

the production of new characteristics by over-feeding and the fixation of these characters. The time during which these experiments have been in progress has been divided into three periods. During the first period, which includes the time immediately following the starting of the culture, the form of the head varied very widely under the new food conditions, but it soon returned to the original form when original conditions were restored. In three to four months after the culture was started, the form of the head was more regular and there were fewer aberrant individuals. Young females returned more slowly to the original head form when changed to original environment. The third period began almost two years after the culture was started and it was found that the young no longer returned to the original helmet form when original conditions were restored. A larger helmet persisted, thus showing a tendency toward the fixation of a new helmet form.

All of these experiments are still in progress and a more extended report on the results is promised at some future date.

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SCIENTIFIC JOURNALS AND ARTICLES

The American Journal of Science for September contains the following articles: "Use of the Grating in Interferometry," by C. Barus; "Fox Hills Sandstone and Lance Formation ('Ceratops Beds') in South Dakota, North Dakota and Eastern Wyoming," by T. W. Stanton; "New Occurrence of Hydrogiobertite," by R. C. Wells; "New Occurrence of Plumbojarosite," by W. F. Hillebrand and F. E. Wright; "Heat of Formation of the Oxides of Cobalt and Nickel," and sixth paper on the "Heat of Combination of Acidic Oxides with Sodium Oxide," by W. G. Mixter; "Mosesite, a New Mercury Mineral from Terlingua, Texas," by F. A. Canfield, W. F. Hillebrand and W. T. Schaller; "Researches upon the Complexity of Tellurium," by W. R. Flint; "Gravimetric Estimation of Vanadium as Silver Vanadate," by P. E. Browning and H. E. Palmer; "Brachiopod Genus *Syringothyris*

in the Devonian of Missouri," by C. Schuchert; "George Frederic Barker."

SPECIAL ARTICLES

THE INFLUENCE OF EXTERNAL CONDITIONS UPON THE LIFE CYCLE OF HYDATINA SENTA

THE search for the factors which regulate the production of the parthenogenetic and the sexual phases in the life history of the rotifer, *Hydatina senta*, has been conducted for some time. Maupas concluded that temperature regulated these two phases, while Nussbaum found that the controlling factor was food. Punnett and the writer¹ were unable to confirm these results. Recently Shull² has claimed that the absence of certain chemicals in the culture water causes the sexual phase to be produced, while the presence of these chemicals prevents the appearance of the sexual phase. This suggestion is probably partially true, but it does not seem to express the whole truth, nor solve satisfactorily the whole problem.

During the past two years I have kept pedigree strains or families of these rotifers continually in the laboratory and have made some observations which may lead to a clearer understanding of the conditions which control the production of the sexual and parthenogenetic phases in the life cycle of this rotifer. A general food culture for rotifers is usually made by adding about one hundred and fifty grams of fresh horse manure to about two thousand cubic centimeters of ordinary water and allowing this mixture to stand at room temperature after being inoculated with a miscellaneous lot of microorganisms. It is readily noticeable that in large jars of such newly made food cultures in which rotifers have been placed, that sexual females (females capable of producing either males from small parthenogenetic eggs or females from large fertilized eggs) appear quite abundantly for a few days or weeks, then gradually disappear and only parthenogenetic females remain in the cultures as they become older.

In June, 1909, several general cultures

¹ *Journ. Exp. Zool.*, Vol. 5, pp. 1-25.

² *Amer. Nat.*, Vol. 44, pp. 146-150.

of rotifers in jars which had been standing in the laboratory from four to twelve weeks were examined. Lots of five to eight hundred individuals were selected at random from each of these jars. In some of these lots three to five males were found, while in lots from other jars no males at all were found. In jars of new food cultures five to ten days old and stocked with individuals from these old culture jars, sexual females constituted as high as thirty per cent. of the individuals present.

In some experiments made this summer in which a few parthenogenetic females were placed in vials containing a small green flagellate, *Chlamydomonas*, and put into direct sunlight in order that the flagellates might remain active and serve as food for the rotifers, and also to aerate the culture water in the vials, males appeared either on the third or fourth day and on the immediately following few days. Then the males disappeared entirely and the parthenogenetic females increased in numbers to fifteen hundred to two thousand in each vial in the course of a week and a half to two weeks. These experiments were repeated several times with always the same result—that as the culture water became older the sexual females disappeared and the parthenogenetic females increased in numbers.

However, in the summer of 1909 in a pedigree strain of rotifers supplied entirely with water and food from an old culture jar, a line of parthenogenetic females was produced for twenty-six generations, but as the old culture water became low in the jar and the liquid very near the bottom was used, males appeared. This food culture was about eleven or twelve weeks old at this time and had been producing only parthenogenetic females for seven to eight weeks, and then under apparently the same conditions, suddenly produced sexual females.

This failure of an old culture water to produce continuously parthenogenetic females, and the high percentage of sexual females found in new culture water, suggested the possibility that the production of sexual females might be due to the *presence* of definite chemical substances in the culture water.

The possibility of this suggestion was strengthened by several cases of male epidemics which have occurred in my cultures.

In new horse-manure cultures at a temperature of 18–22° C., sexual females occur sometimes as high as thirty per cent., as has already been stated. In May, 1909, a large culture about two to three weeks old, containing rotifers, was standing in a south exposed window where it received the direct sunlight for a few hours each day and had its temperature thus raised to 28–30° C. Several lots of large eggs were selected at random from this culture during a period of three to four days, and placed at room temperature in some of the culture water in which the eggs were laid. In some of these lots of thirty to forty eggs, ninety-three per cent. developed into sexual females. Soon the parthenogenetic females began to increase and the sexual females to decrease in numbers, so that about a week later only about five per cent. of the individuals in the jar were sexual females. In another jar of newly made culture, in June, 1909, which was under the same conditions as the former jar, practically the same results were obtained again.

In November, 1909, the laboratory was closed one Sunday and the steam heat left on. The temperature rose to 26° C. or more. In three pedigree strains of rotifers which happened to be subjected to this great change of temperature, sexual females appeared in each strain in the following generations in greater numbers than they had appeared since the preceding May and June. It may be recalled that Maupas obtained a very high percentage of sexual daughter females when he subjected the adult mothers to a temperature of 26–28° C. Of course, when these females were at the high temperature the culture liquid in which the females were living was at the same temperature.

From a consideration of these general observations, it is conceivable that in newly made cultures of horse manure and water during the great chemical changes that are taking place in the decomposition that occurs during the first two weeks, definite but transitory chemical compounds are formed which so act

upon the parthenogenetic female as to cause her to produce sexual daughter females. These chemical compounds may not be final products of decomposition, but break up into or form other products which possess different properties. When these compounds are forming, a higher temperature under certain conditions augments them and consequently they appear in greater abundance suddenly, and thus act upon the parthenogenetic females and cause male epidemics in the third generation.

As the culture water becomes older the decomposition rapidly decreases and the special chemical compound which causes sexual females appears in inappreciable quantities, if at all, throughout the liquid. However, in the bottom of old culture jars decomposition may not have ceased, as was evident in the breaking up of the parthenogenetic pedigree strain into sexual females at the end of twenty-six generations. This chemical substance is evidently something which appears more or less abundantly at first in decomposition and then later disappears or its influence is counteracted by other substances.

Whether the production of parthenogenetic females is sometimes brought about by the action of a different chemical compound or sometimes by the mere absence of the sexual female producing chemical compound is not as yet altogether clear, but the latter possibility seems more probable.

On January 16, 1910, I began feeding two pedigree strains of rotifers with the small flagellates, *Polytoma*, which grew in a culture of about one hundred grams of fresh horse manure and five hundred cubic centimeters of tap water, that had been steam sterilized for about one hour. The *Polytoma* grew very quickly in these cultures and in 24-48 hours immense numbers of them were produced. These new food cultures of *Polytoma* ranging from 24-96 hours old were diluted, one part culture water to two parts tap water, and used to grow the rotifers in. Such diluted *Polytoma* cultures, none of which were over 96 hours old and in which the culture water was cooked, have been used from January 16 to August 13. In each gen-

eration of these two strains ten daughter females were isolated from one to three mothers. Each individual was placed in a separate watch glass and kept at room temperature. In these two strains of a hundred generations and consisting of one thousand individuals in each strain no sexual females have ever appeared during a period of about seven months.

These long parallel series of parthenogenetic females are similar to Punnett's pure female strains. However, the parthenogenetic females of both strains in the generations between the ninetieth and the one hundredth, have produced sexual daughter females when placed in very little dilute culture solution and fed upon the green flagellate, *Chlamydomonas*, thus showing that these are not pure parthenogenetic female strains, and that the production of sexual females has been suppressed since January by some condition of the culture water.

These results are similar to those obtained by Shull who used old culture water, although produced by using newly made cooked culture water and extending over a longer period of time. At the end of some starvation experiments in which Shull used dilute culture water, he makes this general concluding statement, "The larger proportion of sexual forms in the starved families is not due to lack of food, but to the absence of chemicals which, in the well-fed families, prevent the appearance of the sexual forms."

In February, 1910, I had in the laboratory some pure cultures of a colorless flagellate, which seemed to be a species of *Peranema*. These flagellates were very resistant and could live and swim about normally several hours in distilled water, and were readily eaten by the rotifers. These flagellates were taken in quantities, put into large test-tubes, placed on a large electric centrifuging machine, and collected in the bottom of the tubes by centrifugal force. The old culture water was removed, clean tap water added, and the contents thoroughly mixed. Then it was centrifuged again and the protozoa collected at the end of the tubes. This process was repeated

four or five times until the protozoa were thoroughly washed, and no trace of the old culture water remained.

Several parthenogenetic female rotifers were also washed by dropping them into four or five successive dishes of tap water. Then series of watch glasses were prepared containing five cubic centimeters of distilled water in which there were large numbers of the *Peranema*, and amounts of cold culture water varying from one drop to four cubic centimeters. In pure distilled water the rotifers soon died and also in the dishes containing very small quantities of the old culture water, while in the dishes containing larger amounts of the old culture water the rotifers lived and reproduced normally. Under these varying conditions three generations were reared, but no sexual females were produced in any of the dishes.

These experiments in which the quantity of old culture water varied from zero to four cubic centimeters and only parthenogenetic females were produced seem to indicate that the substance which causes sexual females to be produced was absent altogether in this old culture water. If this is true, then perhaps the mere absence of the substance which causes the sexual females to be produced is always sufficient to cause the production of parthenogenetic females and it is unnecessary to look for a specific substance which causes their production.

Newly made uncooked cultures of horse manure and water in which rotifers can live readily are more or less dilute, but as they grow older they become more concentrated by the end products of decomposition. If mere dilution of substances in the culture water, as Shull seems to maintain, produces sexual females, then epidemics of males ought to occur in culture water during the very first days when such culture water is most dilute, and not several days later as it becomes more concentrated by the end products of decomposition. However, the epidemics of males that occurred in my two general culture jars which were between two and three weeks old were preceded by a production of males which

did not exceed thirty per cent. during a period of at least a week.

In some pedigree families of rotifers that I observed in 1907 and 1908, it was found that in any single family of forty to fifty daughters, if there were any sexual daughters they occurred among the first half of the family. When a mother was isolated she was fed and then remained in the same food culture water without the least change until all of her daughters were produced. Sometimes she would produce ten or more sexual daughters in succession, which were often preceded by several parthenogenetic sisters and always followed by parthenogenetic sisters.

It is plain that dilution of the culture water did not occur to cause these series of sexual daughters and it is conceivable that the chemical substance that produces males, in some cases, when sexual females occurred among the first daughters, was present in the culture water at the time of isolation of the mother, and in other cases, when the sexual females appeared between the tenth and twenty-fifth daughter, this substance was formed some time after the mother was isolated and had laid some of her eggs. In all cases its influence disappeared as the culture became older and no sexual daughters appeared in the last half of the family.

It seems evident from all the observations that some culture waters totally lack the power to cause sexual females to be produced, others lack this power at first, but acquire it later, while still others possess it as the cooked new cultures and some old cultures, but are unable to use it unless the culture water is diluted.

In a summary I would maintain that there seems to be a definite, but transitory, chemical substance produced in appreciable quantities in the decomposition processes in newly made horse manure cultures that can so act upon the parthenogenetic females as to cause them to produce sexual daughter females. When this substance is absent no sexual females are ever produced, but only parthenogenetic females are produced, and when this substance is present in culture water which is too concentrated its influence is counteracted and no

effect is produced on the parthenogenetic females.

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August 15, 1910

A REPORT ON THE FRESH-WATER PROTOZOA OF
TAHITI¹

THE following brief report is a result of some work done upon the protozoa of Tahiti during the months of July and August, 1908.

An oceanic island is always an interesting field for the investigation of the higher forms of animals, because of its faunal peculiarities or deficiencies, but a valid question arises, namely, may we expect these same peculiarities or deficiencies to exist with respect to animals of the lowest rank?

So far as the writer has been able to determine, no list of fresh-water protozoa of a mid-ocean island has previously been reported.

Tahiti, the largest of the Society group, is situated $17\frac{1}{2}^{\circ}$ south latitude, $149\frac{1}{4}^{\circ}$ west longitude. It is a high island of over 400 square miles, of volcanic origin, more or less densely covered with a tropical vegetation. Mountains of the interior reach a height of nearly 8,000 feet, and numerous streams of fresh water flow down the valleys across the narrow plain to the sea.

During the winter months of July and August, in Tahiti, the small streams are in a low condition, it being the dry season, and many of them are choked with plants of low orders, which would apparently be a fit condition for the presence of a rich microscopic fauna.

During the brief time allotted to the study of the protozoa, collections were made in many places from the waters of this border zone a few yards from the sea and from its level to a few feet above.

In all, forty-four species were observed and studied with considerable care. Of these, thirty-six were positively identified, and eight referred to their proper genera, but the species undetermined.

¹Read before the Central Branch of the American Society of Zoologists at Iowa City, April, 1910.

Of the thirty-six species identified, nine were of the class Sarcodina, six of the class Mastigophora, and twenty-one of the class Infusoria. Of the undetermined species, two were rhizopods and six ciliates.

All of the thirty-six species studied in Tahiti are more or less common in the waters of this state; twenty of them have been reported from Boulder, Col., by Cockerell; and nearly all of them from Connecticut by Conn.

Penard, in the *American Naturalist* for December, 1891, lists thirty-six species of rhizopods found in the Rocky Mountains near Caribou, Col., at a height of 10,000 feet, and thirteen species at 12,000 feet. Of the thirty-six species listed by Penard at an elevation of 10,000 feet, six species were found in tropical Tahiti within a few feet of the level of the sea. Of the thirteen species listed by Penard found at an elevation of 12,000 feet, one *Difflugia pyriformis*, is a rather common rhizopod in Tahiti at sea-level.

Penard calls attention to the fact that the rhizopoda of higher altitudes are those with lobe-like pseudopodia, the forms with ray-like pseudopodia being absent. It may be added that the predominating rhizopods of the sea-level are also of the lobose type, and a majority are protected by shells. Only one species of rhizopoda with ray-like pseudopodia was found in Tahiti.

Taking as a basis the list of protozoa reported by Stokes in 1888, the list reported by Conn in 1905, and the list of the writer in 1906, it is quite safe to say that the relative proportion of the protozoa for the United States of the three classes (sporozoa not included), is approximately as follows: Sarcodina, 15 per cent.; Mastigophora, 25 per cent., and Infusoria, 60 per cent.

Taking forty-four species of Tahiti as a basis, the proportion is as follows: Sarcodina, 25 per cent.; Mastigophora, 14 per cent.; Infusoria, 61 per cent.

It would seem from these observations that the proportion of infusoria reported in the oceanic island holds true to that of the United States, a variance appearing in the case of the other groups.

Among the pseudopodia-bearing forms, *Ar-*