AMER. ZOOL., 25:197–206 (1985)

Vitellogenesis and Its Control in Malacostracan Crustacea[1]

HÉLÈNE CHARNIAUX-COTTON

Laboratoire de Sexualité et Reproduction des Invertébrés, Université Pierre et Marie Curie, 75230 Paris, Cedex 05, France

SYNOPSIS. Vitellogenesis in the amphipod *Orchestia gammarella,* in natantian decapods and probably in other Malacostraca involves two phases, primary and secondary vitellogenesis. Oogenesis from oogonia to the end of primary vitellogenesis is a continuous process. The primary proteic vitellus is endogenous. The secondary vitellogenesis takes place during the reproductive season. Fully grown primary follicles are surrounded by a permanent follicular tissue (secondary folliculogenesis). The resulting secondary follicles grow synchronously until ovulation. A tubular network has been observed in secondary follicle cells of prawns. The prominent feature of secondary vitellogenesis is the uptake of vitellogenin by the oocytes. This vitellogenin is synthesized by the fat body. Secondary vitellogenesis is controlled by neurohormones that have not yet been isolated and whose modalities of action still remain unclear. In Decapoda, the classical gonad-inhibiting hormone or GIH released by the sinus glands inhibits secondary vitellogenesis and the synthesis of vitellogenin. In Isopoda, vitellogenesis-inhibiting hormone is synthesized by neurosecretory cells in the median part of the protocerebrum. In Amphipoda, neurosecretory cells of this region secrete a neurohormone inducing secondary folliculogenesis and triggering secondary vitellogenesis. The neurohormones would control the formation of functional secondary follicles during the reproductive season.

Other hormones are involved in control of secondary vitellogenesis. In Peracarida molting hormone is necessary for a normal synthesis of vitellogenin and the uptake of this protein by growing oocytes. In the amphipod *Orchestia gammarella* secondary folliculogenesis requires a low titer of ecdysteroids which characterizes postecdysis. The correlation between molt cycle and secondary vitellogenesis would be established in this way. After secondary folliculogenesis, the secondary follicle cells become endocrine and secrete the vitellogenin-stimulating ovarian hormone (VSOH).

INTRODUCTION

Malacostracan Crustacea continue to molt after reaching puberty. Only some crabs and most Oxyrhyncha, as in the Insecta, become sexually mature after a terminal molt. In many malacostracans, chiefly in peracarids and natantian decapods, spawning is obligatorily preceded by a molt.

During the period of genital activity, the number of spawnings varies according to the species. Except for penaeid prawns, malacostracan Crustacea incubate their eggs, and the time interval between two spawnings is longer than the time of egg incubation. In many decapods, the number of incubated eggs is about several thousands, and in species which lay few eggs (about twenty in *Orchestia gammarella*), the oocytes grow to a large size; therefore, the reproduction of malacostracans requires a large amount of yolk.

Beams and Kessel (1963) carried out the first electron microscope studies on the origin of yolk in crayfish oocytes. They observed proteinaceous yolk in the cisternae of the ooplasmic granular endoplasmic reticulum. They concluded—as others did later—that the oocyte is capable of synthesizing the massive store of yolk present in the egg at the end of oogenesis. It is now well established that the origin of yolk is dual, intra-ooplasmic and extra-ooplasmic. As in insects, the fat body of malacostracans synthesizes a precursor (vitellogenin) of the major egg yolk protein (vitellin). It may be possible that the amount of proteinaceous yolk required for each spawning is greater than each source could produce alone. During the first stage or primary vitellogenesis, the vitellins are endogenous. During the second stage, or secondary vitellogenesis, oocytes take up vitellogenin by endocytosis.

In this paper, special attention is devoted to the phenomenon of secondary follicu-

[1] From the Symposium on *Advances in Crustacean Endocrinology* presented at the Annual Meeting of the American Society of Zoologists, 27–30 December 1983, at Philadelphia, Pennsylvania.

logenesis (Charniaux-Cotton, 1974) that ensures the synchronous growth of oocytes during secondary vitellogenesis and which appears peculiar to malacostracans.

Experimental data have been obtained concerning endocrine control of vitellogenin synthesis and of secondary vitellogenesis. Although in Insecta the hormonal control of vitellogenin gene expression is well investigated (review in Chen and Hillen, 1983), in Malacostraca, to my knowledge, no comparable study has been published. Data concerning the endocrine mechanisms which connect molting and secondary vitellogenesis are not yet available. Some recent reviews are related to vitellogenesis and its control (Charniaux-Cotton, 1978, 1980; Meusy, 1980; Blanchet-Tournier, 1982; Legrand *et al.*, 1982; Meusy and Charniaux-Cotton, 1984).

Stages of Oogenesis

The various stages of oogenesis in Malacostraca have been mainly studied in the amphipod, *Orchestia gammarella,* and in some natantian decapods (Zerbib, 1980; review in Charniaux-Cotton, 1980).

From oogonia to the end of primary vitellogenesis

Oogonia are located in the germinative zone, a longitudinal structure which lasts the whole life of the female and each oogonium is completely surrounded by mesodermal cells. Oogonial mitosis takes place exclusively in the germinative zone. Some oogonia continually leave this zone by an unknown mechanism and undergo meiotic prophase up to diakinesis (they are then primary oocytes).

I have called the next stage of oogenesis previtellogenesis. Ribosomes are formed at a high rate and a rough endoplasmic reticulum (RER) develops in primary oocytes. Primary follicle cells, which probably originate from the germinative zone, surround each oocyte (Rateau and Zerbib, 1978).

The RER vesicles become numerous and filled with a granular material, the primary yolk; Dhainaut and De Leersnyder (1976) have called this vitellogenesis primary vitellogenesis. Histochemical studies performed by these authors in the crab *Eriocher sinensis* and by Zerbib (1976) in *O. gammarella* indicated that this material is formed by glycoproteins. Oocytes grow to a maximal size which is typical for the species.

Oogenesis from oogonia to the end of primary vitellogenesis (Fig. 1A) is a continuous process whose possible variations with the seasons have not been reported.

Secondary vitellogenesis and secondary folliculogenesis

Secondary vitellogenesis is characterized by the synchronous growth of the oocytes to a large size through endocytotic uptake of vitellogenin. The RER continues to synthesize glycoproteins which are immunochemically different from vitellogenin (see Meusy, 1980), and lipid droplets appear at the periphery of ooplasma and invade it. Each oocyte is surrounded by a single layer of secondary follicle cells, and oocyte microvilli cross the vitelline layer.

At the end of secondary vitellogenesis, the microvilli retract. The breakdown of the germinal vesicle (egg nucleus) occurs some hours before ecdysis in *O. gammarella* (Mathieu-Capderou, 1980) and in the prawn *Palaemon serratus* (Clédon, unpublished data).

In *O. gammarella* (Charniaux-Cotton, 1974), *Palaemonetes argentinus* (Schuldt, 1980), *Macrobrachium rosenbergii* (Fauvel, 1981), and the crab *Cancer pagurus* (Charniaux-Cotton, unpublished data), the secondary follicle tissue does not degenerate after ovulation. In histological sections, it appears as a folded epithelium bordered by a basal lamina (Fig. 1A), and it collapses around each fully grown primary follicle. The mechanisms of tissue movements and of primary follicle cell integration into a one layer secondary epithelium are not known. During genital rest, the oocytes remain blocked at the end of primary vitellogenesis but during the reproductive period, the oocytes of newly constituted secondary follicles undergo synchronously secondary vitellogenesis (Fig. 1B).

Secondary folliculogenesis, which is probably a general phenomenon in Malacostraca, separates the oocytes into two populations: (1) oocytes surrounded by pri-

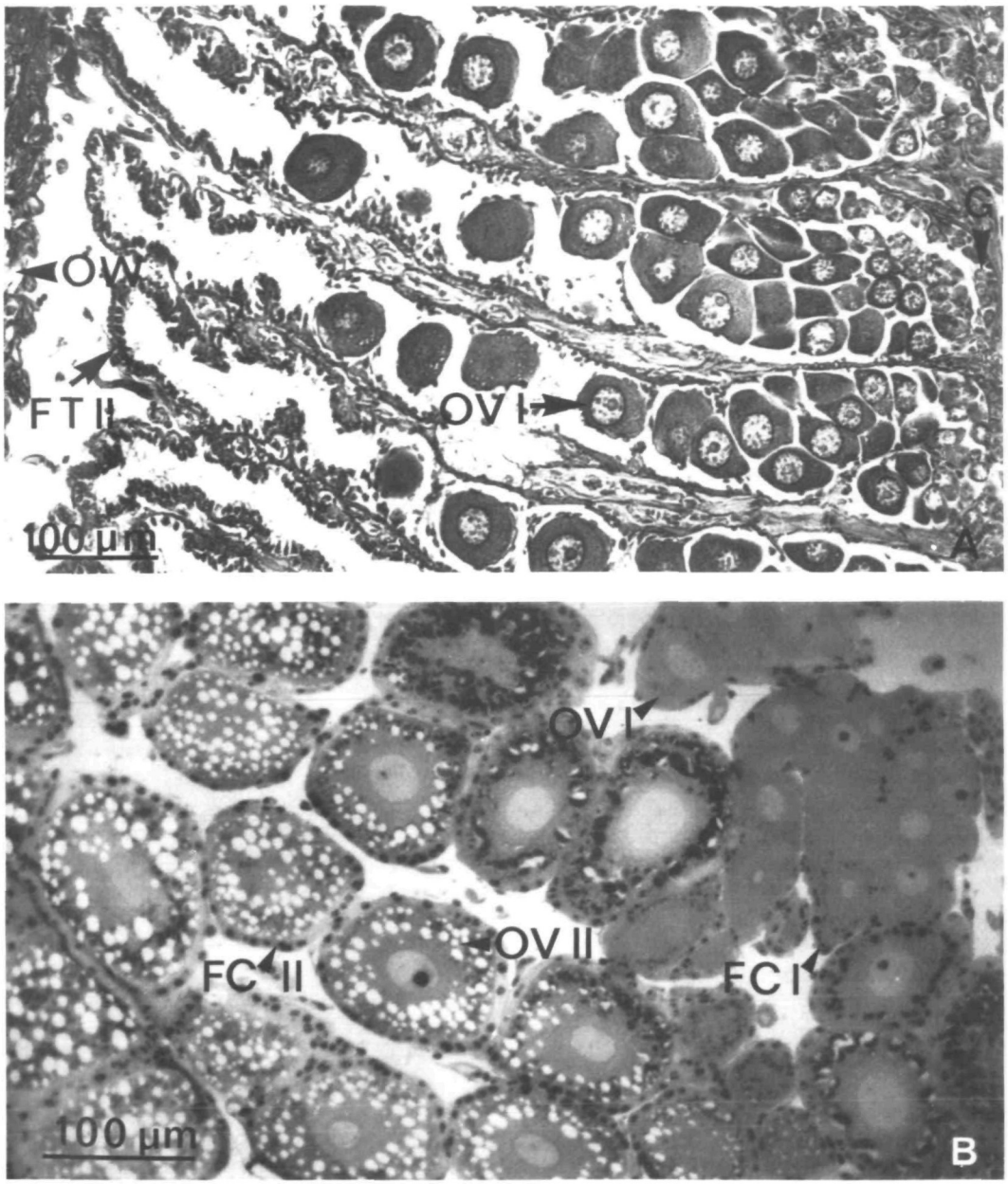

FIG. 1. *Macrobrachium rosenbergii.* Section of ovary. A. Ovary after ovulation. The secondary follicle envelope (FT II) is empty. All stages of oogenesis from oogonia to the end of the primary vitellogenesis are present. B. Ovary after secondary folliculogenesis. All the oocytes in secondary vitellogenesis have the same size. FC I: primary follicle cells; FC II: secondary follicle cells; G: oogonia; OV I: oocyte in primary vitellogenesis; OV II: oocyte in secondary vitellogenesis; OW: ovarian wall.

mary follicle cells and stratified in zones corresponding to the different stages of oogenesis up to the end of primary vitellogenesis and (2) oocytes, surrounded by secondary follicle cells, all of which are at the same stage of secondary vitellogenesis. It is a strategy which allows for the accumulation of fully grown primary follicles

during both the period of genital rest and the time when secondary vitellogenesis is occurring in oocytes of secondary follicles and for synchronous development of these oocytes. I have observed that the secondary follicle tissue is not developed in juvenile females of *O. gammarella, Palaemon serratus, Carcinus maenas.*

Tubular Network of Secondary Follicle Cells

In our laboratory, Jugan and Zerbib (1984) have described a tubular network in the secondary follicle cells of *Macrobrachium rosenbergii* (Fig. 2). The tubules (0.15 μm in diameter) open into all extracellular spaces around the follicle cells. They are filled with a finely granular electron dense material. Some oocyte microvilli penetrate deeply into the tubule lumen. After incubation in peroxidase, the visualization of this protein by electron dense reaction product in the follicle intercellular spaces, in tubular network, in vitelline envelope and in pinocytotic vesicles, suggests that the tubular structure could be implicated in the transportation of vitellogenin from the haemolymph to the oocytes. This structure is not visible after ovulation, during the secondary folliculogenesis and during genital rest. The tubular network appears as a characteristic feature of secondary follicle cells during secondary vitellogenesis.

A tubular network has also been observed in the prawns *Palaemonetes varians* and *Palaemon serratus* (Jugan, unpublished), and Arcier and Brehelin (1982) mentioned tubules in follicle cells of *Palaemon adspersus.* According to these authors, the tubules communicate only with the space between basal lamina and follicle cells and would take a part in endocrine secretion.

Vitellogenin and Its Synthesis

The existence of a female specific protein (vitellogenin) was reported in haemolymph of several species of malacostracans (review in Meusy, 1980). This substance is a lipoglycocarotenoprotein, immunologically not distinguishable from the major egg yolk protein (or vitellin).

By using tritiated vitellogenin in *O. gammarella,* Zerbib and Mustel (1984) obtained proof that secondary follicle oocytes take up the vitellogenin by endocytosis during the reproductive period.

The fat body has been shown to be the site of vitellogenin synthesis in the Isopod *Porcellio dilatatus* (Picaud and Souty, 1980), in *O. gammarella* (Croisille and Junéra, 1980), and in *Palaemon serratus* (Meusy *et al.,* 1983*a*). Immunoenzymatic techniques and injection of tritiated leucine indicate that the fat body of *O. gammarella* and *P. serratus* contains and releases vitellogenin only when the ovaries are in secondary vitellogenesis (Croisille and Junéra, 1980; Meusy *et al.,* 1983*a, b*). In *O. gammarella* (Meusy *et al.,* 1974), *P. dilatatus* (Picaud and Souty, 1981) and *Idotea balthica basteri* (Souty and Picaud, 1981), where secondary vitellogenesis and molt cycle are correlated, vitellogenin synthesis is low at the beginning and at the end of these cycles.

Control of Vitellogenin Synthesis

Some results concerning the control of vitellogenin synthesis have been obtained in Peracarida, which differ between Amphipoda and Isopoda. Few data are available on Decapoda.

Amphipoda

In *O. gammarella,* experiments have proved the existence of a vitellogenin-stimulating ovarian hormone (VSOH) (Junéra *et al.,* 1977). After they removed androgenic glands from males, spermatogenesis stopped but oogenesis and vitellogenin synthesis did not take place. An ovary implanted in these andrectomized males triggered vitellogenin synthesis. In a complementary experiment, after removal of the ovaries from vitellogenic females, vitellogenin synthesis stopped after about 5–8 days but synthesis was restored by implanting an ovary. The secondary follicle cells are probably the source of VSOH. It must be recalled that the ovaries of *O. gammarella* control the formation of oostegites

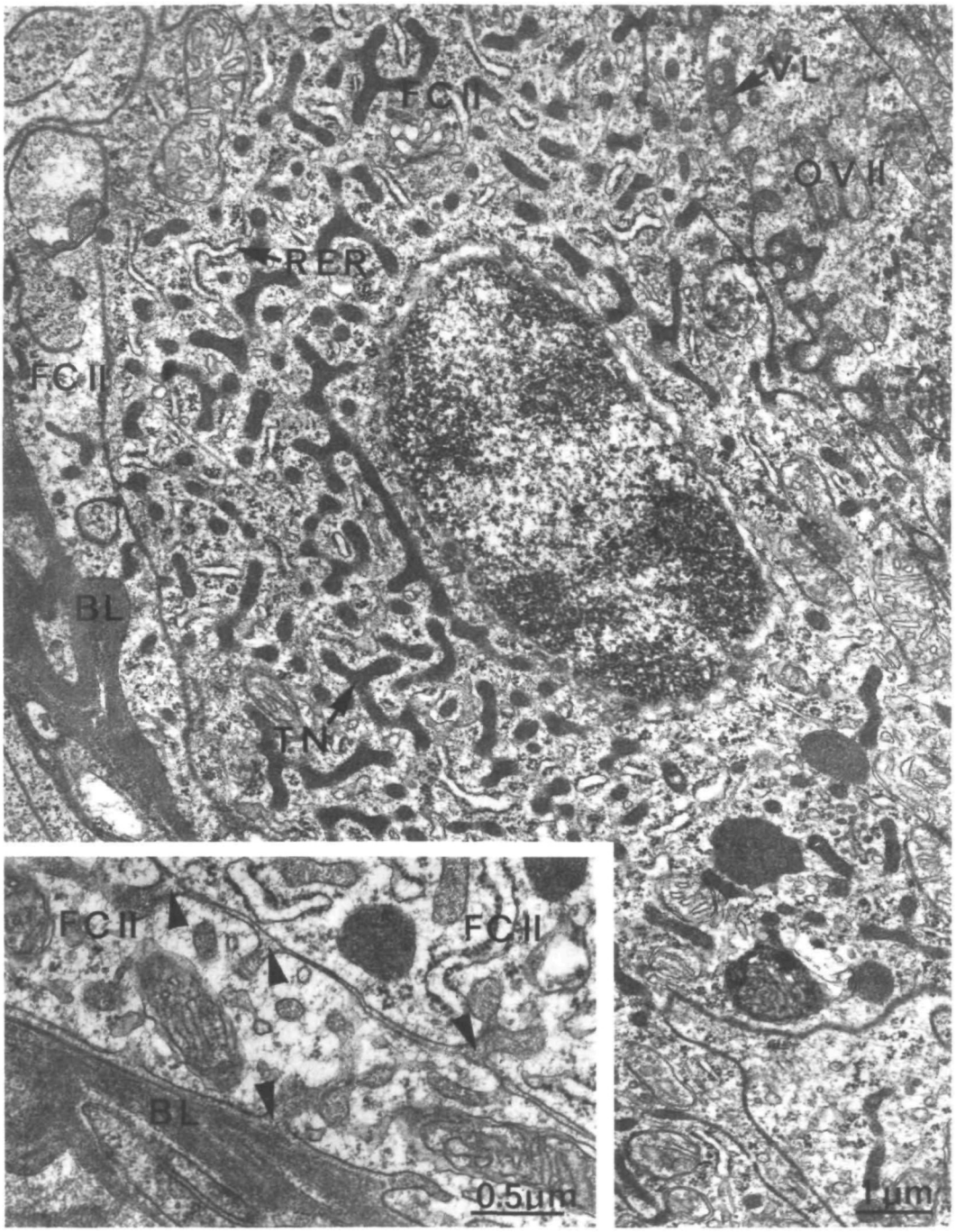

FIG. 2. *Macrobrachium rosenbergii.* Electron micrographs of secondary follicle cells during secondary vitellogenesis. The tubular network (TN) is well developed. The arrows in the insert show the openings of the tubular network into the extracellular space. BL: basal lamina; FC II: secondary follicle cell; OV II: oocyte in secondary vitellogenesis; RER: rough endoplasmic reticulum; VL: vitelline layer. (From P. Jugan and C. Zerbib, 1984.)

and ovigerous setae (Charniaux-Cotton, 1957).

Cauterization experiments of ecdysone-producing glands (Y-organs) in *O. gammarella* show that molting hormone (ecdysteroid) is necessary for vitellogenin synthesis (Meusy *et al.*, 1977; Blanchet-Tournier, 1982). So, in *O. gammarella,* the simultaneous presence of both ovarian and molt hormone is necessary for vitellogenin synthesis. Consequently, VSOH is different from 20-OH-ecdysone (ecdysteroid). Another proof is that, during genital rest, the fat body does not synthesize vitellogenin although the molt cycles continue (Meusy *et al.*, 1974, 1983*a, b*).

Isopoda

In *Porcellio dilatatus,* ovariectomy experiments and incubation of male fat body show that vitellogenin synthesis is not dependent on an ovarian hormone. It is regulated by a feed-back mechanism in females and inhibited by androgenic hormone in males (Picaud, 1983; Gohar and Souty, 1983).

In Isopods, as in *O. gammarella,* molting hormone is necessary for vitellogenin synthesis (Souty *et al.*, 1982; Picaud, 1983).

Decapoda

In the female prawn *Palaemon serratus,* there is no vitellogenin synthesis during genital rest. Removal of eyestalks (source of the well-known gonad-inhibiting hormone or GIH) triggers vitellogenin synthesis, but it is not known whether the target organ of GIH is the ovary or the fat body (Meusy *et al.*, 1983*a*).

No data are available on relations between vitellogenin synthesis and the molt hormone. Oxyrhynchans and some species of brachyrhynchans enter a terminal anecdysis at the puberty molt. Vitellogenin synthesis occurs after this last molt, that is to say, while the concentration of ecdysteroids in haemolymph is low (see Soummoff and Skinner, 1983). So, in some decapods, as in most insects, vitellogenin synthesis appears not to be dependent on high levels of molting hormone.

CONTROL OF SECONDARY VITELLOGENESIS

Gonad-inhibiting hormone

Panouse (1943) discovered that *Palaemon serratus* females in genital rest develop their ovaries after eyestalk or sinus gland removal. If the operation is performed at the beginning of a molt cycle, oviposition occurs after the next ecdysis, but if the females are operated on later in the cycle, spawning occurs only after the second post-operative ecdysis. Eyestalk removal has no such effect in juvenile females (Panouse, 1946). Later, the sinus gland was demonstrated as a neurohormone-releasing organ; the neurohormone-inhibiting ovarian activity has been called gonad-inhibiting hormone or GIH (see review in Charniaux-Cotton and Kleinholz, 1964).

We have observed in prawns that GIH is responsible for genital rest by inhibiting secondary vitellogenesis (Charniaux-Cotton, 1978, 1980). During the reproductive season, GIH can modulate the rate of secondary vitellogenesis. For example, in *P. serratus,* secondary vitellogenesis lasts three molt cycles during winter in Brittany. If the eyestalk removal is performed during the first or second molt cycle, egg laying occurs at the post-operative ecdysis (Faure *et al.*, 1981). Klek-Kawinska and Bomirski (1975) pointed out that the "ovary-inhibiting hormone activity" decreases during the annual reproductive cycle of female *Crangon crangon.* The manner in which GIH controls secondary vitellogenesis is unknown. Juvenile females do not respond to destalking, probably because of the non-development of secondary follicle tissue. Some experiments performed on *P. serratus* indicate that the GIH secretion might be controlled by the medulla externa X-organ (MEX); after MEX removal, secondary vitellogenesis stops (Faure *et al.*, 1981).

Partial purification of the gonad-inhibiting hormone

Until now, three attempts at purification of GIH have been described.

(1) From eyestalks of male *Cancer magister* with female *Crangon crangon* as test ani-

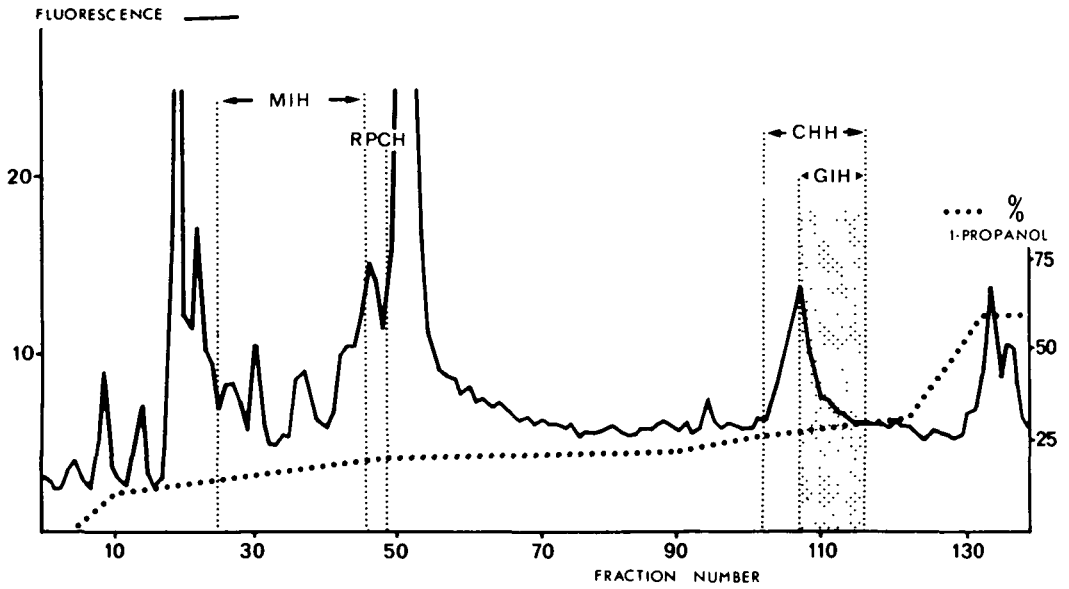

FIG. 3. HPLC filtration of hydrochloric extracts from 40 *Homarus gammarus* sinus glands. CHH: crustacean hyperglycemic hormone; GIH: gonad-inhibiting hormone; MIH: molt-inhibiting hormone; RPCH: red-pigment concentrating hormone. *Column:* Spherisorb 10 ODS. *Elution gradient:* formic acid 0.5 *M*–pyridine 0.14 *M* (pH 3.0)/1-propanol. (From J. E. van Deynen, D. Soyez, and F. van Herp, unpublished data.)

mals, the hormone is described as having a mol wt of approximately 2,000. (Bomirski *et al.*, 1981).

(2) From eyestalks of the spiny lobster *Panulirus argus* with the crab *Uca pugilator* as test animals, the hormone is attributed a mol wt of about 5,000 (Quackenbush and Herrnkind, 1983).

(3) From sinus gland of *Homarus gammarus* with *Atyaephyra desmaresti* as test animals, the mol wt is above 5,000 (Van Deynen, Soyez and Van Herp, unpublished data).

The inhibition of ovarian activity in eyestalkless females by eyestalk extracts of *Cancer* and *Panulirus* was evaluated using ovarian weight. Weight probably is not a good criterion because, in eyestalkless females, oogenesis, up to the end of primary vitellogenesis, is probably modified by the absence of hormones other than GIH.

The activity of sinus glands from *Homarus* was bioassayed using the diameter of oocytes in secondary vitellogenesis. As seen above, growth of these oocytes is controlled by GIH and therefore oocyte size is a good parameter for estimating gonad-inhibiting activity.

Separation of GIH and three other hormones from sinus glands of *Homarus* by filtration by Reversed Phase HPLC has been performed (Fig. 3). The gonad-inhibiting activity has been eluted in fractions showing also crustacean hyperglycemic hormone (CHH) activity. This zone is far from those containing the molt-inhibiting hormone (MIH) and red-pigment concentrating hormone (RPCH).

Other gonadotropic hormones

A gonad-inhibiting hormone appears to be released by the median zone of the protocerebrum in Isopoda since removal of this zone triggers vitellogenesis (see review in Legrand *et al.*, 1982).

In *O. gammarella*, this zone would release a secondary folliculogenesis-stimulating hormone (Blanchet-Tournier, 1982). Implantation of thoracic ganglion into immature female of the crabs *Potamon* and *Paratelphusa* and of the spider crab *Libinia*,

supports the existence of a gonad-stimulating hormone in this ganglion (Adiyodi and Adiyodi, 1970; Hinsch and Bennet, 1979).

Ecdysteroid titers in the haemolymph

In *O. gammarella* and Isopoda, the presence of molting hormone is necessary not only for vitellogenin synthesis but also for secondary vitellogenesis since secondary vitellogenesis stops after Y-organ cauterization (see review in Blanchet-Tournier, 1982). On the contrary, in Oxyrhyncha, secondary vitellogenesis occurs in the absence of ecdysteroids, after the terminal molt (puberty molt).

In species where secondary vitellogenesis and molt cycle are synchronous, the correlation persists after destalking (Charniaux-Cotton and Touir, 1973; Faure *et al.*, 1981), probably being regulated by the pattern of ecdysteroid titers during the molt cycle. The low titer at the beginning of the molt cycle is presumably responsible for the onset of secondary vitellogenesis. Actually, introduction of 20-OH-ecdysone into a female just after ecdysis inhibits the onset of secondary vitellogenesis in the prawn *Lysmata seticaudata* and in *O. gammarella* (Touir and Charniaux-Cotton, 1974; Blanchet *et al.*, 1975; Blanchet-Tournier, 1982).

Discussion

Secondary vitellogenesis takes place only during the reproductive season and leads to spawning. This complex phenomenon consists of several processes; uptake by the secondary oocytes of vitellogenin synthesized by the fat body, continuation of the endogenous vitellogenesis (of varying import according to the species), entrance of lipids, roles of follicle cells.

The existence of a permanent secondary follicular tissue seems peculiar to malacostracan Crustacea. The follicular tissue recognizes the fully grown primary follicles and surrounds them, constituting the secondary follicles. In *Orchestia gammarella,* it acquires an endocrine function and secretes the vitellogenin-stimulating ovarian hormone (VSOH) and the hormone inducing the temporary breeding characters. In prawns, a tubular network appears inside the secondary follicular cells; this structure is probably the route for vitellogenin from the hemolymph to the oocytes. At the end of secondary vitellogenesis, the follicular tissue ensures ovulation by means of its own movements.

The endocrine systems controlling secondary vitellogenesis are still not well known and seem to be different among the various crustacean orders. In Decapoda, the Gonad-Inhibiting Hormone or GIH—released by the sinus gland—inhibits secondary vitellogenesis during the rest season. In *Palaemon serratus,* the inhibitory effect of the GIH on vitellogenin synthesis has pointed out. The mechanisms of actions of the GIH remain unclear, as does the nature of the target organ (ovary, fat body?). Several attempts to purify GIH have been undertaken. The occurrence of an ovarian hormone is not yet demonstrated. In the shrimp *Lysmata seticaudata* where secondary vitellogenesis and molt cycle are synchronous, secondary vitellogenesis takes place during the reproductive season—that is to say in the absence of GIH—at the beginning of each molting cycle. This event seems related to the low level of ecdysteroids present at this time.

In the amphipod *Orchestia gammarella,* secondary vitellogenesis takes place during each molting cycle during the reproductive season. The occurrence of a secondary vitellogenesis-inhibiting neurohormone has not yet been proved. On the other hand, a hormone from the protocerebrum is required for secondary folliculogenesis. This folliculogenesis would be also determined by a low level of ecdysteroids, a characteristic of the postecdysis stage. The secondary follicular cells acquire an endocrine function and release VSOH; but another hormone is necessary to vitellogenin synthesis, the molting hormone. The latter seems to be involved also in vitellogenin uptake since this process stops after removal of the molt gland, the Y-organ.

In Isopoda, the occurrence of a vitellogenic-inhibitory hormone originating in the protocerebrum has been proved. Vitellogenin synthesis is not ovary dependent. After ovariectomy, the level of hemolym-

phatic vitellogenin increases as a consequence of a feed-back mechanism. As in amphipods, 20-OH ecdysone is required for vitellogenesis.

The hormones controlling the secondary vitellogenesis in malacostracan Crustacea are incompletely known. Except for the 20-OH ecdysone, not one has been isolated.

REFERENCES

Adiyodi, K. G. and R. G. Adiyodi. 1970. Endocrine control of reproduction in decapod Crustacea. Biol. Rev. 45:121–165.

Arcier, J. M. and M. Brehelin. 1982. Etude histologique et ultrastructurale du tissu folliculaire au cours des cycles de développement ovarien chez *Palaemon adspersus* (Rathke, 1837). Arch. Biol. 93:79–97.

Beams, H. W. and R. G. Kessel. 1963. Electron microscope studies on developing crayfish oocytes with special reference to the origin of yolk. J. Cell. Biol. 18:621–649.

Blanchet-Tournier, M. F. 1982. Quelques aspects des interactions hormonales entre la mue et la vitellogenèse chez le Crustacé Amphipode *Orchestia gammarellus* (Pallas). Reprod. Nutr. Dévelop. 22: 325–344.

Blanchet, M. F., H. Junéra, and J. J. Meusy. 1975. La mue et la vitellogenèse d'*Orchestia gammarellus* (Crustacé Amphipode): étude de la synthèse de la fraction protéique femelle après implantation de cristaux d'ecdystérone. Experientia 31:865–867.

Bomirski, A., M. Arendarczyk, E. Kawinska, and L. H. Kleinholz. 1981. Partial characterization of crustacean gonad-inhibiting hormone. Int. J. Invert. Reprod. 3:213–220.

Charniaux-Cotton, H. 1957. Croissance, régénération et déterminisme endocrinien des caractères sexuels d'*Orchestia gammarella* Pallas (Crustacé Amphipode). Ann. Sci. Nat. 19:411–560.

Charniaux-Cotton, H. 1974. Données nouvelles concernant la vitellogenèse des Crustacés Malacostracés obtenues chez l'Amphipode *Orchestia gammarellus* (Pallas): folliculogenèse à partir d'un tissu permanent; action du busulfan; action inhibitrice de l'hormone juvénile. C.R. Acad. Sci. Paris 279: 563–566.

Charniaux-Cotton, H. 1978. L'ovogenèse, la vitellogénine et leur contrôle chez le Crustacé Amphipode *Orchestia gammarellus* (Pallas). Comparaison avec d'autres Malacostracés. Arch. Zool. Exp. Gén. 119:365–397.

Charniaux-Cotton, H. 1980. Experimental studies of reproduction in malacostraca Crustaceans. Description of vitellogenesis and of its endocrine control. *In* W. H. Clark, Jr. and T. S. Adams (eds.), *Advances in invertebrate reproduction*, pp. 177–186. Elsevier, North Holland.

Charniaux-Cotton, H. and L. H. Kleinholz. 1964. Hormones in invertebrates other than insects. *In* G. Pincus, K. V., Thimann, and E. B. Astwood (eds.), *The hormones*, Vol. 4, pp. 135–198. Academic Press, New York.

Charniaux-Cotton, H. and A. Touir. 1973. Contrôle de la prévitellogenèse et de la vitellogenèse chez la Crevette hermaphrodite *Lysmata seticaudata* Risso. C.R. Acad. Sci. Paris 276:2717–2720.

Chen, T. T. and L. J. Hillen. 1983. Review article: Expression of the vitellogenin genes in insects. Gamete Research 7:179–196.

Croisille, Y. and H. Junera. 1980. Recherche du lieu de synthèse de la vitellogénine chez le Crustacé Amphipode *Orchestia gammarella* (Pallas). Démonstration, à l'aide d'anticorps spécifiques, de la présence de la vitellogénine dans le tissu adipeux sous-épidermique des femelles en vitellogenèse secondaire. C.R. Acad. Sci. Paris 290: 1487–1490.

Dhainaut, A. and M. De Leersnyder. 1976. Etude cytochimique et ultrastructurale de l'évolution ovocytaire du Crabe *Eriocher sinensis*. I. Ovogenèse naturelle. Arch. Biol. 87:261–282.

Faure, Y., C. Bellon-Humbert, and H. Charniaux-Cotton. 1981. Folliculogenèse et vitellogenèse secondaires chez la Crevette *Palaemon serratus* (Pennant); contrôle par les pédoncules oculaires et l'organe X de la medulla externa (MEX). C.R. Acad. Sci. Paris 293:461–466.

Fauvel, C. 1981. Etude de l'ovaire de la crevette d'eau douce *Macrobrachium rosenbergii* (de Man) au cours du cycle de reproduction. Première description de la folliculogenèse secondaire chez un Crustacé Décapode. C.R. Acad. Sci. Paris 292: 547–552.

Gohar, M. and C. Souty. 1983. Mise en évidence *in vitro* d'une synthèse et d'une libération de vitellogénine dans le tissu adipeux mâle de *Procellio dilatatus* Brandt (Crustacé Isopode terrestre). C.R. Acad. Sci. Paris 297:145–148.

Hinsch, G. W. and D. C. Bennet. 1979. Vitellogenesis stimulated by thoracic ganglion implants into destalked immature spider crabs, *Libinia emarginata*. Tissue and Cell 11:345–351.

Jugan, P. and C. Zerbib. 1984. Follicle cell tubular system in the prawn *Macrobrachium rosenbergii*: A route for exchanges between haemolymph and vitellogenic oocytes? Biol. Cell. 51:395–398.

Junéra, H., C. Zerbib, M. Martin, and J. J. Meusy. 1977. Evidence for control of vitellogenin synthesis by an ovarian hormone in *Orchestia gammarellus* (Pallas), Crustacea Amphipoda. Gen. Comp. Endocrinol. 31:457–462.

Klek-Kawinska, E. and A. Bomirski. 1975. Ovary inhibiting hormone activity in shrimp (*Crangon crangon*) eyestalks during the annual reproductive cycle. Gen. Comp. Endocrinol. 25:9–13.

Legrand, J. J., G. Martin, P. Juchault, and G. Besse. 1982. Contrôle neuroendocrine de la reproduction chez les Crustacés. J. Physiol. Paris 78:543–552.

Mathieu-Capderou, C. 1980. Relation entre la maturation ovocytaire et l'exuviation chez le Crustacé Amphipode *Orchestia gammarellus* (Pallas). C.R. Acad. Sci. Paris 290:1495–1498.

Meusy, J. J. 1980. Vitellogenin, the extraovarian precursor of the protein yolk in Crustacea: A review. Reprod. Nutr. Develop. 20:1–21.

Meusy, J. J., M. F. Blanchet, and H. Junéra. 1977. Mue et vitellogenèse chez le Crustacé Amphipode *Orchestia gammarellus* Pallas. II. Etude de la synthèse de la vitellogénine ("fraction protéique femelle" de l'hémolymphe) après destruction des organes Y. Gen. Comp. Endocrinol. 33:35–40.

Meusy, J. J. and H. Charniaux-Cotton. 1984. Endocrine control of vitellogenesis in malacostraca Crustaceans. *In* W. Engels *et al.* (eds.), *Advances in invertebrate reproduction* 3. Elsevier Science Publishers B.V.

Meusy, J. J., H. Junéra, P. Clédon, and M. Martin. 1983*a*. La vitellogénine chez un Crustacé Décapode Natantia, *Palaemon serratus* Pennant. Mise en évidence, comparaison immunologique avec les vitellines, site de synthèse et rôle des pédoncules oculaires. Reprod. Nutr. Dévelop. 23:625–640.

Meusy, J. J., H. Junéra, and Y. Croisille. 1974. Données sur la synthèse de la fraction protéique femelle chez *Orchestia gammarellus* (Pallas) (Crustacé Amphipode) au cours de l'intermue et chez les femelles en repos sexuel. C.R. Acad. Sci. Paris 279:587–590.

Meusy, J. J., C. Zerbib, F. Dacheux, and M. P. Dubois. 1983*b*. Subcellular localization of vitellogenin in crustacean adipocytes by the unlabelled antibody enzyme method. Tissue and Cell 15:301–310.

Panouse, J. B. 1943. Influence de l'ablation du pédoncule oculaire sur la croissance de l'ovaire chez la Crevette *Leander serratus*. C.R. Acad. Sci. Paris 217:553–555.

Panouse, J. B. 1946. Recherches sur les phénomènes humoraux chez les Crustacés. L'adaptation chromatique et la croissance ovarienne chez la Crevette *Leander serratus*. Ann. Inst. Oceanogr. Monaco 23:65–147.

Picaud, J. L. 1983. Les protéines spécifiques femelles des crustacés isopodes. Constitution, synthèse et certains aspects du contrôle de leur synthèse et de leur libération. Univ. Poitiers, Fac. des Sciences Fondamentales et Appliquées.

Picaud, J. L. and C. Souty. 1980. Démonstration par immunoautoradiographie de la synthèse de la vitellogénine par le tissu adipeux de *Porcellio dilatatus* Brandt (Crustacé Isopode). C.R. Acad. Sci. Paris 290:1019–1021.

Picaud, J. L. and C. Souty. 1981. Approche quantitative de l'influence de l'ovariectomie sur la synthèse de la vitellogénine chez *Porcellio dilatatus* Brandt (Crustacé Isopode). C.R. Acad. Sci. Paris 293:479–482.

Quackenbush, L. S. and W. F. Herrnkind. 1983. Partial characterization of eyestalk hormones controlling molt and gonadal development in the spiny lobster *Panulirus argus*. J. Crust. Biol. 3:34–44.

Rateau, J. G. and C. Zerbib. 1978. Etude ultrastructurale des follicules ovocytaires chez le Crustacé Amphipode *Orchestia gammarellus* (Pallas). C.R. Acad. Sci. Paris 286:65–68.

Schuldt, M. 1980. La estructura ovarica de *Palaemonetes argentinus* Nobili, 1901 en relacion con aspectos actuales de la morfologia functional en crustaceos superiores. Neotropica 26:155–162.

Soumoff, C. and D. M. Skinner. 1983. Ecdysteroid titers during the molt cycle of the blue crab resemble those of other crustacea. Biol. Bull. 165: 321–329.

Souty, C. and J. L. Picaud. 1981. Vitellogenin synthesis in the fat body of the marine crustacean Isopoda, *Idotea balthica basteri*, during vitellogenesis. Reprod. Nutr. Develop. 21:95–101.

Souty, C., G. Besse, and J. L. Picaud. 1982. Stimulation par l'ecdysone du taux hémolymphatique de la vitellogénine chez le Crustacé isopode terrestre *Porcellio dilatatus* Brandt. C.R. Acad. Sci. Paris 294:1057–1059.

Touir, A. and H. Charniaux-Cotton. 1974. Influence de l'introduction d'ecdystérone sur l'exuviation et le démarrage de la vittelogenèse chez la Crevette *Lysmata seticaudata* Risso. C.R. Acad. Sci. Paris 278:119–122.

Van Deijnen, J. E., D. Soyez, and F. Van Herp. 1984. Simultaneous purification and preliminary characterization of some neurohormones: CHH (Crustacean Hyperglycemic Hormone), RPCH (Red Pigment Concentrating Hormone), GIH (Gonad Inhibiting Hormone) and MIH (Moult Inhibiting Hormone) of the sinus gland in the lobster (*Homarus americanus*). Annls. Soc. R. Zool. Belg. 114 suppl. (1):176–177.

Zerbib, C. 1976. Nature chimique des enclaves vitellines de l'ovocyte du Crustacé Amphipode *Orchestia gammarellus* (Pallas). Ann. Histochim. 21:279–295.

Zerbib, C. 1980. Ultrastructure observation of oogenesis in the Crustacea Amphipoda *Orchestia gammarellus* (Pallas). Tissue and Cell 12:47–62.

Zerbib, C. and J. J. Mustel. 1984. Incorporation de la vitellogénine tritiée dans les ovocytes du Crustacés Amphipode *Orchestia gammarellus* (Pallas). Int. J. Invert. Reprod. 7:63–68.