AN ANALYSIS OF THE MAINTENANCE AND
CONTROL OF A POLYMORPHISM
IN THE LIMPET
ACMAEA DIGITALIS ESCHSCHOLTZ

by

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INTRODUCTION

Crypsis is a common phenomenon in many animal groups. *Cepaea nemoralis* and *Cepaea hortensis* are excellent cases in point (Lamotte 1950, Carter 1967, Clarke 1962, Cain and Sheppard 1952, 1960, 1961, Cain and Currey 1963, Owen 1965, Sheppard 1961). Diver (1940) early suggested that the pattern composition of *Cepaea* populations is not affected by natural selection but rather is controlled primarily by random processes. But Cain and Sheppard (1950) found the colors and patterns of *Cepaea sp.* have real selective value and suggested that while random factors such as genetic drift, (a sampling effect, Wright 1931), must exist (some populations are relatively small), drift is not of a magnitude to mitigate selection. They showed that selective value is dependent upon environment, different morphs being differentially selected both physiologically and by predators.

Later Sheppard (1951) found that the song thrush *Turdus ericetorum* selects differentially patterned and colored snails from the same areas at different seasons. Thus selection was shown to be cyclical, the relative advantage of a morph changing with time.

Clarke (1962) found in this regard that *C. hortensis* shows no correlation between distribution of phenotypes and background, pink and brown morphs being at a disadvantage everywhere. He suggested that selection for characters other than those of physical appearance is important in this species.

European robins, hedge sparrows, and other species selectively prey on the moth Biston betularia, the efficacy of selection being dependent on the ability of the moth to rest on cryptic substrates (Kettlewell 1955). Moths, on an individual basis, may actively select a matching background (Kettlewell 1955, Sargeant 1966). A similar suggestion was made in the case of Cepaea (Lamotte 1955).

Owen (1965) and later Carter (1967) found evidence for frequency-dependent selection in large populations of Cepaea. Theoretically, frequency-dependent "apostatic" selection is based on the existence in a polymorphic population of one morph which becomes more common than the others. The visual predator, for example a bird, then forms a search image for the more common form (DeRuiter 1952) preying on it until its frequency becomes relatively small at which time the predator establishes a new search image for another, now common, form. In this way a highly diverse polymorphism is maintained, the establishment of new forms being made possible by the comparatively low predatory selection against them.

Colton (1916) describes a polymorphism in Thais lapillus, the dog whelk, correlates it subjectively with background, and invokes weak natural selection as being responsible for reducing natural variety to a complex of inconspicuous forms. Moore (1936) found that the color of the whelk is a function of pigment quality of the animals' food.

In the limpet Acmaea digitalis that I have studied, two form complexes exist. The one, subjectively, is characteristic of beds of the gooseneck barnacle Pollicipes polymerus Sowerby. The other is found on the rock faces of the high intertidal.

The polymorphism in Acmaea digitalis is the result of genetic and environmental factors. More specifically, I shall demonstrate that color in these animals is first a function of their genotype and second, of their substrate, that there is a genetically based, nonrandom, substrate preference in the population which mediates the balance between genetic and environmental variance. I shall also show that substrate-seeking behavior is correlated with the genetic component of pattern and that the degree of correlation is modified by predation and by physical factors. The degree of modification is directly related to the estimated intensity of predation during a generation. Disruptive selection will be suggested to control both pattern frequency and pattern-behavior relationship and to result in the genesis of two distinct subpopulations.

STUDY AREAS

The study was pursued at middle cove at Cape Arago State Park, and the southernmost rocky area of Sunset Bay State Park. Both areas are on the southern coast of Oregon at 43°18' N. latitude, 124°25' longitude.

Middle cove is partially protected to the west by a long reef paralleling the shore. The reef is emergent at all but the highest high tides. To the north is a headland separating middle cove from north cove. The cove is open to wave attack from the southwest, a direction of prevailing winds in the winter.

The study area at Sunset Bay is partially protected from wave action by a large stack lying nearby to the west.

The Pollicipes colonies at Cape Arago are extensive and large, up to 45 cm diameter. Individual barnacles are generally large, reaching a maximum size, measured by scutum length, of 18 mm. They occur in close association with the mussel Mytilus californianus, sometimes forming integrated beds with this species. They also commonly extend above the Mytilus zone, occurring as much as two feet above the highest mussels. At high intertidal levels the colonies are compact and well-defined and, although they might follow irregularities in the rock face, are often round. The rock face in the vicinity of the high colonies is commonly smooth and inhabited rather sparsely by a small acorn barnacle Balanus glandula Darwin. The lower colonies inhabit an area complicated by lush growth of Endocladia muricata Agardh, a red alga, mussels, and large acorn barnacles. The rock surface is pocked, almost never being smooth.

In the Sunset Bay study area the Pollicipes are much less common and occur in smaller colonies than at Cape Arago. Individual barnacles are smaller (maximum size, 10 mm). The Pollicipes occur in the same associations as at Cape Arago although they are relatively uncommon in the high intertidal.

The goose barnacles in both areas are parasitized by Dydimella conchae Bonar. As a result they have relatively rough, nonreflective scute surfaces. Their shells are generally white to grey.

EXPERIMENTAL ANIMALS

Two forms of Acmeae digitalis were studied. The first, with habitat of rock faces of the high intertidal, is the familiar brown, patterned limpet. The second, found in close association with Pollicipes, typically has a white rim, grey apex and subapex, and striping patterns variable both in degree and morphology. A vast amount of variation is present in this form which, since it is subjectively characteristic of Pollicipes colonies, will be called Pollicipes-type. The more common rock-type animals have tan to light brown apex and tan to white rim tending to pure black or brown depending on the amount of striping present. There is overlap in degree of striping between rock-type and Pollicipes-type animals although the rock-types generally have heavier and more numerous stripes.

Taxonomy

The probable taxonomic relationships are worthy of consideration. Fritchman (personal communication to Peter Frank) feels that the white animals are definitely a form of A. digitalis. The radulae of white Pollicipes-type animals are indistinguishable from those of the more common A. digitalis.

Karyotype studies, using primary and secondary spermatocytes, showed that all animals studied had 9 chromosome pairs. Because of the extremely small nuclei and chromosomes, it was not possible to characterize these animals beyond chromosome number. This of course is not conclusive evidence of taxonomic relationship. Patterson (1967) reports that all of the Patellacea have 9 chromosome pairs.

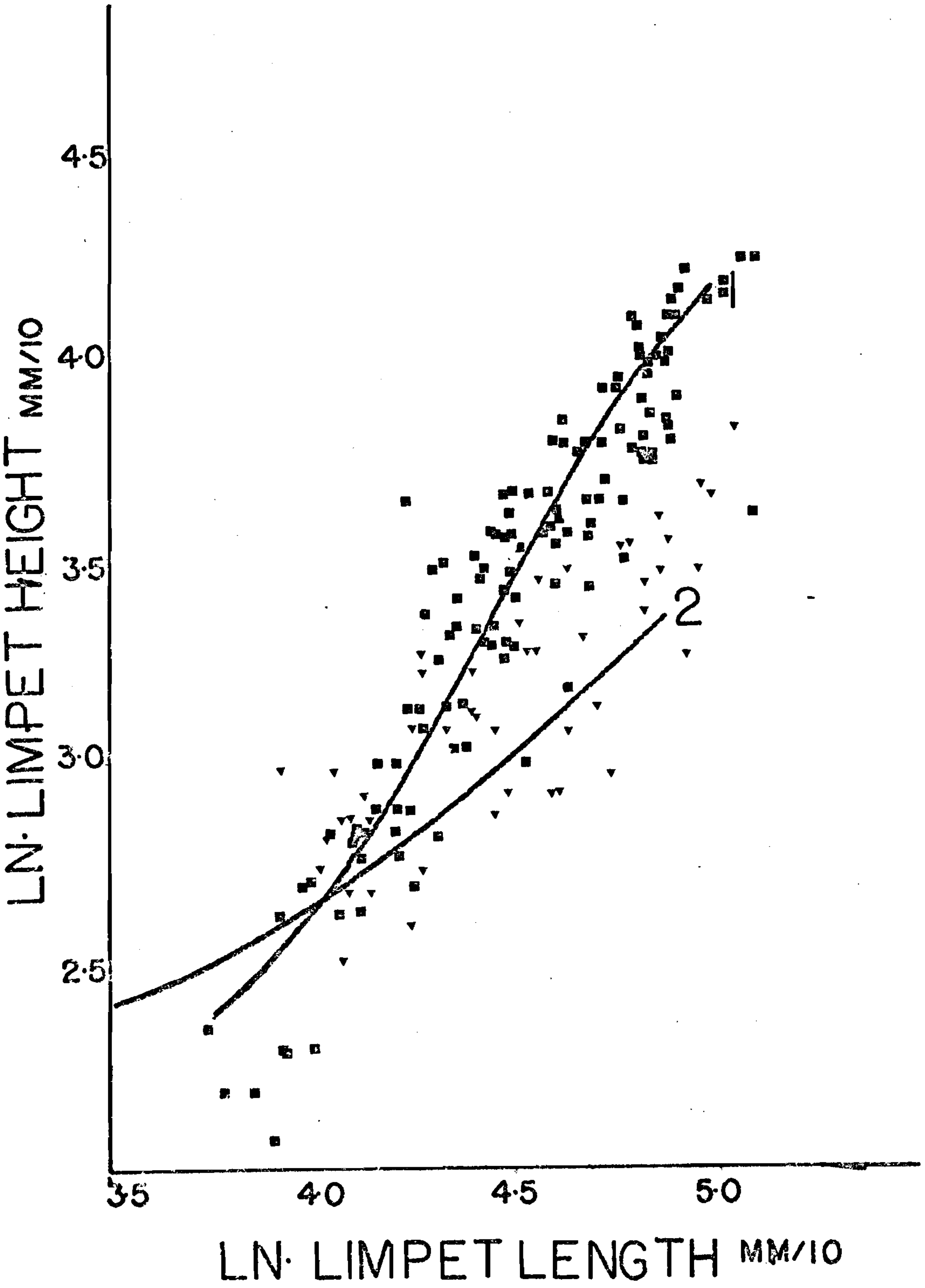
Censuses of young animals of both types show a unimodal distribution of pattern frequencies (see below, Fig. 6B) with intermediate patterns frequently represented. That some darker Pollicipes-type animals when placed on rock assume rock-type pattern, color, and allometry (see p. 17,18) also suggests that only one species is present.

The young of many species of Acmaea are superficially indistinguishable. Those of A. pelta are very polymorphic and can be mistaken for A. digitalis (McLean, personal communication 1967). It is therefore possible, although not probable because of differences in the vertical ranges of the Acmaea species, that samples of young limpets consist of members of many species. This problem should be kept in mind but probably is of no concern.

Allometry

The allometric length-height relationships of rock-type and Pollicipes-type A. digitalis differ strikingly

Figure 1. Relationship of ln height to ln length in *Acmaea digitalis*. Curve 1, *Pollicipes*-type: $y = 13.12 - 5.53x + .748x^2$. Curve 2, rock-type: $y = 3.47 - 1.1x + .23x^2$.


4·5
4·0
3·5
3·0
2·5
LN· LIMPET HEIGHT MM/10
1
2
3·5
4·0
4·5
5·0
LN· LIMPET LENGTH MM/10

(Figure 1). The ln-ln relationship should approximate a straight line or, failing that, should be reducible to two or more straight lines. Each of the segments of a non-linear relationship might correspond to a changed set of environmental influences on the differential growth relationship (Huxley 1934). Both the *Pollicipes*-type animals and the rock-type animals seem to exhibit good approximations to a straight line relationship although higher degree polynomials show significant improvement over their respective first degree equations. Large limpets of both types have similar height-length relationships; curves 1 and 2 will eventually intersect again. However, since 15 mm (ln = 5.0) is, in general, the largest size attained by a *Pollicipes*-type limpet, the intersection may have little meaning.

Pollicipes-type limpets are taller at any length than are rock-type limpets. The difference in height of the two forms increases with increasing length to a length of 15 mm where *Pollicipes*-type limpets are, on the average, 2 mm taller than rock-type limpets.

Price-Jones (1930) has shown *Cepaea nemoralis* to be increasingly squat with size, whereas Oldham (1928) found that _Arianta arbustorum_ when deprived of lime grows a lighter weight, and in some cases less peaked, shell. A substrate composed of _Pollicipes_ which is high in calcium might provide a better source of calcium than the rock substrate. Since _Pollicipes_-type and rock-type limpets are of the same species it is likely that they possess similar capabilities for assimilation of calcium. _Pollicipes_-type limpets might be able, because of their substrate, to utilize better their potential for calcium acquisition. Thus calcium availability might be a factor contributing to the greater height of _Pollicipes_-type limpets.

Russell (1907) found that _Patella_ is flatter, smaller, and thicker in exposed than in sheltered areas. Limpets exposed to great desiccation have taller shells than do those living in a more desiccation free environment; a tall shell may provide a water reservoir (Test 1945). This explanation is not particularly attractive in this case since desiccation is probably less on _Pollicipes_ than on the more exposed rock face.

Frank (personal communication) suggests that lateral growth of _Pollicipes_-type limpets may be limited by the size of the animals' resting locus, a _Pollicipes_ scutum plate. This may also be a possible explanation (see below,

Figure 3) of greater height of Pollicipes-type limpets since limitation of lateral growth might be expected to result in increased vertical growth.

METHODS

General data on pattern frequency distribution and size frequency distribution were gathered from a number of *A. digitalis*-inhabited *Pollicipes* colonies, arbitrarily chosen, which were marked so that they could be regularly and repeatably censused at semi-monthly and monthly intervals. All visible limpets on each of these colonies and within 20 cm of the colony were censused. The pattern scores used for all data collected throughout 1967 and 1968 make direct use of field notes on degree of striping and color.

Pattern Scoring

In the field animals were scored numerically from 0 to 6 depending on a subjective estimate of the amount of striping. They were also scored for grey, white, or brown apex, white, grey, brown, or black rim, and for presence or absence of mottling of shell surface. Mottling occurred only on the *Pollicipes*-type animals and is probably a result of injury. It is most likely a feature of rebuilt shell material or of the underlying apical brown nacreous layer characteristic of *A. digitalis*.

In the final analysis mottling was given a score of 1;

the basic colors, for rim and apex, were scored as follows: white = 0, grey = 1, brown = 2. Final score was calculated by adding scores for striping, apex color, rim color, and mottling. Since shade is a minor function of shell wetness, an attempt was made to census on dry days. Using this system, the Pollicipes-type limpets scored in the low ranges whereas rock-type animals received higher scores.

It should be understood that the above scoring method has merely a qualitative visual basis. Numbers used are ranks. Manner of manipulation of the raw scores to arrive at final pattern score was purely a matter of choice and convenience. Limpet types are illustrated in Appendix B.

Available methods of pattern scoring give rise to a possible statistical problem. The distribution of scores comprises a variable, the intervals between the values of which are not definitely known to be equal. Such a variable is ordinal and is amenable only to non-parametric tests for comparison of population means such as sign test, Wilcoxon rank test, etc. (Tate and Clelland 1957, Stevens 1968). Savage (1957), Humphries (1964), and Anderson (1961), however, feel that the interval question is of no real importance. After consultation with statisticians, I concluded that the t test is sufficiently robust to withstand small amounts of skewness of data.

The distribution of pattern scores of Pollicipes-type limpets is approximately normal (Figure 6A, p. 56, and 6B, p. 58). An approach to normality is also shown in Figure A (Appendix), a normal probability plot of pattern scores of the limpets of one of the standard sample colonies. That the distribution does not include enough dark limpets is probably of no concern. Therefore, it is likely that the _t_ test and the other parametric statistics used in the following analyses are valid.

Measurement

Length, and in some cases width and height, of _Acmaea_ were measured to the nearest 0.1 mm using a knife-edged vernier caliper. Length was measured with the animal _in situ_.

Data on pattern and size were used in analysis of selection intensity and, with some modifications, in behavior experiments. These modifications, as well as more specialized methods and statistical tests used in data analysis, are discussed under appropriate sections of results.

EXPERIMENTS AND RESULTS

Shell Color

Shell pigments in archeogastropods are probably porphyrins (Morton 1958, Fox in Wilber and Yonge 1964, Hyman 1967). Food quality controls the colors of some gastropods (Ino 1940, Leighton 1961, and Moore 1937).

In an attempt to answer the question of control of shell color in rock- and *Pollicipes*-type limpets, the following experiment was performed. All visible *A. digitalis* were removed from a number of *Pollicipes* colonies. These limpets were then marked, by color coding with paint, for their original pattern score. Next their shells were broken in such a way that approximately one spring's growth was removed from the anterior half of the shell. Treated animals were placed on a rock on which there were no *Pollicipes*, and animals were censused at two week intervals until new growth of an extent sufficient for pattern scoring was present.

Regrown shell material was highly variable in pattern relative to that found in untreated shell. Light animals most often regrew shell not substantially different from the original, whereas the new shell of darker forms tended to be the same as or darker than, but never lighter than, the original. Color and pattern, then would seem to be in the first degree genetically determined although there must be a fairly strong environmental component of variance.

It may be assumed that the algal flora of the sandstone differs qualitatively from the epiphytes of a Pollicipes polymerus colony. It is also obvious that different ions will be available on the alternative substrates. The response was more variable in originally dark animals. This means that genetically determined bounds of phenotype vary with original phenotype. However, basic color must be affected by substrate composition and/or food quality.

About 1% of the limpet shells found on the rock habitat have light posteriors and dark anteriors. In all cases noted, the light area comprised about one third of the shell surface and extended in the expected triangle from apex to base. This pattern of variation within a shell is found repeated in the obverse in a small proportion of Pollicipes-type animals. The presence of these possibly mosaic animals may lend credence to the conclusion that pattern is under primary genetic control. Unfortunately there is no way at present to determine if the above is really genetic mosaicism.

Behavior

Although light-colored Acmaea occurred almost invariably on Pollicipes colonies, on two occasions during the summer of 1966, the animals of a Pollicipes colony on a damp, rainy, or foggy early morning were found on the colony in much lower frequency and on the surrounding rock in much higher frequency than normal. As the rocks dried, the animals were observed returning to the Pollicipes colony.

The light Acmaea in this study seem to have a much more well defined home locus than normally reported for Acmaea digitalis (Frank 1964, Galbraith 1965). Since a rather thorough knowledge of their home range was deemed a necessary adjunct to other phases of the study, an attempt was made to quantify this behavior.

Individual Pollicipes and their resident limpet were color coded with drops of paint. These 162 animal pairs were censused first at daily, and later at semi-monthly intervals. At each census location of each limpet relative to its original position was noted. Each was scored as being on its original Pollicipes, within two animals of the original, or away from the original. "Away" in this case means either that the animal had died or that it was not within two Pollicipes of the marked Pollicipes.

Although over one half of the marked pairs separated rather quickly, in only a few cases did an Acmaea leave the colony on which it was originally found. In these cases the animals were located on an adjacent Pollicipes colony which, with the original colony, was part of an extensive mussel, Mytilus californianus, bed. In the remainder of cases, when a marked pair did separate, the Acmaea was usually found within two Pollicipes of the original, on an animal of similar size to that vacated.

Other information substantiates the contention that Pollicipes-type limpets move little, if at all, from a home Pollicipes colony. The visible limpet population of a colony was removed, placed on adjacent rock, and marked. Also marked were the limpets found on all Pollicipes colonies located in the immediate vicinity of the experimental colony. The colony was later found to be repopulated both by its own marked animals and by unmarked animals. None of the animals that had been marked on the adjacent colonies had moved to the cleared colony. However, it is probable, since there is no obvious wandering population, that the "new" animals originated within the experimental colony and were overlooked at the start of the study; repeated removal of visible limpets from a given colony results in a rapid decrease of limpets on that colony. In no case were limpets native to the rock face found to have moved onto the vacated colonies. Thus, ignoring mortality, the A. digitalis population of a Pollicipes colony is relatively stable and, therefore, periodic samples of the standard colonies referred to later are comparable. Numbers of limpets "invisible" at any sampling time are probably low since the repopulation noted above was of small numbers of limpets.

The above and transplant experiments in which limpets did not remain in the "new" area suggest that the two types of limpets are limited behaviorally to their respective substrates. A comparative study of movement was therefore made.

Three experiments were conducted. In the first, Pollicipes colonies were haphazardly chosen for the first test of the series (August 1967), an attempt being made to use colonies representative of all intertidal levels and all degrees of background heterogeneity within the range of Pollicipes. Colonies used in the later (January, February 1968) experiments differed in that they were chosen specifically to test predictions based on the August experiments. Some of these colonies were shielded by projecting rock ledges; two colonies were selected to approximate the habitat of sample colonies mentioned later.

In the experiments, all visible limpets were removed from the Pollicipes colonies. The snails were placed on the rock face at a distance of 8 cm from the home colony and were marked with drops of colored paint. In the August 1967 and January 1968 studies, a census was taken only after 24 hours. The third experiment, February 1968, was begun in the same manner as the above but differed from them in that censusing was done every day for 4 days.

At census, pattern scores and sizes were noted for all visible marked limpets. The limpets were either on Pollicipes, on rock, or close to or under the colony. Animals were individually characterizable by size, pattern, and marking so that it was possible to identify those that had returned to the colonies on a given day. The results of the three experiments are presented below in Tables 1 through 6.

Table 1 consists of the results of Student's t tests computed on the mean pattern scores of returnees to the colonies from which they were removed against those of animals which did not return to their colonies. It was

Table 1. Comparison of mean pattern score of limpets remaining on rock vs. limpets which move to *Pollicipes* colonies.

Colony	Description	N off	N on	t	P
1	high, isolated, exposed	4	18	9.36	<.001
2	high, exposed, *Mytilus*	6	27	12.45	<.001
3	low, large *Balanus*	4	24	3.21	<.01
4	high, isolated, exposed	2	5	2.69	<.05
5	low, large *Balanus*	2	17	2.27	<.05
6	low, unexposed, *Balanus*	7	3	1.24	>.10
7	high, bare rock, deep cleft	4	4	1.39	>.10
8	high, wet	9	7	3.16	<.01

Eight other colonies had complete first day return.

constructed from data collected in August 1967.

Considering the high exposed or exposed colonies, 1, 2, 3, 4, and 5, one sees in Table 1 that proportion of return was invariably high and that those animals which did not return were different (darker) from those which returned. Conversely, the animals of low or sheltered colonies (6, 7, and 8) returned in lower frequency (heterogeneity χ^2 for number off:number on = 28.1; d.f. = 7; $P < .005$). In colonies 6 and 7 returnees were not significantly lighter than nonreturnees. The presence of large Balanus, colonies 3, 5, and 6, at low intertidal elevation may also reduce the degree of differentiation of returnees' and nonreturnees' pattern scores. Table 1 represents only 8 of 15 original colonies; the other 7 had complete first day return. These latter colonies were all located in exposed areas.

Frequency of return and differences in mean pattern scores of limpets of a colony seems not to be a function of intertidal level per se. Colony pairs (2, 7) at high and pairs (6, 5) at low levels exhibit differences in return which must be independent of intertidal level.

Further information is available from Tables 2 and 3 in which variances of pattern scores of limpets which returned during the first time interval are compared between colonies. For all colonies, d.f. = 14, Bartlett's test

Table 2. Variances about the means of shell pattern scores of limpets which returned to *Pollicipes* colonies.

Colony	s^2	N	Description
1	0.96	18	exposed, homogeneous background, high
4	0.29	5	"
14	0.74	4	"
15	0.35	44	"
8	0.43	7	wet
13	0.63	14	*Mytilus*, low
11	0.41	11	high, homogeneous background
12	1.33	28	*Mytilus*, large *Balanus*, low
2	0.98	27	vertical face
3	0.78	24	low, large *Balanus*
5	1.30	17	"
6	0.99	3	"
9	3.00	5	overhung by rock ledge
10	3.18	8	low, exposed

gives $\chi^2 = 45$, $P < 0.0001$, indicating that the variances in question are not homogeneous. From Table 2 and its colony descriptions, one can see that colonies either exposed and located on areas of relatively homogeneous rock (1, 15) or colonies associated with *Mytilus* (11, 13), with no other disturbing factors, exhibit low variances of returnees' pattern scores. Conversely, those colonies which are either associated with *Balanus* or are located under ledges (2, 3, 5, 10, 12) show large amounts of variability in pattern score.

The heterogeneity of variances has two probable origins. First, initial variances of animals within colonies were different; second, pattern-specific returning response of transplanted animals differed between colonies. These two factors obviously confound one another. Probably both contribute to the observed differences. It will be shown later that these two factors are related.

Table 3. Colonies in column 2 are negatively associated with regard to variances about mean pattern score of limpets returned by F variance ratio test with colonies in column 1. P (1 tailed) $<.025$.

Arbitrary colony	Negatively associated with arbitrary colony
10	15, 11, 13, 3, 8
1	10
15	10, 3, 1, 2, 12, 5

The construction of Table 3 made use of the variances in Table 2. Three colonies were arbitrarily chosen. Of these, 1 and 15 were high and exposed, and 10 was low with a heterogeneous background of Balanus, algae, pocked rock, etc. The variances of pattern scores of returned limpets of each of these colonies were compared in turn with this value for each of the other colonies in the study. An *F* variance ratio test was used. Colonies found to have variances significantly different, $P < .025$, from each of the chosen colonies are grouped in Table 3 opposite the colonies with which they were compared. By referring to Table 2, the colonies of limpets are shown to be divisible in this way into the two categories: high, exposed colonies with low variances and low, sheltered colonies of heterogeneous background with high variances. This indicates that there are differences both in the Acmaea colony pattern composition and in variability of Acmaea behavior. These differences are related to differences in habitats of the home Pollicipes colonies.

Table 4 is of frequencies of return within 24 hours of limpets of each of several pattern scores. There is a negative relationship between pattern score and Pollicipes-seeking homing propensity.

Table 4. *Pollicipes* affinities (measured by the probability of return to a colony within 24 hours) as related to pattern score.

Pattern score	# on	Total	% on
0	31	33	94
0.5	23	27	85
1.0	47	55	85
1.5	36	42	86
2.0	53	62	85
2.5	12	16	75
3.0	4	7	57
3.5	4	6	67
4.0	0	2	0
4.5	2	4	50
5.0-6.5	0	0	-
7.0	0	2	0

The above results are confirmed in Table 5 in which mean pattern score of returnees and mean pattern score of animals remaining on rock are compared for both large animals only and for all animals of each colony treated. Data are from the January 1968 experiment in which limpets had been moved from *Pollicipes* to adjacent rock. Student's *t* test was used as the method of comparison; the significance level was $P < .05$. Also compared for each colony in Table 5 are a number of variance ratios.

Colonies T, M, and B are a vertical series in that order from top to bottom. M and B were on a vertical surface. Thus in column 2 the highest colony, T, shows complete return, but there was no return in the lowest colony, B. The colony in the middle, M, showed intermediate results. M2, M3, and M4, also high and exposed, give results similar to those for M. M2 and M3 are both higher and more exposed than M. M5, part of the same group of colonies as M4, but lower, is the only colony with mean pattern score "on" unequal to mean pattern score "off" Pollicipes.

It is interesting to note that inclusion of small ($<$7.0 mm) limpets completely obliterates differences in mean pattern scores of limpets on and off Pollicipes.

There may be seasonal differences in response since proportion of removed limpets returning to Pollicipes was lower in January than in August (Table 1). This may merely be a function of the smaller average size of limpets in January (see below, Figure 4).

Data of the third experiment (February 1968) were treated with multiple regression analyses (IBM Scientific Subroutine "Regrel"), an analysis of size of limpet, return time, and pattern score being completed for each colony in question and for subjectively grouped colonies. The analyses were completed using the IBM 360-50 computer at the University of Oregon. Table 6, for each of 11 colonies, presents regression coefficients of the time-pattern segment of the multiple regression analysis and significance level. Mean pattern scores on and off Pollicipes are compared in the t_{mean} column; N_{on} and N_{off} and first day return frequency are also noted.

Table 6 must be interpreted in the light of both re-

Table 5. Numbers of returnee (r) and nonreturnee (n) limpets whose means and variance of pattern score were tested. *denotes a significant difference between mean (t-test) or variance (F-test) of pattern score among returnees vs. nonreturnees.

Colony	Test of means		Tests of Variance				Proportion r
	No.large (7 mm) (n, r)	No. all animals (n, r)	No.large vs. small r only	Large; no. n vs. r	Small; no. n vs. r	All animals n vs. r	
T	--	2,9					0.81
M	14,7	14,7	no small	14,7	no small	14,7	0.33
B	--	7,0					0
M2	24,3*	37,7	3,4	24,3	13,4	37,7	0.16
M3	12,7*	25,14	7,7	2,7	13,4	25,14	0.36
M4	0,6	12,8	6,5	0,6	9,5	12,8	0.60
M5	--	6,13*					0.68
SS	20,7*	25,9	7,2*	20,7*	4,2	25,9	0.27
SY1	3,5	23,6	5,3	3,5	3,18	23,6	0.20
SY2	15,0						0
SY3	6,3*	10,4	3,1	6,3*	4,1	10,4*	0.29

Table 6. Relationships between pattern score and return time. Mean pattern score of returnees has been compared with that of limpets remaining on rock by student's "t". Significance of the relationship is given in column 6. Regression coefficients of pattern on time are given in column 7. Their significance is given in column 8.

Colony (1)	Description (2)	N off (3)	N on (4)	1st day prop. (5)	s means (6)	r (7)	s (8)
1	low, het. back	39	9	0.19	ns	0.06	<0.01
2	vertical face	31	9	0.23	ns	0.02	<0.05
3	high, cliff face	33	11	0.25	< 0.05	0.44	ns
4	low, het. back	40	22	0.25	ns	0.01	ns
5	low, sheltered	17	8	0.32	ns	0.01	ns
6	high, exposed	15	9	0.38	ns	---	--
7	high	20	7	0.26	<0.05	-0.01	ns
8	series of high colonies	12	8	0.40	<0.05	-0.10	ns
9	high, small colonies	16	6	0.27	<0.10	0.03	ns
10	high, vertical	12	8	0.40	ns	-0.01	ns
11	low, open, homogeneous back.	77	15	0.16	<0.05	0.02	<0.10

turn frequency and first day t test of mean pattern of returnees vs. mean pattern of nonreturnees. The latter compare favorably with t tests in Table 1 in that significance is exhibited in high or low, exposed colonies, 3, 7, 8, 11. Colonies on vertical faces (A2), those with heterogeneous environs (A1, A4), and isolated colonies (A9) show, as did similar colonies of Tables 1 and 5, no difference between mean score off and mean score on Pollicipes. Return frequency increases with increased degree of exposure.

When only large limpets are considered, significant regressions are found only in the cases of low, heterogeneously-environed colonies. In high colonies or low, exposed colonies, there is no correlation between an animal's pattern score and the time taken to return to Pollicipes. Rather, as with Table 1, the relationship is more or less absolute, almost all animals returning the first day.

In animals of less than 8 mm length, pattern and behavior are correlated, $y = .237 + .72x$, $P < .05$; return time increases with limpet darkness.

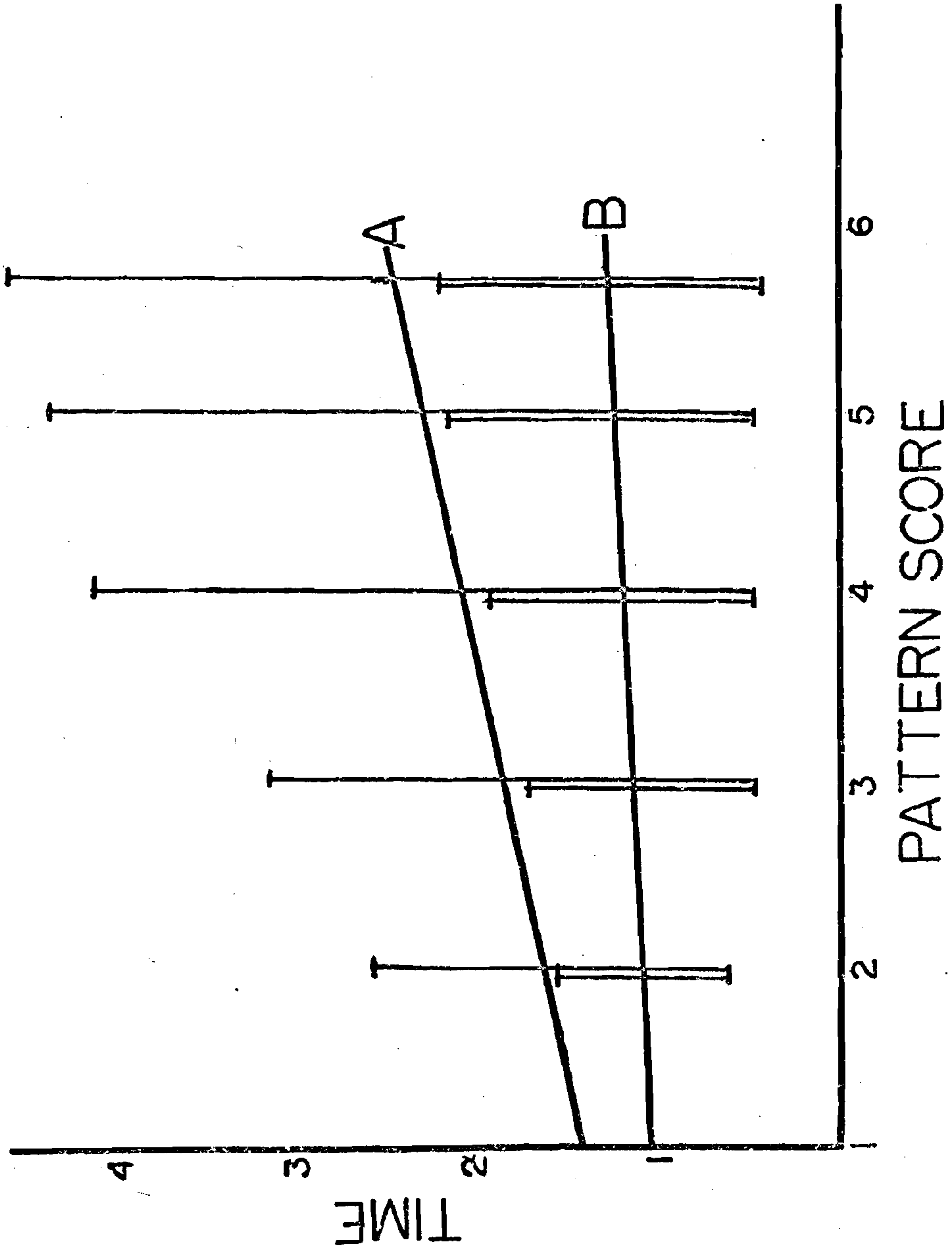
Although there seems, at first, to be a conflict between t test, variance, and regression analysis (one might expect a significant t test to be associated with a significant regression coefficient rather than with an insignificant regression coefficient, a logical explanation exists.

One can construct a model of the above situation. Consider Figure 2, line A, as representing a population of young *A. digitalis*. Time of return (or *A. digitalis-Pollicipes* affinity) is regressed on *Acmaea* pattern. Since both characters are undoubtedly polygenic in origin, a relatively great amount of variance in the proposed relationship must be included. Two separate situations must now be considered. In the first, line A, the model colony is low in the intertidal resulting in less time for predation and/or less exposure to predation. Selection is therefore roughly at random with regard to pattern and to the pattern-*Pollicipes* affinity interaction. As a result, the original relationship, shown in Figure 2, increased time of return with larger pattern score, remains unchanged.

In the second case, line B, the model colony is high and exposed. Predation is intense and directed toward those animals which do not fully utilize behaviorally their potential inconspicuousness. As a result dark *Pollicipes*-type animals with small affinity for *Pollicipes* are selected. The result is that *Pollicipes*-type animals of all patterns have equal affinity for *Pollicipes*.

The model fulfills conditions found in the real population of young animals. Return time is linearly correlated with pattern, and means can, because of large variance be apparently the same. The model also explains the different mean pattern scores but insignificant *r* values (pattern,

Figure 2. Model of possible relationships of pattern score and *Pollicipes* affinity of *Pollicipes*-type limpets. Plotted are hypothetical regressions of *Pollicipes* affinity on limpet pattern. Vertical lines indicate standard error; line A represents young limpets or low intertidal, sheltered, relatively unselected limpets; line B represents exposed, selected limpets.


TIME
1
2
3
4
PATTERN SCORE
2
3
4
5
6
A
B

time), the situation in high, exposed colonies. According to the model (line B) there are few limpets with low affinity for *Pollicipes* left after selection. These are mostly dark and account for first day mean score on being different from mean score of limpets off *Pollicipes*. However, since first day return is 90 to 100% and since, as the model states, affinities over the pattern range are approximately equal, too much variance exists about day 1 for a regression coefficient to have significance.

The composite picture is one in which large *Pollicipes*-type *Acmaea digitalis* in high or otherwise exposed areas exhibit rapid homing to *Pollicipes* regardless of pattern score. At low tidal levels, there is still (Table 6) a suggestion of a relationship of homing propensity to pattern darkness.

Evidence from another population (Sunset Bay) may confirm the hypothesis that selection acts at different levels of intensity to control the above pattern-behavior relationship. Here, the *Pollicipes* colonies spread in a north-south linear array from a center of concentration at the north end of the area to an area of very low population density in the south. In the case of the colony closest to the population center, SY3, Table 5, mean pattern of limpets on *Pollicipes* differs from mean pattern off when large limpets only are considered. There was very little or no return of *Acmaea* to *Pollicipes* in the other colonies considered.

Considering colony SY 3, the pattern of *F*-test results is also interesting. In the case of the *F*-test of *large on-small on*, the variance of patterns of small animals is greater, $P < .05$. Considering *F large on-large off* and *F all on-all off*, variance of animals off the colonies is greater, $P < .05$. *Small on-small off* variances are equal. These first day results seem to fit the above model and correspond to the case of a population of young unselected limpets or of a population under low selection pressure. The picture presented by colony SS is similar to that shown by SY3. Only 27% of the limpets of SS returned on day 1. There seems, qualitatively, to be no difference between rock- and *Pollicipes*-type limpets in the case of SS. Movement appears to be at random.

SY 1 and SY2 are small, remote colonies; there seems little or no affinity for *Pollicipes* in these cases.

In low areas large *Balanus cariosus* are associated with the *Pollicipes* and *Mytilus.* Small *A. digitalis* often establish an association with the *Balanus* which have a grey parasitized apex and a clear white base. The *Acmaea* in such a situation may cue to color rather than shape or some other substrate attribute such as periostracal protein.

Large *A. digitalis* (over 8 mm in length) were never observed in association with these animals.

Size-Behavior Relationships

There is probably a relationship between animal size and propensity to seek *Pollicipes* which is confounded by pattern-behavior correlation. A regression of *Acmaea* size on time taken by that animal to return to a *Pollicipes* colony, using data of the low colonies of the March study, gave the equation $y = 141 - 22.83x$, $F = 1.913$, d.f.(F) = 1,201, $P < .80$. Thus the results are not significant. It might be suggested that selection favors *Pollicipes* affinity in *Pollicipes*-type limpets.

Size *Acmaea*-Size *Pollicipes* Relationship

It was early noticed that the size of limpets seems to be positively related to the size of the *Pollicipes* on which they live. Figure 3 shows a curvilinear relationship between *Acmaea* size and home locus size. As limpet size (length) increases so does the size (depth of scutum) of the *Pollicipes* on which the *Acmaea* is found. See Table 7.

The size of a limpet is obviously related to the size of the *Pollicipes* on which it is resident. That the relationship is not linear demonstrates that the relationship of limpet to *Pollicipes* size is critical to large limpets

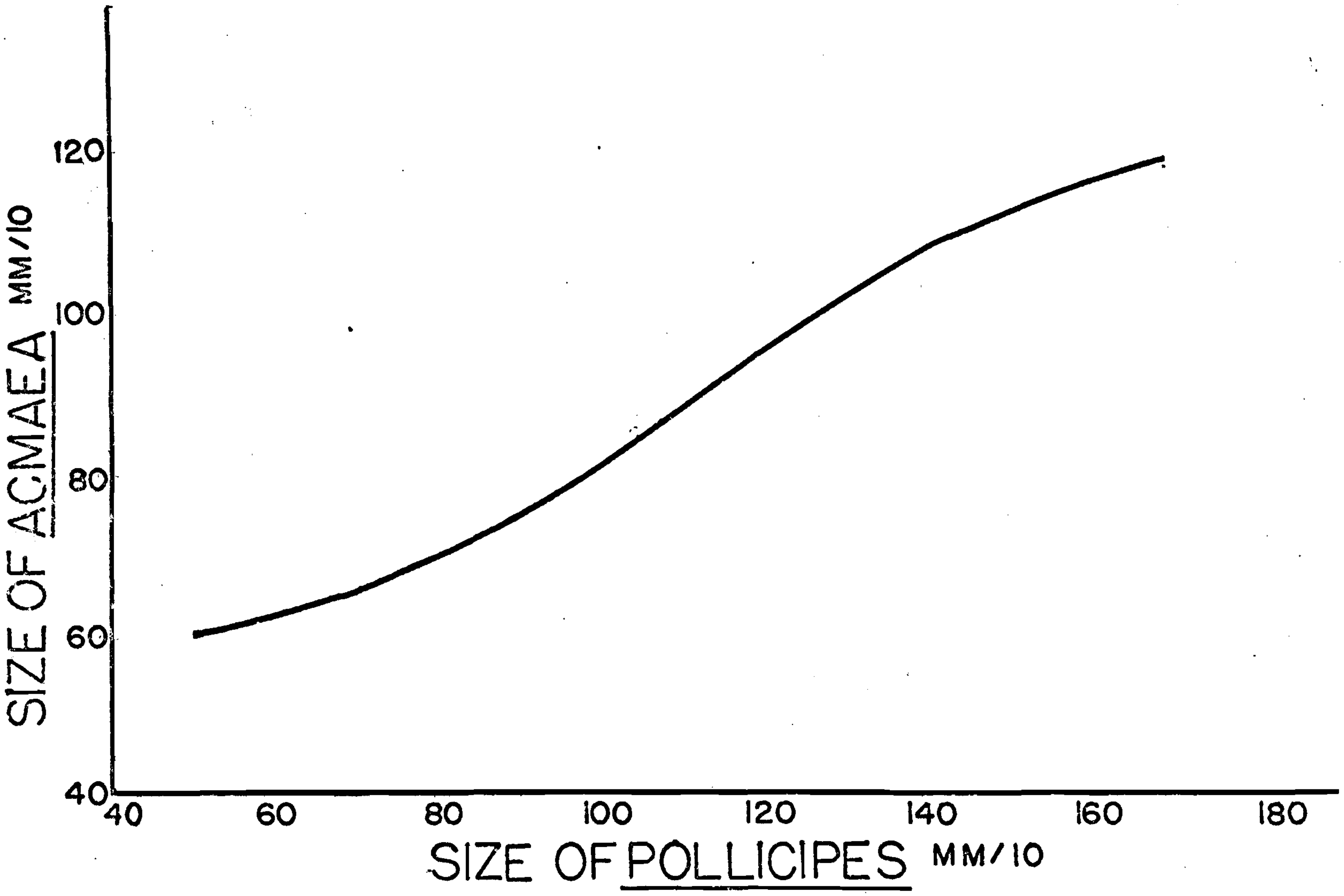
Table 7. Analysis of variance for the regression of Figure 3.

$$y = 86.713 - 1.23887 + 0.01697x^2 - 0.00005x^3$$

	d.f.	Sum of squares
Due to degree 1 regression	1	68013
Due to degree 2 regression	1	6519
Due to degree 3 regression	1	2604
Deviation about regression	185	119768
Total		196903

$y = \frac{mm}{10}, x = \frac{mm}{10}$

Figure 3. Regression of size of limpets on size of _Pollicipes_ inhabited by the limpets. Measurements are of limpet length and _Pollicipes_ scutum length. Units are mm/10.



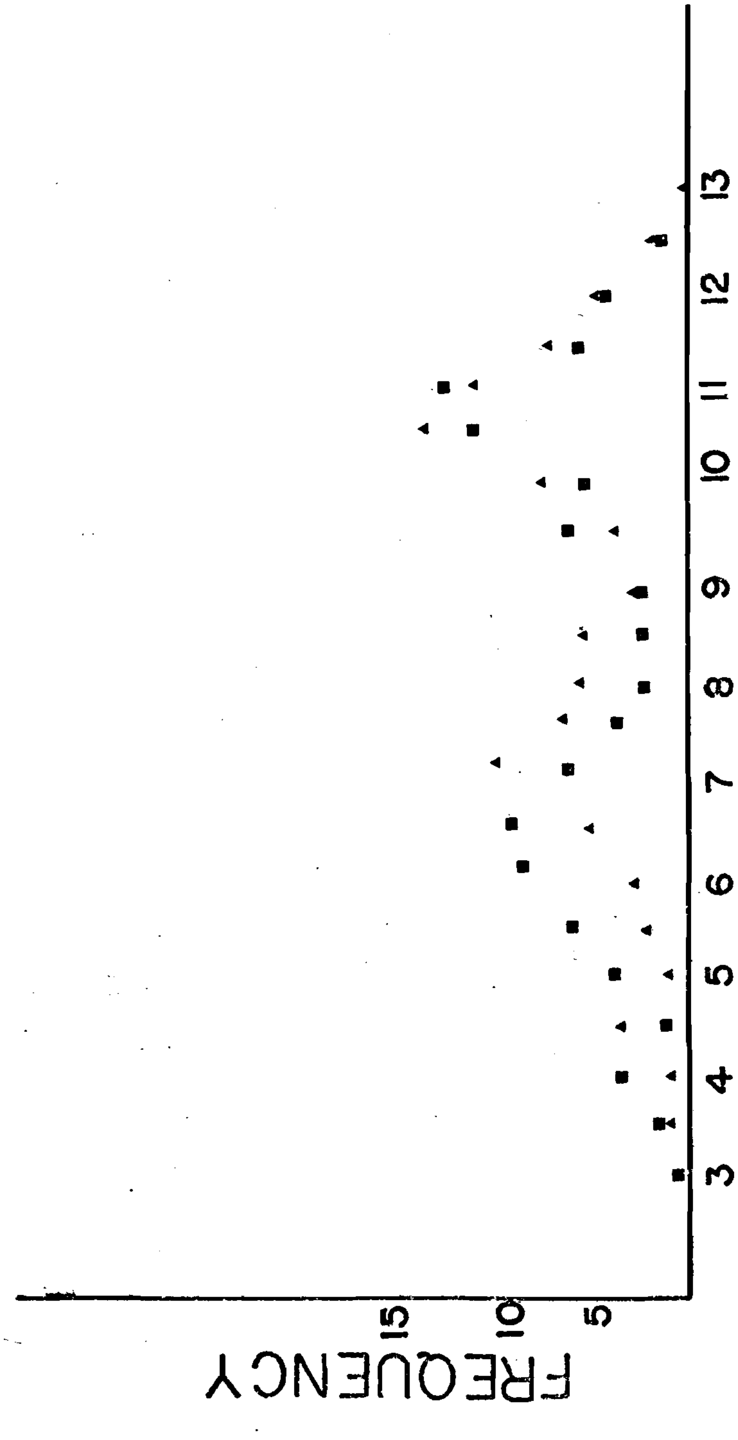
SIZE OF ACMAEA MM/10
120
100
80
60
40
40
60
80
100
120
140
160
180
SIZE OF POLLICIPES MM/10

and that large limpets require as homes proportionately larger barnacles than do smaller limpets. In the Cape Arago region *Pollicipes* seldom exceed 17 mm in scutum length. Thus, especially since the limpet size-*Pollicipes* size curve becomes steeper with increased size of *Pollicipes*, but, at *Pollicipes* size 15mm, noticeably becomes less steep, the size of limpets on *Pollicipes* must be limited by the size of the largest available barnacle. Maximum limpet size must be about 14.0 to 14.5 mm length. In fact, limpets larger than this are seldom found on *Pollicipes*; *Pollicipes*-type limpets larger than 14 mm are relatively common on the rock in sheltered or low areas.

There tends to be a linear relationship between the size of the commensal scale worm *Arctonoë vitata* and the size of its host *Diodora aspera* (Palmer 1968). Similarly, the hermit crab, *Pagurus* sp. chooses homes, mollusc shells, appropriate to its size. One might expect the *Pollicipes* form of *Acmaea digitalis* also to choose *Pollicipes* individuals of an optimum size on which to live. This expectation is based on the observation that *A. digitalis* lives in the high intertidal zone (Rickets and Calvin 1962) and must therefore be subject at low water to rather extensive desiccation (Frank 1965). Other animals of the zone, when located in crevices in the rock are able to exist at higher levels than their counterparts living on the open rock face (Kinsler 1967). Similarly, rock-type *A. digitalis* and *A. paradigitalis* tend to aggregate in the depressions in the rock where they presumably are subject to less severe desiccation than otherwise (Frank 1965, Millard 1968). *Pollicipes*-type *A. digitalis* have a tall shell with a small aperture and a complete rim. It is easy to hypothesize that the complete rim, in conjunction with a mucus-producing foot and mantle edge, when combined with the relatively smooth plates of a *Pollicipes* results in a fairly watertight seal against desiccation. The effectiveness of this seal can only be optimized by an appropriate relationship between the limpet's aperture size and the average size of the *Pollicipes* plates on which it lives.

Pollicipes polymerus may live for about 20 years and grows slowly during this span (Barnes and Reese 1960); growth is about 0.5 mm per year. *Acmaea digitalis* has a life span of about 4 to 6 years, growing rapidly (Frank 1965). Using Figure 4, consisting of plots of frequencies of sizes of *Pollicipes*-type limpets for January 14 and May 1, 1968, it is possible to determine that limpets of up to about 8 mm grow about 0.5 mm/3.5 months. *Pollicipes*-type limpets probably grew very slowly in the summer as

Figure 4. Size frequency distribution of _Pollicipes_-type _A. digitalis_. ■ is of January 1968; ▾ is of May 1968.


FREQUENCY
5
10
15
3
4
5
6
7
8
9
10
11
12
13
LIMPET LENGTH MM

was found (Frank 1965) for rock-type limpets. Therefore, yearly growth rate may be calculated on the basis of 10 months and equals about 1.5 mm per year. This rate is considerably less than Frank found for small rock-type *Acmaea digitalis* in a nearby area.

Size frequency is an extremely poor method of growth estimation. The above rate is therefore meaningless except for purposes of qualitative comparison. The great difference noted does, however, suggest a real difference in growth rates of the two forms.

Pollicipes-type limpets appear to have a growth rate adjusted to size of *Pollicipes*, their host; food is probably not limiting since these snails feed on rock as do faster-growing limpets. *Pollicipes*-type limpets larger than 14 mm (the largest size common on *Pollicipes*) inhabit sheltered areas of the rock face. Since few intermediate-sized *Pollicipes*-type limpets are found on the rock, these large limpets may represent either an unselected population or animals which have outgrown the *Pollicipes* substrate. Since Figure 3 shows size of limpets as an increasing function of size of *Pollicipes*, the latter explanation is more likely.

In *Partula*, the snail, a limitation on shell size is inherited (Murray 1968). Murray has shown shell size to

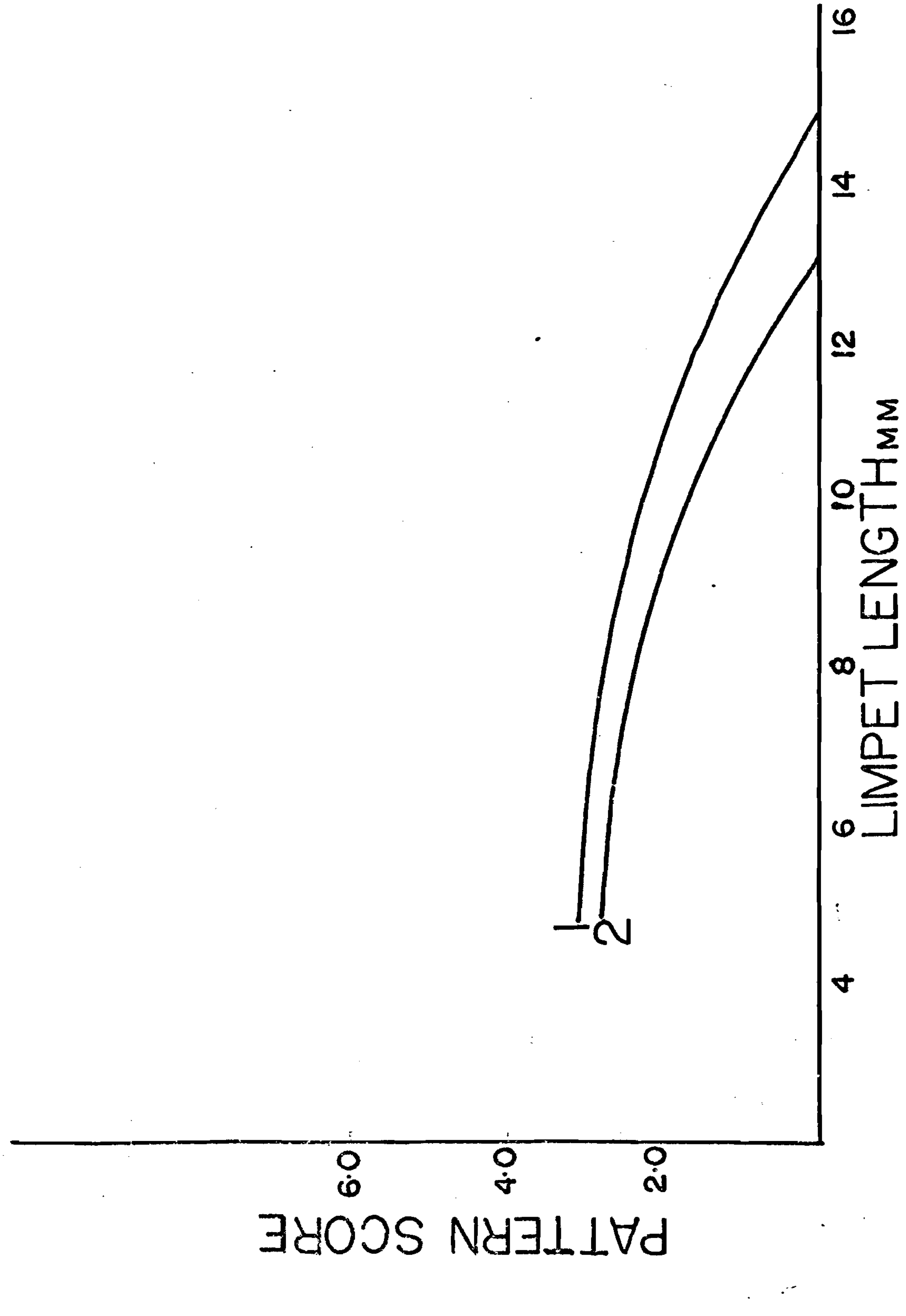
Table 8. Analysis of variance for the regression of Figure 5, curve 2.

$$y = 15.92061 + 0.4024x - 0.00281x^2$$

	d.f.	Sum of squares
Due to degree 1 regression	1	1858
Due to degree 2 regression	1	518
Deviation about regression	204	30496
Total		32873

$x = \frac{mm}{10}$

Figure 5. Relationship of pattern score and length in Pollicipes-type *Acmaea digitalis*. Curve 1: low, sheltered colonies; curve 2: high exposed colonies.


PATTERN SCORE
6.0
4.0
2.0
LIMPET LENGTH MM
4
6
8
10
12
14
16
1
2

have a high heritability. Similarly, Cook (1965, 1967) has shown shell size in <u>Arianta</u> <u>arbustorum</u> and <u>Cepaea</u> <u>nemoralis</u> to have heritability of about 60 to 70%. It seems reasonable that growth rate must also have high heritability. Certainly selection pressure for small size and/or slow growth must be quite high in the <u>Pollicipes</u>-type limpets.

Figure 5 is a plot of a curvilinear regression of pattern score on size of <u>Pollicipes</u>-type limpets. Data were again taken from the March homing study. As in all other nonlinear regressions presented, IBM Scientific Subroutine POLRGL was used. See Table 8. Pattern score remains relatively high up to limpet size 10 mm. In larger animals there is a rather rapid decrease in frequency of dark animals found on <u>Pollicipes</u>. Curve 1 is of low, sheltered colonies, whereas curve 2 represents high, open colonies. There is no significant difference between the two curves.

Average pattern score is a nearly linear, decreasing function of size of limpets, dark animals becoming less common. Two explanations present themselves. First, the limpets respond to the <u>Pollicipes</u> substrate, all animals on <u>Pollicipes</u> becoming lighter as they grow. Second, the developmental effect is unimportant and dark animals are being selected against. One might expect that if the first explanation were real, the relationship between size and score would be linear. Nonlinearity lends more credence to the second explanation than the first. The trend of the difference between the two curves may confirm the idea of differential selection pressure against dark *Pollicipes*-type limpets in sheltered as compared to exposed areas.

An attempt in late May to repeat the collection of data for regression analysis of time vs. size and pattern failed. All animals in all degrees of protection returned to the *Pollicipes* colonies during the first day's interval. The analysis was again attempted in early July. High, exposed and sheltered *Pollicipes*-type limpets returned immediately; 75% (N = 90) of those living lower in the intertidal had returned by the end of the first census period. Mean scores were 2.9 "off" and 3.01 "on."

Young Animals: Movement onto Pollicipes

Acmaea veligers are pelagic (Thorson 1950). The smallest animals found on the intertidal substrate were about 2.4 mm long. All of these animals were found on the rock faces. Cursory examination indicates that lightly colored limpets settle in the vicinity of *Pollicipes* colonies; limpets measuring less than 3.5 mm are not found on *Pollicipes* colonies, but larger, lightly colored limpets

Table 9. Percent *Acmaea digitalis* on rock.

		Pattern Score															
			lighter													darker	
	Size	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0	8.5
I.	3.0-3.9 mm	25	50	88	29	81	95	88	80	100	100	71	83	100	—	100	—
	N	(32)	(4)	(16)	(7)	(26)	(21)	(16)	(10)	(7)	(4)	(7)	(6)	(6)	—	(2)	—
II.	4.0-4.9 mm	27	58	42	70	38	60	59	63	81	94	75	100	71	100	100	—
	N	(15)	(12)	(12)	(20)	(29)	(35)	(37)	(16)	(47)	(17)	(28)	(9)	(17)	(4)	(7)	—
III.	5.0-5.9 mm	25	18	15	27	23	42	39	71	84	86	80	95	93	100	50	100
	N	(12)	(11)	(27)	(26)	(53)	(48)	(67)	(41)	(43)	(21)	(55)	(19)	(41)	(3)	(10)	(1)
	Change*																
	I to II	0	0	-	0	-	-	-	0	0	0	0	0	0	0	0	0
	II to III	0	-	-	-	-	-	-	0	0	0	0	0	0	0	0	0

*χ^2 test: 0 = not significant; - = significant difference, $P < .01$.

are characteristic of this habitat.

Rapidity of return of young animals to _Pollicipes_ is correlated positively with pattern score. Juvenile movement trends were studied as follows: Data collected monthly from October, 1967, to March, 1968, were first arranged by size classes (3.0-3.9, 4.0-4.9, 5.0-5.9 mm) and within these classes, by pattern score. Substrates were noted in the raw data. Percentage of total animals found on rock substrate was calculated for each pattern score within each size class. Results are summarized in Table 9.

Clearly, the primary locus of very young limpets is the rock face. However, greater proportions of 3.0-3.9 mm light limpets are found on _Pollicipes_ than rock. Comparison of 3.0-3.9 mm and 4.0-4.9 mm classes indicates little if any change occurring in animals of pattern scores 1 to 2.5, but does suggest a movement toward _Pollicipes_ of lighter animals in the 3.0 to 4.0 mm classes. Changes seen in 4.5 to 8.0 animals are insignificant. Comparison of the first two classes with the 5.0 to 5.9 mm size class shows rather dramatic changes in pattern scores up to and including class 4.0. These larger, light limpets have become largely associated with _Pollicipes_. There has been no _Pollicipes_-directed movement of the darker limpets. Thus, rate of movement from rock to _Pollicipes_ is correlated with pattern. Data are not good enough to distinguish whether this relationship is linear or quantal although a definite discontinuity is seen at pattern score 4.5 indicating that the largest amount of differentiation as to substrate occurs in animals of approximately 4.5 mm.

Population Changes

Through time various attributes of *Pollicipes*-type *A. digitalis* populations, such as behavior-pattern correlation and mean population pattern score, change. The latter observation suggests that the frequency distribution of pattern scores might change temporally.

The histograms in Figure 6A are of frequencies of pattern scores. Animals on *Pollicipes*, solid bars, and on rock, striped bars, were treated separately. The rates of change represented by comparison of the histograms for January 26 and April 29, 1967, suggest that the months of February through April encompass a period of relatively great change in the distribution of pattern frequencies of the populations.

Figure 6B presents histograms of pattern score for 1967-1978. The population is considered in two groups, limpets found on rock and limpets living on *Pollicipes* colonies. Frequencies of pattern scores of limpets were calculated separately for each group and histograms were constructed as above. There is a set of histograms for

Figure 6A. Frequencies of pattern scores of A. digitalis. Striped bars are of limpets found on Pollicipes, solid bars of limpets found on rock. Limpets included were 4.0 to 8.0 mm in length. January through June 1967.

Number of Limpets

Month	On Pollicipes	On rock
January	93	98
March	70	83
April	63	92
June	52	75

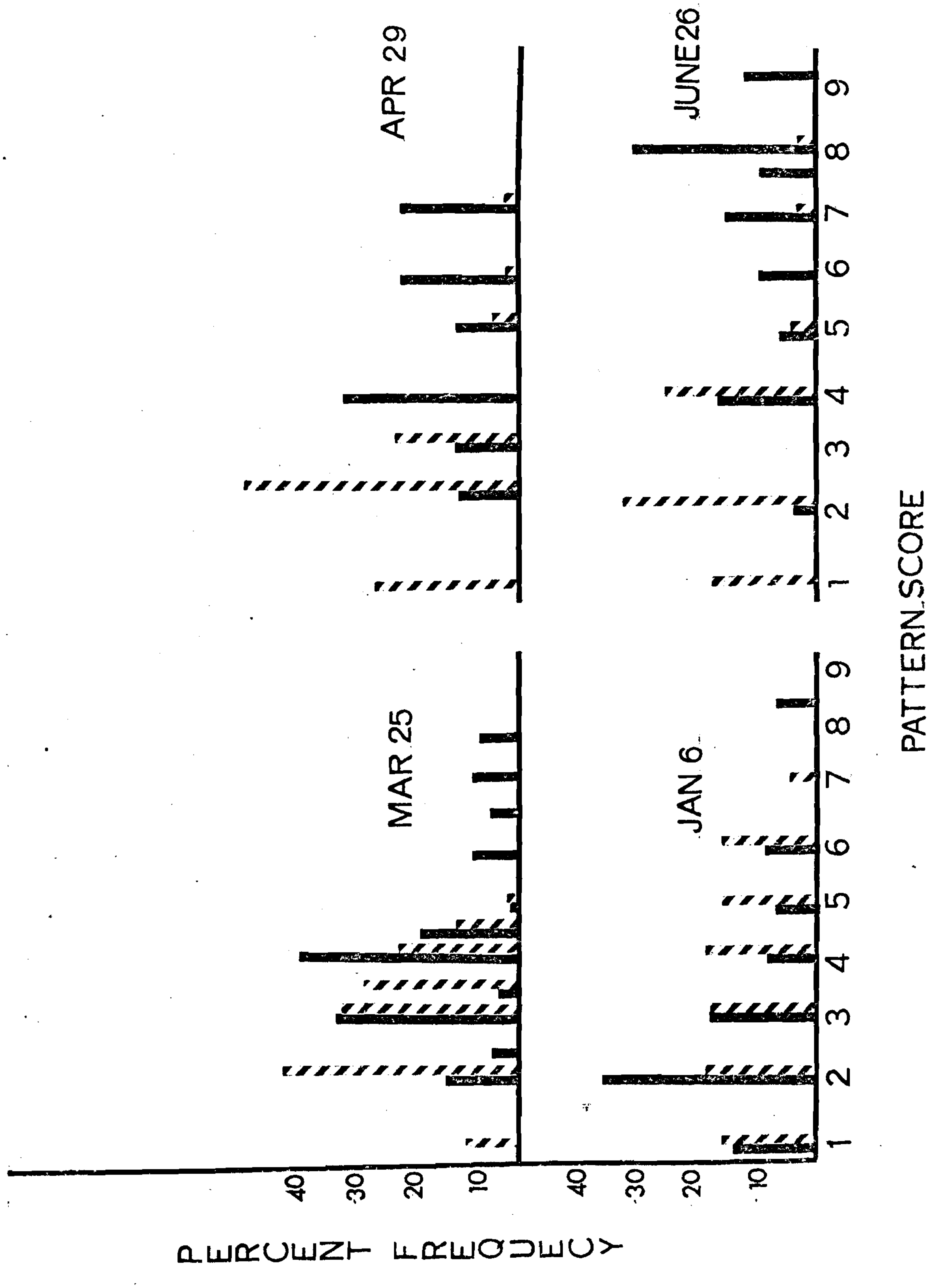
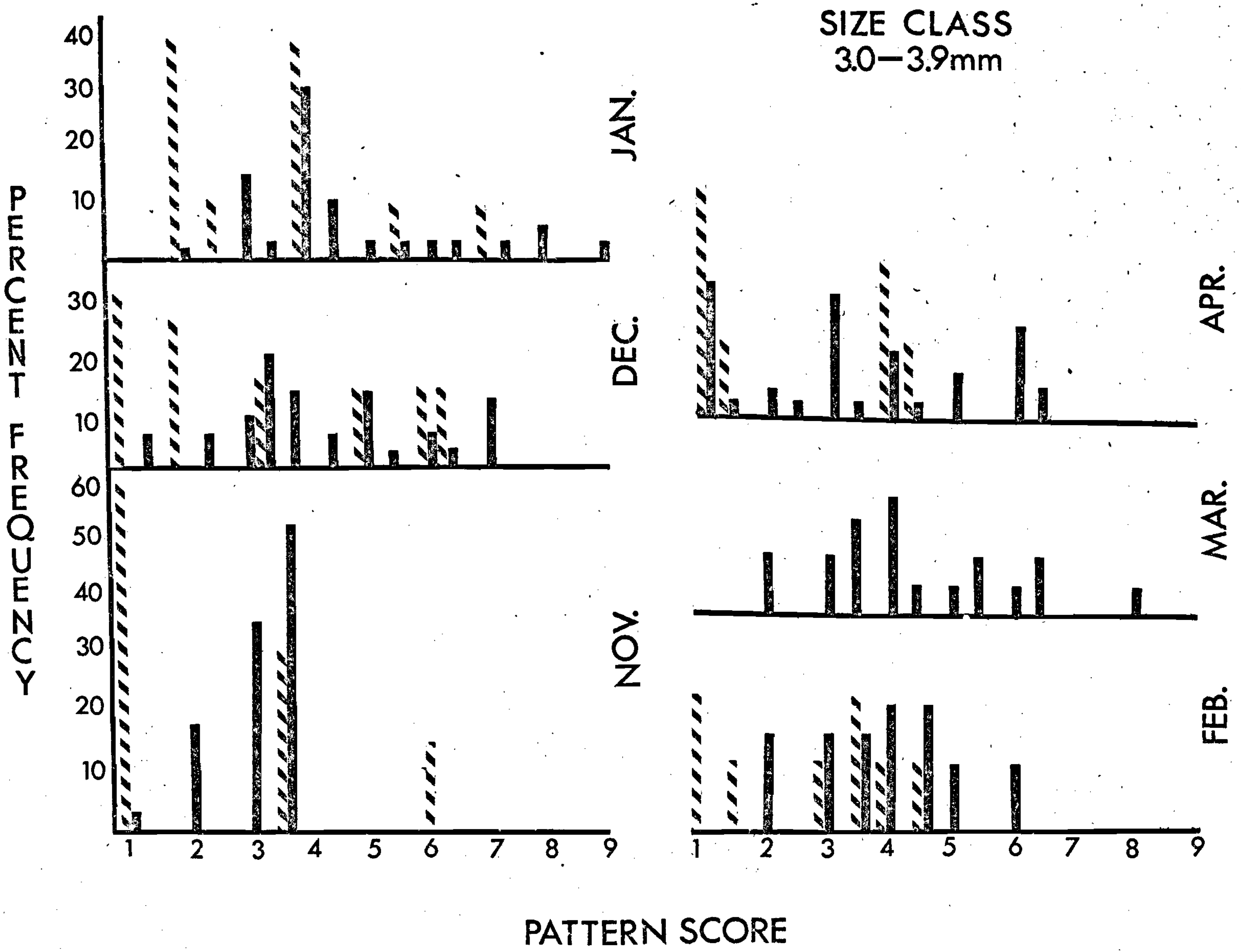

MAR 25
APR 29
JAN 6
JUNE26
PERCENT FREQUECY
40
30
20
10
40
30
20
10
1
2
3
4
5
6
7
8
9
PATTERN SCORE

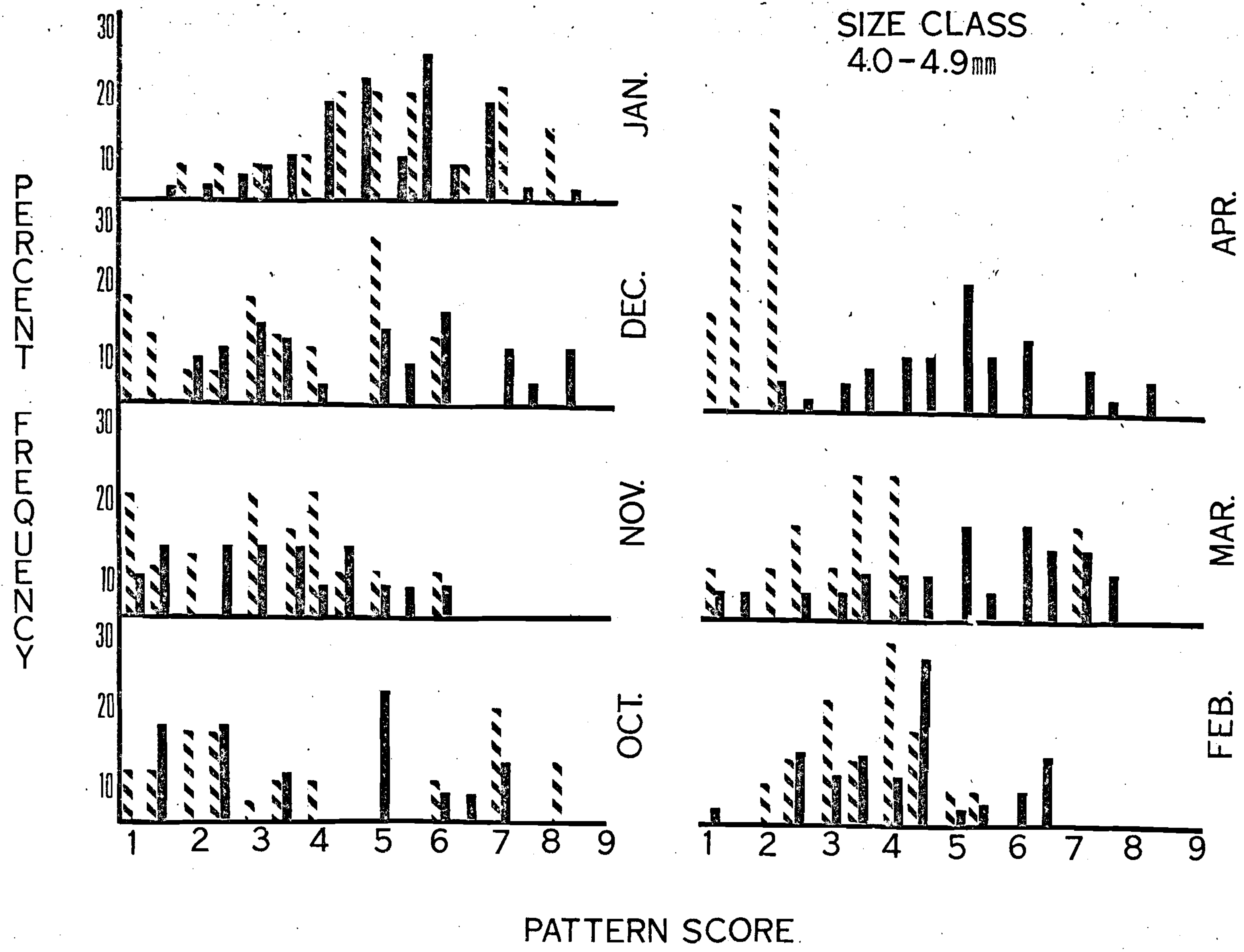
Figure 6B. Frequencies of pattern scores of limpets. Included are months of November 1967 through April 1967 and sizes 3.0-3.9, 4.0-4.9, 5.0-5.9, and 6.0-6.9 mm. Striped bars are of limpets found on Pollicipes, solid bars of limpets found on rock.

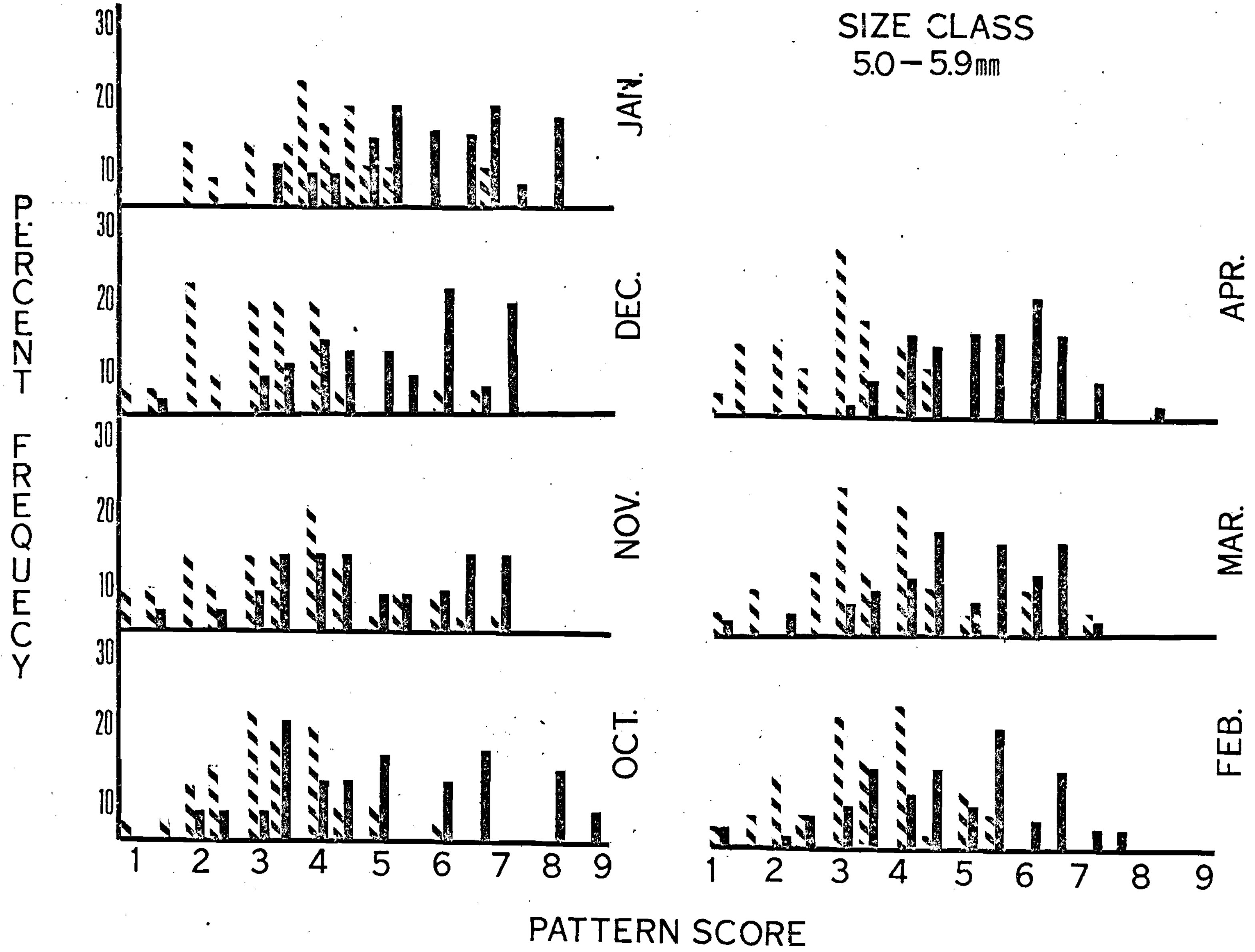
Number of Limpets

Month	Size in mm							
	3.0-3.9		4.0-4.9		5.0-5.9		6.0-6.9	
	P*	R	P	R	P	R	P	R
October	-	-	16	18	19	32	12	10
November	7	6	15	18	42	31	29	24
December	8	32	18	32	32	48	47	14
January	11	35	17	51	30	56	59	32
February	9	21	20	44	42	59	65	33
March	-	19	13	27	29	53	59	25
April	8	38	6	43	25	51	45	24

*P: on-Pollicipes; R: on-rock


SIZE CLASS
3.0—3.9mm
PERCENT FREQUENCY
40
30
20
10
30
20
10
60
50
40
30
20
10
JAN.
DEC.
NOV.
APR.
MAR.
FEB.
1
2
3
4
5
6
7
8
9
PATTERN SCORE


SIZE CLASS
4.0-4.9mm
PERCENT FREQUENCY
PATTERN SCORE
JAN.
DEC.
NOV.
OCT.
APR.
MAR.
FEB.
30
20
10
1
2
3
4
5
6
7
8
9


SIZE CLASS
5.0 – 5.9mm
PERCENT FREQUECY
30
20
10
JAN.
DEC.
NOV.
OCT.
APR.
MAR.
FEB.
1
2
3
4
5
6
7
8
9
PATTERN SCORE

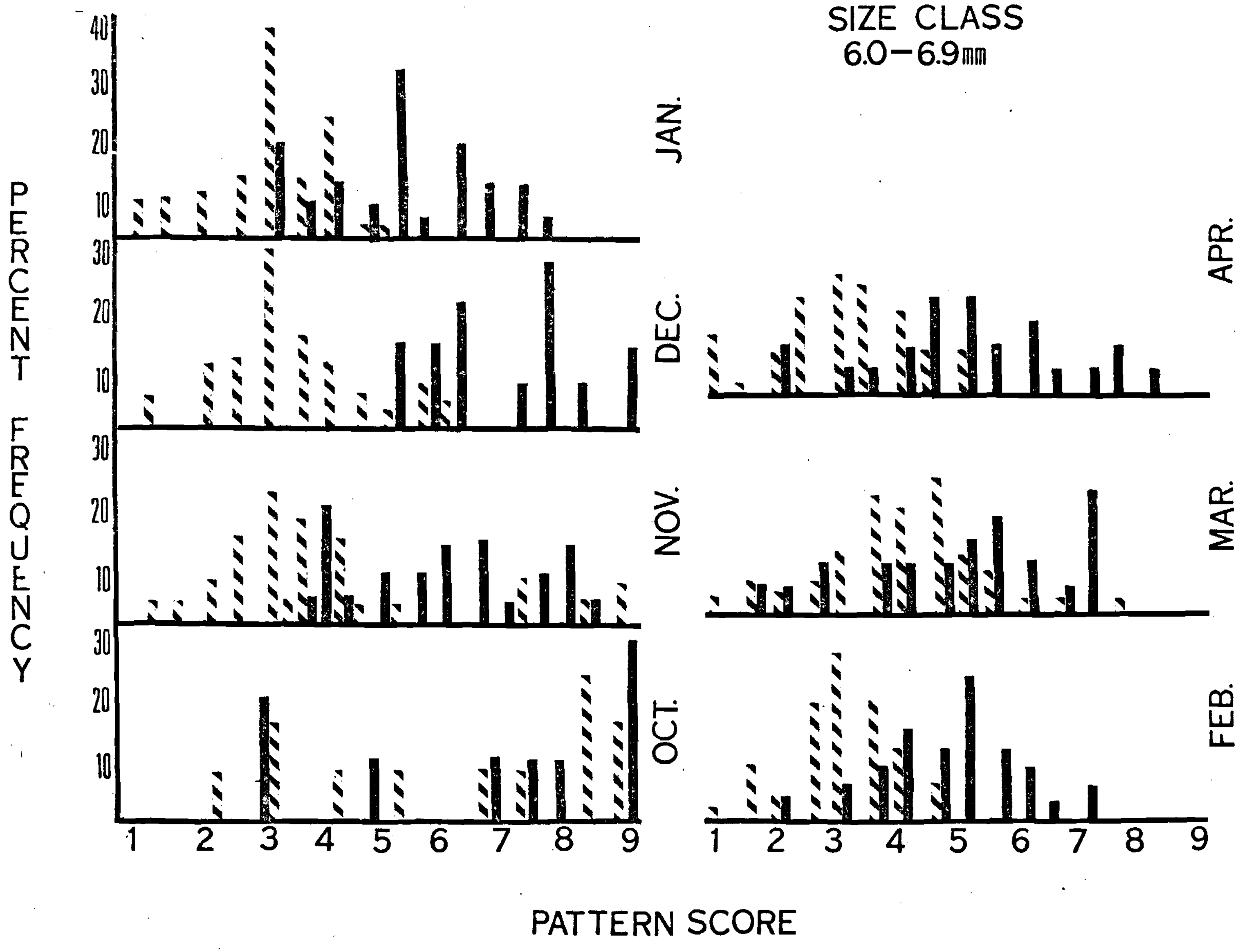
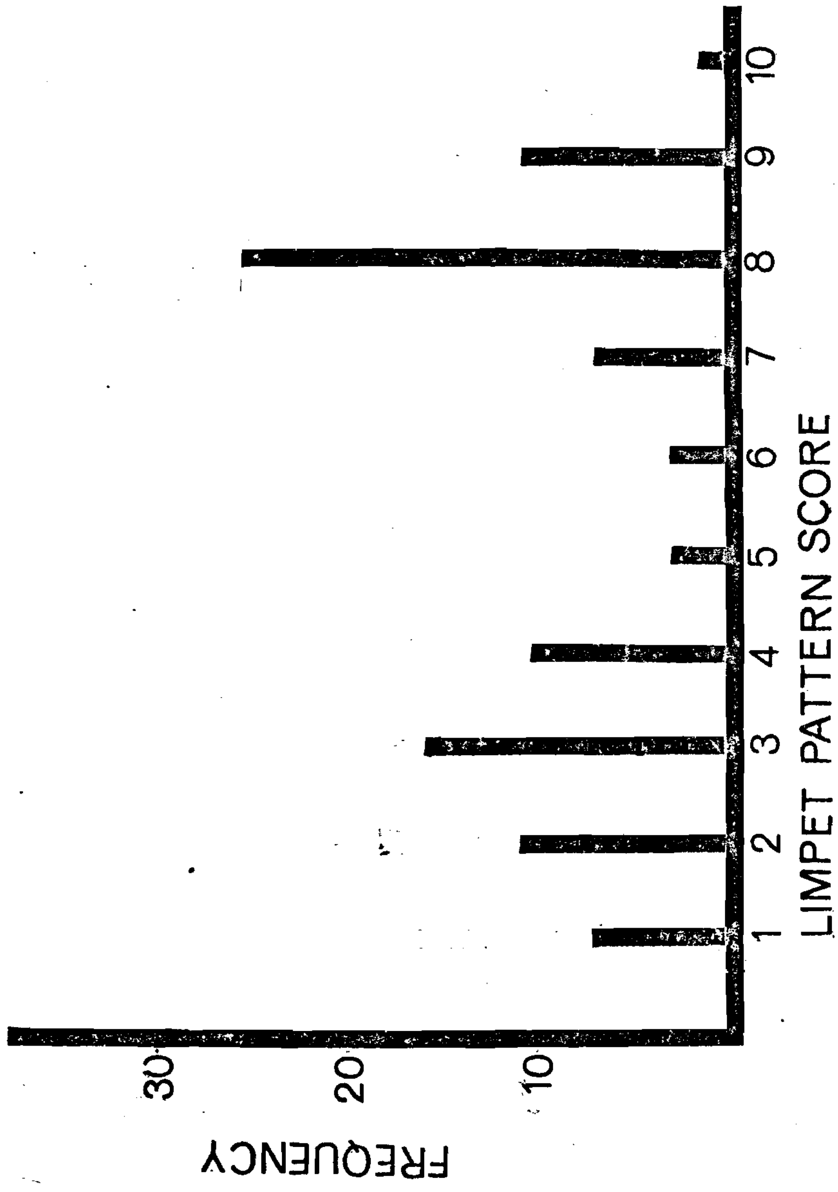

SIZE CLASS
6.0−6.9mm
PERCENT FREQUENCY
40
30
20
10
JAN.
DEC.
NOV.
OCT.
APR.
MAR.
FEB.
1
2
3
4
5
6
7
8
9
PATTERN SCORE

Figure 6C. Frequencies of pattern scores of all limpets larger than 10.0 mm. N = 279.


FREQUENCY
30
20
10
1
2
3
4
5
6
7
8
9
10
LIMPET PATTERN SCORE

each month from November through June. Each set contains histograms for animals of 3.0-3.9, 4.0-4.9, 5.0-5.9, and 6.0-6.9 mm.

In the case of very small animals, less than 4.0 mm, there is very little difference between the pattern frequencies of animals on rock and on Pollicipes. Gradually in time and with increase in animal size, however, a rock-type population and a Pollicipes-type population become established. Pollicipes-type limpets most frequently score from 1 to 4; rock-type animals tend to be darker. Animals of over 10 mm can be divided into two pattern classes: less than 4.5 and greater than 7 (Figure 6C).

As mentioned above, most movement of animals from rock to Pollicipes seems to occur at about 4.5 mm. The histograms show clearly that a bimodality with respect to habitat develops, without regard to time of year, in animals of 4.0 to 6.0 mm. The frequency distribution of pattern scores of limpets on Pollicipes is displaced relative to animals on rock. This becomes more pronounced with increased limpet size and, to a certain extent (February to April), with the passage of time. As the animals grow and as time passes, the proportion of all types of limpets on Pollicipes increases relative to that of animals on the rock face. (Time and size are obviously inextricably confounded by this method.) This latter comparison of course holds only in the immediate vicinity of a Pollicipes colony. In young animals of both types, less than 5.0 mm, the pattern frequency distribution is essentially unimodal and normal. Later, when only animals on and near Pollicipes are considered, the distribution becomes skewed to the right with a great decrease in darker animals, the majority of these being located on the rock face.

Since this type of histogram is useless as a quantitative means of estimating proportional changes in a population, a more quantitative means of measurement was attempted. A value I, similar in conception to Haldane's (1961) selection intensity, I = ln (proportion surviving of optimum) - ln (proportion surviving entire population), was derived and calculated. Haldane's I was derived for an essentially different situation. He dealt with a population of known absolute size, which enabled him to calculate proportions on the basis of N_{t+1}/N_t. My samples, and indeed, I suspect, those derived from most field work, are characterized by randomly fluctuating numbers of individuals. Thus this study demanded an I which could be calculated regardless of temporal differences in total sample size. Haldane's I gives selective advantage, mine, selective disadvantage. $I = \ln (1 - (prop_t/prop_{t-1}))$ where $prop_t$ is the proportion of the entire population of a given pattern class at time t, and $prop_{t-1}$ is a similar value for the same pattern class at time *t-1*. Thus the proportionate decrease over unit time of a given pattern class was calculated. The reciprocal was taken to make the value obtained directly proportional to the observed change. *I* increases with increase of apparent mortality. A logarithmic function normalized the values. Coefficients were calculated (using IBM 360-50 computer) for each of 7 size classes, limpets >5 mm, >6 mm, ... >11 mm, for nine months, November through June, and for 17 pattern scores, 0.5 increments from 1 through 9. Size classes are inclusive of all limpets larger than the indicated size; thus the smallest size class included all animals and the largest only limpets greater than 11 mm. Appendix C contains data used for calculation of *I*'s. The program is available on request.

In order to better define pattern-specific *I* values, a mean *I* value, $\bar{I}$, was calculated for each of the pattern classes, for all sizes of limpets at all periods.

$\bar{I}$ values for each pattern, 1-8, are listed in Table 10, column 1. Note that, for example, -0.01 = ln0.99 and -4.605 = ln 0.01. Numbers of animals of some high pattern scores were so low that the derived statistics are meaningless. Column 3 differs from column 2 in that column 3 considers only the period February-March through April-May and sizes >6 to >10 mm. The latter treatment was suggested by the results of Figures 6A and B which showed relatively great

Table 10. Mean I values of pattern scores. Column 1 calculated using data from all months and size classes. Column 2 calculated using only data from the months of February through May, and size classes >6 mm. (See Appendix C for N.)

	Mean I	
Pattern score	All months and sizes	Feb.-May, >6 mm
1	-0.83	-1.42
1.5	-0.18	-0.77
2	-0.52	-0.69
2.5	-0.64	-1.15
3	-0.69	-1.33
3.5	-0.76	-0.91
4	-1.33	-1.05
4.5	-0.29	-0.12
5	-0.42	-0.21
5.5	-0.10	-0.33
6	-0.79	-0.46
6.5	-0.40	-1.77
7	-0.59	-0.73
7.5	-0.40	-0.53
8	-0.12	--

change in distribution of patterns of the population in February through April and in medium sized limpets.

It is obvious that the $\bar{I}$ values of Table 10 result from more than one source of variability; sampling error and selection are the two most obvious components of the variability. The selection may be either nonpattern-specific, i.e. physiological and that caused by physical factors, or based on the survival value of particular pattern scores. Sampling error can probably be assumed to be evenly distributed over the samples and the various pattern scores. Although pattern may be pleiotropically involved with other parameters open to selection, this is not likely to be a cause of error. Where studied previously, mortality in A. digitalis was about evenly distributed over the course of a year (Frank 1965); unusually high mortality can probably be directly related to particularly rigorous climatic conditions.

In column 3 the demarcation between $\bar{I}$ values for pattern classes 4.5 - 6 and the other pattern scores is very clear; all other pattern classes except 7 and 7.5 show lower mortality or intensity of selection (i.e. greater, though negative, values of $\bar{I}$). Differences are not so clear in column 2 where, although pattern classes 4.5 to 6 have relatively low I values, the general picture is one of less difference in $\bar{I}$ among the patterns. This might be interpreted, considering that column 2 is of $\bar{I}$ for a selected sample of months, that in the time intervals November to March and June-July, and in limpets <6 mm and >11 mm, $\bar{I}$ is varying randomly with respect to pattern scores. However, during March through June in limpets >6 mm to >10 mm the intermediate patterns 4.5 to 5.5 are more subject to selection. Columns 2 and 3 of Table 10 thus may be thought of as effecting some separation of random selection from that related to limpet pattern.

There was little relationship between pattern and selection intensity in very small animals. However, dark animals in close proximity to Pollicipes colonies decreased in frequency with size; their density is about $108/m^2$. In areas of low Pollicipes density, large rock-type animals were abundant ($600/m^2$) and dark (pattern score 5 to 9, Figure 6C).

Table 11. $\bar{I}$ values and numbers of limpets of all sizes found on six colonies of Pollicipes from December 1967 to July 1968.

	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July
N	363	378	445	369	208	275	308	289
$\bar{I}$	-.41	-.27	-.58	-.67	-.82	-.72	-.61	-.53

Table 11 of $\bar{I}$ values and numbers of animals found on Pollicipes at each of 8 census periods shows rather drastic changes in population sizes. Since sampling error was probably great, only the larger changes will be considered. $\bar{I}$ values in this table are the monthly means for all sizes and pattern classes. Large intensities in months December, January, June, and July must be unrelated to pattern specific changes as in Table 10.

The January-February increase in numbers coincides with a major period of *Pollicipes*-directed movement of young animals. The large overall decrease from February through April seems to correspond with high pattern-specific I values and changes in pattern frequency distribution noted during these months. The April through June increase may be partially explained by movement of young limpets.

MISCELLANEOUS OBSERVATIONS

An observational trip was made to Pacific Grove, Bodega Bay, Abalone Point, California, and various rocky intertidal areas north of Abalone Point. The California coast at Pacific Grove and Bodega Bay is characterized by a granite rather than sandstone substrate. In general A. digitalis behavior and distribution are probably no different from those at Cape Arago. One might have expected differences due to the white and black peppered substrate.

At Mussel Point and Point Pinos (Pacific Grove) the white forms of Acmaea digitalis were common on Mytilus as well as Pollicipes. The rocky intertidal in this region seems to be more compressed than further north; the Mytilus-Pollicipes association is more intense. Tides cover less range here than they do further north and probably, together with more direct insolation, are the prime cause of the compression. The shorter times of exposure and the habitat heterogeneity resulting from the apparent compression may result in low intensity of bird predation.

At Bodega, light limpets were not found on Mytilus as they were at Pacific Grove. Limpet behavior seemed at all intertidal levels to be similar to the behavior of low level limpets at Cape Arago.

Distribution of pattern frequencies of limpets appears to differ systematically along the coast; limpets are in general lighter in areas of low Pollicipes density.

Settlement

The larvae of numerous intertidal animals do not settle upon their substrate at random but rather rely on both chemical and tactile stimuli to locate a favorable area for settlement. Barnacle larvae for instance seem to be attracted by a periostracal protein present in the adult (Crisp and Meadows 1962, 1963). Molluscan forms are reported to behave similarly (Crisp 1967, Cole and Knight-Jones 1949).

It is possible that the white forms of A. digitalis are stimulated to settle by the presence of Pollicipes. Young of these forms are found only proximal to Pollicipes. The white limpets may react differentially depending on concentration of Pollicipes. In areas of low barnacle concentration the lighter forms of the limpets are found in greatest frequency, whereas in areas of Pollicipes commonness, all scores (0-4) are in approximately equal frequency. This is explicable if one considers the differential behavior of young limpets to Pollicipes (Table 7). If presumptive pattern score and behavior were also correlated in post-larval settling stages, it might be suggested that these respond to different concentrations of an exudate of the barnacle. Presumptively light limpets would react to lower concentrations of the product than would limpets destined to be darker, and would thus be expected to settle in high frequency in areas of low *Pollicipes* concentration. Settling response to *Pollicipes* concentration is thus suggested to be a function of pattern.

DISCUSSION

It is probable that the light and dark forms of *Acmaea* either belong to the same species or are morphs of a highly introgressed hybrid. Interbreeding of these forms is probably possible; their gonad cycles, which were followed at two week intervals throughout the year, are similar enough to allow interbreeding. The karyotypes of the two forms are similar. However, all Patellaceae which have been studied have 9 chromosome pairs (Patterson 1967). The unimodal frequency distribution of patterns in the young limpets suggests that the two forms are extremes of a distribution of patterns and forms.

All of the dark and light colored limpets probably settle from the plankton (Thorson 1950) to the rock substrate. The lighter colored forms then, in order of their lightness, (Table 7) move to *Pollicipes*. This movement occurs in lightest limpets at about 3.5 mm length and is completed by darker limpets of pattern score 4 to 4.5 by the time they attain a length of 5 to 6 mm. The relationship between behavior and pattern, presented in Tables 1 to 6 and discussed later, extends to young limpets, mediating the time of their movement from the rock to

Pollicipes.

Pollicipes-type limpets are behaviorally confined to *Pollicipes* colonies; reciprocal transplants showed that limpets from the rock prefer the rock substrate. The relationship between limpet pattern score and intensity of substrate preference seems rather complex (Tables 1-6); a model explaining differences in correlation of pattern score and substrate preference was proposed. The data fit the model well.

Reiterating, in small (<7 mm) *Pollicipes*-type limpets there is a positive linear relationship of pattern score to affinity for *Pollicipes*, darker limpets taking longer to return to *Pollicipes* than do light animals. In larger animals this relationship is maintained in sheltered areas. However, the times taken to return to *Pollicipes* of dark and light *Pollicipes*-type limpets (originally found on *Pollicipes*) are indistinguishable in exposed areas, almost all limpets returning on the first day. Those limpets having less affinity for *Pollicipes* have a greater probability of remaining on a selectively unfavorable substrate at low tide. These will be, in a probabilistic sense, conspicuous for longer periods of time than will their counterparts which have high affinities for substrates rendering them inconspicuous. They will thus be visually selected at greater intensity.

Selection will be intense in high areas where the limpets are exposed for long periods of time and where incidence of barnacles, macroalgae, and other factors causing background heterogeneity is low. In low areas with correspondingly short times of exposure, or in areas of great background heterogeneity, which seems to render a prey less conspicuous to a predator, selection intensities will be low.

Since the visible aspects of the polymorphism are the color and patterns of limpets in the population, it is necessary to determine whether the origin of the existing patterns is primarily genetic or primarily environmental.

The genesis of variable shell color and pattern in gastropods has been the subject of a rather large amount of study and of some controversy. It is generally considered (Hyman 1967) that shell pigments in archeogastropods are porphyrins, metabolic products of food pigments or of odontophore and shell-muscle hemoglobin. Thus shades of brown and black pigments, among others, characterize most archeogastropods.

Ino (1940) found that food quality has a controlling influence on the shell color of Turbo cornutis, and Leighton (1961) reports that shell color of the red abalone, Haliotis rufescens, is greatly affected by the species of algae on which the animal has been recently feeding. Moore (1963) studying Thais lapillus found that shell color was correlated with the shell pigments of the prey. Thus *Mytilus*-eating *Thais* have a dark brown to black shell, but those feeding on *Balanus* have lightly-colored shells.

Test (1937) discounts the effect of food on shell color in the *Acmaeidae* stating that infection of shells by the ascomycete *Didymella conchae* Bonar (1936) is of primary importance in determining the color and pattern of the infected animal. Among the commonly infected animals are the limpets *A. digitalis* and *A. pelta*. The fungus penetrates the shell destroying the conchin matrix and rendering the shell honeycombed and thick owing to compensatory regrowth of shell material by the animal. Test further states that, upon removal of the fungus infection, the shells no longer bear a resemblance to their substrate.

She feels that the fungus infection is in complete control of the pattern and cryptic abilities of infected limpets. She has indeed suggested an obligate (for the infected limpet) symbiosis, the relationship having been strongly selected. Thus only those animals which are infected and which therefore bear a remarkable resemblance to their substrate survive. *Didymella conchae* infection certainly is modificatory, tending to reduce shell color intensity and quality and having a muting and dulling effect on shell tone. Since *Pollicipes* scutes and the rock are both dull and rough-surfaced, the dullness and roughness of the limpet shells may be of selective advantage.

Periostracal material, which is pigmented (Hyman 1967), is largely destroyed by the fungus. The partial destruction and consequent muting and diffusion of stripes may be the partial cause of diffuse brown and grey background colors. Original periostracal and prismatic layer material is either brown or white. Since striping is continuous throughout both prismatic and periostracal layers and is little affected by grinding, which removes the fungus, Test's (1945) statement that the fungus is responsible for the pattern of limpet shells cannot be correct.

The phenotypes of the unparasitized limpet shells are of course functions of the interaction of genetic and environmental variance (Fisher 1930). Although better definition of the relative values of these components of phenotypic variance was attempted in the experiment in which <u>Pollicipes</u>-type limpets were transferred to a rock having no <u>Pollicipes</u>, information obtained was only qualitative. The importance of the environmental component increased with increasing pattern score (to 4), i.e. light animals were little affected by the change in substrate, retaining, essentially, their original striping and color. Dark limpets (scores 3-4) exhibited a range of responses but in general became still darker when confined to the rock substrate. Allometry was a similar function of substrate, new growth of darker limpets being more flattened than the original shell. Dark _Pollicipes_-type _Acmaea_ _digitalis_ seem to be capable of becoming similar in appearance to rock-type animals.

After movement to _Pollicipes_ the color and shape of young limpets probably become respectively lighter and higher. The presence of a full range of light to dark young (3-3.5 mm) limpets on the rock face strongly suggests that environmental effects on color and pattern are of little qualitative importance. Populations of young limpets, after choosing one of the two available substrates, progressively diverged in frequency distribution of patterns. Animals of intermediate pattern, light pattern on-rock, and dark pattern on-_Pollicipes_ gradually disappeared leaving two essentially normal pattern frequency distributions of large rock-type and large _Pollicipes_-type limpets of mean pattern scores about 2.5 and 6.5 respectively.

This divergence could possibly be explained partially by _Pollicipes_-types becoming lighter and rock-types becoming darker during development. However, very dark limpets disappear from _Pollicipes_ as do very light limpets from rock. Further, the divergence of pattern frequency distribution is decidedly seasonal, greatest pattern specific changes occurring in February-May (Figure 6A and B, Table 10) During these months limpets of intermediate pattern more frequently have high rates of disappearance than do limpets of lighter or darker patterns (Table 10). Changes in these patterns are relatively greater in size classes 3 to 6 than in size classes 1, 2, and 7. Thus from February to May limpets of intermediate sizes of intermediate pattern scores disappear at a relatively high rate. At other times of year animals of other pattern scores suffer decreases in frequency. In the vicinity of _Pollicipes_ colonies dark limpets are relatively common when small, but they greatly decrease in frequency with increased size.

The behavior, pattern, and shape polymorphism described above is suggestive of at least two selection mechanisms. Clarification of the genetic systems controlling these characters is necessary before consideration of further control of the polymorphism.

It is possible to hypothesize both polygenic and multiple allelic control of limpet affinity for _Pollicipes_. Both possibilities could result in a normal distribution of variants of the trait. There is probably little environmental modification of behavior, especially in young animals. Since the behavior is demonstrably not quantal, control by a single diallelic locus is not likely. However, since Table 6 shows a discontinuity in response of young animals at about pattern score 4.5, a "switch gene" (Ford

1964) with modifiers may be implicated.

Pattern and shape probably have large environmental components of variance. Pattern with its apparently normal non-quantal distribution of expression is likely under polygenic control.

Regardless of the genetics of the individual characters, the good correlations between pairs of two of the three variables (Table 6, Figures 3, 4, and 5) indicate a rather low rate of recombination among the variables (pattern, shape, behavior). As R. A. Fisher (1930) pointed out, "Two factors, the alternative genes in which may be represented by Aa and Bb will maintain each other mutually in genetic equilibrium if the selective advantage of A over a is reversible when B is substituted for b or _vice versa_."

Bodmer and Parsons (1962) feel, ". . . if . . . (it) were such that A was advantageous in the presence of B . . . then natural selection would favor closer linkage between the two factors A and B."

Fisher and Diver (1934) found that the genes for pink and unbanded shell in _Cepaea nemoralis_ are linked. This linkage in _Cepaea nemoralis_ and more especially in _C. hortensis_ must be the result of strong selection (Ford 1955).

That linkage can be selected for has been amply demonstrated by Parsons (1958), Bodmer and Parsons (1962), Nei (1967), Mukherjie (1961, and Detlefsen and Roberts (1929).

The selective advantage of linkage for behavior and pattern in *A. digitalis* is obvious if one considers that any recombination between these factors would result in light colored limpets behaviorally relegated to the dark rock substrate and *vice versa*. Such limpets would not be cryptically colored and would therefore be subject to predation.

Mechanically tight linkage can be established in at least two major ways. First, as Ford (1964) observed, by means of small overlapping inversions groups of genes may be established, the members of which are close enough together that crossing over between them is precluded.

Second, genes have been found which act to modify recombination rate. Levine and Levine (1964) found that different combinations of the chromosome III inversions in *Drosophila pseudoobscura* differentially affect rates of crossing over in the X chromosome. In addition, they detected gene complexes within the inversions which modify cross over rates.

A model of the evolution of a system using a modifier locus to change apparent recombination rate has been proposed by Masatoshi and Imaizumi (1968). They show that alleles of a recombination modifier for a particular set of loci are selected simultaneously as the alleles of the primary loci are selected. Thus selection of recombinants results in increased frequency of the modifier allele favoring recombination.

That linkage in general is being selected for in the limpet populations is strongly suggested by the divergence in variance values of the Sunset Bay populations from those of the Cape Arago populations. Members of the Sunset Bay populations were much less canalized as to substrate preference. Variance of means of pattern scores of returnees and nonreturnees were larger than at Cape Arago. Large variance could be expected if pattern and behavior were not tightly linked. The Cape Arago population, thought to be subject to high selection intensity, exhibits apparently tight linkage. Selection might be less at Sunset Bay simply because the limpets are far smaller (mean length 7.5 mm) than at Cape Arago (mean length 10.5 mm). The small animals might be largely ignored by an efficient predator (Slobodkin 1960).

The first explanation of maintenance of the polymorphism postulates two reproductively isolated populations, Pollicipes-type and rock-type. Each population is normally distributed, but distributions overlap in young limpets by an amount such that both curves together simulate a single normal curve. Selection against the limpets of patterns falling within the area of overlap would result in the complex dimorphism found in adults. Generation of an apparently monomorphic population of young limpets from the two adult populations would be dependent upon recombination solely within each parent complex. However, directional selection relative to each population would tend, in the long run, to move the mean of the distribution of each population progressively away from the monomorphic mean, resulting in divergence of the phenotype distributions of the two populations. Heterozygote superiority might, in the multilocus case, be expected to slow the resultant population divergence. However, divergence would obtain eventually. This model is, of course, dependent on reproductive isolation of the two parent populations. Since it seems likely that the populations do interbreed, since radulae are indistinguishable, and since Fritchman (personal communication to P. Frank), the specialist in this group, considers them one species, it is doubtful that the model applies.

The second possible model proposes an interbreeding population. Circumstantial evidence from gonad cycles indicates a high degree of panmixis to be likely in this case. A roughly normal frequency distribution of each variable, pattern, shape, and behavior, would be generated by interbreeding after which disruptive selection might be expected to result in the observed two polymorphic complexes (Figure 6). Such a system would be self-perpetuating so long as a high degree of interbreeding obtained. Population divergence through disruptive selection (Mather 1953) can occur in the face of 25 to 50% interbreeding (Thoday and Boam 1959, Millicent and Thoday 1960).

Interbreeding is sometimes decreased as a side effect of disruptive selection (Thoday and Boam 1959); various forms of reproductive isolation develop concomitantly. The spawning cycles of Pollicipes-type and rock-type A. digitalis may be slightly divergent (Giesel, unpublished), the divergence probably being a result of disparate levels of important physical factors (insolation, desiccation) associated with the two substrates.

Available information on gonad cycles, karyotypes, the unimodal form of the distribution of patterns in young animals and the form of distribution of selection intensity on pattern (Table 10) suggests that disruptive selection, with inbreeding, is the most likely means by which the frequency distribution of the characters mentioned is maintained. The first, two population, model cannot be ruled out, however, since the time differential in spawning of the two forms could result in positive assortative mating.

Apparently seasonal differences in frequency of return of limpets to Pollicipes were noted in a series of behavior experiments. Return frequency was highest in the May, July, and August experiments and low in January and March. These differences may be explained by considering three possible emphases of natural selection: predation, desiccation, and productivity maximization.

Certain shorebirds such as oystercatchers, surfbirds, (Bent 1927, Dawson 1923), and perhaps gulls (Frank 1965) prey on the Acmaea and other inhabitants of Pollicipes-Mytilus beds. These birds are probably the main pattern and color selective predators of the limpets. Lewis (1954) reports that oystercatchers preyed on his marked Patella. Surfbirds seem to feed both while hovering to the side of vertically disposed Pollicipes colonies and while alighting on the colonies. Oystercatchers walk over the surface of the rocks, picking limpets both from the rocks and from the barnacles. Live painted limpets disappear rather quickly. Their empty shells are found overturned on and in the vicinity of the barnacle colonies.

Migrations of oystercatchers pass the Cape Arago area in March through April and again in late July and August (Bent 1927, Dawson 1923). Small populations are present along the Oregon coast through the winter months and occasional birds have been observed in the summer. Peak populations of the two species of birds were observed in the spring and fall. Greatest pattern specificity of selection at Cape Arago was in February through May in 1968 and in April through May in 1967 (Figures 6A and B). This coincidence of high density of oystercatchers and surfbirds and the time of greatest disruptive selection (Table 10), supports the feeling that these birds are the primary visual predators of the limpets.

The limpets of the Sunset Bay area, where Pollicipes colonies are subjectively neither as common nor as extensive as at Cape Arago, are less canalized behaviorally to Pollicipes than are those at Cape Arago. Lightly colored limpets are in some cases common on the rock at Sunset Bay. At Cape Arago large dark limpets are relatively uncommon on the rock in the vicinity of Pollicipes beds, whereas they are extremely common distant from Pollicipes. Colony 9 (Table 6), somewhat isolated and of low Pollicipes density, exhibits pattern-behavior relationships characteristic of low, sheltered colonies. These differences are explained if one concludes that Pollicipes beds serve as a focus for the feeding activities of limpet-eating birds. A successful predator must be able to outdisperse its prey (Andrewartha 1961), or similarly, to locate its prey easily. It can be assumed that an efficient predator will choose feeding areas of high apparent prey density. Emlen (1968) states, ". . . it may be presumed that experience with any given food increases the efficiency with which the predator forages for it. . . . the feedback relation between a food use and its value leads toward increased specialization." It may be that high Pollicipes density is a strongly reinforced feeding stimulus to the predaceous birds. The low Pollicipes density at Sunset Bay may make the area not particularly attractive to avian predators.

Bird predation may also explain the observation that large dark Pollicipes-type limpets can generally only be found on the undersides of or buried deep within Pollicipes colonies. This phenomenon was particularly noticeable at the April 1968 sampling when the population size of one colony was found to be greatly decreased. Extant animals were all either light and located on the surface of the colony or dark and located on the lower shell plates or on the black necks of the Pollicipes. The lower shell plates are small, polygonal, and sharply demarcated by a network of heavy dark lines.. They thus present a background very similar to the shell pattern of a heavily striped Pollicipes-type A. digitalis. The degree of such within-colony subdivision of the limpet habitat is a function of the location of the colony. Low colonies located in heterogeneous backgrounds subjectively exhibit this definition to a lesser degree than do higher, more exposed colonies.

Desiccation is an important cause of mortality in

Acmaea digitalis (Frank 1965). Analyses of the yearly pattern of times of low tides show that these follow a cycle the result of which, when combined with prevalent weather, is desiccation of high intertidal animals is likely highest in May to July. In May to July, generally bright warm weather combines with low midmorning to early afternoon tides. The result is long exposure to high insolation and consequently great desiccation. The low tides of August to November occur in the very early morning and late evening. Those of December to March, although occurring during daylight hours, are combined with weather conditions which preclude much effective desiccation. It has been suggested that Pollicipes provides a relatively desiccation-free home to Pollicipes-type limpets. The limpets have a smooth, unscalloped shell and are probably able to form a relatively watertight seal between the Pollicipes plates and their shells.

The productivity of micro-algae indigenous to the rock substrate (blue greens, diatoms) is greatest during the early spring when desiccation is low and effective insolation is high (Castenholz, personal communication), and may also be related to seasonal fluctuations in behavior.

If genetic fitness is a function of these three largely temporally separated factors and if evolution may be assumed to be toward a maximization of the fitness of the morph, the limpets must follow a strategy of maximization of feedin (productivity) and minimization of mortality from desiccation and predation. Theoretically the interaction of two or more different modes of selection, i.e. disruptive, apostatic, directional, results in the establishment of a point or points of equilibrium of frequencies of the genotypes (or phenotypes) concerned (James 1962). In this case the intensities of the three selective forces proposed probably differ with time of year and with size of the limpets resulting in interaction that is at least partially discrete. The forces are obviously antagonistic in their influence on behavior of the limpets and would be expected to result in a behavior pattern and variability of behavior optimal for the population.

Relative time spent on the rock face, where the limpets are thought to feed, is probably reflected by the return frequencies shown in Tables 1 to 6 of results of transplant experiments. If so, the limpets feed most in early spring when algal productivity is greatest and when gonad generation and spawning are occurring. Thus the limpets may be making optimal use of algal production. In late spring and summer, however, when algal productivity is low and when desiccation is maximally effective, selective premium appears to be on remaining in the moistest areas available, e.g. <u>Pollicipes</u> colonies. In August, with decreased effective desiccation, the fall gonad build-up, and high fall growth rates (Frank 1965), selective emphasis may again become attached to productivity. The selective effects of bird predation must overlie and interact with those of the other two factors.

There is probably a counterforce militating against total assumption of highly Pollicipes-oriented behavior. This force is maximization of food intake and eventually of fitness through increased growth rates and gamete production. Food intake of a Pollicipes-type limpet is probably a direct function of the time that animal is able to spend on the rock rather than on Pollicipes. At low levels, where selection is probably less intense, there may thus be a reservoir of limpets which may be able to maximize use of producer level productivity.

Although free interbreeding of high and low intertidal level populations cannot occur at certain times of year, there is certainly some opportunity for interbreeding during the balance of the year. This interbreeding of low intertidal limpets with limpets from more heavily selected levels would serve to balance the effects of selection by visual predators, much as migration rate is able to balance selection (Wright 1931).

General conclusions emerge. This particular polymorphism in Acmaea digitalis seems at least partially the result of bird predation which separates an originally unimodal polymorphic population of limpets into two pattern and shape complexes. Apparently different intensities of predation result in different degrees of within-population differentiation. Pattern-correlated behavior seems to be at the root of the visible polymorphism. Thus rock- and *Pollicipes*-type animals are separated originally and maintained separate by behavioral differences in light and dark limpets. Selection can thus act in a disruptive rather than a directional manner.

BIBLIOGRAPHY

Anderson, N. H. 1961. Scales and statistics, parametric and nonparametric. Psychol. Bull. 58: 305-316.

Andrewartha, H. G. 1961. Introduction to the study of animal populations. The Univ. of Chicago Press. 281 p.

Barnes, H. 1960. The behavior of the stalked intertidal barnacle Pollicipes polymerus J. B. Sowerby, with special reference to its ecology and distribution. J. Animal Ecol. 29: 169-185.

Bent, A. C. 1927. Life histories of North American shore birds. U. S. Natl. Mus. Bull 146: Parts 1 and 2.

Bonar, L. 1936. An unusual ascomycete in the shells of marine animals. Univ. Calif. Pub. Botany 19: 187-192.

Bodmer, W. F. and L. A. Parsons. 1962. Linkage and recombination in evolution. Advances in Genetics 11: 1-87.

Cain, A. J. and J. D. Currey. 1963. Area effects in Cepaea. Phil. Trans. Roy. Soc. Lond. 246: 1-81.

Cain, A. J. and P. M. Sheppard. 1950. Selection in the polymorphic land snail, Cepaea nemoralis. Heredity 4: 275-294.

_____ 1952. The effects of natural selection on body color in the land snail, Cepaea nemoralis. Heredity 6: 217-231.

_____ 1961. Visual and physiological selection in Cepaea. Am. Nat. 95: 61-64.

Carter, M. A. 1967. Selection in mixed colonies of Cepaea nemoralis and Cepaea hortensis. Heredity 22: 1

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

Colton, H. S. 1916. On some varieties of Thais lapillus in the Mount Desert region. A study of individual ecology. Proc. Acad. Nat. Sci. Philadelphia 68: 440-454.

Cook, L. M. 1965. Inheritance of shell size in the snail *Arianta arbustorum*. Evol. 19: 86-96.

_____ 1967. The genetics of *Cepaea nemoralis*. Heredity 22: 397-410.

Crisp, D. J. 1968. Chemical factors in the settling of *Crasostrea virginiaca*. J. Anim. Ecol. 36: 329-337.

Crisp, D. J. and P. S. Meadows. 1962. The chemical basis of gregariousness in cirripeds. Proc. Roy. Soc. B 156: 500-520.

_____ 1963. Adsorbed layers: The stimulus to settlement in barnacles. Proc. Roy. Soc. B, 158: 364-387.

Dawson, W. L. 1923. The birds of California, vol. 2. South Moulton Co.

Detlefsen, J. A. and E. Roberts. 1921. Studies on crossing over. I. The effects of selection on crossover values. J. Exp. Zool. 32: 333-354.

De Ruiter, L. 1952. Some experiments on the camoflage of stick caterpillars. Behavior 4: 222-232.

Diver, C. 1940. The problem of closely related species living in the same area. *In* The new systematics, ed. J. S. Huxley. Clarendon Press. 583 p.

Dixon, W. J. and F. J. Massey. 1951. Introduction to statistical analysis. McGraw-Hill. 370 p.

Emlen, J. M. 1968. Optimal choice in animals. Am. Nat. 102: 385-389.

Fisher, R. A. 1930. The genetical theory of natural selection. Clarendon Press.

Fisher, R. A. and C. Diver. 1934. Crossing over in the land snail *Cepaea nemoralis* L. Nature Lond. 133: 834-835.

Ford, E. B. 1964. Ecological genetics. John Wiley and Sons Inc.

Fox, D. L. 1964. Pigmentation of molluscs. *In* Physiology of mollusca, vol. 2, ed. K. M. Wilber and C. M. Yonge. Academic Press.

Frank, P. W. 1964. On home range of limpets. Am. Nat. 98: 99-104.

_____ 1965. Growth of three species of Acmaea. The Veliger 7: 201-202.

_____ 1965. The biodemography of an intertidal snail population. Ecol. 46: 831-844.

Fretter, V. and A. Graham. 1962. British prosobranch molluscs. Royal Society, London.

Galbraith, R. T. 1965. Homing behavior in the limpets Acmaea digitalis and L. gigantia. Am. Midl. Nat. 74: 245-246.

Hewatt, W. G. 1940. Observations on the homing limpet Acmaea scabra Gould. Am. Midl. Nat. 24: 205-208.

Humphries, L. 1964. For research trainees. Comtemp. Psychol. 9: 76-80.

Huxley, J. S. 1932. Problems of relative growth. The Dial Press.

Hyman, L. 1967. The invertebrates IV: Mollusca I. McGraw Hill.

Ino, T. 1940. The effect of food on growth and coloration of the top shell, Turbo cornutus Solander. J. Mar. Research 8: 1-5.

James, J. W. 1962. Conflict between directional and centripetal selection. Heredity 17: 487-499.

Kettlewell, H. B. D. 1955. Recognition of the appropriate background colors by the pale and black phase of Lepidoptera. Nature Lond. 175: 943-944.

Kinsler, C. B. 1967. Desiccation resistance of intertidal crevice species as a factor in their zonation. J. Anim. Ecol. 36: 391-407.

Lamotte, M. 1959. Polymorphism of natural populations of Cepaea nemoralis. Cold Spring Harbor Symposium Quant. Biol. 24: 65-86.

Leighton, D. L. 1961. Observations of the effect of diet on shell coloration in the red abalone, Haliotis rufescens Swainson. The Veliger 4: 29-32.

Levene, H., O. Pavlovsky, and T. Dobzhansky. 1954. Interaction of the adaptive values of polymorphic experimental populations of Drosophila pseudoobscura. Evol. 8: 335-349.

Levine, R. P. and J. I. Dickinson. 1952. The modification of recombination by naturally occurring inversions in Drosophila pseudoobscura. Genetics 37: 599-600.

Levine, R. P. and E. E. Levine. 1954. The genotypic control of crossing over in Drosophila pseudoobscura. Genetics 39: 677-691.

Lewis, J. R. 1954. Observations on a high level population of limpets. J. Anim. Ecol. 23: 85-100.

Mather, K. 1953. The genetical structure of populations. Symp. Soc. Exp. Biol. 7: 66-95.

_____ 1955. Polymorphism as an outcome of disruptive selection. Evol. 9: 52-61.

Millard, C. S. 1968. The clustering behavior of Acmaea digitalis. The Veliger 11: suppl. 45-52.

Millicent, E. and J. M. Thoday. 1960. Gene flow. I. Divergence under disruptive selection. Sci. 131: 1311-1312.

Moore, H. B. 1936. The biology of Purpura lapillus and shell variation in relation to environment. J. Mar. Biol. Ass. U.K. 21: 69-81.

Morton, J. E. 1958. Molluscs. Hutchinson University Library.

Mukherjie, A. S. 1961. Effect of selection on crossing over in the males of Drosophila ananassae. Am. Nat. 95: 57-59.

Murray, J. and B. Clarke. 1968. Inheritance of shell size in Partula. Heredity 23: 189-199.

Nei, M. 1967. Modification of linkage intensity by natural selection. Genetics 57: 625-641.

Nei, M. and Y. Imaizumi. 1968. Efficiency of selection for increased or decreased recombination. Am. Nat. 102: 90-94.

Newell, G. E. 1964. Physiological aspects of the ecology of intertidal molluscs. In Physiology of Mollusca, vol. 2, ed. K. M. Wilber and C. M. Yonge. Academic Press.

Oldham, C. 1928. The influence of lime on the shell of Arianta arbustorum L. Proc. Malac. Soc. Lond. 18: 143-144.

Owen, D. F. 1965. Density effects in polymorphic land Snails. Heredity 20: 313-315.

Palmer, J. B. 1968. Ph.D. dissertation, University of Oregon.

Parsons, P. A. 1957. An effect of gene rearrangement on the recombination fraction in Drosophila melanogaster. Heredity 11: 117-127.

_____ 1958. Selection for increased recombination in Drosophila melanogaster. Am. Nat. 92: 255-256.

Patterson, C. M. 1967. Chromosome numbers and systematics in Streptoneuran snails. Malacologia 5: 111-127.

Price-Jones, C. 1930. The relations of diameter and height to the squatness of shells (Cepaea nemoralis). Proc. Malac. Soc. Lond. 19: 146-152.

Rickets, E. F. and J. Calvin. 1962. Between Pacific Tides, rev. J. W. Hedgpeth. Stanford Univ. Press.

Russell, E. S. 1907. Environmental studies in the limpet. Proc. Zool. Soc. Lond. 1907.

Sargent, T. D. 1966. Background selection of geometrid and noctuid moths. Sci. 154: 1674-1675.

Savage, I. R. 1957. Nonparametric statistics. J. Am. Statist. Ass. 52: 331-344.

Sheppard, P. M. 1951. Fluctuations in the selective value of certain phenotypes in the polymorphic land snail, Cepaea nemoralis. Heredity 5: 125-134.

Slobodkin, L. B. 1960. Ecological energy relationships at the population level. Am. Nat. 94: 213-236.

Steinburg, A. G. 1936. The effect of autosomal inversions on crossing over in the X chromosome of <u>Drosophila</u> <u>melanogaster</u>. Genetics 21: 615-624.

Stern, C. 1936. Somatic crossing over and segregation in <u>Drosophila</u> <u>melanogaster</u>. Genetics 21: 625-730.

Stevens, S. S. 1968. Measurement, statistics, and the schemapiric view. Sci. 161: 849-856.

Tate, M. W. and R. C. Clelland. 1957. Nonparametric and shortcut statistics. Interstate Publishers and Printers, Inc.

Test, A. R. 1945. Ecology of California <u>Acmaea.</u> Ecol. 26: 379-405.

Thoday, J. M. 1958. Effects of disruptive selection: The experimental production of a polymorphic population. Nature 181: 1124-1125.

Thoday, J. M. and J. B. Boam. 1959. Effects of disruptive selection. II. Polymorphism and divergence without isolation. Heredity 13: 205-218.

Thoday, J. M. and J. B. Gibson. 1962. Isolation by disruptive selection. Nature Lond. 193: 1164-1166.

Thorson, G. 1950. Reproduction and larval ecology of marine bottom invertebrates. Biol. Revs. Cambridge Phil. Soc. 25: 1-45.

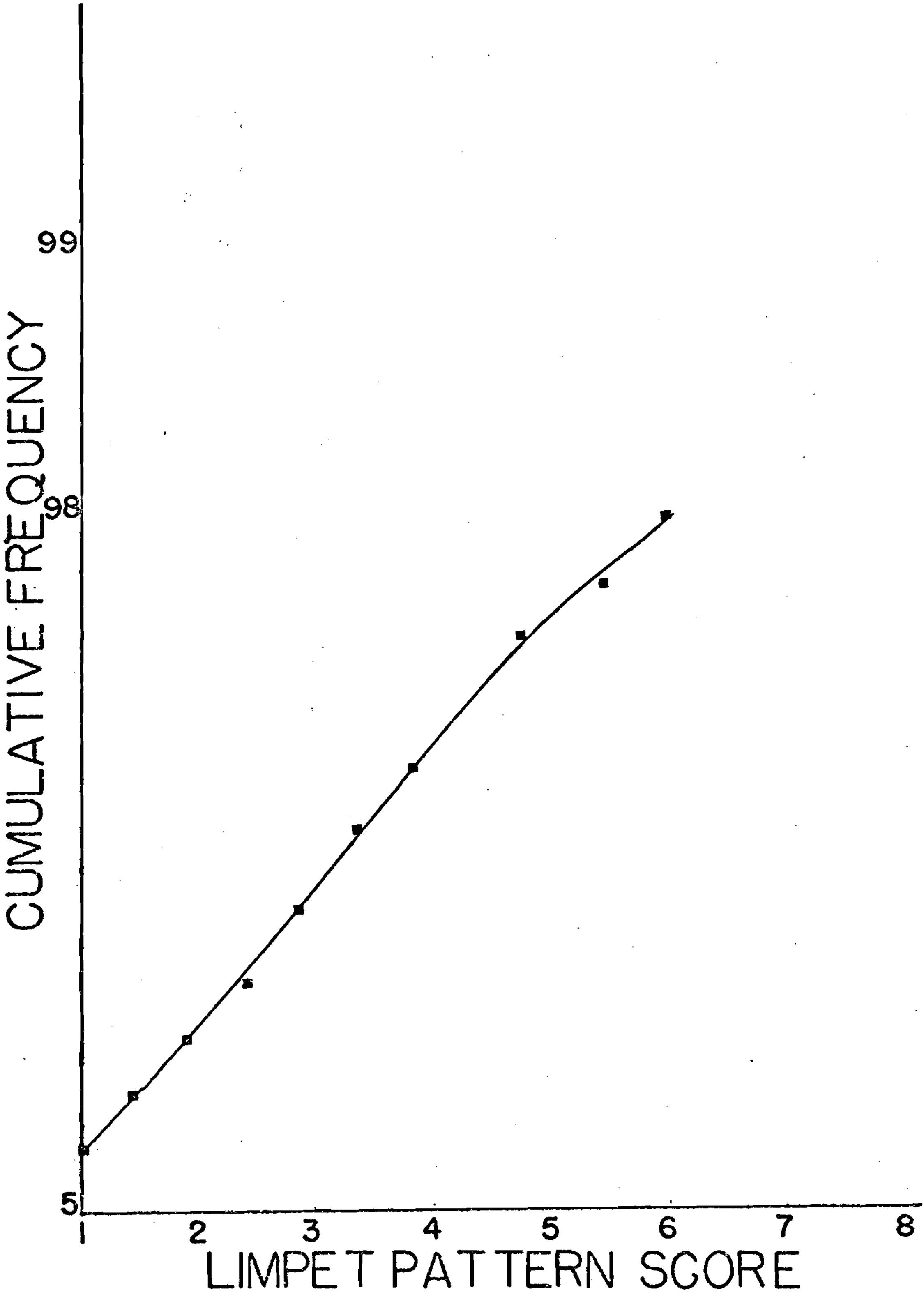
Villee, C. A. and T. C. Groody. 1940. The behavior of limpets with reference to their homing instinct. Am. Midl. Nat. 24: 190-204.

Wright, S. 1931. Evolution in Mendelian populations. Genetics 16: 97-159.

Appendix A

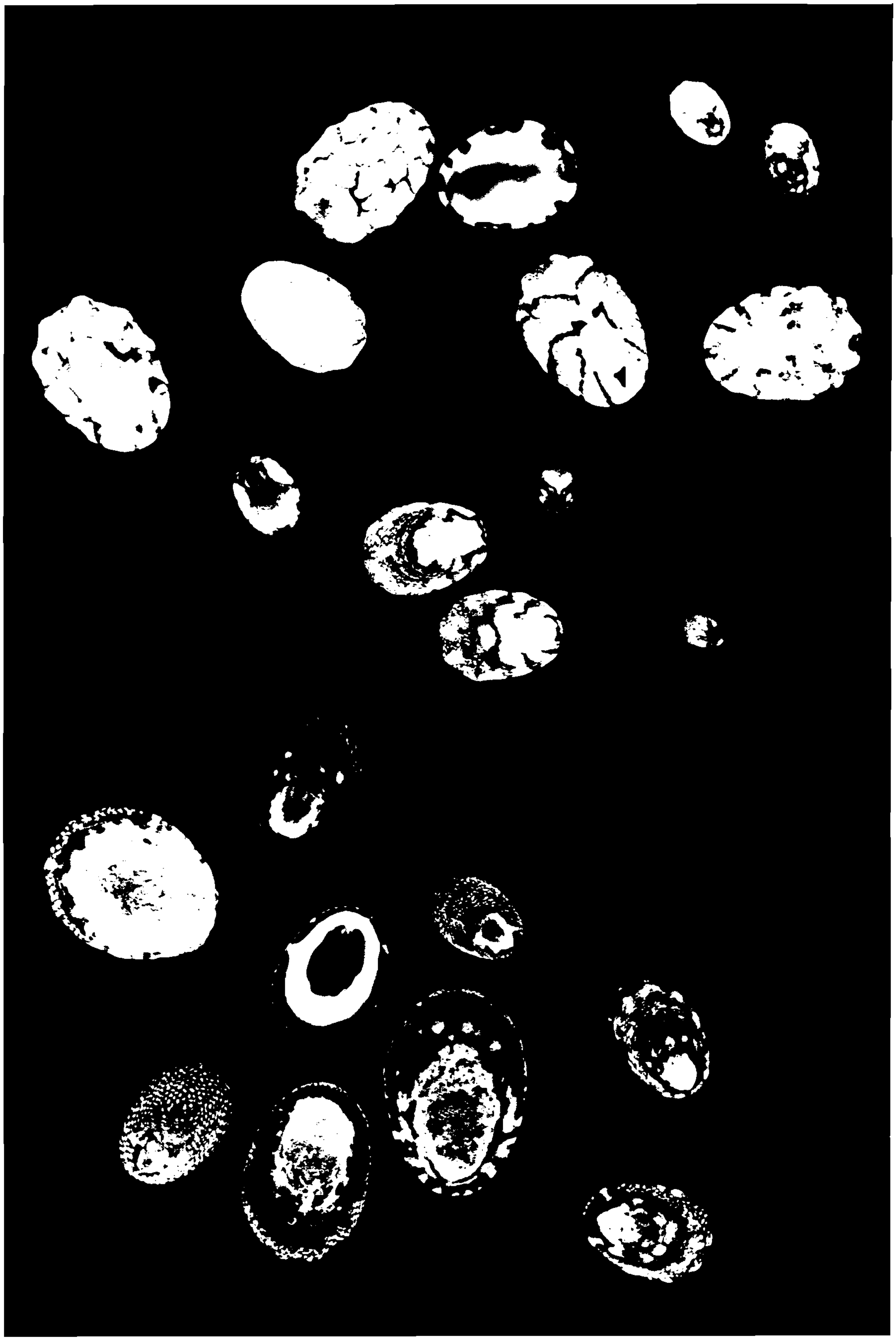
Normal probability plot of frequencies of pattern scores of a sample of Pollicipes-type limpets.

N=131


CUMULATIVE FREQUENCY
99
98
5
1
2
3
4
5
6
7
8
LIMPET PATTERN SCORE

APPENDIX B

Representative *Pollicipes*-type (top), rock-type (bottom), and intermediate (middle) limpets.



APPENDIX C

Numbers used in the calculation of *I* values.

```
                          MONTH  1
SIZE CLASSES 1-7   VERTICAL AXIS
                        PATTERN CLASS NUMBERS
  1   2   3   4   5   6   7   8   9  10  11  12  13  14  15  16  17
 12  20  20  34  34  39  31  19  16  14  20   9  17   1  11   0   0
  9  19  15  25  25  30  17  14   9  12  13   6  12   1   7   0   0
  8  18  12  22  17  22  14  11   6   6  10   4   6   0   5   0   0
  7  15  11  21  17  21  12   8   4   4   6   3   3   0   5   0   0
  6  15  11  17  14  18  12   8   4   3   7   2   2   0   3   0   0
  5  15   9  16   9  13  13   5   2   3   4   2   0   0   2   0   0
  4   9   7   7   4   7   7   3   0   2   3   1   0   0   1   0   0
```

```
                         MONTH  2
SIZE CLASSES 1-7    VERTICAL AXIS
                      PATTERN CLASS NUMBERS
 1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17
 41 21 54 44 67 55 66 28 28 14 28  8 28  1 13  0  2
 41 18 41 41 57 42 55 22 19 11 20  7 14  2 13  0  2
 38 18 39 30 39 36 46 15 15  8 15  1 10  2  9  0  2
 37 18 33 28 36 30 40 13 13  8 10  2  6  2  8  0  1
 34 18 31 28 34 28 34 12 12  8 10  2  4  2  4  0  0
 30 16 29 21 30 22 24 12 11  3  8  2  3  1  4  0  0
 18 15 25 11 18 10 18  8  9  4  5  0  2  2  0  0  0
```

```
                           MONTH  3
SIZE CLASSES 1-7    VERTICAL AXIS
                        PATTERN CLASS NUMBERS
 1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17
  48 17 26 30 89 45 62 13 28  6 39  3 19  0  4  0  1
  44 17 26 25 77 38 51  8 28  4 27  5  9  0  4  0  0
  39 13 20 20 52 34 38  6 19  3 23  2  5  0  4  0  0
  41 12 21 13 40 27 27  4 13  3 11  1  4  0  1  0  1
  37 12 21 13 37 27 20  4 13  2 10  1  4  0  2  0  0
  34  9 15 14 34 19 16  2 10  0  6  0  4  0  0  0  0
  19  7  9  7 15  9  8  1  6  1  5  0  2  0  0  0  0
```

```
                          MONTH  4
SIZE CLASSES 1-7   VERTICAL AXIS
                    PATTERN CLASS NUMBERS
 1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17
 45 32 33 47 99 64 80 25 33 23 37 12 18  7  3  0  4
 42 31 21 44 75 55 62 17 18 16 27  9  7  5  3  0  3
 41 29 18 32 72 38 51 15 16 12 21  4  4  4  2  0  2
 38 26 19 26 52 36 37 10 14 10 15  2  3  4  3  0  1
 37 20 16 25 47 28 33  9 11  8 11  1  0  2  1  0  1
 33 22 16 20 39 18 24  5  6  7  7  1  0  2  1  0  0
 16  9  7 12 10  7 10  4  4  5  5  1  0  1  1  0  0
```

```
                           MONTH   5
SIZE CLASSES 1-7    VERTICAL AXIS
                     PATTERN CLASS NUMBERS
  1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17
 35 22 17 35 77 47 63 27 39 15 30  6 25  9  6  0  2
 34 19 14 37 72 41 50 21 29 10 17  6 14  4  4  0  2
 34 17 11 23 61 31 37 17 22  4 13  4  7  3  2  0  2
 32 15 10 22 51 26 24 13 16  4 10  1  4  2  1  0  2
 31 14 10 16 39 21 21 10 13  3  9  0  2  1  1  0  1
 28 12  8 12 33 17 15  7  9  2  8  2  1  1  0  0  1
 15  3  3  6 13  3  6  3  8  0  4  0  0  0  0  0  1
```

```
                        MONTH  6
SIZE CLASSES 1-7   VERTICAL AXIS
                     PATTERN CLASS NUMBERS
  1   2   3   4   5   6   7   8   9  10  11  12  13  14  15  16  17
 45  30  42  40  70  38  50  23  28  14  29  14  25   6   6   0   1
 43  28  38  38  65  30  45  16  22   8  16   6  16   5   3   0   1
 37  26  34  32  50  23  36  12  14   5  12   4  10   4   2   0   0
 33  25  24  23  43  17  30   9  13   4   8   1  10   3   2   0   0
 35  21  20  21  30  13  24   5  11   4   5   0  10   2   2   0   0
 33  22  14  16  25   9  20   4  10   3   5   0   9   2   1   0   0
 17   5   4   8   8   2   5   3   5   2   5   0   4   2   1   0   0
```

```
                         MONTH  7
SIZE CLASSES 1-7   VERTICAL AXIS
                      PATTERN CLASS NUMBERS
  1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17
 31 26 41 23 55 36 52 18 38 15 31 10 21  4 24  1  3
 29 25 36 24 49 32 50 13 30  9 18  5 12  4 11  0  3
 27 24 31 22 42 26 41  9 20  6 12  5  9  4 12  0  1
 26 23 24 16 30 19 31  8 17  3  9  5  5  1  6  0  0
 22 23 24 13 25 13 23  7 15  2  8  1  3  1  6  0  0
 22 21 22  9 22  8 19  7 10  2  7  0  2  1  4  0  0
 15 10 11  4  7  4  7  2  4  1  5  0  1  0  2  0  0
```

```
                        MONTH  8
 SIZE CLASSES 1-7   VERTICAL AXIS
                      PATTERN CLASS NUMBERS
  1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17
  43 24 53 39 58 25 67 16 49 12 29  7 24  1  8  0  0
  41 24 51 37 53 25 54 10 34 11 25  4 13  1  6  0  0
  39 22 48 32 48 21 47 10 25  5 16  3  7  0  5  0  0
  34 18 45 30 44 21 39  6 18  1 10  1  5  0  4  0  0
  29 16 35 20 32 14 27  5 14  1  6  1  3  0  4  0  0
  25 12 27 14 19 10 16  3 11  1  1  0  2  0  2  0  0
  13  2 13  6  9  6  7  1  2  0  1  0  2  0  1  0  0
```

```
                             MONTH  9
SIZE CLASSES 1-7   VERTICAL AXIS
                          PATTERN CLASS NUMBERS
  1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17
 36 13 52 35 81 22 72  7 43  4 33  1 21  0 19  0  5
 35 14 54 34 80 18 68  8 35  2 29  1 16  0 13  0  4
 33 14 46 26 72 18 58  7 25  2 22  0 12  0 12  0  4
 31 10 43 25 69 17 46  4 20  2 16  0  8  0  7  0  1
 25  8 36 24 60  8 43  4 15  0 11  0  7  0  5  0  1
 21  7 17 19 35  6 23  1 10  0  5  0  3  0  3  0  1
 15  4  6  8 19  3  9  1  7  0  5  0  1  0  2  0  0
```

Typed by

Betty Jean Giesel