

THE INFLUENCE OF LOW TEMPERATURES ON THE ZONAL
DISTRIBUTION OF ALPHOPODA DURING SUMMER AND WINTER.

By

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DEPT. OF ZOOLOGY.

MCGILL UNIVERSITY.

(BEING A STUDY FROM THE STATIONS OF THE BIOLOGICAL
BOARD OF CANADA.)

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INTRODUCTION

Of the physical factors which determine the activity, abundance, and distribution of plant and animal life, that of the environmental temperature is one of the most important. The temperatures to which ^aplants ~~or~~ animals ~~is~~ subjected in nature ~~are~~ ^{is} not of a very wide range, and yet many forms of life cannot withstand the extremes of even that range. The distribution on our globe of many forms of life is greatly determined by their ability to withstand low or high temperatures, and this is true of vertical as well as of horizontal distribution, especially in the case of fresh and salt water life.

During the summer of 1929 the author attempted to show the influence of low temperatures on the vertical distribution of various species of Amphipoda. The work was done at the Atlantic Biological Station, St. Andrews, N. B., under the direction of Prof. A. G. Huntsman.

In the study of the distribution of animals as regulated by temperature, more ^{experimental} work has been done with insects than with other animals. This is because of the economic aspect of the matter, and probably also because insects are more numerous and therefore more easily obtained than other kinds of animals.

Although Newport (1837), Bachmetjew (1901) and Pirsch (1923) found that insects do have a slow control of their body temperatures under different environmental conditions, yet poikilothermic animals, as a rule, cannot ordinarily withstand ~~extreme~~ temperatures. The ability of a species to withstand low temperatures will necessarily constitute a limitation on its distribution. A species may not be lethally affected by the summer temperature range of a locality, but the winter temperature range of that locality may be low enough to kill the species unless it migrates to warmer climates or is specially adapted to low winter temperatures. In many non-migratory insects this adaptation lies in hibernation. (The explanations advanced for this phenomenon are given elsewhere in this paper). Kirby and Spence (1818) noted that insects when hibernating will resist extreme low temperatures, although these temperatures are fatal when the insects are in the active (summer) state. Gueyrlard and Portier (1916) found that the larvae of some insects can survive -20 C. in winter but not in summer. This periodicity in the lethal point of certain bugs and insects has also been reported by Bachmetjew (1901), Duval and Portier (1921), Knight (1922), Ludwig (1928) and Payne (1926 and 1927d).

Animals normally exposed to a temperature range which does not vary much develop cold hardiness in proportion to the lowest temperatures of that range. Semper (1881) calls the inactivity of animals when at low temperatures "chill-coma" and notes that the critical temperature for chill-coma is widely different in various animals. Those animals which live in colder climates have lower critical temperatures for chill-coma than those of warmer climates. Mayer (1914) working on *Aurelia aurita* found that those specimens taken from Halifax, N. S., cease movements at -1.0 C., whilst those taken from Portugas, Florida, become inactive at 7.75 to 11.8 C. Payne (1926), working on oak borers which normally are subjected to temperature extremes, and on aquatic insects which are never exposed to temperatures lower than 0 C., found a correlation between environmental temperature and the lethal temperature--the oak borers withstand lower temperatures than do the aquatic insects. Brown (1929) investigated the ability of various species of Cladocerans to withstand sudden immersion in water at or near 0 C., and found that the southern species became inactive in one to four minutes, whilst northern species remain active indefinitely. Cameron (1914), working on the effects of low temperatures on the frog, found that the species which lives in western Manitoba (*Rana pipiens*) has an actual death temperature of only a few tenths of a degree lower than two other species which occur distinctly further south (*R. clamitans* and *R. sphenoccephala*).

~~RESULTS~~, SUMMER EXPERIMENTS.

In the experiments on the relationship of low temperatures to the distribution of Amphipoda the following species were used:- Orchestia agilis Smith, Gammarus locusta (Lin.), Corophium volutator (Pall.), Unciola irrorata Say, Ampelisca macrocephala Lilljeborg, and Orchomenella pinguis (Boeck).

It has been shown that marine invertebrates cannot maintain any difference between the osmotic pressure of their body fluids and that of the water medium. When the osmotic pressure of the surrounding/medium is varied by either concentrating or diluting the sea-water, the body fluid will adapt itself by undergoing a similar change in concentration. This will, of course, affect the freezing point of the animal, as is cited by Philip (1925) in the case of the crab, Maia verrucosa.

<u>Medium</u>	<u>Sea-water</u>	<u>Body fluid</u>
Normal sea-water	-2.3 C.	-2.3 C.
Concent. "	-2.98	-2.9
Dilute "	-1.38	-1.4

Consequently, since the simplest and cheapest way of cooling an object is by immersing it in an ice-salt mixture, precautions must be taken that the salt concentration of the freezing mixture does not alter the salt concentration of any tissues immersed in it. The following method was thought most expedient, and was used in these experiments:-

The animals were collected in small jars and brought into the laboratory. Most of the experiments were performed right away. Those animals not immediately used were kept in small groups in jars at the temperature of sea water. The animals were dried by carefully pressing them in filter paper and were then pressed individually in small waxed paper bags measuring 2 cms. X 5 cms. The waxed paper was very thin and did not interfere to any appreciable extent with the conduction of heat. The bags were made water-tight by means of small spring clips and were immersed in ice-water mixtures of different temperatures. The ice-water solution pressed the waxed paper evenly on all sides against the animal, so that all parts of the animal were simultaneously cooled.

The animals were subjected to different temperatures for different lengths of time. After being ^{taken} ~~extracted~~ from the bags they were placed in small petri dishes containing sea water at a low temperature and were examined under the microscope at intervals. The failure of the heart to resume pulsation was taken as the final criterion of death. The temperature at which 50% died when subjected for a given time was taken as the lethal temperature for that time. The same animals were never used twice for these experiments. Temperature was read by means of small mercury thermometers which were calibrated nearly every fortnight by Prof. H. B. Hochey.

Not all specimens of even the same species recovered at the same rate. Recovery took place in progressive stages. Generally the breathing processes were resumed first, to be followed by pulsations of the heart and movements of the thoracic legs. The heart beat usually was at first irregular. In a number of cases the heart beats would die out after a few feeble attempts, though irregular twitchings of the legs and occasionally of the antennae would continue for a few hours. It is interesting to note that Brown (1929a) observed that the Cladocerans which he used in his experiments "become inactive by progressive stages, that is, the bodily activities disappear slowly so that the end point is not sharply defined, but when they recover from 'chill-coma' they do so 'all at once'".

The results obtained for more than two hundred experiments involving nearly one thousand five hundred animals tend to show a marked relationship between the normal living temperature and the temperature at which death occurs upon cooling. The lethal temperature of a species corresponds to its habitat depth—a species which lives in the intertidal zone where the temperature varies to a great extent can withstand low temperatures better than a species which lives in shallow water where the temperature fluctuations are only moderate.

Species	Summer Habitat	Relative Habitat Temperature.	Lethal Temperature.
<u>Orchestia</u> <u>agilis</u> Smith.	Above high tide	Coldest.	-7.0 °C.
<u>Corophium</u> <u>volutator</u> (Pall).	Mud flats exposed at low tide.	Cold.	-5.8 °C.
<u>Gammarus</u> <u>locusta</u> (Lin.)	Seaweeds exposed at low tide.	Cold.	-5.2 °C.
<u>Unciola</u> <u>irrorata</u> Say.	Shallow water.	Medium.	-3.8 °C.
<u>Ampelisca</u> <u>macrocephala</u> Lilljeborg.	Shallow water.	Medium.	-3.5 °C.
<u>Orchomenella</u> <u>pingvis</u> (Boeckl).	Deep water.	Warmer.	-3.0 °C.

It should be observed that the lethal temperature and the freezing temperature need not necessarily be one and the same. The lethal temperature of an animal may be higher than the freezing point of the body, as in the jellyfish from the vicinity of Florida which dies from cold at 7.75°C. to 11.8°C. (Mayer 1914). Or, as is shown by these experiments, the lethal temperature may be below the freezing point. The body-fluids of Orchestia agilis probably freezes around -2.5°C. (as does sea-water isotonic with the body-fluids), yet the animal can recover from lengthy exposure to -6.5°C.

From Newton's law of cooling, that the rate of cooling of a body is proportional to the temperature difference between the body and its surroundings, it is evident that an animal taken from 5°C. and exposed to -15°C. will reach its lethal temperature sooner than it would if exposed to -12°C. ~~In other words, an animal may survive exposure to extreme low temperatures for a short period of time but not for a long period of time.~~ This is shown by the accompanying graph (Fig. 1)

And as the temperature of an animal will depend on the length of time it has been exposed to a low temperature,

The average size of Gammarus locusta is 27mm. and the average size of Uniola irrorata is 15 mm. G. locusta can withstand lower temperatures than can U. irrorata. To show that this ability is not a function of size but of the environmental temperature of each species, a number of small G. locusta (9mm.) were submitted to the same process. The curve obtained does not vary much from that for the average sized G. locusta.

Fig. #1. Here

Huntsman and Sparks (1924) found that a relationship existed between the normal habitat temperature of amphipods and the temperature at which death occurred on heating. They found that all individuals of the same species ceased breathing movements at the same temperature but that each species had its own lethal high temperature. Similar results were obtained ~~to hold~~ for lamellibranchs by Henderson (1926). The chart (Fig. 2) combines the results of Huntsman and Sparks with ~~manel~~ ~~present~~ and shows graphically the temperature range which different species of amphipods can withstand and ^rcorrelates this with the relative habitat depth and habitat temperature of each species.

Fig # 2 Here.

~~RESULTS~~ WINTER EXPERIMENTS.

It has been shown that the amphipoda of the vicinity of St. Andrews, N. B., can endure temperatures which are only a few degrees below zero. Since the air temperature during the winter is often as low as -25 C., and the tidal pools entirely frozen up, it was thought interesting to see the relationship between the amphipoda and these low winter temperatures. Through the kindness of the Director, Prof. A. G. Huntsman, the author was enabled, in February 1930, to revisit the Atlantic Biological Station at St. Andrews and to investigate the situation.

Fig # 3. Here

Fig. 3 represents diagrammatically an area in which many G. locusta and O. agilis were found during the summer. This same plot was observed for nearly two weeks during February. The region A was throughout this time covered with ice. The following observations were made:-

February 19th. Average air temperature 1.4° C., minimum -1.1. Average water temperature -0.58. Region A covered with ice and soil well frozen. No amphipoda of any kind under seaweed or in small pools in intertidal region B. A few Orchestia in larger pools. Many Orchestia in shallow water C together with a few Gammarus. Many

Gammarus and a few Orchestia (small ones) in deeper water D.

February 20th. Average air temperature 1.1°C ., minimum -1.1 . Average water temperature -0.11 . No amphipoda in B. Many Orchestia and few Gammarus in C. Many Gammarus and few Orchestia in D.

February 21st. Average air temperature 3.05°C ., minimum -5.5 . Average water temperature -0.16 . Three Orchestia found in B; these never recovered. No Gammarus. Many Orchestia and few Gammarus in C. Many Gammarus and few Orchestia in D.

February 22nd. Average air temperature 0.05°C ., minimum -5.0 . Average water temperature 0.25 . No amphipoda in B. Many Orchestia and few Gammarus in C. Many Gammarus and few Orchestia in D.

February 24th. Average air temperature 2.95°C ., minimum -0.94 . Average water temperature -0.72 . A few Orchestia and two or three Gammarus in B. Many Orchestia and a number of Gammarus in C. Many Gammarus and very few Orchestia in D.

February 26th. Average air temperature -6.2°C ., minimum -9.1 . Average water temperature -0.51 . No amphipoda in B. Very few Orchestia and no Gammarus in C. Many Orchestia and very many Gammarus in D.

February 27th. Average air temperature -4.1°C ., minimum -9.7 . Average water temperature -0.5 . Conditions same as on previous day.

February 28th. Temperature of air in morning was -8.0°C . Temperature of water at that time -0.53 . No amphipoda in B. A few Orchestia and no Gammarus in C. Many Orchestia and many Gammarus in D.

From the observations recorded above, it would seem that *Orchestia* goes into water on the approach of cold, and that *Gammarus* leaves the intertidal pools for deeper waters where the temperature is higher and less variable. When the air becomes warmer than water--about 3.5 --the *Orchestia* and *Gammarus* leave the water for the warmer pools. It is interesting to note that Tait (1925) observed that the European/species of *Orchestia* (~~is~~ *O. littorea*) seem to be very unhappy in water and cannot bear submersion very well. What they do on the approach of cold is puzzling.

The conditions under which *Corophium volutator* exist was also studied. During summer they inhabit the mud flats which are left exposed at low tide. It was observed that during winter a great part of these flats were covered with ice cakes when the tide was low and that during cold days the mud gave higher temperature readings than the little tide pools which froze up. On extremely cold days most of the *Corophium* were found in shallow water, though a good number were also found under the ice of the larger pools and a few buried in the mud. When the small pools did not freeze *Corophium* were often found in them. It might be said that, on the average, those *Corophium* which were found in small tide pools were larger than those found in the mud, and that these in turn were larger than those found in the bigger pools or below water level.

Experiment No. 1.

A bucket containing 15 *G. locusta* in sea-water at 0°C. was placed outdoors for 5 hours. The average temperature of the air was -7°C. and for the water was -2.5°C. A coat of ice 5 cms. thick formed on the top. All 15 animals survived.

Experiment No. 2.

A bucket containing 27 *G. locusta* was left outdoors for nearly 22½ hours. The initial temperature of the water in the bucket was 0.17°C. and the final temperature was -2.7°C. The average temperature of the air for the period was -4.1°C., with -9°C. as the minimum. An ice coat of 9 cms. thickness was formed on the surface. Six *Gammarus* were found encased in this ice; when gradually thawed out all lived. The other animals were inactive on the bottom of the bucket, and three of them were found to be dead on thawing up to 4.0°C. Percentage dead = 10.

Experiment Nos. 3 to 12.

These ten experiments involved the determination of the lethal temperature ^{of} 36 *G. locusta*, 14 *C. volutator*, and 28 *O. agilis* by the methods already outlined. The results may be compared with those for the summer:-

Species	Lethal Temp.		Difference
	Summer	Winter	
<i>O. agilis</i>	-7.0	-6.7	0.3
<i>C. volutator</i>	-5.8	-6.3	0.5
<i>G. locusta</i>	-5.2	-5.9	0.7

It will be seen that there is a different lethal temperature in winter than in summer. Two possible explanations for this suggest themselves:- In the winter experiments the initial temperature of the animals before subjection to cold was lower than the initial temperature of these animals used in summer. This precooling would increase the resistance of the animal to cold. This would explain the increase in the resistance (lowering of lethal point) of *Gammarus* and *Corophium* but not the decreased resistance of *Orchestia*. A second explanation seems more likely. Just as in summer the lethal temperature of a species is correlated with the environmental temperature of that species, so also in winter the lethal temperature is proportional to the environmental temperature. In summer the three species mentioned have different habitats and different environmental temperatures. But it has been seen that during winter the habitats of these species are all very much of the same nature, and that the habitat temperature is also very much similar in each case. It is, therefore, possible that the approximation towards a common winter lethal temperature is due to the somewhat common winter environment.

Experiments Nos. 13 to 21.

Since *Orchestia agilis* lives during the summer months under the masses of seaweed which grow just above high-tide mark, it was thought possible that these animals might, on the approach of winter, dig into the soil for warmth. In winter this region is usually covered with extensive ice formations.

Samples from this region, consisting of ice, the frozen seaweed underneath it and the substrata of small stones and sand were taken into the laboratory and the whole was carefully thawed out and washed. Of the nine tests made, each from a different representative area, not one showed any sign of any kind of amphipod, either living or dead. It is to be concluded, therefore, that O. agilis does not burrow into the soil to hibernate there during winter.

Experiments Nos. 22 to 27.

In consideration of the facts presented above, it was thought desirable to determine experimentally the temperatures at which this migration of ~~O. agilis~~ and G. locusta takes place. These experiments were performed at the Biological Station and at the Zoological Laboratories, McGill University.

Fig # 4 Here

An oblong galvanized-iron box (Fig. 4) having two compartments separated by a partition was used. The following experiments were made.

No. 22. Fifteen G. locusta were placed in the larger compartment (marked "cold") in sea-water at 0° C. The portion "D" was kept in the shade, whilst the rest of the box was in the light. G. locusta is negatively phototropic. The animals collected at "D" after a few minutes. Hot water was then poured into the smaller compartment (marked "hot") without disturbing the box. In this way, a temperature gradient was established in the "cold" compartment. Temperature readings were taken at "L" and at "D".

Temperature C°		Behaviour.
L	D	
0	0	All at rest in the shade.
2	0.25	Ditto.
3.25	2	2 leave D for L, one touches partition and remains, the other returns to shade D.
5	2.5	From 7 to 10 move back and forth from D to L.
9	3.25	9 are permanently within L.
10	4.25	About 4 within L. The others seem to prefer D.
14	9	All move about actively within D, a few go into L but very soon return.

No.23. The above experiment was repeated with 9 G. locusta, but this time the whole box was well illuminated. The animals were placed at D, they stayed there quietly. Then the temperature gradient was set up as before.

Temperature C°		Behaviour.
L	D	
0	0	All at rest in D.
2	1	Ditto.
3	1.5	3 in L
3.25	1.75	5 in L. All seem to hug the partition.
4.75	2.5	7 in L. All in close vicinity of partition.
5.5	3.25	All move about in L.
8	4.25	Animals move back and forth, in both L and D.
12	6	Ditto.
16	10.5	Ditto.

No.24. Experiment No.23 was repeated but this time the G. locusta were placed in L.

Temperature C°		Behaviour.
L	D.	
.25	.25	All at rest in L.
2.5	1	Most of them move around in vicinity of L.
3.5	2	Ditto.
5	3.5	All move back and forth in both L and D.
7	4.25	Ditto.
11	5.25	Ditto.

No. 25. G.locusta placed in L. Whole box kept in dark. Temperature gradient set up as before.

Temperature C°		Behaviour.
L.	D.	
0	0	All at rest in L.
2	0.5	Ditto.
3	1.5	Most of them move around in vicinity of L.
4.75	3.0	Move back and forth in both L and D.
6	3.75	Ditto.
8.75	4.25	Ditto.
11.5	6	Ditto.

No. 26. 8 G.locusta placed in D which was kept shaded whilst L was kept in the light. No temperature gradient was established as in the previous experiments, instead the whole box was gradually warmed from underneath.

Temp. C° for D. and L.	Behaviour.
-0.25	All at rest in D.
1	Ditto
2.5	Move about within limits of D.
3.5	Ditto.
4.75	Ditto.
6	Ditto.
9	Ditto.

No.27. 6 G. locusta placed in L which was kept in the light. D was shaded. Box gradually heated as in experiment No.26.

Temp. C°, for D and L.	Behaviour.
0.5	At rest in L.
1.5	Ditto.
2.75	Move about in L.
3.75	Migrated into shaded region D.
5	Move about within limits of D.
6.5	Ditto.
12	Ditto.

From the results of the above six experiments it would seem that G. locusta begins to migrate into warmer water during autumn when the water and air of the intertidal region reaches 3° to 5°C. The movement back to the intertidal region probably takes place when the water is 3° to 5°C. and the intertidal region is warmer, i.e., in spring. This seasonal migration may be represented by the following diagram; (Fig. 5):-

Fig # 5
Here

NATURE AND CAUSE OF DEATH.

The various explanations advanced on the nature and cause of death due to ^{low temperatures} ~~freezing~~ may be considered as falling into three classes:-(I) as due to formation of ice crystals with resulting mechanical injury, (II) as due to loss of water ~~due to~~ ^{through} changes in cell-membrane permeability thus concentrating the salts up to a toxic point, and (III) as due to irreversible precipitation of the protoplasmic colloids.

It has been observed that in nearly all cases death of vegetable cells due to freezing is associated with formation of ice crystals. Pfeffer (1904) and Kylin (1917) showed that death at a given temperature will occur if ice crystals are formed, but that if no crystals are formed the cells will survive exposure to that temperature. These crystals, if formed, appear either within or outside of the cell. Sachs (1864) found that the water was withdrawn from the cell and froze mostly in the intercellular spaces. McKay and Weld (1926) working on the muscle cells of hake and cod noticed the formation of ice crystals on freezing, and Weld (1927) showed that the size of these crystals depended on the rate of freezing (the quicker the freezing, the smaller the crystals), on the thickness of the muscle (this is really an expression of the rate of conduction), and on the locality from which the muscle is taken (samples from the periphery show smaller crystals than those taken from the depths of the fish). All these

workers claim that the ice crystals, whether within the cell or in the intercellular spaces, will, if large enough, rupture the cell wall. McKay and Weld (1926) and Weld (1927) showed that if the tissue is frozen very rapidly, only very small ice crystals will form and will not cause rupture. Nägeli (1861) calculated the expansion on freezing of the total water content of a (plant) cell and found that the resulting volume would be insufficient to cause rupture of the cell wall.

If it is the formation of ice crystals and the resulting rupture of the cell which causes death, then if the tissue is dehydrated sufficiently to ensure the non-formation of ice crystals, the tissue ought to be able to survive those temperatures which normally cause death. Kühne (1864) dehydrated some *Tradescantia* hairs and rapidly froze them at -14°C . After exposure of five minutes he found that protoplasmic streaming was resumed ten minutes later. It has often been shown that seeds which have been dehydrated can be subjected for various lengths of time to temperatures below -100°C . and yet not be killed. Rahm (1921) found that nematodes and rotifers which were dried were not killed by a temperature as low as -271.8°C . Payne (1927b) dehydrated the larvae of the Mediterranean meal moth with calcium chloride, and found that they can endure -15°C ., whereas normally they cannot endure 0°C . Miss Payne also showed (1927b) that the Japanese Beetle will withstand -28°C . if experimentally dehydrated to half its body weight,

whereas Ludwig (1928) showed that the normal individual cannot survive the freezing process. Most non-migratory insects of northern districts spend the winter months in hibernation at temperatures which would kill them if they were in the active state (Kirby and Spence, 1818)". Breitenbecher (1918) has shown that hibernation in the potato beetle is associated with low moisture content. Payne (1926, 1927a, 1927b) has been able to induce hibernation during summer by desiccating the insect and finds that the desiccated insect has the same lethal point that it has during winter. An objection to the theory that death by cold is due to ice crystal formation is that in many animals death will occur when the temperature has not been lowered sufficiently for ice crystals to form. The jelly-fish from Florida dies when it is cooled to only 7.75 C. (Mayer, 1914)".

Muller-Thurgau (1880) held that there is no freezing to death without the formation of ice crystals, and that death follows crystal formation. He concluded (1880 and 1886) that as ice forms, the salt concentration of the rest of the protoplasm increases until the lethal point is reached. Molisch (1897) supported this theory. Maximow (1914) holds the same theory but adds that the ice crystals produce a mechanical effect on the colloids of protoplasm. He (1912) noticed injury of the plasma membrane which was due to alterations in permeability at time of freezing. Molliard (1902) and Zacharawa (1926) describe the formation of vacuoles in both the cytoplasm and the nucleus of cells killed by cold. These vacuoles are possibly due to increased salt content,

but they are also

but they may also be due to the formation of minute ice crystals. The Muller-Thurgau theory seems to be **opposed** by the work of Payne and others (see above). It has been shown that the resistance of an animal to death by cold is increased by ^edessicating the animal. This process will increase the salt concentration in the cells to a point as great ^{as} if not greater ~~than,~~ [↑] ~~as~~ by the formation of ice crystals. And yet this concentration is not toxic. On the other hand the formation of ice crystals in a cell will lower the freezing point by the resulting increase in the salt concentration.

Gorke (1906) **believed** that death by cold is due to the irreversible precipitation of proteins. Lidforss (1907) and shortly later Schaffrit (1910) noted that cells exposed to low temperatures show an increase in sugar concentration. They held that this acts to protect the proteins from precipitation. However, the increase of sugar will also lower the freezing point. It is possible that death from cold exposure is due only to increased viscosity and subsequent coagulation. This would be the case of death of tissues at temperatures above the freezing point of the cell contents, Heilbrunn (1926) found that the viscosity of Cumingia eggs decreases with temperature until about 2 C. is reached, after which further lowering suddenly increases the viscosity. He believes this is due to a physical change of the cell lipoids. Knight (1922) repeatedly refroze and thawed the same batch of bugs and found that the freezing point is gradually raised in each subsequent freezing. This would seem to indicate that death is of a colloidal nature, since this duplicates the well-known phenomena of setting of colloids. If death is really due to a coagulation of the

protoplasmic colloids, one would expect the tissue to be rigid and stiff. The author has been unable to find any reference to such a condition in the literature consulted. In my own experiments the animals on being gradually thawed out showed, in the cases where they did not survive, no such rigidity.

SUMMARY.

- 1/ The lethal temperatures of various species of amphipoda occurring in the vicinity of St. Andrews, N.B., were determined. The results show a marked relationship between the normal habitat temperature and the temperature at which death occurs upon cooling.
- 2/ Observations during winter show that *O. agilis*, a shore form, and *G. locusta*, an intertidal form, both migrate into warmer water for the winter. *C. volutator*, another intertidal form, was also studied.
- 3/ The lethal temperatures of the above three species during winter differ from those for summer. The death temperatures for winter lie closer together; it is suggested that this is due to the somewhat common winter environment.
- 4/ *O. agilis* does not dig into the soil to hibernate during winter.
- 5/ *G. locusta* probably begins to migrate to warmer regions when the intertidal zone reaches 3 to 5 C.
- 6/ Three theories on the nature and cause of death by cold are given.

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EXPLANATIONS OF THE DIAGRAMS AND FIGURES.

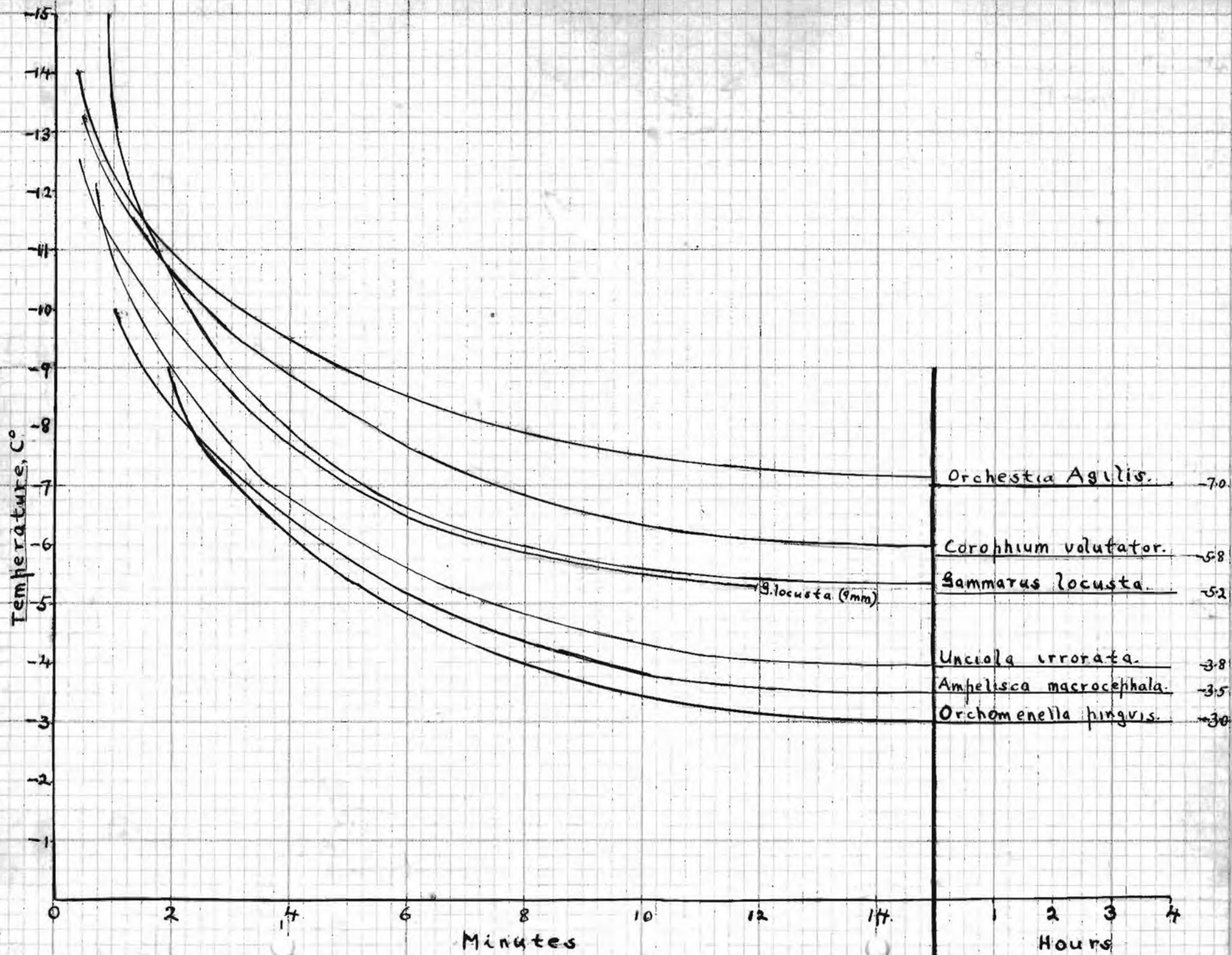
Fig.1. Shows temperature at which 50% die when subjected for a given time to that temperature. Shows that the ability to resist low temperatures is a function of the environment and not of size.

Fig.2. Correlating the temperature range which different species of amphipoda can withstand with their relative habitat depth and habitat temperature. The black represents the author's results, the dotted blocks represent the results of Huntsman and Sparks (1924), The lined block for O.locusta is only a probable one based on experiments on other species, the true lethal temperature (high) was not been determined

Fig.3. Cross-section of a region studied in February. The zone "A" was constantly covered with ice.

Fig.4. Temperature gradient box.

Fig.5. Showing the seasonal migration of G.locusta. The arrows show the direction of movement. The animals cross and recross the low water mark when the intertidal temperature reaches 3° to 5°C. (Spring and Autumn).

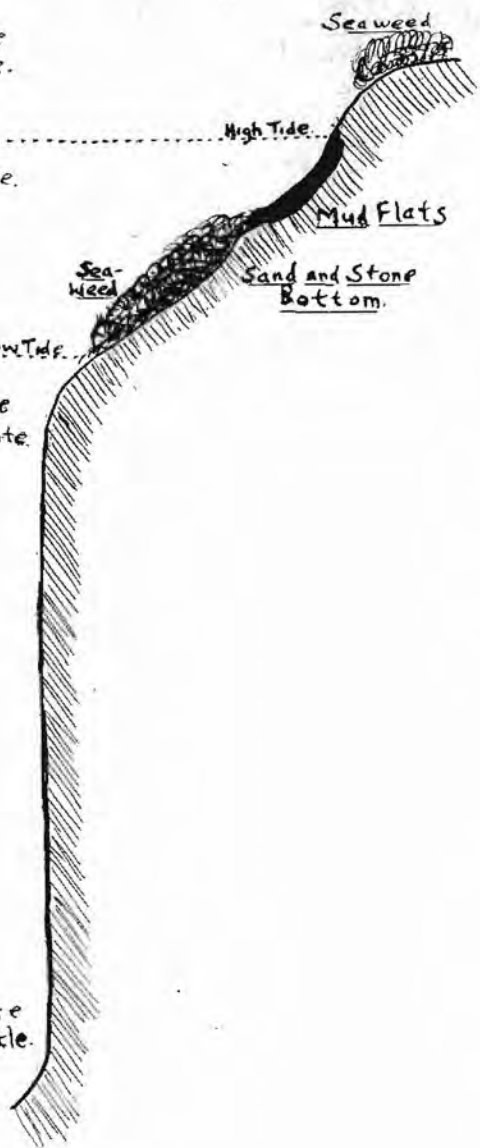


Temperature
Very Variable.

Temperature.
Variable.

Temperature
Range Moderate.

Temperature
Varies Little.



-7.00

0°C.

42.00?

Orchestia agilis

-5.80

Corophium volutator

37.2

-5.21

Gammarus locusta

33.3

-3.84

Unciola irrorata

29.0

-3.58

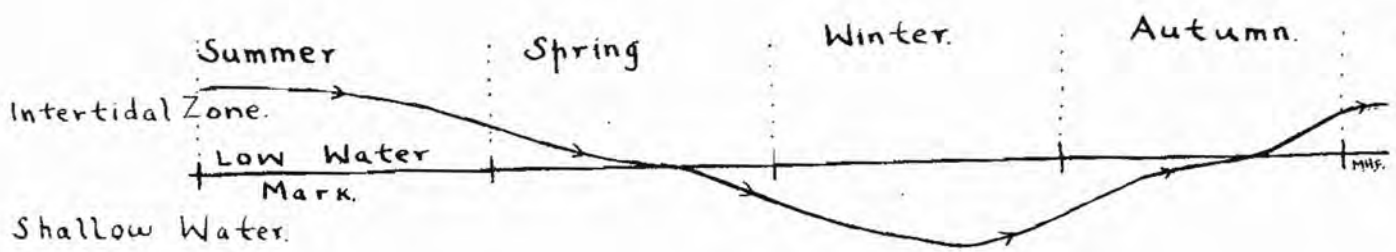
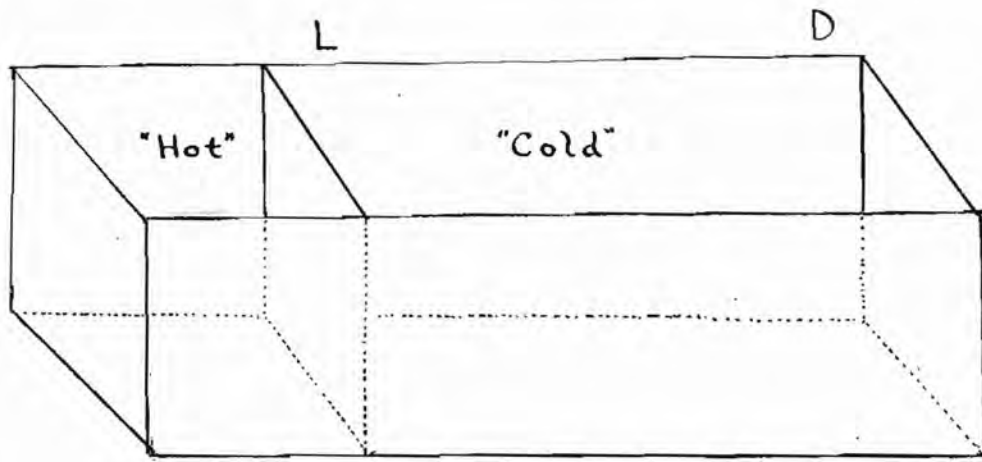
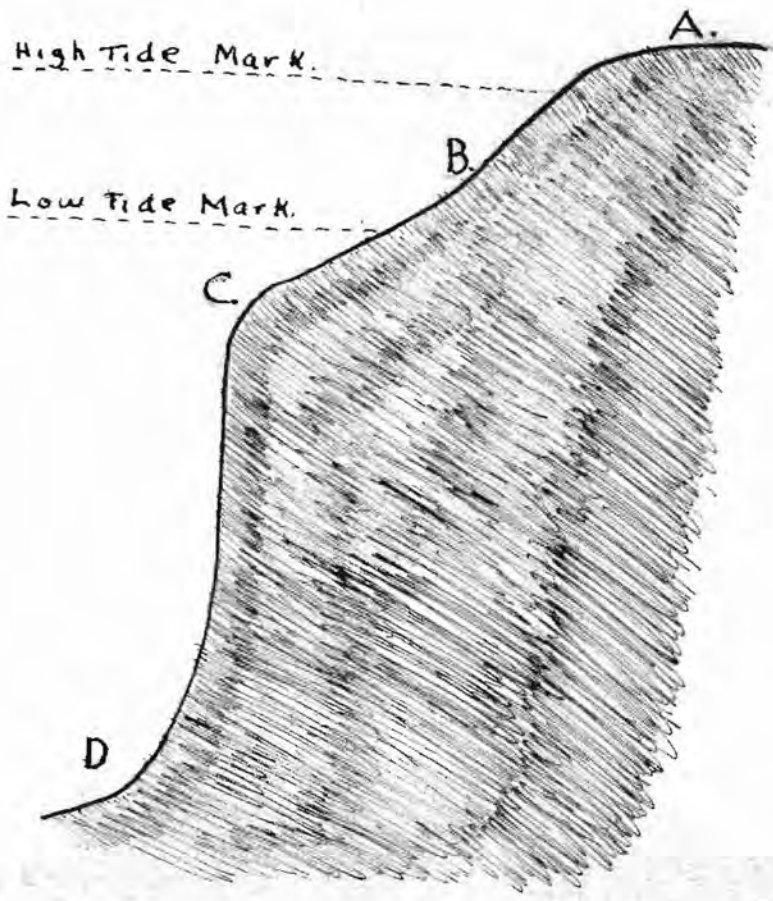
Ampelisca macrocephala

27.6

-3.00

Orchomenella pinguis

27.5



Showing the Seasonal Migration of *G. locusta*.