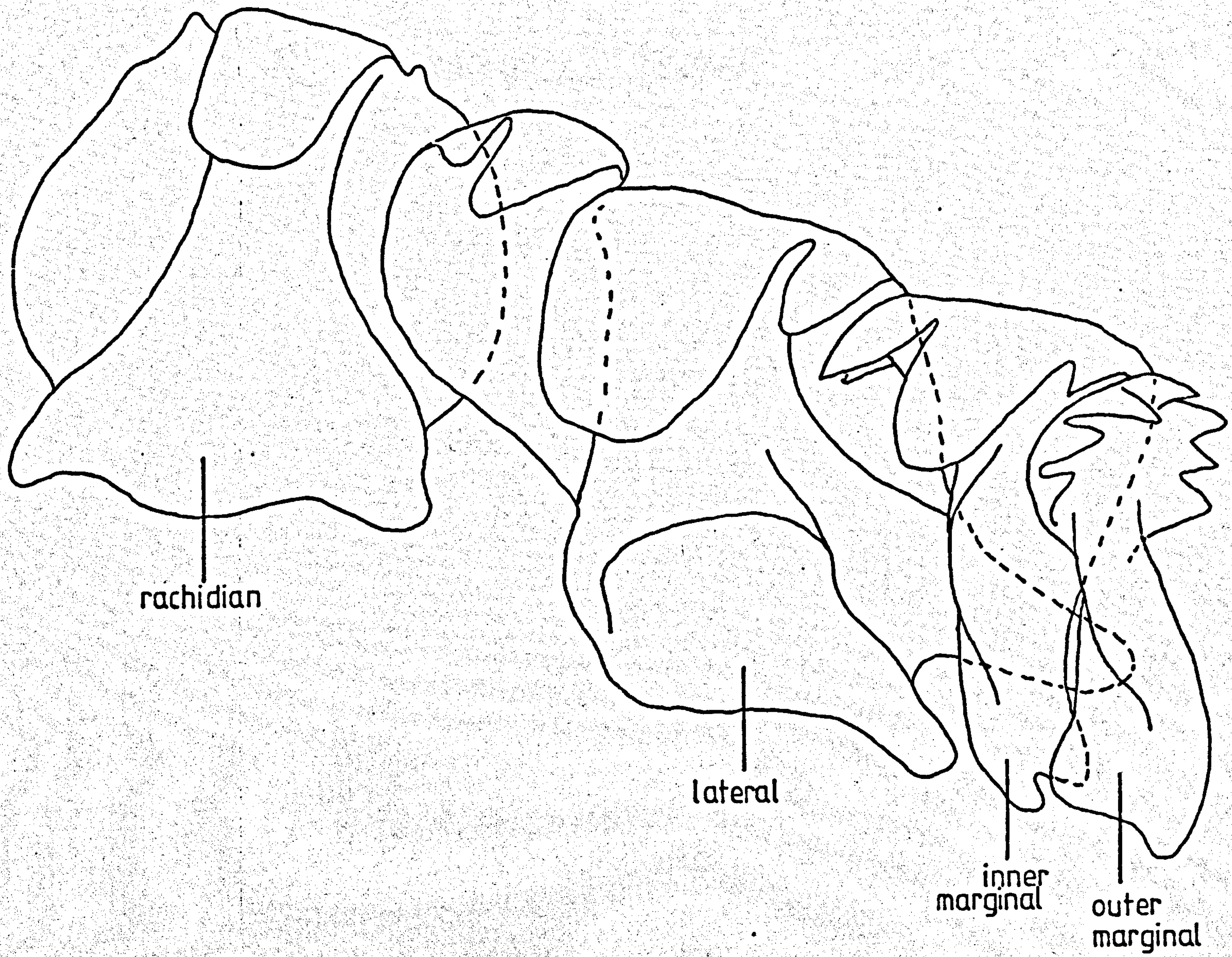
The Littorinids have the taenioglossan pattern of radular structure. The bilaterally-symmetrical central tooth, the rachidian, is fairly large and robust. It is flanked on either side by three other teeth, a large lateral, a smaller inner marginal and a delicate outer marginal tooth (fig. 19). Structurally the inner marginal tooth seems much closer to that of the lateral than to the outer marginal. The main points of affinity are the existence of a bifurcated base and the similarity of the size sequence of the cusps.

The usefulness of the littorinid radula as a taxonomic character has been assessed very differently. At one extreme, Rosewater (1970) has suggested that it is of little value below the generic level, and at the other extreme Sars and Teilman-Friis (quoted in Johansen 1901) separated littorinid species on minor differences such as cusp number. The scanning electron microscope (SEM) has proved particularly useful for the study of inter- and intra- specific variation in molluscan radulae. Bandel (1974) applied it to the examination of inter-specific variation in a world-wide range of Littorinid species and he was able to distinguish all the 18 species he studied on their radula characteristics, thus re-establishing the littorine radula as a useful taxonomic character.

Of the species which occur in Britain, Bandel looked at littorea, saxatilis, obtusata (sensu lato) and neritoides and he noted several species-specific differences. In view of the recent

Figure 19.

Radular teeth of Littorinids.


rachidian
lateral
inner marginal
outer marginal

changes in British littorinid taxonomy it was thought that a similar survey on the eight currently recognised species might provide not only information of species-specific differences (potentially useful in specific identification especially of immature or otherwise difficult individuals) but also show differences that might be correlated with feeding habits.

1.6.1. Preparation of the Radula

1.6.1.1. Dissection and cleaning of radulae

The buccal mass is dissected out complete with attached radula, enveloping tissues and radula sac. It is then cut into three pieces. One cut is made posterior to the buccal mass and the other anterior to the coiled region of the radula (see fig. 20). This separates the older worn teeth and the younger immature teeth from the central section which bears fully formed and unworn teeth, required for study of cusp shape. It is essential to use only these fully mature teeth, preferably from freshly killed specimens. Plate 11 includes scans of a piece of nigrolineata radula from a specimen, which had been preserved for several years in formalin, from the more immature part of the central section. The teeth show marked abnormal back-bending.

The tissues which envelope the radula and obscure the teeth are removed by chemical maceration. The macerating agents most frequently used by previous authors are KOH and NaOH (e.g. Solem 1972, Radwin 1970, Meeuse 1950, Carriker 1943). However, a solution of .5% w/v sodium hypochlorite and 8.25% w/v sodium chloride was found to be superior; it is faster at room temperature

PLATE 11.

SEM photomicrographs.

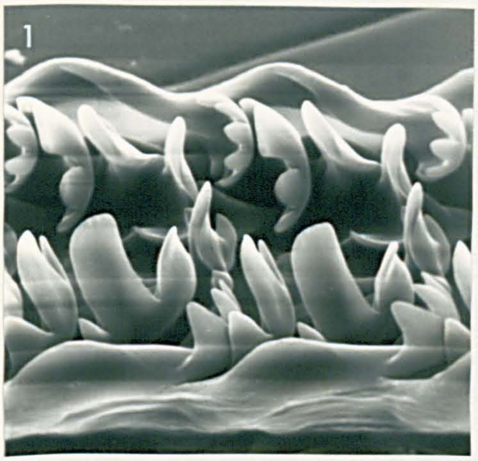
1). & 2). Abnormal backbending of cusps in a radula of *L.nigrolineata* which had been preserved for a long while in formalin.

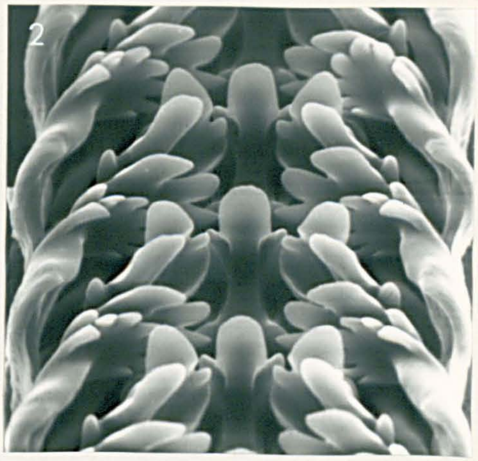
3). & 4). *L.neritoides*, adult outer marginal teeth.

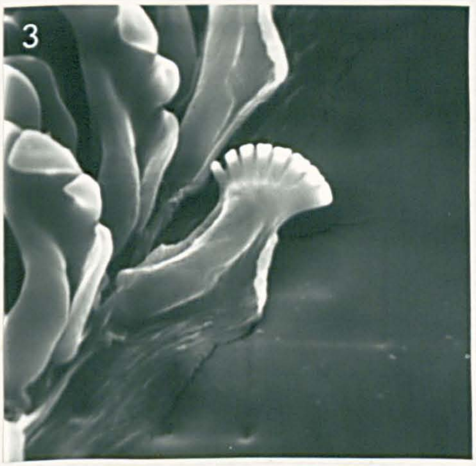
5). *L.nigrolineata*, adult outer marginal teeth.

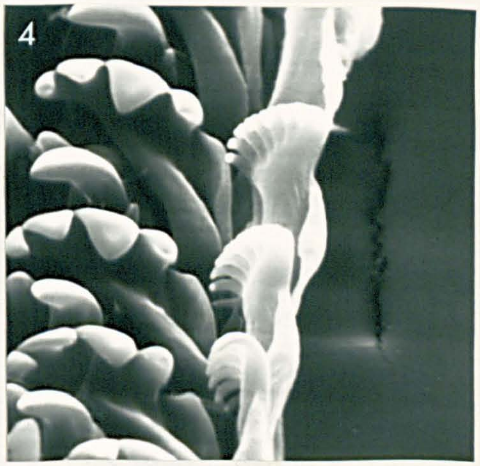
6). Apical cusps on the rachidian tooth in *L.neritoides*

7). & 8). Similar, but less developed, apical cusps in juvenile *L.arcana* and *L.obtusata* respectively

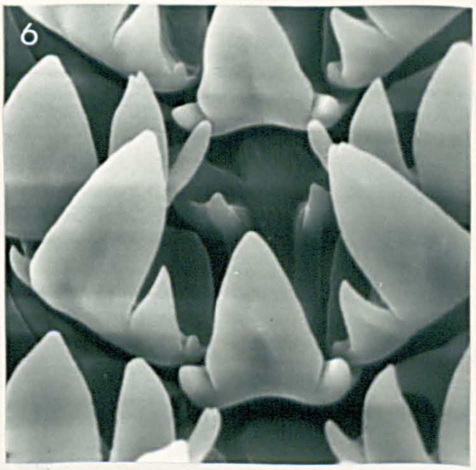

1

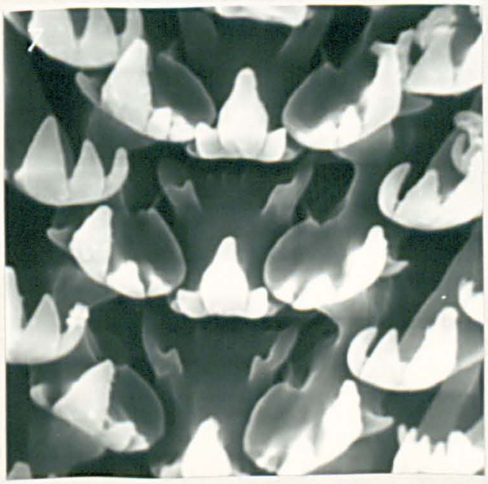

2

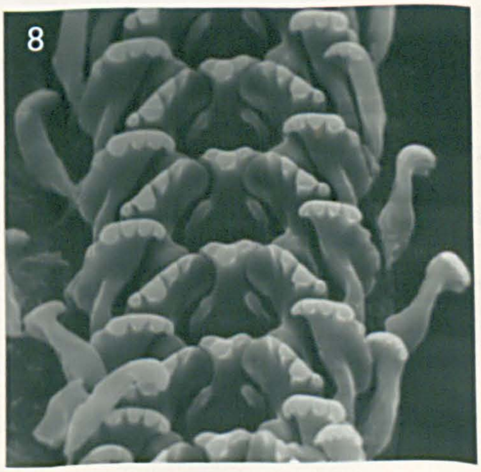

3


4


5


6


7


8

(5-30 minutes depending on size of radula) and it is not caustic. Its main disadvantage is that it is very corrosive to metal, including stainless steel. The solution is obtainable at most chemists under the trade name of Milton sterilizing fluid (which requires dilution by 50%). The use of this macerating agent was suggested to me by Dr. J.D. Taylor (British Museum, Natural History). The radula should not be left in the diluted Milton solution longer than required to macerate the surrounding tissues since the radula membrane is softened after prolonged ($\simeq 1\frac{1}{2}$ hours) immersion. The radula is then rinsed in a jet of water and placed in 70% alcohol in which it can be stored until needed.

1.6.1.2. Mounting the radula for SEM

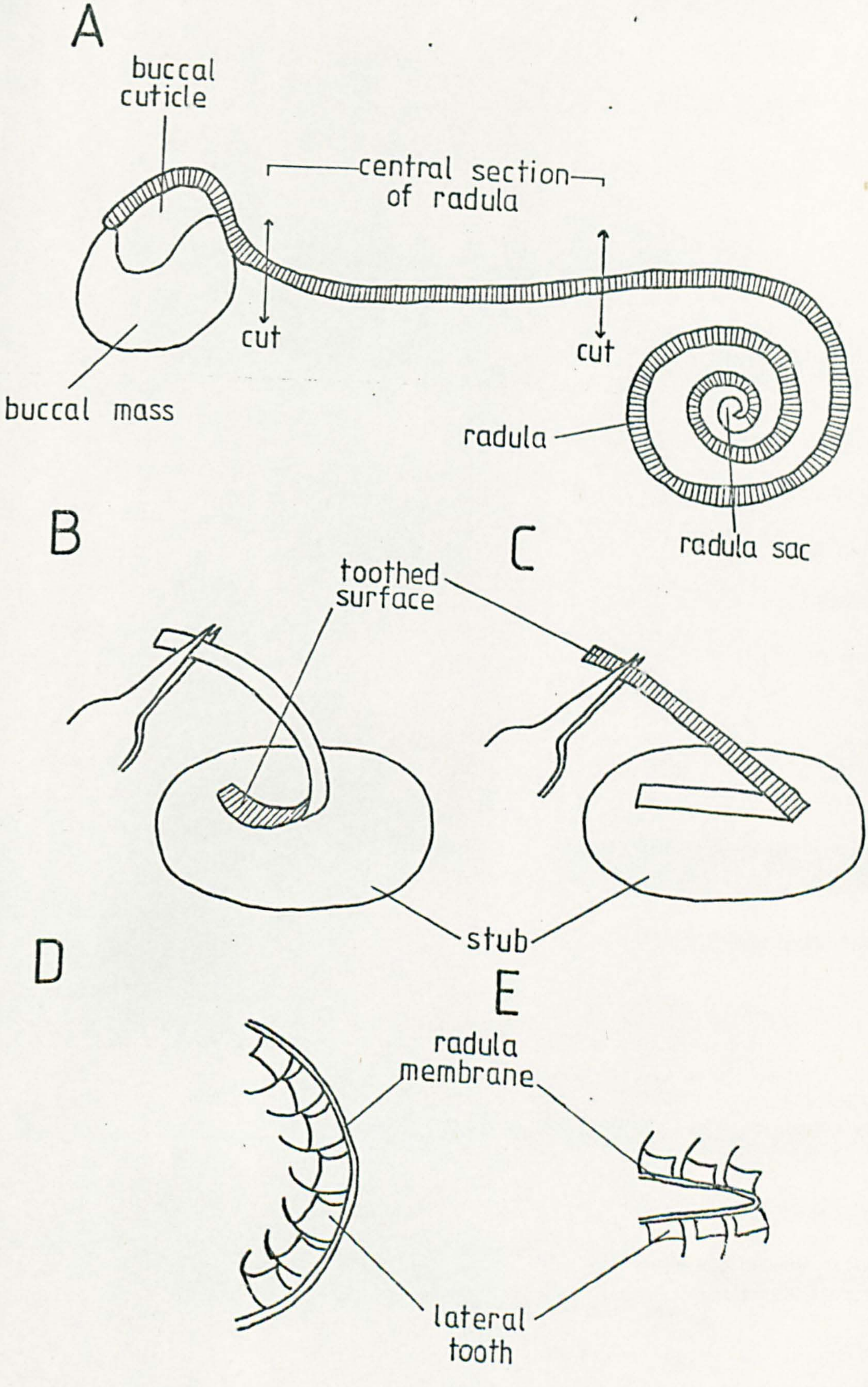
A dab of Durafix glue placed on the stub is flattened and left briefly to become sufficiently tacky so that the radula placed on it sticks to it but does not sink into it. The radula is most easily mounted on the stub under a binocular microscope. Often, especially with very small radulae it is difficult to see which side of the radula membrane bears the teeth. However, this can be ascertained by a quick test: the radula ribbon is held at one end with fine forceps and the free end is pressed against the stub; it will then bend either into an arc (fig. 20B) in which case the teeth are on the inside surface or fold back on itself (fig. 20C).

This effect is primarily due to the shape of the lateral teeth which prevent the radula folding back on itself when the teeth are on the inside surface (fig. 20D) but not when they are on the outside (fig. 20E).

The mounted specimens and stubs were then coated with

Figure 20.

Mounting radulae for SEM work.


A
buccal cuticle
central section of radula
cut
cut
buccal mass
radula
radula sac
B
C
toothed surface
stub
D
E
radula membrane
lateral tooth

gold-palladium and examined with a Cambridge Mk IIa Stereoscan.

1.6.1.3. Scanning

The use and usefulness of the SEM for radula studies has been discussed at length by Solem (1972). The SEM proved particularly useful in the study of radulae from very small specimens. The appearance of each radula was recorded from six standard positions a) anterior b) anterior oblique c) lateral d) posterior oblique e) posterior (these five positions viewed from $\simeq 45^\circ$ to the radula) f) directly above ($\simeq 90^\circ$). This permitted accurate counting of the number of cusps on each tooth and gave a good idea of the shape of the cusps.

1.6.1.4. Preparing radulae for light microscopy

Though most radulae were examined using the SEM, some were examined using a light microscope and the number of cusps per tooth recorded.

Counting numbers of cusps under a light microscope is facilitated by dissociating the teeth of the radula, since if the radula is mounted whole, neighbouring teeth obscure the cusp number of individual teeth. Dissociation is achieved by leaving the radula in Milton solution for several hours, rinsing, and subjecting the softened radula to an ultrasonic bath.

1.6.2. Ontogenetic Changes Affecting Radula Characters

The intensive study of the radulae of British Littorinids covered the age range of each species and, as well as giving information about specific characteristics (or lack of them) also led to the unexpected discovery of ontogenetic change affecting such characteristics as cusp shape and cusp number. These ontogenetic changes affect the usefulness of the radula for specific identification.

1.6.2.1. Ontogenetic changes in cusp number

In all eight species some decline in cusp number with age was found, particularly in the marginal tooth. The lateral teeth were similarly affected in both species of flat winkle. The observations are summarized in Table 2, and shell height against outer marginal tooth cusp number is plotted (figs. 21-23) for each species. The data are given in Appendix A, table 3. Intra-radula variation in cusp number was frequently observed; such radulae were omitted from the data plotted in the graphs.

Decline in cusp number affected particularly *arcana* and *obtusata*.

1.6.2.2. Ontogenetic changes in cusp shape

Radulae from the youngest to the oldest available specimens of each species were examined using the SEM and their appearance recorded from six standardised positions (see section 1.6.1.3).

Photographic records of eight specimens of each species, selected to cover as wide a range of ages as possible, are reproduced in Plates 12-27. In the case of *L. rudis* radula 1 is from

TABLE 2.

Range of variation in cusp number observed

Species	Cusp formula R	L	IM	M	Number of specimens examined	Number of specimens showing intra-radula variation in marginal tooth cusp number
L. arcana	3	4	4	5-10	47	3
L. rudis	3	4	4	5-8	20	3
L. nigrolineata	3	4	4	5-8	47	2
L. neglecta	3	4	4	6-8	20	0
L. neritoides	3+2	3	4	5-8	30	1
L. littorea	3	4	4	4-7	23	4
L. obtusata	3or5	4-6	5-6	5-10	26	0
L. mariae	3or5	4-6	5-7	6-9	18	2

NOTE M :- outer marginal

Figure 21.

Ontogenetic decline in cusp number.
I. L.arcana, L.rudis and L.nigrolineata

ONTOGENETIC DECLINE IN CUSP NUMBER. I.

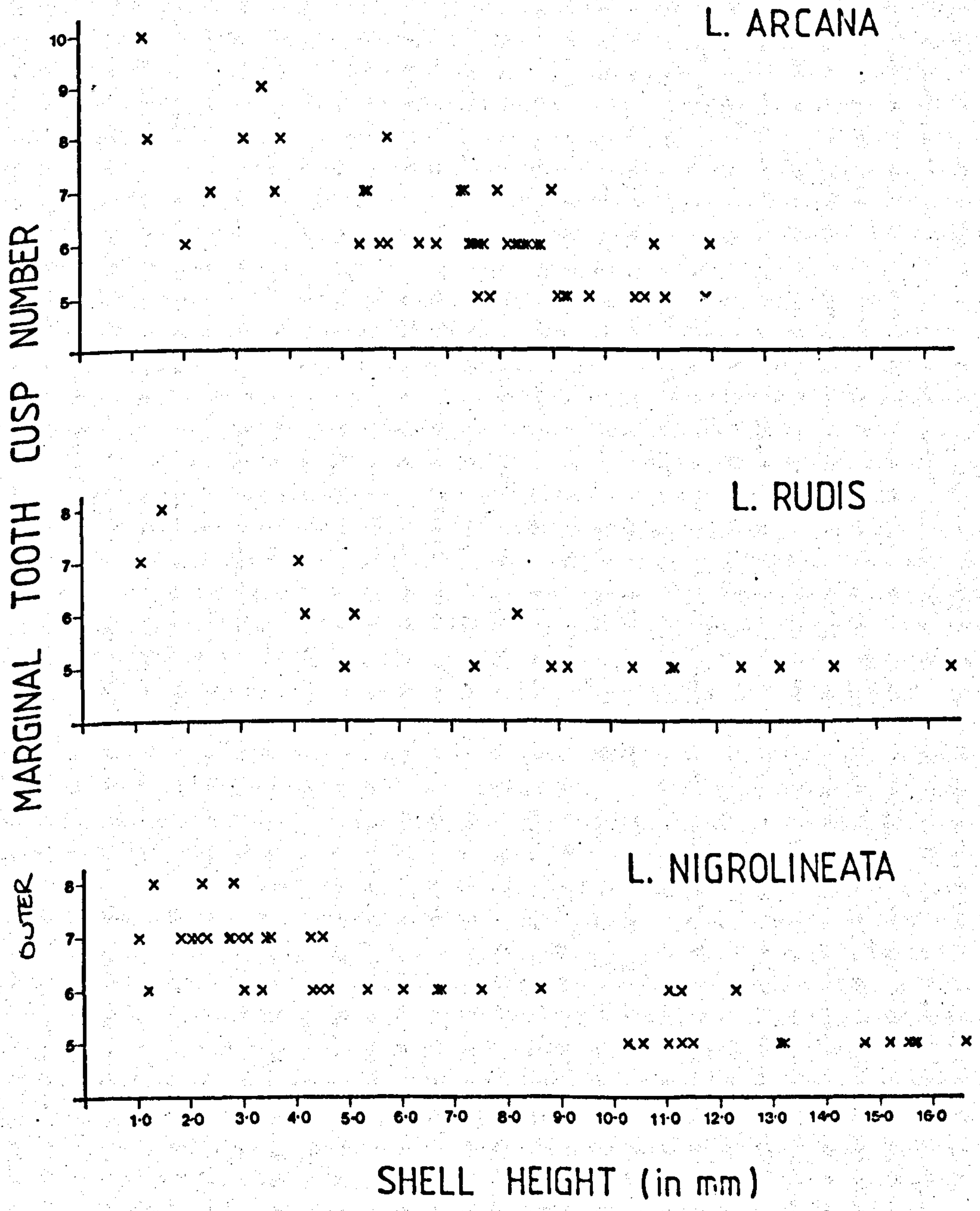


Figure 22.

Ontogenetic decline in cusp number.

II. *L.neglecta*, *L.neritoides* and *L.littorea*

ONTOGENETIC DECLINE IN CUSP NUMBER. II.

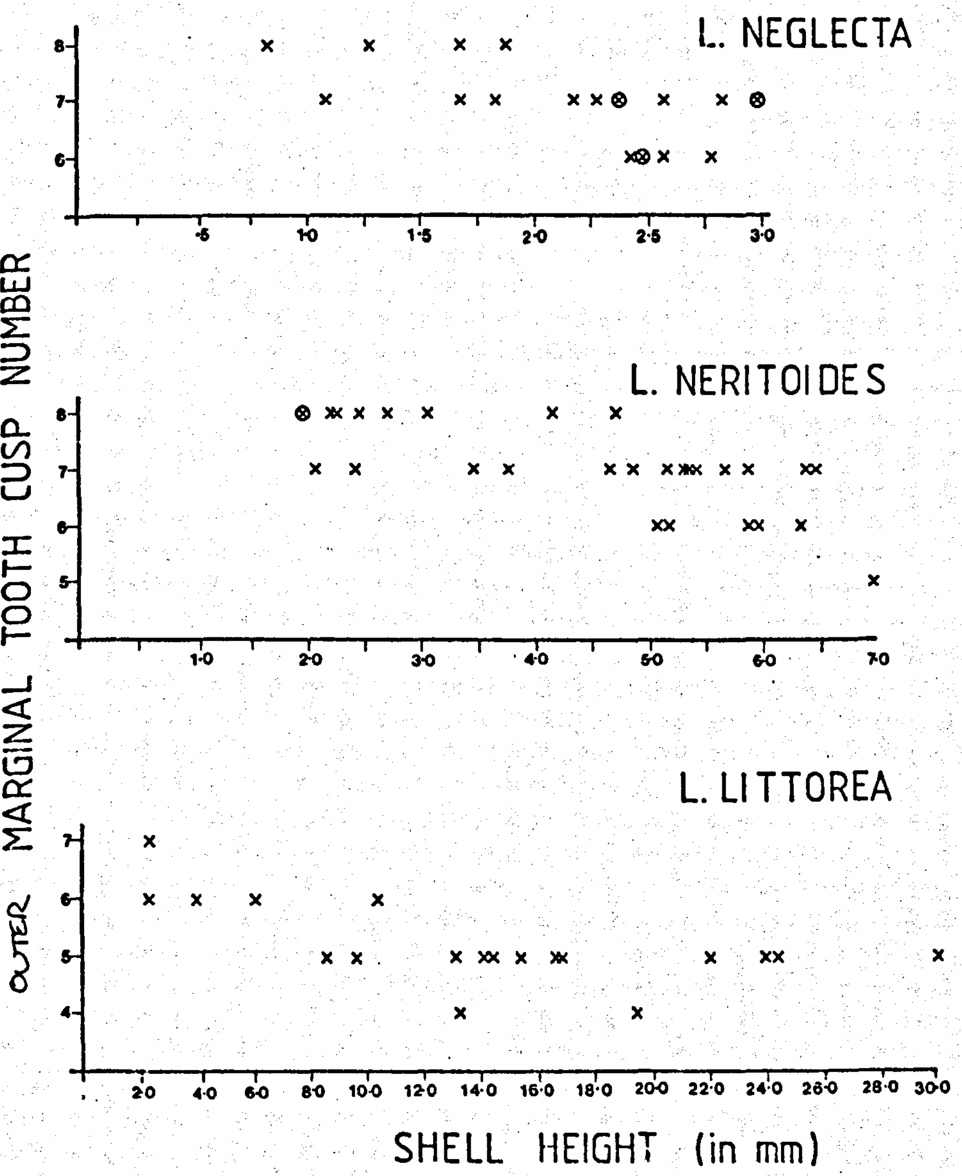
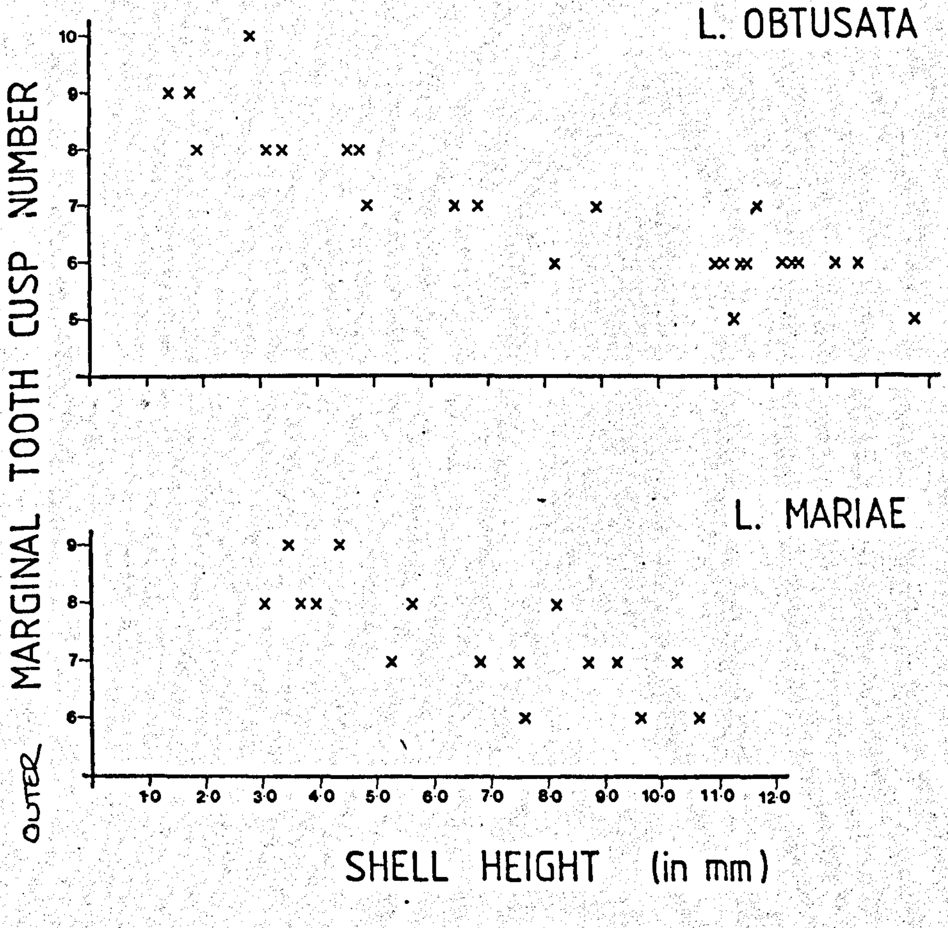


Figure 23.

Ontogenetic decline in cusp number.
III. *L.obtusata* and *L.mariae*

ONTOGENETIC DECLINE IN CUSP NUMBER. III.



a specimen extracted from the brood pouch.

In five of the eight species, *arcana*, *rudis*, *nigrolineata*, *obtusata* and *mariae*, there is a distinct change in cusp shape from juvenile radulae, with sharp-pointed cusps, to adult radulae, with blunt cusps. This change is particularly marked in *obtusata*, *mariae* and *nigrolineata*. In the three other species, *littorea*, *neritoides* and *neglecta* the sharp-pointed cusps are retained throughout life.

1.6.2.3. Possible reasons for ontogenetic changes in radulae and functional significance of cusp shape.

The sharp pointed cusps of the radulae of juveniles were at first thought to be an adaptation for escaping from the egg membrane. The radula is used by juveniles of oviparous and ovoviviparous species to cut through the egg membrane and so escape. Plate 6 shows such a young winkle in the act of hatching. However, since sharp-pointed cusps are produced for a considerable period after hatching this suggests that a different explanation is needed.

If the premise is accepted that the shape of the cusps is not merely an expression of phylogenetic difference (as Bandel, 1974, suggests) but is important in efficiency of food gathering, it therefore seems likely that sharp and blunt cusps are adapted for different purposes, either for collecting different types of food, or for collecting food from different textures of surface. Taking the first suggestion, if the blunt cusps, of adult *nigrolineata*, *mariae* and *obtusata*, are interpreted as shovels and the sharper cusps, of juveniles and other species, as rakes, a difference in type of food captured might be expected. The ontogenetic change in cusp shape might then indicate a difference in food source and might be interpreted as avoidance of intraspecific competition for food either through collection of different foods from one area or through occupation of different areas which support different foods. Alternatively in the second suggestion, sharper cusps may be advantageous for collecting food from rough surfaces and blunt cusps from smooth surfaces. The very sharp cusps common to juveniles of all species might then be interpreted as a reflection of the relative sizes of the animal and the texture of the food-bearing surface. The tiny, sharp cusps might be able to extract food from the micro-crevices into which the larger cusps of the adult radula will not fit. Ontogenetic change might therefore be related to size of the animals relative to the texture of the rock surface.

Furthermore, it can be argued from the original premise that since arcana and rudis, which are of comparable size and habitat, have very similar radula patterns and show similar ontogenetic changes, they are feeding on similar foods and/or substrates. Hence the possibility of niche separation of these species through food specialisation seems unlikely.

1.6.3. Assessment of the Usefulness of the Radula in Specific Identification

Few species - specific characteristics were found and ontogenetic changes complicate the use of the radula for specific identification. In general, radulae can only be described as of a type which might be exhibited by a number of species.

The radulae fall into three groups

a) the type exhibited by nigrolineata, rudis, arcana, neglecta and littorea, with spoon-like outer marginal teeth and major cusps longer than wide. With the possible exception of adult nigrolineata, which have square-ended cusps, the radulae are nearly indistinguishable.

b) the type exhibited by obtusata and mariae which have spoon-like outer marginal teeth and major cusps wider than long. Differences in the lateral tooth cusp number and shape of marginal tooth cusps have been used as a taxonomic character by Reimchen (1974). However, as shown in section 1.6.2 both species show variation in cusp number of the lateral tooth. Variation was also found by Goodwin and Fish (1977). Also, though the shapes of the cusps of the marginal teeth do seem to be different in the adults of the two species, the change in shape with age makes it a difficult character to use.

c) the type exhibited only by neritoides which has comb-like marginal teeth and major cusps longer than wide. The difference in marginal tooth shape is illustrated in Plate 11. Also rachidian teeth of neritoides have apical cusps (see Plate 11) which Bandel (1974) indicates do not occur in other British species. However, similar, though less well-defined cusps appear in radulae of other species, though only in very young individuals.

Hence the radula character is not particularly useful in specific identification.

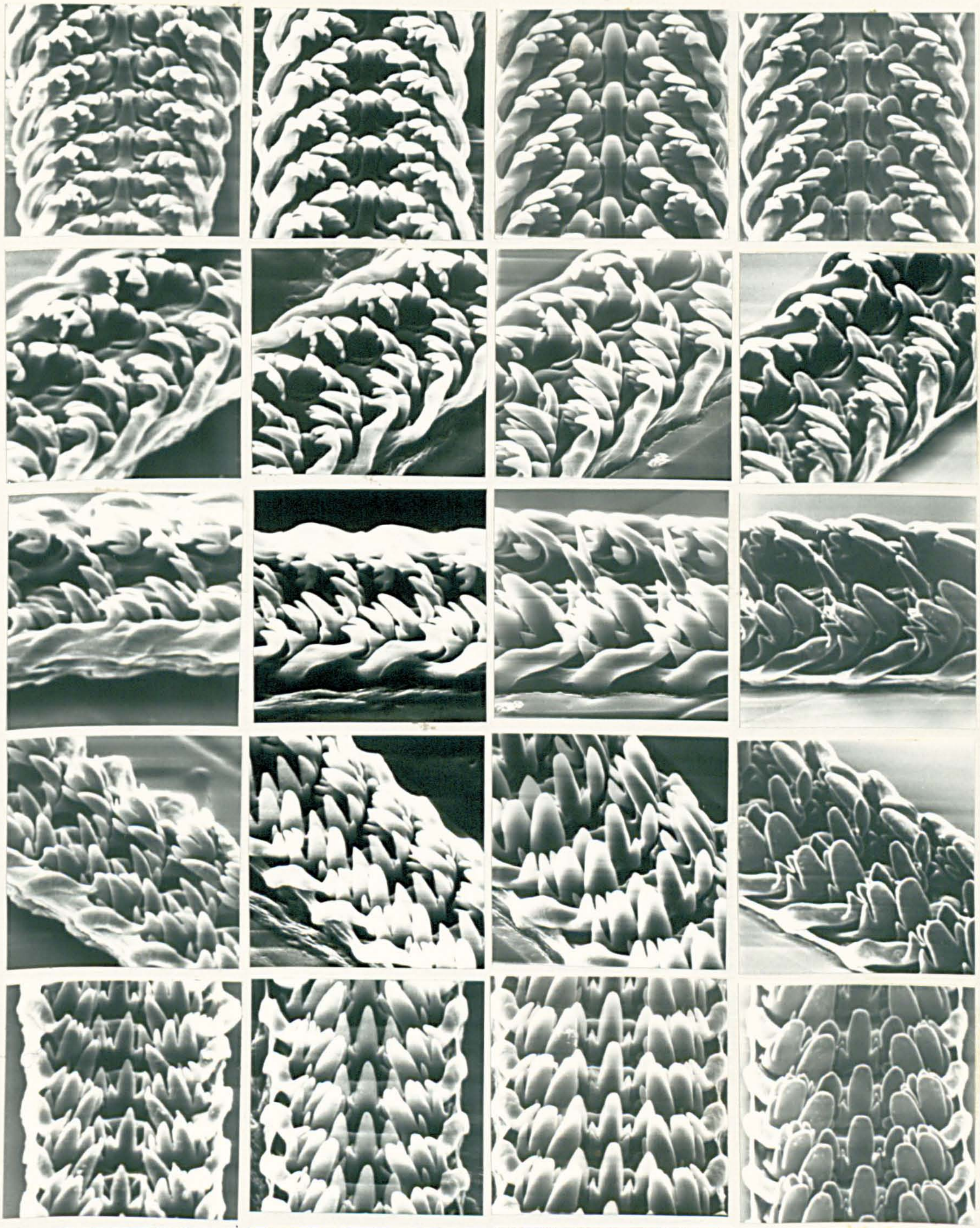
PLATES 12 - 27

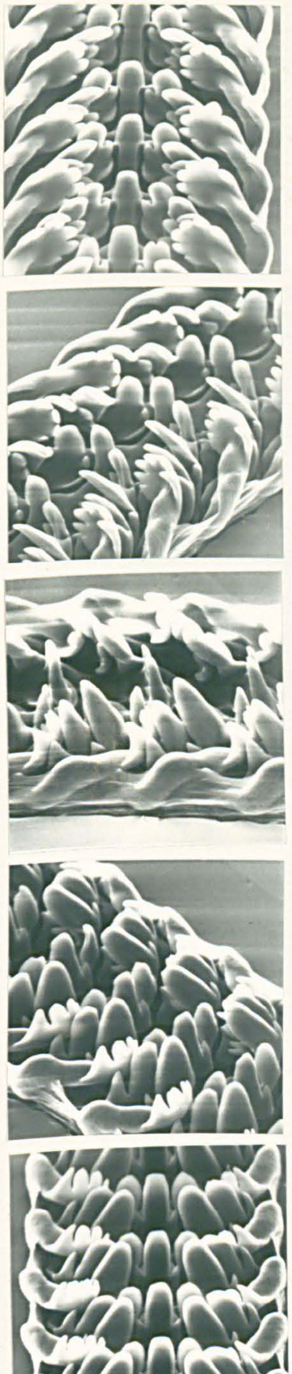
SEM photomicrographs of radulae: Each radula is photographed from six standard positions (see p.36). Eight radulae of each species have been selected to illustrate ontogenetic changes and species characteristics. The shell heights, of the individuals from which the radulae illustrated came, are given in the preceeding plate legend.

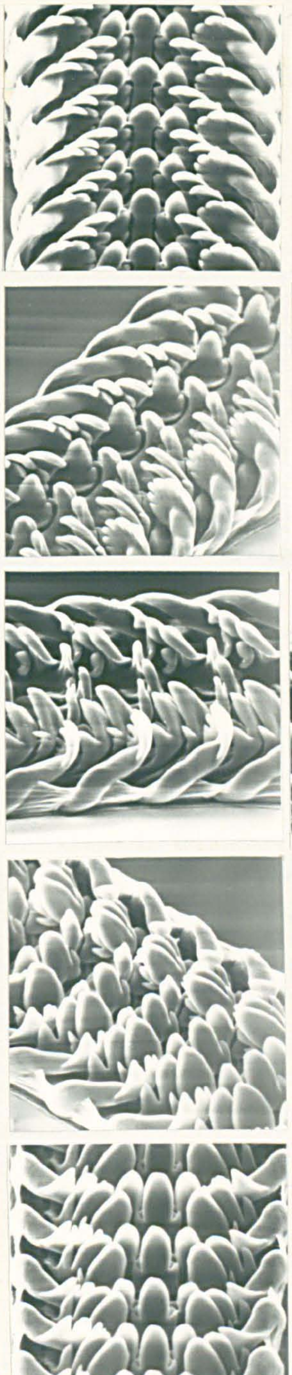
PLATES 12 & 13.

L. arcana radulae

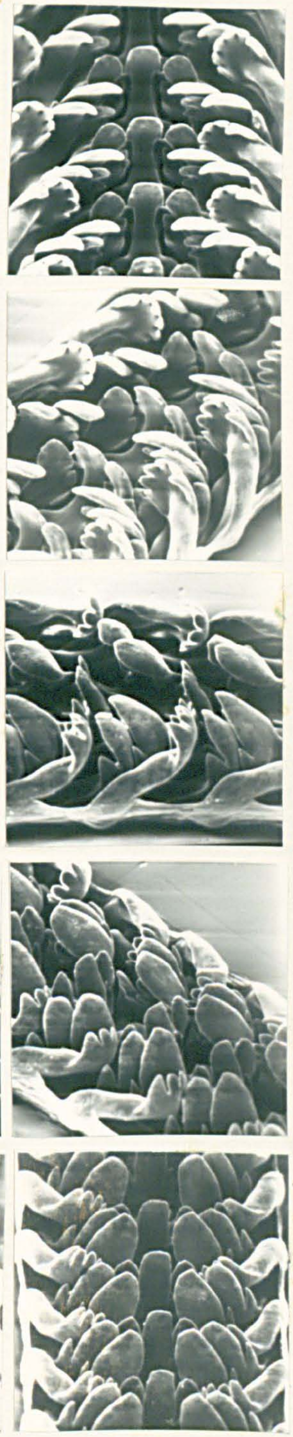
Radula No.	Shell heights
1	1.3mm
2	2.0mm
3	3.8mm
4	5.2mm
5	7.45mm
6	8.95mm
7	9.55mm
8	11.95mm


1
2
3
4


5


6

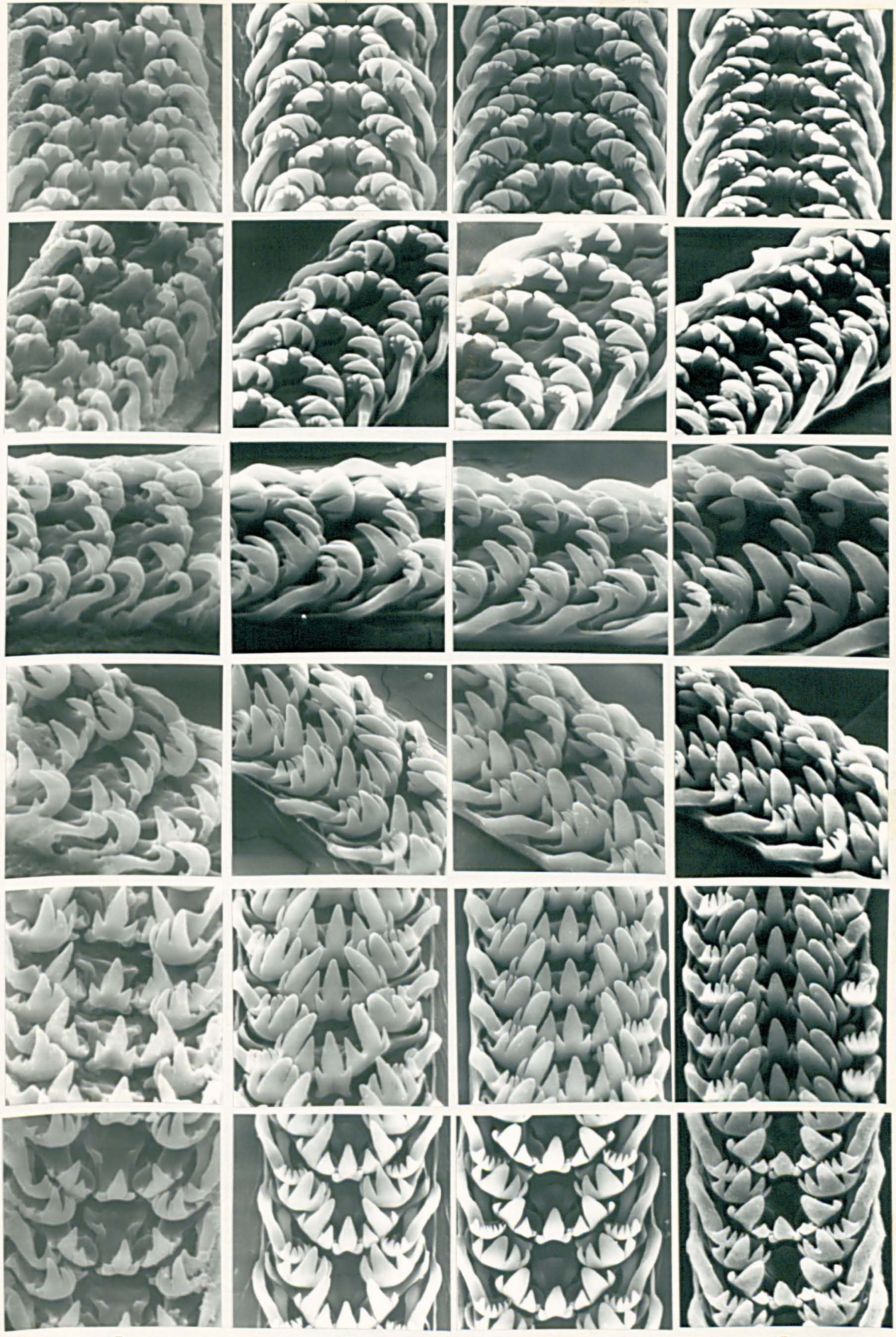

7


8

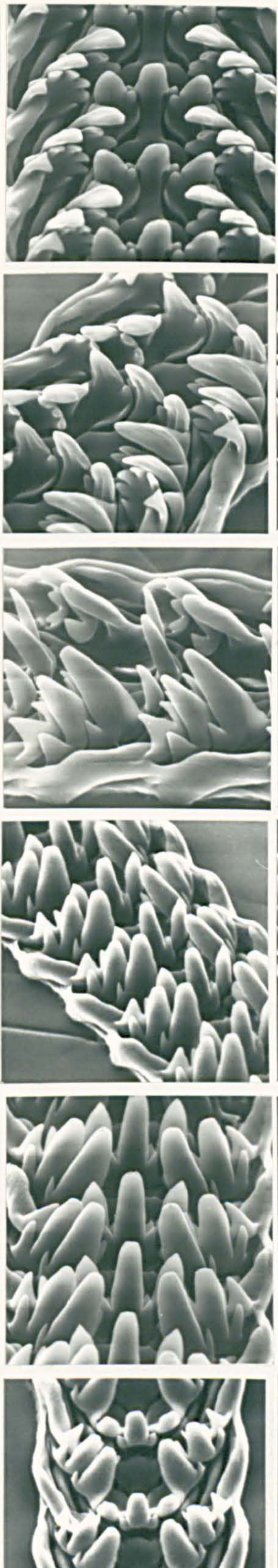
PLATES 14 & 15.

L. rudis radulae.

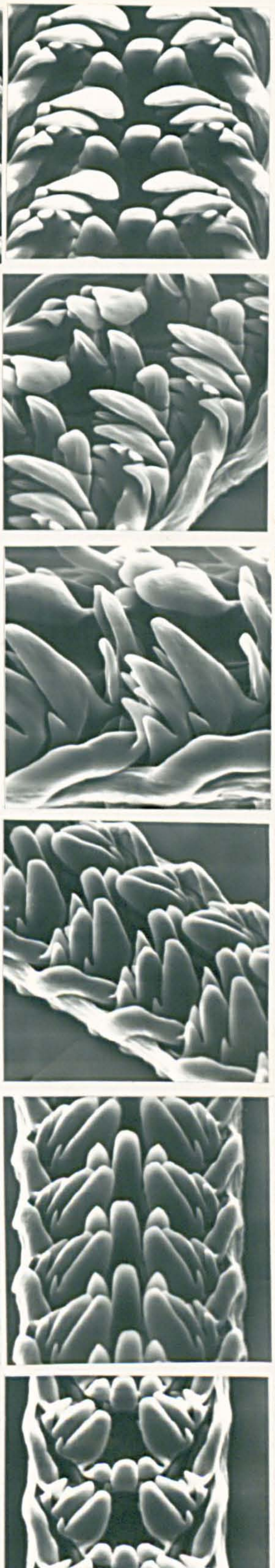
Radula No.	Shell heights
1	.6mm
2	1.1mm
3	1.5mm
4	4.1mm
5	5.15mm
6	7.4mm
7	11.15mm
8	13.2mm


1
2
3
4

5



6



7

8

PLATES 16 & 17.

L.nigrolineata radulae.

Radula No.	Shell heights
1	1.2 mm
2	2.2 mm
3	2.8 mm
4	6.0 mm
5	6.7 mm
6	8.55mm
7	10.5 mm
8	11.0 mm


1


2

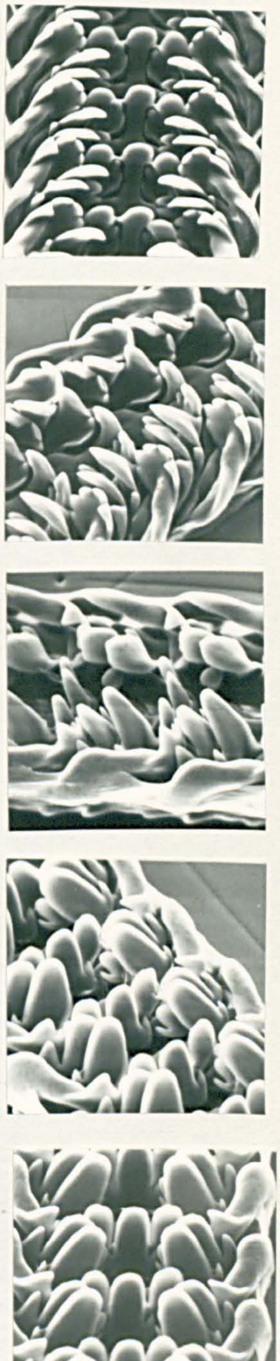

3


4

5

6

7

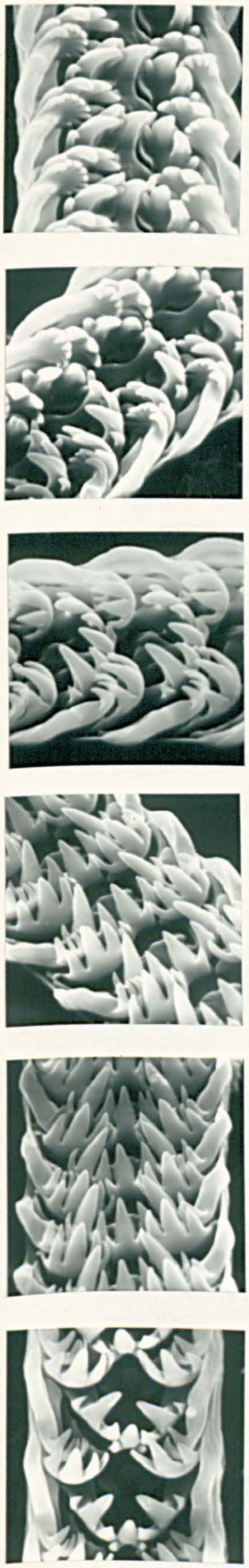


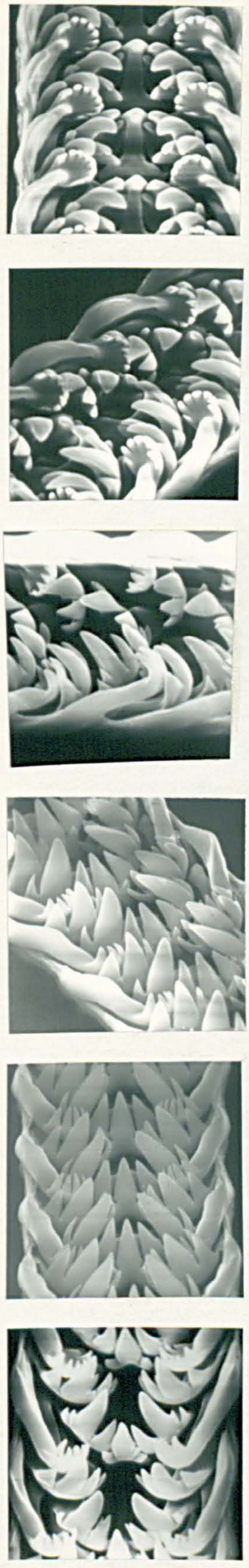
8

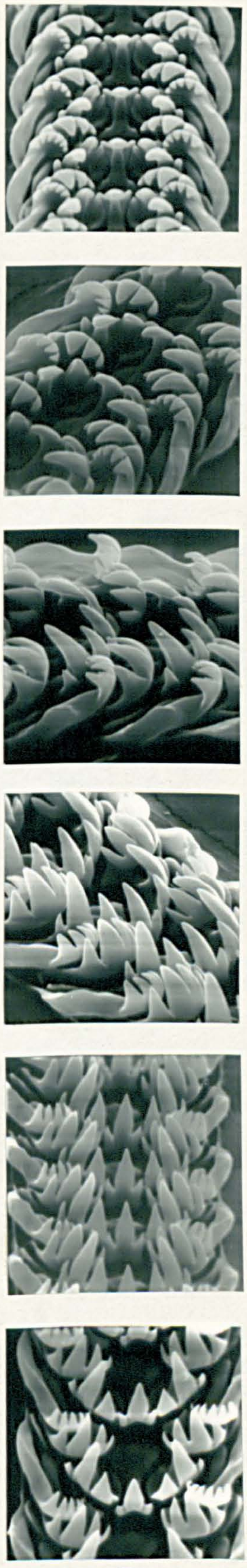
PLATES 18 & 19.

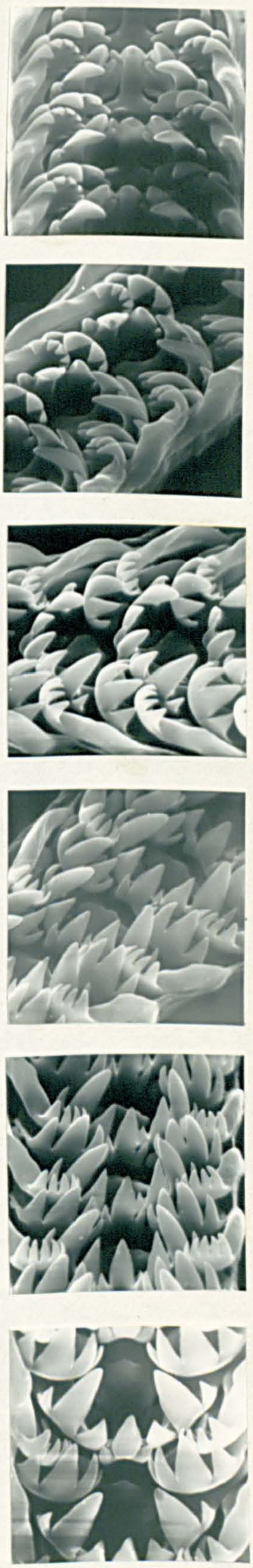
L.neglecta radulae.

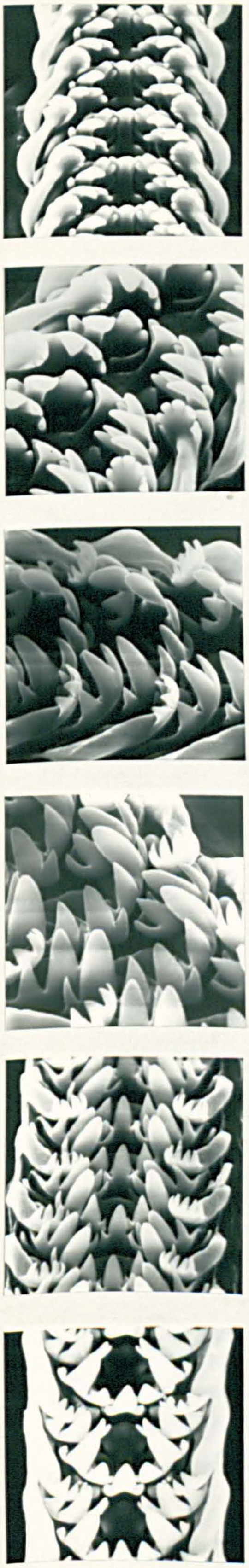
Radula No.	Shell heights
1	1.3 mm
2	1.9 mm
3	2.2 mm
4	2.3 mm
5	2.5 mm
6	2.6 mm
7	2.8 mm
8	3.0 mm


1

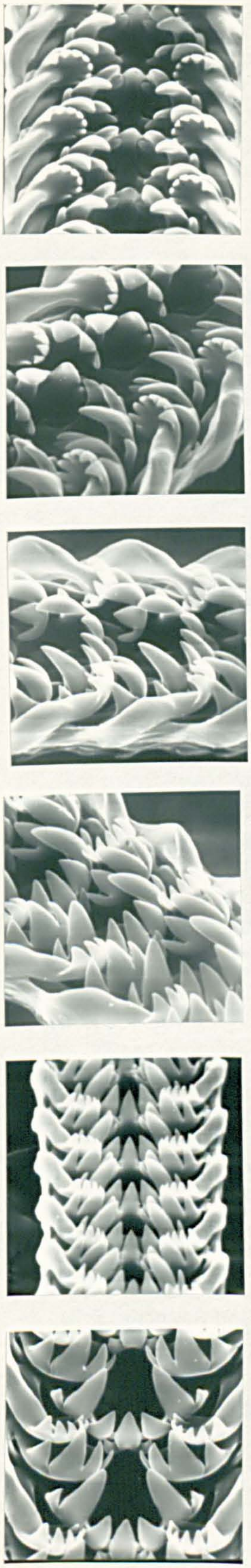

2


3

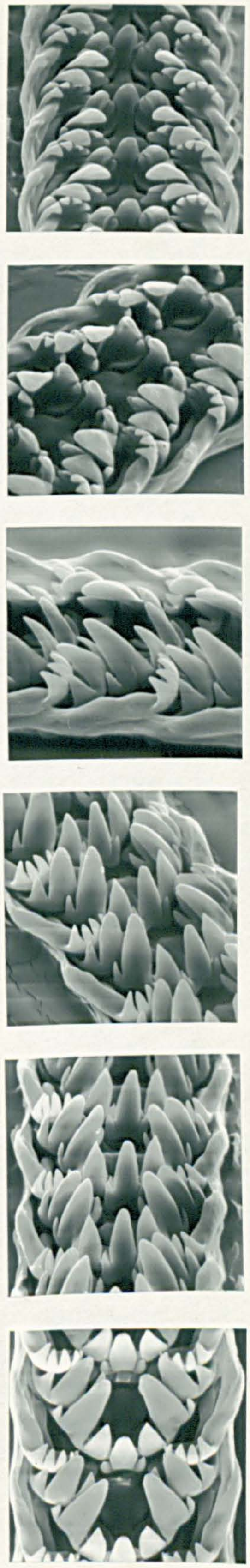

4



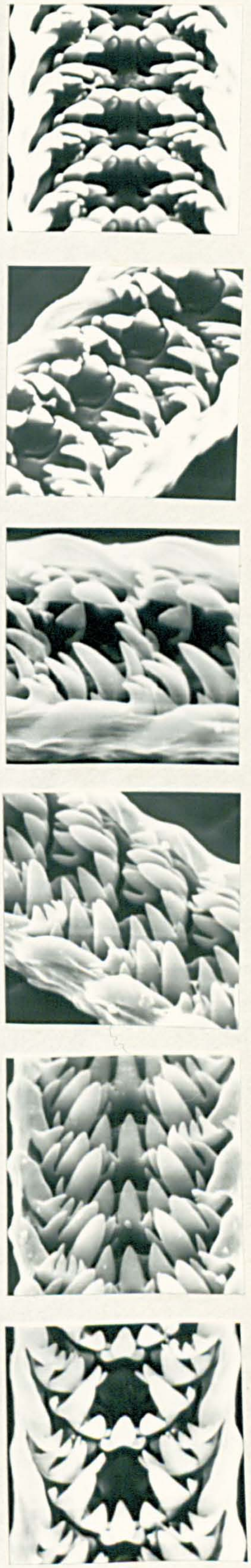
5



6



7

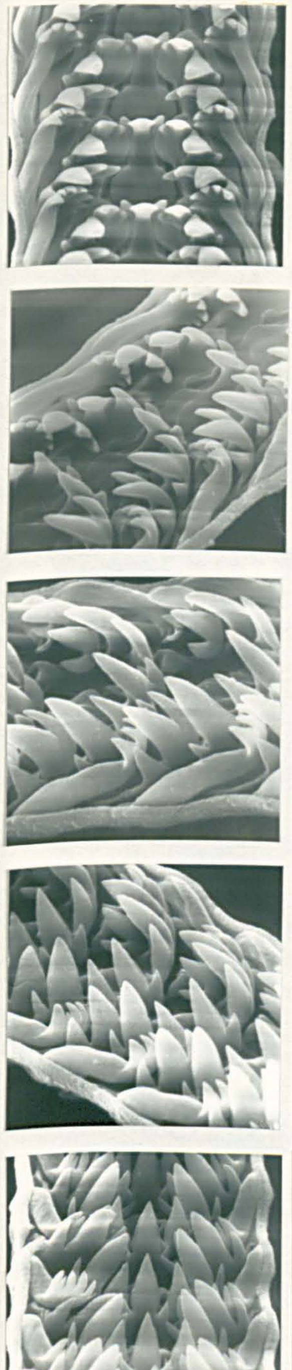


8

PLATES 20 & 21.

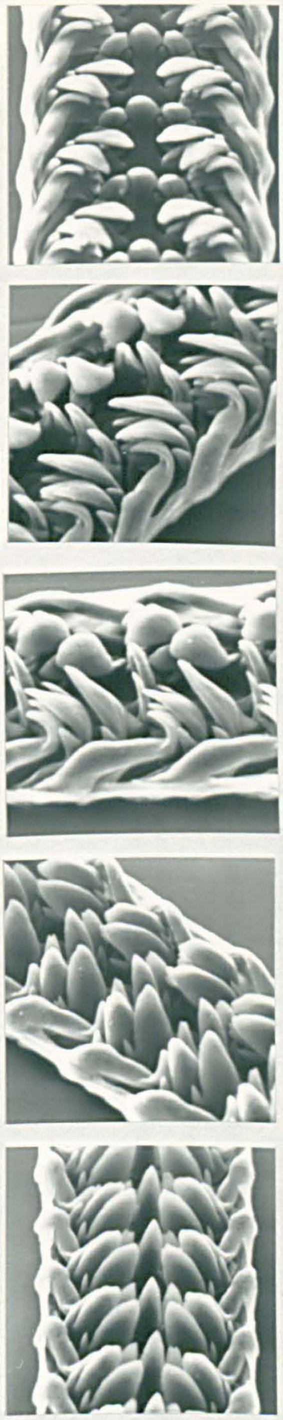
L.littorea radulae.

Radula No.	Shell heights
1	2.2 mm
2	2.3 mm
3	6.2 mm
4	7.5 mm
5	9.7 mm
6	13.15mm
7	17.3 mm
8	22.05mm



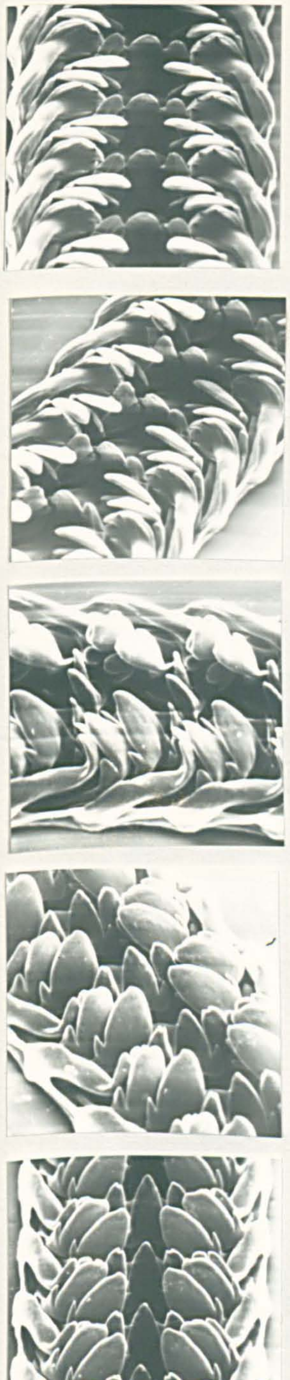
1

2



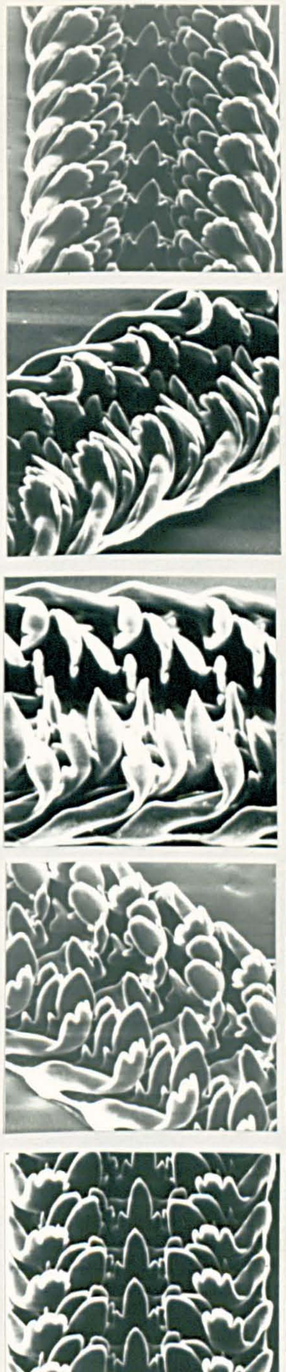
3

4

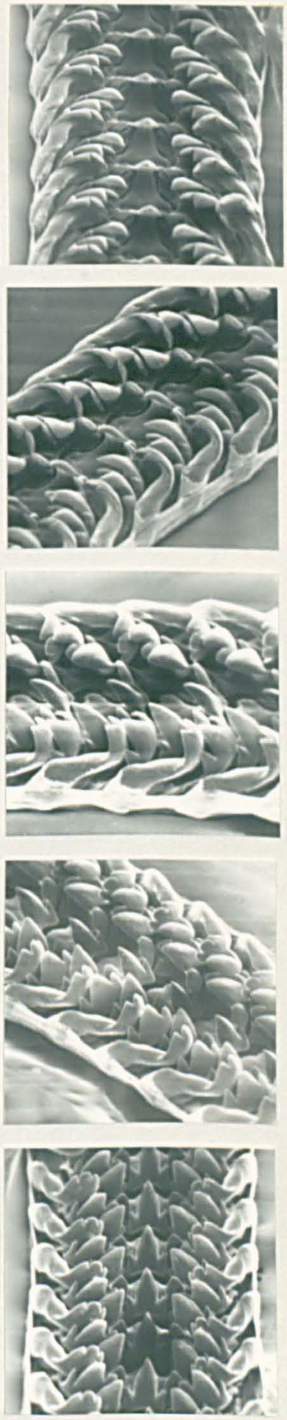


5

6



7

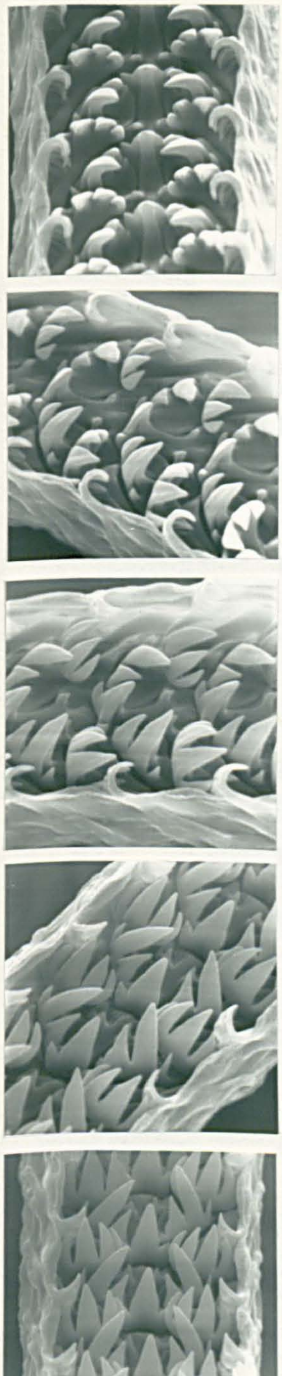


8

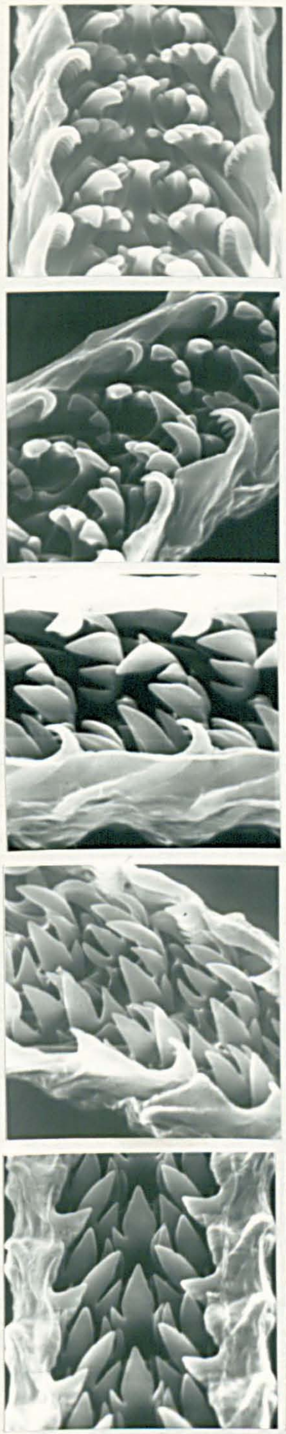
PLATES 22 & 23.

L.neritoides radulae.

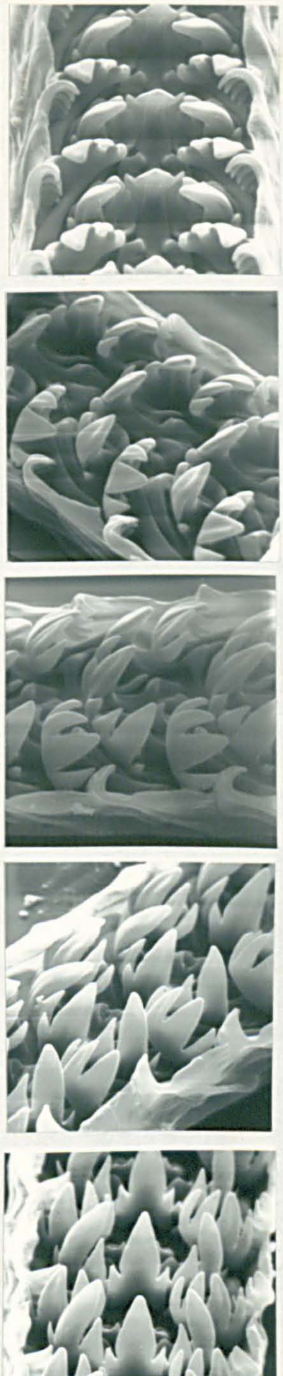
Radula No.	Shell heights
1	1.9 mm
2	2.0 mm
3	2.9 mm
4	3.1 mm
5	3.5 mm
6	5.2 mm
7	6.4 mm
8	7.0 mm



1



2



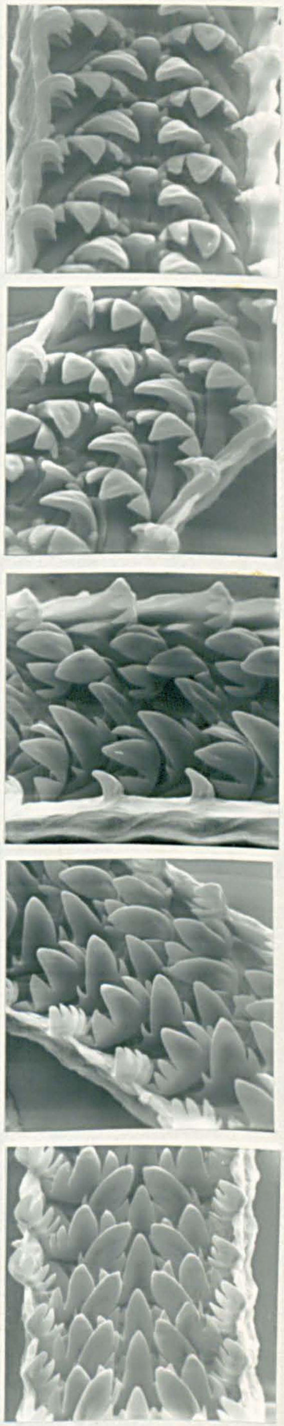
3

4

5

6

7



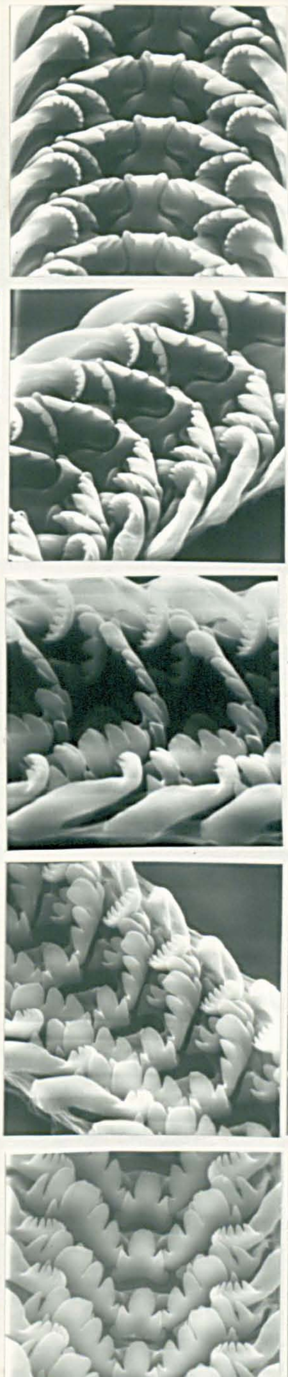
8

PLATES 24 & 25.

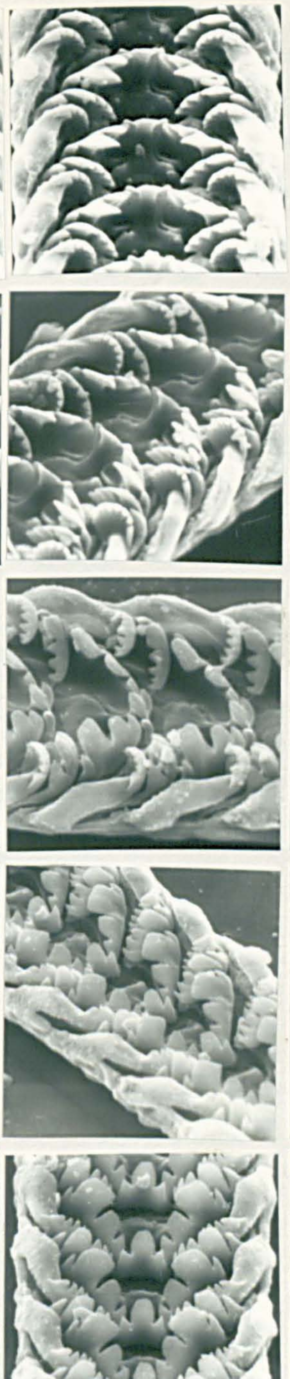
L. obtusata radulae.

Radula No.	Shell heights
1.	2.0 mm
2	3.5 mm
3	4.85mm
4	4.95mm
5	6.9 mm
6	11.05mm
7	13.6 mm
8	14.6mm

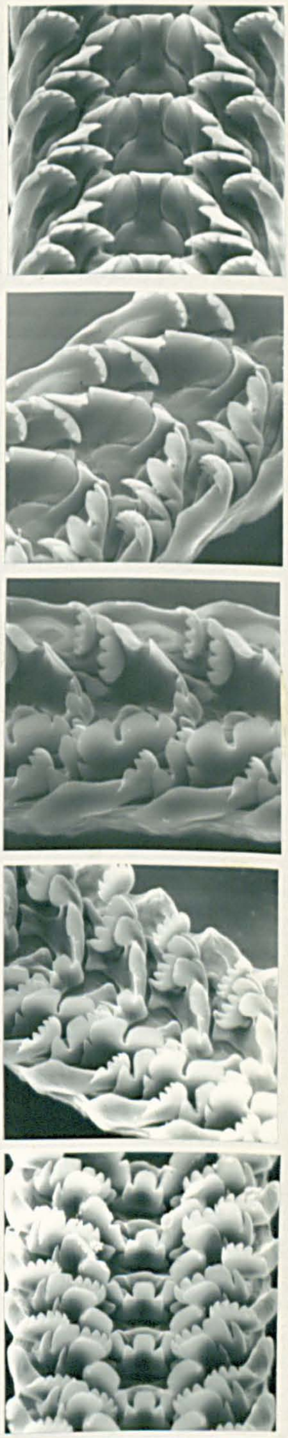
1



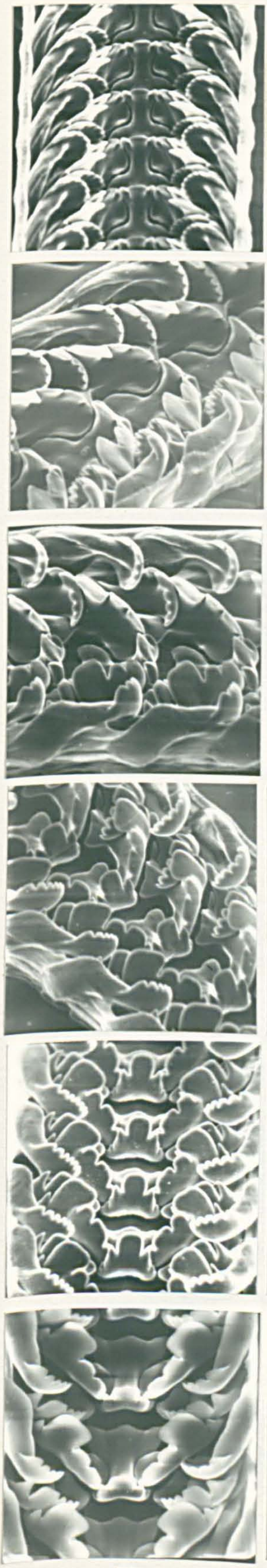
2



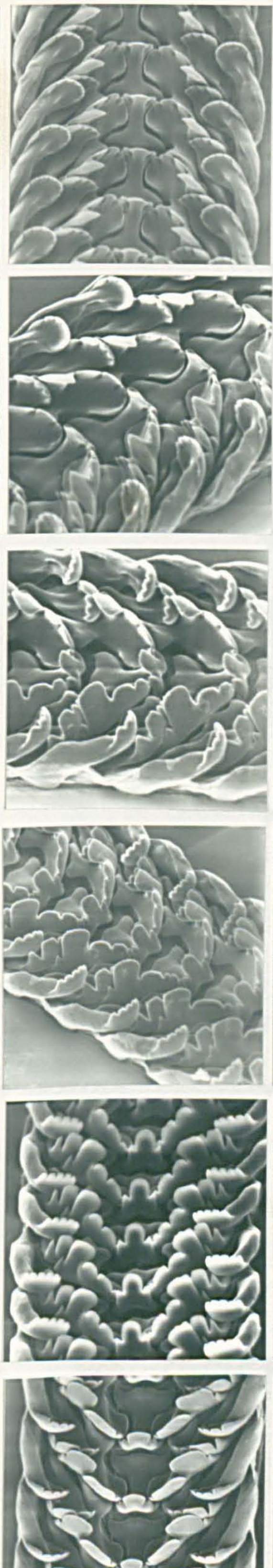
3



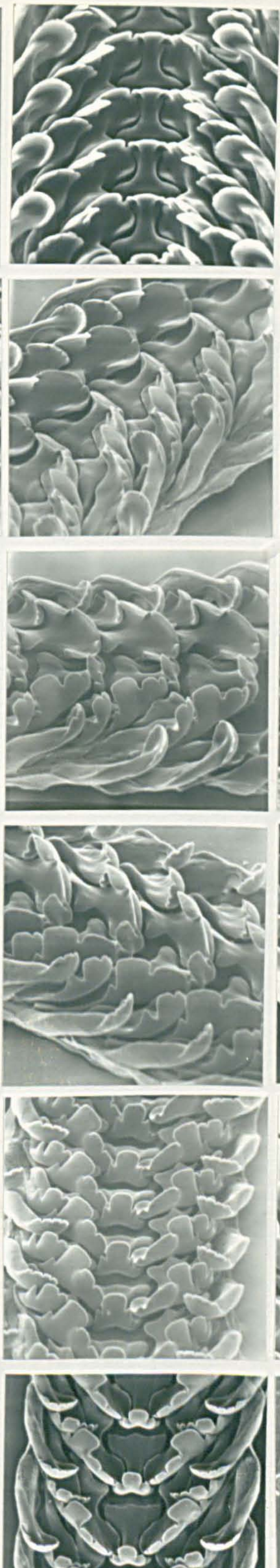
4



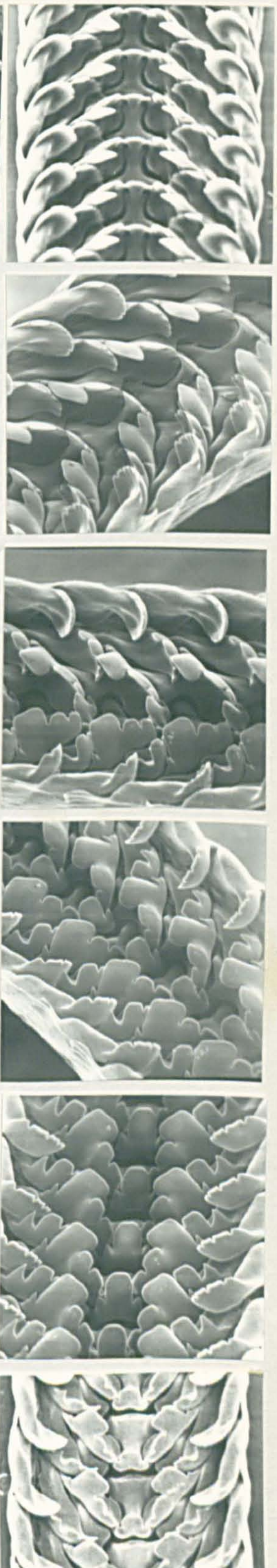
5



6



7

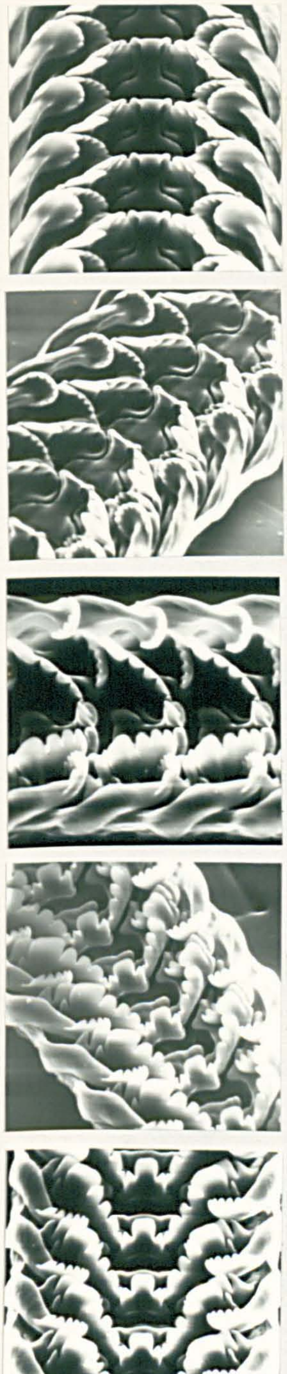


8

PLATES 26 & 27.

L.mariae radulae.

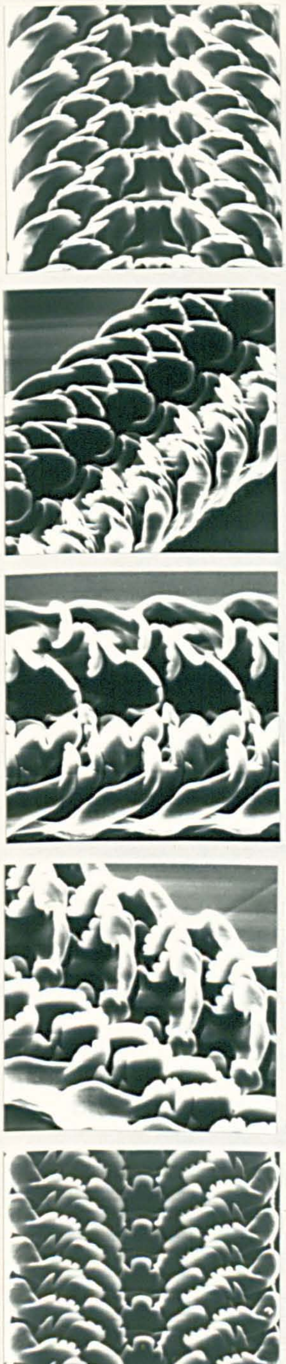
Radula No.	Shell heights
1	3.7 mm
2	4.4 mm
3	5.7 mm
4	6.7 mm
5	8.2 mm
6	8.8 mm
7	10.35mm
8	10.7 mm


1


2


3


4



5

6

7

8

1.7. SHELL CHARACTERISTICS

The shell by virtue of its convenience and permanence has previously been given a too prominent role in the identification of winkle species. Earlier classifications e.g. Jeffreys, 1865, Dautzenburg & Fischer, 1912, were based primarily on shell characteristics and did not distinguish species whose shells are very similar, notably *obtusata* and *mariae*, *arcana* and *rudis*. This pre-eminence of the shell in classification also led to the grouping together of *nigrolineata*, *neglecta*, *arcana* and *rudis* as one species *saxatilis*, in which variability of the shells was a byword; any shell which was not of the *littorea*, *neritoides* or *littoralis* types was labelled *saxatilis*. Thus the distinctive natures of the shells of *nigrolineata* and *neglecta* were largely obscured both by the variability encountered within the shells of these two species and by the extraordinary variability of the shells of *arcana* and *rudis*. This *arcana* - *rudis* variation approached the shell types of *neglecta* and *nigrolineata* sufficiently closely for taxonomists, even as recently as 1968 (James, 1968a) to set up series of *saxatilis* shells grading from one form to another, which in fact traversed species.

Though overemphasis on shell appearance has led to serious errors in littorinid taxonomy in the past, the shell nevertheless provides characters useful in specific identification. It is particularly important in identifying juveniles (in which the reproductive system is undifferentiated) and in identifying animals which are required alive for experiments. It is therefore important that the limits of reliability of shell characteristics are known i.e. which shell characteristics are diagnostic of which species at which shell heights.

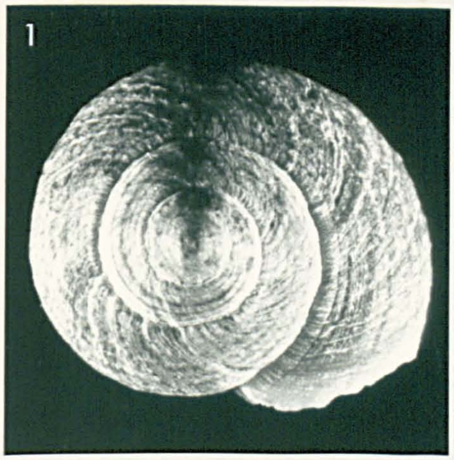
Prior to the work for this thesis the shell characters of the adults of most species had been described, and it has been shown that in the majority of species the adult shells have species - specific characteristics, e.g. the sculpture pattern of nigrolineata shells (Heller, 1975a). However there were two gaps in the descriptions available. One was the description of the shell of the recently diagnosed species, arcana. I have published this description in 1978, but for completeness reproduce it here, as it is important that the similarity of the shells of arcana and rudis (applying to both adult and juvenile shells) should be fully appreciated. The second gap concerns the lack of descriptions of juvenile shells for most species. Like the radula, the shells of certain species show ontogenetic changes, and some, e.g. littorea juveniles, would be unidentifiable from descriptions of adult shells. By working through size series of shells it was possible to connect the juvenile shell form to the adult. The use of the juvenile shell for identification of species was particularly important for the work on the ecology of small winkles (section 3.3).

1.7.1. Shell Characteristics of Adult L. arcana compared with Adult L. rudis and Assessment of the Usefulness of their Shells in Identification

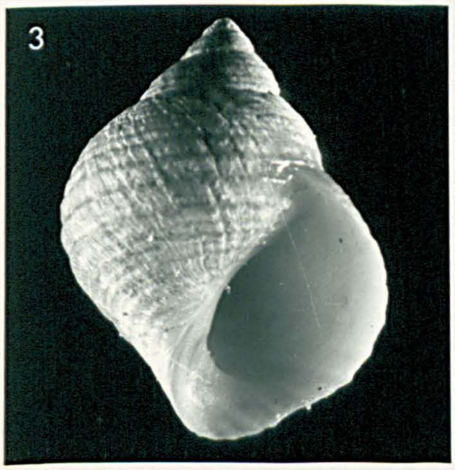
The shell of L. arcana is very like that of L. rudis and is equally variable. There is no diagnostic character that distinguishes shells of this species from shells of L. rudis and no shell

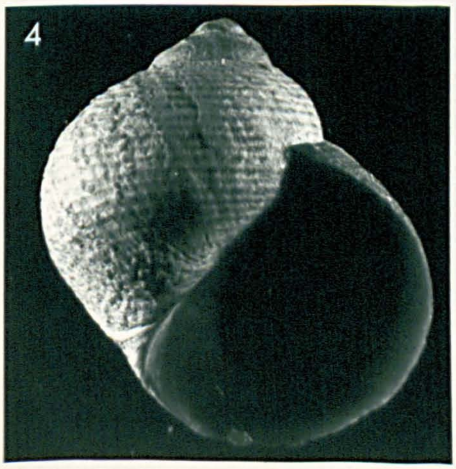
PLATE 28.

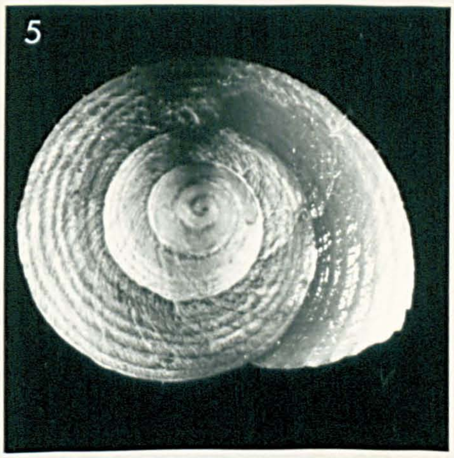
Shells, nos. 1 - 4, L.rudis, 5 - 8, L.arcana

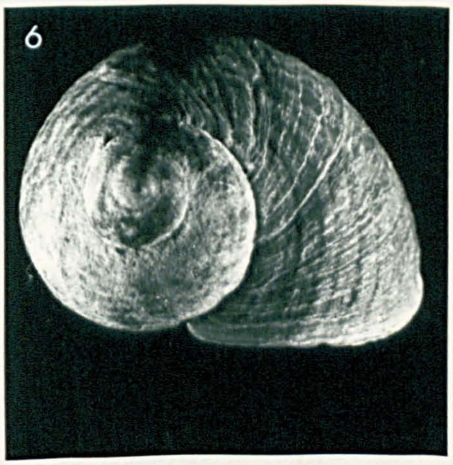

1

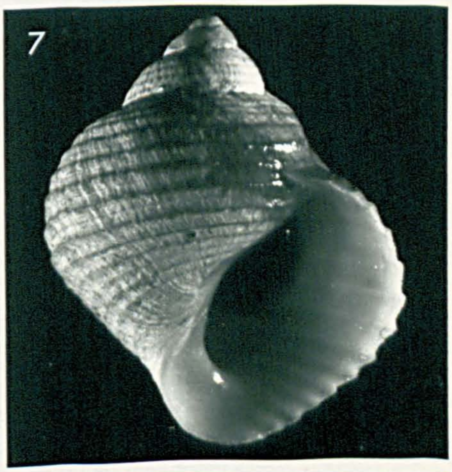

2


3


4


5


6


7

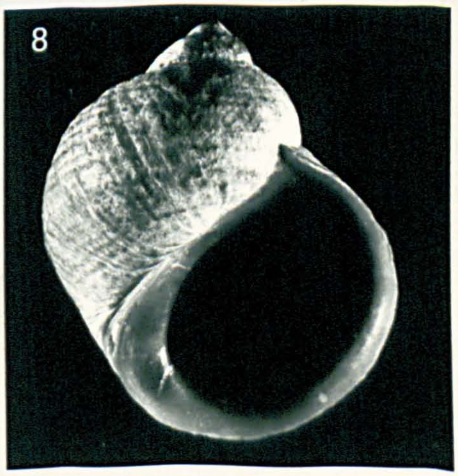
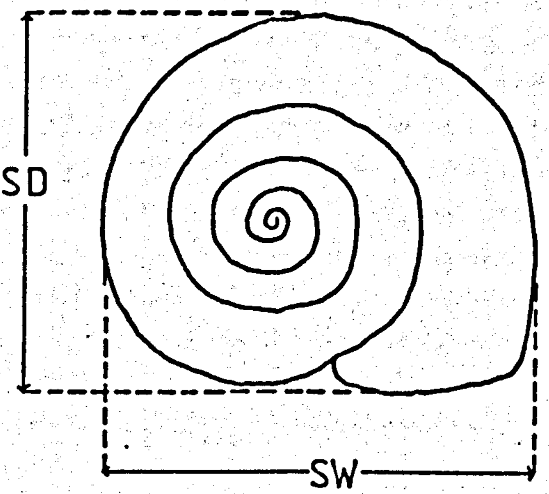
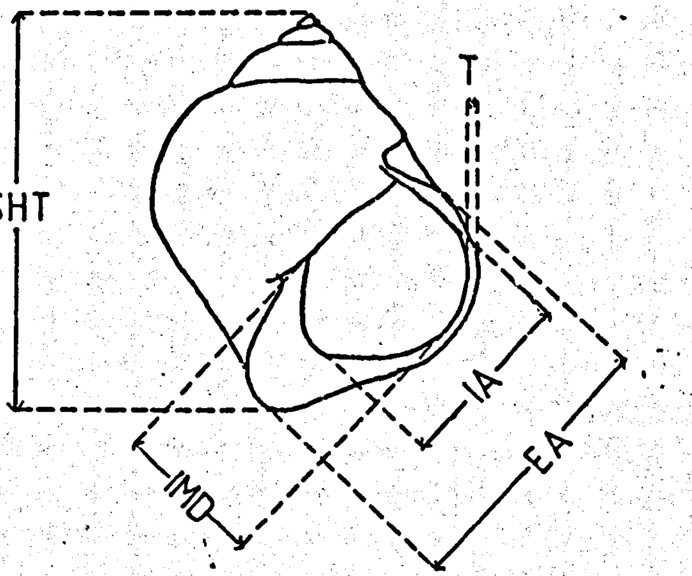

8

Figure 24.

Measurements of shell dimensions.


SD
SW


SHT
T
IA
EA
IMD

variety seems to be peculiar to one species or the other. Plate 28 shows the similarity of shells in the two species. All these shells were from the same site.

At many sites it is noticeable however, that the shells of L. arcana exhibit, more strongly than those of L. rudis, patulous shell characteristics. The patula shell type is currently regarded (Heller, 1975a) as exhibiting the characteristics described by Jeffreys (1865) i.e. short-spired, asymmetrical in spire view and with a wide, round aperture, showing little expansion and angulation below. The typical rudis shape is medium-spired, symmetrical in spire view, with an oval aperture considerably expanded and angulated below. These aspects of shell shape can be estimated respectively by the ratios shell height/shell width, shell diameter/shell width, internal mouth diameter/internal aperture length and internal aperture length/external aperture length (fig. 24). Measurements were therefore made on shells of animals of both species collected from mixed populations at five localities and ratios were derived from these data to indicate the degree of expression of patulous characters in the two species at each locality.

Comparisons between the ratios of the two species are given in Table 3 and it can be seen that shells of L. arcana are significantly more short-spired and asymmetrical than L. rudis at all sites. At four sites out of five the angulation and expansion of the lip is significantly less developed but only at one site are the shells of L. arcana more commonly rounder-mouthed than those of L. rudis. Overall the data confirm the suggestion that

TABLE 3

Comparison of shell shape measurements of _L. arcana_ with _L. rudis_ using the Mann-Whitney _U_ test.

		L. arcana			_L. rudis_				
Ratio	Site	n	median	range of observations	n	median	range of observations	_z_	two-tailed p
SHT/SW	P.S.	92	1.092	.983-1.188	90	1.095	1.012-1.171	1.71	.0872
	A.B.	67	1.080	1.014-1.186	61	1.091	1.000-1.177	1.00	.3174
	B.R.	75	1.112	1.041-1.243	63	1.156	1.074-1.260	5.54	<.00006
	Y.W.	80	1.070	.966-1.175	80	1.120	1.018-1.200	6.20	.0046
	D.	73	1.080	.955-1.183	68	1.099	.959-1.230	2.84	.0046
SD/SW	P.S.	92	.634	.584-.677	90	.664	.610-.697	6.94	<.00006
	A.B.	67	.608	.574-.674	59	.648	.606-.708	7.13	<.00006
	B.R.	75	.641	.594-.639	63	.667	.626-.693	7.65	<.00006
	Y.W.	80	.618	.537-.699	80	.661	.610-.712	9.34	<.00006
	D.	73	.618	.568-.690	68	.653	.586-.712	6.88	<.00006
IA/EA	P.S.	104	.662	.577-.832	87	.661	.568-.722	.62	.5352
	A.B.	52	.708	.608-.798	56	.551	.580-.719	5.96	<.00006
	B.R.	75	.632	.576-.748	63	.595	.552-.648	6.87	<.00006
	Y.W.	50	.636	.595-.696	50	.625	.553-.650	3.07	.00022
	D.	70	.692	.590-.795	69	.644	.569-.727	6.03	<.00006
IMD/IA	P.S.	103	.771	.705-.859	85	.781	.687-.926	1.95	.0512
	A.B.	67	.608	.574-.674	59	.648	.606-.708	7.13	<.00006
	B.R.	74	.787	.714-.861	63	.787	.690-.855	.11	.9124
	Y.W.	50	.797	.703-.878	50	.782	.714-.874	1.04	.2984
	D.	70	.780	.680-.862	68	.771	.681-.878	1.32	.1868

Sites:- (See Table 1)

Measurements:- E.A. - external aperture; I.A. - internal aperture;
I.M.D. - internal mouth diameter; S.D.- shell diameter;
S.H.T. - shell height; S.W. - shell width

L. arcana is more strongly associated with the patulous (*sensu* Jeffreys) shell shape than is *L. rudis*. Nevertheless both species can exhibit shells with either *rudis* or *patula* characteristics and as the overlap in ratios observed in the two species indicate, it is not possible to separate the species reliably by the shape of the shell.

In spite of this the shell can be used in identification under certain circumstances. At many sites it is possible to find that a particular shell shape or colour is always associated with one or other species. For example at Abraham's Bosom shells of *rudis* are thick, smooth and orange and those of *arcana* are thin, heavily ridged and multicoloured with tessellations or bands. Once such an association of species with a particular shell colour or sculpture has been identified (through killing a representative sample and examining the anatomy), then the shell can be made use of in the field. However at many sites no such association of shell characteristics and species can be found.

1.7.2. Identification of Juveniles from Characteristics of the Shell

The following descriptions are based mainly on Porth Swtan material but appear to apply to other Anglesey sites. Below a shell height of about 1.0mm it is not often possible to identify species from the shell.

1.7.2.1. L. nigrolineata

Juveniles' shells, like adults', have a distinctive ridging pattern, but unlike the wide flat ridges and narrow grooves of the adults', the ridges and grooves are triangular in cross-section. Triangular ridges are also seen in shells of juvenile arcana, rudis and littorea,but unlike these species there are no minor ridges running parallel to the main ridges (compare nigrolineata 5 & 6, Plate 29, with rudis and arcana 1-4, Plate 29 and littorea 1, Plate 30), so the ridges appear clean cut. Also unlike arcana and rudis the ridging pattern follows a graded size sequence, small at the suture of the body whorl with the previous whorl, increasing in size to the widest part of the whorl and declining in size to the columella. The shell appears glassy compared with rudis and arcana shells, and is often sufficiently translucent for black pigment on the body to be seen through it. As the animal ages the ridges change from triangular to rounded and ultimately become flat, (see fig. 25). The aperture is comparatively round (Plate 30,6) and the columella usually white.

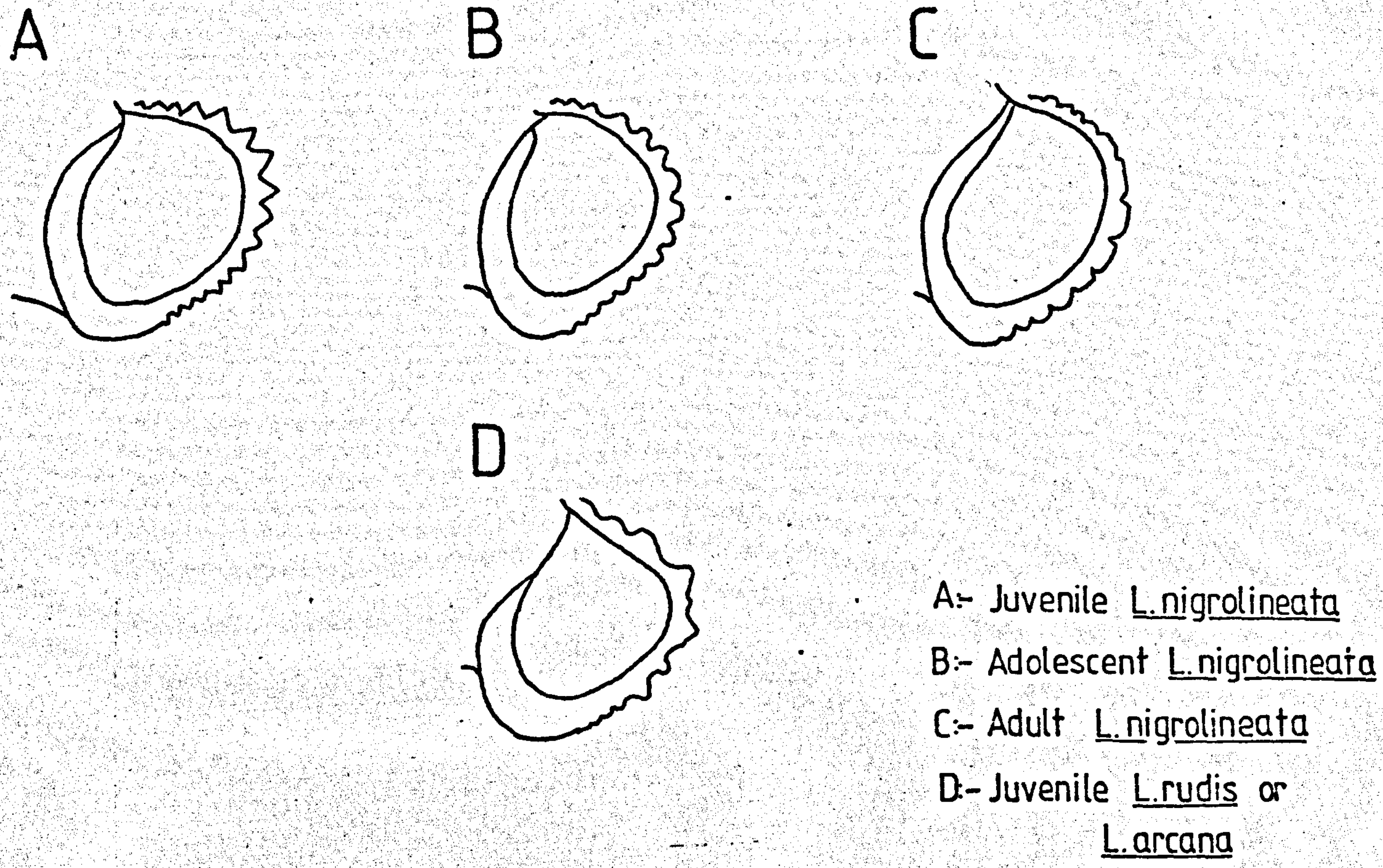
1.7.2.2. L. rudis and L. arcana

These are not distinguishable at Porth Swtan. The shell is usually ridged to some degree (Plate 29, 1-4). Ridges can be triangular in cross-section or more rounded, often with small secondary ridges. They are usually irregularly spaced and are narrower than the grooves, and do not show a regular size sequence (fig. 25). The aperture is often oval or slightly angled (Plate 30, 5), and the columella usually has some pigment. The

<u>Figure 25.</u>

Shell sculpture at the aperture.

SHELL SCULPTURE AT THE APERTURE



A:- Juvenile *L. nigrolineata*

B:- Adolescent *L. nigrolineata*

C:- Adult *L. nigrolineata*

D:- Juvenile *L. rudis* or *L. arcana*

shells show a considerable range of thicknesses. Some are sufficiently thin for black body pigment to be seen through it.

1.7.2.3. L. neglecta

Juvenile shells of this species resemble those of the adults and are usually smooth shelled, though a few slightly-ridged specimens have been collected. The body whorl is usually expanded and the spire comparatively small and short (Plate 29, 7). The majority of individuals from Porth Swtan can be identified by the distinctive *neglecta* colour pattern, with a wide dark band near the columellar lip and tessellations or bands between it and the suture (see Heller, 1975a and fig. 31). Some specimens from Porth Swtan have black or near-white shell colours. These are identifiable by the smooth shell, the shape, and the opaque quality of the shell; also the accessory character of the sub-opercular pattern is useful here.

1.7.2.4. L. littorea

Specimens of this species of 4mm shell height or less have a very characteristic shell shape and sculpture (Plate 30, 1). The shell is extremely high-spired and heavily ridged. The shells are pale coffee coloured, and darker pigment is sometime laid down in the ridges (giving a reverse pattern to nigrolineated *nigrolineata*).

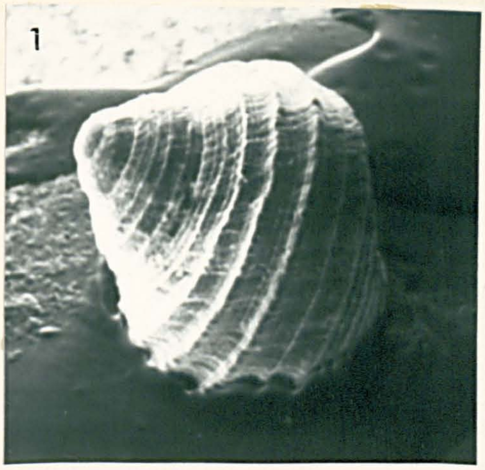
1.7.2.5. L. mariae and L. obtusata

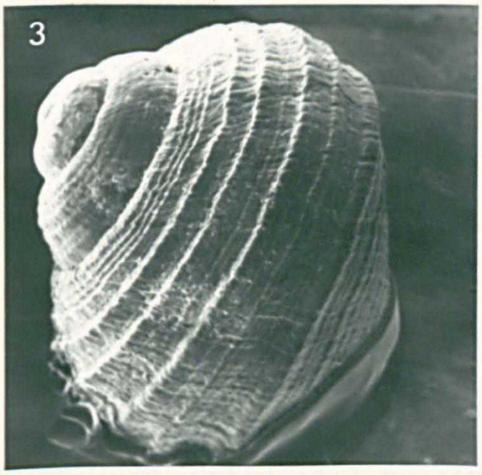
Juveniles of both species have the flattened spire and so are easily separated from juveniles of other winkle species. Reimchen

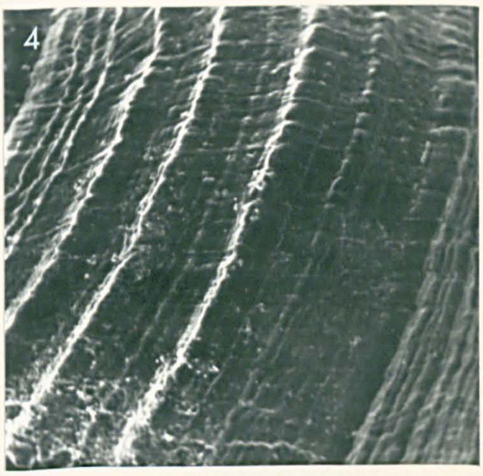
PLATE 29.

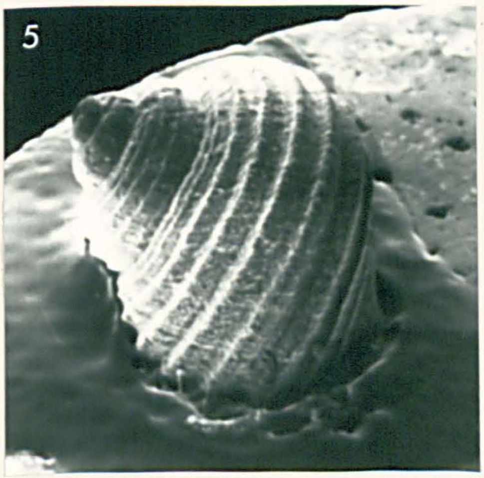
SEM photomicrographs: Juvenile shells and close-up of sculpturing pattern in

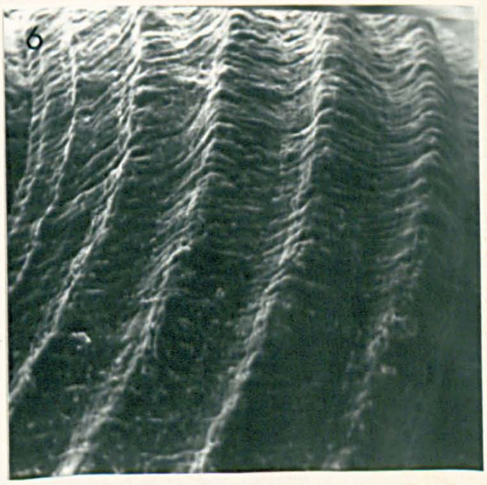
1). & 2). *L.arcana*
3). & 4). *L.rudis* C
5). & 6). *L.nigrolineata*
7). & 8). *L.neglecta*


1


3


4


5


6


7

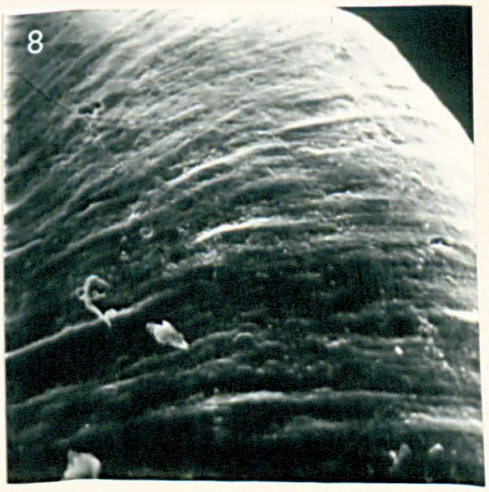

8

PLATE 30.

SEM photomicrographs:

1). *L.littorea*, juvenile shell.

2). *L.neritoides*, juvenile shell.

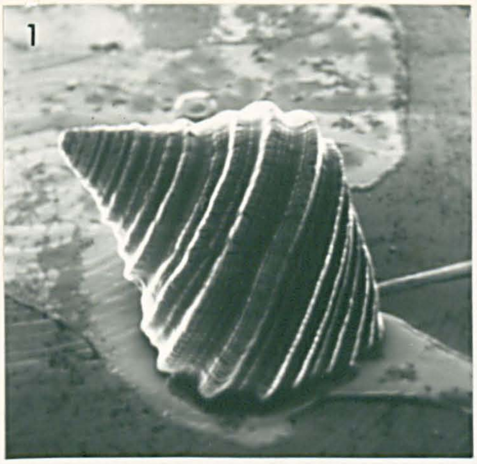
3). Close-up of the periostracal sculpture of *L.obtusata*.

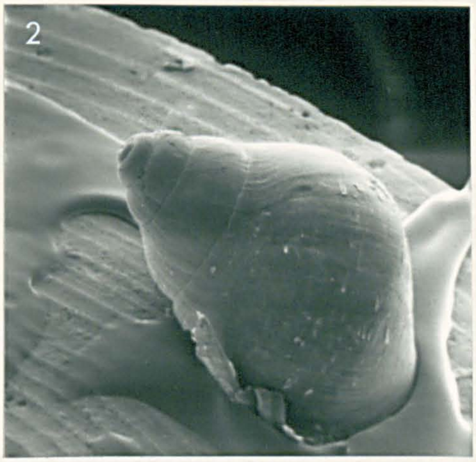
4). Close-up of the periostracal sculpture of *L.mariae*.

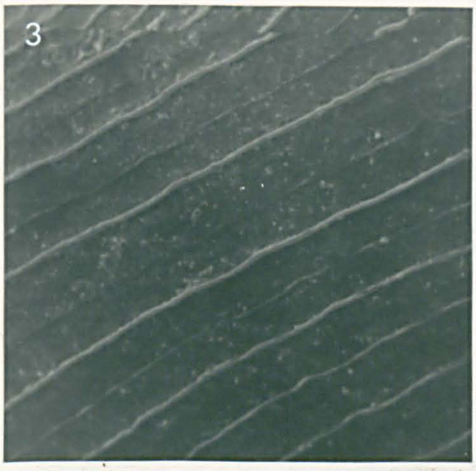
5). Aperture view of the shell of a *L.arcana* juvenile.

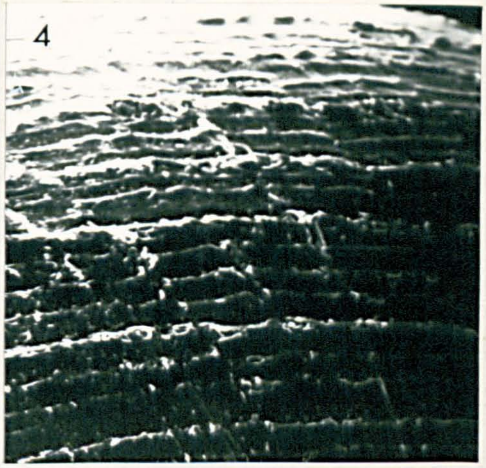
6). Aperture view of the shell of a *L.nigrolineata* juvenile.

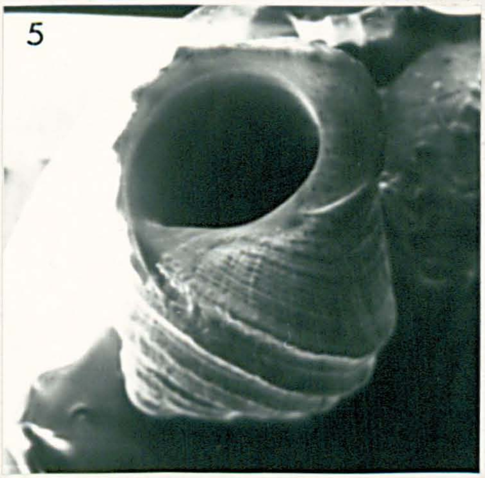
7). & 8). Protoconch of juvenile *L.arcana*.


1

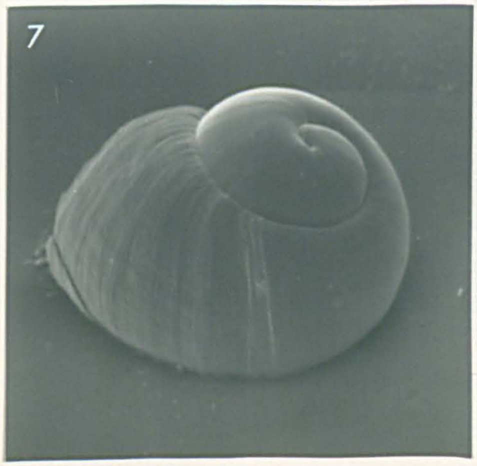

2

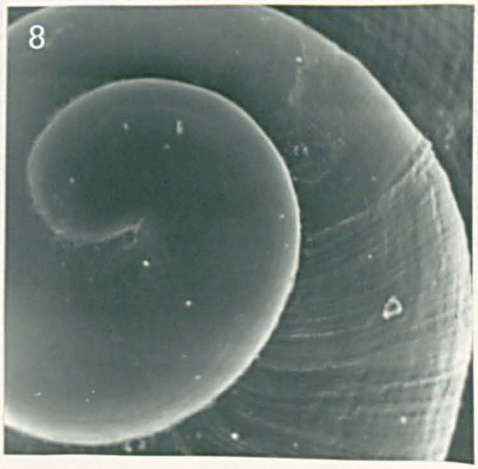

3


4


5


6


7


8

(1974) has pointed out a difference in the periostracal ridging pattern; obtusata juveniles have alternate high and low ridges running parallel to the suture and mariae juveniles have high ridges separated by several low ridges. (Plate 30, 3 &4).

1.7.2.6. L. neritoides

Juvenile shells of this species resemble the adults (Plate 30,2).

SECTION 2: ASPECTS OF THE AUTECOLOGY OF ROUGH WINKLE SPECIES

Methods of assessment of maturity, shell dimorphism, reproductive seasonality, age-related colour pattern changes (in neglecta) and population structure (neglecta only) are discussed in this section.

The majority of the work in this section was done on Porth Swtan material and the results may not therefore be of general applicability. The same species at sites of different character eg. in substrate, exposure, etc. may show quite different adaptive characteristics. However, concentration at this site has allowed a more detailed analysis than would otherwise have been possible and the results may provide a basis for future comparative work at a wider range of sites.

At Porth Swtan there are two populations of rudis which differ in many respects and samples from them have been given separate treatment in the analyses below. One population consists of animals with large, thick, smooth, narrow-mouthed shells. This population is associated with an unstable boulder/pebble habitat and these animals are referred to as rudis B (boulders). The other population is associated with the crevices of stable rock masses and consists of animals with small, thin, ridged, wide-mouthed shells. These are referred to as rudis C (crevice). A fuller consideration of the differences between these two rudis populations is given in section 4.

2.1. METHODS OF ASSESSING MATURITY

In many winkle species the adults have a cycle of reproductive activity and when winkles go out of reproductive condition the reproductive organs regress (Bergerard 1971a). Distinguishing the reproductive state of the animals was important for both the taxonomic and ecological work undertaken.

Three terms, juvenile, immature and mature are used here to describe the reproductive condition of the animals. Juvenile animals are young animals in which the reproductive tract is insufficiently differentiated for the sex to be identified. Immature animals are those in which sex is identifiable but either have not yet bred or have gone out of reproductive condition. These very different groups could not be distinguished reliably. Mature animals are those which are in breeding condition. The distinction between immature and mature animals is discussed in greater detail below; males and females are considered separately.

2.1.1. Definition of maturity in male rough winkles

The definition of a male winkle as mature or immature is based on the state of the penis. Both qualitative and quantitative methods can be used and are compared here. On a qualitative basis a visual assessment of the state of the penis can be made and if it is well-formed and the penial glands are turgid then it is scored as mature. On a quantitative basis maturity can be defined using the relative penis length, PL/SHT (penis length, PL, is defined in fig. 10,and shell height, SHT, in fig. 24). A comparison of visual estimate and PL/SHT ratio is given in table 4 and plotted in fig. 26A, using data from nigrolineata (equivalent data were obtained for arcana and rudis). It is obvious that the ratio frequencies are distributed in two groups and those with larger ratios correspond with mature animals. The strong bimodality emphasizes the comparative rarity of animals with penes in a transitional state, ie. if an animal has a developed penis then it is likely to have a fully developed one. Thus it seems that the qualitative, visual assessment method is as satisfactory as the quantitative method and has the added advantage of being quicker to use. Visual assessment of maturity is therefore used in subsequent analyses.

2.1.2. Definition of maturity in female rough winkles

In ovoviviparous species (rudis and neglecta), mature females are defined as those with embryos in the brood pouch. In oviparous species the distinction between immature and mature females is not so easily made. As in the males, qualitative or quantitative methods can be used and these are compared below. On a qualitative basis, females were defined as mature if the oviduct was turgidly fleshy (see Plate 2) and on a quantitative basis maturity can be defined on the basis of relative oviduct size which can be obtained from the ratio c+w/SHT

TABLE 4: Comparison of visual estimate of maturity of males with PL/SHT ratio

PL/SHT ratio size class		.0-.09	.1-.19	.2-.29	.3-.39	.4-.49	.5-.59	.6-.69	.7-.79	.8-.89	.9-.99	1.0-1.09
Visual assessment	Immature	13	104	64	15	11	3	2	-	-	-	-
	Mature	-	-	-	-	-	3	21	33	49	30	8
Total in size class		13	104	64	15	11	6	23	33	49	30	8

TABLE 5: Comparison of visual estimate of maturity of females with c+w/SHT ratio

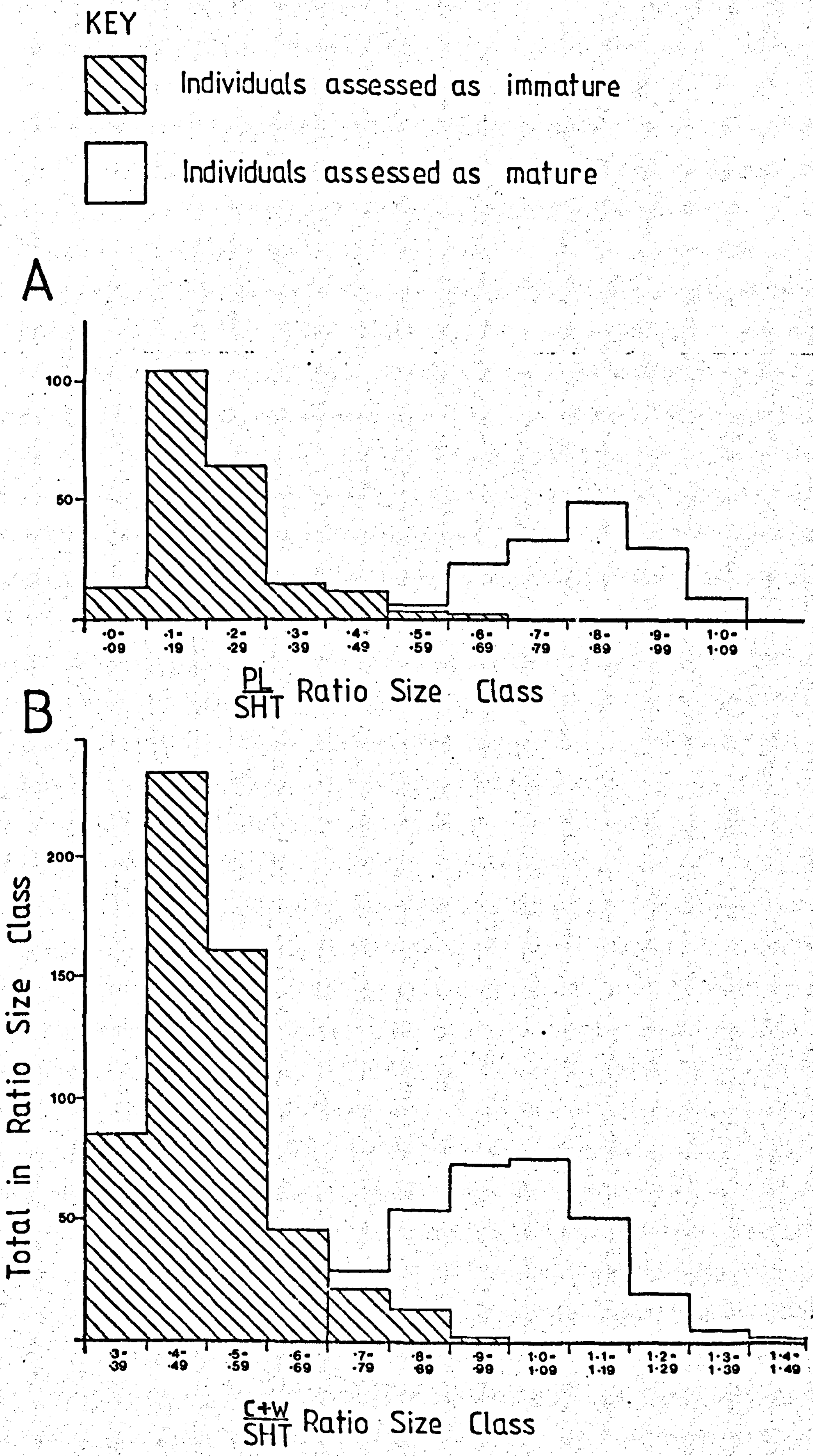
c+w/SHT ratio size class		.3-.39	.4-.49	.5-.59	.6-.69	.7-.79	.8-.89	.9-.99	1.0-1.09	1.1-1.19	1.2-1.29	1.3-1.39	1.4-1.49
Visual assessment	Immature	85	235	161	46	22	11	2	-	-	-	-	-
	Mature	-	-	-	-	7	43	71	76	51	20	5	2
Total in size class		85	235	161	46	29	54	73	76	51	20	5	2

Figure 26.

Comparison of qualitative and quantitative assessments of maturity:

A). Male *L. nigrolineata*

B). Female *L. nigrolineata*


KEY
▨ Individuals assessed as immature
☐ Individuals assessed as mature
A
100
50
·0-·09
·1-·19
·2-·29
·3-·39
·4-·49
·5-·59
·6-·69
·7-·79
·8-·89
·9-·99
1·0-1·09
PL/SHT Ratio Size Class
B
Total in Ratio Size Class
200
150
100
50
·3-·39
·4-·49
·5-·59
·6-·69
·7-·79
·8-·89
·9-·99
1·0-1·09
1·1-1·19
1·2-1·29
1·3-1·39
1·4-1·49
C+W/SHT Ratio Size Class

(oviduct length, c, and width, w, are defined in fig. 27, shell height, SHT, in fig. 24). A comparison of ratio frequency distribution and the visual assessment is made in table 5 and fig. 26B, using data from <u>nigrolineata</u>; comparable data were obtained for <u>arcana</u>.

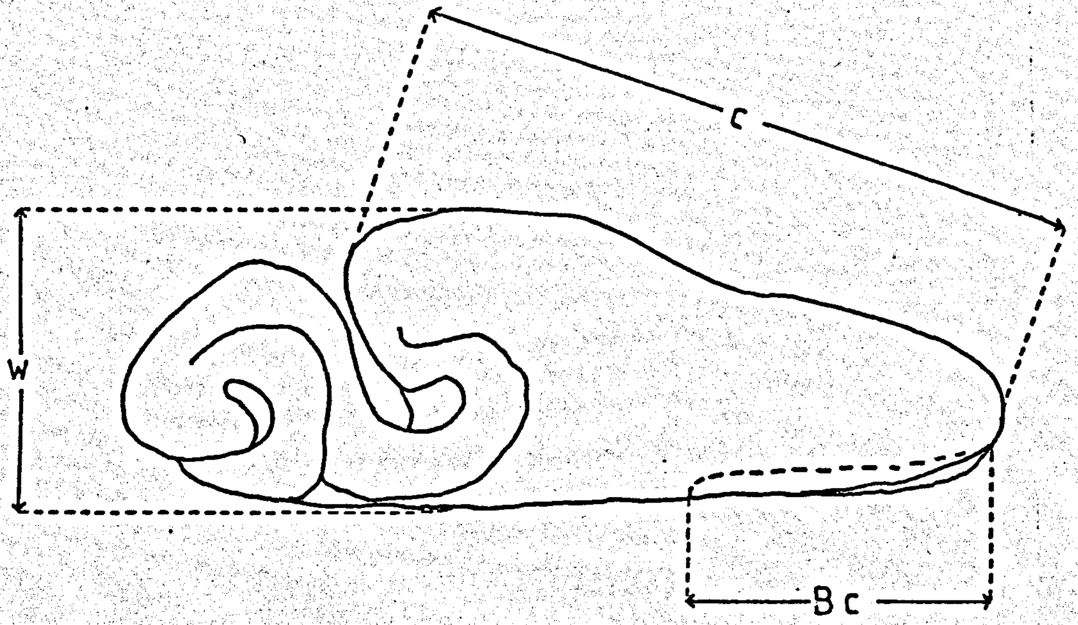
As in the males, the frequency distribution of the ratio shows marked bimodality which is strongly associated with the visual assessment. Visual assessment, being quicker to use, was therefore used in the following analyses to distinguish mature and immature females and as the data indicate comparatively few specimens were collected which appeared to be in a transitional state.

Figure 27.

Measurements of oviduct dimensions

Key: Bc:- bursa copulatrix length
c:- jelly gland or brood pouch length
w:- jelly gland or brood pouch width

MEASUREMENTS OF OVIDUCT DIMENSIONS



2.2. SHELL DIMORPHISM

It was noticeable in many samples, from a range of sites, that sexual dimorphism in shell size occurred in all the rough winkle species. Such dimorphism has previously been reported in mariae, obtusata and nigrolineata.

In mariae and obtusata, sexual dimorphism of the shell has been examined extensively. Sacchi (1968) recorded that in both species, females were larger and more globular than males, the differences being more pronounced in mariae, and with increased exposure in obtusata. Reimchen (1974) in an intensive study of British populations agreed that in both species, females are larger than males and that the differences are greater in mariae; frequently sexual dimorphism was not apparent in populations of obtusata. He showed that dimorphism was most pronounced in populations of mariae with relatively small adult size. Goodwin and Fish (1977) also showed that sexual dimorphism was present in both species though was less pronounced in obtusata than mariae. They likewise agreed that it was greater in populations of mariae from sheltered shores where adult size was relatively small.

In nigrolineata Naylor (1978, unpublished report) has noted sexual dimorphism of shape. As in the flat winkles, females are more globular. No other previous study of dimorphism in the recently diagnosed species of rough winkles has been made.

Three aspects of sexual dimorphism in the rough winkle species at Porth Swtan are examined in sections 2.2.1. - 2.2.3.. The data used were amalgamated from samples taken over a period of at least a year which avoided possible seasonal influences. In *nigrolineata* at Porth Swtan not only was sexual dimorphism in shell size apparent but also size dimorphism related to colour polymorphism. This is explored in section 2.2.4.

2.2.1. Dimorphism in shell height related to sex

Shell heights of males and females, collected over a long period (one year, except for *nigrolineata* which was collected over 22 months) were compared. In this comparison no account was taken of the reproductive condition of the individuals. Brandt and Snedecor's formula for a contingency table with n columns and 2 rows (Bailey, 1959) was used to compare the observations against the null hypothesis that the ratio of males to females in each size class is homogeneous with the ratio of total numbers of males to total numbers of females.

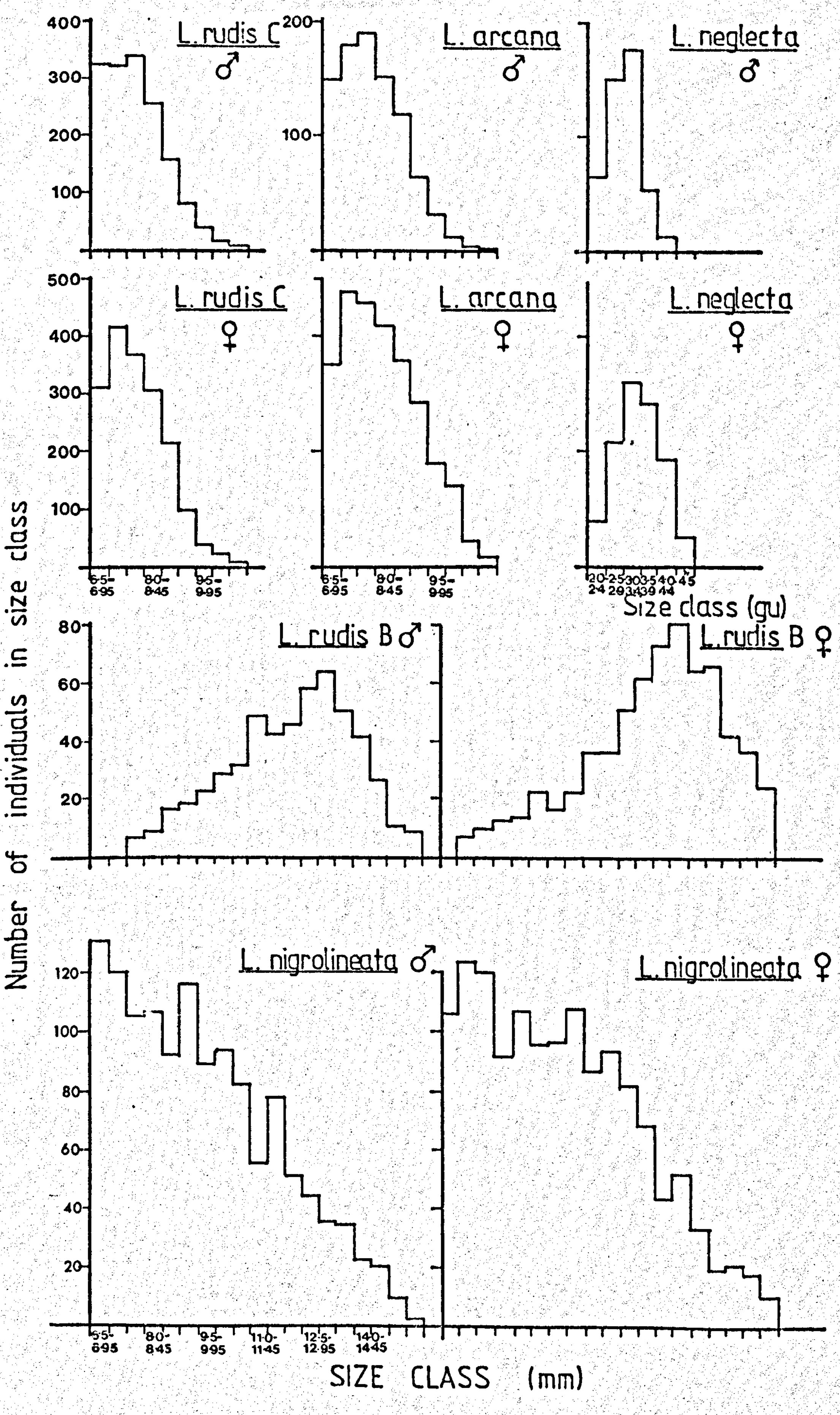
Results

The data for the analyses appear in table 1, Appendix B, and the results are given below:

Species	χ^2	Degrees of freedom	Significance
L.rudis C	12.63	8	Not significant
L.rudis B	40.86	18	Sig. at 1.0%
L.arcana	61.68	8	Sig. at 0.1%
L.nigrolineata	30.64	19	Sig. at 5.0%
L.neglecta	134.37	5	Sig. at 0.1%

Figure 28.

Sexual dimorphism in shell height.

L. rudis C ♂
L. arcana ♂
L. neglecta ♂
L. rudis C ♀
L. arcana ♀
L. neglecta ♀
L. rudis B ♂
L. rudis B ♀
L. nigrolineata ♂
L. nigrolineata ♀
400
300
200
100
200
100
500
400
300
200
100
80
60
40
20
120
100
80
60
40
20
6·5-6·95
8·0-8·45
9·5-9·95
6·5-6·95
8·0-8·45
9·5-9·95
20-24
25-29
30-34
35-39
40-44
45
Size class (gu)
6·5-6·95
8·0-8·45
9·5-9·95
11·0-11·45
12·5-12·95
14·0-14·45
Number of individuals in size class
SIZE CLASS (mm)

In four species, rudis B, arcana, nigrolineata and neglecta, the null hypothesis can be rejected. Examination of the data, which are plotted in fig. 28, shows that males do not grow as big as females.

2.2.2. Dimorphism in shell height related to reproductive maturity.

A similar χ^2 test was used as in the previous analysis and a comparison made of mature males to females in each size class with the ratio of total mature males to females.

Results

The data for the analysis are given in table 2, Appendix B and the results are given below:

Species	χ^2	Degrees of freedom	Significance
L.rudis C	43.20	8	Sig. at 0.1%
L.rudis B	33.03	13	Sig. at 1.0%
L.arcana	78.36	8	Sig. at 0.1%
L.nigrolineata	35.79	19	Sig. at 5.0%
L.neglecta	209.65	5	Sig. at 0.1%

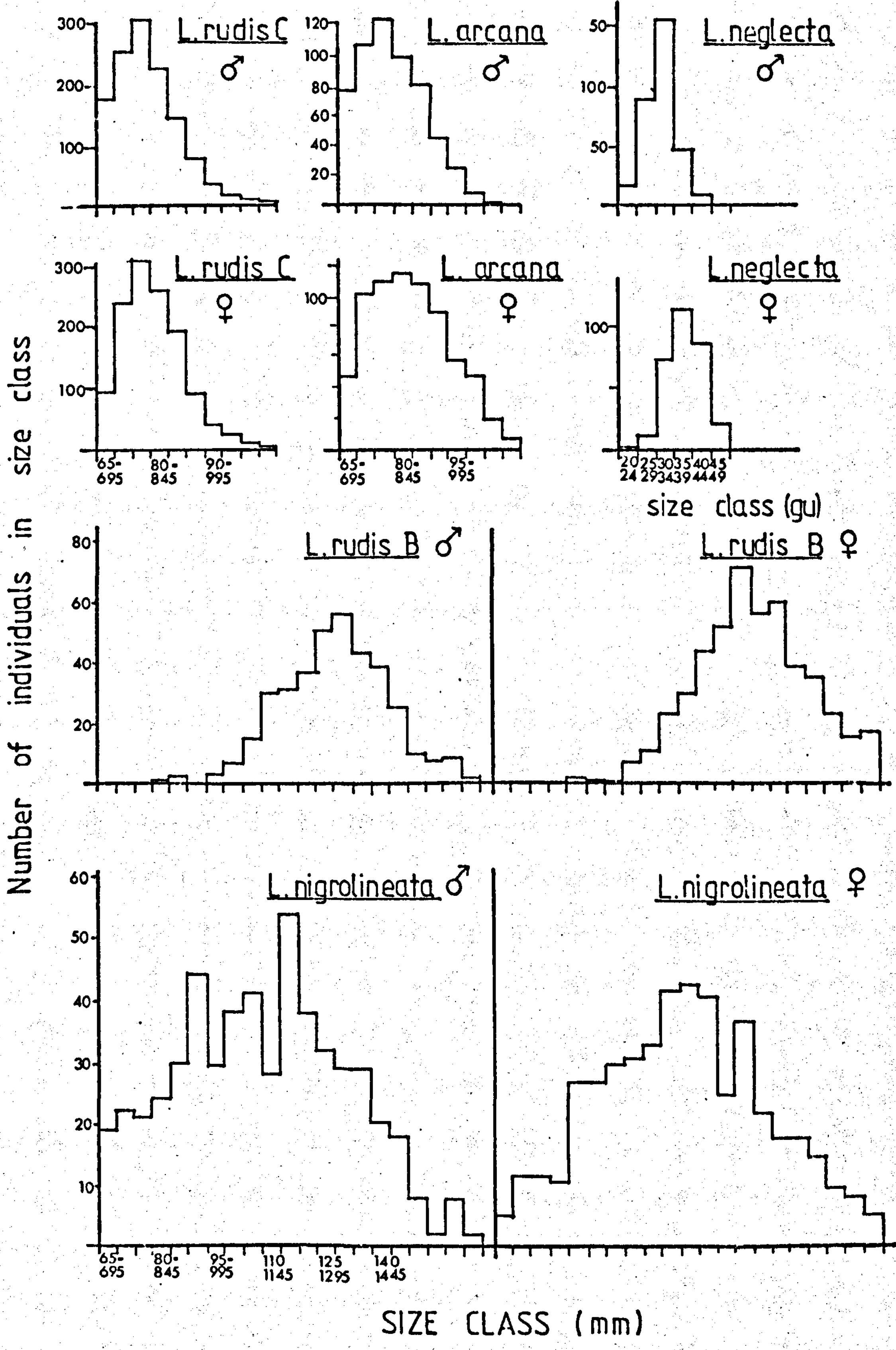
The null hypothesis can be rejected for every species. Examination of the data plotted in fig. 29, shows that males mature at a smaller size than females.

2.2.3. Dimorphism in shell shape related to sex

This analysis compares dimorphism of shape measured by the ratio shell height/shell width (SHT & SW are defined in fig. 24) between males and

Figure 29.

Sexual dimorphism in shell height at maturity.


L.rudis C ♂
300
200
100
L.arcana ♂
120
100
80
60
40
20
L.neglecta ♂
50
100
50
L.rudis C ♀
300
200
100
65-
695
80-
845
90-
995
L.arcana ♀
100
65-
695
80-
845
95-
995
L.neglecta ♀
100
20 25 30 35 40 45
24 29 34 39 44 49
size class (gu)
Number of individuals in size class
L.rudis B ♂
80
60
40
20
L.rudis B ♀
L.nigrolineata ♂
60
50
40
30
20
10
65-
695
80-
845
95-
995
110
1145
125
1295
140
1445
L.nigrolineata ♀
SIZE CLASS (mm)

females. The Mann-Whitney U test was used to compare the shell shape of males with that of females of a species. To avoid errors which might result from changes in shell shape with age, animals of a restricted size range were used. The shell heights of the males and females were compared with a t-test to check that the samples were compatible (see table 3, Appendix B).

Results

The results are summarized in table 6. The null hypothesis, that male and female shell shape do not differ, can be rejected for arcana, nigrolineata and rudis B. In each of these cases examination of the data indicates that females are more globose than the males.

Discussion of sections 2.2.1. and 2.2.2.

The data obtained confirm the existence of size dimorphism related to sex in all species of rough winkle; males maturing at smaller shell heights than females and, with the exception of rudis C, not growing as large as females. In three cases, rudis B, arcana and nigrolineata, females are generally more globular than males and it is notable that nevertheless males of these species are significantly shorter than females. Theoretically if females are more globular this will tend to reduce their shell height, relative to males of equivalent soft tissue size, ie. females, if only shape dimorphism occurred, would appear shorter. Thus the size dimorphism, with males shorter than females, is more pronounced than the data reveal.

Why does size dimorphism occur?

TABLE 6. Comparison of male and female shell shape using Mann-Whitney U test.

Species	Number of individuals ♂	Number of individuals ♀	Range of observations ♂	Range of observations ♀	z	two-tailed p	Significance
L. rudis C	71	66	1.013-1.164	1.007-1.150	.009	.9362	Not significant
L. rudis B	70	75	1.082-1.325	1.075-1.278	2.230	.0258	Sig. at 5%
L. arcana	69	70	1.007-1.188	1.014-1.167	2.680	.0074	Sig. at 1%
L. nigrolineata	59	58	1.083-1.258	1.048-1.223	2.790	.0052	Sig. at 1%
L. neglecta	85	59	.926-1.167	1.000-1.133	.340	.7320	Not significant

For winkles size is a critical factor, known to affect mortality, fecundity and reproductive strategy. Heller (1976) and Elner & Raffaelli (1980) have shown that winkle size is an important factor in crab predation, small winkles being more susceptible to attack. Faller-Fritsch (1977) has proposed that physical causes of mortality, such as crushing or burial (in unstable habitats) and desiccation can be expected to act differently on winkles of differing size, with small winkles at greater risk. In addition the food resources available to a winkle may vary with its size. Smaller winkles presumably are able to penetrate deeper into crevices than larger ones and also get into smaller crevices. They may therefore have access to food resources inaccessible to larger winkles. Larger winkles may have the compensation of being able to range further from crevice shelter during feeding periods.

Raffaelli (1976) has shown that the number of embryos a female carries is influenced by her size, - the larger the female the greater the number of embryos. Size structure of populations of <u>rudis</u>, <u>neritoides</u> and <u>neglecta</u> are known to be affected by microhabitat characteristics notably crevice size and availability (Emson & Faller-Fritsch 1976, Raffaelli and Hughes 1978, Raffaelli 1978a). In turn population structure is related to reproductive strategy in terms of size at maturation, etc. Faller-Fritsch (1977) has shown that exposed populations on stable rocks tend to consist of small individuals which mature at a small size, and he has suggested that maturation is relatively delayed in sheltered, unstable boulder or pebble habitats because energy is devoted to somatic growth, allowing individuals to outgrow the size ranges in which high mortality rates occur.

Since size is undoubtedly an important factor in reproductive success and mortality, differences in rate and pattern of growth in males and females might be correlated to reproductive strategy. Alternative hypotheses eg. that size is merely neutrally linked to sex, or that males are smaller because they are more sensitive to turbulence (Sacchi, 1968) or that females are more globular to accomodate the reproductive organs (Naylor, 1978, unpublished report) seem rather unlikely in view of the dominant importance of size in the survival and reproductive rate of winkles, and the inconsistency of expression of sexual dimorphism.

2.2.4. Dimorphism related to colour polymorphism in L.nigrolineata

At Porth Swtan L.nigrolineata exhibits shell colour polymorphism. It is either yellow- or white-shelled. There are no intermediates and although the yellow shells show a range of shades from very pale to primrose yellow, the distinction between white and yellow is easily made. Of the 2906 nigrolineata collected at the site between June 1976 and February 1978, 222 (or 7.64%) were the rarer white-shell morph.

It was noticeable that the commonness of the white-shell morph depended on size class; there were relatively more white-shelled animals in the smaller size classes than in the larger size classes. It was also apparent that white-shelled animals were frequently mature when yellow-shelled animals of equivalent shell height were not. These aspects are explored in sections 2.2.4.1. and 2.2.4.2. below.

2.2.4.1. Size dimorphism between colour morphs.

The size structure of the yellow-shelled morph population was compared with that of the white using a χ^2 test (Brandt & Snedecor's formula). The observed ratios of white and yellow shells in each size class were tested for homogeneity with the overall ratio of white- to yellow-shelled morphs.

Results

The data are given in table 4, Appendix B and are plotted in fig. 30. In brief, $\chi^2 = 73.34$ with degrees of freedom = 15, which is significant at .001 (or .1%). As fig. 30 illustrates, there are relatively fewer white-shelled morphs in the larger size classes. The difference is highly significant.

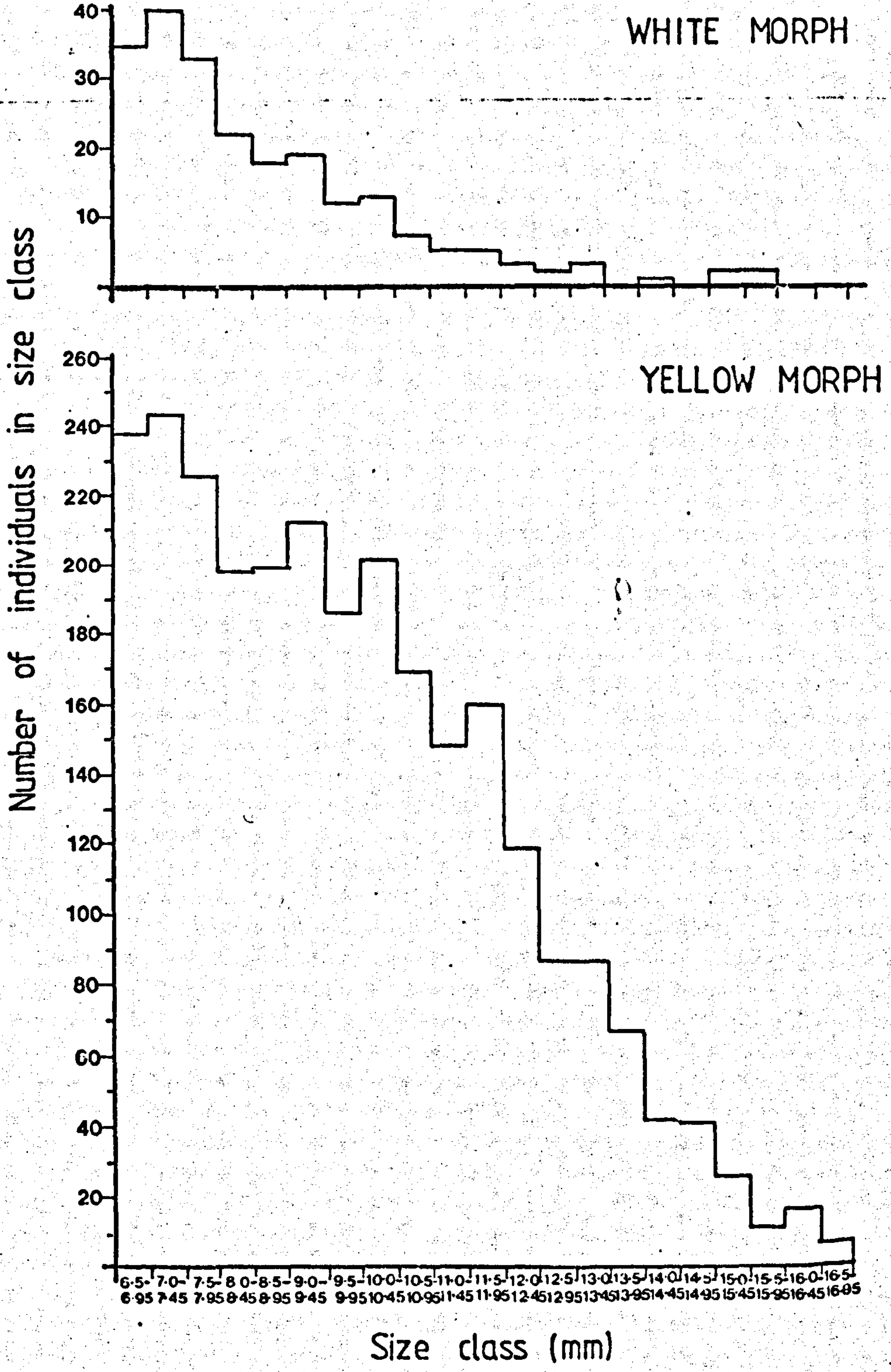
2.2.4.2. Comparison of size at maturity of white and yellow morphs.

Yellow- and white-shelled animals were scored for maturity on the basis outlined in section 2.1. and the number of mature males and females and the overall number (females + males) in successive size

Figure 30.

Size dimorphism of L.nigrolineata colour morphs.

SIZE DIMORPHISM OF L.NIGROLINEATA COLOUR MORPHS



classes were recorded. The percentage mature in each size class for the two colour morphs was then calculated.

Results

The data are given in table 7 and it is clear, by comparing the percentage mature of white- with yellow-shelled animals in successive size classes, that white morphs are much more likely to be in a mature condition at a smaller shell height than yellow morphs, for instance in the size range 6.5 -7.45mm about 25% of white-shelled animals were mature compared to about 12% of yellow-shelled.

Discussion

From the data it seems that white-shell is linked to maturation at a small size. This presumably results from shell colour being linked to growth, either since white-shelled animals are slower growing (maturation occurring at a set age) or since early maturation (with its concomitant demand for energy) retards growth. It would be interesting to compare growth rates of white- and yellow-shelled juveniles.

Why this association of shell colour and size occurs has not been studied. However since size and fecundity are related, the differences between white- and yellow-shelled populations will affect their relative reproductive success. Theoretically white-shelled animals, tending to be smaller, will be less fecund (fewer juveniles produced per unit time) than their yellow counterparts. Thus if the white-shelled

TABLE 7. Size at Maturity of *L. nigrolineata* Colour Morphs

Size class (mm)		6.5- 7.45	7.5- 8.45	8.5- 9.45	9.5- 10.45	10.5- 11.45	11.5
White-shelled	Mature	19	19	16	12	9	15
	Total	75	55	37	25	12	18
	% Mature in size class	25%	35%	43%	48%	75%	83%
Yellow-shelled	Mature	58	68	128	128	145	481
	Total	486	422	412	383	317	664
	% Mature in size class	12%	16%	31%	33%	46%	72%

animals are not to be out-competed and so rendered extinct, there must be some selective force(s) acting differentially on the morphs; always assuming that the colour polymorphism is stable and the white morph is not in the process of becoming extinct. This selective force must act in such a way that yellow morphs suffer higher mortality than white morphs.

So, though a selective force can be postulated since reproductive tactics of white- and yellow-shelled populations differ, what the selective force is can only be guessed at. Possibly the colours have no direct significance, eg. if colour was related to strength of the shell and white shells were stronger than yellow but required more energy to construct, so that the extra protection would reduce the risk of mortality and so compensate for reduced size and reproductive rate by increasing reproductive life. However, Raffaelli (1976) has shown that for L.rudis there is no difference in thickness of differently-coloured shells. Alternatively, the visual polymorphism might be of direct importance, white- and yellow-shelled animals may be subject to different predator pressures on account of their shell colour and the two morphs may be maintained either as they are cryptic on different backgrounds or through apostatic selection (Clarke, 1962). The difference in size structure and size at maturity might be further responses to predator pressure eg. if large white-shelled animals were selectively predated, this should encourage early maturation, probably with concomitant slowing of growth.

2.3. AGE-RELATED CHANGES IN L. NEGLECTA SHELL COLOUR PATTERNS

In Anglesey neglecta shells have commonly a pale ground colour with dark brown pigmented patterns. More rarely all-over colours, white, off-white or black, are seen. The most constant feature of patterned shells is a wide dark brown band that runs parallel to the suture, in the lower half of the body whorl. Between this band and the body whorl suture, pigment can be laid down in a variety of patterns, notably as bands, bands and tessellations or tessellations. There is a wide variation in shell ground colour white, biscuit and dull yellow are common and the marking pigment can be any shade of brown (usually) or yellow (less commonly). Mixtures of differently coloured marking pigments on one shell are frequent.

Casual observations suggested that with patterned shells the pattern is age-related. Small shells are frequently observed to have only complete bands, larger shells to have only tessellations in addition to the wide band. On some shells gradations from one pattern to another were seen e.g. complete bands were apparent on the oldest part of the shell, younger parts had mixtures of bands and tessellations. An investigation of possible ontogenetic change in pattern was made.

Method

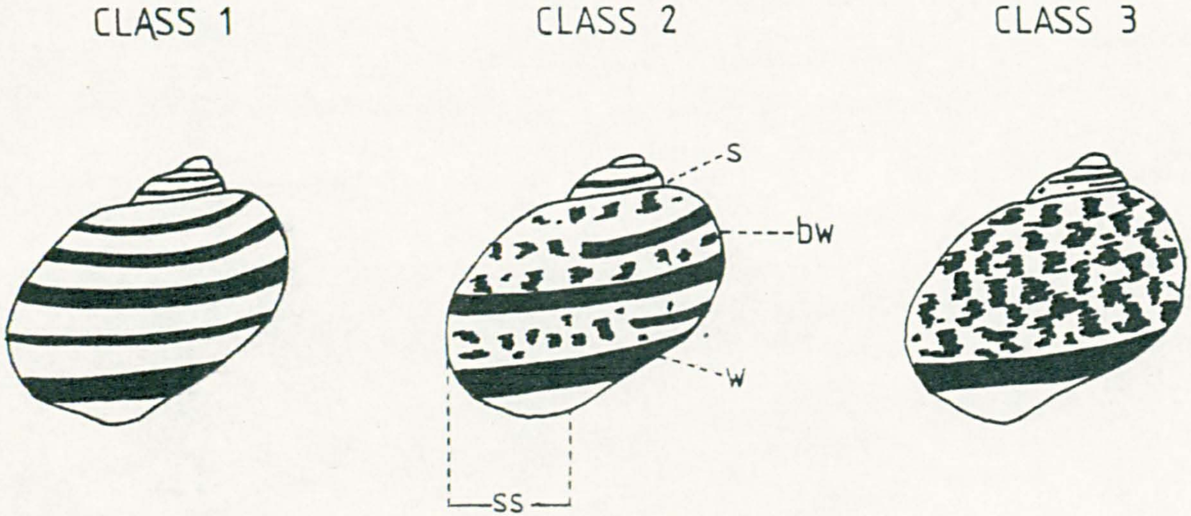
Several hundred neglecta were collected from Porth Swtan and Porth Diana. For each individual, shell height was measured in graticule units (g.u.) and the marking pigment colour scored into on the following classes (see fig. 31):-

Figure 31.

Classes of colour pattern in L.neglecta.

Key: ss:- scored section
bw:- body whorl
w:- wide band
s:- suture

CLASSES OF COLOUR PATTERN IN L. NEGLECTA



	CLASS
i) completely banded	1
ii) mixed bands (excluding wide band) and tessellations	2
iii) completely tessellated (excluding wide band)	3

Of the body whorl only the youngest part (scored section, ss, fig. 31) was scored for pigment pattern.

Results

The data appear in Appendix B, tables 5 and 6. The frequency of each pattern class in each size class is plotted in fig. 32. Graphs G and H illustrate the percentage of each pattern in each size class, omitting size classes with less than 20 individuals.

At both sites class 1 markings (all-over bands) are commonest in the smaller size classes, class 2 (mixed bands and tessellations) in the middle range of size classes and class 3 (all-over tessellations) in the larger size class.

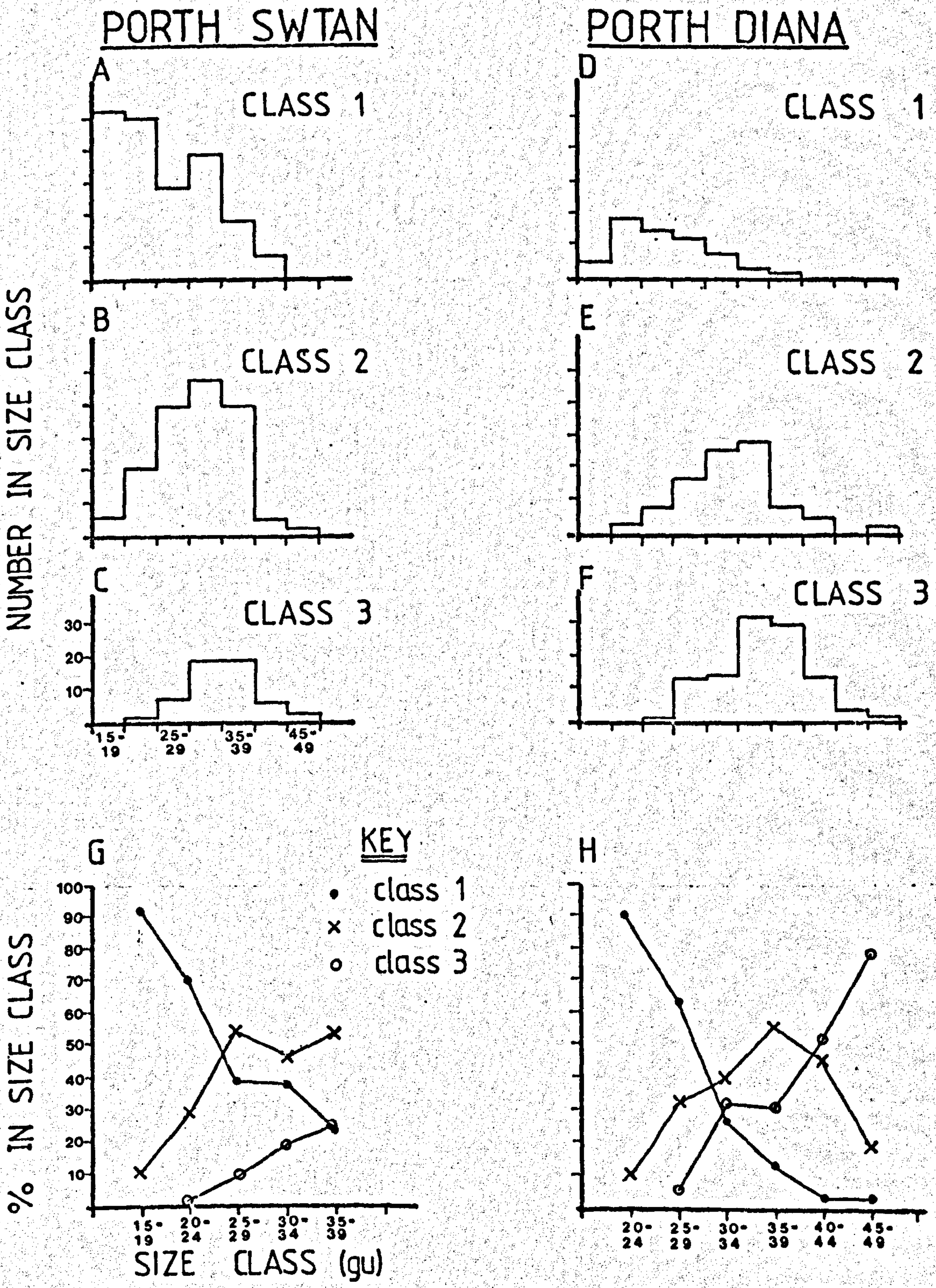
DISCUSSION

Theoretically, the association of size and colour pattern could arise through differential mortality related to colour patterns at particular sizes. However, the scope for such an effect is restricted by the very compressed size range exhibited by *neglecta*. The alternative hypothesis, that the pattern of pigment laid down changes with age, seems more likely, in view of the marked association of pattern type with shell height recorded here and the frequent occurrence of shells which have bands on older parts,

<u>Figure 32</u>.

Frequency of colour patterns in *L.neglecta*.

FREQUENCY OF COLOUR PATTERNS



and tessellations on younger (the reverse situation has never been observed). It is of course possible that both differential mortality and shell pattern change occur.

Marked changes in size structure and maturity structure of the neglecta population at Porth Swtan indicate that this species is an annual (see section 2.5). Since size structure of the population changes seasonally the frequency of the colour patterns 1, 2 and 3 must also be expected to change. Thus the overall percentage of striped shells in a population must be expected to alter over the year. Anderson (1974) when correlating percentage of striped nigrolineata with percentage striped neglecta and other environmental factors was unaware of this factor (pers. comm.).

2.4. REPRODUCTIVE SEASONALITY IN ROUGH WINKLES

A common feature in the family Littorinidae is an annual cycle of sexual maturity marked by the seasonal ripening and regression of the genital organs. For L.saxatilis, periodicity of reproductive activity has been noted by Bergerard (1971a & b), Berry (1961) and James (1968b). Bergerard found a decline in sexual activity in the summer, and Berry in the months of May, June and July and a smaller decline in February which he correlated with the particularly cold conditions at that time. James suggested that there were two periods of high reproductive activity, January and February, and July and August. These studies were almost certainly based on rudis. No previous studies have been made of reproductive periodicity in nigrolineata, neglecta or arcana.

2.4.1. Variables of the samples used for the assessment of reproductive seasonality

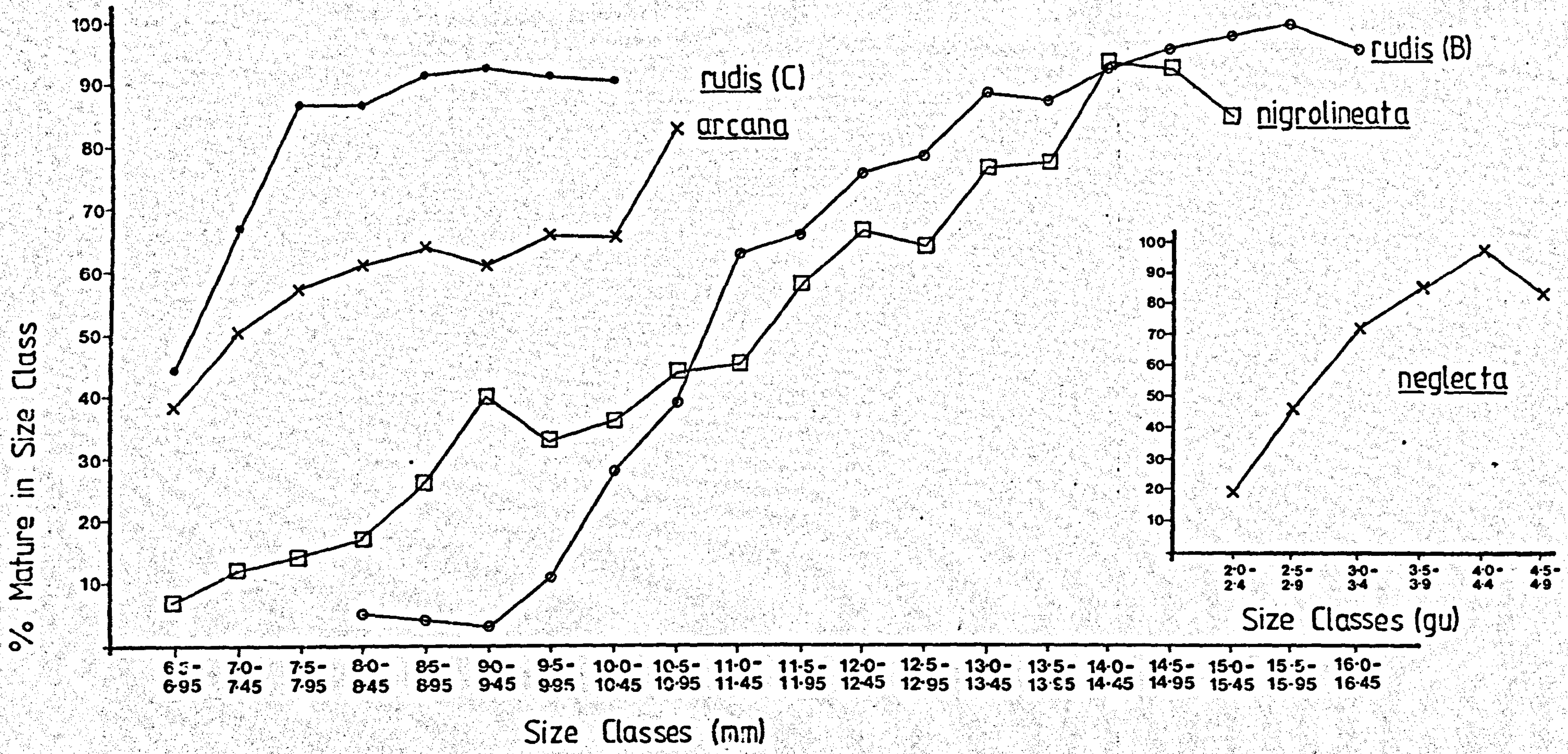
Approximately every four weeks (for precise dates see table 8, Appendix B) samples of arcana, rudis B, rudis C, nigrolineata and neglecta were collected at Porth Swtan. For each animal, shell height, sex, maturity or immaturity (based on the visual assessment as outlined in section 2.1.) and the size of the reproductive organs (PL in males, see fig. 10; c and w in females, see fig. 27) were recorded; the size of the reproductive organs in neglecta, excepted.

It was noticeable that size influenced the likelihood that an animal would be collected in a mature condition; the larger the animal the more likely it was to be mature. Fig. 33 shows the percentage mature in each size class for each species. Since the data for this figure were amalgamated from animals collected over a year (March 1977 - February 1978) the graphs indicate that the larger winkles are in reproductive condition for a longer part of the year. A similar situation has been noted in littorea by Williams (1964). In view of this, and also because the sampling is not free of bias related to size, it was necessary to confine the examination of reproductive periodicity to animals of some defined size range and not to use the monthly samples in their entirety. It is impracticable, if not impossible to avoid size bias when sampling at Porth Swtan (with the probable exception of neglecta) because of the nature of the environment in which the winkles live. The crevices of a rocky shore, though accessible to winkles are not always so to winklers and the conditions at the time of collection can affect the size structure of the sample. If it is hot and dry the winkles often move more deeply into crevices and consequently it becomes more difficult to collect them, and disproportionately more difficult to collect smaller winkles as they can move further into crevices than larger animals. The environment from which samples of neglecta were collected differs in that there are few if any crevices in the transect area chosen and the dead

<u>Figure 33</u>.

Percentage of mature individuals in successive size classes.

PERCENTAGE MATURE IN SUCCESSIVE SIZE CLASSES



shells of barnacles, which they favour for the shelter they provide, are easily broken open and the winkles collected. It is therefore rare for an individual of neglecta to be inaccessible.

Since sampling bias is unlikely in neglecta the monthly sample of this species was used in its entirety. For the other species, appropriate size ranges were selected and the assumption made that immature individuals of size x were as likely to be collected as mature individuals of the same size. The size ranges used were:-

L.arcana	7.0 - 8.95mm
L.rudis C	7.0 - 8.95mm
L.rudis B	11.0 - 14.45mm
L.nigrolineata	8.5 - 12.95mm
L.neglecta	$\geqslant$2.0gu (1mm = 1.7gu)

Two methods of assessing reproductive periodicity were explored, a) percentage of animals, in selected size range, in mature condition b) reproductive organ size frequency structure of animals in the selected size range.

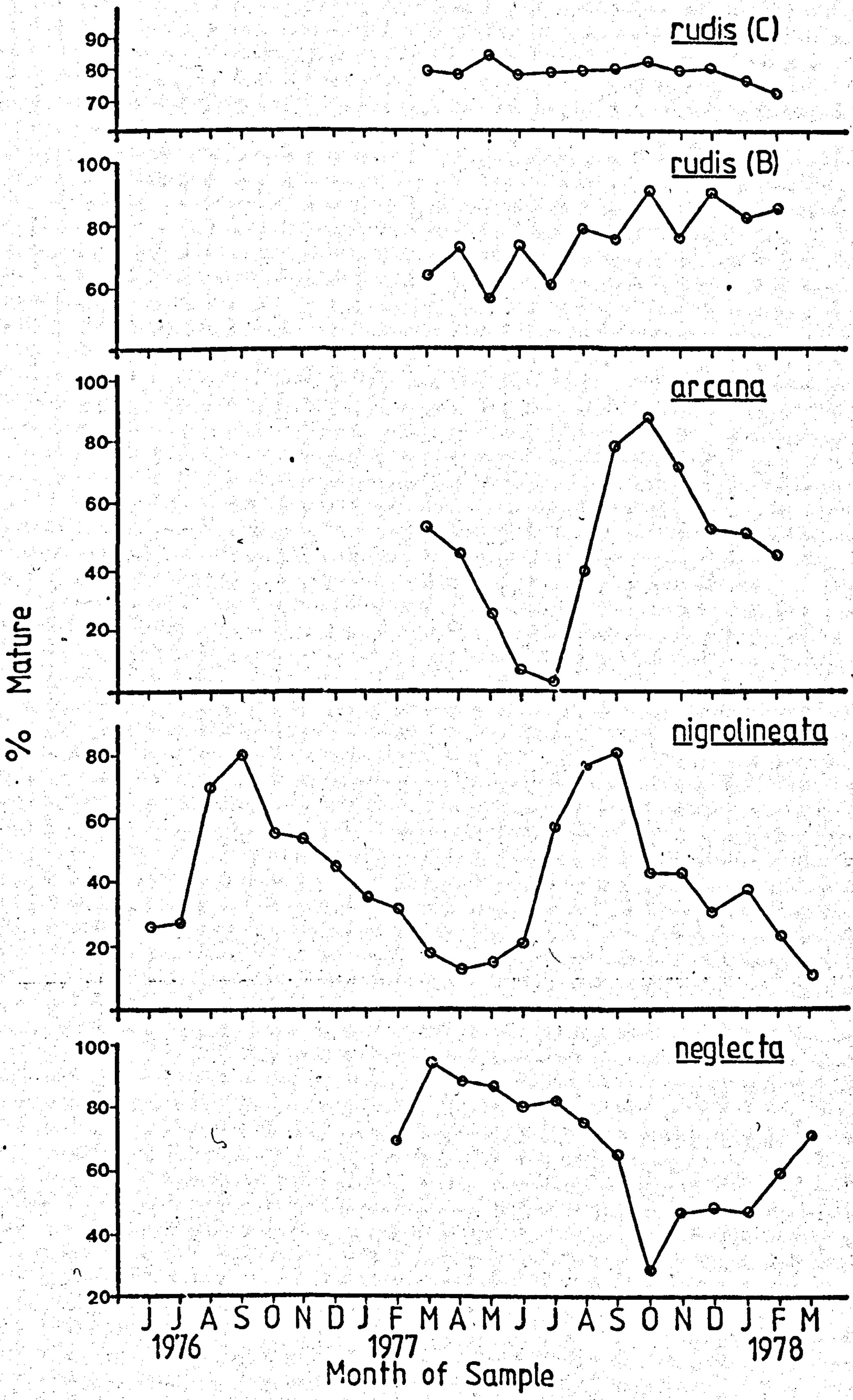
2.4.2. Examination of reproductive periodicity based on percentage of mature individuals.

In the chosen size range the percentage of mature animals, for each month and species was found. Data from males and females were combined. The results are plotted in fig. 34 and the data are given in table 8, Appendix B.

<u>Figure 34</u>.

Reproductive periodicity.

REPRODUCTIVE PERIODICITY



Results

a) *L.rudis* C

No indication of any cycle of reproductive activity was apparent. The population maintained a fairly constant and high level of reproductive activity throughout the year with about 80% of individuals in the 7.0-8.95mm size range being mature in each month.

b) *L.rudis* B

The results from this ecotype were erratic and no clearly defined pattern was apparent. It is notable that the lowest levels of activity were recorded in May and July, corresponding with Berry's findings for Whitstable. The population he studied lived in a habitat more similar to that of the *rudis* B population than the *rudis* C population. Like *rudis* C, the *rudis* B population generally maintained a high level of reproductive activity throughout the year.

c) *L.arcana*

A marked cycle of reproductive activity was apparent in this species, with low percentages in reproductive condition (less than 30%) in May, June and July and higher percentages (more than 70%) in September, October and November. Though data from the previous year were unavailable for this analysis (as the method of collection was affected by migration,(see section 3.2.2.) observations indicated that a similar cycle occurred in the previous year, 1976-1977.

d) L.nigrolineata

Data for this species were obtained for a period of 22 months and, as in arcana, cyclic activity was apparent. Peaks of activity, with more than 70% of the population mature, were observed in August and September 1976 and 1977. Activity fell below 30% in June and July 1976, May to July 1977 and May 1978.

e) L.neglecta

Cyclic reproductive activity was apparent in this species also. The period of high activity was comparatively long; from March to August (1977) more than 70% of the population were in reproductive condition. The lowest level of activity was recorded for October 1977.

2.4.3. Examination of reproductive seasonality based on monthly frequency distribution of relative reproductive organ size.

In successive months, Berry (1961) looked at the numbers of embryos in the brood pouch of 20 rudis females of 10 - 11mm shell height and was able show a saesonal cycle of embryo production. In rudis the numbers of embryos is paralleled by the size of the brood pouch (Hart, 1978, unpublished report) the bigger the brood pouch the more embryos contained (also, the converse). Thus the relative size of the reproductive organ may be useful as a measure of the reproductive rate ie. a larger oviduct may indicate a higher rate. A similar relationship could apply in the oviparous species, and perhaps an equivalent relationship may occur in males.

For females the relative size of the reproductive organs was defined as the length of the brood pouch or jelly gland (c) plus width (w) (see fig. 27) divided by the shell height. For males this was defined as penis length (PL, see fig. 10) divided by the shell height.

Results

The monthly frequency distributions of these ratios were plotted as histograms (figs. 35 - 42) for each species and sex. The data appear in table 9, Appendix B. As in the previous section the data used were derived from individuals in the selected size range as the sampling was not free of size bias. The data presented have not been standardised on a percentage basis as this would obscure the relative reliability of the data collected in a month; in some months comparatively few animals, in the size range ultimately adopted for these analyses, were collected as the effect of shell height on likelihood of maturity was underestimated. This particularly affects the results presented for rudis B and nigrolineata, which both exhibit a wide range of shell heights so what seemed to be a large sample was not so satisfactory as appearance suggested. The histograms generally show bimodality which is due to the reproductive organs not increasing in size in step with the growth of the animal but rather showing a spurt of growth from immature state to mature, and an equally marked regression at the end of the breeding season (see section 2.1.). Variation in the position of the second, larger, ratio peak is of particular interest as this may reflect differences in reproductive rate which may not be apparent from calculating the percentage mature in successive months.

a) <u>L.rudis</u> C

In this species PL/SHT ratios $\geqslant .5$ and c + w/SHT ratios $\geqslant 1.0$, generally correspond with animals scored as mature. For both males and females no well-defined movement of the size frequency distribution is seen, though there is some spreading of the second peak, particularly in the females, in the months of January and February. This may indicate some lowering of the reproductive rate at this time of year.

b) <u>L.rudis</u> B

In this species PL/SHT ratios $\geqslant .4$ and c + w/SHT $\geqslant 1.0$ generally correspond with animals scored as mature. The data obtained from this species were few so the results are questionable. In both sexes there is a tendency for a shift towards lower ratios (in mature specimens) during the spring/summer months April-August compared to the autumn/winter months of October-January.

c) <u>L.arcana</u>

In this species PL/SHT ratios $\geqslant .4$ and c + w/SHT ratios $\geqslant 1.0$ generally correspond with animals scored as mature. The data obtained from this species showed a well-defined sequence of change of the

Figure 35.

L.rudis C: Monthly frequency distribution of penis size.

L. RUDIS (C): MONTHLY FREQUENCY DISTRIBUTION OF PENIS SIZE

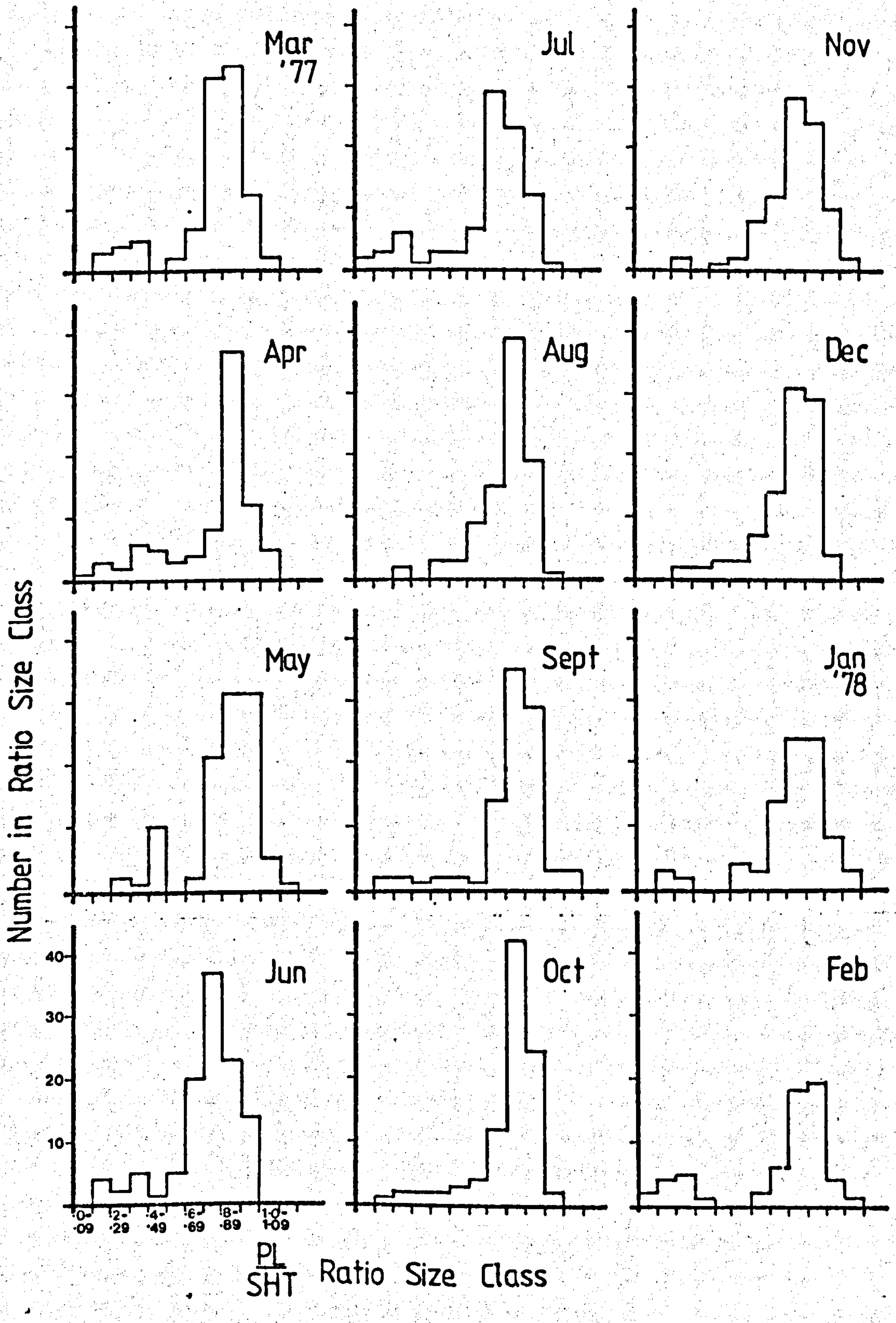


Figure 36.

L. rudis C: Monthly frequency distribution of pallial oviduct size.

L. RUDIS (C): MONTHLY FREQUENCY DISTRIBUTION OF PALLIAL OVIDUCT SIZE

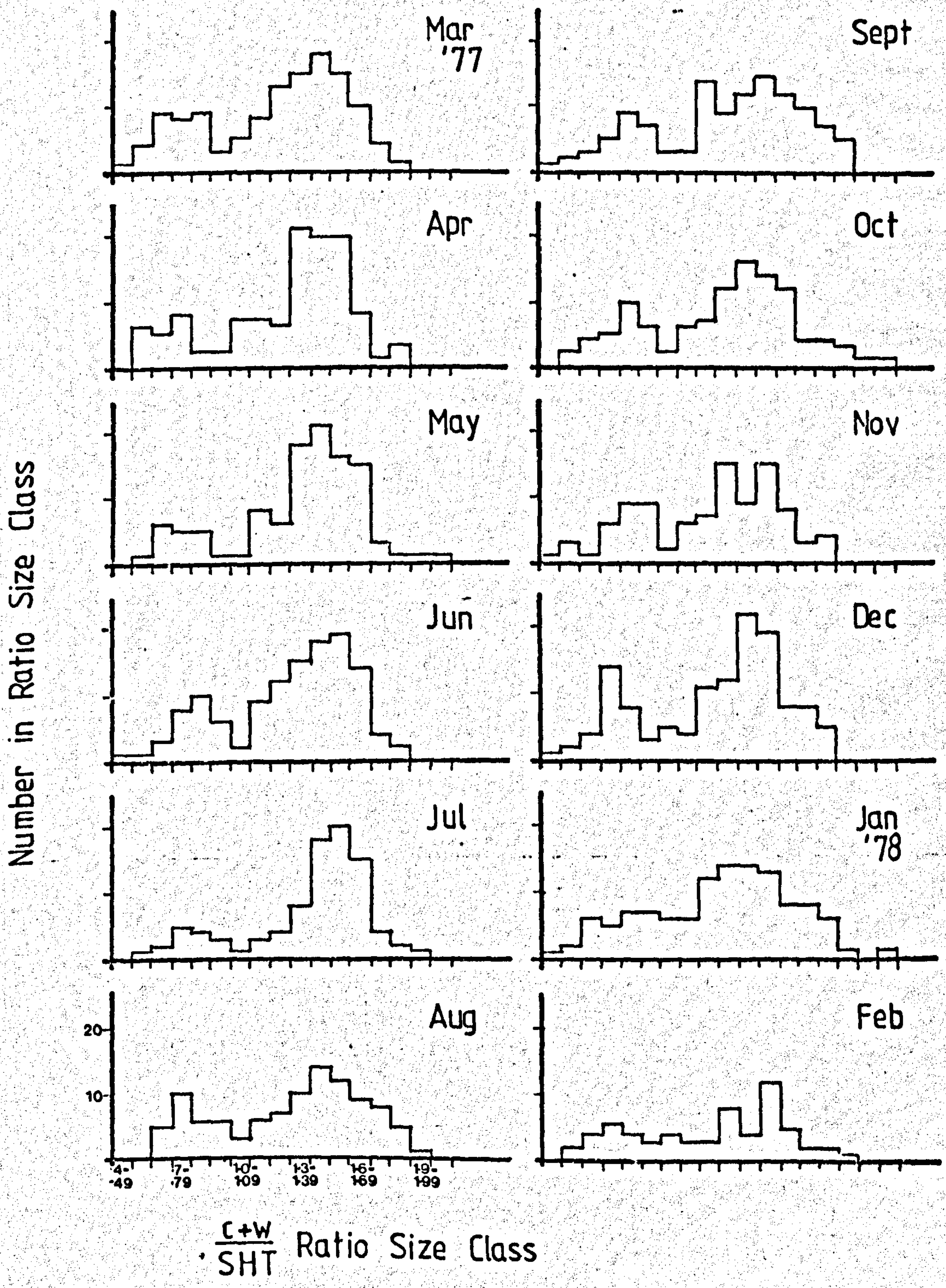


Figure 37.

L.rudis B: Monthly frequency distribution of penis size.

L. RUDIS (B): MONTHLY FREQUENCY DISTRIBUTION OF PENIS SIZE

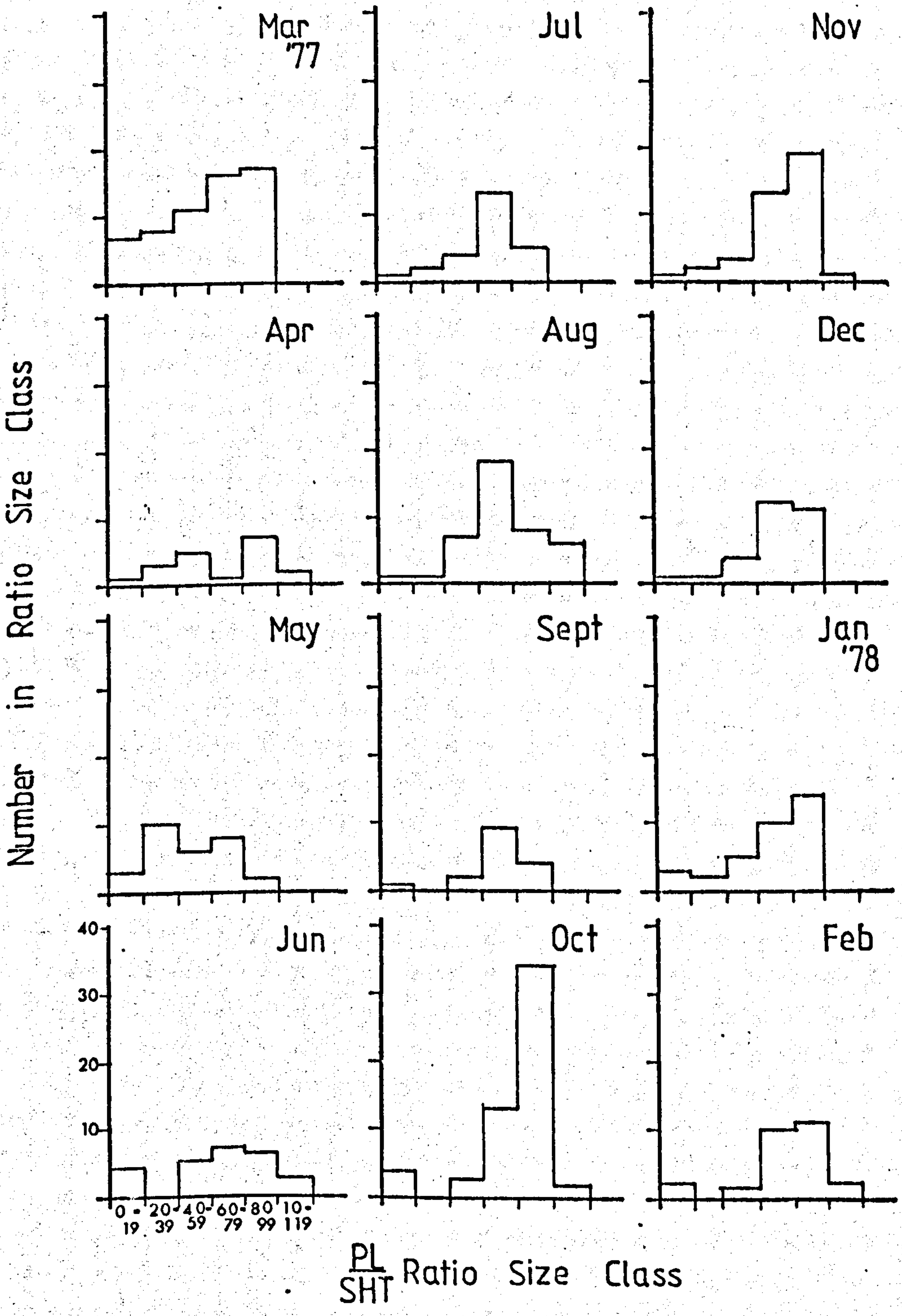


Figure 38.

L. rudis B: Monthly frequency distribution of pallial oviduct size.

L. RUDIS (B): MONTHLY FREQUENCY DISTRIBUTION OF PALLIAL OVIDUCT SIZE

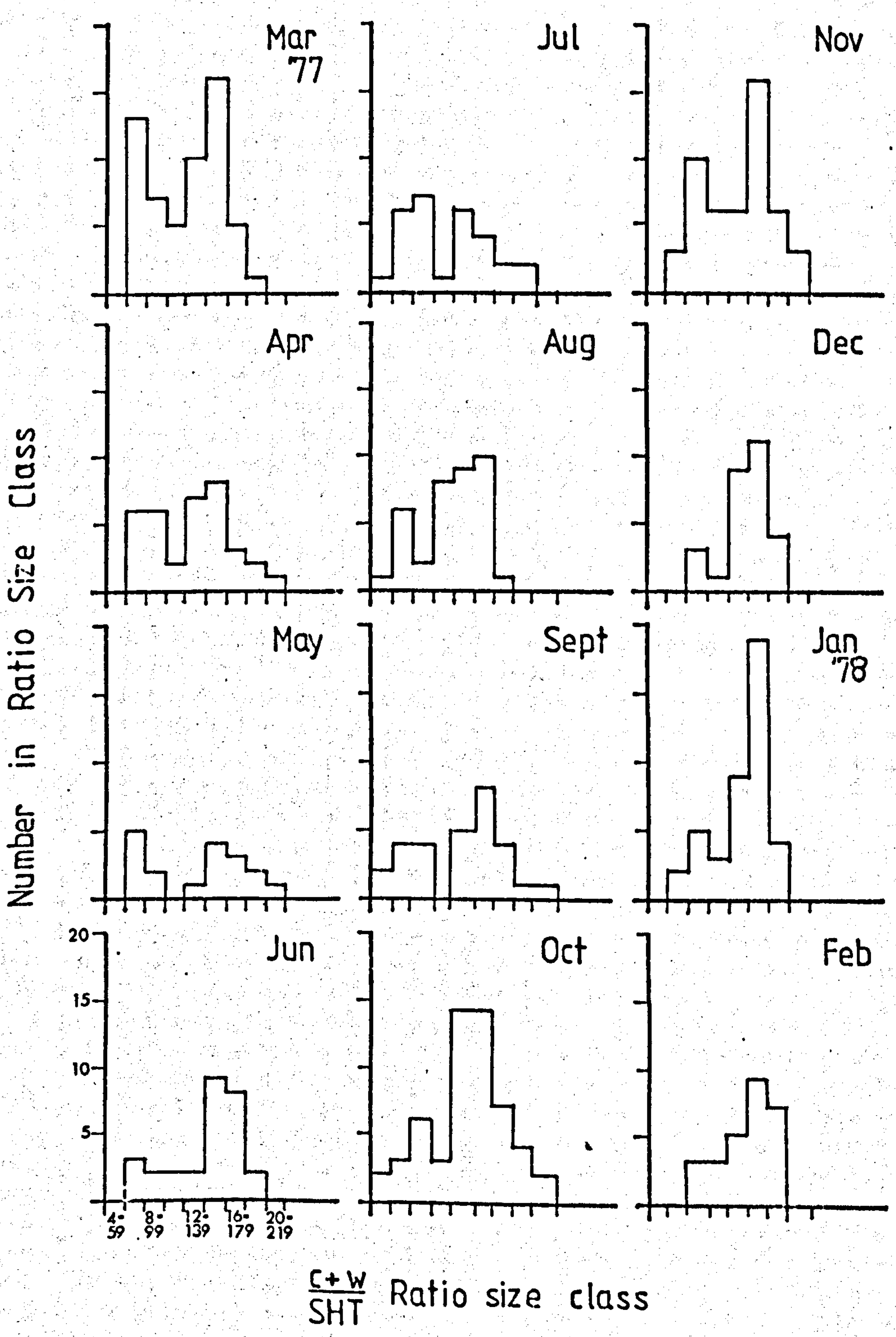


Figure 39.

L.arcana: Monthly frequency distribution of penis size.

L.ARCANA: MONTHLY FREQUENCY DISTRIBUTION OF PENIS SIZE

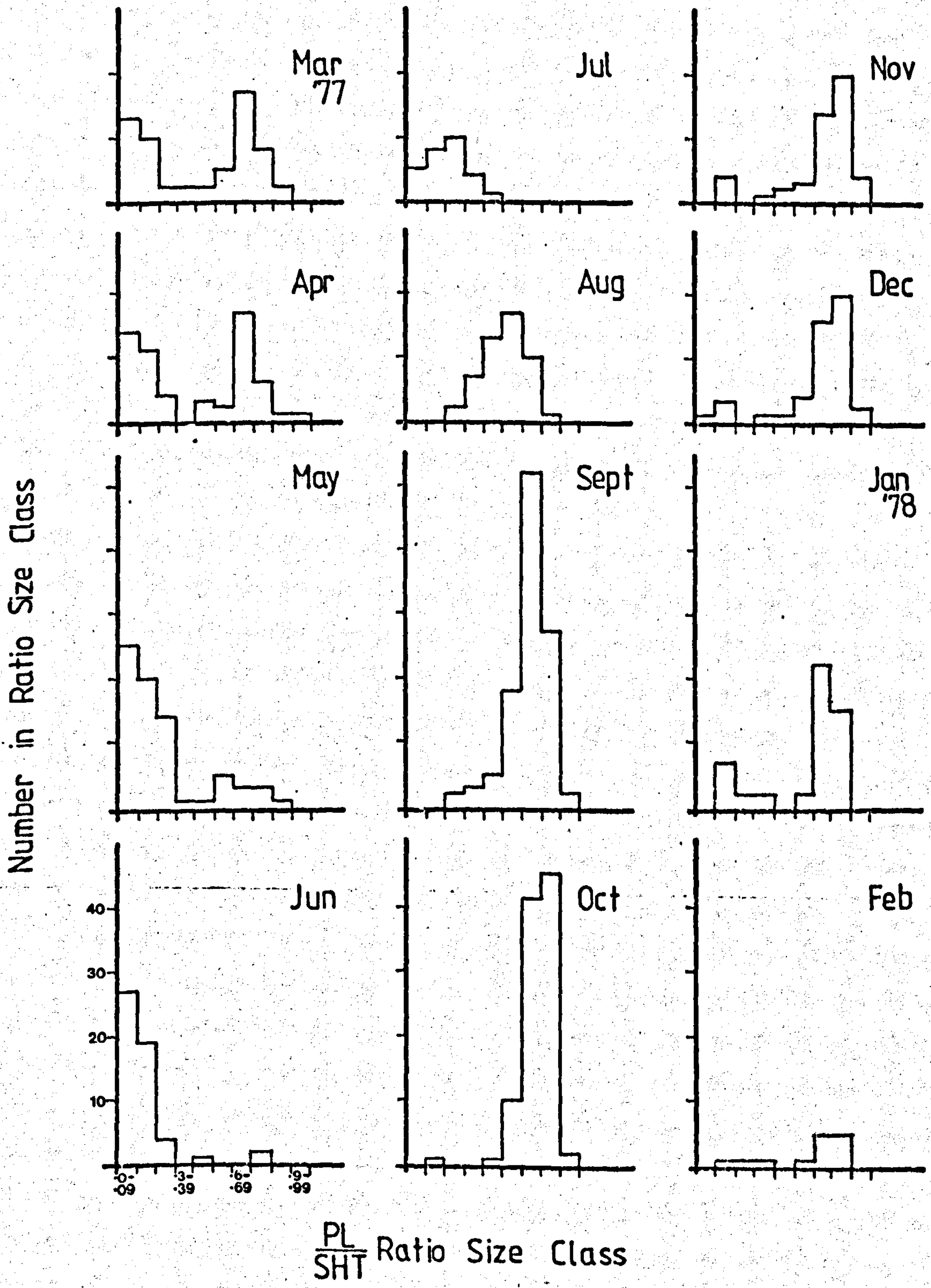


Figure 40.

L.arcana: Monthly frequency distribution of pallial oviduct size.

L.ARCANA: MONTHLY FREQUENCY DISTRIBUTION OF PALLIAL OVIDUCT SIZE

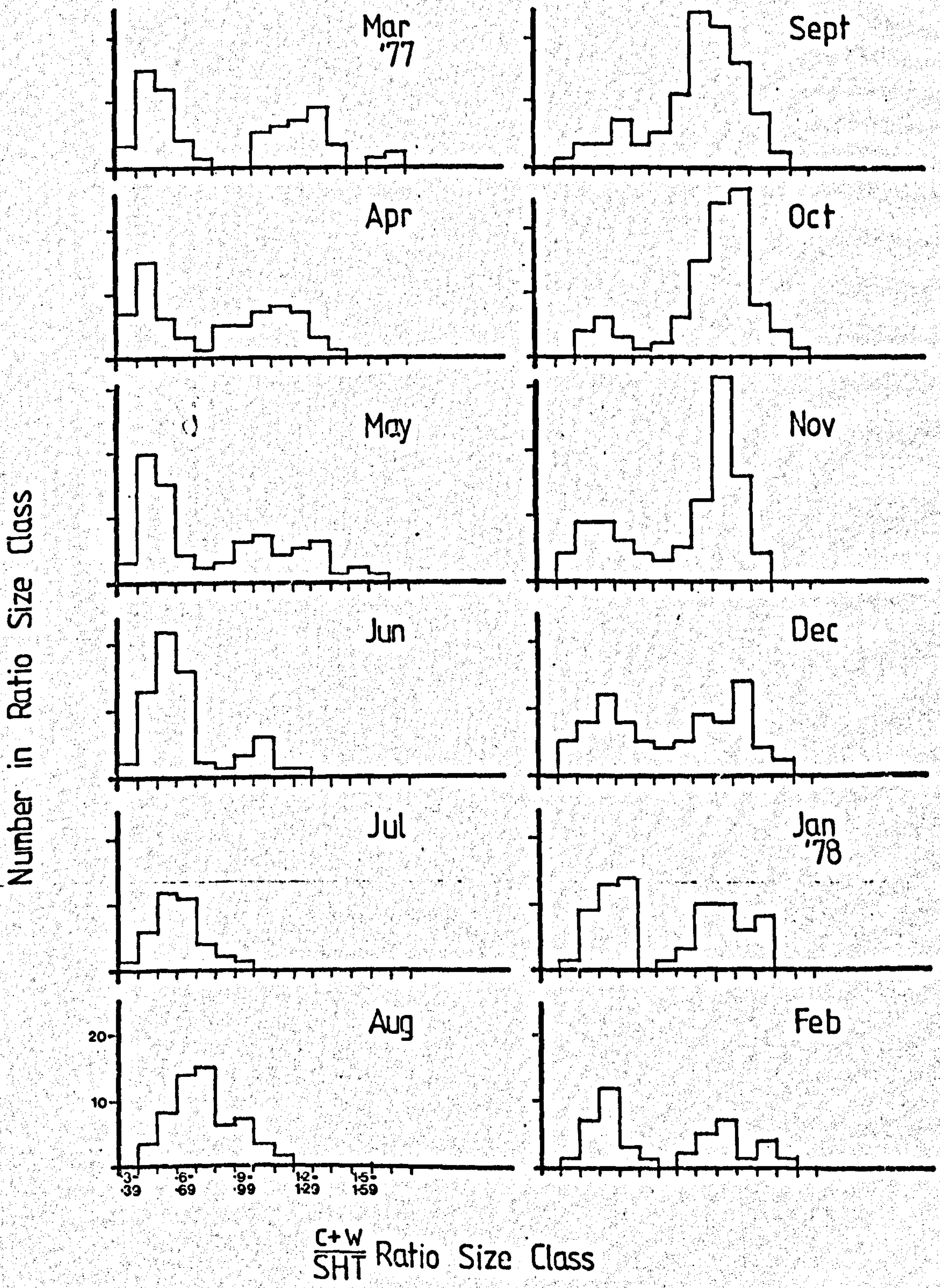


Figure 41.

L.nigrolineata: Monthly frequency distribution of penis size.

L. NIGROLINEATA: MONTHLY FREQUENCY DISTRIBUTION OF PENIS SIZE

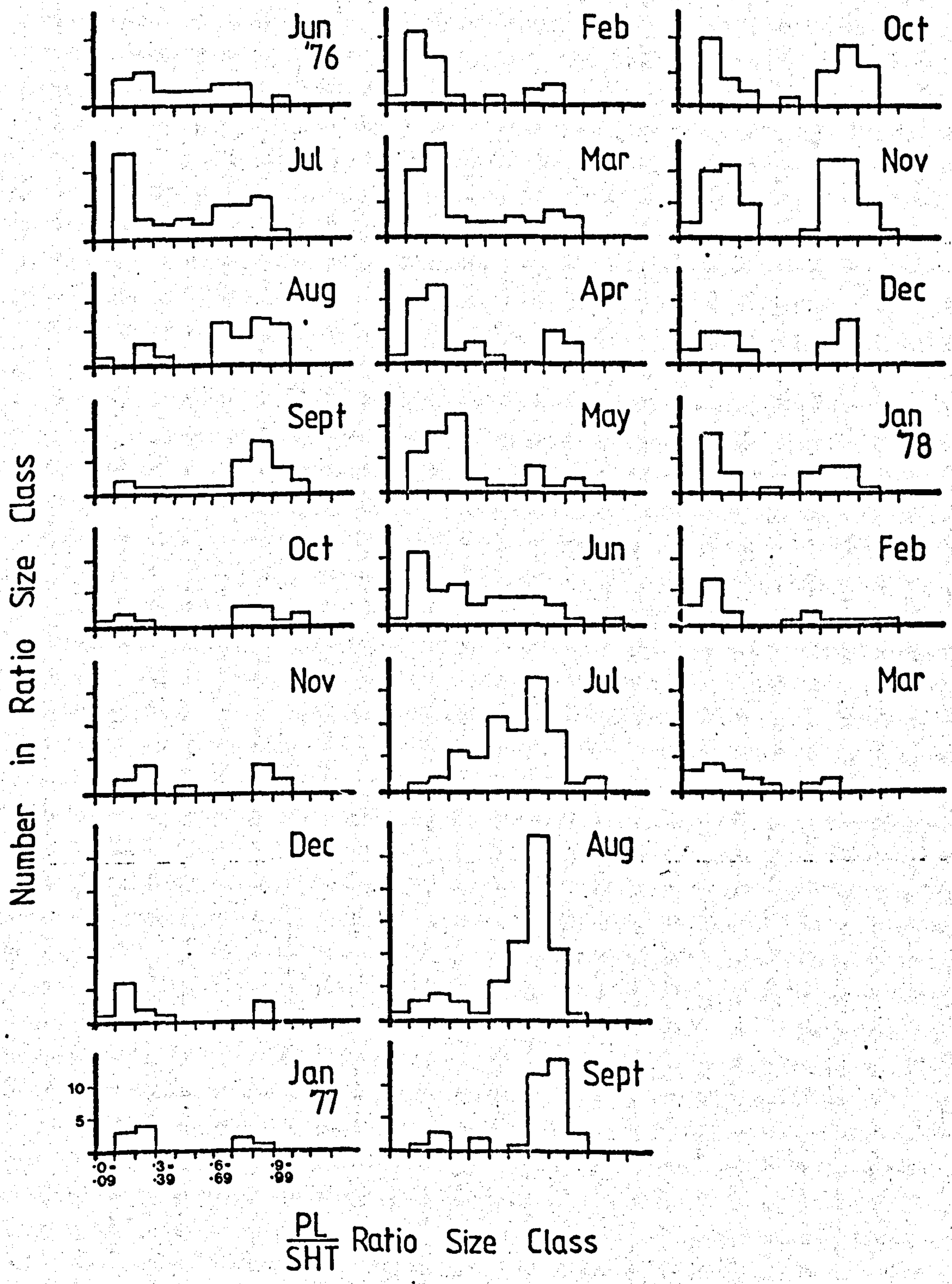
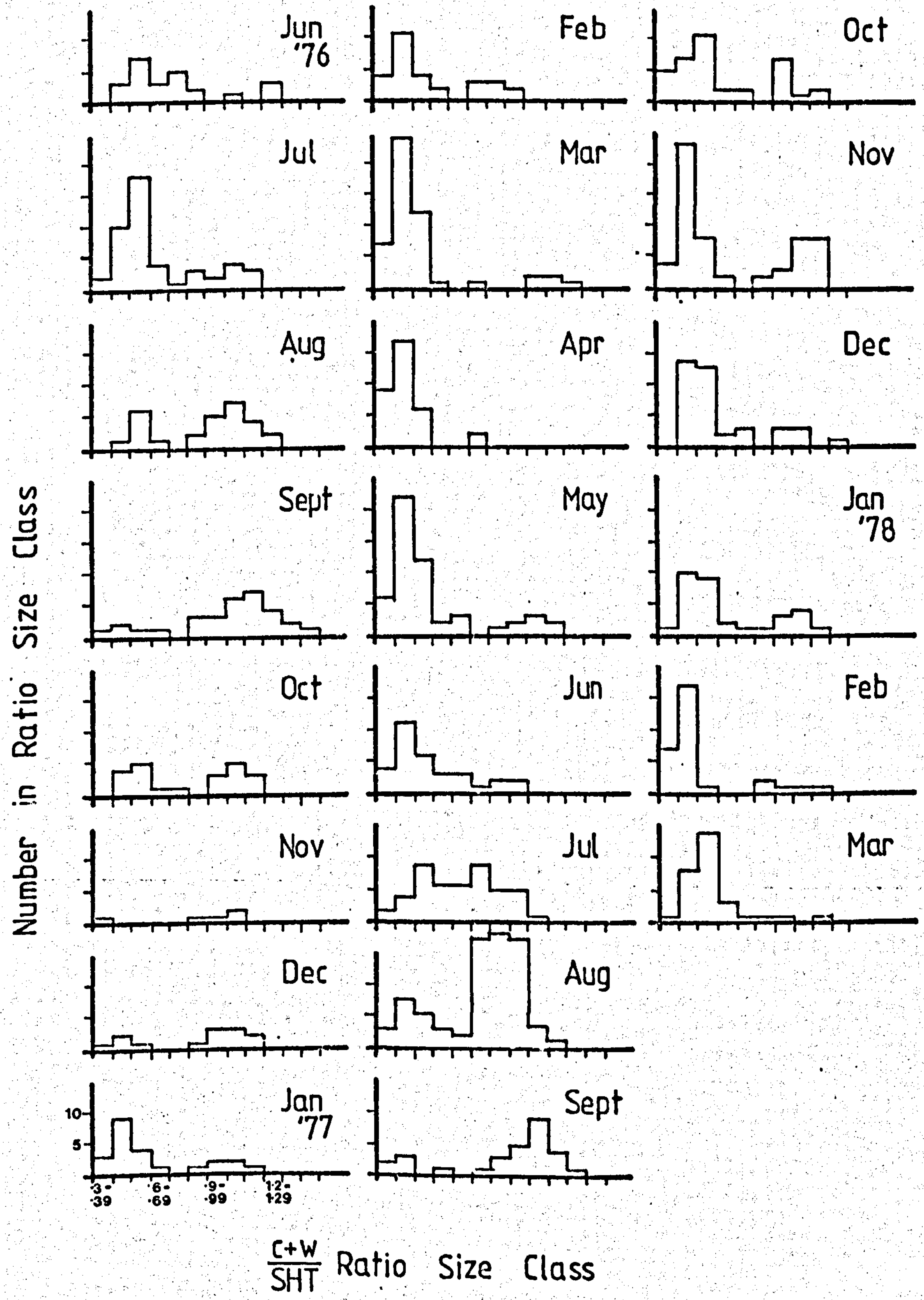


Figure 42.

L.nigrolineata: Monthly frequency distribution of pallial oviduct size.

L. NIGROLINEATA: MONTHLY FREQUENCY DISTRIBUTION OF PALLIAL OVIDUCT SIZE



size frequency distribution indicating a marked seasonality of reproduction. It is also notable that the males are coming into reproductive condition in August, a month prior to the females, and are mostly out of condition in June, whereas some females are still mature at this time, ie. the periodicity of males and females is very slightly different.

d) L.nigrolineata

In this species PL/SHT $\geqslant .6$ and c + w/SHT ratios $\geqslant .8$ generally correspond with animals scored as mature. As in arcana the marked seasonality is readily apparent and also males are coming into reproductive condition sooner than the females. However the data do not suggest that they also go out of reproductive condition earlier than females, unlike arcana.

Discussion of section 2.4.

The slight staggering of reproductive periodicity in the two sexes of both oviparous species is of particular interest, males of both species coming into season before the females and in arcana males also going out of season earlier. This is probably related to differences in male and female reproductive strategy (see also section 2.5., outlining a similar occurrence in neglecta). Males, by coming into season relatively early will miss fewer opportunities of mating with females that come into season earlier than the main mass.

In the two rudis populations there is no comparable reproductive periodicity; both ecotypes exhibit a fairly high level of activity throughout the year. However there is some suggestion of slightly lower rates of activity in January and February in *rudis* C and from April to August in *rudis* B, but for the latter ecotype the data are limited.

The other ovoviviparous species, *neglecta*, shows a marked cycle of reproductive activity with high activity in the summer months. It is notable that all the ovoviviparous species have the capability of breeding in the hotter months of the year and that these months are avoided by the oviparous species. This might be traceable to the difference in breeding method. Egg masses of *nigrolineata* and *arcana* are probably fairly susceptible to desiccation. The ovoviviparous method of reproduction probably protects the developing young from this.

2.5. CHANGE IN THE POPULATION STRUCTURE OF L.NEGLECTA OVER A YEAR.

The sources of major inaccuracy in random sampling of populations of arcana, rudis and nigrolineata, notably the interactions of habitat characteristics (eg. crevice number and size), animal size and weather conditions at time of sample, do not apply in the case of neglecta at Porth Swtan. The rock surface in its zonal range is so eroded as to be effectively crevice-less and barnacle shells, which provide the main protective shelter for neglecta, are no protection from sampling. Also no migration to zones outside the sample area has been detected in this species. L.neglecta is therefore unique amongst rough winkles in its availability for studies of population structure since sampling can be unbiassed in respect of sex, maturity or size, the only restriction to sampling being that individuals less than about 1mm were difficult to collect as their shells are so small and fragile.

Data from the transect 2 samples (see section 3.2. for details of collecting) were combined to give an overall picture for each month. Every individual of neglecta (over 2.0gu shell height, 1mm = 1.7gu) was measured and scored for sex and reproductive status. The overall changes in population size structure and frequency are plotted in fig. 43. For fig. 44 the sample was broken down into subsections, mature males, mature females, immature males, immature females and juveniles and the frequency and size structure for each subsection in

Figure 43.

L.neglecta: Population size structure changes for the year April 1977 to March 1978.

L.NEGLECTA: POPULATION SIZE STRUCTURE CHANGES FOR THE YEAR APRIL '77 TO MARCH '78

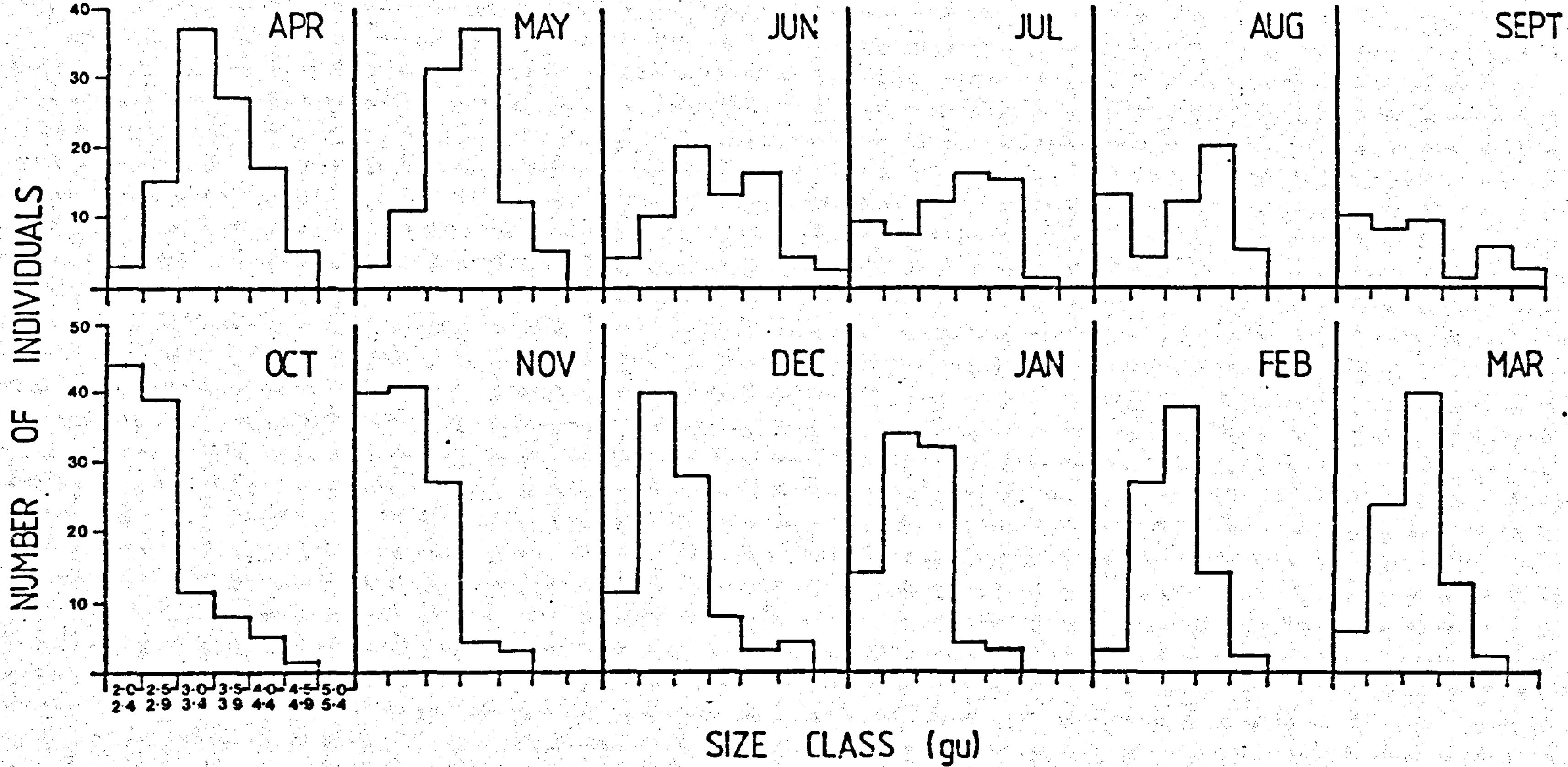
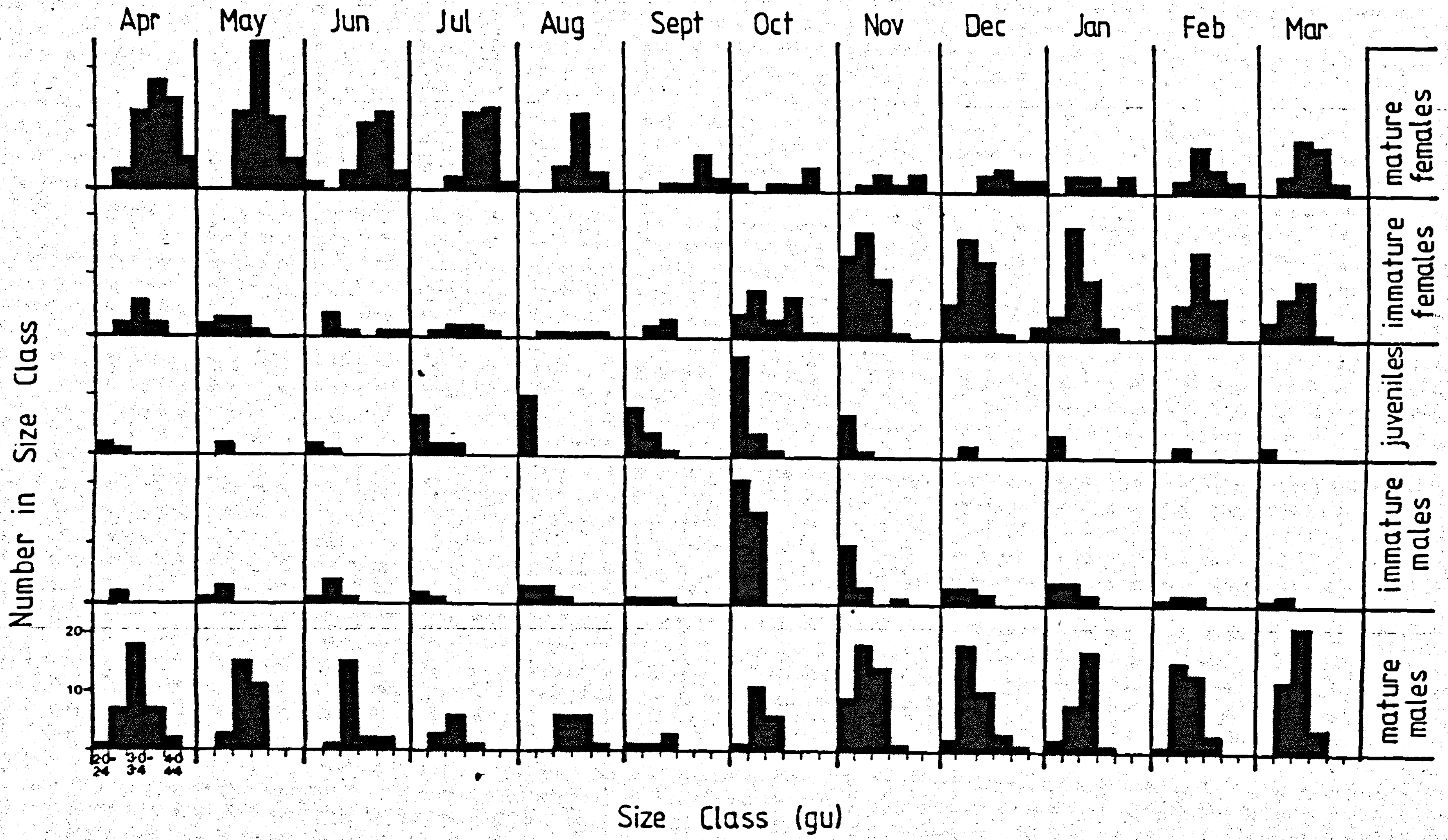


Figure 44.

Frequency and size structure changes in subsections of the *L. neglecta* population.


Apr
May
Jun
Jul
Aug
Sept
Oct
Nov
Dec
Jan
Feb
Mar
mature females
immature females
juveniles
immature males
mature males
Number in Size Class
20
10
20-24
30-34
40-44
Size Class (gu)

successive months were plotted. The data are given in table 10, Appendix B. The changes over the year in each subsection are outlined below.

2.5.1. Mature females.

Mature females were at their maximum frequency in May. In the following months not only did the frequency decline indicating that mortality exceeded recruitment but also the sub-population increased generally in shell height ie. increasingly high numbers were recorded in larger size classes compared to smaller size classes. Low levels of recruitment were apparent from October to January increasing in February and March.

2.5.2. Immature females.

Very few immature females were collected from April to September (inclusive). Recruitment to this sub-population was apparent from October to January when numbers in the smallest size classes were comparatively high; thereafter numbers declined as the animals attained reproductive maturity and were recruited to the mature female sub-population or alternatively were killed.

2.5.3. Juveniles

Few juveniles were collected from April to June (inclusive). From July to November recruitment is higher and this sub-population reaches a maximum frequency in October. From December to March few individuals were collected. The greater part of the juvenile sub-population was too small to collect, notably those below 2.0gu shell height, which would nearly exclusively be juvenile and so no changes in size structure were apparent.

2.5.4. Immature males.

Few immature males were collected from April to September and in February and March of the following year. Recruitment to this sub-population occurred principally from October to January, most especially in October, and thereafter frequency declined as the animals attained reproductive maturity or were killed. A slight change in size structure was apparent as animals, recruited in the autumn, grew larger.

2.5.5. Mature males.

Mature males were at a fairly high frequency in April. In the following months, through to September numbers declined, indicating that recruitment was less than mortality. Recruitment increased in October and reached a maximum in November and December when large numbers in the smaller size classes were recorded. In subsequent months frequency remained fairly high though there were changes in the size structure towards relatively more individuals in the larger size classes.

2.5.6. Overall features.

It is notable that recruitment from the juveniles to the immature male sub-population takes place earlier in the year than the equivalent recruitment to the immature female sub-population, and similarly to the mature male sub-population. This results in the main mass of males coming into reproductive condition before the females. This is probably an important aspect of reproductive strategy as presumably a male that matures earlier than the main body of the females will be well placed to copulate with any female that comes into reproductive condition earlier than average. It is probable that the attaining and maintaining of sexual maturity early by males is achieved at the cost of growth and this may be the basis of the sexual dimorphism observed in this species.

Since male neglecta mature at a smaller shell height than females and the reproductive organs become sufficiently differentiated to detect this sex at a significantly smaller shell height than in females, probably many of the juveniles recorded in October are in fact genotypically female.

Though the period of high production of young in neglecta can be expected to coincide with the period of high numbers of mature females, the data appear to indicate a lag between the period of maximum mature females and the period of high recruitment to the juvenile population. This is probably an artefact due to the non- recording of very small neglecta (those below 2.0gu).

The pattern of change in size structure, frequency and composition of the neglecta population (see figs.43 & 44) strongly suggests that this species is an annual, individuals leaving the brood pouch, reaching maturity, reproducing and dying or being killed within the space of a year. The majority of the population seems to be at approximately the same stage of the life cycle at any one time, and this is therefore the basis of the cycle of reproductive activity recorded in section 2.4.. The other rough winkle species are thought to be perennial and the cycle of activity in these will be in part due to individuals' cycle of activity.

SECTION 3: ZONATION AND NICHE SEPARATION OF ROUGH WINKLE SPECIES

The winkles of British shores have long been held up as examples of how closely-related species on the beach show vertical zonation differences, this fitting neatly with the ecological theory of competitive exclusion. Observations of the distribution of rough winkle species at various localities suggested to me that vertical zonation differences were not in fact so well-defined and two instances of major zonational overlap were frequently seen. Firstly, at the majority of sites where rudis and arcana occurred they appeared to be truly sympatric; not only was the vertical zonation apparently similar, but also no evidence of habitat specialisation could be detected. i.e. one species was not associated in particular with, for example, rock pools, aspect or a species of alga. Secondly, neglecta populations were not inhabiting the microcrevices created by barnacles (both dead and alive) by themselves; alongside them, and often outnumbering them, were juveniles of other species, most frequently nigrolineata.

The following section explores aspects of niche separation at one site, Porth Swtan, where a more detailed study of the inter-relationships was made. It was hoped that by concentrating on one community at least some of the mechanisms of niche separation might be understood.

3.1. GENERAL SITE DESCRIPTION OF PORTH SWTAN.

After visiting a number of sites Porth Swtan was chosen as a suitable locality for the studies proposed.

Porth Swtan, Grid Ref. SH 299892, is on the north-west coast of Anglesey. It is a wide sandy bay facing west, with rocky shores north and south. A particular rock (see Plate 31) in the southern rocky area was selected as the study site. It is also the type locality of L.arcana.

The rock is large, about 4m at the highest point and 15m square at the base. It is approximately wedge-shaped. The landward wall faces east and is nearly vertical, the side walls slope steeply and the seaward side slopes gently down to a barnacle-encrusted rock platform which extends some way down the beach. The majority of the upper part of the rock lies in the Pelvetia and Verrucaria zones though a small pinnacle, the highest part of the rock's surface, bears yellow lichen. The populations of rudis C and arcana which occupy these zones are isolated from similar populations in crevices on nearby rock masses by both the wide barnacle belt, which clothes the base of the rock, and by broad belts of unstable large pebbles on the landward and northern sides.

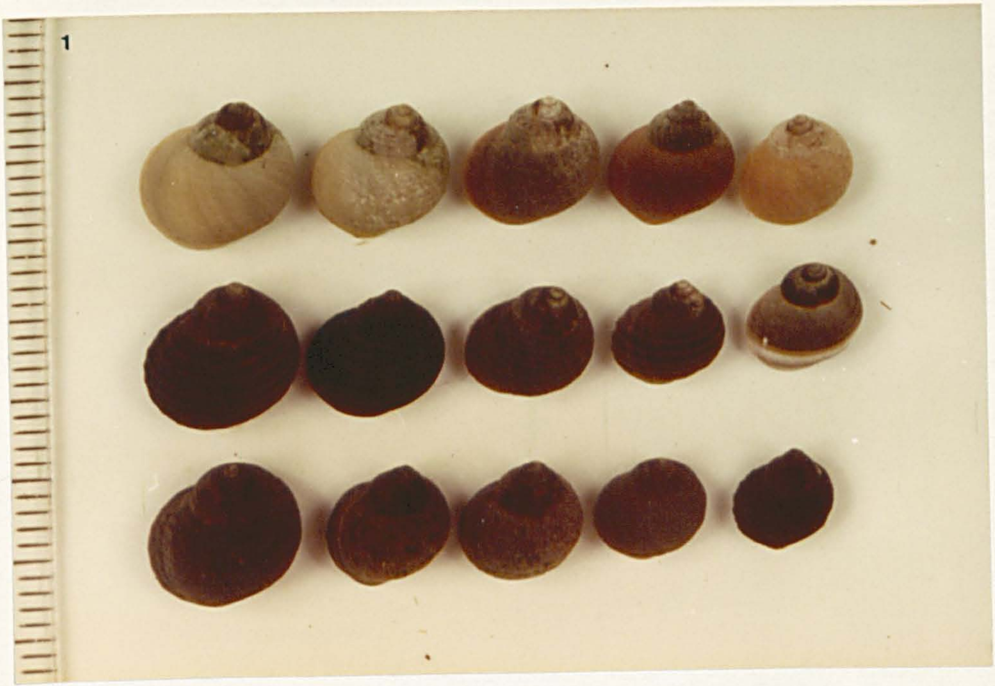
The rock combines several other unusual and useful features which particularly fitted it as the main study site of the interactions of species of the

PLATE 31.

1). Shells from Transect 1 samples, top two rows <u>L.arcana</u>, bottom row <u>L.rudis</u> C.

2). Porth Swtan, transects were sited on the rock mass centre, middle distance.

1



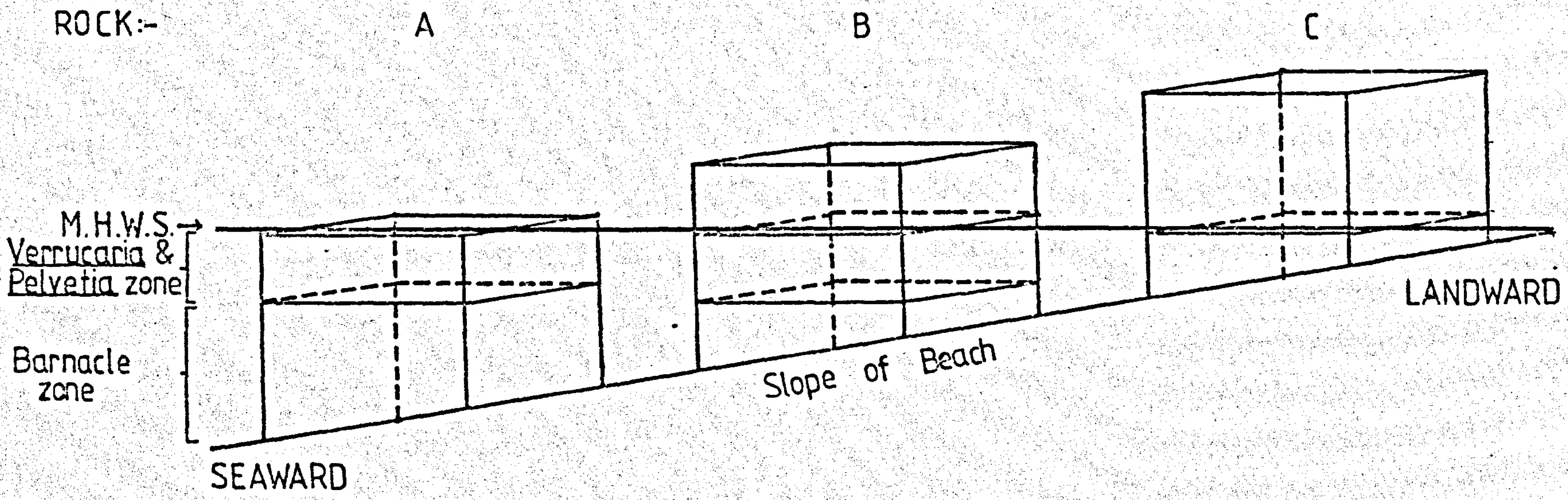
2

rough winkle community. The combination of its position low down the shore, with its height and approximately flat-topped shape determines not only that most of its surface is submerged at high tide, but also that a peculiarly large surface area lies in the *Verrucaria/ Pelvetia* zone (see fig. 45). Correspondingly this rock supports an unusually large population of *rudis* C and *arcana* which was a pre-requisite for a long-term study involving removal of large samples at regular intervals. Other useful features are 1). all four species of rough winkle were common on the rock, 2). the crevices were comparatively shallow, so that the winkles were fairly easy to collect, 3). the landward wall of the rock, being nearly vertical (see Plate 32), provided a good site for work on zonation in the barnacle belt.

Figure 45.

Effect of a rock's position on surface area in major ecological zones.

EFFECT OF A ROCK'S POSITION ON SURFACE AREA IN MAJOR ECOLOGICAL ZONES



Relative surface area in *Pelvetia*/*Verrucaria* zone: A:B:C = 21 : 12 : 9·5

Relative surface area in Barnacle zone : A:B:C = 21·5 : 9·5 : 0

PLATE 32.

1). Porth Swtan; foreground, unstable boulder/pebble habitat of L.rudis B; middle distance, stable rock mass, habitat of L.rudis C.

2). Sampling transect 2.

1

2

3.2. ASPECTS OF NICHE SEPARATION OF L.ARCANA AND L.RUDIS

Attempts to find a definable difference in spatial habitat between rudis and arcana were unsuccessful. No generally applicable pattern emerged from the study of a range of sites, though at a few, tantalising sites some pattern was seen, e.g. at Abraham's Bosom where a rock ridge provided two distinct habitats (one side facing N.W.& nearly sheer, the other facing S.E. & stepped), one supported an almost pure population of arcana and the other almost pure rudis (respectively); also at various sites some vertical zonation differences seemed to exist but there was no consistency in the relative zonations of the species. At the majority of sites the populations of rudis and arcana seemed to be intermingled and truly sympatric. Thus the mechanism of niche separation of these species seems to be more subtle than the expected adaptation to different exposure levels and might even differ from site to site, as the observation of habitat differences can be made at some sites (eg. Abraham's Bosom) but not at others (eg. Porth Swtan).

My efforts were concentrated on understanding the mechanism(s) of niche separation of the rudis C and arcana of the study population at Porth Swtan. The possibility of seasonal differences in vertical zonation patterns was the principal aim of the study since initial (1976-1977) work, aimed primarily at defining the new species, arcana and studying reproductive periodicity, had turned up evidence of possible migratory activity in arcana related to its reproductive activity. A fixed transect (number 1) was established and visited monthly in order to follow the long-term movements of the rudis and arcana populations.

3.2.1. Transect 1:- Siting and sampling

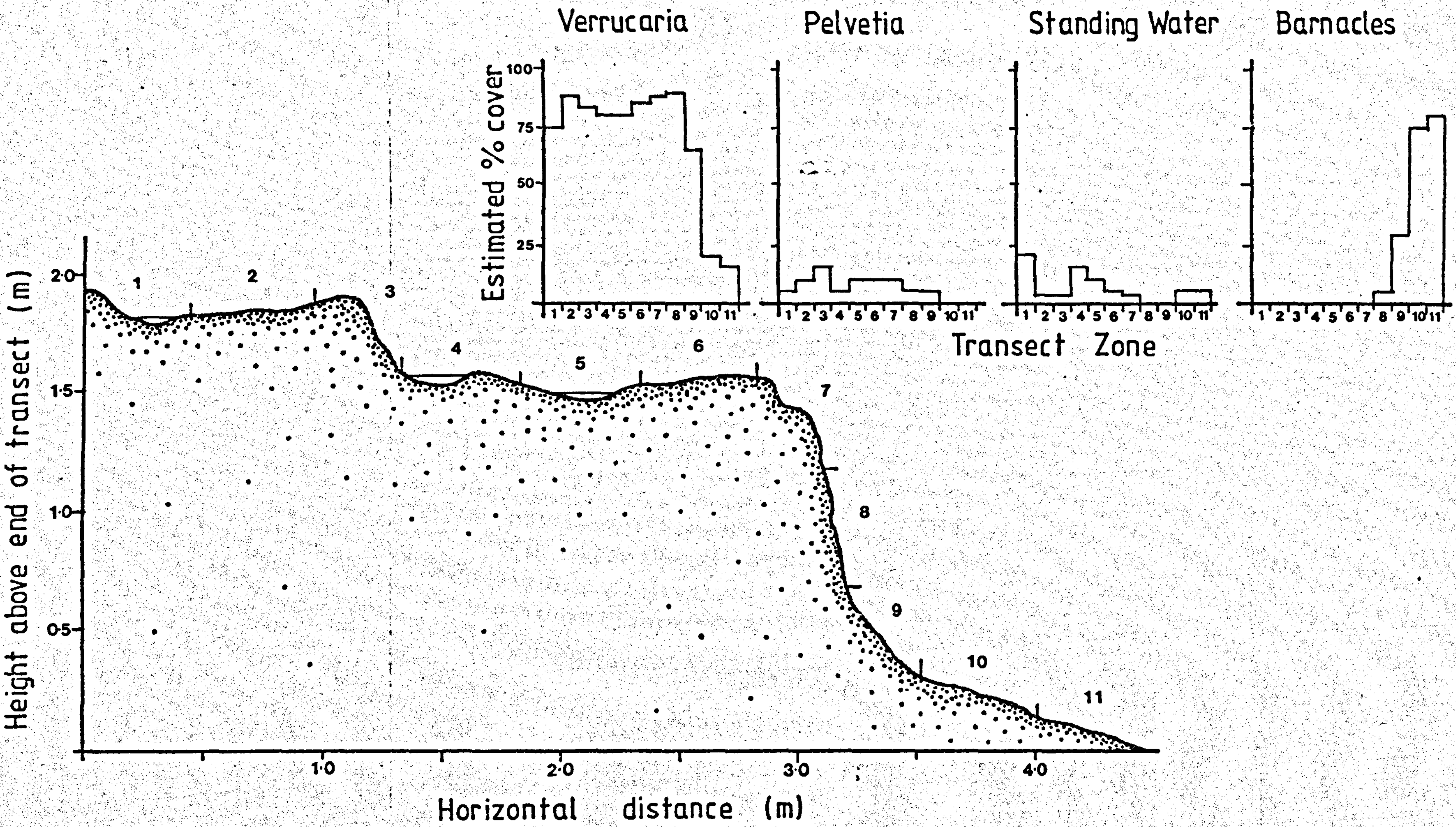
Transect 1 was sited for sampling the winkle population in the Verrucaria, Pelvetia and upper barnacle zones. The Pelvetia did not form a distinct belt. Its distribution was patchy, the plants usually occurring on the damper parts of the rock though not actually in the small permanent rock pools.

A string marked at 50cm intervals, defining zones of the transect, was laid between fixed points on the rock in a straight line, at right angles to the top of the barnacle belt line. The transect zones were numbered successively. In all, there were eleven zones; zone 1 was at the top of the Verrucaria belt and zone 11 in the barnacle belt. Winkles larger than ≃6.0mm shell height within a zone up to 25cm on either side of the guide line were collected using forceps and put into appropriately numbered plastic bottles. The profile of the transect along the guide line is shown in fig. 46, with the estimated % cover of Pelvetia, Verrucaria, standing water and barnacles for each zone.

The transect was sampled at approximately four week intervals. The considerable and unavoidable variation in topography in each zone meant that the sur-

Figure 46.

Profile of Transect 1.


Verrucaria
Pelvetia
Standing Water
Barnacles
Estimated % cover
100
75
50
25
1 2 3 4 5 6 7 8 9 10 11
Transect Zone
Height above end of transect (m)
2·0
1·5
1·0
0·5
1
2
3
4
5
6
7
8
9
10
11
1·0
2·0
3·0
4·0
Horizontal distance (m)

face area sampled in each zone was not equal. Analysis of population structure and density was therefore not possible. Furthermore the height of each zone above chart datum was not evenly sequential (compare with transect 2). Between visits large winkles in the transect were replenished by recolonisation (in the short term) or growth of the remaining smaller winkles (in the long term). However, it was noticeable in the final two months of collection that the numbers of large winkles appeared to be dropping, probably as a result of the depletion, through prolonged sampling, of the population.

3.2.2. Zonational patterns of L.rudis C and L.arcana along transect 1.

The analyses below examine the zonational patterns of mature and immature, male and female animals of rudis C and arcana along the transect over the period March 1977 to February 1978. Since size and maturity are related and the sampling cannot be free of size bias (see section 2.4.1.), the analyses concentrate on animals of a particular size range, 7.0-8.95mm shell height for both species. Records of all specimens collected in the transect, shell dimensions, reproductive status, etc. were made and the data obtained were used in various other analyses.

Results:-

3.2.2.1. Overall zonation patterns

By combining the data collected over the year for each sub-population (male or female, mature or immature in the size range 7.0-8.95mm shell height) an overall pattern of the zonation of each sub-population of rudis and arcana can be obtained. The data are given in table 1, Appendix C and are plotted in fig. 47.

a) L.rudis C

Both mature and immature animals of both sexes generally decline in numbers down the transect. The decline in mature males seems more marked than in mature females for zones 1 - 7.

b) L.arcana

In both sexes, mature animals zone much lower than immature animals. Also mature females seem to zone generally slightly lower than males.

Discussion

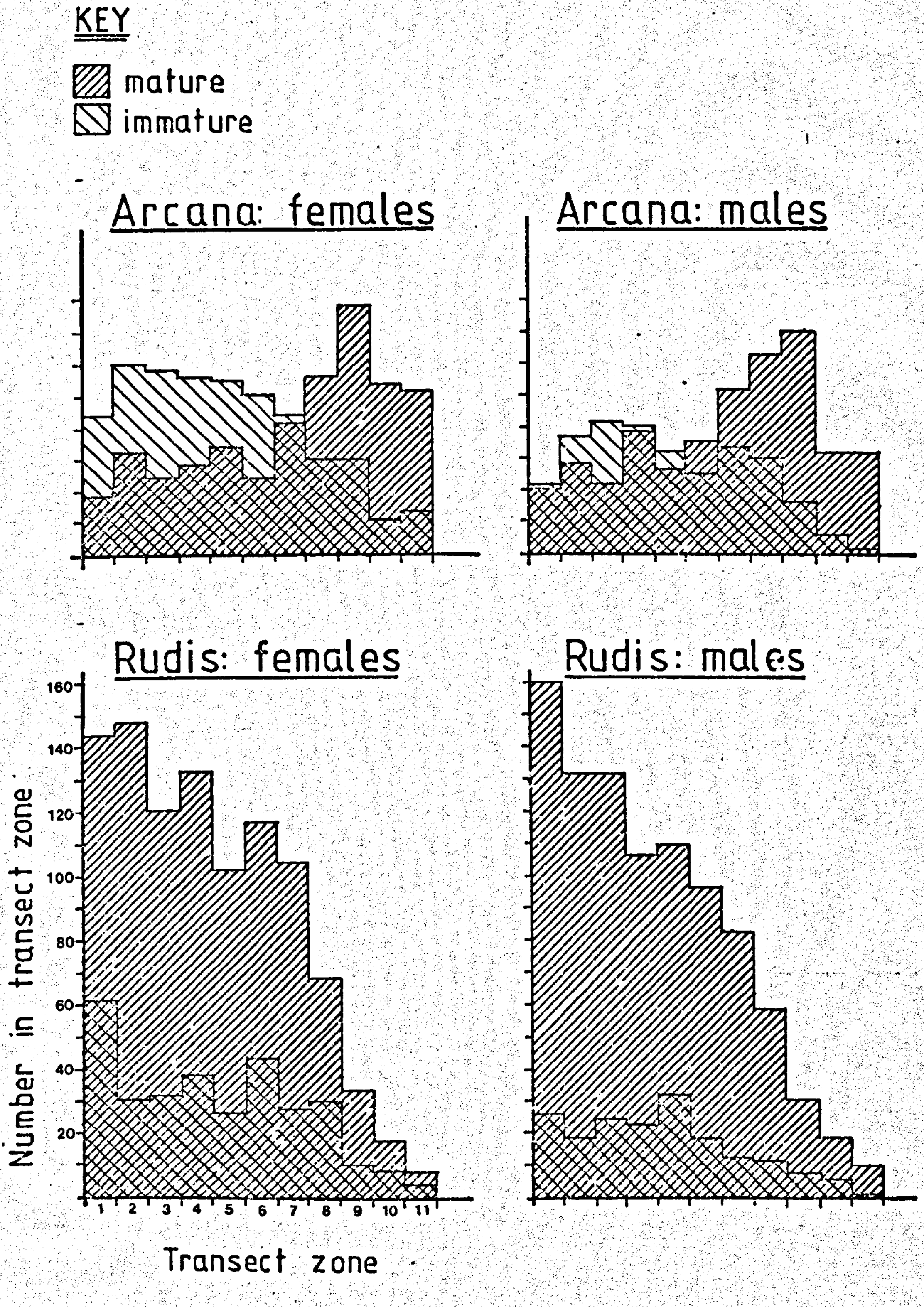
The numbers collected in successive transect zones in individual months tended to be comparatively low and were erratic in their distribution. Combining the data for the year shows more clearly the zoning patterns for the species sub-populations.

The overall zonation pattern of rudis C indicates that this is an ecotype of the Pelvetia/Verrucaria zone, comparatively few being collected from the barnacle belt. No distinctive difference in zonation of mature and immature animals is apparent. In contrast the mature and immature arcana each prefer quite distinct sections of the transect, immature animals being commonest in the upper transect zones and mature animals in the lower zones (concentrating just above and in the top of the barnacle belt). Since mature and immature

Figure 47.

Average zonation patterns for mature and immature, male and female subpopulations of *L.rudis* C and *L.arcana*.

AVERAGE ZONATION PATTERNS



animals are similar in all respects except their reproductive state it seems reasonable to suggest that migration related to the attainment of maturity occurs.

In both species the data suggest that mature males zone slightly above mature females. A similar situation is seen in neglecta (section 3.3.). This zonational difference between the sexes may be related to the difference in costs and benefits of living in different zones which might vary with the sexes, e.g. females may gain a reproductive advantage if their young are released into the lower zones, if by this survival of the young is increased.

3.2.2.2. Monthly zonation patterns.

The monthly pattern of zonation of each sub-population of each species was also examined. The data appear in table 2, Appendix C and are plotted in figs. 48 - 51.

a) L.rudis C sub-populations

The zonation patterns are plotted in figs. 48 & 49. The variation in the pattern of zonation for each sub-population seems to be fairly erratic and in view of the numbers collected from each zone, there is no meaningful deviation from the average pattern of zonation.

b) L.arcana sub-populations

The zonation patterns are plotted in figs. 50 & 51. Clear changes in the patterns can be seen. In March, April and May mature females are fairly common and congregate in the lower zones of the transect. Immature

Figure 48.

Monthly zonation patterns: *L. rudis* C females

MONTHLY ZONATION PATTERNS: RUDIS FEMALES

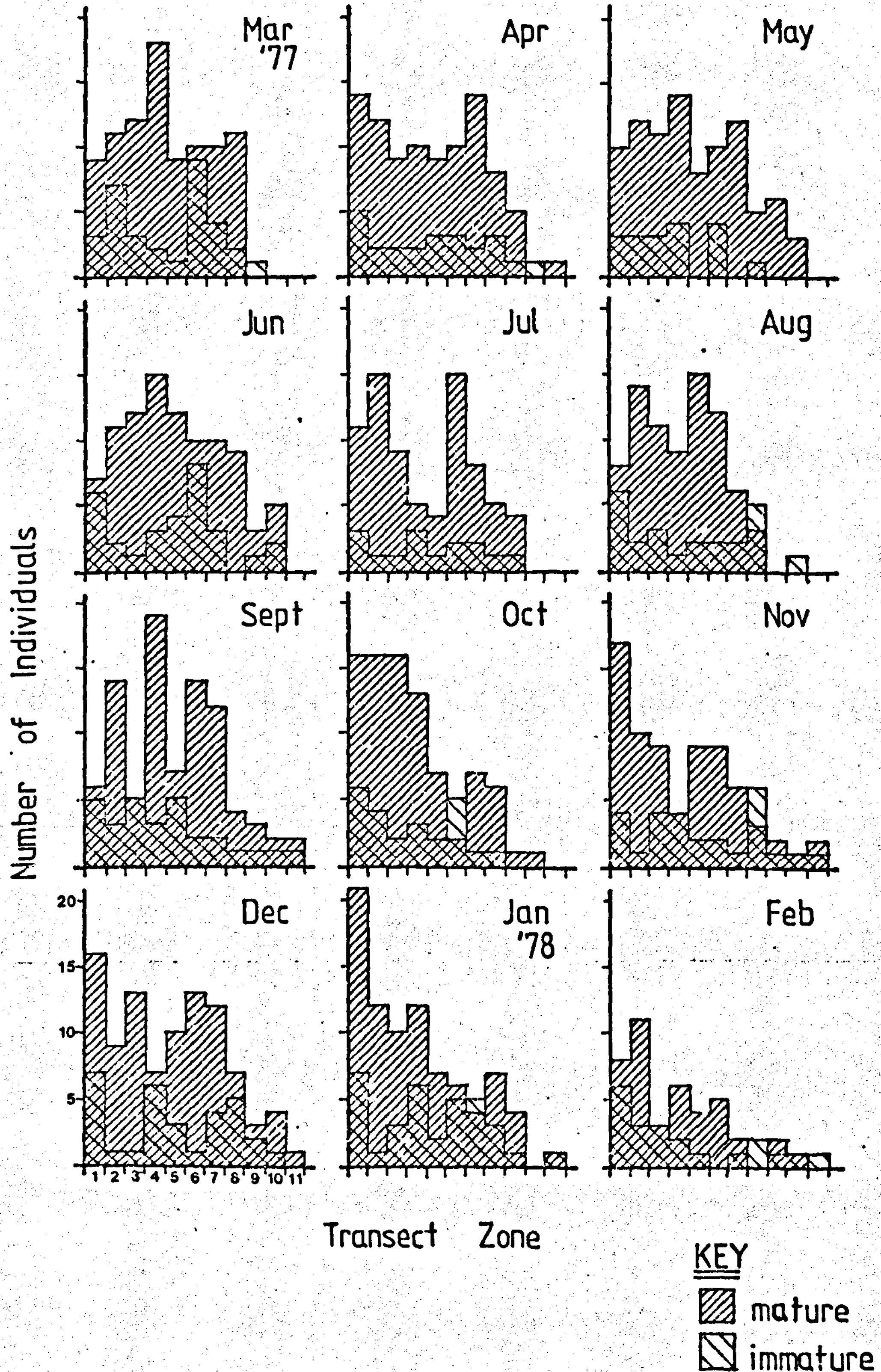


Figure 49.

Monthly zonation patterns: *L.rudis* C males

MONTHLY ZONATION PATTERNS
RUDIS MALES

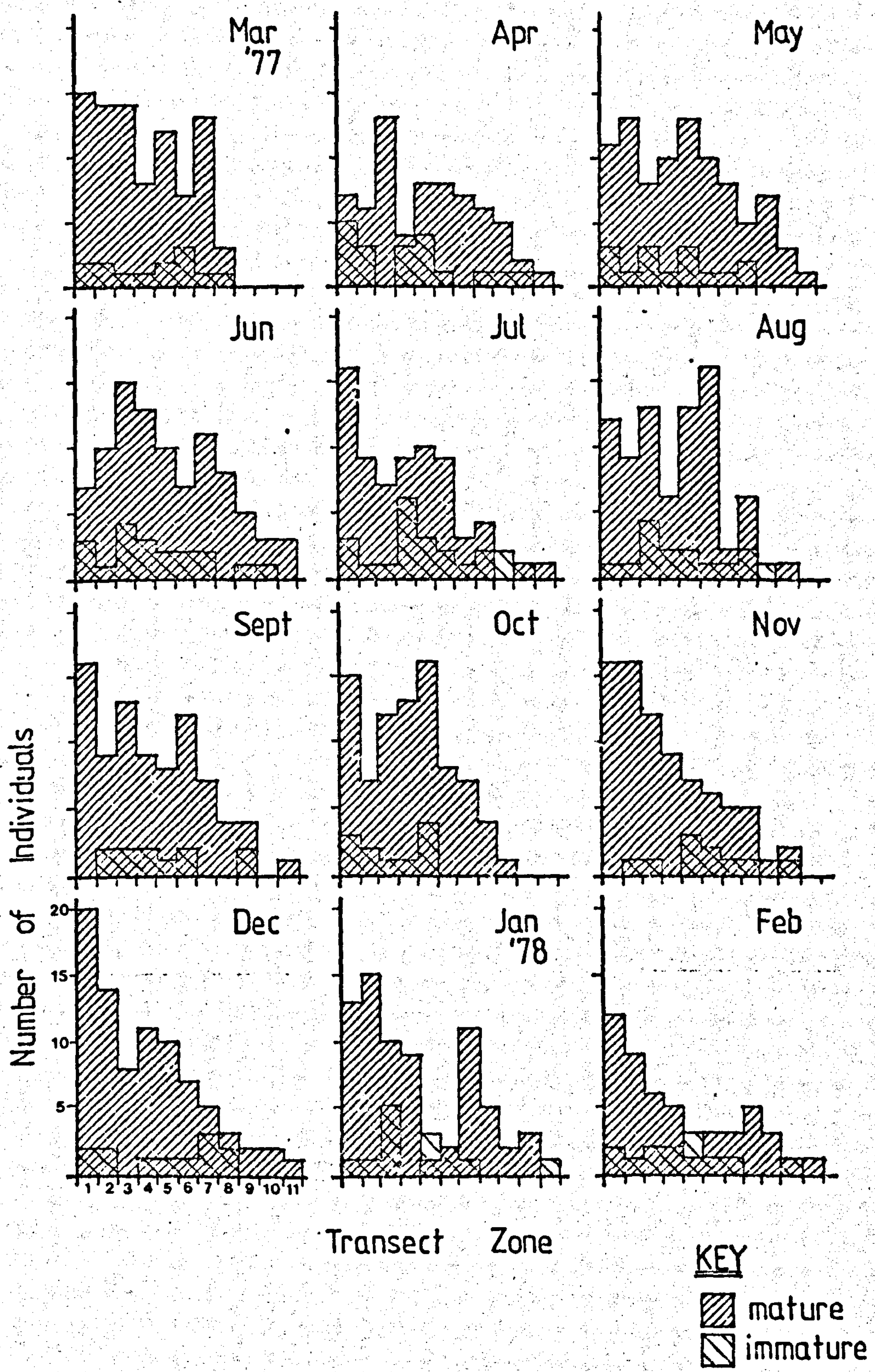


Figure 50.

Monthly zonation patterns: *L. arcana* females.

MONTHLY ZONATION PATTERNS: ARCANA FEMALES

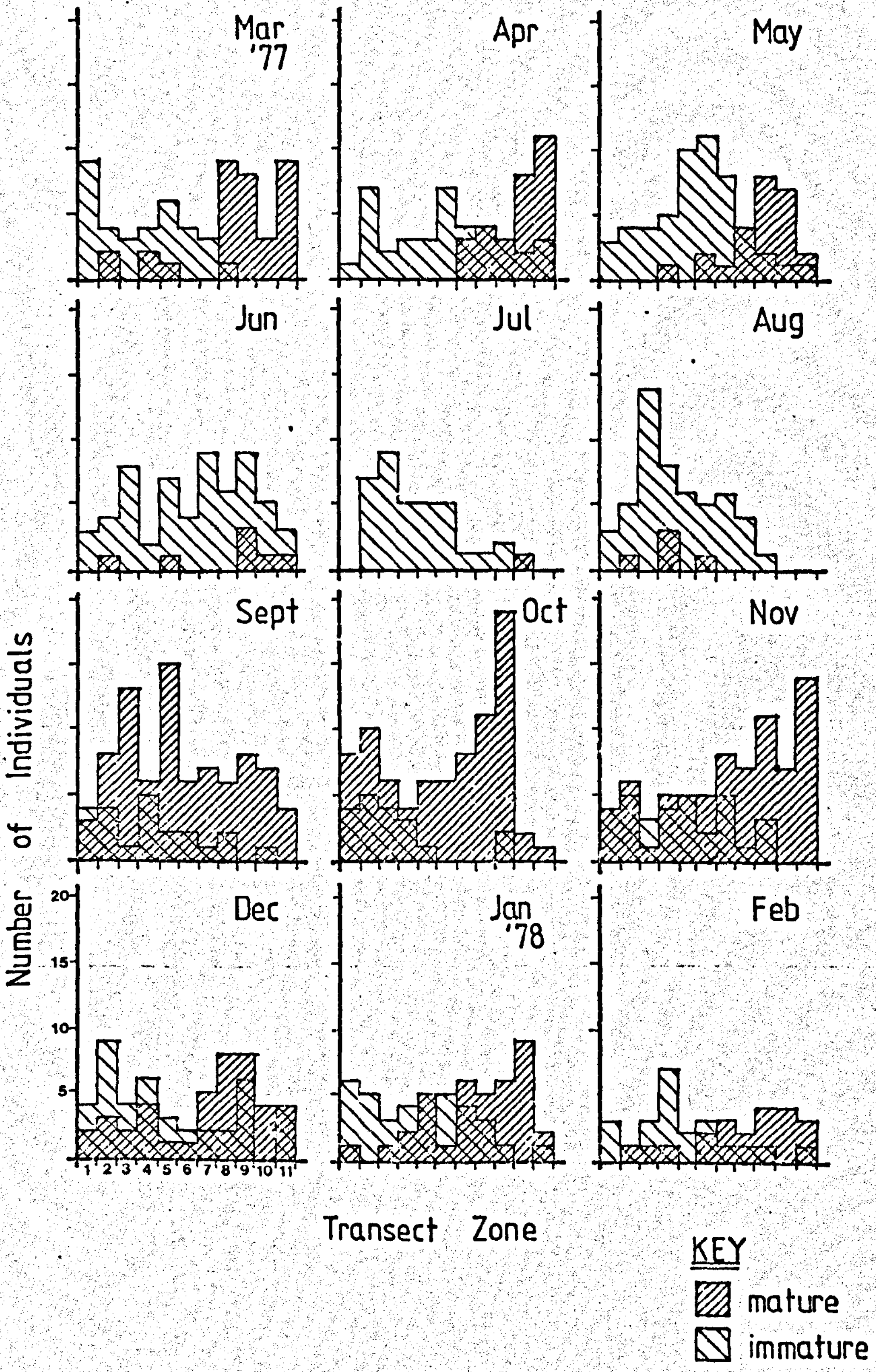
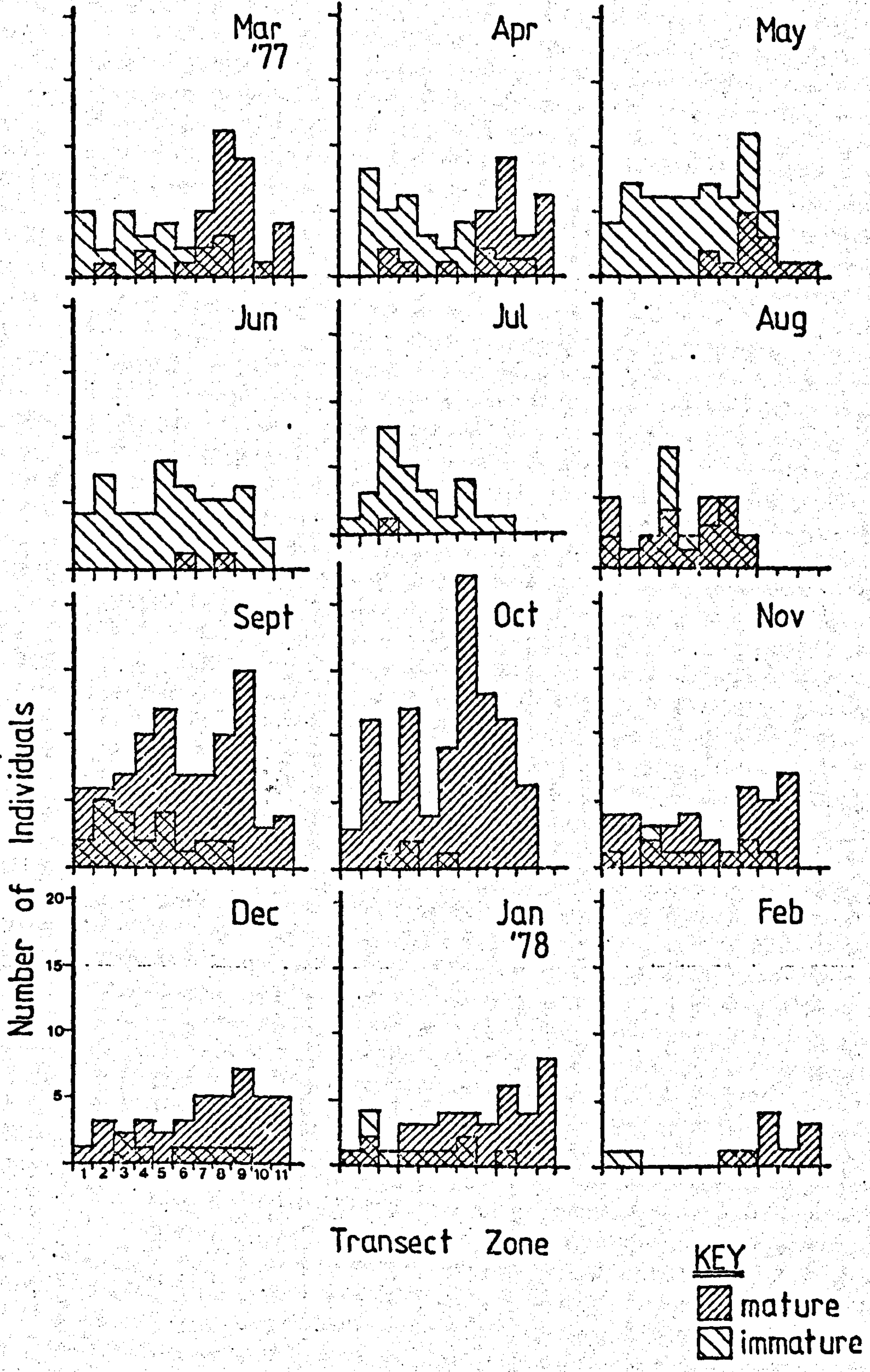


Figure 51.

Monthly zonation patterns: L.arcana males

MONTHLY ZONATION PATTERNS: ARCANA MALES



females, in contrast, are congregated in the middle and upper zones.

In June mature females are relatively fewer and immature females are spread throughout the transect.

In July and August mature females are very scarce, and the immature females are congregated in the upper and middle transect zones.

In September there is a marked increase in numbers of mature females accompanied by a decline in immature female numbers and the mature females are spread throughout the transect.

In October mature females are abundant and their zonation pattern has changed towards higher numbers lower down the transect.

From November '77 to February '78 mature females are commoner in the lower zones, their numbers declining in successive months. Immature females are commoner in upper transect zones. This pattern is similar to the months of March to May '77.

The changes in the zonation patterns of the male sub-populations are similar to those observed for females.

Discussion

In rudis C there is no indication of vertical seasonal movement. In arcana, however, a clear pattern of vertical movement associated with reproductive activity is apparent.

Mature arcana females congregate in the

lower, and presumably damper, zones of the transect where their egg masses will be less exposed to the risk of desiccation. Immature females seem to prefer the higher zones.

In June the numbers of mature females show a marked decline. Mortality may play a part in this but probably more significant is the regression of the reproductive organs when animals that were reproductively active enter a phase of reproductive inactivity. Possibly as a result of this, immature animals (which includes both pre-reproductives and animals out-of-season) appear to be distributed throughout the transect. In July and August probably as a result of upward migration the familiar pattern of immature animals preferring upper zones is re-established; zones 10 and 11 are devoid of arcana. In September many animals regain or attain reproductive maturity and these are spread throughout the transect; as downward migration begins, zones 10 and 11 are re-occupied. In October and November the downward migration of mature females is more apparent and by December the majority of mature females are again collected from the lower zones of the transect.

The changes in zonation of male arcana follow a similar pattern to the females, though they seem to mature slightly earlier. However, in this instance, this could be due to sexual dimorphism; males in the size range 7.0-8.95mm are probably somewhat older than females of a similar size. It has been shown (section 2.4.1.) that older animals have a longer breeding season than younger ones.

3.2.2.3. Relative zonation of L.arcana and L.rudis.

The data obtained show that at Porth Swtan the zonal range of the two species is very similar; both species occur in all the zones of the transect. This was typical of the intermingling of the two species seen at many sites. However by the detailed examination of the zonation at Porth Swtan over the year a vertical zonation difference has been shown to exist, notably that during its breeding season mature arcana individuals migrate down the transect and occupy the lower levels. This zonational difference disappears when arcana is no longer breeding. This may account for observations of specific zonational differences, noted at some sites, since the observations were made at different stages of the arcana breeding cycle. The reason for the migration is probably traceable to the method of reproduction. The risk of desiccation to the egg masses is probably reduced by laying them lower down the rock where they will be exposed for a shorter period. However, this reproduction-related migration is unlikely to be the only niche separation mechanism operating and further research is needed in this field. One possible line of study would be examination of arcana-rudis communities for further zonational differences which may exist between the juveniles of the species.

PLATE 33.

1). & 2). Mixed L.rudis and L.arcana in zono 2 of transect 1.

1

2

3.3. ASPECTS OF NICHE SEPARATION OF L.NEGLECTA

The identification of the barnacle belt as the habitat zone of L.neglecta was made by Heller in his paper (1975a) in which he also raised this form to specific rank. Before this date, no reliable ecological information is available as the species cannot be fully correlated to the subspecies L.saxatilis neglecta as used by James (1968a) and previous authors. Raffaelli (1976) confirmed the barnacle belt habitat for this species at Llanddwyn Island.

A pilot study at Porth Swtan indicated that the barnacle belt was the habitat not only of neglecta but also of substantial numbers of juveniles of other winkle species. Thus it appeared that neglecta was co-existing with closely-related and ecologically similar species. The principle of competitive exclusion predicts that no two species can occupy permanently the same ecological niche. From this it can be suggested that there should be some part of the niche,e.g. temporal or spatial, unique to neglecta which might be revealed on closer examination. Defining this area of non-overlap with other species i.e. defining that part of the niche which was unique to neglecta, was therefore of particular interest.

The possibility of food specialisation was considered. However it seemed unlikely that the spatial overlap apparently observed could be explained thus. There is no direct evidence for or against selectivity of food items by any of the rough winkle species, but it can be inferred from the indirect evidence of the similarity of the shape of the radula cusps of neglecta and of juveniles of other species (see section 1.6.) that little food specialisation occurs (though the possibility of differential digestion remains). Also the change in shape of cusps in nigrolineata, from the neglecta-like juvenile form to the flat-edged adult form can be interpretted as positively indicating that juveniles are adapted to taking a similar food to neglecta.

The other possibilities of resource partitioning considered included differences in specific zonation within the barnacle belt, size zonation effects and dovetailing of the population size-structure changes. The investigation was concentrated at the Porth Swtan site and for a year an intensive study of the relationships of the barnacle belt small winkle population, along a transect (transect 2) was made.

3.3.1. Transect 2: Siting and sampling techniques

The nearly smooth, vertical, landward wall of the rock mass, on which transect 1 was also sited, provided a naturally suitable site for transect work on the barnacle belt. Since the surface of the rock was worn comparatively smooth throughout the depth of the barnacle belt and for a little way above, there were no inaccessible crevices into which winkles might retreat. This made unbiassed collecting possible. Also since the surface was nearly vertical the transect sequence was equivalent to an exposure gradient. If a site had been chosen on the shallower gradient of the seaward side the hummocks and pools would have resulted in mixed ecological areas in a single transect zone, or alternatively a non-continuous series of samples would have been necessary.

The line of the transect was fixed and divided into eight zones running successively from zone 1 (above the barnacle belt) to zone 8 at the foot of the rock. Each zone was 20cm vertically by 60cm horizontally. Since the rock surface was nearly smooth, the area sampled in each transect zone was roughly equal (unlike transect 1). The transect site rated about 5 on Ballantine's exposure scale.

The percentage cover of barnacles in each zone of the transect was obtained by using a point quadrat technique on photographs of a 20cm square portion of the transect zone. A grid was laid over the photographic enlargements and random number tables used to fix the co-ordinates of 100 points. Presence or absence of a barnacle at each point was recorded to give an estimate of percentage cover, and the results are plotted in fig. 52 (see table 3, Appendix C for the data).

Once a month all the small winkles in each zone were collected with fine forceps and placed in appropriately numbered tubes. Samples were taken to the laboratory and killed by brief immersion in boiling water.

Individuals were identified and shell length was measured in graticule units with an eyepiece graticle and binocular microscope. Specimens smaller than 2.0gu (=1.18mm, which were too small to collect easily) and larger than 6.4gu (=3.76mm, which were thought not to be in direct competition with neglecta) were discarded from the analyses. Information on the sex and reproductive state of neglecta individuals was also recorded.

Juveniles of all species (except mariae which was absent) were collected as well as neglecta. Individuals were identified to species where possible, using the characteristics described in section 1.7. Separation of arcana and rudis was largely impossible for juvenile individuals at this site because of the similarity of shell colour. However, within the rudis/arcana category fragile, ridged, black shells stood out from the rest which ranged in colour, white, yellow, fawn, brown, grey and intermediate colours being represented. Juveniles of the rudis/arcana types were kept in aquaria until they had grown sufficiently for the species to be identified. Twelve of the black shell type grew and all proved to be rudis. Of the individuals with other-than-black shells, 48 grew, of which three were arcana and the rest rudis. On the basis of shell colour it seemed likely the black-shelled juvenile rudis were progeny of the similarly coloured rudis C population, common in the zones above the transect, and the other-than-black shelled juvenile rudis were thought to be the progeny of the rudis B population, common on the pebble habitat at the foot of the rock. Since arcana juveniles and rudis juveniles with other-than-black shells were indistinguishable they are treated as one type in the following analyses and as the majority are likely to be rudis B juveniles (the aquarium experiment suggested that

Figure 52.

Percentage cover of barnacles along transect 2.

PERCENTAGE COVER OF BARNACLES IN TRANSECT 2

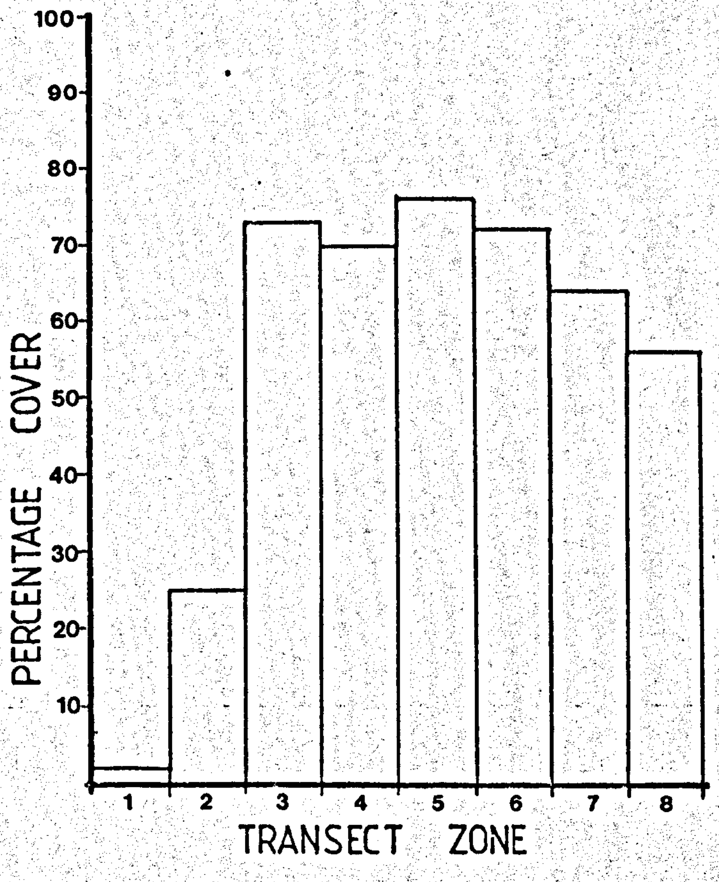
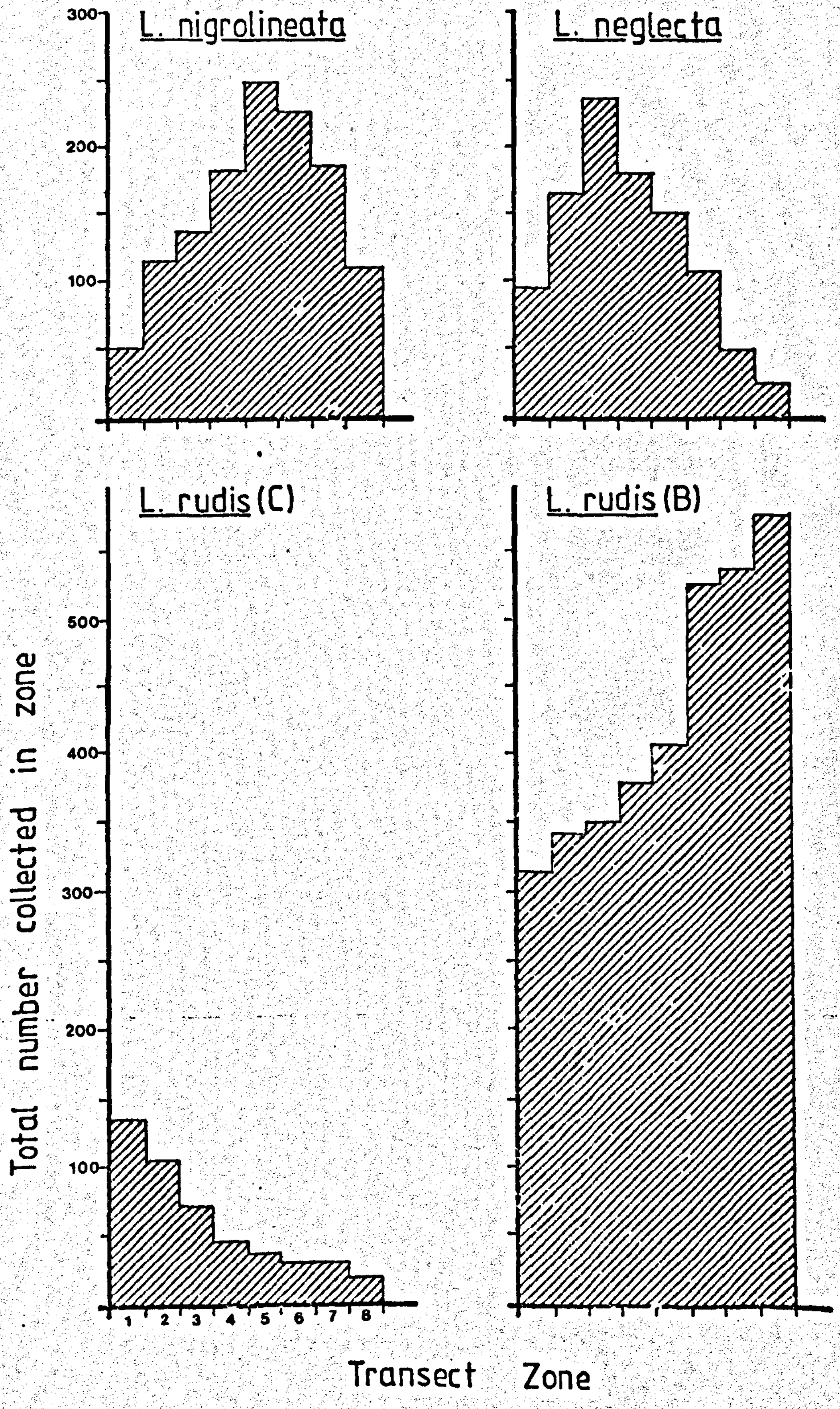


Figure 53.

Average zonation of species along transect 2.

SPECIFIC ZONATION PATTERNS
ALONG TRANSECT 2



probably only 6.25% were arcana) they are for convenience given this name.

3.3.2. Zonation of species within the barnacle belt

Initial observations on the winkle population of the barnacle belt suggested that juveniles of many winkle species (as well as juvenile and adult neglecta) were common in the barnacle belt, indeed this habitat seemed to be a winkle 'nursery' ground. The data obtained in transect 2 were examined to see if there was zoning of the species within the belt, and to see if neglecta occupied a distinct spatial habitat.

Results

By combining the data obtained from transect 2 (see table 4, Appendix C) over the year April '77 to March '78 the average zonation pattern for each species can be more easily appreciated (fig. 53).

Discussion

Comparison of figs. 52 and 53 shows that neglecta are congregated above, and in the upper part of, the barnacle belt. Juvenile nigrolineata show a peak of frequency below neglecta and well within the barnacle belt. The two groups of rudis juveniles, split on the basis of shell colour, most interestingly show very different zonation patterns. Those with black shells are commonest above the barnacle belt and the other group are commonest in the lowest zones of the transect. The difference in zonation pattern seems to confirm the suggested difference in origin, black-shelled juveniles invading the transect from above and other coloured juveniles from below. It is probable that the inescapable error resulting from the mixing of arcana juveniles with non-black rudis juveniles will differentially affect the results, the higher transect zones (1-4) probably having a higher percentage of arcana than the lower zones (5-8) as, like rudis C juveniles, their adults live in zones above the transect. However if allowance is made for a probably differential arcana-error the preference of rudis B juveniles for the lower zones would only be accentuated.

Overall, it seems that the barnacle belt, which has often been regarded as a distinct and uniform ecological zone from a winkle viewpoint, is neither distinct (neglecta, for example, are as common in zone 2, above the belt as in zone 4, within it) nor uniform, as some differential distribution is seen. The zonation of neglecta, however, is not as a distinct band sandwiched between other species of winkle; neglecta, even in the zone in which it is most common, is outnumbered 2 to 1 by other species juveniles. Thus it seems that spatial zonation difference cannot be the only mechanism of niche separation.

3.3.3. Dovetailing of population size structure changes

Size is an important element in competition for food and space resources. Smaller winkles, by virtue of their size can exploit the shelter and food resources of tiny crevices which are inaccessible to larger winkles. However, though this may reduce intra- and inter-specific competition between winkles of different sizes there will still be the potential for competition between winkles of similar sizes as they are probably exploiting similar resources. Rough winkles of all species begin an independent existence at approximately the same size (circa 0.6mm shell height ± 0.8gu). Hence competition between neonates can be expected for all species and neglecta juveniles have no period when they are smaller than other species in which they might have access to resources unavailable to other species.

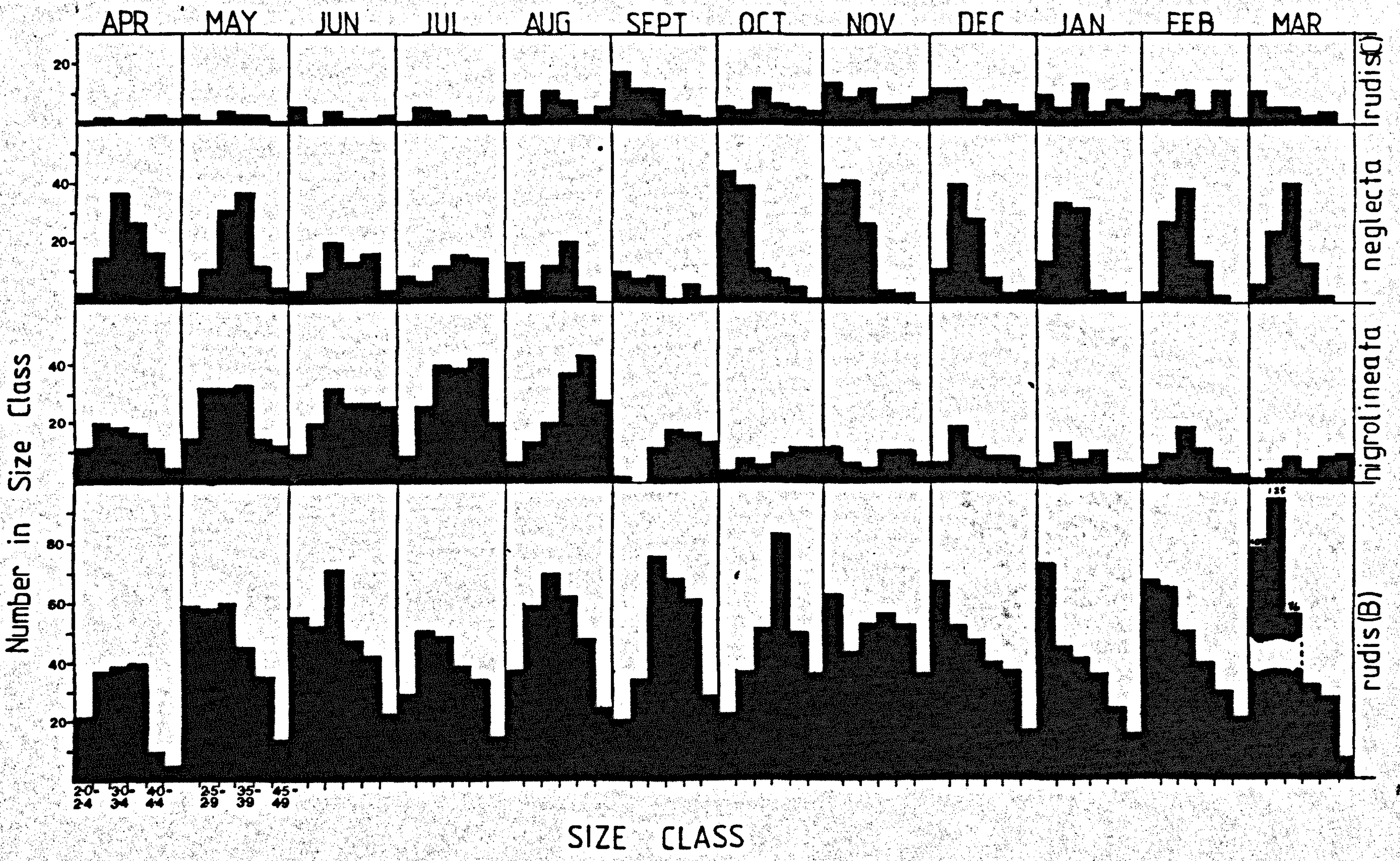
One way in which interspecific competition between winkles of the same size in the barnacle belt might be reduced is through the staggering of the breeding season, and it has been noted previously (section 2.4.) that neglecta and nigrolineata do have seasonal, and different, breeding periods; rudis populations at Porth Swtan however do not appear to be markedly seasonal. Such a staggering of breeding season might be expected to result in reduction of juvenile interspecific competition, juveniles of one species growing larger before juveniles of another appear on the beach. The population size structures of the main types of small winkle in the barnacle belt were examined to see if any dovetailing occurred.

Results

Figure 54 shows the monthly size structure of the populations of the four main types of small winkle

Figure 54.

Population size structure changes of small winkles in the barnacle belt.


APR
MAY
JUN
JUL
AUG
SEPT
OCT
NOV
DEC
JAN
FEB
MAR
rudis(C)
neglecta
nigrolineata
rudis (B)
Number in Size Class
20
40
20
40
20
20
40
60
80
135
16
20-24
30-34
40-44
25-29
35-39
45-49
SIZE CLASS

over the year April '77 to March '78. The data are given in table 5, Appendix C.

The population size structure of rudis C does not show any clear changes or dovetailing with the other species in the barnacle belt. However it is probable that the main bulk of the juveniles of this ecotype occur in zones above the transect and so are not sampled. Also the population is comparatively small, so it is probably not very important as a competitor to neglecta.

The neglecta population shows a marked change in size structure with an influx into the smallest recorded size classes in the months of October and November. It is possible to follow this age class as it grows larger through subsequent months.

The juveniles nigrolineata population also exhibits a change in size structure increasing in numbers and size from April to August, the numbers falling off afterwards. The age class responsible for modal numbers in sequential size classes can be followed through the months until August when they presumably grow out of the size range included in the analysis. Comparatively few nigrolineata in the size class 2.0-2.4gu were collected and this is thought to be due to the fragility of the shells of this species in this size class which probably results in more being crushed on collection. The shells of rudis B, rudis C and neglecta are very much more robust at this size.

The juvenile rudis B population suggests that there might be some reproductive seasonality in this species or alternatively survival varies over the year. Two pulses of recruitment are seen in May-June and November-February.

Overall the data obtained suggest that some dovetailing does occur. The peak of numbers in the smallest size classes for neglecta comes in October shortly after the marked decline in numbers of nigrolineata (which affects all the size classes) and when numbers of rudis B in these small size classes is particularly low.

Discussion

The dovetailing of the size structure of the populations of neglecta and the juveniles of other species suggests that temporal effects may be important in the niche separation. It is particularly interesting that the population structures of nigrolineata and neglecta are so distinctly dovetailed as at many sites these are the main species in the barnacle belt; the site at Porth Swtan is somewhat atypical in the high numbers of rudis juveniles present. The dovetailing presumably results from the difference in breeding season, neglecta breeding in the spring and summer (March to September) and nigrolineata principally in August and September (see section 2.4.). The difference in breeding method results in further staggering of the appearance of young on the beach, as the eggs of nigrolineata may take an appreciable time to reach hatching point (an arcana egg mass took 73 days to reach hatching at 5°C). .

3.3.4. Intraspecific zonation patterns in L.neglecta

Competition occurs not only between species but also within them. From the view point of the individual winkle its greatest competitors will be members of its own species. The possibility of such competition in neglecta being reduced by different components of the population (young or old, male or female) occupying different niches was examined.

3.3.4.1. Zonation of male and female L.neglecta

The effect of sex on zonation stategy was examined in neglecta using data from transect 2 for the year April 1977 to March 1978. The ratio of males and females in each zone of the transect was compared to the overall ratio of males to females using Brandt and Snedecor's formula for χ^2.

Results

χ^2=21.47, degrees of freedom =7 which was significant at 1%. Hence the null hypothesis that male and female neglecta zone similarly can be rejected. There is a significant difference in the zonation of the two sexes. Examination of the data (table 6, Appendix C) and figure 55 indicates that males tend to zone above females, males preferring zones 2 & 3 and females zones 3 & 4 and to a lesser extent zone 5 also.

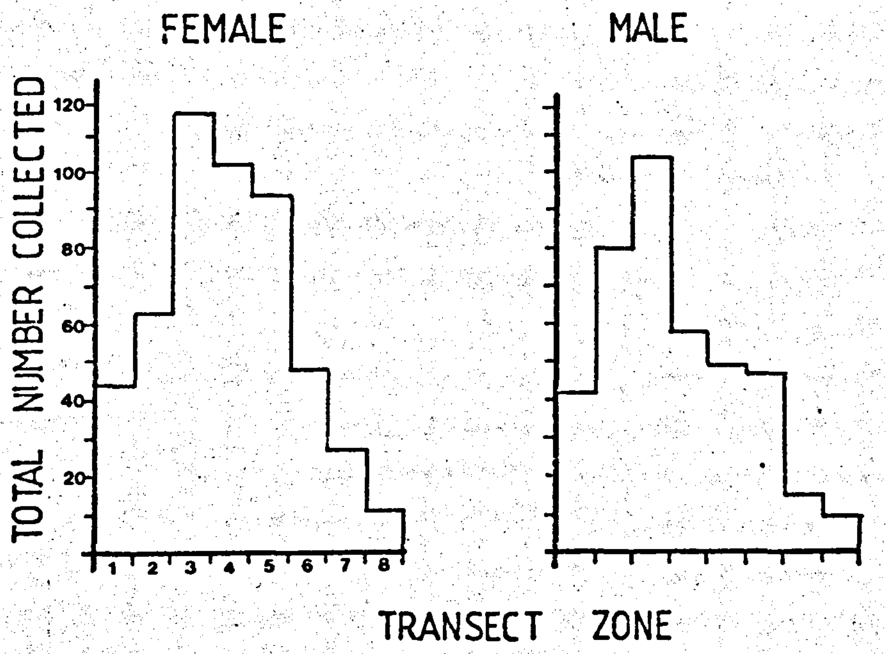
Discussion

The results show that there is a significant difference in zonation pattern of males and

Figure 55.

Zonation of male and female *L.neglecta*.

ZONATION OF MALE & FEMALE NEGLECTA



females. This situation may have arisen in several ways. One possibility is that it is associated with difference in male and female reproductive strategy. If juvenile survivorship is improved lower down (perhaps as mortality due to desiccation is reduced) and adult survivorship is greater higher up, it may be advantageous to females to zone lower where the greater risk is more than offset by the number of surviving young.

Alternatively it may be that size is important, and larger _neglecta_ have a better chance of survival lower down. Female _neglecta_ since they grow larger than males would therefore zone below the males and thus the apparently sex-defined zonation, which has been observed, may arise.

3.3.4.2. Size zonation in L.neglecta

If a size zonation is imposed on _neglecta_ by a size gradation in its habitat, notably the size of dead barnacles as suggested by Raffaelli's work (1978), then males and females of the same size range should zone similarly, the zonation related to sex observed could then derive from the marked sexual dimorphism.

The ratio of males and females in each zone for each size class was compared to the overall ratio of males to females in the size class using Brandt and Snedecor's formula for χ^2. The data from transect 2 for the year were combined for this analysis.

Results

The data are given in table 7, Appendix C and the results are summarised below:-

Size class (gu)	χ^2	Degrees of freedom	Significance
2.0-2.4	0.621	3	Not significant
2.5-2.9	4.76	6	Not significant
3.0-3.4	28.63	6	Sig. at 0.001
3.5-3.9	13.64	4	Sig. at 0.01

The results show that there is no difference in zonation of males and females in the size classes 2.0-2.4gu and 2.5-2.9gu, but significant differences in zonation of males and females occur in the larger size classes.

Discussion

The possibility of the difference in male and female zonation arising indirectly through the combined effect of sexual dimorphism and gradation of sizes with exposure level, can therefore be rejected. The observed difference in zonation of the sexes appears to be wholly a function of sex.

SECTION 4: VARIATION IN L.RUDIS.

The species name L.rudis currently covers all the large ovoviviparous females (i.e. of greater size than neglecta) and all the males of similar size and shells, with penes which have tips longer than the width of a penial gland. It occurs abundantly in an unusually great variety of habitats. It can be found living in the extremely exposed conditions of Rockall, for example, (S. Smith, pers. comm.) yet may also be found on sheltered beaches such as Beddmanarch Bay (SH 275806) and in saltmarshes , eg. Aberlady Bay (NT 437768). The appearance of the winkles living in such different habitats varies enormously.

Examination of the winkles from a number of sites in Anglesey gave the impression that two varieties commonly occurred. These varieties could be recognised at a population level though not neccessarily on an individual basis. One variety generally had robust, tall-spired, murky-coloured shells and was typically found on more sheltered boulder-pebble shores. The other variety had more fragile, globose, clear-coloured shells and was typically found on more exposed shores in the crevices of stable rock masses. Whether the correlation of variety with habitat is a correlation with substrate type or exposure is uncertain as substrate and exposure level are themselves usually correlated in Anglesey.

Unusually, at Porth Swtan both substrate types are present and support these different varieties of *L.rudis*. In spite of their proximity (which can be measured in inches) the two varieties seem to maintain their respective identity. The following section looks at some of the differences, particularly conchological, that distinguish these two varieties at Porth Swtan and compares the situation with that at another site, Rhoscolyn. At this site a cline, between two forms that paralleled the Porth Swtan populations, seemed to exist. The possibility of a further taxonomic division is briefly considered in the light of the information obtained from these two sites.

4.1. SITE DESCRIPTIONS AND SAMPLING.

4.1.1. Porth Swtan

The Porth Swtan site is described, in general terms, in section 3.1. At this site two distinct substrates exist, stable rock masses and unstable boulders and pebbles. These offer dissimilar habitats for winkles. The areas of unstable boulders and pebbles lie between the outcroppings of rock (see Plate 32) so that the two habitats abut. Although they are side by side it is probable that the two substrates experience different degrees of exposure. The boulder-pebble habitats seem to be protected from the full force of the waves by the rock masses, so the exposure levels experienced in the unstable habitat may be equivalent to that on a more obviously sheltered shore. This substrate also remains fairly damp at low tide. It is comparatively low-lying and shallow pools are often present. In dry weather the winkles frequently move down through the interstices between the pebbles and boulders, at low tide. They apparently seek out the damper regions and avoid the desiccating heat of the upper surfaces. In contrast the rock masses offer fewer damp refuges and even crevices under Pelvetia (which are generally damp) may become dry during sunny weather. Little free water is generally retained on the rock surface.

The boulder-living rudis population (rudis B) was sampled monthly by collecting about 200 individuals starting at a point and working outwards. These were taken from the same locality in the boulder-pebble habitat. The crevice-living rudis population (rudis C) was sampled along a transect (see section 3.2.1.) and the data were amalgamated. For the following analyses the data were obtained from samples of the two populations taken over the year, March 1977 to February 1978, or from the November 1977 samples only. The data from the single month's samples were used in those analyses in which less data was required.

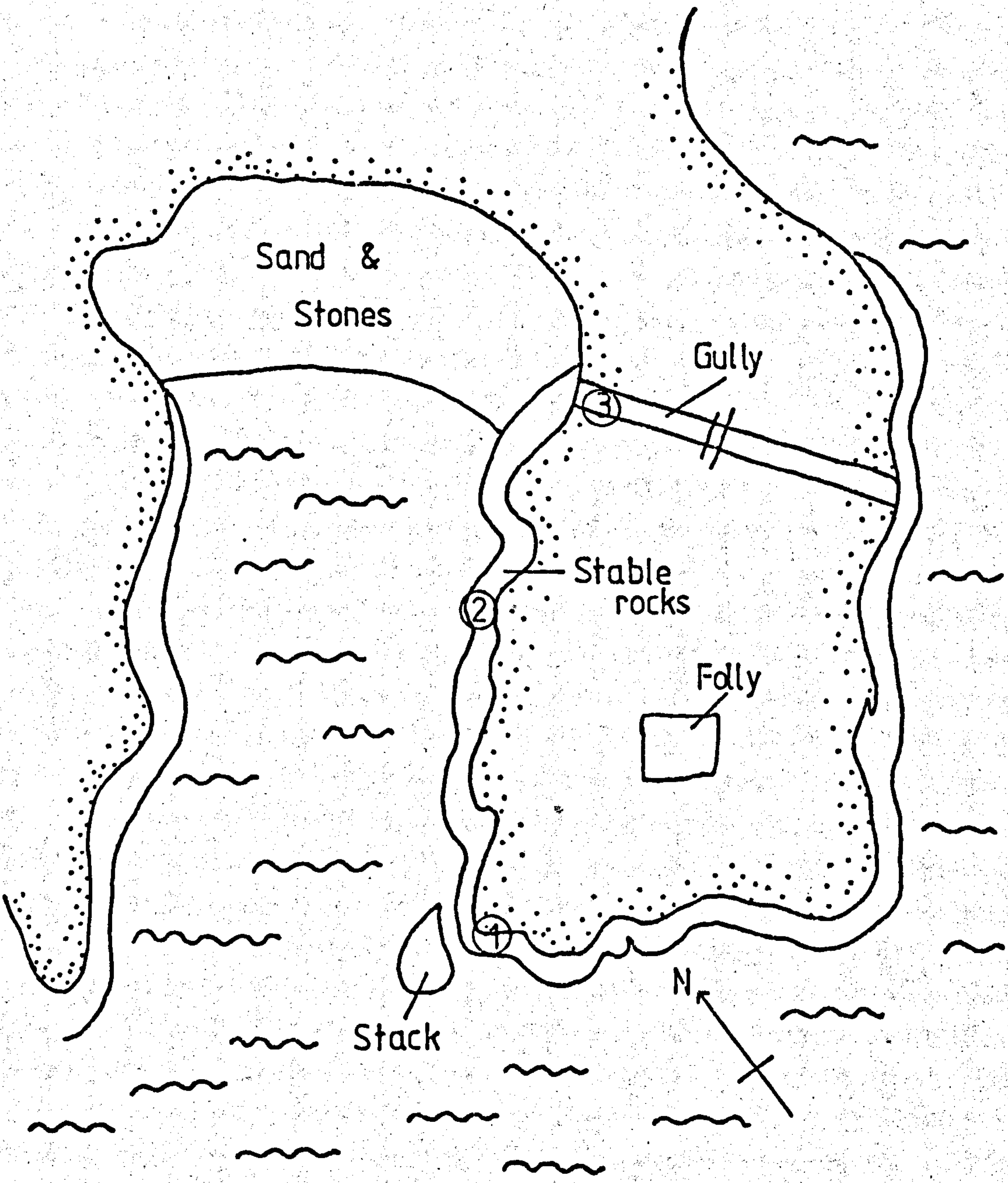
The particular populations from which the samples were taken were, in fact, not directly juxtaposed because of a belt of barnacles. This was atypical of the site in general. Most rock masses at Porth Swtan were positioned higher up the beach and lacked the belt of barnacles. However, adults (and juveniles, see section 3.3.) of both populations overlapped in the barnacle belt on the rock mass used, and since the crevice-living population was similar to the populations on the other rock masses, where the two habitats and rudis varieties directly abutted, it was felt unneccessary to repeat the work already done on these study populations.

4.1.2. Rhoscolyn.

At Rhoscolyn there is an inlet(Grid Ref. SH 291747) formed by a right-angled spit of land (see sketch map, fig. 56). The rock strata of the spit lie nearly vertically, and in the intertidal zone the softer strata have been worn away leaving large, deep clefts. The rock surface has many small crevices which are of appropriate sizes to harbour the abundant winkle population. The mouth of the inlet is moderately exposed, rating

Figure 56.

Sketch map of the Rhoscolyn site.



SKETCH MAP OF RHOSCOLYN SHOWING SAMPLE AREAS

4 on Ballantine's exposure scale. The stable rocks here support a large population of *rudis* (and also *arcana*) which appear to be similar to *rudis* C at Porth Swtan, and these are termed *rudis* c. Moving from the mouth to the base of the inlet (where sand and small pebbles have been deposited) the conditions become progressively more sheltered. The rocks along the side of the inlet are geologically similar to those at the mouth and offer a comparable topography to the winkle community. However, they lie in more sheltered conditions, rating 6 on Ballantine's scale. They support a fairly large population of *rudis* (but very few *arcana*) which appear intermediate between *rudis* c-type and *rudis* b-type(see below) and are termed *rudis* I. Transecting the spit is a gully, apparently man-made. This is floored with small boulders and pebbles and, at low tide, shallow pools remain. This substrate resembles the unstable boulder-pebble habitat at Porth Swtan and supports a population of rudis (but no *arcana*) which resemble *rudis* B and are termed *rudis* b.

These three populations were sampled in the same manner as the *rudis* B population at Porth Swtan, in November 1977.

4.2. COMPARISON OF SHELL CHARACTERISTICS

The nature of the shell secreted by an animal is undoubtedly profoundly important in its life, since its survival depends, in part, on various characteristics of its shell: Shell colour and patterning are probable factors in predation (Reimchen 1974, Heller 1975b), its size, a factor in how it can exploit crevice shelter (Emson & Faller-Fritsch 1976, Raffaelli & Hughes 1978), its mouth shape, in protecting it from crab predation (Heller 1976) and its thickness in protecting it from crushing and attack by crabs (Raffaelli 1978b).

The differences in the shells of the boulder-living and crevice-living *rudis* populations are striking. The following section is aimed at quantifying some of these differences and at examining whether or not the shells of the sheltered-crevice-living *rudis* population (*rudis* I) are in fact intermediate between the b-type and c-type. The characteristics of the shell that are examined here are shape, thickness and colour.

4.2.1. Shell shape.

In general the shells of the boulder-living *rudis* seem to have a smaller apical angle and larger spiral pitch than those of the crevice-livng *rudis*, i.e. the former have taller-spired shells than the latter. Furthermore, the columellar lip seems to be better developed and the aperture narrower in the boulder-living populations. The following analyses test whether there are significant statistical differences in shell characters of the rudis populations studied.

A number of measurements of the shells of the animals collected were taken, using dial calipers accurate to .05mm. These were shell height (SHT), shell width (SW), shell diameter (SD), internal aperture length (IA), external aperture length (EA) and internal mouth diameter (IMD). These measurements are defined in fig.24. Ratios describing shell shape characters were calculated from them viz:- SHT/SW, SW/SD, IA/EA, IMD/IA. These can be used to give an assessment of the relative tallness, symmetry (in spire view), degree of expansion of the columellar lip, and mouth shape respectively. The ratios were calculated on shells in the height range 8.5-9.95mm for Rhoscolyn. Thus, if shell shape varies with shell height, possible size bias in sampling should not affect the comparison. This was not possible for Porth Swtan material as the shell height ranges of rudis B and rudis C in the samples collected were so disparate. The shell height ranges used were 8.5-9.95mm for rudis C and 11.5-12.95mm for rudis B. The data were obtained from the samples taken in November at Porth Swtan.

Results.

The results are summarised in table 8.

a) Porth Swtan.

The results indicated that rudis B shells were significantly taller-spired and more asymmetrical, and showed more expansion of the columellar lip than rudis

TABLE 8. Comparison of shell shape measurements using Mann-Whitney U test.

PORTH SWTAN

Ratio	*L. rudis* C			*L. rudis* B					
	n	median	range of observations	n	median	range of observations	z	two-tailed p	Significance
SHT/SW	87	1.095	1.012-1.171	57	1.185	1.090-1.292	9.19	.00006	Sig. at .01%
SW/SD	87	.664	.610- .697	57	.714	.690- .829	10.10	.00006	Sig. at .01%
IA/EA	87	.661	.568- .722	57	.562	.511- .646	9.9	.00006	Sig. at .01%
IMD/IA	87	.781	.687- .926	57	.786	.689- .845	.85	.3954	N.S.

RHOSCOLYN

	L. rudis c			*L. rudis* I					
SHT/SW	57	1.102	1.029-1.208	82	1.147	1.021-1.283	5.76	.00006	Sig. at .01%
SW/SD	57	.675	.605- .708	82	.689	.833- .742	3.38	.0006	Sig. at .01%
IA/EA	57	.605	.523- .718	82	.617	.511- .678	1.41	.1586	N.S.
IMD/IA	57	.792	.714- .970	82	.795	.641- .959	.16	.8728	N.S.

	L. rudis I			*L. rudis* b					
SHT/SW	82	1.147	1.021-1.283	50	1.192	1.090-1.329	6.38	.00006	Sig. at .01%
SW/SD	82	.689	.633- .742	50	.709	.677- .756	6.60	.00006	Sig. at .01%
IA/EA	82	.617	.511- .678	50	.562	.489- .652	8.02	.00006	Sig. at .01%
IMD/IA	82	.795	.641- .959	50	.773	.722- .861	2.33	.0198	Sig. at 2%

	L. rudis c			*L. rudis* b					
SHT/SW	57	1.102	1.029-1.208	50	1.192	1.090-1.329	7.88	.00006	Sig. at .01%
SW/SD	57	.675	.605- .708	50	.709	.677- .756	7.84	.00006	Sig. at .01%
IA/EA	57	.605	.523- .718	50	.562	.489- .652	6.91	.00006	Sig. at .01%
IMD/IA	57	.792	.714- .970	50	.773	.722- .861	2.59	.0096	Sig. at 1%

NOTE:
SHT: - shell height
SW: - shell width
SD: - shell diameter
IA: - internal aperture length
IMD: - internal mouth diameter
These measurements are defined in figure 24.

N.S.: - not significant

C shells, but there was no significant difference in the shape of the aperture.

b) Rhoscolyn.

At this site rudis b were significantly taller-spired, more symmetrical & showed more expansion of the columellar lip. In these characters they, therefore, paralleled those seen between rudis B and rudis C at Porth Swtan. However, they were, in addition, narrower-mouthed than either rudis I or rudis c, though the difference is not so marked in mouth shape as in the other characteristics. The I-population are taller-spired and more symmetrically-shelled than rudis c but are similar in mouth shape and degree of columellar lip expansion. Thus the shell shape characteristics of the rudis I population seem to be intermediate between the b and c forms.

4.2.2. Shell thickness.

The degree of protection from crushing (either by boulders/pebbles or crabs) afforded by a shell will depend, in part, on the thickness of the shell over the surface of the animal. Observations suggested that the shells of the crevice-living variety were relatively thinner than those of the boulder-living type,

In comparing the thickness of the shells produced by different populations several factors must be taken into account. Firstly, in general, shell thickness increases with age; older animals have thicker shells than younger ones. This relationship is, however, unlikely to be linear and furthermore, it does not neccessarily mean that the relative investment in shell production increases with age. Thus in comparing shell thickness between populations the age or size of the animals must be taken into account. Secondly, since one of the differences between the boulder-living and crevice-living varieties is a difference in shell shape, winkles of the same height from the two populations will probably be of different tissue weights. Or, put another way, a larger animal from the short-spired, crevice-living population could have the same shell height as a smaller animal from the tall-spired, boulder-living population as they lay down shells with different apical angles and spiral pitch. Thus the relationship between the size of the animal and the shell height is likely to differ in the two populations.

In the following assessment of the relative thickness of the shells produced by the five rudis populations studied, the ratio used is AL/T (AL=animal length, defined in fig. 10, and T=shell thickness measured about 2mm inside the lip and through a ridge on ridged shells, defined in fig. 24). The use of the measurement AL avoids the problems involved in using shell dimensions (discussed above) but assumes that the proportion of the head mass to the overall size of the animal (ie. soft tissue) is equivalent in all the rudis populations examined. The relative shell thickness is assessed from measurements of individuals in the shell height range 8.5-9.95mm from Rhoscolyn and 7.5-8.95mm for material collected in November from Porth Swtan. Mann-Whitney U tests were used to

Table 9. Comparison of shell thickness ratios using Mann-Whitney U test

PORTH SWTAN

L. rudis C			*L. rudis* B					
n	median	range of observations of AL/T	n	median	range of observations of AL/T	z	two-tailed p	Significance
67	9.67	7.14-19.0	51	8.09	5.62-11.55	5.89	.00006	Sig. at .1%

RHOSCOLYN

L. rudis c			*L. rudis* b					
82	10.29	6.4 -19.0	50	7.45	5.55-10.86	6.27	.00006	Sig. at .1%
L. rudis b			*L. rudis* I					
50	7.45	5.55-10.86	100	10.0	6.67-16.2	7.52	.00006	Sig. at .1%
L. rudis c			*L. rudis* I					
82	10.29	6.4 -19.0	100	10.0	6.67-16.2	.54	.5892	N.S.

compare the ratios from the different populations.

Results.

The results of the comparisons between populations at the two sites are summarized in table 9.

a) Porth Swtan.

The comparison of the ratio AL/T from the two populations showed that rudis C shells are significantly thinner than rudis B shells.

b) Rhoscolyn.

At this site the shells of the rudis b population are significantly thicker than those of either rudis c or rudis I. The shells of rudis c and rudis I are of similar thicknesses.

4.2.3. Shell colour.

At the risk of generalising, it often seems that the ecotypes of rudis exhibit rather different qualities of shell colour. The crevice-living rudis often seem to have a variety of well-defined, attractively-coloured shells while the boulder-living rudis often have rather murky and dull coloured shells. The frequencies of shell colours in the different rudis populations studied are given in table 10. No attempt has been made to score quantitatively the shell colours of the populations considered (as has been done by Pettitt, 1973a) as a descriptive list of colours and frequency seemed to convey the point more simply here.

Table 10:- Frequencies of shell colours.

	L.rudis population:				
Colour of shell.	Porth Swtan		Rhoscolyn		
	B	C	b	I	c
Bright yellow	-	-	-	67	121
Pale yellow	-	-	7	44	21
Yellow-white	-	-	1	42	18
Yellow with (1 or 2) brown bands	-	-	-	6	10
White	-	-	-	36	5
Grey-white	-	-	5	3	1
White with purple bands	-	-	-	-	2
Fawn-white	4	-	6	5	-
Pink-white	-	-	1	4	-
Brown	-	-	-	4	-
Pale brown	-	-	-	16	1
Grey-brown	6	-	3	6	-
Fawn-brown	6	-	8	-	-
Dark brown	2	12	-	-	-
Fawn	38	-	52	-	-
Yellow-fawn	2	-	16	3	-
Fawn with brown bands	1	-	-	-	-
Grey	10	-	-	-	-
Dark grey	45	-	-	-	-
Dark grey with white stripe	2	-	-	-	-
Black	-	107	-	-	-
Orange	-	-	-	1	-
Brown-purple	-	-	3	-	-
Peach	-	-	-	9	3
Pale purple	-	-	-	-	1

Results

a) Porth Swtan.

The rudis C population exhibits a very limited range of colours with only black and dark brown shells (see Plate 31). The rudis B population shows a wider range of typically dull colours, most common of which are fawn and dark grey.

b) Rhoscolyn.

The colours of the rudis c and rudis I populations are very similar. In both populations yellow and white are the commonest shell colours and in both there are individuals with the dramatically-coloured, brown-banded yellow shells. In contrast the rudis b population tend to have dull-coloured shells which are predominantly fawn and yellow-fawn. None of the animals taken from this population had shells that were bright yellow, white or yellow with brown bands.

SUMMARY OF SECTION 4.2.

The data collected support the observed difference in shells of crevice-living and boulder-living varieties of rudis. Boulder-living rudis tend to have thicker, taller-spired, more symmetrical, duller-coloured shells with more expanded columellar lips than the crevice-living rudis. A significant difference in mouth shape was, however, observed only at Rhoscolyn and not at Porth Swtan. At Rhoscolyn the shells of the intermediate population, rudis I, bridge some, but not all, of these differences. They tend to be intermediate in shape characteristics, but in both colour and shell thickness they show greater affinity to the crevice-living population. The interpretation of these results is discussed in section 4.5.

4.3. PENIS CHARACTERISTICS.

Significant differences in the form of the reproductive organs are good species-markers, and penial form often differs between species of winkles. With L.littoralis the initial indication that this was a mixture of two species (obtusata and mariae) was the observation of two penial forms (Sacchi, 1966). Between the crevice-living and boulder-living rudis however, there appears to be little difference in penis form, except in overall size; the boulder-living form, being generally larger, has correspondingly larger penes.

Comparisons were made of the penial proportions between the populations at each site using the Mann-Whitney U test. The ratios compared were relative tip length (TL/PL), relative size of the penial gland region (PGL/PL), relative base length (BL/PL) and relative size of the penis (PL/AL). The measurements TL,PGL,PL,BL and AL are defined in fig. 10. The frequencies of penial gland number and arrangement were compared for the Porth Swtan populations. With the Rhoscolyn population,only the frequency distributions of penial gland numbers were compared as the samples were too small for comparison of frequencies of arrangement pattern. The data given for Porth Swtan populations were derived from material collected over the period of a year.

Results.

The results of the comparisons of penis proportions are summarised in table 11 and the frequency

Table 11: Comparison of penis proportions using Mann-Whitney U test

PORTH SWTAN

Ratio	*L. rudis* C			*L. rudis* B					
	n	median	range of observations	n	median	range of observations	z	two-tailed p	Significance
PL/AL	100	1.120	.776-1.568	78	1.100	.750-1.577	1.38	.1676	N.S.
TL/PL	94	.236	.134- .365	80	.171	.061- .369	7.19	.00006	sig. at .01%
PGL/PL	95	.606	.375- .780	80	.620	.230- .842	.77	.77	N.S.
BL/PL	93	.171	.000- .275	80	.220	.000- .500	2.89	.0038	sig. at 1.0%

RHOSCOLYN

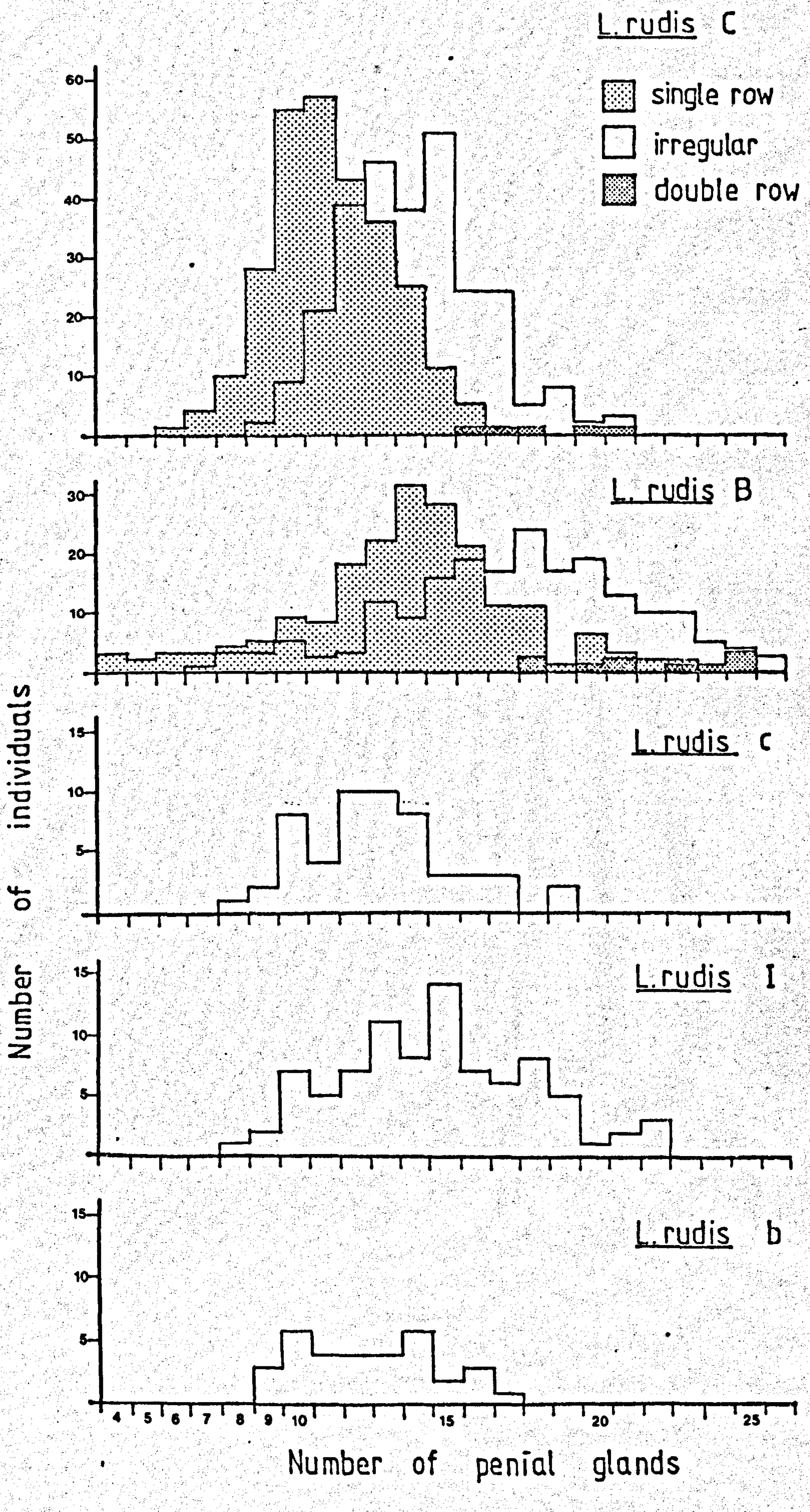
	L. rudis c			*L. rudis* I					
PL/AL	38	1.059	.781-1.328	69	1.055	.754-1.397	.48	.6312	N.S.
TL/PL	38	.253	.191- .339	69	.224	.128- .304	3.48	.00046	sig. at .01%
PGL/PL	38	.618	.495- .759	69	.627	.461- .786	.13	.8966	N.S.
BL/PL	38	.117	.000- .253	69	.150	.000- .333	1.89	.0588	N.S.

	L. rudis I			*L. rudis* b					
PL/AL	69	1.055	.754-1.397	27	1.061	.890-1.441	.71	.4778	N.S.
TL/PL	69	.224	.128- .304	27	.224	.155- .319	.31	.7566	N.S.
PGL/PL	69	.627	.461-.786	27	.625	.439- .775	.07	.9442	N.S.
BL/PL	69	.150	.000- .333	27	.151	.000- .337	.25	.8026	N.S.

	L. rudis c			*L. rudis* b					
PL/AL	38	1.059	.781-1.328	27	1.061	.890-1.441	.06	.9522	N.S.
TL/PL	38	.253	.191- .339	27	.224	.155- .319	2.38	.0172	sig. at 2%
PGL/PL	38	.618	.495- .759	27	.625	.439- .775	.24	.8104	N.S.
BL/PL	38	.117	.000-.253	27	.151	.000- .337	1.42	.1556	N.S.

Figure 57.

Frequencies of penial gland number (and arrangement where applicable) in the L.rudis populations at Porth Swtan and Rhoscolyn.

L. rudis C
single row
irregular
double row
60
50
40
30
20
10
0
L. rudis B
30
20
10
L. rudis c
15
10
5
L. rudis I
15
10
5
L. rudis b
15
10
5
4
5
6
7
8
9
10
15
20
25
Number of individuals
Number of penial glands

distributions of penial gland numbers and arrangement (the latter for Porth Swtan material only) are given in table 1, Appendix D, and plotted in fig.57.

a) Porth Swtan.

The penes of the rudis C individuals had relative tip lengths that were significantly longer and relative basal lengths that were significantly shorter than those of the rudis B individuals. There were no significant differences in the relative length of the penial gland region, nor in the proportions of penis to animal size. Though the frequencies of the different modes of penial gland arrangement are closely similar in the two populations it appears that males of the rudis B type tend to have more penial glands.

b) Rhoscolyn.

The penes of the rudis c individuals had tips that were significantly longer than those of rudis I or rudis b individuals. No other significant differences in proportion were noted. Only penial gland numbers were compared between the populations (fig. 57) as too little material was available for a comparison of arrangement to be made. The data suggest that the rudis I population may have more penial glands than either c or b populations but the difference is relatively slight and is probably not significant.

Discussion

Overall, the differences noted, which mainly concern the Porth Swtan populations, are far less extensive than those seen between rudis and arcana (see table 3, p.19 onwards) and are not sufficiently marked to be regarded as indicative of separate species. It is interesting that the percentage of single and irregular penial gland arrangements are, in the *rudis* B population, 49% and 49% and in the *rudis* C population 50% and 49% (respectively). Thus the distribution of patterns of arrangement is nearly identical though *rudis* B individuals tend to have more penial glands (see fig. 57). Hence the arrangement of the penial glands is not purely a function of number as suggested by Raffaelli (1979). Since the relative length of the penial gland region is no different in the two populations, this suggests that the relative size of the glands may be slightly different.

4.4. SIZE FREQUENCY AND AVERAGE MATURATION SIZE.

The average size above which winkles are generally capable of reproducing varies in different populations and appears to be related to the size frequency of the population. It is noticeable that winkles of boulder-living *rudis* populations seem to grow much larger than crevice-living *rudis*, and are generally larger when they come into reproductive condition. These population characteristics can be seen by looking at a) the size frequency and size range of the population, and b) at the percentage mature in successive size classes. Only animals greater than 6.5mm (shell height) were considered, since small animals (less than about 6.0mm) are disproportionately more difficult to collect. For the Porth Swtan populations data collected over the year were used for the comparison.

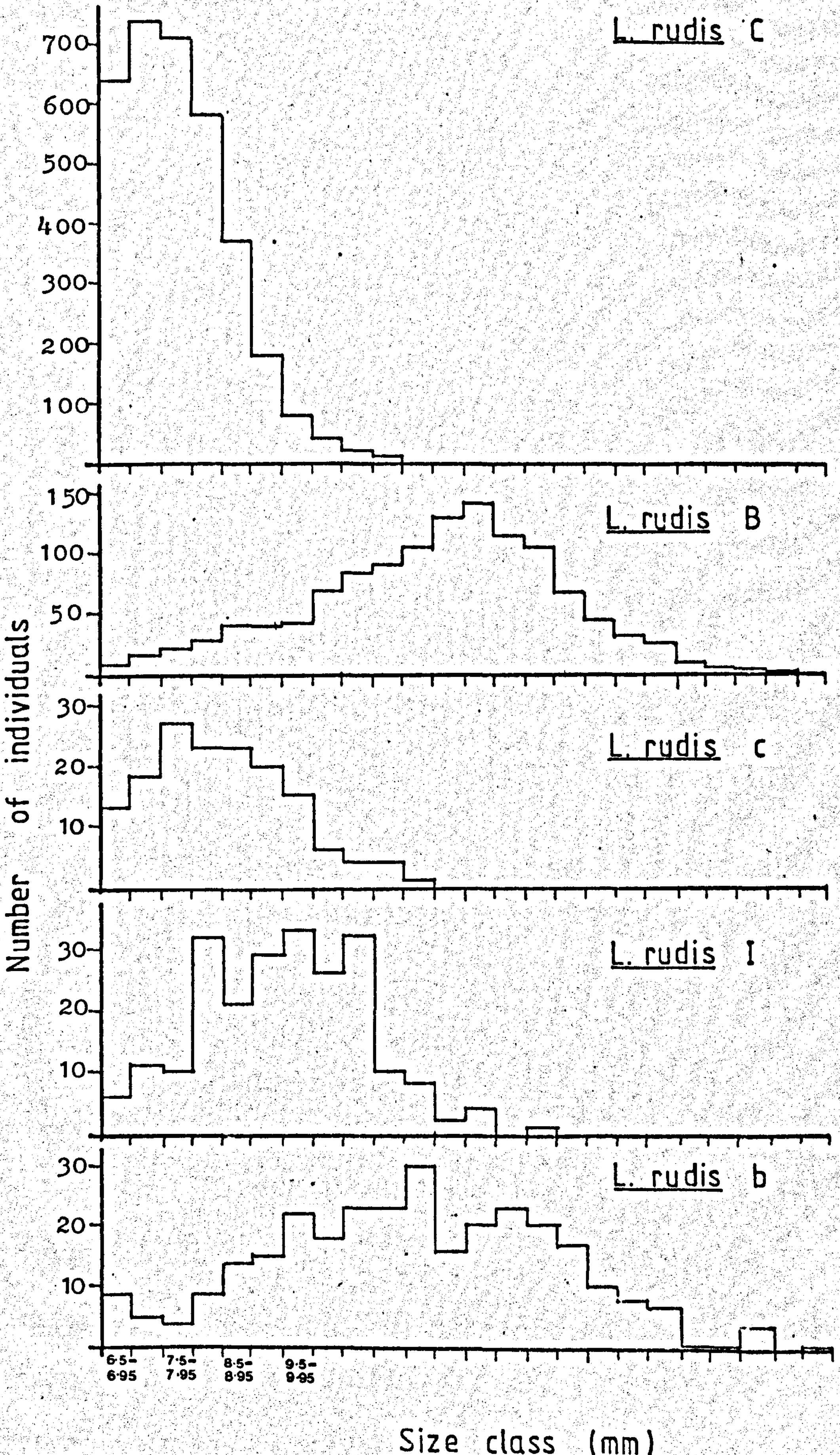
Results.

The data are given in table 2, Appendix D. The size frequencies of animals from the different populations are plotted in fig.58. and the % mature in successive size classes are plotted in fig. 59. The smallest size class in which 50% or more are mature is:-

POPULATION		Smallest size class in which 50% or more are mature.
Porth Swtan:	*L.rudis* C	7.0-7.45mm
	L.rudis B	11.0-11.45mm
Rhoscolyn	*L.rudis* c	7.5-7.95mm
	L.rudis I	8.5-8.95mm
	L.rudis b	9.5-9.95mm

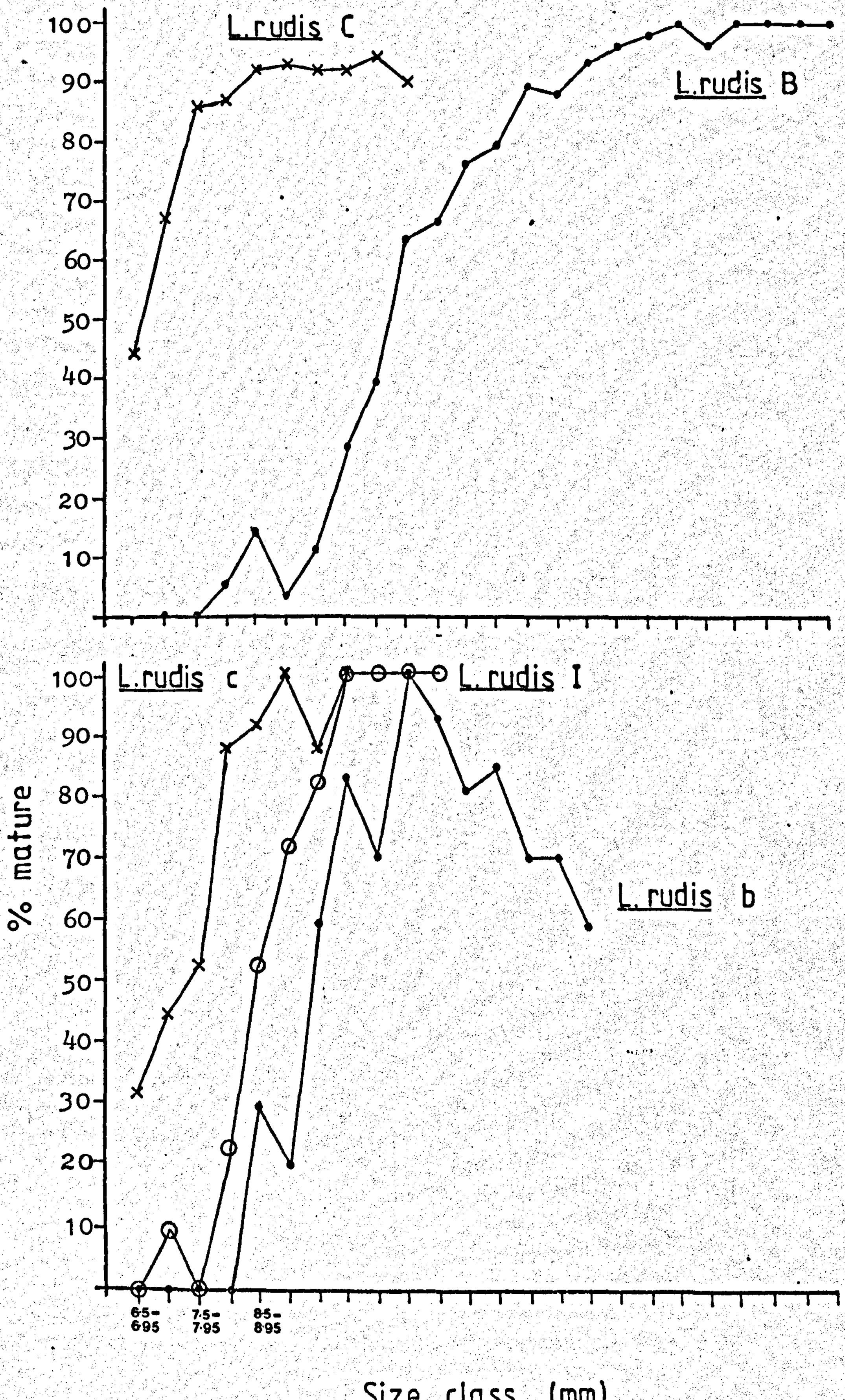
Figure 58.

Size frequencies in L.rudis populations B,C,b,c & I.


L. rudis C
700
600
500
400
300
200
100
L. rudis B
150
100
50
L. rudis c
30
20
10
L. rudis I
30
20
10
L. rudis b
30
20
10
Number of individuals
6·5–6·95
7·5–7·95
8·5–8·95
9·5–9·95
Size class (mm)

<u>Figure 59</u>.

Percentage of mature individuals in successive size classes.


100
90
80
70
60
50
40
30
20
10
L.rudis C
L.rudis B
L.rudis c
L.rudis I
L.rudis b
% mature
6·5–6·95
7·5–7·95
8·5–8·95
Size class (mm)

a) Porth Swtan.

Individuals of the <u>rudis</u> C population tend to mature at a much smaller size than <u>rudis</u> B individuals. The data also show (see fig.58) that <u>rudis</u> B individuals generally grow much larger than <u>rudis</u> C and may achieve nearly twice the shell height of even the tallest <u>rudis</u> C individual.

b) Rhoscolyn.

Individuals of the <u>rudis</u> c population tend to mature at smaller shell heights than either <u>rudis</u> I or <u>rudis</u> b. The difference is less marked between b and c types than between the comparable Porth Swtan populations, C and B. The maximum size is largest in <u>rudis</u> b, less in <u>rudis</u> I and least in <u>rudis</u> c. The lower values for % mature in size class 11.5-11.95mm and above for the <u>rudis</u> b population could be due either to a local reproductive cycle effect at the time of collection, or to senescence of these large animals. Parasitism is not suspected, as parasitized animals were not included in the data used.

<u>Discussion</u>.

These differences noted in the size frequencies and average size of maturation suggest important differences in the ecology of the populations living in the different habitats. Several suggestions have been made, as to how such differences may have arisen, by previous authors. Raffaelli and Hughes (1978) working on similar populations of <u>rudis</u>, and also <u>neritoides</u>, have shown that as crevice size increases, so does the maximum size of the winkles sheltering in them. These authors have suggested that in exposed crevice habitats, larger winkles will have an increased risk of mortality since they are less likely to find suitable crevices in which to shelter, and this will produce a selection pressure for maturation at comparatively small shell heights. On sheltered shores they found that size frequencies of L.rudis and crevices were not related. Emson and Faller-Fritsch (1976), working on rudis populations from exposed and sheltered boulder habitat shores have suggested that larger size is selected for especially in exposed conditions. They argue that in such habitats small winkles are at greater risk of crushing or predation and they have suggested that the winkles are selected for rapid growth to large sizes, thus outgrowing size ranges at greater r risk before becoming mature. Thus the selective pressures of the habitat could be of considerable importance in affecting the size frequency and maturation size of the animals at a locality. The authors of these studies have suggested that there is a genetic basis for the adaptation of rudis to different environments. However, it is also possible that the differences may result from phenotypic adaptation, or through heavy selection.

4.5. DISCUSSION AND TAXONOMIC IMPLICATIONS OF SECTION 4.

At Porth Swtan the habitat types are clearly demarked. The substrate for winkles is either stable rock masses or unstable boulders and pebbles. The populations of rudis which inhabit these different substrates are shown to differ markedly in shell and population structure characteristics and there are no obvious intermediate populations. At Rhoscolyn, populations that parallel those at Porth Swtan can be seen. However, there appears to be a clinal situation between these forms at this site since, in an area intermediate, both geographically and in exposure level, there are rudis exhibiting intermediate characteristics.

Several interpretations could be put upon the observations made. The data could support arguments for the existence of a). sympatric species (it could be suggested that the intermediate population at Rhoscolyn represents a sheltered form of the stable-habitat type and the differences between I and c types could be interpreted as phenotypic or as the result of differing selective pressures), b). genotypically-adapted forms (ecotypes), c). phenotypically-adapted forms or d). as the result of heavy selection against inappropriate forms in each habitat.

Certainly L.rudis is a species in which sympatric speciation could easily occur. Its biology fits the theoretically-predicted optimum requirements

(Maynard Smith, 1966) for sympatric speciation to occur,viz:-

a) It lives in a heterogeneous environment in which density-dependent factors regulating population size operate independently in the two niches.

b) There is likely to be a low interchange of genetic material (the size difference at Porth Swtan probably makes it mechanically difficult for inter-varietal matings to occur).

c) The species is one in which the females lay eggs in the habitat in which they, themselves, grow up.

In view of the lack of anatomical differences which the animals, themselves, might use to recognise conspecifics (which are especially likely to exist in sibling species when they are sympatric, Cain,1978) the interpretation, that the variation is a reflection of distinct species, seems unlikely. Raffaelli & Hughes (1978) have suggested that these factors (a-c) in the biology of <u>rudis</u> have resulted in an ecotypic situation, which could also account for the variation observed. However, the other possibilities cannot be discounted. Clearly the simple examination of the physical appearance of the winkles cannot provide a solution to the question of how the variation arises. Critical experimentation, designed to answer this question is needed.

Since any formal taxonomic recognition of these forms would imply a genetic basis for the variation, to describe the forms studied herein, as for example, varieties, would be premature. However, some nominal recognition of the variation in <u>rudis</u> would be helpful as an aid to ecologists in view of the pronounced differences in biology that can be observed.

GENERAL DISCUSSION.

TAXONOMIC AND FUNCTIONAL ASPECTS.

Heller (1975a) split the rough winkle L.saxatilis into four species, patula, rudis, neglecta and nigrolineata, using primarily the shell and penial form as diagnostic characters. For each species he also noted the breeding habit, esterase pattern, radula appearance and habitat.

It was initially intended to examine, in greater detail, the ecology of these four sibling species. However, it was found that two of the species, rudis and patula had been described from mixed-species material (Hannaford Ellis, 1979) and furthermore that the current descriptions were inadequate for identifying the juveniles and immature individuals. Thus it was necessary to alter the taxonomy of the rough winkles (Hannaford Ellis, 1978 & 1979) and to expand the descriptions of the species to include juveniles and immature animals before examination of the ecology of the species could be undertaken.

Those aspects of the identification of rough winkles which had not been described adequately by other authors are described in section 1. Thus particular attention was paid to defining the characters that could be used to identify juveniles (in which sex cannot be discerned)and immature individuals of the four species, and the adults of rudis and arcana. It was unnecessary to define the characteristics of adult nigrolineata and neglecta as descriptions of the characters used for identifying them were already available (Heller, 1975a, Sacchi, 1975).

When samples of juvenile rough winkles were collected, juveniles of the other winkle species, mariae, obtusata, littorea and neritoides, were often mixed with them and so these had to be identified before the rough winkles were examined. Despite the fact that some ontogenetic changes are seen in obtusata and mariae (Reimchen, 1974) and in littorea (section 1.7.2.) all four species have species-specific shell characteristics at all ages which can be used to identify them.

Specific differences, which would allow separation of the four rough winkle species, were looked for in the shell, pallial oviduct, penis, prostate, ciliated field, sub-opercular pattern and radula. The ciliated field and prostate were only examined in rudis and arcana since the more readily quantifiable shell and penial characters were not diagnostic. By using combinations of these characters all individuals except juvenile arcana and rudis, which defied separation from each other, could be identified.

Besides their taxonomic application, the functional aspects of the structures are important and observations made on them have posed many questions that need to be answered. Both taxonomic and functional aspects of each structure are considered below.

1). Pallial oviduct

The appearance of the mature pallial oviduct has been shown to distinguish *rudis* and *nigrolineata* females by Sacchi (1975). Though he, and other authors (Linke, 1933, Fretter & Graham, 1962) have figured the external appearance of the mature oviducts, these figures have not made clear the anatomy of this complex structure. The structure of both immature and mature oviducts in *arcana* (oviparous) and *rudis* (ovoviviparous) were examined in detail and the differences noted. The oviducts of these species are different in immature as well as mature states and these differences can be used to diagnose the females.

Since reproductive mode is the same in *rudis* & *neglecta* (ovoviviparous) and in *arcana* & *nigrolineata* (oviparous) general oviduct anatomy is insufficient to distinguish between these pairs of species. However, since the shell characters of both *nigrolineata* and *neglecta* are diagnostic, pallial oviduct differences are not needed to identify them.

Examination of the oviduct may also point to inter- and intra-specific differences in life history tactics. When comparing interspecifically the oviduct contents of *neglecta* and *rudis* it was noted that the former have fewer, relatively larger embryos (pers. obs.). This suggests, if the rate of embryo development is similar in the two species, that *neglecta* has a lower percentage mortality to reproductive age. Comparable intraspecific differences have been found in different populations of rudis. Faller-Fritsch (1977) examined boulder-living populations and found that in shelter, females produced fewer, larger embryos, and Hart (1978) comparing crevice- and boulder-living populations, found that crevice-living females produced fewer, larger embryos. It is possible that these may represent an ecotypic difference as these authors suggest, but as Stearns(1976) points out until the genetic basis of the differences has been established through laboratory studies, any assertion that a difference observed in the field has evolved and is an adaptation must be viewed with suspicion.

2) Penis.

Penial form can be a useful taxonomic character as Sacchi & Rastelli (1966) showed for obtusata and mariae. However, of the rough winkles only nigrolineata has a diagnostic penial form and this has been described by both Heller (1975a) and Sacchi (1975). The use of the penis in identification is further complicated by the regression (Bergerard, 1971a &b) or shedding (Pettitt, 1973b) of the penis which occurs when the animal goes out of breeding condition. Thus for rough winkles penial form is not particularly useful taxonomically.

Cain (1978), drawing on such examples as mariae, obtusata and nigrolineata has suggested that penial form may play an important role in the recognition of conspecifics and the prevention of hybridisation between related species. Since a) the penes of rudis and arcana appeared to be so similar in form, b) the species are widely sympatric and c) they are of similar size, it would be reasonable to suppose that the chance of hybridisation is greater for these species than for any other pair of winkle species. Furthermore, rudis and arcana may on occasion attempt copulation as, in general, winkles seem to be fairly indiscriminate and do not always copulate only with conspecifics; Raffaelli (1977) reviewed observations by other authors and gave examples of males of rudis and nigrolineata with their penes engorged and inserted into the mantle cavity of conspecific males and individuals of other species, apparently attempting copulation. However, he also noted that reduced time was spent in attempts at copulation between 'unnatural' pairs and suggested that sperm release does not occur. Since rudis-arcana hybrids have not been observed (pers. obs.) it seems that reproductive isolation is maintained (or hybrids are so uncompetitive that they do not survive) despite the possibility of interspecific coupling. The mechanism of isolation has still to be clearly defined and in spite of the apparent similarity of the penes, it may have a mechanical basis. For example, the length of the bursa copulatrix may be an important stimulus (section 1.2.2.1.).

3) Prostate.

The prostate has not been used previously in diagnosis of winkle species. It was examined here only in arcana and rudis and its appearance was found to differ slightly in these two species. It is generally more puckered and of greater width relative to length in arcana. The differences are not, however, diagnostic so it is only of use as an ancillary character in separating the males of these species.

In rudis and arcana histological examination of the prostate showed that at least two sorts of secretory product are present in the cells. The functions of these are unknown.

4) Ciliated field.

The ciliated field in winkles has not been recorded by previous authors. It can be a difficult character to use in identification as it is obvious only in freshly-killed animals. The structure was examined in detail in rudis and arcana and specific differences in size and position of the field were seen. These differences are particularly useful for diagnosing male rudis and arcana.

The function of the ciliated field remains obscure. The direction of beating of the cilia would serve to drive something out of the mantle cavity and there are many possible candidates for expulsion, e.g. water, particles and faecal pellets, but there is no evidence to indicate which, if any, are expelled. Any hypothesis suggesting its function should, however, take into account the variation in size (and therefore, presumably, importance) in the different species and why it appears to be absent in other winkles such as littorea (Dr.V. Fretter, pers. comm.).

5) Sub-opercular pattern.

This character was noted and used in identification of winkles for the first time in this thesis. It was examined in the rough winkles and also littorea and neritoides. Of the rough winkles only the sub-opercular pattern in neglecta was significantly different from those of other species. L.rudis, arcana and nigrolineata patterns are very variable and in these species a similar range of patterns are seen. The pattern of neglecta can, however, be useful as an aid in distinguishing it from juveniles of other species.

The reason for its precise form in neglecta, littorea and neritoides and why these specific differences occur still needs to be elucidated. It is feasible that it has a function in camouflage but there is no evidence to support this or any other speculative suggestion.

6) Radula.

The radula is the least accessible of all the structures that were considered as a basis for making identifications. Its application to diagnosis is complicated by both the ontogenetic changes and the general variability of cusp shape and number that have been found (section 1.6.). These results indicate that the littorinid radula is not such a reliable taxonomic character as Bandel (1974) suggested, and that his key to a worldwide range of littorinid species, which extensively employs cusp number is unworkable, certainly for the species that occur in Britain.

Only in the radula of nigrolineata adults (their radular cusps are blunt-edged) is there any significant difference from other species. This species can, however, be identified much more easily on other characters such as the shell and reproductive organs.

The shape of radula cusps can give clues to the general nature of the diet of a species (Solem, 1974, provides an overall account). It would be interesting if the finer differences in cusp shape and the ontogenetic changes could similarly be correlated to finer specialisations. The changes in the radula, which are most marked in nigrolineata, mariae and obtusata may possibly indicate some important ecological differences in the feeding of juveniles and adults. Comparison of the gut contents may reveal differences in food resource exploitation which could be important in the ecology of these species and would be an interesting field for future research.

7) Shell.

The shell characters of nigrolineata and neglecta are diagnostic (Heller, 1975a) so individuals of these two species can generally be identified by their shells alone. The penial form and pallial oviduct type are useful to confirm the identification especially when the shells are eroded or otherwise damaged.

The shells of *rudis* and *arcana*, though different from those of other rough winkle species, are not sufficiently different from each other to be used diagnostically. The lack of diagnostic shell differences between these species and the observation of ontogenetic changes (section 1.7.) serves to underline the caution that is required in the sole use of the shell in taxonomy and palaeontology.

The functional aspects of the shell deserve detailed examination. Like the radula, the shell can undergo ontogenetic changes which may reflect changing demands, perhaps mechanical or energetic, on the shell as the animal grows. The most dramatic ontogenetic change is seen in the development of *littorea*. The juveniles have extraordinarily tall-spired and heavily-ridged shells and the adults have shells which are medium-or short-spired and almost smooth. The other impressive ontogenetic change is in the ridging of *nigrolineata* shells. The juveniles have markedly triangular-ridged shells and the adults have low, flat ribs. The adaptive significance of such changes may be related to the hydrodynamics of waterflow over the animals and needs to be explored.

Shell colour in winkles is a line of study that has long fascinated researchers (e.g. Fischer-

Piette, Gaillard and James, 1963, Heller, 1975b, Pettitt, 1973a, Raffaelli, 1976, Reimchen, 1974). Unfortunately most of these studies were probably based on mixed species and so the results must be regarded with suspicion. Much remains to be discovered about how colour is controlled. Reimchen (1974) in breeding experiments on <u>mariae</u> showed that there is a genetic basis for shell-colour in this species, and that dark reticulata was dominant to citrina. Whether shell colour in other species, particularly the extraordinarily variable colours of <u>rudis</u> and <u>arcana</u> shells, is genetically controlled remains untested. There is also much to be discovered of the importance of the variation in shell colour. In some cases the colour appears to camouflage the animal and protect it from visual predation. In an experiment on predation of <u>mariae</u> by the fish, <u>Blennius pholis</u>, Reimchen (1974) demonstrated that <u>both</u> citrina and reticulata were camouflaged by their colour on fucoids and avoided predation by the fish, depending on whether the light shone through the lamina of the alga or was reflected from it. He also showed that white-shelled juveniles appeared to avoid visual predation as their colour and size mimicked that of the tubes of the polychaete, <u>Spirorbis</u>.

In rough winkles frequent examples of camouflage are also seen, e.g. Heller(1975b) describes the occurrence of high proportions of red-shelled <u>rudis</u> and <u>nigrolineata</u> on red rocks, whilst in neighbouring populations on other coloured rocks there were few red-shelled animals. However, by no means all populations of rough winkles are cryptically camouflaged. For instance, on the grey and dull purple rocks of Llanddwyn Island (SH 386628) the majority of _rudis_ found had brilliant orange or yellow shells, and at Lligwy Bay (SH 497878) the rocks are deep red and all the _nigrolineata_ bright yellow (pers. obs.). Why some populations of a species are camouflaged and others are not, is not apparent.

A comparison of the shell colours that occur in _arcana_ and _rudis_ may also aid in discovering the significance of shell colour. These two species are commonly sympatric, yet the range of colours exhibited is quite different at some sites but not at others (Hannaford Ellis, 1979). If colour is selected through visual predation, as suggested by Heller (1975b) for _nigrolineata_ and _rudis_, the colour differences between these very similar-shelled species at some sites is therefore particularly intriguing. Furthermore, since the range of shell colours in these two species is often different, colour can be of use in identification, once the colour differences at a site are known. For example, at Abraham's Bosom (SH 215813) the species have no shell colours in common and so can be distinguished on this basis.

ECOLOGICAL ASPECTS.

Littorinids are one of the commonest molluscs of rocky shores and the genus is represented throughout the littoral zone. They play an important role in littoral food webs: Their herbivorous habit can influence the community structure of the algae (Castenholz, 1961) and they are a food source for many species of bird, fish, crab and mammal (Pettitt, 1975). Thus understanding winkle ecology is important to the comprehension of the littoral ecosystem. Such information could have wider applications; for example, in estimating the rate at which radio-active waste, disposed of in coastal waters, might be returned to the land (Odum, 1971).

Winkles live in a rigorous environment and have to be capable of surviving the extremes of physical conditions experienced in the intertidal zone, as well as the biological pressures exerted by other organisms. Apart from the predation pressure, they have also to compete for the resources of their habitat. For individual winkles competition pressure is likely to be greatest from other winkles, both of the same species and other species and how the effects of this pressure are reduced or avoided are critical questions in understanding their ecology.

The comparatively recent identification of the species of rough winkles has introduced an extra dimension into the problem of understanding the processes of rocky shore ecology. There is much to be learnt of the differences in biology and ecology of each species.

Various aspects of the biology of rough winkles were studied, notably a) dimorphism, b) ontogenetic changes in neglecta shell colour c) reproductive seasonality and d) the population composition and size structure changes of neglecta. In addition, since these species are commonly sympatric and appear to have similar requirements for shelter and food, how these resources are partitioned are important features of their ecology and some aspects of niche separation were examined. Spatial and temporal inter- and intraspecific zonation differences were found which could be important in resource partitioning. Each aspect of the ecology of the rough winkles that was investigated is discuussed below.

1) Dimorphism.

The observation of sexual dimorphism in all the rough winkle species parallels similar results recorded from flat winkles, by Sacchi (1968) Reimchen (1974) and Goodwin & Fish (1977). The common occurence of dimorphism in size in winkles (females growing larger than males) may be related to differences in male and female reproductive strategy. In neglecta, as a consequence of its apparently annual life cycle (section 2.5.) a difference in rate of maturation of the sexes can be seen; males reach maturity sooner than females. If males are diverting energy from growth to reproduction at an earlier age than females, this may be the underlying cause of dimorphism. This assumes that no unusual factors are involved, such as the sex of the progeny being related to parental age, with males being produced early in the season and females later. There may be equivalent differences in rate of maturation of males and females in the other species (which are perennial) and this may result in the observed dimorphism. An alternative possibility, that cannot be dismissed, is that males and females may differ in their efficiency of food utilisation.

In crevice-living species smaller size may be advantageous. Raffaelli & Hughes (1978) and Emson & Faller-Fritsch (1976) have shown that larger winkles are barred from taking advantage of the shelter of a greater proportion of crevices than smaller ones, because of their size and so, having a smaller shelter space they have a higher risk of mortality. Hence in being smaller, males may have a lower risk of mortality than females. For females the risk of larger size could be more than offset since larger females probably produce more offspring (Faller-Fritsch, 1977). Thus the dimorphism may be seen as the sexes occupying different positions in a size continuum, controlled by selection for increasing reproductive success and decreasing mortality. In addition dimorphism may reduce intra-specific competition through partitioning the resource of living space.

Dimorphism has an extra dimension in <u>nigrolineata</u> at Porth Swtan where there is also a dimorphic difference between the yellow- and white-shelled morphs; the rarer white morphs tending to be smaller and to mature at smaller shell heights. The co-existence of the morphs makes comparison of their biology particularly interesting as they are living in the same habitat, yet appear to show differences in their biology which seem to be mediated by their shell colour (section 2.2.4.). The smaller size of the white morph suggests that its females might be less fecund than those of the yellow morph. If so, it would be interesting to know how the white morph is maintained in the population. It is possible that lower fecundity may be offset if its smaller size gives it more possible sheltering spaces and so enables it to survive for longer which would effectively increase its reproductive life. Alternatively, selective predator pressure may prove to be the important factor, either through apostatic selection, in which predators adopt specific search images and tend to ignore other prey (the mechanism described by Clarke, 1962, for the origin and maintenance of distinct visual types in populations of Cepaea) or through crypsis, the white morph being possibly better camouflaged. Zaret (1972a & b) has shown how a less fit, small-eyed form of the cladoceran, Ceriodaphnia cornuta is maintained in the population by the disproportionate predation pressure of the fish Melaniris chagresi, which selectively predates the more fit, large-eyed form.

2) Ontogenetic change in the shell colour of L.neglecta.

An ontogenetic change in the shell colour pattern of neglecta was found, Juvenile neglecta often have banded patterns and adults tessellated patterns. No comparable change was seen in any of the other rough winkles though shell colour was occasionally seen to have altered from the older to the younger parts of the shell in other species (pers. obs.). Reimchen (1974) also noted that in mariae, juveniles in a population were often white whilst adults were either the citrina or reticulata colouring and so suggested that ontogenetic changes may occur in this species. For neglecta the significance of the change in pattern is not readily apparent. One possibility is that it aids in predator avoidance, but there is no evidence to support this suggestion.

3) Reproductive seasonality.

Very marked seasonal cycles of reproduction were seen in neglecta, nigrolineata and arcana (section 2.4.) Interestingly, although there were indications of a cycle in rudis this was less marked than that observed by other authors (Bergerard, 1971a & b, Berry, 1961, James, 1968). Possibly there may be differences in the expression of a reproductive cycle from population to population, which would be worth exploring. The well-defined cycles of the other species can have important implications for their ecology. The different timings of reproductive effort in nigrolineata and neglecta, species that co-occur in the barnacle belt, contribute to reducing interspecific competition between juveniles for food and living space (section 3.3.).

4) Population composition and size structure changes in L.neglecta.

The data obtained, though limited by only being collected for a one year period, suggested that neglecta may have an annual life cycle with most of the animals reaching maturity, breeding in the summer months and then disappearing. This contrasts with rudis which is known to be iteroparous, i.e. capable of living and breeding over several years (Bergerard, 1971a &b). If, as the data suggests, the adult neglecta disappear from the population, it is possible that this species is semelparous, i.e. the adults invest so much energy in reproduction that they cannot survive. Raffaelli (1978a) has suggested that neglecta evolved from a rudis-type species. If this is so, then these observations suggest that the life history tactics have also diverged.

5) Zonation differences.

The basic vertical zonation pattern of rough winkles, with arcana and rudis in the Pelvetia-Verrucaria belt (or equivalent high zones on more sheltered beaches) and nigrolineata and neglecta in the barnacle-fucoid (lower) zones, is complicated by factors such as a) the seasonal migration of arcana adults (see section 3.2.), b) a vertical zonation difference between neglecta and juvenile nigrolineata (see section 3.3.) and c) temporal effects in nigrolineata and neglecta juveniles (see section 3.3.). Indeed it is probable that these are only part of the niche differences between these related sympatric species.

For rudis and arcana the zonational patterns of the adults only were compared and they were found to be sympatric except during the arcana breeding season when mature arcana migrate down the beach. The sympatry and similarities of size and radula of these two species suggest that they are competing for food and space resources. However, this is not necessarily the case. The work of Emson & Faller-Fritsch (1977) showed that crevices, and not food, were limiting the rudis population at Newhaven. Since the study rock at Porth Swtan is crevice-rich, there may be sufficient crevice and food resources to support both adult populations, if their population sizes were independently controlled at the juvenile stage. This would occur if juveniles of the two species zoned differently and so encountered different selective forces. Such a zonation difference is possible and is made more likely by the observed segregation of the breeding females. Hence the zonation of juveniles could be critically important in the ecology and niche separation of these species.

INTRASPECIFIC VARIATION.

The general occurence of different forms in winkle species is of note. Particular attention has been paid here to two forms of rudis that were common in Anglesey, one living in boulder habitats, the other in crevice habitats. L.rudis however, exhibits many more types (some of which have been given variety names by S. Smith, 1978). Comparable variation can be seen in other species, e.g. the dwarf and normal forms of mariae described by Reimchen (1974), crevice- and boulder-living types of nigrolineata (Naylor, 1978), and crevice- and boulder-living types of arcana (pers. obs.). The hypothesis generally put forward by previous authors (the above, and also Faller-Fritsch,1977, Raffaelli & Hughes, 1978) is that these forms are the result of localised adaptation of the genome resulting from different selective pressures in the different habitats. However, this is still to be critically tested. It is possible that the differences may have no genetic basis or that a complex of sibling species are still being confused.

The existence of different forms in different habitats makes understanding the ecology of the winkle species even more complex, since what may be true of one form will not necessarily apply to another. Some nominal recognition of the different forms would be useful as an aid in ecology, although much more work is needed on the basis of the variation before any taxonomic recognition of the different forms can be contemplated.

ACKNOWLEDGEMENTS

There are a great many people whom I would like to thank for helping me to produce this thesis. First and foremost of them is my supervisor, Professor A.J.Cain. To him I owe the inspiration for this project and thanks for his stimulating conversations and his help with drafting this work.

I would also like to express my appreciation of the technical help given by Mr.K. Veltkamp (scanning electron microscopy), Mr.F. Schaeffer (histology), Mr.P. Chell and Mr. G. Green (macrophotography), Mr.J. Smith and Dr. J. Chubb (microphotography) and Dr. M. Nicholls (statistics).

There were many people who helped me with encouragement, suggestions and discussion, Dr.M. Begon, Mr.R. Cowie, Dr.R. Faller-Fritsch, Dr.V. Fretter, Dr.P. Gill, Mrs.P. Hargreaves, Dr.P. Mordan, Dr.J.D. Taylor, Prof. Sir J.E.S. Smith, Dr.S.Smith, and Dr. T. Warwick, to name but a few. I would also like particularly to thank Dr. M.V. Angel for his assistance and constructive criticism, Mr.A. Madgwick for his help in producing the colour plates, Mrs.P. Talbot who typed part of this thesis and my husband, whose patience, encouragement and practical help as chauffeur and winkle-picker played a major role.

This work was sponsored by a N.E.R.C. research studentship.

APPENDIX A

TABLE 1: Number and arrangement of penial glands in <u>L.rudis</u>

Site	Yuys Wellt				Bell Rock				Porth Swtan				Abraham's Bosom				Dunbar				Overall totals			
Penial gland arrangement	S	I	D	T	S	I	D	T	S	I	D	T	S	I	D	T	S	I	D	T	S	I	D	T
Penial gland number																								
4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	1	1	-	-	1
5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	-	-	-	-	2	-	-	2	1	-	-	1	1	-	-	1	-	-	-	-	4	-	-	4
7	2	-	-	2	-	-	-	-	4	-	-	4	3	1	-	4	-	-	-	-	9	1	-	10
8	1	-	-	1	1	-	-	1	10	-	-	10	-	-	-	-	2	-	-	2	14	-	-	14
9	5	-	-	5	5	2	-	7	28	2	-	30	7	1	-	8	1	-	-	1	46	5	-	51
10	5	1	-	6	10	2	-	12	55	9	-	64	5	1	-	6	2	-	-	2	77	13	-	90
11	8	-	-	8	9	7	-	16	57	21	-	78	3	4	-	7	3	3	-	6	80	35	-	115
12	4	2	-	6	10	3	-	13	43	39	-	82	4	2	-	6	2	4	-	6	63	50	-	113
13	4	2	-	6	1	2	-	3	36	46	-	82	1	7	-	8	5	3	-	8	47	60	-	107
14	1	2	-	3	1	2	-	3	25	38	-	63	-	5	-	5	-	-	-	-	27	47	-	74
15	-	1	-	1	-	3	-	3	11	51	-	62	-	2	-	2	-	4	-	4	11	61	-	72
16	-	1	-	1	-	2	-	2	5	24	1	30	-	-	-	-	1	3	-	4	6	30	1	37
17	-	1	-	1	-	1	-	-	1	24	1	26	-	4	-	4	-	2	-	2	1	32	1	34
18	-	-	-	-	-	-	-	-	-	5	1	6	-	1	-	1	-	3	-	3	-	9	1	10
19	-	-	-	-	-	-	-	-	-	8	-	8	-	-	-	-	-	2	-	2	-	10	-	10
20	-	-	-	-	-	-	-	-	-	2	1	3	-	-	-	-	-	2	-	2	-	4	1	5
21	-	-	-	-	-	-	-	-	-	3	1	4	-	-	-	-	-	1	2	3	-	4	3	7
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	1	-	1	-	1
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
26	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	1	-	-	1	1
	30	10	-	40	39	24	-	63	276	272	5	553	24	28	-	52	17	28	3	48	386	362	8	756

Penial gland arrangements:- S= single row
I= irregular row
D= double row

T= total

APPENDIX A

TABLE 2: Number and arrangement of penial glands in L.arcana

Site	Ynys Wellt				Bell Rock				Porth Swtan				Abraham's Bosom				Dunbar				Overall totals			
Penial gland arrangement	S	I	D	T	S	I	D	T	S	I	D	T	S	I	D	T	S	I	D	T	S	I	D	T
Penial gland number																								
4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	-	-	-	-	-	-	-	-	1	-	-	1	-	-	-	-	-	-	-	-	1	-	-	1
7	-	-	-	-	-	-	-	-	1	-	-	1	-	-	-	-	-	1	-	1	1	1	-	2
8	-	-	-	-	1	-	-	1	1	-	-	1	-	-	-	-	-	-	-	-	2	-	-	2
9	-	-	-	-	1	1	-	2	6	1	-	7	-	-	-	-	-	-	-	-	7	2	-	9
10	4	-	-	4	2	1	-	3	6	5	-	11	4	-	-	4	2	-	-	2	18	6	-	24
11	3	1	-	4	3	1	-	4	12	4	-	16	2	-	-	2	3	4	-	7	23	10	-	33
12	2	4	-	6	2	6	-	8	17	19	1	37	3	4	-	7	1	9	-	10	25	42	1	68
13	1	3	-	4	3	3	-	6	12	30	1	43	1	2	-	3	-	4	-	4	17	42	1	60
14	-	6	1	7	-	8	1	9	7	38	1	46	-	7	-	7	-	5	-	5	7	64	3	74
15	2	5	-	7	1	6	-	7	6	51	2	59	-	8	-	8	-	2	2	4	9	72	4	85
16	-	4	1	5	1	4	1	6	7	49	5	61	1	1	-	2	-	3	1	4	9	61	8	78
17	-	1	-	1	-	7	-	7	6	45	6	57	-	8	-	8	-	3	1	4	6	64	7	77
18	-	3	2	5	-	3	1	4	-	33	2	35	-	6	-	6	-	1	1	2	-	46	6	52
19	-	4	1	5	-	4	-	4	-	25	7	32	-	3	2	5	-	2	1	3	-	38	11	49
20	-	3	-	3	1	1	1	3	-	10	4	14	-	1	1	2	-	-	-	-	1	15	6	22
21	-	2	1	3	-	2	1	3	-	8	3	11	-	-	1	1	-	1	-	1	-	13	6	19
22	-	2	1	3	-	3	-	3	-	8	3	11	-	1	-	1	-	-	-	-	-	14	4	18
23	-	-	-	-	-	2	1	3	-	2	5	7	-	-	1	1	-	-	1	1	-	4	8	12
24	-	-	2	2	-	1	-	1	-	3	4	7	-	-	1	1	-	-	2	2	-	4	9	13
25	-	-	-	-	-	-	1	1	-	1	2	3	-	-	-	-	-	-	-	-	-	1	3	4
26	-	-	-	-	-	1	1	2	-	1	3	4	-	-	-	-	-	-	-	-	-	2	4	6
27	-	-	-	-	-	-	-	-	-	-	1	1	-	-	1	1	-	-	-	-	-	-	2	2
28	-	-	-	-	-	-	-	-	-	-	2	2	-	-	-	-	-	-	1	1	-	-	3	3
29	-	-	-	-	-	-	-	-	-	-	1	1	-	-	-	-	-	-	-	-	-	-	1	1
30	-	-	-	-	-	2	-	2	-	-	2	2	-	-	-	-	-	-	-	-	-	2	2	4
31	-	-	-	-	-	1	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	1
32	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	1	-	-	-	-	-	-	1	1
33	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
34	-	-	-	-	-	1	-	1	-	-	-	-	-	-	1	1	-	-	1	1	-	1	2	3
	12	38	9	59	15	58	8	81	82	333	55	470	11	41	9	61	6	35	11	52	126	505	92	723

Penial gland arrangements:- S= single row
I= irregular row
D= double row

T= total

APPENDIX A

TABLE 3: Ontogenetic decline in cusp number

L. arcana		L. rudis		L. nigro-lineata		L. neglecta		L. neritoides		L. littorea		L. obtusata					L. mariae				
SHT	M	SHT	M	SHT	M	SHT	M	SHT	M	SHT	M	SHT	R	L	IM	M	SHT	R	L	IM	M
11.95	6	10.4	5	16.6	5	3.0	7	7.0	5	30.8	5	14.6	3	5	5	5	10.7	3	4	5	6
11.85	5	14.2	5	15.65	5	3.0	7	6.5	7	24.5	5	13.6	3	4	5	6	10.35	3	4	6	7
11.1	5	14.2	5/6	15.6	5	2.85	7	6.4	7	24.05	5	13.2	5	4	5	6	9.7	3	4	5	6
10.9	6	13.2	5	15.55	5	2.8	6	6.35	6	22.05	5	12.55	3	4	5	6	9.3	3	4	5	7
10.7	5	12.45	5	15.2	5	2.6	7	6.15	5/7	19.55	4	12.4	3	4	5	6	8.8	3	4	5	7
10.55	5	11.2	5	14.7	5	2.6	6	6.0	6	16.95	5	12.25	3	4/5	5	6	8.2	3	4	5	8
9.65	5	11.15	5	13.15	5	2.5	6	5.9	6	16.75	5	11.8	3	5	5	7	7.65	3	4	5	6
9.55	6/5	10.4	5	13.1	5	2.5	6	5.9	7	15.5	5	11.6	3	5	5	6	7.55	3	4	5	7
9.2	5	9.15	5	12.25	6	2.45	6	5.7	7	14.5	5	11.5	3	4	5	6	6.9	3	4	5	7
9.15	5	8.85	5	11.45	5	2.4	7	5.45	7	14.1	5	11.5	3	4	5	6	6.7	3	4	5	7/8
9.05	5	8.5	5/6	11.25	6	2.4	7	5.4	7	13.3	4	11.35	3	4	5	5	5.7	3	4	5	8
8.95	7	8.2	6	11.25	5	2.3	7	5.35	7	13.15	5	11.2	3	4	5	6	5.35	5	4/5	6	7
8.7	6	7.4	5	11.0	6	2.2	7	5.2	7	10.5	6	11.05	3	4/5	5	6	4.45	3	5	6	9
8.65	6	5.15	6	11.0	5	1.9	8	5.2	6	10.0	5/6	9.0	3	5	5	7	4.0	3	5	6	8
8.45	6	4.95	5	11.0	5/6	1.85	7	5.1	6	9.7	5	8.25	3	5	5	6	3.75	5	5	6	8
8.4	6	4.2	6	11.0	5	1.7	8	4.9	7	8.6	5	6.9	3	5	5	7	3.7	3	6	6	8/9
8.3	6	4.1	7	10.5	5	1.7	7	4.75	8	7.5	5/6	6.5	3	4	5	7	3.55	5	5	7	9
8.25	6	2.3	7/8	8.55	6	1.3	8	4.7	7	6.2	6	4.95	3	4	5	7	3.1	5	4	6	8
8.1	6	1.5	8	8.1	6/7	1.1	7	4.2	8	5.8	6/7	4.85	3	6	5	8					
7.9	7	1.1	7	7.45	6	.85	8	3.8	7	5.65	6/7	4.65	3	4	5	8					
7.75	5			6.7	6			3.5	7	4.1	6	3.5	3	4	5	8					
7.65	6			6.6	6			3.1	8	2.4	6	3.2	3	5	6	8					
7.55	6			6.0	6			2.75	8	2.4	7	2.9	5	6	6	10					
7.5	6			5.35	6			2.5	8			2.0	3	5	5	8					
7.45	6			4.6	6			2.45	7			1.85	3	6	6	9					
7.4	6			4.45	6			2.3	8			1.5	3	5	5	9					
7.3	7			4.3	6			2.25	8												
7.25	7			4.25	7			2.1	7												
7.25	6/5			3.5	7			2.0	8												
6.75	6			3.45	7			2.0	8												
6.45	6			3.35	6																
5.85	8			3.05	7																
5.85	6			3.0	6																
5.7	6			2.9	7																
5.4	7			2.8	8																
5.35	7			2.7	7																
5.3	6			2.7	7																
3.8	8			2.3	7																
3.7	7			2.2	8																
3.45	9			2.1	7																
3.1	8			2.0	7																
2.5	7			1.8	7																
2.0	6			1.3	8																
1.7	7/8			1.2	6																
1.3	8			1.0	7																
1.2	10																				

NOTE:- SHT = shell height in mm.
M = outer marginal tooth cusp number
IM = inner marginal tooth cusp number
R = rachidian tooth cusp number
L = lateral tooth cusp number

APPENDIX B

TABLE 1 SEXUAL DIMORPHISM IN SHELL HEIGHT

Shell Height	L. rudis C				L. rudis B				L. arcana				L. nigrolineata			
Class (mm)	♂ (a)	♀	Total (n)	$\frac{a^2}{n}$	♂ (a)	♀	Total (n)	$\frac{a^2}{n}$	♂ (a)	♀	Total (n)	$\frac{a^2}{n}$	♂ (a)	♀	Total (n)	$\frac{a^2}{n}$
6.5-6.95	325	309	634	166.6	-	-			149	175	324	68.52	131	106	237	72.41
7.0-7.45	322	413	735	141.07	-	6	21	1.72	179	237	416	77.02	120	123	243	59.26
7.5-7.95	339	369	708	162.32	6	9			190	228	418	86.36	105	120	225	49.0
8.0-8.45	251	306	557	113.11	8	12	20	3.2	151	206	356	63.87	106	92	198	56.75
8.5-8.95	155	211	366	65.64	15	13	28	8.04	117	179	296	46.25	92	107	199	42.53
9.0-9.45	80	97	177	36.16	17	22	39	7.41	63	142	205	19.36	116	96	212	63.47
9.5-9.95	38	39	77	18.75	22	16	38	12.74	32	87	119	8.61	89	97	186	42.59
10.0-10.45	15	23	38	5.92	28	22	50	15.68	12	68	80	1.8	93	108	201	43.03
10.5-10.95	8	8	16	4.0	31	36	67	14.34	2	22	32	.28	82	87	169	39.79
11.0-11.45					48	36	84	27.43	1	7			55	93	148	20.44
11.5-11.95					42	50	92	19.17					78	82	160	38.02
12.0-12.45					45	61	106	19.10					51	68	119	21.86
12.5-12.95					58	72	130	25.88					44	43	87	22.25
13.0-13.45					63	80	143	27.76					35	52	87	14.08
13.5-13.95					50	63	113	22.12					34	33	67	17.25
14.0-14.45					41	65	106	15.86					22	20	42	11.52
14.5-14.95					26	41	67	10.09					20	21	41	9.76
15.0-15.45					10	36	46	2.17					9	18	27	3.0
15.5-15.95					8	23	31	2.06					2	10	12	.33
16.0-16.45					9	17	26	3.12					8	9	24	4.17
>16.5					2	18	20	.20					2	5		
TOTALS	1533	1775	3308	713.57	529	698	1227	238.09	896	1351	2247	372.07	1294	1390	2684	631.51
$\frac{A^2}{N}$				710.43				228.07				357.28				623.86
χ^2	$\frac{3.14}{.248031}$	= 12.63			$\frac{10.02}{.245239}$	= 40.86			$\frac{14.79}{.239799}$	= 61.68			$\frac{7.65}{.249676}$	= 30.64		
Degress of Freedom	8			N.S.	18		sig. at .01		8			sig. at .001	19		sig. at .05	

Shell Height	L. neglecta			
Class (g.u.)	♂ (a)	♀	Total (n)	$\frac{a^2}{n}$
2.0-2.4	64	38	102	40.16
2.5-2.9	150	107	257	87.55
3.0-3.4	177	160	337	92.96
3.5-3.9	53	143	196	14.33
4.0-4.4	11	92	103	1.17
>4.5	-	25	25	0
TOTALS	455	565	1020	236.17
$\frac{A^2}{N}$				202.97

$$\chi^2 = \frac{33.2}{.247084} = 134.37$$

d of F = 5
sig. at .001

NOTE: N.S. = not significant
sig. = significant

APPENDIX B

TABLE 2 SEXUAL DIMORPHISM: - SHELL HEIGHT AT MATURITY

Shell Height	L. rudis C				L. rudis B				L. arcana				L. nigrolineata			
Size Class (mm)	Mature ♂ (a)	Mature ♀	Total (n)	$\frac{a^2}{n}$	Mature ♂ (a)	Mature ♀	Total (n)	$\frac{a^2}{n}$	Mature ♂ (a)	Mature ♀	Total (n)	$\frac{a^2}{n}$	Mature ♂ (a)	Mature ♀	Total (n)	$\frac{a^2}{n}$
6.5-6.95	186	93	279	124.0	-	-			78	46	124	49.06	19	5	24	15.04
7.0-7.45	256	236	492	133.2	-	-			107	101	208	55.04	22	12	34	14.24
7.5-7.95	303	307	610	150.51	-	-			125	111	236	66.21	21	12	33	13.26
8.0-8.45	226	256	482	105.97	1	-			99	117	216	45.37	24	11	35	16.46
8.5-8.95	145	190	335	62.76	2	2	24	7.04	81	110	191	34.35	30	27	57	15.79
9.0-9.45	74	90	164	33.39	-	1			44	86	130	14.89	44	27	71	27.27
9.5-9.95	33	38	71	15.34	3	1			22	57	79	6.13	29	30	59	14.25
10.0-10.45	14	21	35	5.6	7	7			7	46	53	.92	38	31	69	20.93
10.5-10.95	8	7	24	7.04	15	11	26	8.65	2	18	26	.15	41	33	74	22.72
11.0-11.45	5	4			30	23	53	16.98	-	6			28	42	70	11.2
11.5-11.95					31	30	61	15.75					54	43	97	30.06
12.0-12.45					37	44	81	16.90					38	41	79	18.28
12.5-12.95					51	52	103	25.25					32	25	57	17.96
13.0-13.45					56	71	127	24.69					29	37	66	12.74
13.5-13.95					44	56	100	19.36					29	22	51	16.49
14.0-14.45					39	60	99	15.36					20	18	38	10.53
14.5-14.95					25	39	64	9.77					18	18	36	9.0
15.0-15.45					10	35	45	2.22					8	15	23	2.78
15.5-15.95					8	23	31	2.06					2	10	12	.33
16.0-16.45					9	16	25	3.24					8	8	23	4.35
>16.5					2	18	20	.20					2	5		
TOTAL	1242	1250	2492	637.81	370	489	859	167.47	565	698	1263	272.12	536	472	1008	293.78
$\frac{A^2}{N}$				627.01				159.37				252.75				285.02
χ^2	$= \frac{10.8}{.249996} = 43.20$				$\frac{8.1}{.245239} = 33.03$				$\frac{19.37}{.247191} = 78.36$				$\frac{8.76}{.244764} = 35.79$			
Degress of Freedom	8	sig .001			13	sig .01			8	sig .001			19	sig .05		

Shell Height Class (g.u.)	L. neglecta Mature ♂ (a)	Mature ♀	Total (n)	$\frac{a^2}{n}$
2.0-2.4	18	2	20	16.2
2.5-2.9	92	12	104	81.38
3.0-3.4	156	75	231	105.35
3.5-3.9	49	116	165	14.55
4.0-4.4	11	87	98	1.23
>4.5	-	21	21	0

$$\chi^2 = \frac{52.39}{.2499} = 209.65$$

d of F = 5
sig .001

APPENDIX B

TABLE 3

Species	Shell Height Range		Mean $\bar{x}$	SE	n
L. rudis C	7-7.95 mm	♂	7.41	.035	71
		♀	7.45	.039	66
L. rudis B	11-12.95mm	♂	11.94	.072	70
		♀	12.12	.063	75
L. arcana	7-7.95 mm	♂	7.42	.032	69
		♀	7.44	.033	70
L. nigrolineata	8-12.95mm	♂	10.36	.219	59
		♀	10.33	.155	58
L. neglecta	2.5-3.5gu	♂	2.98	.073	85
		♀	3.08	.075	59

APPENDIX B

TABLE 4

Size Class µm	Yellow (a)	White	Total (n)	$\frac{a^2}{n}$
6.5-6.95	237	35	272	206.50
7.0-7.45	243	40	283	208.65
7.5-7.95	225	33	258	196.22
8.0-8.45	198	22	220	178.20
8.5-8.95	199	18	217	182.49
9.0-9.45	212	19	231	194.56
9.5-9.95	186	12	198	174.73
10.0-10.45	201	13	214	188.79
10.5-10.95	169	7	176	162.28
11.0-11.45	148	5	153	143.16
11.5-11.95	160	5	165	155.15
12.0-12.45	119	3	122	116.07
12.5-12.95	87	2	89	85.04
13.0-13.45	87	3	90	84.1
13.5-13.95	67	-	67	67.0
14.0-14.45	42	1	151	141.17
14.5-14.95	41	-		
15.0-15.45	27	2		
15.5-15.95	12	2		
16.0-16.45	17	-		
≥16.5	7	-		
TOTALS	2684	222	2906	2484.11
$\frac{A^2}{N}$				2478.96

$$^2 = \frac{5.15}{.070224} = 73.34$$

d of F = 15

sig. at .001

APPENDIX B

TABLE 5 Porth Swtan (Date of sample collection: 13.8.77)

Size Class	Number in Pattern Class			Total in	% in Size Class		
(g.u.)	1	2	3	Size Class	1	2	3
1.5-1.9	52	5	-	57	91	9	-
2.0-2.4	50	20	1	71	70	28	1
2.5-2.9	28	39	7	74	38	53	9
3.0-3.4	38	47	19	104	37	45	18
3.5-3.9	17	39	19	75	23	52	25
4.0-4.4	6	4	6	16			
4.5-4.9	-	1	2	3			

TABLE 6 Porth Diana (Date of sample collection 17.7.78)

Size Class	Number in Pattern Class			Total in	% in Size Class		
(g.u.)	1	2	3	Size Class	1	2	3
1.5-1.9	4	-	-	4			
2.0-2.4	18	2	-	20	90	10	-
2.5-2.9	14	7	1	22	64	32	5
3.0-3.4	11	16	13	40	27.5	40	32.5
3.5-3.9	6	25	14	45	13	56	31
4.0-4.4	2	28	32	62	3	45	52
4.5-4.9	1	7	29	37	3	19	78
5.0-5.4	-	4	13	17			
5.5-5.9	-	-	3	3			
6.0-6.4	-	1	1	2			

APPENDIX B

TABLE 7 % MATURE OF TOTAL IN SIZE CLASS (DATA AMALGAMATED FROM SAMPLES COVERING YEAR APRIL 1977 TO MARCH 1978)

Shell Height Size Class (mm)	L. rudis B				L. rudis C				L. arcana				L. nigrolineata			
	I	M	T	%M	I	M	T	%M	I	M	T	%M	I	M	T	%M
6.5-6.95	-	-	-		352	280	632	44	200	124	324	38	159	12	171	7
7.0-7.45	6	-	6		240	489	729	67	209	208	417	50	155	21	176	12
7.5-7.95	15	-	15		94	614	708	87	180	238	418	57	146	24	170	14
8.0-8.45	19	1	20	5	70	469	539	87	141	216	357	61	102	21	123	17
8.5-8.95	24	4	28	4	30	334	364	92	106	190	296	64	84	29	113	26
9.0-9.45	38	1	39	3	13	164	177	93	74	132	216	61	79	52	131	40
9.5-9.95	34	4	38	11	6	69	75	92	40	79	119	66	84	42	126	33
10.0-10.45	36	14	50	28	3	30	33	91	27	53	80	66	80	45	125	36
10.5-10.95	41	26	67	39	1	16	17		4	20	24	83	60	48	108	44
11.0-11.45	31	53	84	63	1	9	10		2	6	8		57	47	104	45
11.5-11.95	31	61	92	66	-	2	2						47	66	113	58
12.0-12.45	25	81	106	76									28	57	85	67
12.5-12.95	27	103	130	79									24	43	67	64
13.0-13.45	16	127	143	89									15	49	64	77
13.5-13.95	13	100	113	88									11	39	50	78
14.0-14.45	7	99	106	93									2	31	33	94
14.5-14.95	3	64	67	96									2	27	29	93
15.0-15.45	1	45	46	98									3	17	20	85
15.5-15.95	-	31	31	100									-	9	9	
16.0-16.45	1	25	26	96									1	11	12	
16.5-16.95	-	9	9										-	2	2	
17.0-17.45	-	5	5										-	1	1	
17.5-17.95	-	4	4													
18.0-18.45	-	2	2													

Shell Height Size Class (g.u.)	L. neglecta			
	I	M	T	%M
2.0-2.4	82	19	101	19
2.5-2.9	132	109	241	45
3.0-3.4	86	207	293	71
3.5-3.9	26	139	165	84
4.0-4.4	4	85	89	96
4.5-4.9	4	18	22	82
5.0	-	2	2	

KEY
I = Immature
M = Mature
T = Total
%M = Percentage mature ($\frac{M}{T}$ x 100)

APPENDIX B

TABLE 8 REPRODUCTIVE PERIODICITY: % MATURE IN SUCCESSIVE MONTHS

Date of Collection	L. rudis B			L. rudis C			L. arcana			L. nigrolineata			L. neglecta		
	M	I	%M	M	I	%M	M	I	%M	M	I	%M	M	I	%M
25th June 1976										13	33	28			
11th July										25	62	29			
9th August										40	16	71			
5th September										46	11	81			
3rd October										20	15	57			
1st November										10	8	56			
6th December										12	14	46			
16th Jan 1977										8	25	36			
12th February										13	42	31	42	19	69
19th March	74	42	64	176	45	80	68	61	53	18	75	19	45	3	94
16th April	38	16	73	159	43	78	56	71	44	10	61	14	89	12	88
14th May	32	25	56	180	33	85	38	114	25	16	87	16	84	13	87
12th June	38	14	73	186	49	79	9	111	7.5	17	59	22	53	13	80
10th July	33	21	61	145	37	80	2	63	3	65	45	59	40	9	82
13th August	62	16	79	156	39	80	30	77	39	115	32	78	33	11	75
10th September	34	11	76	171	40	81	170	44	79	53	12	82	15	8	65
11th October	100	10	91	168	35	83	177	23	88	33	42	44	25	61	29
12th November	68	22	76	151	37	80	111	42	73	52	65	44	50	57	47
10th December	52	5	91	178	43	81	83	49	63	20	43	32	45	47	49
7th Jan 1978	63	13	83	155	47	77	76	49	61	22	35	39	38	45	46
4th February	47	8	85	91	34	73	31	27	53	12	37	24	48	34	59
5th March										5	38	12	59	24	71

in chosen shell
height range
(see p. 66)

M = total ♂ + ♀ mature
I = total ♂ + ♀ immature
%M = percentage mature

APPENDIX B

TABLE 9

PL / SHT	*L. rudis* C Month of Sample '77										'78	
Ratio Size Class	M	A	M	J	J	A	S	O	N	D	J	F
.0-.09	-	1	-	-	2	-	-	-	-	-	-	2
.1-.19	3	3	-	4	3	-	2	1	-	-	3	4
.2-.29	4	2	2	2	6	2	2	2	2	2	2	5
.3-.39	5	6	1	5	1	-	1	2	-	2	-	1
.4-.49	-	5	10	1	3	3	2	2	1	3	-	-
.5-.59	2	3	-	5	3	3	2	3	2	3	4	-
.6-.69	7	4	2	20	7	9	1	4	8	7	3	2
.7-.79	31	8	21	37	29	15	14	12	12	14	14	6
.8-.89	33	37	31	23	23	39	35	42	28	31	24	18
.9-.99	12	12	31	14	12	19	29	24	24	29	24	19
1.0-1.09	2	5	5	-	1	1	3	2	10	4	8	4
1.1-1.19	-	-	1	-	-	-	3	-	2	-	3	1
1.2-1.29	-	-	-	-	-	1	-	-	-	-	-	-
	L. rudis B											
.0-.19	7	1	3	4	1	1	1	3	1	1	3	2
.2-.39	8	3	10	-	2	1	-	-	2	1	2	1
.4-.59	11	5	6	5	4	7	2	2	4	4	5	2
.6-.79	16	1	8	7	13	18	9	13	13	12	10	10
.8-.99	17	7	2	6	5	8	4	34	19	11	14	11
1.0	-	2	-	2	-	6	-	1	1	-	-	2
	L. arcana											
.0-.09	13	14	25	27	5	-	-	-	-	1	-	-
.1-.19	10	11	20	19	8	-	-	1	4	3	7	1
.2-.29	2	4	14	4	10	2	2	-	-	-	2	1
.3-.39	2	-	1	-	4	7	3	-	1	1	2	1
.4-4.9	2	3	1	1	1	13	5	1	2	1	-	-
.5-.59	5	2	5	-	-	17	18	10	3	4	2	1
.6-.69	17	17	3	-	-	10	52	41	14	16	22	5
.7-.79	8	6	3	2	-	1	27	45	20	20	15	5
.8-.89	2	1	1	-	-	-	2	2	4	2	-	-
.9-.99	-	1	-	-	-	-	-	-	-	-	-	-

C + W / SHT	*L. rudis* C Month of Sample '77										'78	
Ratio Size Class	M	A	M	J	J	A	S	O	N	D	J	F
.4-.49	1	-	-	1	-	-	1	-	1	1	1	-
.5-.59	4	6	1	1	1	-	2	2	3	2	2	2
.6-.69	9	5	6	3	2	5	3	4	1	4	6	4
.7-.79	8	8	5	8	5	10	5	5	6	14	5	6
.8-.89	9	2	5	10	4	6	9	10	9	8	7	4
.9-.99	3	2	1	6	3	6	7	6	9	3	7	3
1.0-1.09	5	7	1	2	1	3	3	2	2	5	6	4
1.1-1.19	8	7	8	9	3	6	3	6	6	4	6	3
1.2-1.29	13	6	6	12	4	7	14	7	7	11	12	3
1.3-1.39	15	21	18	15	8	10	9	12	15	12	14	8
1.4-1.49	18	20	21	18	18	14	12	16	9	22	14	4
1.5-1.59	15	20	16	19	20	12	15	14	15	19	13	12
1.6-1.69	10	8	15	14	15	9	12	12	8	8	8	5
1.7-1.79	4	1	3	4	4	8	10	4	3	8	8	2
1.8-1.89	1	3	1	2	2	5	7	4	4	5	6	2
1.9-1.99	-	-	1	-	1	1	5	3	-	-	1	1
2.0-2.09	-	-	1	-	-	-	-	1	-	-	-	-
2.1-2.19	-	-	-	-	-	-	-	1	-	-	1	-
	L. rudis B											
.4-.59	-	-	-	-	1	1	2	2	-	-	-	-
.6-.79	13	6	5	3	6	6	4	3	3	-	2	-
.8-.99	7	6	2	2	7	2	4	6	10	3	5	3
1.0-1.19	5	2	-	2	1	8	-	3	6	1	3	3
1.2-1.39	10	7	1	2	6	9	5	14	6	9	9	5
1.4-1.59	16	8	4	9	4	10	8	14	16	11	19	9
1.6-1.79	5	3	3	8	2	1	4	7	6	4	4	7
1.8-1.99	1	2	2	2	2	-	1	4	3	-	-	-
2.0-2.19	-	1	1	-	-	-	1	2	-	-	-	-
	L. arcana											
.3-.39	3	7	3	2	1	-	-	-	-	-	-	-
.4-.49	15	15	20	13	6	3	1	-	4	5	1	1
.5-.59	12	6	15	22	12	8	3	4	9	8	9	7
.6-.69	4	3	4	16	11	14	3	6	9	12	13	12
.7-.79	1	1	2	2	4	15	7	3	6	8	14	3
.8-.89	-	5	3	1	2	6	3	1	4	5	-	1
.9-.99	-	5	6	3	1	7	5	2	3	4	1	-
1.0-1.09	5	7	7	6	-	3	11	6	5	5	3	2
1.1-1.19	6	8	4	1	-	1	24	15	13	9	10	5
1.2-1.29	7	7	5	1	-	-	22	24	32	8	10	7
1.3-1.39	9	3	6	-	-	-	16	26	16	14	6	1
1.4-1.49	3	1	1	-	-	-	8	8	4	4	8	4
1.5-1.59	-	-	2	-	-	-	2	4	-	2	-	1
1.6-1.69	1	-	1	-	-	-	-	1	-	-	-	-
1.7-1.79	2	-	-	-	-	-	-	-	-	-	-	-

APPENDIX B

TABLE 9 CONTD.

<u>L. nigrolineata</u> (yellow-shelled individuals only)

$\frac{C + W}{SHT}$	'76							'77												'78		
Ratio Size Class	J	J	A	S	O	N	D	J	F	M	A	M	J	J	A	S	O	N	D	J	F	M
.0-.09	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-	-	-	-
.1-.19	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-	-	-
.2-.29	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
.3-.39	-	2	-	1	-	1	1	3	4	7	9	6	4	2	3	2	5	4	-	1	7	1
.4-.49	3	10	1	2	4	-	2	9	11	24	17	22	11	4	7	3	7	23	14	10	17	8
.5-.59	7	18	6	1	5	-	1	4	4	12	6	12	6	9	5	-	11	8	13	9	1	14
.6-.69	3	4	1	1	1	-	-	1	2	1	-	2	3	6	3	1	2	2	2	2	-	3
.7-.79	5	1	-	-	1	-	-	-	-	-	-	3	3	6	2	-	2	-	3	1	-	1
.8-.89	2	3	2	3	-	1	1	1	3	1	2	-	1	9	17	1	-	2	-	1	2	1
.9-.99	-	2	5	3	3	1	3	2	3	-	-	1	2	5	18	3	7	3	3	3	1	1
1.0-1.09	1	4	7	6	5	2	3	2	2	-	-	2	2	5	17	5	1	8	3	4	1	-
1.1-1.19	-	3	4	7	3	-	2	1	-	2	-	3	-	1	3	9	2	8	-	1	1	1
1.2-1.29	3	-	2	4	-	-	-	-	-	2	-	2	-	-	1	4	-	-	1	-	-	-
1.3-1.39	-	-	-	2	-	-	-	-	-	1	-	-	-	-	-	1	-	-	-	-	-	-
1.4-1.49	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
$\frac{PL}{SHT}$																						
Ratio Size Class																						
.0-.09	-	-	1	-	1	-	1	-	1	-	1	-	1	-	1	-	-	2	2	-	3	3
.1-.19	4	13	-	2	2	2	6	3	11	10	10	6	11	1	3	1	10	10	5	9	7	4
.2-.29	5	3	3	1	1	4	2	4	7	14	12	9	5	2	4	3	4	11	5	3	2	3
.3-.39	2	2	1	1	-	-	1	-	1	3	2	12	6	6	3	-	2	5	2	-	-	2
.4-.49	2	3	-	1	-	1	-	-	-	2	3	2	3	5	1	2	-	-	-	1	-	1
.5-.59	2	2	-	1	-	-	-	-	1	2	1	1	4	11	6	-	1	-	-	-	1	-
.6-.69	3	5	6	1	-	-	-	-	-	3	-	1	4	9	12	1	-	1	-	3	2	1
.7-.79	3	5	4	5	3	-	-	2	2	2	-	4	4	17	28	12	5	12	3	4	1	2
.8-.89	-	6	7	8	3	4	3	1	3	4	5	1	3	9	11	14	9	12	7	4	1	-
.9-.99	1	1	6	4	1	2	-	-	-	3	3	2	1	1	1	3	6	5	-	1	1	-
1.0-1.09	-	-	-	2	2	-	-	-	-	-	-	1	-	2	-	-	-	1	-	-	1	-
1.1-1.19	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-	-	-
1.2-1.29	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
1.3-1.39	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-

APPENDIX B

TABLE 10 POPULATION SIZE CHANGE AND ECOLOGICAL AGE STRUCTURE

SIZE CLASS (g.u.)	1977									1978		
	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Feb	Mar
Mature ♀												
2.0-2.4	-	-	1	-	-	-	1	-	-	-	-	-
2.5-2.9	3	-	-	-	-	-	-	1	-	3	2	3
3.0-3.4	13	13	3	2	4	1	1	3	3	3	8	9
3.5-3.9	18	25	11	13	13	1	1	1	4	1	4	8
4.0-4.4	15	12	13	14	3	6	4	3	2	3	2	2
4.5-4.9	5	5	3	1	-	2	-	-	2	-	-	-
5.0-5.4	-	-	2	-	-	-	-	-	-	-	-	-
Immature ♀												
2.0-2.4	-	2	-	-	-	-	4	14	6	4	1	3
2.5-2.9	2	3	4	1	1	2	8	18	17	19	6	7
3.0-3.4	6	3	1	2	1	3	8	10	13	10	15	10
3.5-3.9	2	1	-	2	1	-	7	1	1	2	7	1
4.0-4.4	-	-	1	1	1	-	1	-	-	-	-	-
4.5-4.9	-	-	1	-	-	-	1	-	2	-	-	-
Juveniles												
2.0-2.4	2	-	2	7	10	8	17	7	-	4	-	2
2.5-2.9	1	2	1	2	-	4	4	1	2	-	2	-
3.0-3.4	-	-	-	2	-	1	1	-	-	-	-	-
Immature ♂												
2.0-2.4	-	1	1	2	3	1	21	10	3	4	1	1
2.5-2.9	2	3	4	1	3	1	16	3	3	4	2	2
3.0-3.4	-	-	1	-	1	1	-	-	2	2	2	-
3.5-3.9	-	-	-	-	-	-	-	1	-	-	-	-
Mature ♂												
2.0-2.4	1	-	-	-	-	1	1	9	2	2	1	-
2.5-2.9	7	3	1	3	-	1	11	18	18	8	15	12
3.0-3.4	18	15	15	6	6	3	6	14	10	17	13	21
3.5-3.9	7	11	2	1	6	-	-	1	3	1	3	4
4.0-4.4	2	-	2	-	1	-	-	-	1	-	-	-
Overall total in size class												
2.0-2.4	3	3	4	9	13	10	44	40	11	14	3	6
2.5-2.9	15	11	10	7	4	8	39	41	40	34	27	24
3.0-3.4	37	31	20	12	12	9	11	27	28	32	38	40
3.5-3.9	27	37	13	16	20	1	8	4	8	4	14	13
4.0-4.4	17	12	16	15	5	6	5	3	3	3	2	2
4.5-4.9	5	5	4	1	-	2	1	-	4	-	-	-
5.0-5.4	-	-	2	-	-	-	-	-	-	-	-	-

APPENDIX C

TABLE 1. Average zonation patterns of the subpopulations of *L. rudis* C and *L. arcana*.

Transect	*L. rudis* C				*L. arcana*			
zone	♀M	♀I	♂M	♂I	♀M	♀I	♂M	♂I
1	143	61	160	25	18	44	20	21
2	147	30	131	18	32	60	28	37
3	120	31	131	24	24	58	21	41
4	132	38	106	22	28	56	38	40
5	101	26	109	31	34	55	26	32
6	116	43	95	18	24	50	35	25
7	103	27	82	12	41	44	51	33
8	68	30	58	11	56	30	62	30
9	33	10	30	7	78	30	69	16
10	17	8	18	5	53	10	31	5
11	7	4	9	1	51	13	31	1

Note:- M = Mature
I = Immature

APPENDIX C

TABLE 2: Monthly zonation patterns of the subpopulations of L. rudis C and L. arcana

Transect	Month of sample											
zone	Mar '77	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan '78	Feb
	Mature L. arcana ♀♀											
1	-	-	-	-	-	-	3	8	4	2	1	-
2	2	-	-	1	-	1	8	10	6	3	-	1
3	-	-	-	-	-	-	13	6	1	2	1	1
4	2	-	1	-	-	3	6	4	5	4	2	1
5	1	-	-	1	-	-	15	6	5	1	5	-
6	-	-	2	-	-	1	6	6	5	1	1	2
7	-	3	1	-	-	-	7	8	8	5	6	3
8	9	4	4	-	-	-	6	11	7	8	5	2
9	8	3	8	3	-	-	8	19	11	8	6	4
10	3	8	7	1	1	-	7	2	7	4	9	4
11	9	11	2	1	-	-	4	1	14	4	2	3
	Immature L. arcana ♀♀											
1	9	1	3	3	-	3	4	4	4	4	6	3
2	4	7	4	4	7	5	4	5	5	9	5	1
3	3	2	4	8	9	14	1	4	3	4	3	3
4	4	3	5	2	5	8	5	3	4	6	4	7
5	6	3	10	7	5	6	2	1	5	3	5	2
6	4	7	11	4	5	5	2	-	2	2	5	3
7	3	4	8	9	1	6	1	-	5	2	4	1
8	1	4	5	6	1	4	2	-	1	2	3	1
9	-	3	2	9	2	1	-	2	3	6	1	1
10	-	2	1	5	1	-	1	-	-	-	-	-
11	-	3	1	3	-	-	-	-	-	4	1	1
	Mature L. arcana ♂♂											
1	-	-	-	-	-	5	6	3	4	1	1	-
2	1	-	-	-	-	1	6	11	4	3	2	-
3	-	2	-	-	1	2	7	5	2	2	-	-
4	2	1	-	-	-	4	10	12	3	3	3	-
5	-	-	-	-	-	1	12	4	4	2	3	-
6	1	1	2	1	-	5	7	9	2	3	4	-
7	5	-	1	-	-	5	7	22	1	5	4	1
8	11	5	5	1	-	2	10	13	6	5	3	1
9	9	9	3	-	-	-	15	11	5	7	6	4
10	1	3	1	-	-	-	3	6	7	5	4	1
11	4	6	1	-	-	-	4	-	-	5	8	3
	Immature L. arcana ♂♂											
1	5	-	4	4	1	2	2	-	1	-	1	1
2	2	8	7	7	3	-	5	-	-	-	4	1
3	5	5	6	4	8	2	4	1	3	2	1	-
4	3	6	6	4	5	9	2	2	1	1	1	-
5	4	3	6	8	3	2	4	-	1	-	1	-
6	2	2	7	6	1	3	1	1	-	1	1	-
7	2	4	6	5	4	5	2	-	1	1	2	1
8	3	2	11	5	1	2	2	-	2	1	-	1
9	-	1	5	6	1	-	-	-	1	1	1	-
10	1	1	1	2	-	-	-	-	-	-	-	-
11	-	-	1	-	-	-	-	-	-	-	-	-

TABLE 2 Contd.

Transect	Month of sample											
zone	Mar '77	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan '78	Feb
	Mature L. rudis C ♀♀											
1	9	14	10	7	11	8	6	16	17	16	21	8
2	11	12	12	11	15	14	14	16	10	9	12	11
3	12	9	11	12	9	11	5	16	9	13	10	3
4	18	10	14	15	5	9	19	13	4	7	12	6
5	9	9	8	12	4	15	7	7	9	10	7	4
6	10	10	10	10	15	12	14	2	9	13	6	5
7	10	14	12	10	8	6	12	7	6	12	4	2
8	11	8	5	9	5	3	4	6	3	7	7	-
9	-	5	6	3	4	-	3	1	2	3	4	2
10	-	-	3	5	-	-	2	1	1	4	-	1
11	-	1	-	-	-	-	2	-	2	1	1	-
	Immature L.rudis C ♀♀											
1	3	5	3	6	3	6	5	6	4	7	7	6
2	7	2	3	2	1	2	3	4	1	1	1	3
3	3	2	3	1	1	3	5	2	4	1	3	3
4	2	2	4	3	3	1	3	3	4	6	5	2
5	1	3	-	4	1	2	5	2	2	3	2	1
6	9	3	4	8	2	2	2	5	2	1	5	-
7	4	2	-	3	2	2	2	1	1	4	5	1
8	2	3	1	-	1	5	1	1	6	5	3	2
9	1	1	-	1	1	-	1	-	1	2	1	1
10	-	1	-	2	-	1	1	-	1	1	-	1
11	-	-	-	-	-	-	1	-	1	-	1	1
	Mature L. rudis C ♂♂											
1	15	7	11	7	16	12	16	15	16	20	13	12
2	14	6	13	10	9	9	9	7	16	14	15	9
3	14	13	8	15	7	13	13	12	12	8	10	6
4	8	4	10	13	9	6	9	13	9	11	9	5
5	12	8	13	10	10	13	8	16	7	10	1	1
6	7	8	10	7	9	16	12	8	6	7	2	3
7	13	7	8	11	3	2	7	7	5	5	11	3
8	3	6	5	8	4	6	4	4	5	3	5	5
9	-	5	7	5	-	-	4	1	1	2	2	3
10	-	2	3	3	1	1	-	-	2	2	3	1
11	-	1	1	3	1	-	1	-	-	1	-	1
	Immature L. rudis C ♂♂											
1	2	5	3	3	3	1	-	3	-	2	1	2
2	2	3	1	1	1	1	2	2	1	2	1	1
3	1	-	3	4	1	4	2	1	1	-	5	2
4	1	3	1	3	6	2	2	1	-	1	-	2
5	2	4	3	2	3	2	1	4	3	1	3	3
6	3	1	1	2	2	1	2	-	2	1	2	1
7	1	-	1	2	1	1	-	-	1	3	1	1
8	1	1	2	-	2	2	-	-	1	2	-	-
9	-	1	-	1	2	1	2	-	-	-	-	-
10	-	1	-	1	1	-	-	-	1	-	-	1
11	-	1	-	-	-	-	-	-	-	-	1	-

APPENDIX C

TABLE 3. Estimated % cover of barnacles in transect 2 (April 1977)

Transect zone	% cover
1	2
2	25
3	73
4	70
5	76
6	72
7	64
8	56

APPENDIX C

TABLE 4: Zonations in transect 2 in successive months

Transect zone	Month of Collection												Total in zone
	Apr. '77	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan '78	Feb	Mar	
	L. rudis C juveniles												
1	-	2	7	3	12	13	10	18	14	20	19	17	135
2	2	1	4	-	6	14	18	16	15	10	10	9	105
3	3	4	2	4	8	14	8	7	4	11	5	1	71
4	4	2	3	2	7	5	4	3	8	2	3	2	45
5	3	-	-	5	1	3	-	7	1	3	9	-	32
6	-	2	-	1	5	5	4	2	4	1	3	-	27
7	-	3	2	3	2	1	-	6	2	3	1	4	27
8	-	3	3	1	3	-	-	1	3	1	1	1	17
	L. rudis B juveniles												
1	3	32	18	7	35	44	26	25	33	21	27	43	314
2	16	34	24	15	39	30	21	41	24	25	32	41	342
3	21	24	30	22	35	45	27	34	26	24	27	35	350
4	11	38	34	21	35	26	43	44	20	36	29	40	377
5	14	32	48	32	26	34	44	33	37	22	27	56	405
6	35	57	45	34	41	56	34	49	43	27	44	60	525
7	33	22	57	35	46	47	48	39	49	44	50	64	534
8	18	39	44	54	54	29	58	62	44	50	56	67	575
	L. nigrolineata juveniles												
1	-	9	8	13	8	1	6	3	-	1	-	1	50
2	5	20	12	18	22	4	6	6	3	5	4	9	114
3	5	16	20	24	19	13	6	4	9	4	3	13	136
4	18	22	25	36	17	18	8	11	5	6	9	6	181
5	25	33	27	29	42	19	11	10	13	16	13	10	248
6	20	19	32	34	31	20	10	11	13	10	17	6	223
7	17	19	18	31	24	19	11	16	15	6	6	2	184
8	1	10	23	19	16	3	7	6	10	6	5	1	107
	L. neglecta												
1	3	11	7	5	6	6	15	8	6	10	9	8	94
2	14	7	18	15	11	9	21	13	11	12	12	21	164
3	28	20	18	20	10	5	28	29	22	16	22	18	236
4	19	13	16	5	12	12	13	10	23	14	20	22	179
5	22	18	5	6	11	1	9	25	14	20	12	7	150
6	9	16	2	7	4	2	14	17	10	12	6	4	103
7	5	7	1	2	-	-	4	11	8	3	2	4	47
8	4	7	2	-	-	1	4	2	-	-	1	1	22

APPENDIX C

TABLE 5: Population size structures

	Month of collection											
Size class (gu)	Apr '77	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan '78	Feb	Mar
	L. nigrolineata											
2.0-2.4	12	15	10	9	7	2	4	12	7	7	6	2
2.5-2.9	20	32	20	26	14	-	8	7	19	14	10	5
3.0-3.4	19	32	32	40	20	12	6	5	12	8	19	9
3.5-3.9	17	33	27	39	37	18	10	11	9	11	12	5
4.0-4.4	12	15	27	42	43	17	12	11	9	3	5	9
4.5-4.9	5	13	26	20	28	14	12	7	5	3	3	10
5.0-5.4	5	3	17	19	20	18	11	8	5	6	1	7
5.5-5.9	1	3	3	5	8	8	1	4	1	-	-	-
6.0-6.4	-	2	3	4	2	8	1	2	1	2	1	1
	L. rudis C											
2.0-2.4	1	3	6	1	11	17	6	14	12	10	10	11
2.5-2.9	2	1	-	6	3	12	5	9	12	6	9	6
3.0-3.4	1	4	4	5	11	12	12	12	6	14	11	6
3.5-3.9	2	3	2	2	8	5	7	7	8	4	5	3
4.0-4.4	3	3	2	3	3	3	6	7	7	8	11	4
4.5-4.9	2	1	3	1	6	2	5	9	4	6	2	-
5.0-5.4	-	1	-	-	1	3	1	1	1	2	3	2
5.5-5.9	1	1	3	-	-	-	2	1	1	1	-	-
6.0-6.4	-	-	1	1	1	1	-	-	-	-	-	2
	L. rudis B (& *L. arcana*)											
2.0-2.4	21	59	55	29	37	20	22	63	67	73	67	108
2.5-2.9	37	58	52	51	59	34	37	43	52	45	65	125
3.0-3.4	39	60	71	49	70	75	51	53	47	41	50	86
3.5-3.9	40	45	47	39	62	68	83	56	40	36	40	32
4.0-4.4	9	35	42	34	48	61	50	53	37	24	30	28
4.5-4.9	4	14	22	15	24	28	36	36	17	16	21	9
5.0-5.4	1	5	7	4	8	16	15	16	8	8	11	13
5.5-5.9	-	1	4	1	1	7	5	5	5	3	4	4
6.0-6.4	-	1	-	-	2	2	2	-	3	3	4	1
	L. neglecta											
2.0-2.4	3	3	4	9	13	10	44	40	11	14	3	6
2.5-2.9	15	11	10	7	4	8	39	41	40	34	27	24
3.0-3.4	37	31	20	12	12	9	11	27	28	32	38	40
3.5-3.9	27	37	13	16	20	1	8	4	8	4	14	13
4.0-4.4	17	12	16	15	5	6	5	3	3	3	2	2
4.5-4.9	5	5	4	1	-	2	1	-	4	-	-	-
5.0-5.4	-	-	2	-	-	-	-	-	-	-	-	-

APPENDIX C

Table 6. Zonation of male and female L. neglecta

Transect zone	Number of ♀♀ in each zone (a)	Number of ♂♂ in each zone	Total in zone (n)	$\frac{a^2}{n}$
1	44	42	86	22.51
2	63	80	143	27.76
3	119	104	223	63.50
4	102	58	160	65.02
5	94	49	143	61.79
6	48	47	95	24.25
7	27	15	42	17.36
8	11	10	21	5.76
Totals	A = 508	405	N = 913	287.95
			$\frac{A^2}{N}$	282.65

$$\chi^2 = \frac{5.3}{.556 \times .444} = 21.47$$

degrees of freedom = 7

significant at .01

APPENDIX C

TABLE 7

Transect zone	Female				Male				Total in zone			
	Size classes (gu)											
	2.0-2.4	2.5-2.9	3.0-3.4	3.5-3.9	2.0-2.4	2.5-2.9	3.0-3.4	3.5-3.9	2.0-2.4	2.5-2.9	3.0-3.4	3.5-3.9
1	3	9	11	7	7	13	17	5	10	22	28	12
2	5	12	12	10	6	25	39	9	11	37	51	19
3	9	26	27	32	19	35	35	13	28	61	62	45
4	5	22	33	21	5	19	28	5	10	41	61	26
5	6	12	32	31	13	19	11	5	19	31	43	36
6	3	9	14	13	10	18	15	3	13	27	29	16
7	3	5	10	7	3	9	3	-	6	14	13	7
8	-	2	4	7	2	3	5	-	2	5	9	7
	34	97	143	128	65	141	153	40	99	238	296	168

χ^2 tests

Transect zone	$\frac{a^2}{n}$ for each size class 2.0-2.4	2.5-2.9	3.0-3.4	3.5-3.9
1	3.05	3.68	4.32	9.32
2		3.89	2.82	
3	2.89	11.08	11.76	22.76
4	4.17	11.80	17.85	16.96
5		4.65	23.81	26.69
6	1.71	3.0	6.76	24.3
7		2.58	8.91	
8				
$\sum \frac{a^2}{n}$	11.82	40.68	76.23	100.03
$\frac{A^2}{N}$	11.68	39.53	69.08	97.52
$\chi^2 =$	$\frac{.14}{.225351}$	$\frac{1.15}{.241536}$	$\frac{7.15}{.249711}$	$\frac{2.51}{.181356}$
=	.621	4.76	28.63	13.84
degrees of freedom	3 NS	6 NS	6 sig. .001	4 sig. .01

Note

NS: not significant
sig: significant

APPENDIX D

Table 1. Frequency distribution of numbers and arrangements (where possible) of penial glands.

	Porth Swtan						Rhoscolyn		
	L. rudis C			*L. rudis* B			*L. rudis* c	*L. rudis* I	*L. rudis* b
Penial gland arrangement	S	I	D	S	I	D			
Number of penial glands									
4				3					
5				2					
6	1			3					
7	4			3	1				
8	10			4	3		1	1	
9	28	2		5	3		2	2	3
10	55	9		9	5		8	7	6
11	57	21		8	2		4	5	4
12	43	39		18	3		10	7	4
13	36	46		22	12		10	11	4
14	25	38		31	9		8	8	6
15	11	51		28	16		3	14	2
16	5	24	1	21	19		3	7	3
17	1	24	1	11	17		3	6	1
18		5	1	11	24	2		8	
19		8		1	17		2	5	
20		2	1	6	19	1		1	
21		3	1	3	13	2		2	
22				2	10			3	
23				2	10	1			
24					5	1			
25					4	3			
26					2				
Total	276	272	5	193	194	10			
% in each category of penial gland arrangement	50%	49%	1%	49%	49%	2%			

Note: S: - single row
I: - irregular
D: - double row

APPENDIX D

TABLE 2: Percentage mature in successive size classes

	Rhoscolyn (data from a sample taken in November 1977)																		Porth Swtan (data from samples taken over the year March 1977 - February 1978)											
Size class	*L. rudis* c						*L. rudis* I						*L. rudis* b						*L. rudis* C						*L. rudis* B					
	♂M	♀M	♂I	♀I	T	%M	♂M	♀M	♂I	♀I	T	%M	♂M	♀M	♂I	♀I	T	%M	♂M	♀M	♂I	♀I	T	%M	♂M	♀M	♂I	♀I	T	%M
6.5-6.95	3	1	4	5	13	31	-	-	4	2	6	0	-	-	6	3	9	0	186	93	139	216	634	44	-	-	-	-		
7.0-7.45	4	4	5	5	18	44	1	-	4	6	11	9	-	-	1	4	5	0	256	236	66	177	735	67	-	-	-	6	6	0
7.5-7.95	6	8	4	9	27	52	-	-	3	7	10	0	-	-	1	3	4	0	303	307	36	62	708	86	-	-	6	9	15	0
8.0-8.45	8	12	2	1	23	87	5	2	6	19	32	22	-	-	5	4	9	0	226	256	25	50	557	87	1	-	7	12	20	5
8.5-8.95	11	10	1	1	23	91	6	5	4	6	21	52	2	2	3	7	14	29	145	190	10	21	366	92	2	2	13	11	28	14
9.0-9.45	12	8	-	-	20	100	11	10	2	6	29	72	1	2	6	6	15	20	74	90	6	7	177	93	-	1	17	21	39	3
9.5-9.95	5	8	-	2	15	87	12	15	5	1	33	82	10	3	5	4	22	59	33	38	5	1	77	92	3	1	19	15	38	11
10.0-10.45	1	5	-	-	6	100	14	12	-	-	26	100	6	9	2	1	18	83	14	21	1	2	38	92	7	7	21	15	40	28
10.5-10.95	1	3	-	-	4	100	21	11	-	-	32	100	8	8	3	4	23	70	8	7	-	1	16	94	15	11	16	25	67	39
11.0-11.45	2	2	-	-	4	100	5	5	-	-	10	100	12	11	-	-	23	100	5	4	-	1	10	90	30	23	18	13	84	63
11.5-11.95	1	-	-	-	1	100	3	5	-	-	8	100	14	14	2	-	30	93							31	30	11	20	92	66
12.0-12.45							2	-	-	-	2	100	7	6	2	1	16	81							37	44	8	17	106	76
12.5-12.95							4	-	-	-	4	100	7	10	2	1	20	85							51	52	7	20	130	79
13.0-13.45							1	1	-	-	2	100	11	5	3	4	23	70							56	71	7	9	143	89
13.5-13.95							-	-	-	-	-		6	8	2	4	20	70							44	56	6	7	113	88
14.0-14.45							1	-	-	-	1	100	4	6	1	6	17	59							39	60	2	5	106	93
14.5-14.95													-	5	4	1	10	50							25	39	1	2	67	96
15.0-15.45													1	3	-	4	8	50							10	35	-	1	46	98
15.5-15.95													1	2	3	1	7	43							8	23	-	-	31	100
16.0-16.45													-	-	1	-	1	0							9	16	-	1	26	96
16.5-16.95													1	-	-	-	1	100							2	7	-	-	9	100
17.0-17.45													-	2	-	2	4	100							-	5	-	-	5	100
17.5-17.95													-	-	-	-	-								-	4	-	-	4	100
18.0-18.45													-	-	-	1	1	0							-	2	-	-	2	100

Notes:- M: - Mature
I: - Immature
T: - Total

REFERENCES

ANDERSON, A. 1974. Studies in the ecogenetics of Littorina neglecta Bean, a member of the rough winkle aggregate of species. M.Sc Thesis. University of Liverpool.

BAILEY, N.T.J. 1959. Statistical methods in biology. English Universities Press Ltd.

BALLANTINE, W.J. 1961. A biologically-defined exposure scale for the comparative description of rocky shores. Field Studies. 1(3) 1-9.

BANDEL, K. 1974. Studies on Littorinidae from the Atlantic. Veliger 17(2) 92-114.

BEAN, W. 1844. A supplement of new species. In C. Thorpe, British marine Conchology. London Lumley.

BERGERARD, J. 1971a. Cycle sexuel annuel de Littorina saxatilis (Olivi). Haliotis 1(1) 23-24.

BERGERARD, J. 1971b. Facteurs écologiques et cycle sexuel de Littorina saxatilis (Olivi). Cah. Biol. Mar. 12 187-193.

BERRY, A.J. 1961. Some factors affecting the distribution of Littorina saxatilis (Olivi). J. Anim. Ecol. 30 27-45

CAIN, A.J. 1978. Different sorts of species; taxonomic and ecological. Porcupine Newsletter 1 (5) 78

CARRIKER, M.R. 1943. Variability, developmental changes, and denticle-replacement in the radula of Lymnaea stagnalis apressa Say. Nautilis 57(2) 52-59

CASON, J.E. 1950. A rapid one-step Mallory-Heidenhain stain for connective tissue. Stain Technology 25. 225-226.

CASTENHOLZ, R.W. 1961. The effect of grazing on marine littoral diatom populations. Ecology 42 783-794

CLARKE, B. 1962. Natural selection in mixed populations of two polymorphic snails. Heredity 17 319-345

DAUTZENBERG, P. & FISCHER, P. 1912. Mollusques provenant des campagnes de l'Hirondelle et de la Princess Alice dans les Mers du Nord. Résult. Camp. Scient. Princo Albert I. Moll. Hirondelle 37 1-629

DAVIS, G.M. 1966. Notes on Hydrobia totteni. Venus 25(1) pp 27-39.

ELNER, R.W. & RAFFAELLI, D.G. 1980. Interactions between two marine snails, Littorina rudis and Littorina nigrolineata Gray, a predator, Carcinus maenas (L), and a parasite, Microphallus similis Jägerskiold. J.exp.mar.Biol.Ecol. 43 1-10

EMSON, R.H. & FALLER-FRITSCH,R.J. 1976. An experimental investigation into the effect of crevice availability on abundance and size-structure in a population of Littorina rudis (Maton): Gastropoda:Prosobranchia. J. exp.mar.Biol.Ecol. 23 285-297

FALLER-FRITSCH, R.J. 1977. Reproductive strategies of the winkle Littorina rudis in relation to population dynamics and size structure. In 'Biology of Benthic Organisms' Ed. Keegan et al. Pergamon Press.New York.

FISCHER-PIETTE, E., GAILLARD, J.M. & JAMES,B.L. 1963. Études sur les variations de Littorina saxatilis. V. sur des cas de variabilite extrème. Cah.Biol.Mar. 4 1-22

FRETTER, V. & GRAHAM, A. 1962. British Prosobranch Molluscs. Ray Society. London.

GOODWIN, B.J. & FISH, J.D. 1977. Inter- and intraspecific variation in Littorina obtusata and L.mariae (Gastropoda: Prosobranchia). J. Moll. Stud. 43 241-254

GRAY, J.E., 1839. The zoology of Captain Beechey's Voyage. Molluscous animals and their shells. London.

HANNAFORD ELLIS, C.J. 1978. Littorina arcana sp.nov.: A new species of winkle (Gastropoda: Prosobranchia: Littorinidae). J. Conch. 29 304

HANNAFORD ELLIS, C.J. 1979. Morphology of the oviparous rough winkle, Littorina arcana Hannaford Ellis, 1978, with notes on the taxonomy of the L.saxatilis species-complex (Prosobranchia: Littorinidae). J. Conch. 30 43-56

HART, A. 1978. Life history strategies in relation to environmental factors: A study of local populations of Littorina rudis. Unpublished report. University of Liverpool.

HELLER, J. 1975a.The taxonomy of some British Littorina species with notes on their reproduction. (Mollusca: Prosobranchia). Zool. J. Linn. Soc. Lond. 56(2) 131-151

HELLER, J. 1975b. Visual selection of shell colour in two littoral prosobranchs. Zool.J.Linn.Soc. 56(2)153-170

HELLER, J. 1976. The effects of exposure and predation on the shell of two British winkles. J. Zool. 179 201-213.

HUGHES, R.N. 1979. South African populations of Littorina rudis. Zool.J.Linn.Soc. 65 119-126.

JAMES, B.L. 1968a. The characters and distribution of the subspecies and varieties of Littorina saxatilis (Olivi, 1792) in Britain. Cah.Biol.Mar. 9 143-165.

JAMES, B.L. 1968b. The occurrence of Parvatrema homoeotecnum James,1964, (Trematoda: Gymnophallidae) in a population of Littorina saxatilis tenebrosa (Mont.) J.Nat.Hist 2 21-37

JEFFREYS, J.G. 1865. British Conchology, or an account of the Mollusca which now inhabit the British Isles and the surrounding seas. vol. 3. London. van Voorst.

JOHANSEN, A.C. 1901. On the variations observable in some northern species of Littorina. Vidensk. Meddelfaaden Naturh. Foren. Kjøbenhaven. 53 295-307.

LINKE, O. 1933. Morphologie und Physiologie des Genitalapparatus der Nordsee littorinen. *Wiss.Meeres.Abt. Helgoland*. (N.F.) **19**(5): 1-60

MATON, W.G. 1797. *Observation relative chiefly to the Natural History, Picturesque Scenery, and Antiquities of the western Counties of England, made in the years 1794 and 1796*. Salisbury: J.Eaton.

MAYNARD SMITH, J. 1966. Sympatric speciation. *Amer. Nat* **100** (916) 637-650.

MEEUSE, A.D.J. 1950. Rapid methods for obtaining permanent mounts of radulae. *Basteria* **14** 28-43.

NAYLOR, R. 1978. Distribution, isolation and adaptation of *Littorina nigrolineata* (Gray) on Holy Island, Anglesey. Unpublished report. University of Liverpool

ODUM, E.P. 1971. *Fundamentals of Ecology*. W.B.Saunders Co.

OLIVI, A.G. 1792. *Zoologica Adriatica Bassano*

PETTITT, C.W. 1973a. A proposed new method of scoring the colour morphs of *Littorina saxatilis* (Olivi,1792) (Gastropoda: Prosobranchia). *Proc. Malac. Soc. Lond.* **40**(6) 531-538

PETTITT, C.W. 1973b. Possible shedding of the penis by *Littorina saxatilis* (Olivi,1792). *New York Shell Club Notes* **195** 7.

PETTITT, C.W. 1975. A review of the predators of *Littorina* especially those of *L.saxatilis* (Olivi) (Gastropoda Prosobranchia) *J.Conch* **28** (6) 343-357

RADWIN, G.E. 1970. Technique for Extraction and Mounting of Gastropod Radulae. *Veliger* **12** (1) 143-144

RAFFAELLI, D.G. 1976. The determinants of zonation patterns of *L.neritoides* and the *L.saxatilis* species complex. Unpublished Ph.D. thesis. University College of North Wales.

RAFFAELLI, D.G. 1977. Observations on the copulatory behaviour of *Littorina rudis* (Maton) and *Littorina nigrolineata* Gray (Gastropoda: Prosobranchia).*Veliger* **20** 75-77

RAFFAELLI, D.G. 1978a. Factors affecting the population structure of Littorina neglecta Bean. J.Moll. Stud. 44 223-230.

RAFFAELLI, D.G. 1978b. The relationship between shell injuries, shell thickness and habitat characteristics of the intertidal snail Littorina rudis Maton. J. Moll. Stud. 44 166-170

RAFFAELLI, D.G. 1979. The taxonomy of the Littorina saxatilis species complex, with particular reference to the systematic status of Littorina patula Jeffreys. Zool. J. Linn. Soc. 65 219-232

RAFFAELLI, D.G. & HUGHES, R.N. 1978. The effects of crevice size and availability on populations of Littorina rudis and Littorina neritoides. J. Anim. Ecol. 47 71-83

REIMCHEN, T.E. 1974. Studies on the biology and colour polymorphism of two sibling species of marine gastropods (Littorina) Unpublished Ph.D. thesis. University of Liverpool.

ROSEWATER, J. 1970. The family Littorinidae in the Indo-Pacific Part 1. The superfamily Littorininae. Indo-Pacific Mollusca 2 417-506

SACCHI, C.F. 1966. Sur le dimorphisme du penis chez Littorina obtusata (L.) (Gastropoda:Prosobranchia) C.R.Acad. Sci. Paris 262D(22) 2370-2372

SACCHI, C.F. 1968. Sur le dimorphisme sexuel de Littorina mariae Sacchi & Rastelli. C.R.Acad.Sci.Paris 266 2483-5

SACCHI, C.F. 1975. Littorina nigrolineata (Gray) (Gastropoda:Prosobranchia) Cah. Biol. Mar. 16 111-120

SACCHI, C.F. & RASTELLI, M. 1966. Littorina mariae n. sp. les différences morphologiques et écologiques entre 'nains et 'normanaux' chez l'espèce L.obtusata (L)

(Gastropoda:Prosobranchia) et leur signification adaptative et évolutive. Atti. Soc. Ital. Sci. Nat 105 351-369

SESHAPPA, G. 1947. Oviparity in Littorina saxatilis (Olivi).Nature Lond. 160 (4062) 335-336

SESHAPPA, G. 1948. Nomenclature of the British Littorinidae Nature, London 162 (4122) 702-703

SIEGEL, S. 1956. Nonparametric statistics for the behavioural sciences. McGraw-Hill N.Y.

SMITH, S.M. 1978. Winkles in the Uists. Hebridean Naturalist 1 45-49

SNELI, J. A. & van MARION, P. 1979. Nye strandsnegler i norsk fauna. Fauna 32 4-8

SOLEM, A. 1972.. Malacological applications of SEM. II. Radular structure and function. Veliger 14(4) 327-336

SOLEM, A. 1974. The shell makers: Introducing Mollusks. John Wiley & Sons.

STEARNS, S.C. 1976. Life-history tactics: a review of the ideas. Quart. Rev. Biol. 51 3-47

THORPE, C. 1844. British Marine Conchology. London.Lumley

WILLIAMS, E.E. 1964. The growth and distribution of L.littorea (L) on a rocky shore in Wales. J. Anim. Ecol. 33 413-432

ZARET, T.M. 1972a. Predator-prey interaction in a tropical lacustrine ecosystem Ecology 53 248-257

ZARET, T.M. 1972b. Predators, invisible prey, and the nature of polymorphism in the Cladocera (class Crustacea) Limn. and Ocean. 17 171-184