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SPERM TRANSFER IN MARINE INVERTEBRATES: RECORD OF VERMETID GASTROPOD SPERMATOPHORES IN THE PLANKTON

RIASSUNTO

Viene segnalato per la prima volta il ritrovamento di spermatofores dei Gastropoda Vermetidae lungo le coste dell'Isola d'Elba. Tali spermatofores sono riconducibili a quelle della specie *Thylacodes arenarius* (L., 1758) (= *Serpulorbis arenarius* (L., 1767)). Vengono riportate le caratteristiche morfologiche delle differenti spermatofores osservate e sono discussi i risultati alla luce delle particolari strategie riproduttive di tali invertebrati sessili afallici.

SUMMARY

The occurrence of Gastropoda Vermetidae spermatofores is recorded. They correspond to the spermatofores of the species *Thylacodes arenarius* (L., 1758) (= *Serpulorbis arenarius* (L., 1767)). The morphological characteristics of the different spermatofores observed are showed and the records are analyzed in the light of the reproductive strategies of such aphyllid sessile invertebrates.

INTRODUCTION

The interest in autoecology of marine invertebrates increased in recent years, being considered a key to understand ecological systems functioning and development (GIANGRANDE *et al.*, 1994). In particular, the knowledge of life cycle features is necessary to interpret life histories and spatial and temporal distribution of marine organisms (BOERO, 1990). Among life cycles features, those concerning reproductive traits are especially worth of interest because

of their implication in demographic patterns, colonization strategies, community dynamics, plankton-benthos interaction. As regarding the reproductive strategies, in their detailed review GIANGRANDE *et al.* (1994) focused on reproductive effort in term of egg size, larval development modes and dispersal and demographic patterns. Other important traits of reproductive biology concern sperm type, sperm transfer and male copulatory apparatus (FRANZEN, 1956; FRETTER and GRAHAM, 1962). In contrast with the majority of marine invertebrate species, among very few *taxa*, such as Crustacea and Gastropoda, fertilization is always internal. In particular, in some Mollusca Caenogastropoda fertilization cannot be effected by copulation, e.g. Cerithioidea, Vermetoidea, Triphoroidea, Janthinoidea (MORTON, 1966; ROBERTSON, 1989). The Turitellidae have the mantle cavity kept closed by a portcullis of pinnate tentacles against the entry of sediment and thus also of the penis. In some Cerithiidae and Cerithiopsidae the mantle cavity is very narrow and the female opening is too far to be reached by a penis. In the sessile Gastropoda of the family Vermetidae the adult may be out of reach of other individuals. These latter have aphillic males and sperm transfer is assured by means of water currents (SCHEUWIMMER, 1979).

Pelagic spermatophores of Vermetidae are oval and gelatinous drop-shaped sac with three concentric capsules (or spheres) and are filled with a transparent liquid. A tertiary capsule, the outer, is very thin, irregular and elongated. The secondary capsule, very elastic, is often adherent to the primary capsule: this is a swollen capsule linked to the ejaculatory tube (or extrusive tube). This tube is simple or complexly coiled, depending to the species that generate it, and it is connected with the sperm mass (HADFIELD and HOPPER, 1980).

Free spermatophores are released in the water column from the mantle cavity of the males (HADFIELD and HOPPER, 1980) and are caught in the feeding mucous string or inhalant current of the females. When Vermetidae females issue mucus strings to pick food particles, spermatophores may be hauled in to be eaten, the outer sphere is damaged by jaws and radula and the inner sphere is exposed. In this way, the spermatophore is activated, the content of the inner sphere pass through the everted ejaculatory tube, the sperm mass is then released and enters the mantel cavity, driven by the respiratory current (SCHEUWIMMER, 1979; HADFIELD and HOPPER, 1980). Sperm mass contains both euspermatozoa (eupyrenic spermatozoa or "typical spermatozoa") and paraspermatozoa (apyrenic spermatozoa or "giant spermatozoa" or "atypical spermatozoa") arranged in a spiral in the spermatid sac. It can be supposed that paraspermatozoa have a trophic function for euspermatozoa, which are considered to be evolutionarily primitive because of the absence of a sex chromosome system (HAYAKAWA, 2007). The comparatively large and polysaccharidic apyrenic spermatozoa are essential in maintaining viable eupyrenic spermatozoa during the spermatophore planktonic life (HADFIELD and HOP-

PER, 1980; D'ANCONA LUNETTA and DAMIANI, 2002). The life span of spermatophores in laboratory is of 12-20 hours and it may be representative of their survival in the plankton (HADFIELD and HOPPER, 1980).

Adult Vermetidae are sessile animals that live in rocky communities. Some species are solitary or have a low density population (e.g. *Thylacodes arenarius*, *Vermetus granulatus*, *V. cristatus*); other species are gregarious and form dense aggregations often cemented by encrusting red algae (e.g. *Dendropoma petraeum* and *Vermetus triquetrus*). Vermetidae typically colonize the upper infralittoral zone, but they may be also commonly found deeper (D'ANCONA LUNETTA and DAMIANI, 2002; CALVO and TEMPLADO, 2005). Vermetidae are often important bio-constructors between intertidal and upper infralittoral zones: they may build "reef" and "trottoir", large platforms expanded for several meters, that are among the more important bioconstructions of the Mediterranean Sea (CHEMELLO, 2009).

Four genera (*Vermetus*, *Dendropoma*, *Petalocochnus* e *Thylacodes*) and 10 species belonging to Vermetidae are recorded in the Mediterranean Sea (RELINI, 2008). Vermetidae are common in the Mediterranean Sea, nevertheless only few studies regard spermatophores particularly concerning the Spanish coasts (CALVO *et al.*, 1998; CALVO and TEMPLADO, 2004; 2005). Some others studies regard the coasts of Japan (SCHEUWIMMER, 1979), Hawaii, California (HADFIELD and HOPPER, 1980) and New Zealand (MORTON, 1951).

This paper reports the occurrence of Gastropoda Vermetidae spermatophores in the plankton from the Ligurian-Tyrrhenian Sea, with some observations on differences in morphology and on reproductive strategies of such aphyallic sessile invertebrates.

MATERIALS AND METHODS

Plankton samples were collected from April 2010 to February 2014 along the north-western coast of Isola d'Elba (Tuscany), between Marciana Marina and Patresi.

Plankton was collected with net of 100 µm mesh hauled in superficial waters in areas 20-50 m depth. Samples were transported alive to the laboratory to be studied *in vivo*, then they were filmed with digital camera for optical microscope.

The size of each spermatophore ranged from 1.5 to 2.2 mm.

RESULTS AND DISCUSSION

A total of 14 Vermetidae spermatophores were collected along the coasts of

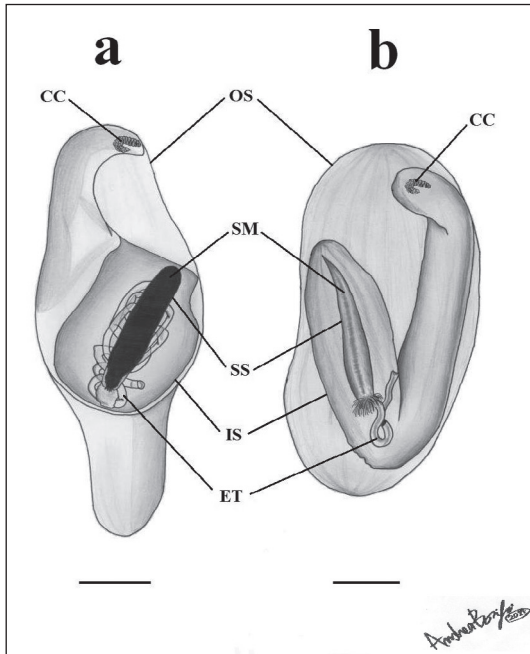


Fig. 1 – Spermatophores of *Thylacodes arenarius* (= *Serpulorbis arenarius*) from Isola d'Elba: a. morph "a"; b. morph "b"; OS: outer sphere; IS: inner sphere; CC: connecting cord; SS: sperm sac; SM: sperm mass; ET: eiaculatory tube. Scale bars: 500 μ m.

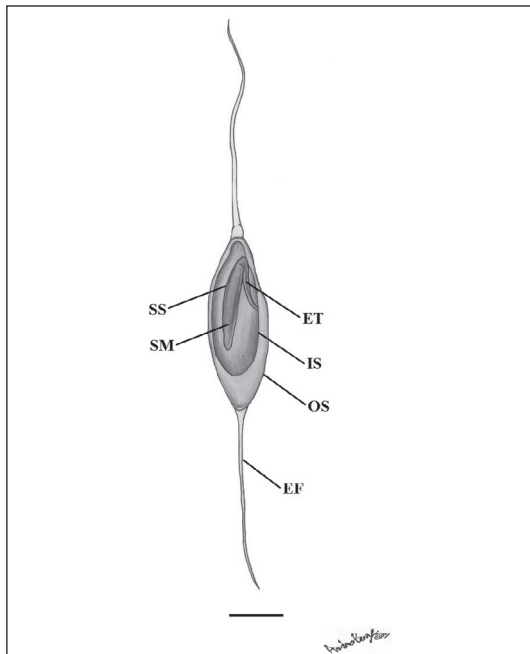


Fig. 2 – Spermatophore of *Vermetus triquetrus*. OS: outer sphere; IS: inner sphere; SS: sperm sac; SM: sperm mass; ET: eiaculatory tube; EF: end filament. Scale bars: 100 μ m. The spermatophore is redrawn from CALVO and TEMPALADO (2005).

Isola d'Elba. They corresponded to those of *Thylacodes arenarius* (L., 1758) (= *Serpulorbis arenarius* (L., 1767)), all lacking of terminal filaments. Two different morphologies have been recognized: ten specimens have been included into the morph "a" (Fig.1a); the other four into the morph "b" (Fig.1b). In Vermetidae species of the genera *Dendropoma*, *Petaloconchus* and *Vermetus* the outer sphere have tapered ends, drawn out into long, thin filaments (Fig.2), while in the genus *Thylacodes* the spermatophores are oblong in shape without the characteristic filaments at each end (CALVO and TEMPLADO, 2005).

All the collected spermatophores correspond both in shape and size to these of *T. arenarius*, in fact, their structure consists in an outer sphere, oval and turgid, containing both secondary and inner sphere. In the morph "b", the inner capsule containing a sperm mass with a pointed apex, which is linked to a not very intricate and coiled ejaculatory tube (Fig.1b). The spermatophores of morph "a" essentially distinguished from those of morph "b" in size and shape of sperm mass and in length of ejaculatory tube: in fact, the inner sphere contains a cylindrical sperm mass with blunted apex, linked to a long and coiled ejaculatory tube (Fig.1a). The sperm mass holds both euspermatozoa and paraspermatozoa (HAYAKAWA, 2007).

The distribution of adults of *T. arenarius* in Ligurian sea and in northern Tyrrhenian sea confirms such record (SCHIAPARELLI and CATTANEO-VIETTI, 1999; SCHIAPARELLI *et al.*, 2003).

Isolated spermatophores of *T. arenarius* morph "b" have been mainly collected in spring and summer: 06/04/2010; 16/07/2011; 08/08/2012; 16/08/2012. On the contrary, isolated spermatophores of morph "a" have been collected in winter and spring: 29/04/2011; 04/01/2012; 02/04/2012; 06/04/2012; 10/04/2012; 13/04/2012; 30/12/2012; 02/01/2013; 24/03/2013; 08/02/2014 (one spermatophore for each date).

T. arenarius is an infralittoral species and its spermatophores may be found isolated or close together in low density aggregates. Adults of such species feed trapping suspended material from the water column by means of mucous feeding net. On the other hand, other Vermetidae gregarious species, such as *Dendropoma petraeum*, feed by a ciliary filter-feeding mechanism without mucous feeding nets (CALVO and TEMPLADO, 2005). For such sessile gonochoric species which retain the characteristic internal fertilization, the sperm transfer mechanism is connected with the nutrition assumption mode.

The production of pelagic spermatophores results a successful reproductive strategy in gregarious, sessile, tubicolous marine invertebrates, in addition to the vermetids, such as some Polychaeta (CAZAUX, 1971; RICE, 1978; HSIEH and SIMON, 1990), Pogonophora (FLÜGEL, 1977) and Phoronida (ZIMMER, 1967). In fact they consist in substantial sperm packets, that may be appropriately received by the females present in the dense aggregations of individuals.

On the contrary, for certain Vermetidae such as for the genus *Thylacodes*, which live in relatively disperse distribution, fertilization by means of pelagic spermatophores is assured by males releasing spermatophores continuously or over a long period of the year. As correlated adaptive strategy, the females have a lot of sperm storing organs to maintain viable sperms for a long time, expecting their breeding period (SCHEUWIMMER, 1979; HADFIELD and HOPPER, 1980). Furthermore, in the gregarious species without a mucous-trap feeding mode, e.g. *Dendropoma petraeum*, the two long filaments at the tapered end of spermatophores increase the likelihood of capture by the entering water currents mucous feeding-net of the females. On the other hand, for the Vermetidae producing the mucous feeding nets, such as *T. arenarius*, these latter act as traps for the pelagic spermatophores by the females: it was estimated that a specimen with a tube diameter of less than 1 cm may catch spermatophores from an area of more than 1 m² (HADFIELD and HOPPER, 1980).

So, pelagic spermatophores are an effective mechanism for sperm transfer among marine invertebrates with sessile and aphyllous adults, which maintain the constraint of internal fertilization.

REFERENCES

- BOERO F., 1990 – Life-cycles, life-histories, and recruitment. *TREE*, 5: 6.
- CALVO M., TEMPLADO J., 2004 - Reproduction and development in a vermetid gastropod, *Vermetus triquetrus*. *Invert. Biol.*, 123: 289 - 303.
- CALVO M., TEMPLADO J., 2005 - Spermatophores of three Mediterranean species of vermetid gastropods (Caenogastropoda). *J. Mollus. Stud.*, 71: 301 - 311.
- CALVO M., TEMPLADO J., PENCHASZADEH P.E., 1998 - Reproductive biology of the gregarious mediterranean vermetid gastropod *Dendropoma petraeum*. *J. Mar. Biol. Ass. U.K.*, 78: 525 - 549.
- CAZAUX C., 1971 – Développement larvaire de *Microspio mecnikowianus*. *Proc. 4th Eur. Mar. Biol. Symp.*, 4: 247-257.
- CHEMELLO R., 2009 - Le bioconstruzioni marine in Mediterraneo. Lo stato delle conoscenze sui reef a vermeti. *Biol. Mar. Mediterr.*, 16: 2 - 18.
- D'ANCONA LUNETTA G., DAMIANI F., 2002 - Spermiogenesis in vermetid gastropod *Dendropoma petraeum* (Gastropoda Prosobranchia). *Eur. J. Histochem.*, 46: 75 - 86.
- FLÜGEL H., 1977 – Ultrastructure of the spermatophores of *Siboglinum ekmani* Jägersten (Pogonophora). *Nature*, 269: 800-801.
- FRANZEN A., 1956 – On spermiogenesis, morphology of the spermatozoon and biology of fertilisation among invertebrates. *Zool. Bidr. Uppsala*, 31: 355-482.
- FRETTER V., GRAHAM A., 1962 – British Prosobranch Molluscs; their functional anatomy and ecology. *Ray. Soc., London*: 755 pp.
- GIANGRANDE A., GERACI S., BELMONTE G., 1994 – Life-cycle and life-history diversity in marine invertebrates and implications in community dynamics. *Oceanogr. Mar. Biol. Ann. Rev.*, 32: 305-333.
- HADFIELD M.G., HOPPER. C.N., 1980 - Ecological and evolutionary significance of pe-

- lagic spermatophores of vermetid Gastropods. *Mar. Biol.*, 57: 315 - 325.
- HAYAKAWA Y., 2007 – Parasperm: morphological and functional studies on non fertile sperm. *Ichthyol. Res.*, 54: 111-130.
- HSIEH H.L., SIMON J.L., 1990 – The sperm transfer system in *Kinbergonuphis simony* (Polychaeta: Onuphidae). *Biol. Bull.*, 178: 85-93.
- MORTON J.E., 1951 - The structure and adaptation of the New Zealand Vermetidae. I. The genus *Serpulorbis*. *Trans. R. Soc. N. Z.*, 79: 1 -19.
- MORTON J.E., 1966 – Molluscs. Hutchinson University Library, London: 243 pp.
- RELINI G. (ed.), 2008 – Checklist della flora e della fauna dei mari italiani (Parte I). *Biol. Mar. Mediterr.*, 15: 256.
- RICE S.A., 1978 – Spermatophores and sperm transfer in spionid polychaetes. *Trans. Amer. Microsc. Soc.*, 97: 160-170.
- ROBERTSON R., 1989 - Spermatophores of aquatic non-stylommatophoran gastropods: a review with new data on *Heliacus* (Architectonicidae). *Malacologia*, 30: 341 – 364.
- SCHEUWIMMER A., 1979 - Sperm Transfer in the Sessile Gastropod *Serpulorbis* (Prosobranchia: Vermetidae). *Mar. Ecol. Progr. Ser.*, 1: 65 - 70.
- SCHIAPARELLI S., CATTANEO-VIETTI R., 1999 – Functional morphology of vermetid feeding-tubes. *Lethaia*, 32: 41-46.
- SCHIAPARELLI S., GUIDETTI P., CATTANEO-VIETTI R., 2003 – Can mineralogical features affect the distribution of sessile gastropods? The Vermetidae case in the Mediterranean Sea. *J. Mar. Biol. Ass. U.K.*, 83: 1267-1268.
- ZIMMER R.L., 1967 – The morphology and function of accessory reproductive glands in the lophophores of *Phoronis vancouverensis* and *Phoronis harmeri*. *J. Morphol.*, 121: 159-178.

