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CULTURE OF DAPHNIA

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DAPHNIA are crustaceans which belong to the same large group of animals as the crayfish and the lobster. They are also Entomostraca (a sub-class name) and belong to the order Cladocera. In addition to Daphnia, other genera of Cladocera are cultivated such as Moina, Simocephalus, Scapholeberis, and Chydorus.

There has been an increased interest in the culture of daphnids for feeding young fish, particularly bass, since the large species, Daphnia magna, was imported from Canada in the spring of 1924. At that time, Dr. Klugh, Professor of Zoology at Queens University, Canada, but residing at Ithaca, N. Y., received several live individuals from Canada. Probably most of the American stock of this species has been obtained from the progeny of these specimens. Daphnia magna occurs naturally in this country, however, and was reported from Maine, North Dakota, Colorado, Nebraska, and California prior to 1918. The maximum size of females of this species is about 5.0 millimeters, while Daphnia pulex is about half that length. Hayford (1927) states that the great advantage of Daphnia magna for feeding bass is that it hatches at least four weeks earlier than the other daphnia. Actually, there probably is not a great deal of advantage in culturing D. magna over P. pulex or other species unless it is that adults of D. magna are larger and are eaten by bass fingerlings until they reach a fairly large size. On the other hand, the adult specimens of D. magna are too large for bass fry to consume when they first rise from their nests. However, there always are plenty of young daphnia of suitable size for bass fry in any thriving culture.

Daphnia pulex is universally distributed and may be found in temporary ponds and sometimes in lakes. Moina inhabits barnyard, brickyard, and other temporary ponds, and it is reported that it may be reared farther south than its larger relative, Daphnia magna.

INDOOR CULTURE

The first problem confronting the prospective daphnia culturist is how to get cultures started. It is advisable to have culture media ready in advance of the collection or the receipt of live individuals or eggs.

Note: - Revision of Memorandum I-131, No. 17, The Progressive Fish Culturist, pp. 1-6, April 1936

Indoor cultures may be prepared as follows: Dr. Arthur M. Banta (1921, pp. 557-558; Needham, et al 1937, pp. 207-210) found that a culture medium could be obtained readily anywhere and at any season of the year by placing two pounds of fine garden soil and six ounces of finely divided fresh (8-15 days old) horse manure in a 9 x 12-inch battery jar and covering with 10 quarts of strained pond water. Pond water is specified because Cladocera are extremely susceptible to the toxic effects of the salts of the heavy metals. Galvanized or copper containers are avoided for the same reason. Tap water should be used with caution until proved innocuous. The mixture is allowed to stand at 59 to 68 degrees Fahrenheit without disturbance for three days, when it is strained through silk bolting cloth (#20) or other very fine material. Dilute with strained pond water one to four parts or one to two, depending upon the degree of density in the appearance of the solution. Stir thoroughly and allow to stand for a short time (about an hour) before stocking with Daphnia. Bacteria constitute the principal food element in this culture medium.

Another simple method is described by Bond (1934, p. 60). He recommends the use of ordinary Fleishman's yeast (in cakes) as a cultural food for Daphnia. About one-quarter fresh yeast cake is mixed into a uniform suspension with from about two to three ounces of water and poured into the aquarium which contains from 60 to 70 quarts of water. The feeding is repeated every fifth or sixth day. He states, however, that it is necessary to have a stream of air bubbling through the medium at all times, probably to remove carbon dioxide gas. This is the unfortunate part of this method, but yeast may be used without this aeration if it is used in smaller quantities than recommended above.

Chipman (1934, pp. 59-60) describes a method in which cottonseed meal is used as follows: one-half ounce cottonseed meal and three ounces of garden soil are stirred in one quart of strained pond water and set aside for five days. The water is then poured off the surface and strained through muslin. One part of this is diluted with 100 parts of filtered water and innoculated with Daphnia after the pH of the diluted solution is adjusted to 7.2 with sodium carbonate. Fresh stock supplies of the cottonseed mixture were prepared each week, a small amount of the old mixture being added each time to insure innoculation with the original bacteria.

Arthur D. Hasler (Needham et al. 1937, pp. 214-215) used 1 teaspoonful of dry sheep manure and about 1/60th ounce (0.5 gram) acid phosphate per gallon of water to which a quart of water filtered through rich garden soil was also added. This mixture was allowed to stand for a day before innoculation with Daphnia.

All sorts of substances have been used in the culture of water fleas indoors (Needham et al. 1937), but it is believed that the described methods will suffice.

Outside pools for large scale production of Daphnia should be set up several days to a week in advance of their innoculation.

LARGE SCALE CULTURAL METHODS IN OUTSIDE POOLS

In the production of Daphnia which are to be used in feeding your bass, for example, concrete ponds about 2 by 5 by 30 feet or 2 by 6 by 20 feet or similar dimensions probably will be found most convenient, although any size

pond may be used. The Daphnia can be removed readily with a seine of small mesh bobinetting or of fine bolting silk (00XX is fine enough), or other fine material, from ponds of these approximate dimensions. At Leetown, West Virginia, we have used our bass fry seine of bobinetting for collecting Daphnia from our ponds, which measure 3 by 8 by 25 feet.

At some fish cultural stations a series of these concrete Daphnia ponds are located at such elevations above the bass nursery ponds that the Daphnia may be flowed into them by gravity. However, such ponds seldom seem to be used in this manner, and there is the added disadvantage that the young bass congregate about the inlet pipes from these ponds where they become easy prey for their faster-growing cannibalistic brothers and sisters. The usual practice is to seine the Daphnia from the ponds and broadcast them here and there about the borders of the larger nursery or feeding ponds.

Embody and Sadler (1934) report experiments the object of which has been to devise methods of producing large quantities of Daphnia throughout the summer. In their earlier experiments efforts to keep a single culture going continuously throughout the summer failed. Very successful cultures were obtained in May and part of June, which ran out in July and came back in late September and October. A temperature of 82 degrees Fahrenheit appeared to be a cause of diminishing numbers of Daphnia. In later experiments, they worked on methods of bringing cultures to their peak production, when the Daphnia were removed, the culture ponds drained and new cultures set up. With 7 concrete ponds, 8' by 12' with a water depth of 12 to 14 inches, a new culture was started every 4th day. Embody and Sadler (1934, p. 207) give a schedule for operating Daphnia ponds using four kinds of fertilizer. This table is given below:

Table 1. -- Amount of fertilizer per 100 cubic feet of water and its apportionment

	Experimental ponds at Ithaca, New York			
	Pond No. 1 : Soybean meal	Pond No. 2 : Cottonseed meal	Pond No. 3 : Dry Buttermilk	Pond No. 4 : Sheep Manure and Soybean meal
Initial dose	1 pint	1 quart	1/2 pint	4 quarts Sh.M 1 pint S.B.M.
Second dose	5th day 1 pt	7th day 1.5 pt	7th day 1/2 pt	14th day 1/2 pt S.B.M.
Third dose	10th day 1 pt	14th day 1 1/2 pt.	14th day 1/2 pt	----------
Fourth dose	15th day 1/2 pt.	----------	----------	----------

Note: Harvest on the 21st day.

According to these authors, the fertilizers which gave cultures averaging ighest were dry buttermilk, soybean meal, and cottonseed meal. Very little ifference between them was noted. It happened that the densest culture, amounting to 2,416.5 grams of daphnia, mosquito and midge larvae, was obtained in 24 ays using one pint soybean meal at the start and one pint every sixth day thereafter, making a total of two quarts for the period. The water temperature varied etween 70 and 80 degrees Fahrenheit.

The population densities of these cultures reached their maxima in from 16 o 26 days; 21 days was the average.

Langlois (1931, p. 109) used six quarts of sheep manure and one quart of superhosphate per 100 cubic feet of water with good results. Daphnia could be reoved daily from these ponds after three weeks and a culture was made to last ix to eight weeks by the periodic addition of more fertilizer.

O. Lloyd Meehean (in a report to the former Bureau of Fisheries now Fish nd Wildlife Service) observed that a good culture of Daphnia magna could be btained by the addition of 24 pounds of sheep manure to each 400 cubic feet f water. Additional amounts of sheep manure were added at the rate of twelve ounds each week unless the water temperature increased, when smaller amounts ere added. He observed that this mixture lacked something (a vitamin or similar ubstance) to give the culture that extra pulse which would make the animals bundant enough to remove in large numbers. This substance was supplied by the ddition of green grass or oats.

Hayford (1927, p. 143) used twenty quarts of solid green cow manure and ixed it in an ash can of water to which four quarts of menhaden meal and four uarts of bean meal were added. This mixture was placed in concrete ponds 2 y 5 by 30 feet. He notes that horse manure is good, but it is not equal to he cow manure. In these same pools containing 225 cubic feet of water, Robert ayford (personal communication, April 1935) reported success using sixty pounds f cow manure just as it comes from the cow stables, straw and manure. When he water begins to clear, only a few pounds of manure are added to keep the ulture going.

Kauffman (1943, p. 56) used one large shovelful of chicken manure in a pool by 10 by 20 feet with good success.

Nelson (1941) records phenomenal production of largemouth bass on daphnia y stocking 100,000 to 150,000 fry in ponds fertilized as follows: 3 tons or 1 ton per acre-foot) of cow-lot manure is piled in the center of the pond. f fertilization is adequate, a good amber color is produced within 24 hours fter the pond is filled. Additional manure is supplied as needed by piling t along the windward side of the pond. Old heaps or windrows are removed after ach season.

The list of fertilizers used in culturing daphnia in aquaria and in ponds is ong. Most of them contain organic matter which can be decomposed with the aid f bacteria. Some of the materials not already mentioned are the following: ardine meal, green bone meal, decayed fish, cooked oatmeal, clam meal, beef earts, salt water mussels (Mytilus edulis), herring meal, and shrimp bran, and hicken manure.

Running water is, of course, not used in daphnia ponds, water being added only to supply that lost by evaporation or seepage.

When a culture is in normal healthy condition, there are all sizes of daphnia from the very young to the adults, eagerly feeding upon the bacteria present. Numerous large daphnia are present with parthenogenic of sexual eggs and young daphnia in their brood chambers. These parthenogenic eggs are usually green in color, visible to the naked eye, and often a half dozen or more are present in an individual. When a culture is in nearly perfect condition, males are not developed so that all, or practically all, individuals in a culture are egg or potential egg producers.

Certain evidence of the decline of a culture or its diminished capacity to produce progeny is in the appearance of individuals bearing one or two eggs in a black saddle on the back. These eggs are sexual or "winter" eggs which require fertilization by male daphnia. The male daphnia are small individuals which become differentiated from potential females when the competition for food is too great, or when some other unfavorable condition has arisen or is impending. These winter eggs are the means by which the species is perpetuated over winter periods and drought. They are also a means of distribution because they can be readily blown about by winds on becoming dried out. Needham (1933, p. 132) cites the experiments of Henry Markus of the former U. S. Bureau of Fisheries who found that these eggs required alternate freezing and thawing for a number of times before they would hatch. This could be done in an electric refrigerator. The dried winter eggs treated in this manner may be transported in the mail from place to place at little cost.

Most daphnia cultures reach their maximum production in about three weeks. If quantities of daphnia are removed and small additional amounts of fertilizer added, the life of the culture may be prolonged, but it is generally not worth while to attempt to maintain them much beyond this time. The best plan appears to be to set up cultures in rotation so that new cultures will be coming on as the old are discontinued. When winter eggs predominate among the individuals in a culture, it is time to drain the pond, clean it out, and start over again.

High water temperatures are not conducive to daphnia culture, particularly that of Daphnia magna. O. Lloyd Meehean (Fish and Wildlife Service) has observed that they cannot be raised in profitable numbers unless the water temperature remains below 75 degrees Fahrenheit. As has already been stated, Embody and Sadler (1934) observed that a temperature above 82 degrees Fahrenheit appeared to be injurious. Shades over pools may be used, at least for a time, to prevent the water from becoming too warm.

Usually during the duration of a culture, particularly if it is maintained for a long time, foreign organisms are attracted to the pool which feed upon the daphnia. Many of these predators are air-breathing insects, which, although they swim about, submerged, eventually come to the surface for a supply of air. Such insects can be destroyed by producing a thin film of oil of a non-toxic nature, such as salmon oil, herring oil, or cod liver oil, over the surface of the pool. This cuts off their air supply, and they die quickly. The number of tadpoles can be kept down by removing the masses of frog eggs before they have an opportunity to hatch.

Only a small amount of information is available on the amount of daphnia, or weight of fish food, produced by a single culture of daphnia. The most accurate data yet observed by the writer is that of Embody and Sadler (1934) who give the actual weight in grams of daphnia, mosquito, and midge larvae produced in several of their experiments. Their densest culture amounted to 2,416.5 grams of daphnia, mosquito, and midge larvae after 24 days using a total of 2 quarts of soybean meal in a concrete pond with a water capacity of 100 cubic feet.

Embody (1942) later records an average production of 0.721 ounce of food organisms per cubic foot of water with soybean meal as compared with 0.43 ounce with cottonseed meal.

O. Lloyd Meehean reports what appears to be a remarkable production from a pond of 400 cubic feet capacity. This pond was set up on May 11, and fertilized with 24 pounds of sheep manure. It was stocked on May 12, with Daphnia magna. Some bundles of green grass or oats were added. On May 17, 12 pounds of fertilizer were added to the culture since a rain the night before had diluted the solution. On May 22, 17 pails of daphnia were removed from the pond. These were Fearnow pails one-quarter to one-half filled with water and the remainder daphnia. On May 25, ephippial or "winter" eggs began to appear and 16 additional large pails were taken out, making a total of 33 pails to the time of the appearance of the ephippial eggs. On May 26, 16 more pails were removed, bringing the total to 49 pails. Two inches of water were added along with 12 pounds of fertilizer. No more daphnia were taken from this pond until June 2, when 3 pails were removed, and on June 3, the pond was drained and cleaned yielding 12 more pails of daphnia. There were altogether 64 pails of daphnia raised in the culture. The temperature of the pond was 64.4 degrees Fahrenheit at the start of the culture but climbed to 79.7 and was 75.2 degrees when the pond was drained.

It is the collection by comparable methods of such quantitative data as the above which will ultimately decide which are the best methods of producing daphnia.

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