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EXTENDED ESSAY TITLE PAGE

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EXTENDED ESSAY TITLE: Microorganisms Association with Halosaccion glandiforme

IB SUBJECT: Biology
BEST LANGUAGE[S]: Arabic
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LANGUAGE B: English B
Approximate number of words: 4200

ABSTRACT

This study involves the microorganisms associated with *H.glandiorme.* The samples of *H.glandiforme* were taken from Race Rocks Island in the Race Rocks Ecological Reserve.

In this study the main purpose was to detect the effect of certain characteristics of the habitat (*H.glandiforme*) on the diversity and population of the species present inside *H.glandiforme*.The results of this study show that the diversity and population of the species present in *H.glandiforme* is sometimes affected by the factors studied.The factors that were studied include total surface area of the sample,the location of the species within *H.glandiforme* and the fact that some samples have their top part open and others closed.

TABLE OF CONTENTS

INTRODUCTION:-

Organisms live in an interrelated ecosystem with each other and with their surroundings.

The relationship between communities of the same habitat can be classified into three types:-

(i) Direct competition, in which organisms compete for the resources available in the habitat.

(ii) Predation, in which one organism uses another's biomass as a source of energy and for building its body.

(iii) Symbiosis, in which organisms live together. This is of three types:-

(a) Parasitism, in which one species benefits and the other is harmed.

(b) Mutualism, in which both species benefit.

(c) Commensalism, in which one species benefits and the other is neutral.

This shows that the reason why species interact with each other in a habitat is to utilize the resources available in the habitat - light, shelter, food, etc. - to continue their metabolic cycle. This will exhibit the dominance of certain species in certain habitats because of their ability to adapt better than others to that habitat.

In this essay the habitat used for this study is the marine algae Halosaccion glandiforme.

H. glandiforme is a red algae which grows on rocky shores in the middle of the intertidal zone, is sausage shaped with a length of up to 30cm and a diameter of 2-3cm(Waalands1977, p. 84). It consists of a hollow sac full of water with its top portion filled with gases that have been trapped as a result of rapid photosynthesis (Waaland 1977, p. 84). The algae has thin walls and the older ones sometimes have their top portion open, which leaves the algae flat. The algae are found along the Pacific shores "from Alaska to Mexico" (Waaland 1977).

This algae was used to study the microorganisms associated with it and try to link certain qualities of this habitat (eg. size, location,...etc.) with the diversity and abundance of those associated organisms.

This is more of a qualitative study for these organisms but some quantitative analysis was necessary to test the hypotheses put forward trying to explain and link some of the factors with each other.

THE PROBLEM:-

- What kind of microorganisms are associated with H. glandiforme?

- How many of these organisms are common to all samples and in what proportion?

-Is the diversity of these organisms dependant upon the location within the habitat and/or upon whether the habitats are "open" or "closed" (i.e. whether the algae's top portion is still intact or not)?

- Is the number of organisms of the same species dependant on the location in the habitat?

- Does the surface area of the habitat affect the diversity of organisms present?

HYPOTHESIS:-

- The location of the species does not affect the frequency of the species present.

- The diversity of the species is not dependant on the surface area of the habitat nor on the type of habitat.

- The average number of organisms of the same species will not be the same in all locations.

- The total number ot all organisms in one sample will not be different from the others in that specific location.

ASSUMPTIONS:-

There are some limitations on these assumptions which will be mentioned later.

1- Species present will show a distribution which is the same or very similar to thé normal distribution.

2- There is no migration or immigration from or to the habitat.

3- The time between collection of samples and collection of data is not so long as to cause any changes in the diversity or population of species or the habitat itself.

4- H. glandiforme shows metabolic activities during the experiment similar to those in its original habitat.

5- There was no change in the nutrient supply to H.

glanditorme during the experiment period.

6- The habitat is an ecosystem.

LIMITATIONS:-

The limitations on this research are:-

1- Time:- this research and the data collection were done at a specific time of the year so the results cannot be generalized.

2- Temperature:- the temperature at which the H. glanditorme was maintained in the laboratory was higher than that of the Ocean.

3- Geographical location:- these samples were collected from Face Rocks island. These samples and the organisms present inside of them may be different from samples taken from other locations.

4- There are some microorganisms that are in the water inside the sac. These were not studied and were not taken into consideration in relation with the ones attached to the walls which were the subject of this study.

5- The microorganisms observed were the ones seen under magnification power 10X. There are some other, smaller microorganmisms that were not studied at all.

6- H. glanditorme is assumed to be an ecosystem. This system is not in isolation from other systems and it is subjected to interactions with other organisms. These interactions may be positive or negative and this will affect the stability of the ecosystem and cause fluctuations in the number and diversity of species present there.

DELIMITATIONS:-

In this study a combination of subjective and objective methods was used to study the populations of the microorganisms present in the *H. glandiforme*. The organisms taken into acccount were the ones attached to the walls that appeared under magnification power 10x. Although there are some smaller microorganisms that cannot be seen under that magnification, it is assumed that these are there all the time and do not affect the organisms studied, or if they do, the effect is negligable.

These experiments took place at Pearson College not very far from Race Rocks and the samples were put in tanks where there was running seawater so the changes in temperature were not sufficient to cause changes in the habitat or the organisms. Because the college is very close to Race Rocks there was negligable change in the geographical location or nutrient supply or other climatic factors.

The time between sample collection and data collection was less than twelve hours, which is considered insignificant to cause any fluctuations in the algae and the organisms.

Although *H. glandiforme* interacts with other species it is assumed that this interaction occurs all the time and it might not have an effect on the organisms. However, if it does, the effect is assumed not to be instantaneous therefore these species live inside *H. glandiforme* under these conditions because of their ability to adapt to these conditions at that particular time of the year.

LITERATURE REVIEW

H. glandiforme, habitat: lives on rocky shores in the middle of the intertidal zone (Waaland 1977, p. 84). This habitat is affected by the movement of the tide during the day and seasonal fluctuation in temperature.

H. glandiforme is a red algae and contains photosynthetic pigments which enable it to photosynthesize at different light wavelengths (Carefoot, 1977, p. 73). This specific character makes it more interesting to study besides the fact that red algae have a comparatively low rate of respiration (Carefoot, 1977, p. 72) which enables them to live at great depth in the water. *H. glandiforme* is able to photosynthesize at various light qualities and intensities so it will not be affected very much by light fluctuations due to tidal movement.

Another factor affecting the habitat, which is very much dependant on the tidal movement, is nutrient supply. The nutrients are brought to *H. glandiforme* from seawater or from air. *H. glandiforme* is capable of photosynthesis so it absorbs CO from air and sometimes the alternative is the CO gas dissolved in water which is found in the form of carbonate or bicarbonate ions. The water sac is assumed to be not very useful in storing nutrients, but it eases the utilization of nutrients that are brouht by the water when the sac is refilling with water

Temperature is a very important factor affecting the habitat. The metabolic rate is mainly affected by temperature fluctuations, since *H. glandiforme* lacks mechanisms of body temperature regulations. (Sumich, 1982, p. 14)

Temperature of seawater is the main factor controlling the metabolic rate of marine life The distribution of organisms living in seawater is associated with this factor (Sumich, 1982).

Organisms have specific requirements for food and space, certain qualities of the space is needed to provide organisms with the supply they need of food, shelter and other requirements. This is the reason why certain organisms are capable of adapting to specific environmental factors.

In marine life some species show seasonal fluctuations in their numbers. This is mainly due to fluctuations in water temperature, light, and availability of nutrients e.g. diatom, which bloom in the spring (Sumich, 1982, p. 15).

The combination of climatic factors, nutrient availablity, and presence of associated organisms make for the diversity of marine life and life on earth in general.

In the samples seen in the experiment there were marine algae samples, zoolplankton, phytoplankton and diatoms. These are mainly primary producers with the exception of zooplankton. the nature of the interaction between these species and H. glandiforme and within these species was not studied but there are some predictions based on a study of some characteristics and requirements for these species.

H. glandiforme creates a good shelter for the protection of the zooplankton and at the same time phytoplankton is abundant there. The type of relationship with phytoplankton is predation because zooplankton uses phytoplankton's biomass as a source of energy and building blocks for its body.

H. glandiforme provides a shelter for the organisms inside. For the primary producers *H. glandiforme* may provide a good source of nutrients either from the water inside the sac or "if there are parasites" from the cells of *H. glandiforme*. To determine the specific type, further study is required.

Competition between species in a habitat is a common type of interaction, but among certain species other types of interactions occur. For example lichens might parasitize bryophates. Fungal components of lichens parasitize bryophates for a short period of time after germination, which may help to bridge the time gap until the algal component is encountered and this may eventually kill the bryophate (During, Van Tooren, 1990). There are many other types of interaction between species but in each case one or both species benefit from this interaction.

Association of species has a wide range of study. Some species may be associated in the form of the support of one species for another, e.g. *Sphagnum* supports whole microcommunities on or inside the plant and similar results for epiphytic algae was reported for aquatic bryophytes (During, Van Tooren, 1990).

Another type of association is observed between insects and flowers. Some insects help in the process of pollination of flowers and transfer pollen grains from one flower to another. This is an essential process for flowers and without the help of these insects pollination will not occur and no reproduction take place.

A typical example of association is found in the forest. The flying squirrel eats sporocaps of hypogeous fungi which contains nutrients, water, viable fungal spores, nitrogen fixing bacteria and yeast, (Maser, 1988). These are digested and the undigested materials are expelled as fecal pellets. These fecal pellets contain spores of hypogenius mycorrtizal fungi, yeast, nitrogen fixing bacteria and some nutrients for these organisms. If these pellets fall near a conifer feeder rootlet and environmental conditions are suitable for the fungal spores to be germinated, a new fungal colony is established. The fungal hyphae form a harmless mycorrhizal symbiosis with the roots. The fungus absorbs nutrients from the soil and translocates them into the host plant. Nitrogen fixing bacteria which are inside the mycorrhiza use the fungal extract as a food and fix the atmospheric nitrogen. The host plant provides simple sugars and other metabolities to the mycorrhizal fungi. (Maser, 1988).

This is a whole chain of associated species which support each other and the whole process is cyclical.

Interaction of species is beneficial for species to allocate the resources available in their habitat. These interactions may take several patterns but in the end one or probably more than one species benefit from the interaction.

METHOD

The data were collected in mid-autumn from a population of *H. glandiforme* from Race Rocks Island. Forty five samples were examined under the microscope at a power of 10X to find out how many and what kind of species were present on the walls of *H. glandiforme* that can be seen under that power of microscope.

The methods used to estimate populations are as follows:-

1) Direct counting if the number of species is small enough to be counted,

2) Approximate counting if the number of species is very high. The number of species per one unit area is counted and the total area covered by the species is estimated and converted to the number of species,

3) Percentage cover if the species are too dense to count or estimate.

Species that are common to all samples and frequently appeared were the ones dealt with. Other rare species were not counted or taken into account.

The samples tested were tested at three locations the top, middle and bottom parts to test if there is variations in species within one sample in different locations.

Species present were difficult to identify but some of them show some characteristics of epiphytic diatoms. Other species are more likely to be marine algae, zooplankton and phytoplankton.

Because of the lack of literature about these species and very few details about them from the experiment, it is very difficult to identify most of them.

Diagrams containing figures for these species and details of some of them are indicated.

All species are dealt with as unknown species and identified as unknown species and referred to by specific numbers.

PROCEDURE

Hallosacions were collected from Race Rocks on the 23rd of October and put into tanks where the water supply is from the sea.

These samples were tested on the same day by dividing each sample into three parts: top, middle and bottom. The surface area of each part was calculated using a transparency of graph paper and recorded.

Using a cork of area $0.2cm^2$ (see figures 1 and 2), five different samples of different locations on each part were obtained on a microscopic slide and examined under magnification power of 10X.

Species present on those samples were observed, drawn, counted and recorded so that the figures on the data sheet are for 1cm and those are the figures for species density at each location.

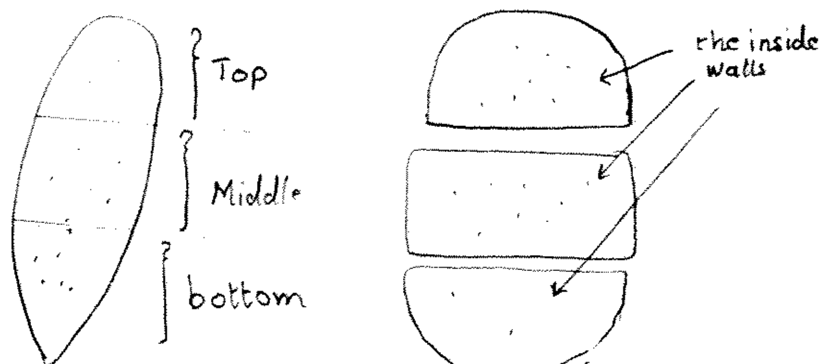


Figure 1 : The 3 locations of *H. glandiforme*

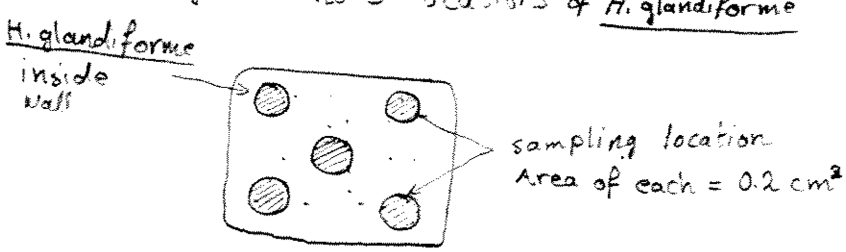


Figure 2 : The sampling methods

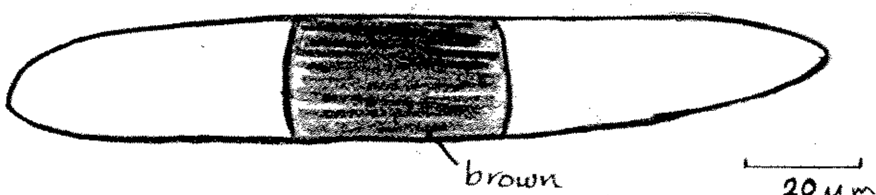
RESULTS

There were about 34 common species to all samples used. These species differ in the numbers and size and of course they were diverse and belong to different classifications.

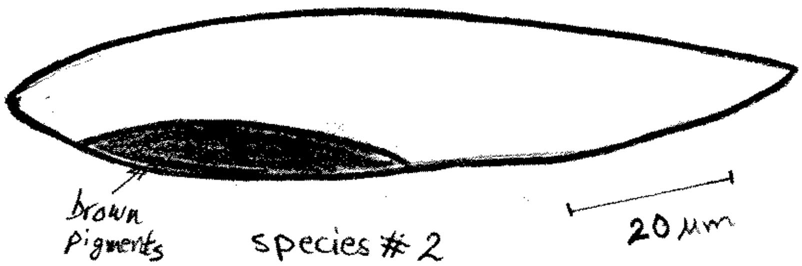
There were some fluctuations in numbers of organisms of the same species. Some of these fluctuations were very high others were very small, but the frequency of these numbers within one species shows a very close distribution to the normal one while others show a great deviation from the normal ditribution. There were very few organisms that appeared in some samples not very frequently and with very few numbers "max. 3" these ones were not studied or included in the study but were recorded and presented.

In different locations of the same sample there seemed to be no differences in the number of species present, but the number of organisms of the same species sometimes show differences with respect to the location of the species.

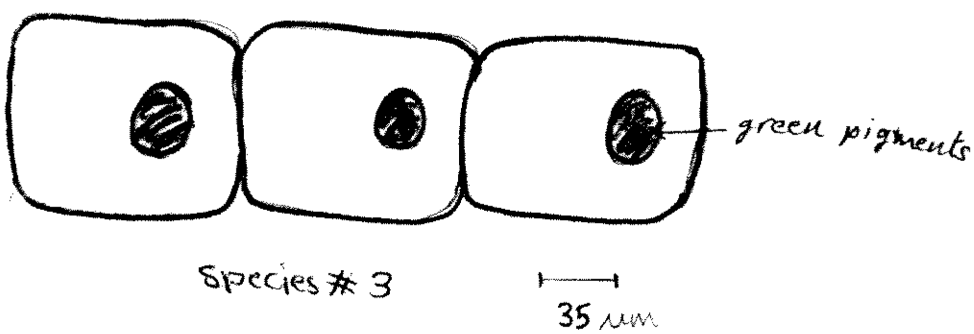
The type of habitat "Hallosaccion" whether it is open or closed seemed not to affect the diversity of species but the total number of species in the closed ones seemed to be larger in general than in the open ones.



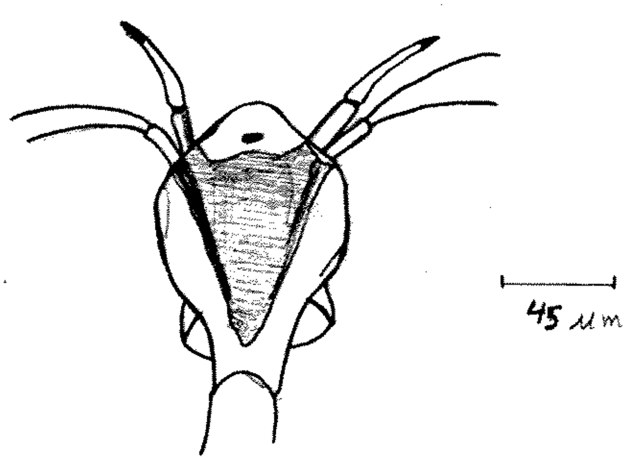
species # 1



species # 2

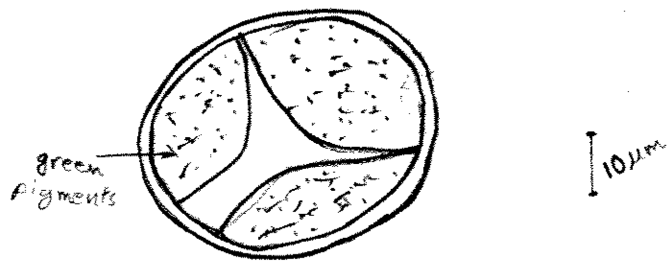


Species # 3



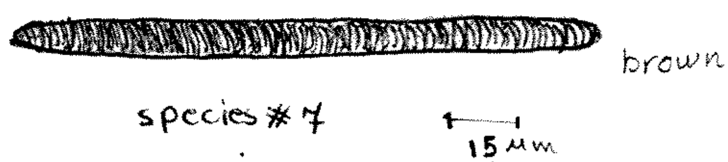
species # 4

Drawings of species found in *[illegible]*

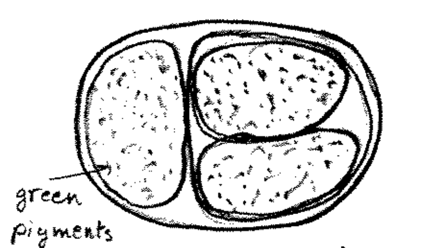


species # 5

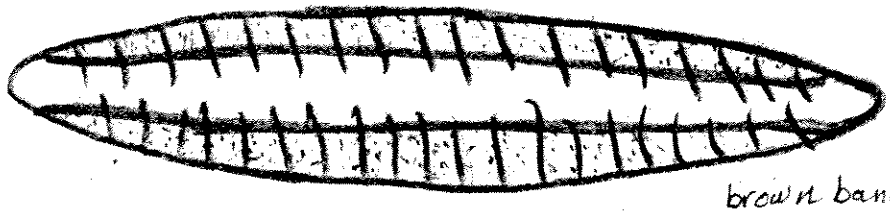
species # 6



species # 7



species # 8

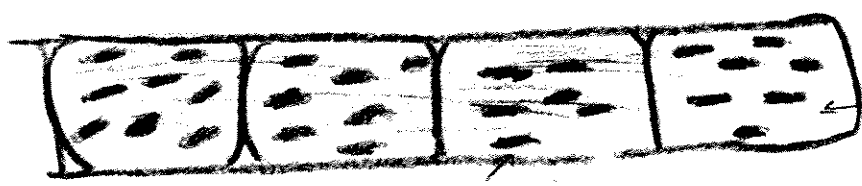


species #9

Probably Navicula sp.

species #10

Phormidium autumnale

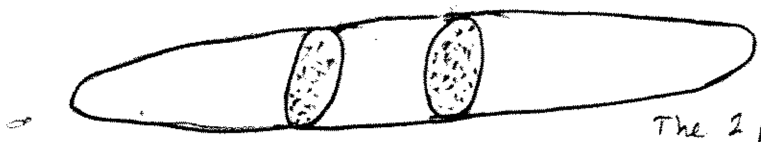


species #11

Melosira sp.

species #12

Gyrosigma or Pleurosigma



species #13

The 2 pigments inside have brownish red colour.

20 μm

species #14

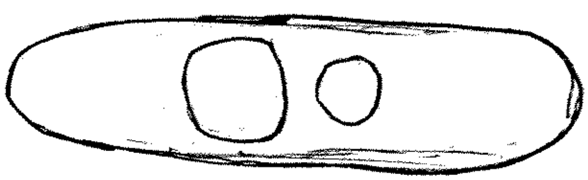
green pigments do not seem to be enclosed by membrane

25 μm

species #15

Pleurosigma

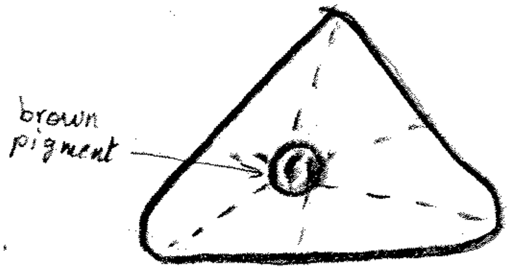
10 μm



Species #16

15 μm

Navicula



species # 17

10 µm

Triceratium americanum

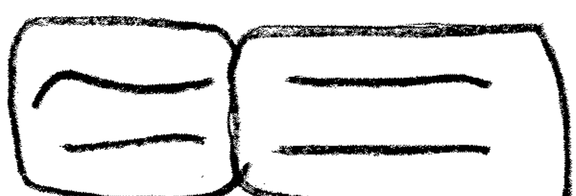
species # 18

50 µm

Nitzschia serrata

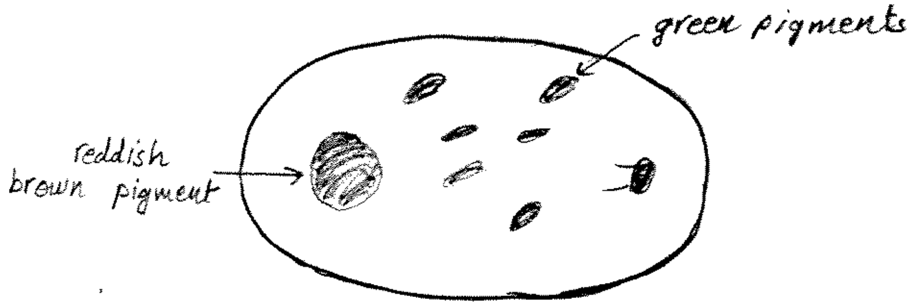
species # 19

10 µm



species # 20

25 µm

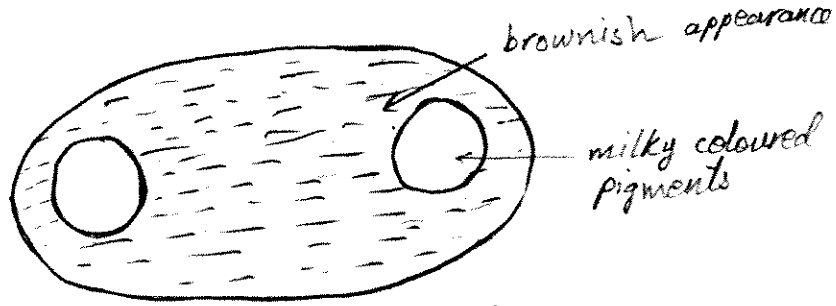


species # 21

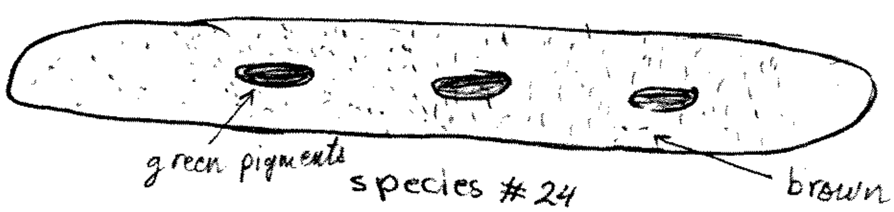
20 μm

species # 22

15 μm

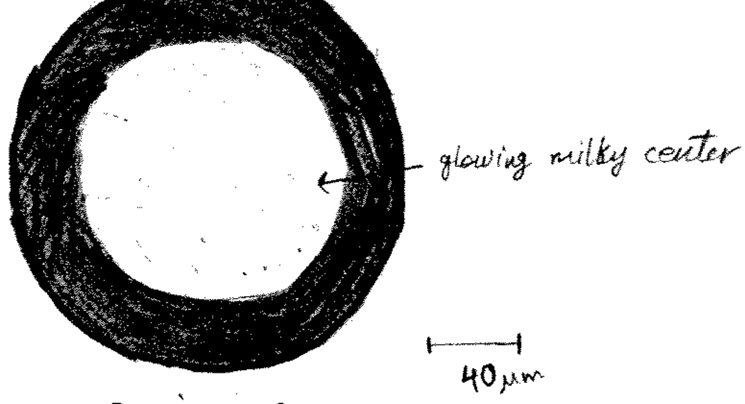


species # 23 30 μm



species # 24

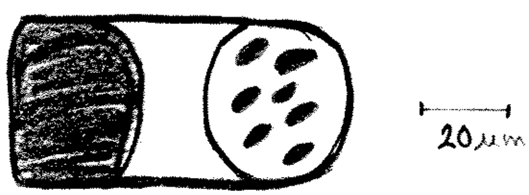
15 μm



species # 29

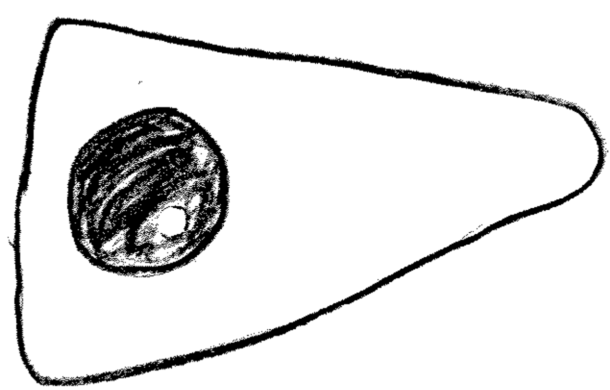
This species moves very fast

species #30



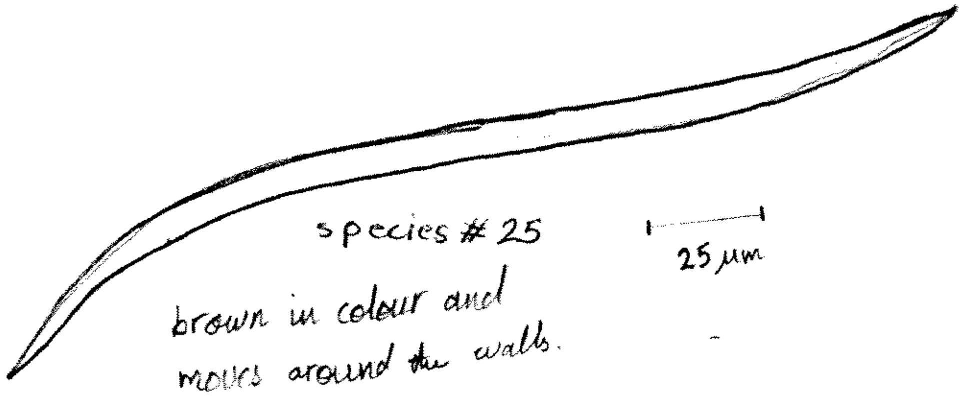
species # 31

Melosira sp.



species # 32 15 µm

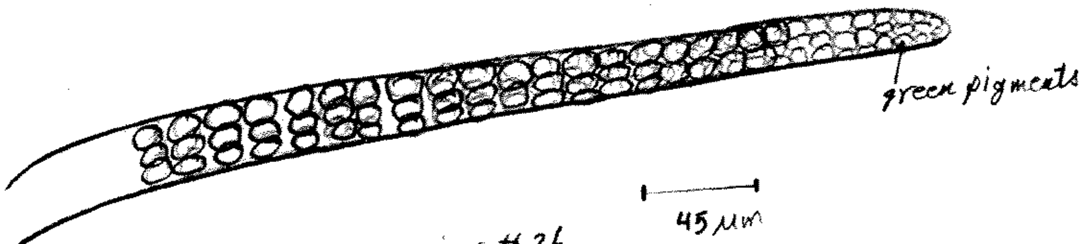
Licmorpha sp.



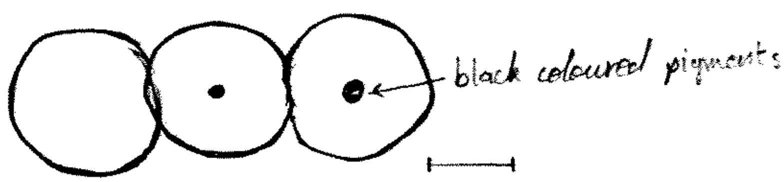
species # 25

brown in colour and moves around the walls.

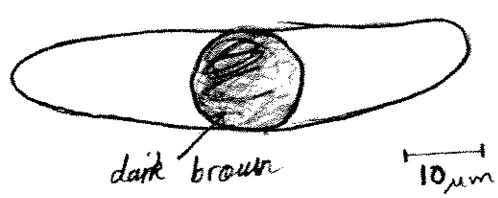
Thalassiothrix longissima



species #26



species # 27



species # 28

DISSCUSSION

The first hypothesis is related to the frequency of species with respect to their location in samples . Graphs (1a - 1c) show the 34 species on the x-axis and their frequency on the y-axis at three different locations in the habitat.The general pattern of the graphs shows that most species do not show very large deviations in their frequencies in these three locations. However, the frequency of some species seem to show rather large deviations in some locations.The hypothesis states that the frequencies of any species in all locations will be more or less the same.

The chi square (χ^2) test was applied to some of the species that show deviation and the result of it appear in table

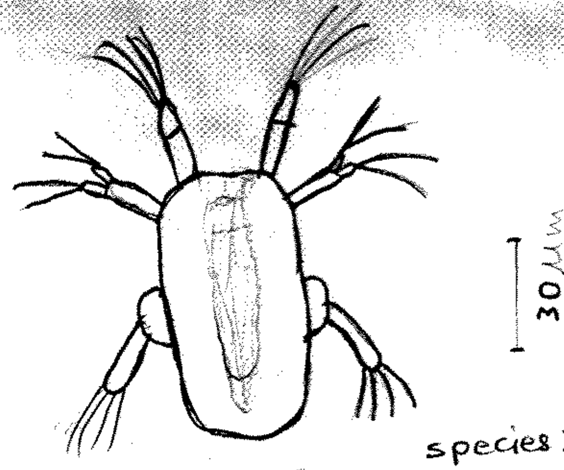
The chi square test is a simple statistical test for the significance of the deviation of an observed value from the expected one .

$$\chi^2 = \sum (O-E)^2/E$$

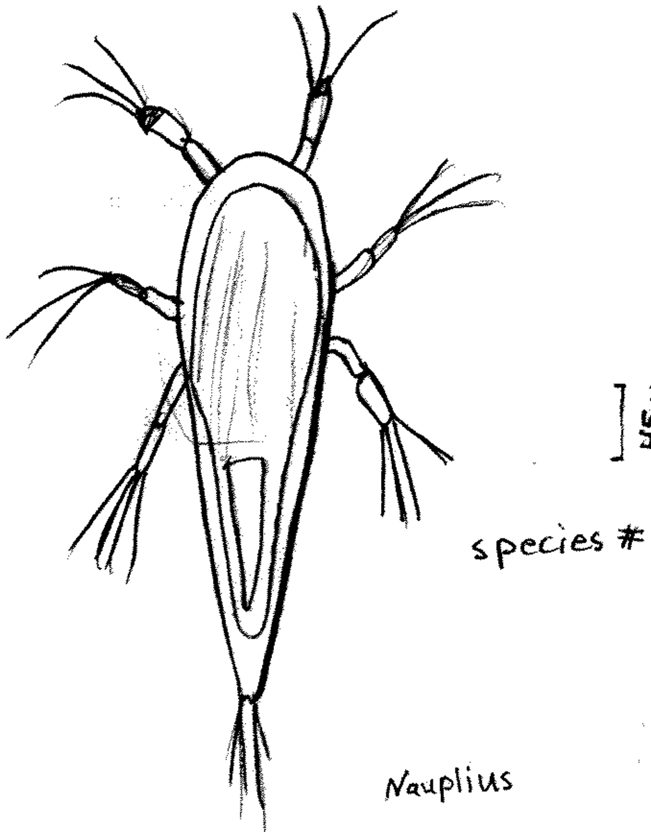
where O is the observed value .

E is the expected value.

The χ^2 values are then compared with critical values under a certain degree of freedom to find out the probability values (P), which then determine the significance of the deviation (See appendix)



species # 33



species # 34

Nauplius

Table I

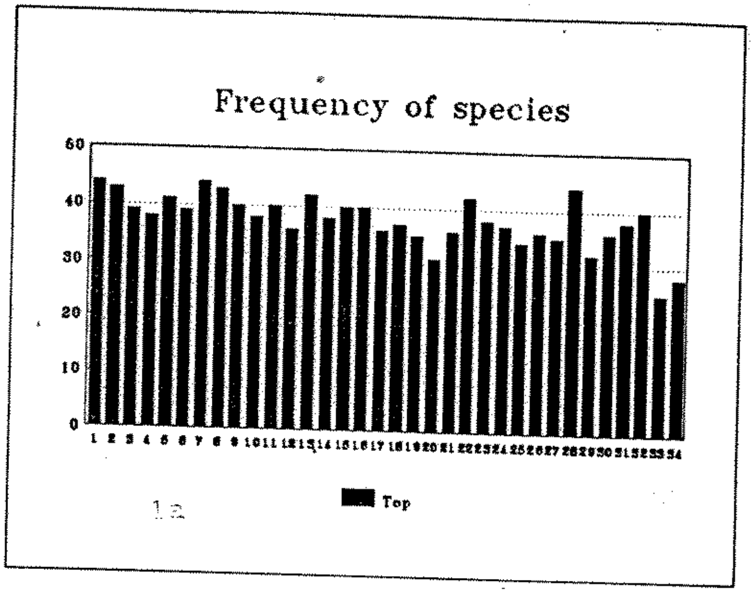
Species	O Top	Middle	bottom	E	χ^2	P
4	38	43	42	41	0.463	0.70
5	41	43	39	41	0.195	0.70
7	44	37	36	39	0.947	0.50
8	43	37	42	40.3	0.523	0.70
9	40	35	38	37.7	0.223	0.80
11	40	37	42	39.7	0.319	0.80
14	38	43	37	39.3	0.526	0.70
17	36	32	37	35	0.4	0.80
18	37	40	42	39.7	0.319	0.80
20	31	37	39	35.7	0.971	0.50
22	42	39	36	39	0.462	0.70
23	38	30	37	35	1.086	0.50
25	34	33	38	35	0.4	0.80
26	36	41	35	37.3	0.554	0.70
27	35	41	43	39.7	0.873	0.50
29	32	39	39	36.7	0.89	0.50
31	38	30	35	34.3	0.953	0.50
32	40	36	37	37.7	0.230	0.80
33	25	30	35	30	1.667	0.30

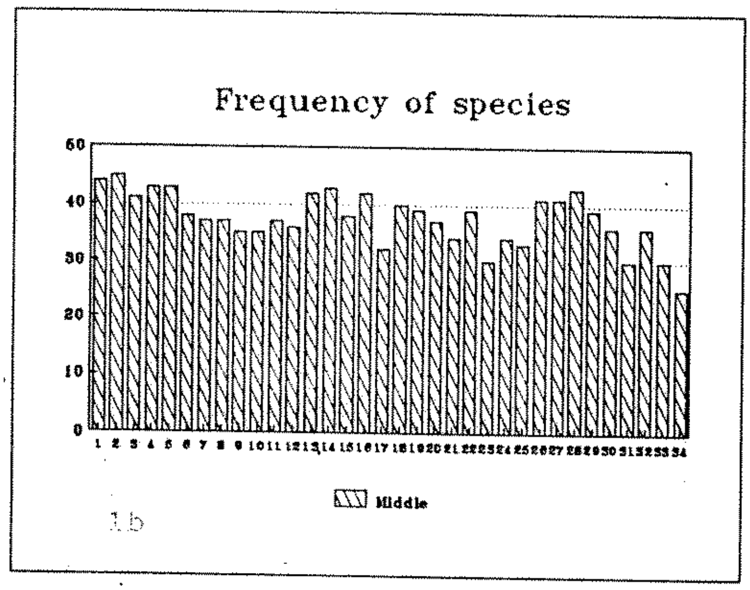
degree of freedom = 2

In this case the expected value is assumed to be the average of the frequencies at three locations (top, middle and bottom). The values in the table show very high probability values which indicates that the deviation is due to pure chance and upon this test, we can accept the hypothesis for the species present.

The second hypthesis deals with the relationship between the average number of different species present in samples and the total surface area of the sample and the type of the sample (open, closed). It states that there will not be any change in the average number of species as the surface area changes.

Graph I   Frequency of species in different locations





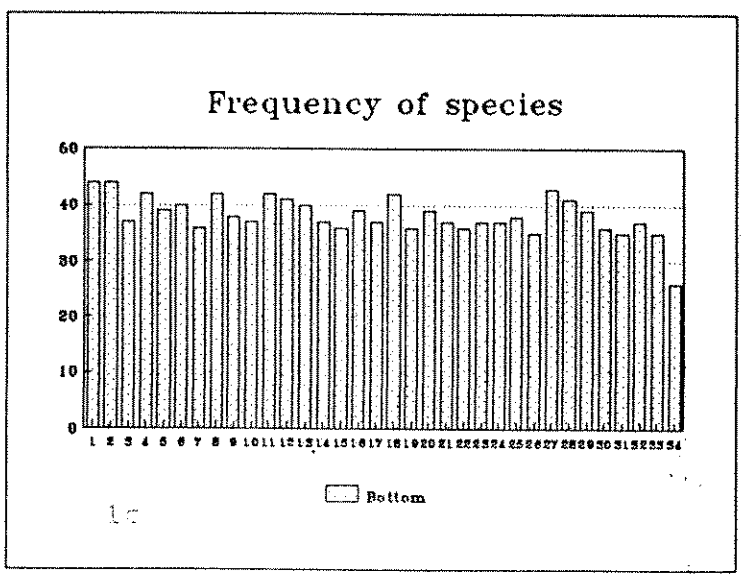


Table I

Species	O			E	χ^2	P
	Top	Middle	bottom			
4	38	43	42	41	0.463	0.70
5	41	43	39	41	0.195	0.70
7	44	37	36	39	0.947	0.50
8	43	37	42	40.3	0.523	0.70
9	40	35	38	37.7	0.223	0.80
11	40	37	42	39.7	0.319	0.80
14	38	43	37	39.3	0.526	0.70
17	36	32	37	35	0.4	0.80
18	37	40	42	39.7	0.319	0.80
20	31	37	39	35.7	0.971	0.50
22	42	39	36	39	0.462	0.70
23	38	30	37	35	1.086	0.50
25	34	33	38	35	0.4	0.80
26	36	41	35	37.3	0.554	0.70
27	35	41	43	39.7	0.873	0.50
29	32	39	39	36.7	0.89	0.50
31	38	30	35	34.3	0.953	0.50
32	40	36	37	37.7	0.230	0.80
33	25	30	35	30	1.667	0.30

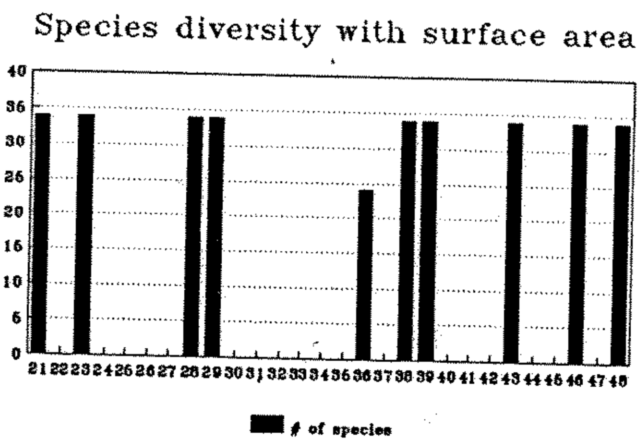
degree of freedom = 2

In this case the expected value is assumed to be the average of the frequencies at three locations (top, middle and bottom) .The values in the table show very high probability values which indicates that the deviation is due to pure chance and upon this test , we can accept the hypothesis for the species present.

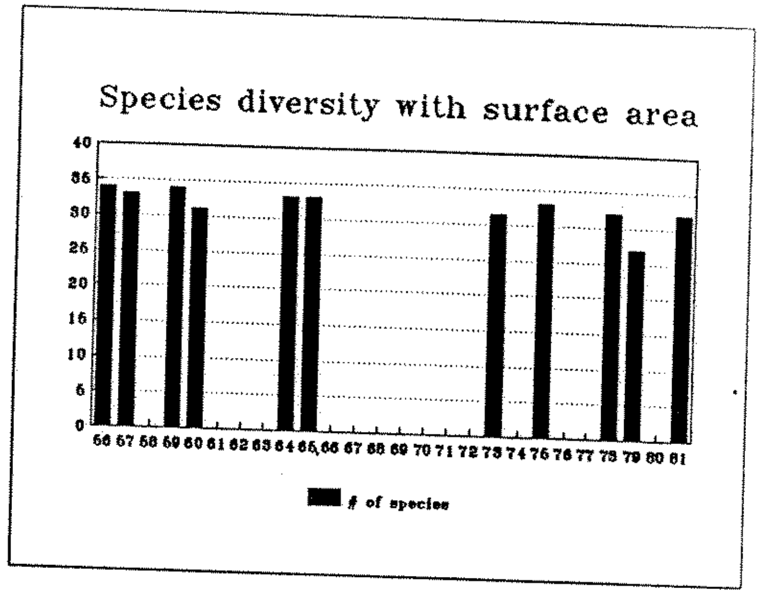
The second hypthesis deals with the relationship between the average number of different species present in samples and the total surface area of the sample and the type of the sample (open, closed) .It states that there will not be any change in the average number of species as the surface area changes .

Graph II

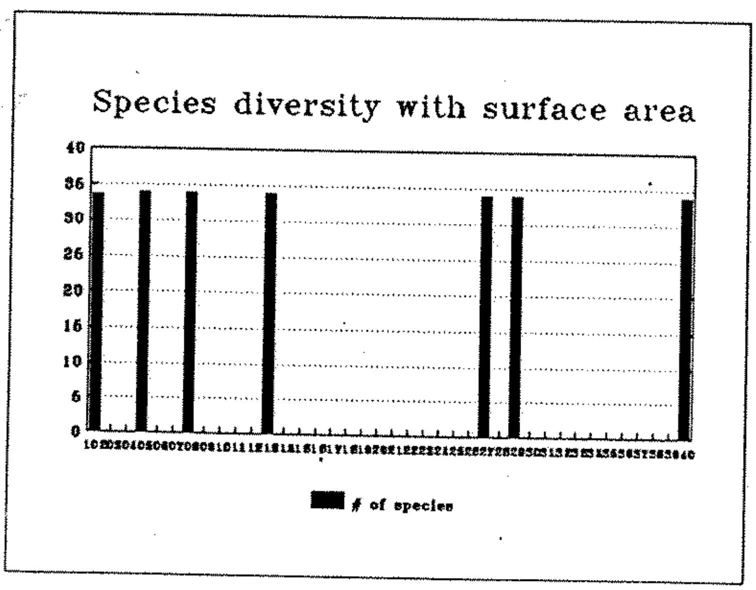
Diversity of species vs surface area



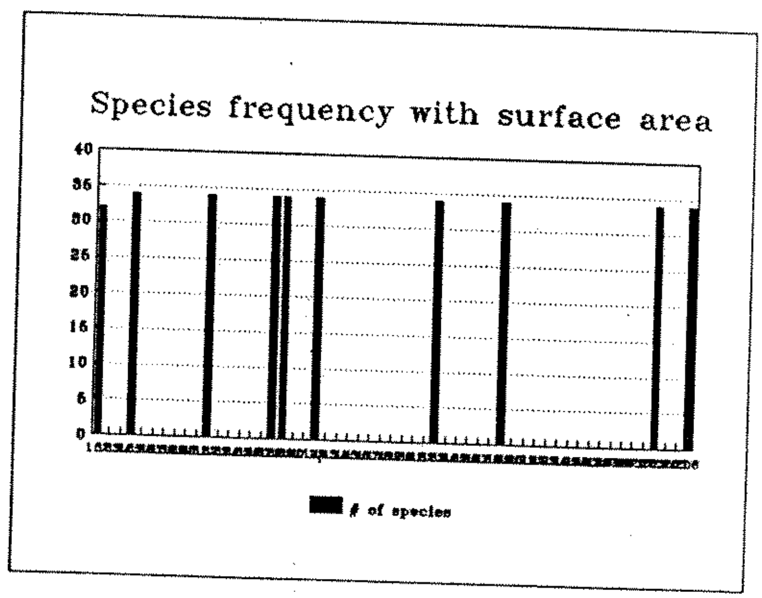
2a



2b



2c



2d

Table III

Species	O			E	χ^2	P
	Top	Middle	Bottom			
1	216	142	132	163.5	25.4	0.005
2	126	95.2	71.9	97.8	15.2	0.005
3	6.2	5.4	5.1	5.6	0.116	0.90
4	6.6	5	3.4	5	1.024	0.50
5	9.7	9.5	10	9.7	0.013	0.975
6	4.2	2.8	3	3.3	0.349	0.70
7	11.5	5.5	4.8	7.3	3.72	0.10
8	9.3	8.9	8.6	8.9	0.028	0.975
9	9.1	5.5	5.2	6.6	1.43	0.50
10	1.7	1.9	1.5	1.7	0.047	0.975
11	4.2	3.6	3.6	3.8	0.063	0.90
12	7.3	5.9	4.6	5.9	0.619	0.70
13	272	182	139	197	47.1	0.005
14	6.8	6	4.2	5.7	0.623	0.70
15	5.1	4.6	3.4	4.4	0.348	0.70
16	20.4	15.5	11.7	15.9	2.393	0.70
17	6.5	5.6	4	5.4	0.854	0.50
18	154	112	109	125	10.1	0.005
19	337	290	213	290	27.8	0.005
20	22.1	16.5	15	17.9	1.57	0.20
21	108	76.6	74	86.1	8.01	0.01
22	4.2	3.7	3.2	3.7	0.135	0.90
23	104	89.4	79	90.9	3.53	0.10
24	7.6	5.9	5.3	6.3	0.452	0.70
25	8.4	6.8	4.7	6.6	1.044	0.975
26	3.5	3.5	3.1	3.4	0.032	0.975
27	5.4	4.8	3.3	4.5	0.52	0.70
28	17.5	14.4	13	15	0.71	0.70
29	62	60	53.1	58.4	0.747	0.50
30	32.2	37	40.5	36.6	1.02	0.500
31	7	8.2	8.6	7.9	0.176	0.900
32	170	135	129	145	6.720	0.025
33	2.1	2.1	2.0	2.10	0.0048	0.995
34	1.7	2.2	1.6	1.80	0.1170	0.900

degree of freedom = 2

Graphs (2a - 2d) show the number of species against surface area of a sample and the general pattern of the graph shows that the number of species is almost the same for all surface areas with few deviations for some species. These species were tested by the χ^2 test as shown in table II :-

Table II

O	E	χ^2	P
33	33.22	0.0015	0.995
32		0.0448	0.995
27		1.165	0.995
24		2.559	0.995
33.5		0.0024	0.995
34		0.0183	0.995

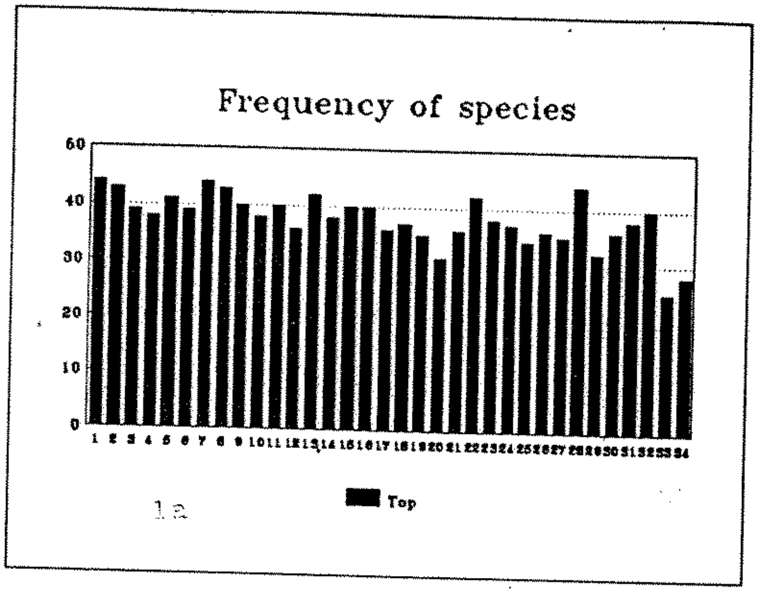
E, in this case, is the average of the number of spoecies found in the 38 possibilities.

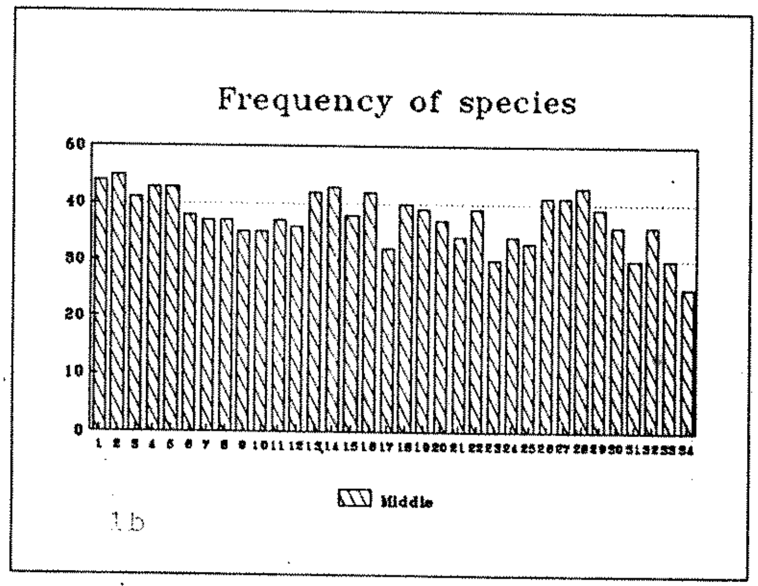
The degree of freedom in this test is 37 since we have 38 different surface areas.

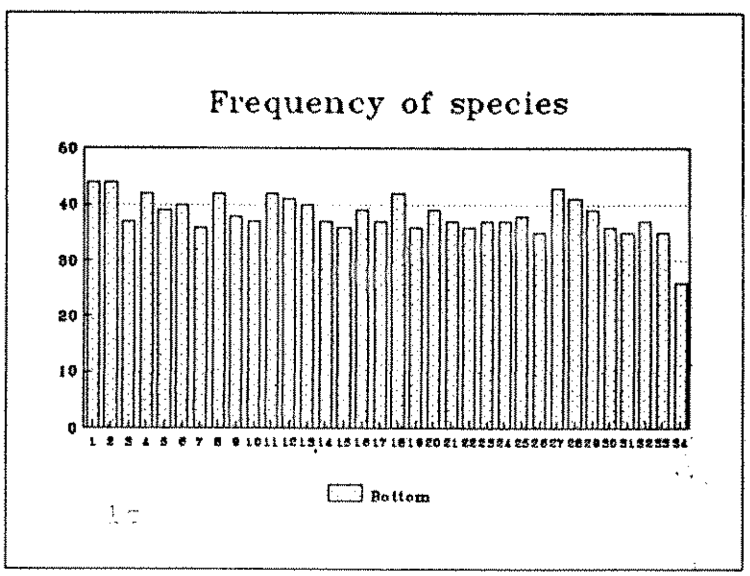
The table shows perfect agreement with the hypothesis. As the deviation is not very significant the first as well as the second part of the hypothesis is accepted since the samples are of both open and closed types. Thus we can say that the hypothesis seems to apply for this particular population.

The third hypothesis deals with the number of organisms of the same species in different locations. According to this hypothesis the number of organisms will be different from one location to another. Graphs (3a - 3d) show the average number of organisms in the three different locations represented on the y-axis and the 34 species on the x-axis were represented by numbers of organisms at three different locations.

Graph I Frequency of species in different locations

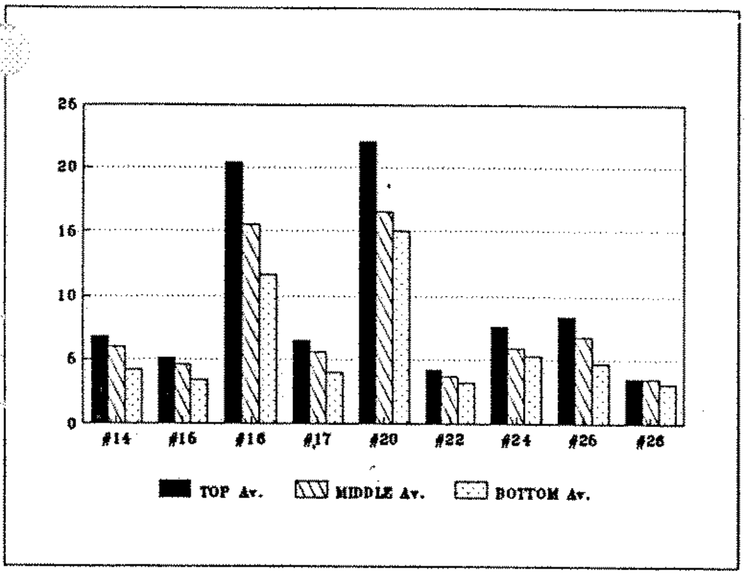




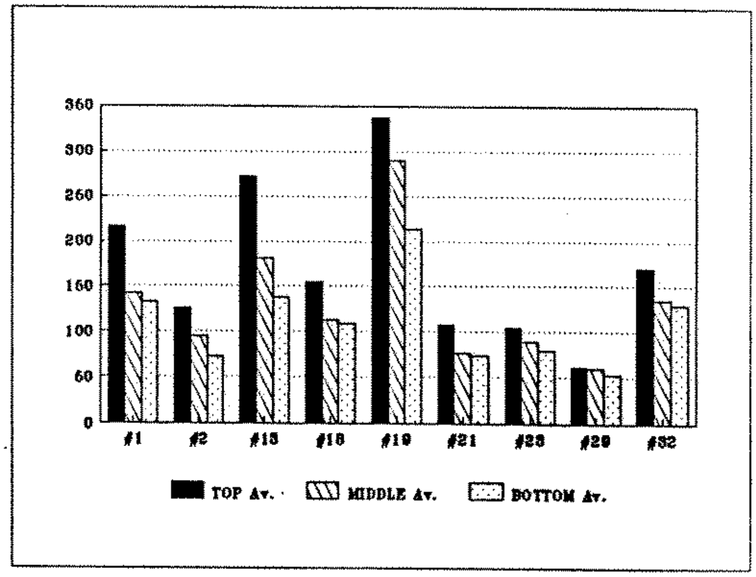


Graph III

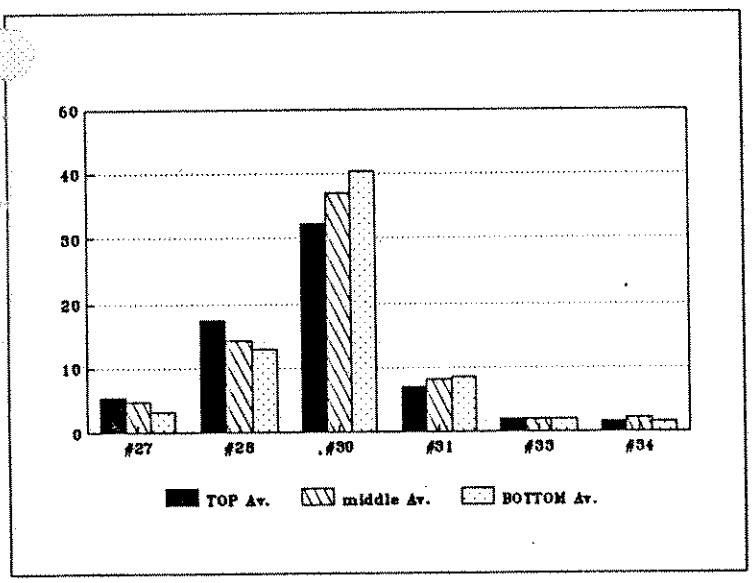
The average number of organisms at different locations



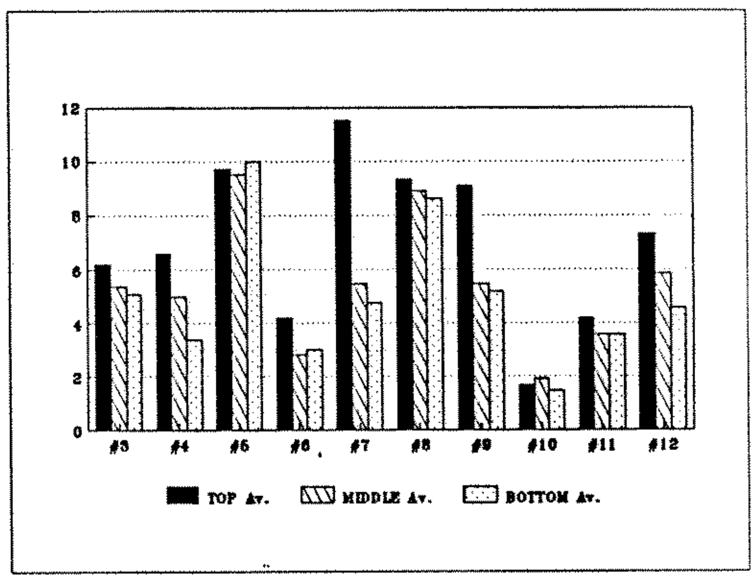
3a



3b



3c



3d

Looking at the graphs, some species seem to appear in large numbers in one location and smaller numbers in others, whereas most species do not show large deviations in number from one of the above locations to another. From this it seems that the alternative hypothesis "The average number of organisms of a particular species will not be different at those three locations" applies better to most species. If this hypothesis is true then the original is false and vise versa.

Table III shows the results obtained from applying the chi square test to the alternative hypothesis.

From the results obtained we can reject the original hypothesis for most species.

However there are some interesting results obtained from the table. The original hypothesis is accepted for most of the species in table 2b. These species appear in large numbers (i.e. over 100). The hypothesis is rejected for two species in that table (#23 and #29). These species appear in rather lower numbers than the others. From this it seems that the hypothesis is accepted only for species which appear in large numbers, whereas the alternative hypothesis is accepted for species that appear in small numbers. From this we can conclude that some species that appear in larger numbers show deviations in number from one location to another.

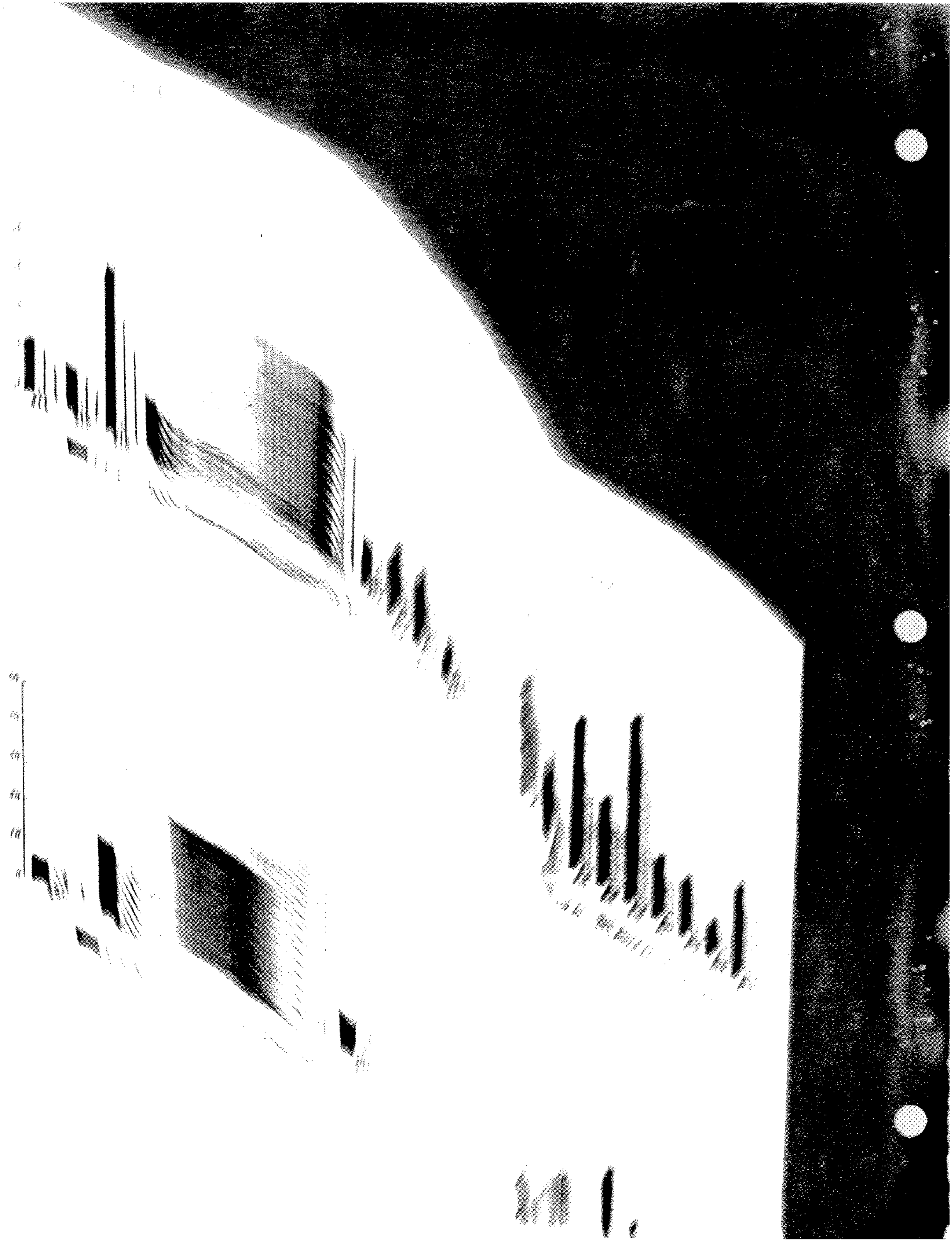
The hypothesis probably should be stated as :"The average number of organisms of most species will not be dependant on the location within the habitat". There may be an exception for species that appear in large numbers but this cannot be stated with great certainity and it still needs further investigation.

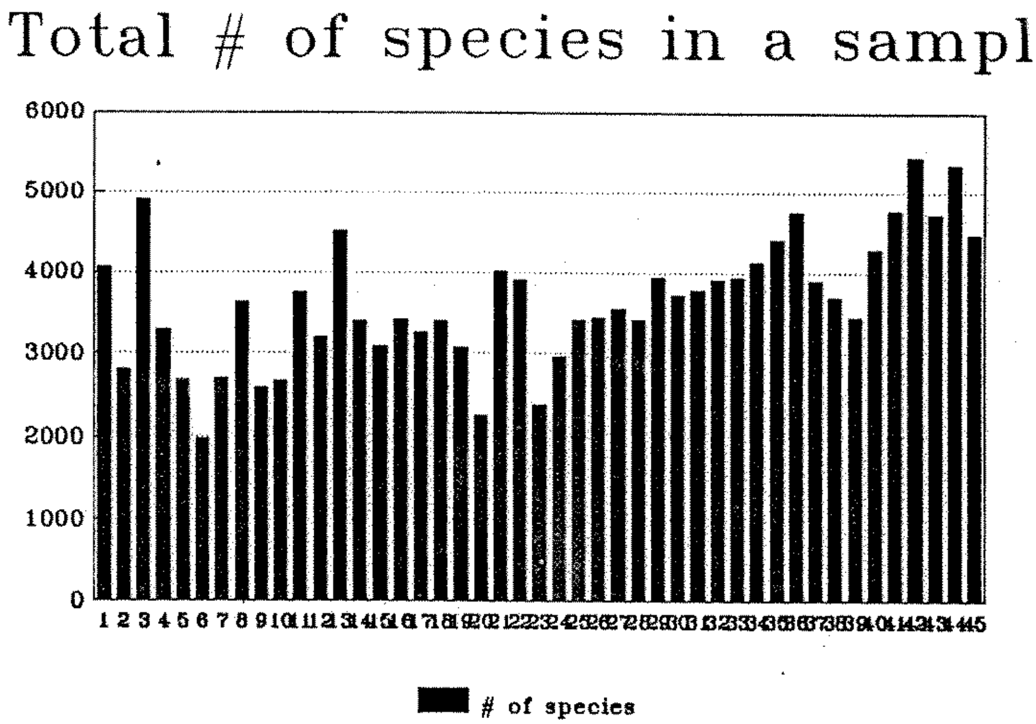
The last hypothesis deals with the total number of organisms of different species in a given surface area. The given surface area is considered a strip of an area 3cm that runs from the top of the sample to the bottom. The total number of species is considered the sum of all organisms of different species in all locations in that sample. Graph 4a shows the total number of species on the y-axis graphed against sample number on the x-axis. The graph does not give a clear idea of how significant the difference is between species. Graph 2b shows the best fitting straight line through these points. The slope of the line is approximately 23°. This is a large slope for a line that the hypothesis expected to be a horizontal line. The chi square test is done for 10 randomely choosen samples out of the 45. These samples show fluctuations which Table IV below expands upon:-

Table IV

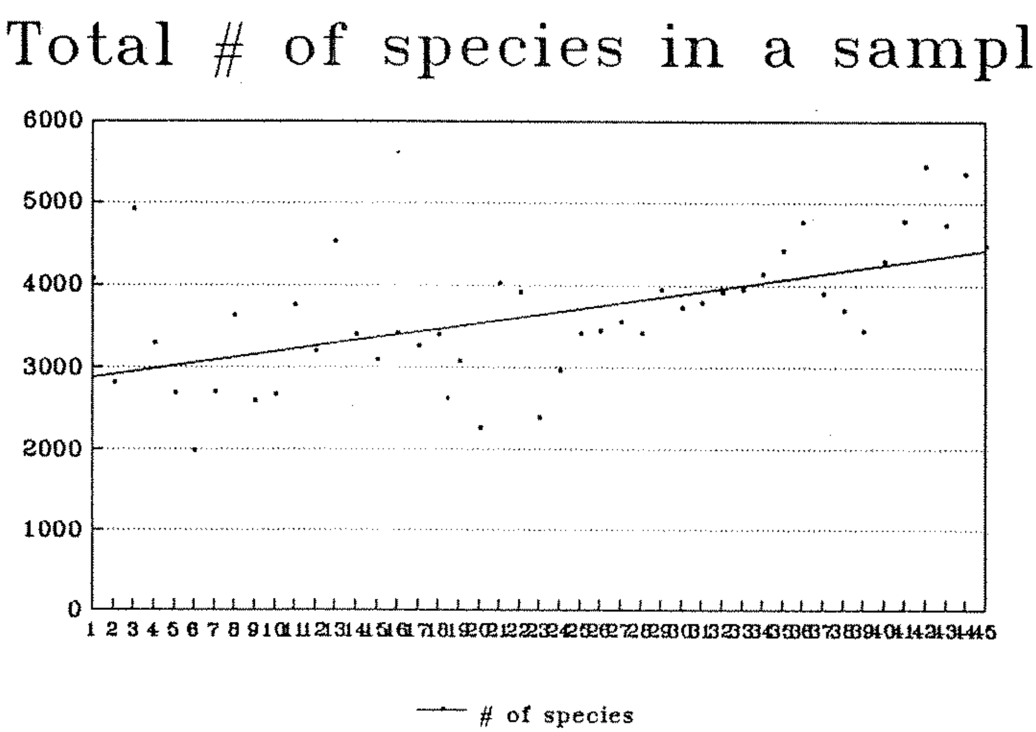
	O	E	χ^2	P
22	3902	3535	37.99	Rejected
12	3186		34.55	
17	3239		24.87	
23	2363		388.8	
43	4706		387.5	
28	3395		5.58	accepted
20	2241		474	Rejected
5	2666		213.8	
41	4759		423.4	
3	4898		525	

Degree of freedom = 9




Total # of species in a sampl
6000
5000
4000
3000
2000
1000
0
# of species

4a


Total # of species in a sampl
6000
5000
4000
3000
2000
1000
0
# of species

4b

The table shows that the total number of organisms in these randomly chosen samples show extremely high deviations. The expected value is chosen to be the average of the numbers in the chosen samples. Only one sample out of the ten showed an accepted probability value. Few samples from the total population will show accepted probability values but most of the samples will show rejected probability values. Upon this test we will reject the hypothesis and accept the alternative hypothesis which states that "The total number of organisms in a given area is not equal in all samples."

The hypotheses stated were in some cases accepted and in others rejected. These hypotheses were based upon some observations seen in some of the samples.

Surprisingly, the type of the habitat did not affect the diversity nor the number of organisms present. It was expected that the diversity of organisms in open habitats would be greater since the chance of a greater diversity of organisms that will be able to enter open habitats is greater. The factor that influenced the expectations of more diversity of species in open habitats is the fact that at low tide some organisms from air might have dropped inside. There are valid possibilities for more species diversity in open habitats than in closed ones but the organisms that might have entered there might have been victims of predation from other species inside or were not able to adapt to the life inside that habitat. Other organisms found in both types of samples have some ecological connections to _H. glandiforme_. The type of association between _H. glandiforme_ and the species inside it is a form of symbiosis and is not affected by the size, type (open or closed) or location (within

H.glandiforme) of the organisms.

Those organisms might not be found in H. glandiforme in different places or at different times of the year so the results obtained are restricted to the samples from Race Rocks at that time of the year.

SUMMARY:-

H. glandiforme is a red intertidal marine algae that has a water sac that fills with water at high tide. It has fluctuations on the metabolic rate related to climatic or chemical changes in the surrounding event.

The algae was studied under the microscope to find what kind of microorganisms that can be seen on the inside wall of the water sac under magnification power of 10X. The data were collected from a population of samples which were collected from Race Rocks Island in mid-Autumn (1990). These samples have different sizes and were of both types (closed and open).

There were 34 common species in those samples . The diversity and population of these species at three locations of H. glandiforme were studied and related to "location, size and type". The data were presented graphically and the related hypotheses were tested by the chi square test. Some of these hypotheses were accepted.

It is found that some of these factors (location, size and type) may have some effect on the diversity and the population of associated species. These results are limited by the factors of time, location and experimental procedure.

CONCLUSION AND IMPLICATIONS:-

The purpose of this study is to link certain characteristics of *H. glaniforme* with the diversity and/or population of the microorganisms found on the wall of its water sac. The experimental procedure emphasizes certain characteristics of the habitat of these microorganisms, namely size, total surface area, location within the habitat and the type of the habitat (open or closed). There are some other factors that may influence the diversity and/or the population of the species present inside *H. glandiforme* which were not studied in this research. These factors may have affected the species and resulted in some of the observations seen with regard to the diversity and population of the species.

The experimental procedure may have resulted in overlooking some other species that might have been there, and this potential flaw would have an impact on some of the hypotheses stated. Also if the observed species were identified, clearer ideas about the type of association between these species and *H. glandiforme* could be obtained.

There are limitations on the study (mentiond earlier) besides the limitations of the experimental procedure and the range of the limitations of the experimental procedure and the range of the factors studied. In the range of these limitations one cannot draw conclusions with great precision for the factors studied and the related hypotheses need further investigation.

It appears that the diversity of the organisms present is independant of the characteristics studied (surface area, location within the habitat and type of habitat). This may be related to some adaptive factors that affect the species present and results in the dominance over others of the species observed. The total surface area of the habitat does not seem to affect the number of organisms present in that habitat. Another result that was found is that the total number of organisms in a given area within the samples show fluctuations in these samples. These particular conclusions cannot be stated with certainty because of the limitations of the experimental procedure which may have resulted in the observations seen.

Although the limitations on this study were taken in account one cannot make generalizations based on this study. Further detailed investigation for H. glandiforme and the related species is needed to acheive a greater degree of certainty in stating conclusions.

SUGGESTIONS FOR FURTHER STUDY:-

H. glandiforme creates a good species for ecological studies. Some observations were noticed while doing this study which make good topics for further studies about H. glandiforme and associated species. Below are some suggestions for further study:-

- Investigation for the microorganisms found in the water inside the water sac.
- Taxonomic study for the different types of species present in the water and on the walls of the water sac.
- Study about the ecological links between H. glandiforme and the associated species.
- Studies about species associated with H. glandiforme from samples taken from different locations and at different times of the year.

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Appendices

- Appendix I :-
  Samples of the data

- Appendix II :
  The probability values for χ^2 test

R.R

Race Rocks samples collected on Wesday Oct. 23rd, 1990

Sample	Length cm	C diameter cm	Surface area cm³ top	middle	bottom	total	Notes
1	13.5	1.90	17.5	30	12	59.5	Open
.2	10.3	1.20	10.5	18	7.1	35.6	s
3	20.1	3.20	46	78	28	152	s
4	15.0	2.50	24	42	14.6	80.6	s
5	15.0	2.30	24	42	13.3	79.3	s
6	16.0	2.60	21	54	17	102	s
7	14.0	2.10	22	39	12	73	s
8	14.5	2.20	23.4	43	11.6	78	s
9	13.5	2.0	19.2	33	11.8	64	s
10	13.0	1.90	16.9	29	10.7	56.5	s
11	21.0	3.60	50.4	89	28.6	168	s
12	13.0	2.0	17.9	30	11.1	59.0	s
13	14.0	2.30	22.6	41	11.4	75	s
14	17.0	2.70	33.9	57	22.1	113	s
15	18.5	2.70	38.7	62	28.3	129	s
16	16.5	2.50	33	55	20	108	s
.17	12.5	1.40	13	20	10	43	s
.18	9.5	1.10	9	16.5	3.5	29	s
19	13.8	2.10	19.6	34	11.4	65	s
20	14.1	2.30	22	38	13	73	s
21	16.3	2.70	31.5	54	19.5	105	s
.22	12.8	1.10	11.4	20	6.6	38	s
23	13.3	1.80	16.8	27.5	11.7	56	s
24	19.0	3.0	45.6	73	33.4	152	Closed
25	18.8	2.90	42	70	28	140	s

Sample	length cm	diameter cm	surface area cm³ top	middle	bottom	total	notes
26	19.1	3.10	47	73	35	155	closed
27	22.0	3.50	57	96	36	189	s
28	23.0	3.50	60.9	102	40.1	203	s
.29	10.8	1.20	9	14	6	29	s
.30	9.6	1.10	6.9	10.6	5.5	23	s
.31	12.8	1.60	13.8	22.3	9.9	46	s
.32	12.5	1.60	13	21	9	43	s
.33	12.0	1.50	11.7	20	7.3	39	s
34	16.0	2.50	31	52	19	102	s
35	18.0	2.80	38.1	63	25.9	127	s
36	23.0	3.60	62.1	105	39.9	207	s
37	19.6	3.10	48.6	85	28.4	162	s
38	22.3	3.30	54.9	96	32.1	183	s
39	21.9	3.20	50.7	90	28.3	169	s
.40	7.9	1.00	6.3	10	4.7	21	s
41	21.9	3.40	51.6	87	33.4	172	s
.42	8.5	1.00	6.9	12	4.1	23	s
.43	9.6	1.10	8.4	14	6.6	28	s
.44	12.9	1.60	15	25	8	48	s
45	15.0	2.50	25	43	13	81	s

species present	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
	533	454	716	356	406	119	156	243	111	48	617	413	1327	299	185
	264	242	337	224	258	244	175	216	31	182	235	168	226	246	200
	10	7	8	7	9	6	28	15	11	6	19	23	29	34	30
	9	9	17	15	28	10	5	13	17	11	21	31	23	26	24
	26%	28%	30%	16%	23%	29%	16%	22%	31%	25%	21%	25%	27%	23%	27%
	2	1	3	9	10	12	11	12	5	9	18	6	12	15	16
	19	16	27	25	23	18	28	35	22	16	31	36	17	21	19
	22%	25%	17%	28%	24%	29%	28%	20%	33%	33%	17%	21%	19%	19%	20%
	31	15	—	—	10	15	14	10	10	12	19	11	14	13	17
	2	4	6	2	2	6	2	3	—	1	3	4	4	6	6
	6	12	—	3	4	4	9	4	7	9	6	8	12	7	13
	11	7	—	7	4	6	15	12	19	7	12	11	10	10	5
	1020	636	1830	800	680	452	741	1551	885	368	659	775	1010	569	1071
	13	13	6	25	—	18	10	9	16	8	20	22	10	12	13
	6	6	2	—	6	13	12	7	9	3	8	7	13	7	10
	33	41	79	48	34	98	42	21	16	27	49	71	30	56	68
	21	—	15	12	—	15	—	16	7	7	9	8	15	10	10
	237	310	383	242	—	140	197	162	50	145	102	184	225	195	108
	563	—	200	599	—	275	411	533	197	878	752	371	562	847	385
	40	38	61	—	—	—	—	40	30	35	61	59	48	39	34
	212	—	136	—	—	—	—	139	376	173	199	181	143	285	13
	6	—	17	4	—	—	11	9	11	12	10	9	11	10	10
	310	—	387	255	305	—	211	—	161	212	272	205	169	258	208
	—	—	10	5	6	7	13	9	20	12	18	17	3	13	24
	10	—	11	11	1	16	—	—	16	—	19	19	12	13	12
	1	—	4	2	13	8	15	16	16	17	6	4	5	13	5
	60	—	12	13	9	19	19	12	25	31	17	24	27	24	26
	30%	30%	40%	18%	23%	35%	37%	18%	26%	28%	35%	45%	60%	60%	25%

species present	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
29	110	133	116	119	141	94	158	51	66	79	115	246	73	47	100
30	70	75	65	82	59	92	56	67	76	41	152	69	92	80	93
31	18	—	11	3	12	18	17	17	19	24	7	19	23	13	18
32	411	790	437	405	638	240	320	393	330	270	283	395	365	200	332
33	—	—	—	9	3	8	8	3	3	2	2	2	3	7	9
34	—	3	2	1	5	7	2	8	9	4	3	6	—	5	1
	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30
28	32%	27%	34%	37%	44%	35%	70%	75%	55%	30%	50%	70%	55%	45%	45%
29	180	70	80	91	58	211	205	150	175	165	163	153	167	243	215
30	47	64	86	98	114	165	112	83	150	134	119	119	98	91	129
31	12	24	11	17	11	26	23	27	16	9	17	14	25	15	32
32	315	346	155	288	173	290	380	140	210	310	380	290	500	530	510
33	5	3	2	2	3	6	7	4	4	11	6	2	3	3	3
34	4	2	2	3	5	1	4	7	1	2	2	3	4	3	2
	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45
28	80%	45%	30%	85%	45%	40%	55%	45%	25%	20%	75%	60%	65%	40%	55%
29	175	200	268	160	270	162	156	150	64	100	110	165	135	135	170
30	71	90	49	77	117	121	38	58	84	126	111	56	71	85	158
31	23	17	21	25	25	20	19	17	23	21	23	10	13	9	17
32	400	530	500	410	600	390	300	150	260	570	470	410	270	350	160
33	4	6	3	4	3	5	4	3	13	3	4	3	2	2	3
34	2	2	2	2	1	6	2	3	4	2	6	1	2	2	3

	species present	Sample 1			Sample 2			Sample 3			Sample 4			Sample 5		
		top	middle	bottom	top	middle	bottom	top	middle	bottom	top	middle	bottom	top	middle	bott
1		200	73	260	162	112	180	273	183	260	121	95	140	132	94	180
2		96	83	85	86	91	65	112	118	107	69	83	72	93	84	81
3		—	10	—	1	6	—	—	8	—	2	5	—	—	9	—
4		6	2	1	7	2	—	12	2	3	8	4	3	12	10	6
5		10%	8%	8%	15%	5%	8%	16%	5%	9%	5%	5%	6%	10%	5%	8%
6		—	—	2	—	—	1	—	—	3	—	6	3	2	—	8
7		16	—	3	12	—	4	19	8	—	23	2	—	18	5	—
8		5%	10%	7%	8%	12%	5%	6%	5%	6%	12%	8%	8%	6%	10%	8%
9		10	8	13	9	3	3	—	—	—	—	—	—	1	6	3
10		—	2	—	1	1	2	3	1	2	—	—	2	1	—	1
11		—	6	—	3	8	1	—	—	—	2	—	1	2	1	1
12		2	8	1	1	4	2	—	—	—	3	3	1	1	2	1
13		600	120	300	300	200	136	725	612	493	—	600	200	300	200	180
14		—	11	2	2	6	5	—	4	2	12	10	3	—	—	—
15		6	—	—	3	1	2	2	—	—	—	—	—	2	1	3
16		20	13	—	23	6	12	45	18	16	20	16	12	16	12	6
17		6	12	3	—	—	—	12	—	3	3	8	1	—	—	—
18		87	90	60	130	107	73	200	121	62	93	62	87	—	—	—
19		360	105	98	—	—	—	—	200	—	312	195	87	—	—	—
20		23	17	—	16	19	3	20	18	23	—	—	—	—	—	—
21		108	81	23	—	—	—	120	—	16	—	—	—	—	—	—
22		6	—	—	—	—	—	3	6	8	2	1	1	—	—	—
23		123	89	98	—	—	—	263	—	124	162	—	93	137	82	96
24		—	—	—	—	—	—	—	8	2	—	3	2	3	2	1
25		6	3	1	—	—	—	10	—	1	8	1	2	—	—	1
26		—	1	—	—	—	—	1	1	2	1	—	1	2	8	3
27		12	31	17	—	—	—	6	—	6	1	12	—	—	6	3
28		15%	5%	10%	20%	5%	5%	21%	6%	13%	10%	—	8%	16%	7%	—

	Sample 1			Sample 2			Sample 3			Sample 4			Sample 5		
species present	top	middle	bottom	top	middle	bottom	top	middle	bottom	top	middle	bottom	top	middle	bottom
29	60	36	14	80	28	25	65	30	21	43	46	30	[illegible]	63	—
30	34	16	20	31	—	24	—	—	65	—	28	54	—	59	—
31	12	6	—	—	—	—	8	2	1	—	—	3	—	—	12
32	200	80	131	600	93	97	185	170	82	192	83	130	252	206	180
33	—	—	—	—	—	—	—	—	—	—	[illegible]		—	1	2
34	—	—	—	—	—	3	—	2	—	—	—		—	2	
35															

species present	16	17	18	19	20	21	22	23	24	25	26	27	28	29	3[illegible]
1	482	652	1065	533	474	542	557	212	258	616	447	275	519	446	44[illegible]
2	209	370	187	364	252	466	247	291	268	199	272	322	263	185	273
3	13	12	21	21	22	11	12	9	12	14	6	5	8	7	7
4	11	16	11	11	6	13	9	6	5	4	11	7	10	14	16
5	27%	20%	12%	16%	18%	20%	13%	21%	23%	24%	30%	26%	14%	8%	32%
6	5	10	11	11	11	3	2	3	6	8	14	5	3	4	6
7	24	20	19	19	20	23	17	20	18	14	20	18	3	26	19
8	22%	16%	10%	13%	18%	13%	21%	16%	15%	21%	19%	34%	32%	36%	34%
9	17	20	14	14	11	23	16	16	17	15	19	26	27	23	7
10	2	3	6	2	4	4	3	4	4	6	3	4	2	3	4
11	11	7	11	8	8	11	11	12	9	11	10	9	7	13	7
12	22	12	11	9	17	18	18	24	31	22	17	13	13	18	21
13	608	334	400	299	106	437	509	488	445	492	307	436	230	587	303
14	13	16	12	12	15	14	9	13	9	20	10	15	16	7	18
15	9	10	7	10	13	8	10	13	19	11	12	15	13	12	16
16	45	12	25	47	70	26	36	45	44	58	40	15	34	38	42
17	9	9	14	12	14	11	15	9	10	7	13	16	24	12	21
18	278	179	302	352	25	396	267	122	195	313	389	480	381	441	499
19	760	442	399	400	344	781	885	576	428	405	693	709	560	677	863
20	42	40	39	32	34	69	25	64	64	34	15	40	23	27	24
21	206	213	183	210	182	237	264	112	226	214	197	269	167	231	167
22	5	8	12	6	7	14	7	9	8	10	12	24	26	14	12
23	207	294	245	192	219	129	204	266	260	248	187	216	231	211	294
24	15	10	16	—	—	25	18	26	24	7	11	23	15	22	8
25	20	23	13	24	8	18	11	10	22	20	13	14	18	22	9
26	9	8	10	12	9	15	13	13	6	7	15	3	10	3	8
27	15	10	8	8	6	[illegible]	6	2	7	8	7	5	2	6	8

	species present	top	middle	bottom	top	middle	bottom	top	middle	bottom	top	middle	bottom	top	middle	b
28		30%	25%	20%	20%	20%	20%	20%	75%	20%	15%	5%	10%	15%	20%	20
29		20	90	—	30	100	35	40	—	95	55	80	—	83	67	2
30		26	45	40	—	30	26	—	28	43	45	—	40	40	61	5
31		8	5	10	3	—	7	4	9	—	1	8	—	3	8	6
32		150	150	170	210	—	200	180	90	—	100	180	70	80	—	8
33		1	3	—	2	1	—	—	1	1	—	—	2	1	1	1
34		3	3	—	1	—	—	1	—	1	1	—	1	—	2	1

Species present	Sample 41 top	Sample 41 middle	Sample 41 bottom	Sample 42 top	Sample 42 middle	Sample 42 bottom	Sample 43 top	Sample 43 middle	Sample 43 bottom	Sample 44 top	Sample 44 middle	Sample 44 bottom	Sample 45 top	Sample 45 middle	Sample 45 bot
	160	100	150	170	200	180	150	300	400	200	360	100	380	400	20
	130	140	160	250	120	150	260	140	100	250	200	130	150	300	10
	6	10	12	11	7	12	10	8	5	4	3	1	—	7	6
	3	7	—	12	3	7	7	7	3	—	3	4	—	6	—
	5%	15%	15%	10%	20%	—	5%	8%	10%	20%	15%	15%	15%	10%	10%
	3	1	1	3	3	1	3	1	2	—	4	3	3	2	3
	4	4	—	6	4	—	3	7	10	11	—	12	6	11	—
	8%	7%	10%	8%	8%	7%	5%	5%	10%	12%	20%	15%	10%	10%	10%
	2	3	—	12	—	15	17	12	10	12	—	—	7	12	—
	3	3	1	—	2	1	2	1	1	—	1	2	1	—	1
	1	2	4	6	3	6	—	4	5	—	3	5	4	4	2
	7	12	3	16	6	8	—	7	1	9	—	3	4	5	6
	200	210	150	250	160	280	150	250	140	230	140	—	140	100	80
	10	8	5	7	11	5	9	11	—	4	10	7	6	9	5
	3	10	3	7	—	4	9	3	9	4	7	—	4	9	8
	18	20	17	9	26	9	16	21	15	7	23	13	12	17	[illegible]
	8	5	9	7	4	[illegible]	[illegible]	7	5	—	3	3	7	[illegible]	4
	200	280	300	210	350	380	—	130	400	360	260	300	—	130	30
	—	400	—	200	600	300	150	680	100	450	650	350	300	260	100
	17	28	32	26	25	20	25	—	21	—	13	24	—	13	17
	250	150	100	100	50	70	120	100	80	130	—	150	100	120	10
	1	5	4	3	5	5	3	4	2	6	3	4	7	3	2
	210	150	210	60	100	200	80		150	60	—	120	70	180	40
	12	4	6	10	5	5	10	4	10	6	5	11	—	10	10
	12	12	11	12	—	13	—	—	10	12	14	7	9	13	12
	1	3	—	[illegible]	1	—	7	2	2	1	3	—	1	4	1
	7	2	4	2	4	1	6	7	1	5	3	5	2	1	2

species present	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45
	645	526	510	470	573	537	312	275	443	228	410	550	850	660	980
	250	250	432	360	190	308	246	316	290	210	450	520	500	580	530
	17	10	4	9	9	15	17	13	16	20	28	30	23	8	13
	20	14	17	9	9	17	7	14	12	11	10	22	17	7	6
	33%	27%	40%	25%	37%	47%	20%	51%	15%	30%	35%	30%	23%	50%	35%
	16	21	20	19	8	6	6	6	6	5	5	7	6	7	8
	12	22	21	26	17	24	17	13	18	27	8	10	20	23	17
	30%	32%	10%	33%	30%	20%	15%	25%	22%	25%	25%	23%	20%	47%	30%
	17	20	19	14	18	22	17	23	22	24	5	27	39	12	19
	4	6	8	8	8	4	4	6	6	5	7	3	4	3	2
	18	17	23	22	31	7	7	9	8	10	7	15	9	10	15
	20	12	16	15	17	15	16	15	18	32	22	30	10	12	15
	270	338	220	300	280	478	449	270	266	212	560	690	540	370	320
	18	19	6	13	10	27	19	20	16	27	23	23	20	21	20
	16	10	12	14	6	10	5	13	17	28	16	11	21	13	21
	23	25	25	35	25	37	57	62	48	49	55	44	52	43	39
	12	16	10	31	14	16	15	6	10	16	22	21	17	6	23
	380	180	530	200	380	555	512	458	243	510	780	940	550	940	430
	800	1150	750	1400	1250	1100	1150	1150	1000	1570	400	1100	930	1450	760
	42	43	52	36	48	87	53	57	39	101	77	71	46	37	30
	250	210	250	320	320	400	165	324	290	190	500	220	300	280	320
	8	7	8	6	8	9	14	10	6	9	10	13	9	13	12
	215	119	100	110	120	300	225	180	150	116	570	380	200	180	290
	15	12	19	17	17	27	11	21	29	17	22	20	24	22	20
	10	9	11	10	12	23	18	25	23	12	35	25	10	33	34
	13	16	7	10	9	6	14	5	4	9	4	2	6	4	6
	2	5	3	8	7	6	5	4	6	12	9	7	10	13	6

Appendix II

TABLE R. Critical values of the chi-square distribution.

ν \ α	0.995	0.975	0.9	0.5	0.1	0.05	0.025	0.01	0.005	ν
1	.000	.000	0.016	0.455	2.706	3.841	5.024	6.635	7.879	1
2	0.010	0.051	0.211	1.386	4.605	5.991	7.378	9.210	10.597	2
3	0.072	0.216	0.584	2.366	6.251	7.815	9.348	11.345	12.838	3
4	0.207	0.484	1.064	3.357	7.779	9.488	11.143	13.277	14.860	4
5	0.412	0.831	1.610	4.351	9.236	11.070	12.832	15.086	16.750	5
6	0.676	1.237	2.204	5.348	10.645	12.592	14.449	16.812	18.548	6
7	0.989	1.690	2.833	6.346	12.017	14.067	16.013	18.475	20.278	7
8	1.344	2.180	3.490	7.344	13.362	15.507	17.535	20.090	21.955	8
9	1.735	2.700	4.168	8.343	14.684	16.919	19.023	21.666	23.589	9
10	2.156	3.247	4.865	9.342	15.987	18.307	20.483	23.209	25.188	10
11	2.603	3.816	5.578	10.341	17.275	19.675	21.920	24.725	26.757	11
12	3.074	4.404	6.304	11.340	18.549	21.026	23.337	26.217	28.300	12
13	3.565	5.009	7.042	12.340	19.812	22.362	24.736	27.688	29.819	13
14	4.075	5.629	7.790	13.339	21.064	23.685	26.119	29.141	31.319	14
15	4.601	6.262	8.547	14.339	22.307	24.996	27.488	30.578	32.801	15
16	5.142	6.908	9.312	15.338	23.542	26.296	28.845	32.000	34.267	16
17	5.697	7.564	10.085	16.338	24.769	27.587	30.191	33.409	35.718	17
18	6.265	8.231	10.865	17.338	25.989	28.869	31.526	34.805	37.156	18
19	6.844	8.907	11.651	18.338	27.204	30.144	32.852	36.191	38.582	19
20	7.434	9.591	12.443	19.337	28.412	31.410	34.170	37.566	39.997	20
21	8.034	10.283	13.240	20.337	29.615	32.670	35.479	38.932	41.401	21
22	8.643	10.982	14.042	21.337	30.813	33.924	36.781	40.289	42.796	22
23	9.260	11.688	14.848	22.337	32.007	35.172	38.076	41.638	44.181	23
24	9.886	12.401	15.659	23.337	33.196	36.415	39.364	42.980	45.558	24
25	10.520	13.120	16.473	24.337	34.382	37.652	40.646	44.314	46.928	25
26	11.160	13.844	17.292	25.336	35.563	38.885	41.923	45.642	48.290	26
27	11.808	14.573	18.114	26.336	36.741	40.113	43.194	46.963	49.645	27
28	12.461	15.308	18.939	27.336	37.916	41.337	44.461	48.278	50.993	28
29	13.121	16.047	19.768	28.336	39.088	42.557	45.722	49.588	52.336	29
30	13.787	16.791	20.599	29.336	40.256	43.773	46.979	50.892	53.672	30
31	14.458	17.539	21.434	30.336	41.422	44.985	48.232	52.191	55.003	31
32	15.134	18.291	22.271	31.336	42.585	46.194	49.480	53.486	56.329	32
33	15.815	19.047	23.110	32.336	43.745	47.400	50.725	54.776	57.649	33
34	16.501	19.806	23.952	33.336	44.903	48.602	51.966	56.061	58.964	34
35	17.192	20.569	24.797	34.336	46.059	49.802	53.203	57.342	60.275	35
36	17.887	21.336	25.643	35.336	47.212	50.998	54.437	58.619	61.582	36
37	18.586	22.106	26.492	36.335	48.363	52.192	55.668	59.892	62.884	37
38	19.289	22.878	27.343	37.335	49.513	53.384	56.896	61.162	64.182	38
39	19.996	23.654	28.196	38.335	50.660	54.572	58.120	62.428	65.476	39
40	20.707	24.433	29.051	39.335	51.805	55.758	59.342	63.691	66.766	40
41	21.421	25.215	29.907	40.335	52.949	56.942	60.561	64.950	68.053	41
42	22.138	25.999	30.765	41.335	54.090	58.124	61.777	66.206	69.336	42
43	22.859	26.785	31.625	42.335	55.230	59.304	62.990	67.459	70.616	43
44	23.584	27.575	32.487	43.335	56.369	60.481	64.202	68.710	71.893	44
45	24.311	28.366	33.350	44.335	57.505	61.656	65.410	69.957	73.166	45
46	25.042	29.160	34.215	45.335	58.641	62.830	66.617	71.201	74.437	46
47	25.775	29.956	35.081	46.335	59.774	64.001	67.821	72.443	75.704	47
48	26.511	30.755	35.949	47.335	60.907	65.171	69.023	73.683	76.969	48
49	27.249	31.555	36.818	48.335	62.038	66.339	70.222	74.919	78.231	49
50	27.991	32.357	37.689	49.335	63.167	67.505	71.420	76.154	79.490	50