

ascertain whether the activated vegetable oil could be separated into similar fractions. A preliminary test showed that the unsaponifiable fraction of cotton seed oil was unable to protect rats which were placed on a low phosphorus rickets-producing diet. the same was found to be true in respect to linseed oil. When, however, irradiated linseed oil was fractioned, it was found that 0.1 gm. of the unsaponifiable fraction was sufficient to confer protection. The criterion of its efficacy was the absence of microscopic lesions at the costochondral junctions. Furthermore the blood of these rats contained more than twice as much inorganic phosphorus as the blood of similar rats which had received the non-irradiated oil. It would seem that irradiation of the vegetable oil had produced a substance similar in its properties to that in cod liver oil.

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Rate of metabolism and sex determination in Cladocera.

By ARTHUR M. BANTA and L. A. BROWN.

[*From the Laboratory of the Station for Experimental Evolution, Cold Spring Harbor, N. Y.*]

Cladocera reproduce mostly by parthenogenesis, and for long periods of time only female young may appear. Sometimes, however, males arise from parthenogenetic eggs, which eggs are indistinguishable from the parthenogenetic eggs ordinarily giving rise to females. At times sexual eggs appear and, if pairing occurs, these larger eggs pass into specially modified egg cases, the ephippia, within which they are cast off, undergo the earlier developmental stages, and enter a resting or latent period. This latent period is much prolonged (probably for months or even years with some forms) unless the eggs undergo desiccation or freezing or both. After desiccation or freezing if returned to water at 10° to 25° C. the sexual eggs develop into female young.

Experience in rearing several species of Cladocera lead the senior writer to conclude¹ that different environmental conditions are responsible for the occurrence of parthenogenetic females, or

¹ Banta, Arthur M., *PROC. SOC. EXP. BIOL. AND MED.*, 1916, xiv, 3.

of males, or of sexual eggs. In the collaborative work (most experiments were with *Moina macrocopa*) it was found that crowding the mothers caused the production of a variable percentage of males, while sisters given identical treatment, but uncrowded, produced only females. It was also found that CO₂ or uric acid treatments slightly increased the percentage of males in crowded or semi-crowded bottles. However by far the majority of treatments (including other excretory or related products), which altered the chemical condition of the medium in crowded or semi-crowded bottles, reduced male production. These experiments suggested the accumulation of excretory products as associated with male production.

Later, low temperature and chloretone were added to the list of treatments inducing male production. Adrenal cortex and alcohol (light dosages), on the other hand, eliminated or greatly reduced male production, whereas control bottles containing the same numbers of mothers produced characteristic male percentages.

The critical period at which the eggs in the ovary of a mother may be caused to develop either into females or males was found to be about two hours and to end about four hours before the egg leaves the ovary and passes into the brood chamber. This is approximately the time at which Kühn found that the maturation spindle is formed. Kühn² also found, as Weismann³ had done before him, that there is a single division without chromatic reduction in the maturation of the parthenogenetic egg. The presumption is strong that environmental conditions, in some manner, alter the chromatic behavior in the single maturation division and thus affect the sex of the forthcoming young Cladocera. According to Miss Taylor,⁴ however, the female and male have the same number of chromosomes (eight).

It was surmised and proven by direct observation and the accumulation of ample data that male production is associated with a lowered rate of metabolism of the mother during maturation of the eggs. This is true whether the male production is associated with crowding, low temperature, or any of the other conditions favoring male production. Hence it is assumed that the rate of metabolism, or some phenomenon associated with it, in

² Kühn, A., *Arch. Zellforsch.*, 1908, i, 1-50.

³ Weismann, A., *Zeit. wiss. Zool.*, 1876-1879, 27-33.

⁴ Taylor, M., *Zool. Anz.*, 1914, xlv, 21-24.

some manner alters the maturation division and thus determines the sex of the young. However it operates, we have found that lowered rate of metabolism is associated with male production in Cladocera; that agents which increase the rate of metabolism reduce or eliminate the occurrence of males; and that treatments which decrease the rate of metabolism induce the production of males. Further, within limits, the percentage of males produced is roughly proportional to the amount of retardation exhibited by the mothers.

Riddle⁵ has suggested that change in rate of metabolism may account for the results obtained by Whitney,⁶ Shull⁷ and others on control of sex in rotifers. It seems questionable whether the factor of *change* in rate of metabolism enters into the present case, if indeed it does in rotifers. It appears rather that a lower rate of metabolism is associated with male determination in the parthenogenetic eggs; and a higher rate of metabolism, such as is usual for ordinary laboratory cultures and in nature, is associated with female determination. It seems probable that also with rotifers, phylloxerans, aphids, paedogenetic diptera, and many other organisms, in which parthenogenesis alternates with gamogenesis, the production of males and the occurrence of sexual females are determined by rate of metabolism.

NOTE: Obreshkove, working in our laboratory, has found evidence that male Cladocera have a higher rate of metabolism than females, a fact previously known for the males of many forms, including man. In our present case *male causation* is associated with a lowered rate of metabolism at time of egg maturation *but the resultant males have the higher metabolic rate characteristic of males in general.*

⁵ Riddle, O., *Am. Nat.*, 1916, 1, 345-410.

⁶ Whitney, D. D., *J. Exp. Zool.*, 1914, xvii, 545-558.

⁷ Shull, A. F., and Ladoff, S., *J. Exp. Zool.*, 1916, xxi, 127-161.