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Vitellogenesis in *Orientocreadium batrachoides* Tubangui, 1931 (Digenea: Allocreadiidae) from *Clarias gariepinus*, an ultrastructural study

A.A. Taeleb [a,*], **G.H. Lashein** [b]

[a] *Zoology Department, Faculty of Science, Zagazig University, Egypt*
[b] *Zoology Department, Faculty of Science, Benha University, Egypt*

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Ultrastructure

Abstract Vitellogenesis in *Orientocreadium batrachoides*, an intestinal parasite of the catfish, *Clarias gariepinus* was investigated by transmission electron microscope. Four stages of vitellocyte development have been distinguished during vitellogenesis: (I) stem cell stage of the gonial type (immature vitellocyte), (II) early and (III) advanced stages of maturing vitellocytes. Both early and advanced stages of maturing vitellocytes involved the initiation of protein synthetic activity, onset of shell globule formation and progressive fusion of single shell globules into large shell globule cluster in their cytoplasm and (IV) mature vitellocyte. Vitellocyte maturation is characterized by: (1) an increase in cell volume, (2) extensive development of parallel cisternae of granular endoplasmic reticulum, (3) development of Golgi complexes and (4) a continuous development of shell globules that fused into shell globule clusters. Mature vitelline cells are characterized by shell globule clusters, which play an important role in egg shell formation, lipid droplets and β-glycogen that accumulated in large amounts in their cytoplasm. Single "lamellar" granules are present in the cytoplasm of the mature vitellocytes.

Introduction

Literature holds little information on the ultrastructural aspects of vitellocytes and vitellogenesis in digeneans. Most of the studies have been carried out on species of economic or medical importance such as *Fasciola hepatica* (Bjorkman and Thorsell, 1963; Thorsell et al., 1966; Irwin and Threadgold, 1970; Hanna, 1976), *Schistosoma mansoni* (Erasmus et al., 1982) and *Paragonimus ohirai* (Fukuda et al., 1983). These studies demonstrated many intact vitellocytes (about 20–30) enclosed within the ovum in large eggs.

It has been thought, for many years, that Mehli's gland provides the egg shell, whereas the vitelline gland provides the nutrient for the developing embryo. Later the vitelline gland has been reported to be implicated in producing the shell materials as well. Irwin and Threadgold (1972) and Grant et al. (1977) reported that the vitelline gland produces vitelline

* Corresponding author.
E-mail address: dr_azza2011@yahoo.com (A.A. Taeleb).

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globules, of which some of them are involved in shell formation and others provide nutrients for the developing embryo within the egg. Bjorkman and Thorsell (1963) reported that the shell globules are formed within the vitelline cells.

The role of the vitelline gland in egg-shell formation in digeneans has been investigated in *Dicrocoelium dendriticum* (Grant et al., 1977), *S. mansoni* (Wells and Cordingley, 1991), *Haploporus benedenii* (Sampour, 2006) and in the cestode *Diplocotyle olrikii* (Poddubnaya et al., 2005). The fine structure of the vitellaria was studied by Bjorkman and Thorsell (1963), Erasmus (1973), Irwin and Magurie (1979) and Holy and Wittrock (1986) who reported that in some primitive species of digeneans (with extensive vitelline glands) many intact vitelline cells are enclosed within the ovum in the large egg, whereas, in some advanced species like Microphallidae, two vitelline cells are enclosed with the ovum during the shell formation.

The very few published ultrastructural studies on vitellogenesis were carried out on digeneans as *Gorgoderina vitelliloba*)Irwin and Magurie, 1979), *Maritrema linguilla* (Hendow and James, 1989), *Haploporus lateralis* (Sampour, 2008), *Maritrema feliui* (Świderski et al., 2011), monogeneans as *Metamicrocotyla macrocauta* (Baptista-Farias, 1998), cestodes as *Paraechinophallus japonicus* (Levron et al., 2007) and aspidogastrean, as *Aspidogaster limacoides* (Levron et al., 2010).

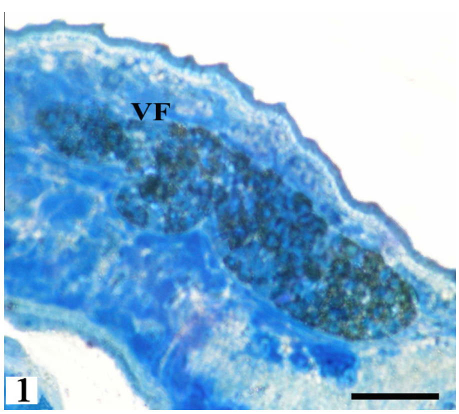

Fig. 1 Semi-thin section of *O. batrachoides* showing vitelline follicles (VF) surrounded by parenchyma. Scale bar = 40 μm.

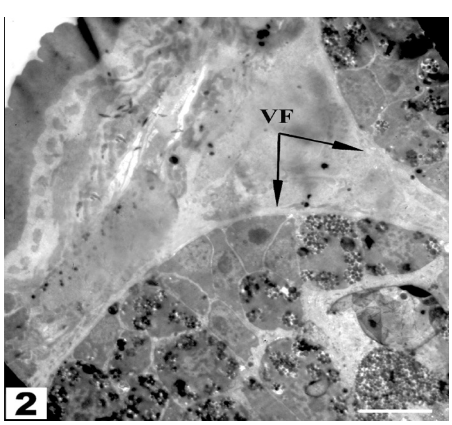

Fig. 2 Transmission electron micrographs of vitellaria showing different stages of vitellogenesis. Vitelline follicles (VF) showing vitellocytes at various stages of development. Scale bar = 10 μm.

All transmission electron microscopic observations on vitellogenesis have recently been considered as useful criteria for phylogenetic and evolutionary studies of the flatworms (Świderski and Xylander, 2000; Świderski et al., 2009). Erasmus (1975), Mehlhorn et al. (1981) and Shaw and Erasmus (1988) reported that the knowledge on vitellogenesis in parasitic flatworms may also have an important applied aspect.

The present study was conducted to describe in detail the ultrastructural aspects of vitellogenesis in *Orientocreadium batrachoides* (Digenea: Allocreadiidae), a parasite of *Clarias gariepinus* in comparison with previous reports on other trem-

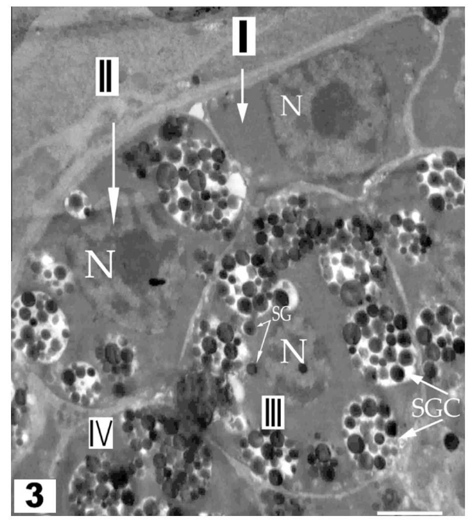

Fig. 3 Vitelline follicles showing four consecutive stages of vitellogenesis within the follicles: (I) immature vitellocyte, (II) early and (III) advanced stages of maturing vitellocytes and (IV) mature vitellocyte. Note nucleus (N), shell globules (SG) and shell globule clusters (SGC). Scale bar = 2 μm.

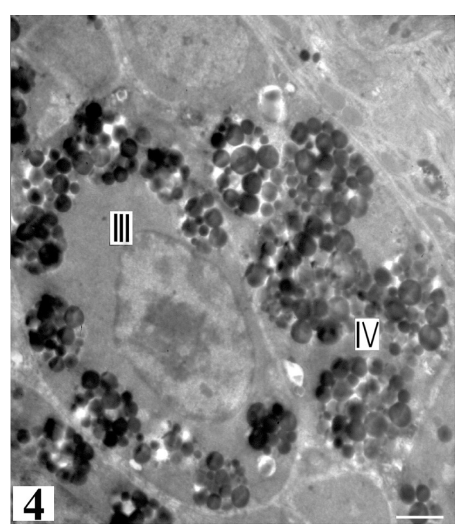

Fig. 4 Vitelline follicles showing four consecutive stages of vitellogenesis within the follicles: (I) immature vitellocyte, (II) early and (III) advanced stages of maturing vitellocytes and (IV) mature vitellocyte. Note nucleus (N), shell globules (SG) and shell globule clusters (SGC). Scale bar = 2 μm.