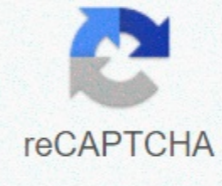




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Water daphnia culture

6.1.1. Biology and life cycle of Dafnie 6.1.2. Nutritional value of Dafnie 6.1.3. Feeding and feeding Dafnie 6.1.4. Mass culture dafnie 6.1.5. Production and use of resting eggs 6.1.6. Use moina 6.1.1. Biology and life cycle Daphnia Daphnia is a frequently used food source in freshwater larvikult (i.e. for different carp species) and in the ornamental fish industry (i.e. guppies, sword tails, black mollies and plattys, etc.) Dafnie belongs to the sub-order cladocera, which are small crustaceans that almost exclusively live in fresh water. The shell surrounds the entire trunk, except for the head and apical spine (if present). The head projected ventrally and somewhat posteriorly in a beak-like snout. Stem additions (five or six pairs) are flattened, leafy structures that are used to feed the suspension (filter feeders) and for movement. The front of the trunk, postabdomen rotates ventrally and forward and carries special claws and spines to clean the shell (Fig. 6.1.). Species of the genus Daphnia are found from the tropics to the Arctic, in habitats of different sizes from small ponds to large freshwater lakes. Currently, 50 dafnia species are reported worldwide, of which only six are usually found in tropical lowlands. The size of the adult is exposed to large changes; when food is abundant, growth continues throughout life, and large adults can have a shell length twice as long as in newly mature individuals. In addition to differences in size, the relative size of the head can gradually change from a round shape to the shape of a helmet between spring and the summer solstice. From the summer solstice to the fall, the head turns back to a normal round shape. These different forms are called cyclomorphic and can be triggered, as in rotifers, by internal factors, or may be the result of interactions between genetic and environmental conditions. Normally there are 4 to 6 instar stages; Daphnia grows from naupliu to mingling through a series of 4-5 molts, the time depends mainly on temperature (11 days at 10 ° C to 2 days at 25 ° C) and the availability of food. Daphnia species reproduce with either cyclic or compulsory parthenogenesis, and populations are almost exclusively female. Eggs are produced in the joints of two to several hundred, and one female can produce several joints associated with the process of blending. Parthenogenetic eggs are produced ameiotically and lead to females, but in some cases males may appear. In this way, the reproductive pattern is similar to rotifers, where parthenogenetic diploid eggs are usually produced. Parthenogenetic eggs (their number can range from 1 to 300 and depend largely on the size of the female and food intake) are laid in the amniotic chamber shortly after edryze and hatch just before the next ekdysis. Embryonic development in kladocerans occurs and larvae are miniature versions of adults. In some cases, the embryonic period does not correspond to the period of the fetus, which means that the larvae are kept in the amniotic chamber even after the completion of the embryonic period, due to postponed oesasis (environmental factors). For different species, the ripening time is remarkably uniform at given temperatures ranging from 11 days at 10 ° C to just 2 days at 25 ° C. Factors, such as a change in water temperature or food deprivation due to an increase in the population, can provoke the production of men. These males have one or two gonopores that open near the rectum and can be modified into a copulatory organ. The male spoils the female with the first antennae and inserts the copulation processes into a single, medium female gonopore. Fertilized eggs are large, and only two are produced in one joint (one from each ovary) and are a thick shell: these resting or dormant eggs are enclosed by several protective membranes, ephippium. In this form, they are resistant to dried, freezing and digestive enzymes, and as such play an important role in the colonization of new habitats or in the restoration of the extinguished population after adverse seasonal conditions. 6.1.2. The nutritional value of Daphnia The nutritional value of Daphnia strongly depends on the chemical composition of their food source. However, because Daphnia is a freshwater species, it is not a suitable prey for marine organisms because of its low content of essential fatty acids, and in particular (n-3) HUFA. In addition, Daphnia contains a wide range of digestive enzymes such as proteinases, peptides, amylase, lipases and even cellulases, which can serve as exo-enzymes in the gut of fish larvae. 6.1.3. Feeding and nutrition Dafnie Filter dafnie is made of specialized thoracic allowances for collecting food particles. Five thoracic limbs act as a suction and pressure pump. The third and fourth pairs of pendants carry large filter screens that filter particles from water. The effectiveness of the filter also allows the intake of bacteria (approx. 1µm). In a study on the quality of freshwater phytoplankton food for the production of cladocerans, that of the spectrum of blue-green, flagellate and green algae, Daphnia performed best on the diet of cryptomonad, Rhodomonas minutae and Cryptomonas sp., containing high levels of HUFA (more than 50% of the fatty acids in these two algae consisted of EPA and DHA, while green algae were characterized by more 18:3n-3). This means that long-term polyused polysaturated fatty acids are important for the normal growth and reproduction of Daphnia. Heterotrophic microflagellates and ciliates up to the size of Paramecium can also be used as food for Daphnia. Even the sut and food can be an important food source, especially if the concentration of food falls below a certain threshold. In this case, the water current produced by the animals that swims to the bottom ejects the material that is eventually infused. Since daphnia appears to be non-selective filter feeders (i.e. they do not distinguish between food particles to taste), high concentrations of suspended material may interfere with food particle uptake. Figure 6.1. Schematic drawing of the internal and external anatomy of Daphnia. 6.1.4. Dafnie mass culture 6.1.4.1. General procedure for tank culture 6.1.4.2. Detrital system 6.1.4.3. Autotrophic system 6.1.4.4. General procedure for pond culture 6.1.4.5. Contamination 6.1.4.1. The general procedure for the cultivation of reservoir dafnie is very sensitive to contaminants, including leaching components from the holding equipment. When using plastic or other polymer vessels, a certain time of leaching will be required to remove toxic compounds. The optimal ion composition of the culture medium for Daphnia is not known, but the use of hard water containing about 250 mg.l-1 CO32-is recommended. Potassium and magnesium levels should be kept below 390 mg.l-1 and 30-240 µg. l-1. Maintaining a pH between 7 and 8 seems important for a successful Dafnie culture. To maintain water hardness and high pH levels, lime is usually added to the tanks. The optimal culture temperature is about 25 ° C and the tank should be gently aerated to keep the oxygen level above 3.5 mg.l-1 (dissolved oxygen levels below 1.0 mg.l-1 are fatal for Daphnia). Ammonia levels must be kept below 0.2 mg.l-1. Vaccination is carried out using adult Daphnia or resting eggs. The initial density is generally in the order of 20 to 100 animals per liter. Normally, the optimal algae densities for Dafnie culture are about 105 to 106 cells. ml-1 (larger dafnia species may support 107 to 109 cells.ml-1). There are two techniques for obtaining the desired algae densities: detrital system and autotrophic system: 6.1.4.2. Detrital system The stable tea breeding system is a culture medium consisting of a mixture of soil, manure and water. Manure acts as a fertilizer to support algae flowers on which dafnides feed. One can use fresh horse manure (200 g), which is mixed with sandy clay or garden soil (1 kg) in 10 l of pond water for a stable stock solution; this solution diluted two to four times can then be used as a culture medium. Other commonly used fertilisers are: poultry manure (4 g.l-1) or cow faeces substrates. This system has the advantage of being self-sustaining and Daphnia are not quickly exposed to deficiencies, due to the wide range of blooming eyelashes. However, the culture parameters in the detrital system are not sufficiently reliable for the cultivation of Daphnia under standard conditions, i.e. they are not reliable enough to cultivate Daphnia under standard conditions, resulting in anoxic conditions and consequently high mortality and/or ephippial production. 6.1.4.3. Autotrophic system Autotrophic systems, on the other hand, use the addition of cultured algae. Cultures of green water (105 to 106 cells.ml-1) obtained from wastewater from the pond are often used, but these systems show large differences in production speed, mainly due to the variable composition of algae species from one wastewater to another. The best control over the culture medium is obtained with the use of pure algae cultures. These can be monocultures such as algae such as Chlorella, Chlamydomonas or Scenedesmus or mixtures of two algae cultures. The problem with these selected media is that they are not able to sustain many generations of Daphnia without adding additional vitamins to Dafnie cultures. A typical mixture of vitamins is represented in Table 6.1. Table 6.1. Vitamin mixture for monospecific culture of Daphnia on Selenastrum, Ankistrodesmus or Chlamydomonas. Add one ml of this stock solution to each litre of algae culture medium (Goulden et al., 1982). Nutrient concentration in stock solution (µg.l-1) Biotin 5 Thiamin 100 Pyridoxine 100 Pyridoxine 3 Calcium panthothenate 250 B12 (as manitol) Nicotinic acid 50 Nicomid 50 Folic acid 20 Riboflavin 30 Inositol 90 Regular sampling of population density must be carried out regularly to calculate daily algae requirements and to estimate harvest time. Harvesting techniques can be non-selective regardless of size or age group or selective (only medium-sized daphnids are harvested, leaving newborns and mature individuals in a culture tank). Mass cultivation of Daphnia magna can also be achieved on inexpensive agro-industrial residues such as cotton seed meal (17 g.l-1), wheat bran (6.7 g.l-1) etc. Rice bran has many advantages compared to other live foods (such as microalgae): it is always available in large quantities, it can be easily purchased at low prices, it can be used directly after simple treatment (micronization, flaking), it can be stored for a long time, it is easy to dose and has no problem with maintaining stocks of algae and cultures. In addition to these benefits, there is also the fact that rice bran has a high nutritional value; rice bran (fat) containing 24% (18.3%) crude protein, 22.8% (1.8%) gross fat, 9.2% (10.8%) fiber and is a rich source of vitamins and minerals. Dafnie can be grown on this food item for an unlimited number of generations without noticeable deficiencies. Fat-fat rice bran is preferred over raw rice bran because it prevents hydrolysis of the fatty acids present and subsequently the yellowing of the product. Micronization of bran into particles less than 60 µm is usually carried out by adjusting the aqueous suspension (50 g.l-1) by manual conciliation and filtering it 60 µm or its industrial preparation in a dry mill. The suspension is given in small quantities for 24 hours: 1 g of fat rice bran per 500 individuals for two days (density: 100 animals.l-1). The ratio of food conversion has an average of 1.7, which means that with less than 2 kg of dry rice bran, approximately 1 kg of wet dafnid material can be produced (with 25% water recovery per week; De Pauw et al., 1981). 6.1.4.4. The general procedure for pond culture Daphnia can also be produced in ponds at least 60 cm in height. To produce 1 tonne of Daphnia biomass per week, a 2500 m3 cultivation pond is required. The pond is filled with 5 cm of dried (for 3 days) soil, to which lime powder is added at a rate of 0.2 kg of lime powder per tonne of soil. After that, the pond is filled with water up to 15 cm. Poultry manure is added to ponds in the .m 4. Fertilization of the pond with organic manure instead of mineral fertilizers is preferred, since cladocerans can use a large part of the manure directly in the form of rubble. On day 12, the water level is raised to 50 cm and the pond is fertilized for the second time with poultry manure (1 kg.m-3). After that, the weekly fertilization rate is maintained at 4 kg of poultry manure per m-3. In addition, fresh cow manure can also be used: in this case, a suspension containing 10 g.l-1 is prepared, which is then filtered through a sief by 100 µm. During the first week, 10 l of extract per tonne of water is consumed daily; fertilization increases in the coming weeks from 20 l.m-3.day-1 in the second week to 30 l.m-3.day-1 in subsequent weeks. Vaccination of ponds is carried out on the fifteenth day at a rate of 10 daphnids per liter. One month after vaccination, flowers of more than 100 g.m-3 can be expected. To maintain water quality in these ponds, fresh hard water can be added at a maximum speed of 25% per day. Harvesting is carried out by concentrating daphnia on a seed of 500 µm. The extracted biomass is concentrated in a per fermented container (< 200 daphnids.l-1). To separate the daphnia from the unsteered substrates, exuviae and fecal material, the contents of the vessel are brought to a seed which is provided with a continuous circular water flow. Un fed particles, exuviae and feces are collected in the center at the bottom of the suction, while dafids remain in the water column. The unwanted material can then be removed using a pipette or suction pump. The harvest may be complete or partial; for partial harvesting, not more than 30 % of the permanent crop may be harvested daily. 6.1.4.5. Cultures of contamination Dafnie are often accidentally contaminated by rotifers. In particular, Brachionus, Conochilus and some bdelloids can be harmful (. B i.e. the commission of the investigation collection activities). Brachionus is simply removed from the culture by rinsing water and using a screen of suitable mesh size, since Dafnia is much larger than Brachionus. Conochilus, on the other hand, can be removed by adding cow droppings to the culture (reducing oxygen levels). Bdelloids are more difficult to remove from the culture because they are resistant to a wide range of environmental conditions and even drought. However, elimination is possible by creating strong water movements that bring bdelloids (which are inhabitants of the bottom) in the water column, and then remove them with the help of a network. 6.1.5. The production and use of resting eggs Resting eggs are an interesting material for the storage, transport and initiation of new dafnie cultures. The production of resting eggs can be started by exposing part of the Daphnia culture to a combination of stressful conditions such as low food availability, animal overcrowding, lower temperatures and short photoperiods. These conditions are generally acquired with an aging population at the end of the season. The collection of ephippia from the wild can be carried out by sampling sediments, rinsing them with a 200 µm sie way and isolating ephippia under a binocular microscope. Normally, these embryos remain at rest and require diapause inhibition to end this condition so they can hatch when conditions are optimal. Possible diapaus termination techniques are exposure to ephippia at low temperatures, darkness, oxygen and high concentrations of carbon dioxide for a minimum of several weeks (Davison, 1969). There's still no standard hatching for Daphnia. In general, the hatching process is stimulated by exposure to ephippi to higher temperatures (17-24 ° C), bright white light (70 W.m-2), longer photoperiods and high dissolved oxygen levels. However, it is important that these shocks are given while resting eggs are still in ephippium. After shock, eggs can be removed from the ephippia. Hatching will then take place after 1-14 days. 6.1.6. The use of Moina Moina also belongs to Cladocera and many biological and cultural properties that have been discussed for Dafnia can be used on Moina. Moina thrives in ponds and reservoirs, but above all inhabits temporary ponds or ditches. The period of achievement of reproductive maturity lasts four to five days at 26 ° C. At maturity, clear sexual dimorphic characteristics in the size of animals and antennula morphology can be observed. Males (0.6-0.9 mm) are smaller than females (1.0-1.5 mm) and have long grips that are used to hold the female during coulation. Sexually mature females carry only two eggs enclosed in ephippia, which is part of the dorsal exoskeleton. Moina is smaller in size than Daphnia, with a higher protein content and comparable economic value. Biomass is successfully used in rainbow trout, salmon, striped redfish and tropical fish hobbyists, who also use it in frozen form to feed more than sixty fresh and salty fish varieties. It has also been reported that partial replacement of moina micrura artemia has a positive effect during the larviculture of freshwater shrimp Macrobrachium rosenbergii (Alam, 1992). Enrichment of Moina can be done by direct method, cultivation on baker's yeast and emulsified fish or cuttlefish liver oils. Experiments have shown that Moina occupies (n-3) HUFA in the same way, albeit slower than rotifers and Artemia nauplii, reaching a maximum concentration of around 40% after a 24 h-feeding period. Period.

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