POLSKA AKADEMIA NAUK
INSTYTUT ZOOLOGII

ANNALES ZOOLOGICI

Tom XXXII Warszawa, 30 X 1975 Nr 16

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# Reproductive maturity in some *Vitrinidae* (*Mollusca, Gastropoda*) from Poland

[With 1 text-figure and 3 plates]

While discussing the annual breeding cycle in palearctic *Vitrinidae* several authors used the terms "juvenile", "semiadult" and "adult" (HESSE 1923; LOHMANDER 1938; FORCART 1955; RIEDEL, personal communication; WALDÉN, personal communication). These terms were not defined, but obviously meant not more than "small", "medium sized" and "big". For a more thorough study of the breeding cycle I felt it necessary to study the maturation of reproductive system first.

## MATERIAL AND METHODS

Specimens of *Vitrina pellucida* (MÜLLER), *Semilimax kotulai* (WESTERLUND) and *Eucobresia nivalis* (DUMONT et MORTILLET), collected in the years 1963–1965 in the valley Dolina Kościeliska, Tatra Mts, Poland, were used. The specimens were dissected, the reproductive system stained in borax carmine and mounted in Cedax microscopic slides. A total of 229 slides was made. Because of the well-known technical difficulties in dissecting out the ovotestis, this organ was excluded from the present study. All measurements were taken from ready slides. Diameter is always understood as the biggest diameter of the organ in question, shell diameter as the major diameter. Total length of genitalia was measured from top of the albumen gland to the genital orifice along all the curves of spermoviduct, free oviduct, vagina and genital atrium. The terms "proximal" and "distal" refer to the genital orifice.

## RESULTS

To describe the continuous process of growth, development and maturation with reasonable clarity, I found it necessary to distinguish among the juvenile animals three successive stages. This distinction being purely arbitrary, it is nevertheless based on some discontinuous events. Among animals, which could be referred to as mature ones, I have discovered profound differences between

those before and after copulation. Accordingly, two mature stages are described. The first mature stage represents animals ready to copulate, the second one animals that have already copulated and are about to lay eggs. This distinction can be done, as the three species in question breed only once in their lifetime and die out afterwards.

### *Vitrina pellucida* (O. F. Müller) (Pl. I)

Juvenile stage I. Individuals of shell diameter up to 1.8 mm and total length of genitalia up to 1.4 mm are assigned here. Genitalia in the form of a simple and straight strand of non-differentiated tissue. Albumen gland is a semicircular vesicle, 0.07–0.1 mm in diameter, without any visible structure. Hermaphrodite duct much wider at the proximal end, so as to form an elongated triangle, its lumen not visible. In the narrow spermoviduct the oviducal and prostatic portions are not distinguishable. Bursa copulatrix not discernible. Judging from later stages, it is adhering closely to the spermoviduct. Vas deferens not visible. Penis short and thick, pear-shaped, penial retractor well-marked.

Juvenile stage II. Individuals of shell diameter 1.9–2.9 mm and total length of genitalia 1.5–2.2 mm. The distinction of this stage is based mainly on the growth and differentiation of the albumen gland, while the rest of reproductive organs change but little. Spermoviduct remains straight and narrow. Hermaphrodite duct is cylindrical, but still completely straight. It is at this stage, that appears the mysterious atrial diverticulum, possibly a brachium copulatorium rudiment (Umiński 1968). Animals at this stage, having the total length of genitalia relatively stable, differ in size and structure of particular organs, warranting the following subdivision:

a. Albumen gland, 0.12–0.17 mm in diameter, is still flat and thin. A spirally coiled duct of 2–3 coils is showing inside. Penis twice or even three times as big as in the preceding stage, but of the same shape.

b. Albumen gland 0.18–0.28 mm in diameter, with the lobular structure beginning and gradually masking the coiled duct (4–5 coils) inside. In the majority of individuals penis is still pear-shaped, thickened at the distal end. In some it starts to change into the final shape, i.e. elongated, almost cylindrical, thickest at the proximal end. Sometimes the bursa copulatrix and its duct are discernible, though it is not possible to separate them, using standard dissecting technique. In still fewer cases vas deferens can be seen, too.

Juvenile stage III. Individuals of shell diameter 2.6–3.9 mm and total length of genitalia 1.8–3.3 mm. Diagnostic characters of this stage are first of all the albumen gland, which gains the typical lobular structure of mature animals. The other typical trait is the spermoviduct with oviducal and prostatic portions clearly distinct, and the whole of it bent to form several loops, as in mature specimens. In the middle run the oviducal part grows considerably, so

as to be relatively wide there, while remaining comparatively narrow at the proximal end and just below the albumen gland. Vas deferens of almost definitive form. A primitive trait, making the difference between this stage and the mature animals, is the still persisting atrial diverticulum. Another one is the duct of bursa copulatrix, which lacks a thickened, glandular basis. The bursa itself is not widened, so that the whole is just a blind tube. The following subdivision is proposed:

a. Diameter of albumen gland about 0.3 mm (0.27–0.48 mm). Spermoviduct looped, but oviducal part not folded.

b. Diameter of albumen gland about 0.5 mm (0.41–0.52 mm). Oviducal part folded.

Mature stage I. This stage is attained by animals of shell diameter 2.6–4.1 mm. Reproductive system is strikingly bigger, its total length being 3.4–6.2 mm, diameter of albumen gland 0.7–1.4 mm. Diagnostic traits are: a. no atrial diverticulum, b. a thick glandular swelling developing around the base of bursa copulatrix duct. It is well marked-off from the rest of the duct as well as from other parts of the reproductive system. Cell nuclei are particularly big here and cells relatively small, so in stained slides this glandular part looks very different from all the other organs.

Bursa copulatrix widens a little, so as to be distinguishable from duct, though the difference remains small. Oviducal part of spermoviduct heavily folded and prostatic part with the typical structure of numerous small folds. As opposed to the next stage, albumen gland is rather flat, reminding of the juvenile stages.

Mature stage II. This stage is reached by animals of shell diameter not less than 2.8 mm, usually much more. In comparison with the preceding stage, albumen gland is very thick. Its volume seems to be twice to three times that of this gland in mature stage I, diameters being equal. It is only at this stage that the hermaphrodite duct shows the typical coiling. The middle, thickened run of spermoviduct is still thicker at this stage. Oviducal part contains large quantities of mucus and is swelling considerably in water. Bursa copulatrix diameter is 3–5 times that of its duct. Inside the bursa there generally is a well-staining concretion of sperm. Glandular part of duct much thickened, its delimitation still more pronounced, than it was in the first mature stage.

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The ontogenic development of reproductive system in *Vitrina pellucida* is at first isometric. All organs grow and develop at about the same rate. Towards the end of juvenile stage III this is changed due to a speedy increase and development of albumen gland and spermoviduct. From this moment on, these two organs markedly overbalance all the remaining ones, the difference still growing during the period of maturity, especially after copulation (i.e. at

the mature stage II). Bursa copulatrix is growing steadily all the time, together with its duct, not exceeding the latter in diameter. It does enlarge suddenly and considerably on reaching mature stage II, an obvious result of the receipt of partners' sperm at the time of copulation. The glandular basis of bursa copulatrix duct appears and develops during the period of maturity. A purely juvenile organ, found only in stages II and III is the atrial diverticulum, budding off the atrium opposite the penis. Fertilization pocket (=vesicula seminalis, =talon) is not to be seen at any stage.

### *Semilimax kotulai* (Westerlund) (Pl. II)

Juvenile stage I. Individuals of shell diameter up to 2.1 mm and total length of genitalia up to 2.2 mm are assigned here. Albumen gland is a simple vesicle, without any visible structures inside. In very young animals it is elongated, tapering into the hermaphrodite duct. Spermoviduct is a straight strand of tissue, oviducal and prostatic parts not distinguishable. Bursa copulatrix and vas deferens not discernible. Penis short and thick, with distal end the thickest.

Brachium copulatorium (copulatory arm), found in the genus *Semilimax* Stabile, is diagnostic not only in taxonomy. It serves equally well in judging the development of reproductive system. Here it is only a primordium. The arms' sheath is a thin-walled vesicle, with the arm proper inside, in the form of a well-staining globose body.

Juvenile stage II. Individuals of shell diameter 2.2–3.2 mm and total length of genitalia 2.2–3.9 mm. Hermaphrodite duct cylindrical, well marked-off from albumen gland, which is round, 0.19–0.38 mm in diameter. In younger animals a coiled duct is visible inside, in older ones it is covered by the beginning lobular structure of the gland. Oviducal and prostatic parts of spermoviduct are clearly distinct. Spermoviduct is somewhat bent, when in natural position. Its middle run is much wider than are both ends. Bursa copulatrix, together with duct, is thin and elongated. Vas deferens still not visible. Penis elongated, cylindrical. The sheath of copulatory arm is a vesicle, still thin-walled, but already elongated. The heavily staining arm is also elongated, sticking a little out from the sheath. In younger specimens it is only the semispherical end, showing on the outside, in older ones it is a longer fragment, bent in the shape of a tobacco pipe.

Juvenile stage III. Individuals of shell diameter 2.6–3.7 mm and total length of genitalia 3.1–4.1 mm. Albumen gland much bigger, 0.48–0.83 mm in diameter, its lobular structure well pronounced. Bursa copulatrix with duct look like an elongated diverticulum, tapering regularly from a rather wide base towards the blind end. Vas deferens visible. Penis elongated, of about the final shape, but still devoid of the glandular cap, covering the distal tip in

mature animals. Inner structures of penis readily visible, resemble the final pattern. Copulatory arm changes considerably. It is now much bigger, twice or three times longer (0.95–1.9 mm) than in the preceding stage. The sheath wall is muscular by now. The arm proper protrudes much more from the sheath, the protruding part being about equal in length and diameter with the sheath.

Mature stage I. This stage is attained by animals of shell diameter 3.2–4.3 mm and total length of the reproductive system 4.6–6.8 mm. Albumen gland bigger, 0.72–1.5 mm in diameter, with final lobular structure, rather flat in comparison with that of the last mature stage. Spermoviduct much bigger, but the difference is quantitative only. Bursa copulatrix with its duct are both much wider, so as to appear quite stocky. A small but distinct constriction is marking the bursa off from the duct. Bursa is empty. Copulatory arm is growing on (length 1.7–3.4 mm), circular folds appearing inside the sheath. The arm proper, thickened at the distal end, is thin and convoluted close to the sheath.

Mature stage II. This stage is reached by animals of shell diameter not less than 3.7 mm and total length of reproductive system 7.3 mm or over. Growth of albumen gland, both relative and absolute, is typical of this stage. With a diameter of 2.2–3.6 mm it is also very thick, in marked contrast with the earlier stage. Similar growth is characteristic of the spermoviduct, particularly of its oviducal part, which is full of mucus, swelling in water. Bursa copulatrix very big, with a sphere of well-staining material inside. Duct narrow, sharply delimited from the bursa. Copulatory arm still bigger, its sheath with a typical pattern of circular folds. The thin proximal part of the arm proper is longer and much more contorted.

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The ontogenic development of reproductive system in *Semilimax kotulai* is to some extent similar to that in *Vitrina pellucida*. At first all organs develop and differentiate at about the same rate, but later the albumen gland and spermoviduct accelerate and by far outdistance all the other ones. Still, disproportion between these two groups of organs is here far less accentuated than in the other two species. This is because in *Semilimax kotulai* the proximal organs, used in copulation, do not deteriorate afterwards, but are growing further, and at the second mature stage are very big. Penis is enormous, when compared with that of the other two species at the same stage. Particularly intriguing is the continued growth of copulatory arm. Judging from the observations of KÜNKEL (1933) on *Semilimax semilimax* (FÉR.) it is used to keep hold of the partner in copula. Such being the case, it is difficult to imagine any reasonable explanation of its prolonged growth after copulation.

***Eucobresia nivalis*** (Dumont et Mortillet) (Pl. III)

Juvenile stage I. Individuals of shell diameter up to 2.4 mm and total length of genitalia up to 2.0 mm are assigned here. Albumen gland is a simple vesicle, 0.12–0.17 mm in diameter, without any visible structure inside. In very young animals it is elongated, tapering gradually into the hermaphrodite duct. Spermoviduct straight, oviducal and prostatic parts distinguishable only in older specimens, bordering on the next stage. Bursa copulatrix and vas deferens not discernible. Vagina[1] elongated, narrow, about the same length as in fully mature animals. Penis long (about 0.50 mm) and thin, contrasting with that in juvenile *Vitrina pellucida* and *Semilimax kotulai*. Its distal end is the thickest.

Juvenile stage II. Individuals of shell diameter 2.2–3.3 mm and total length of genitalia 2.5–3.6 mm. At this stage all reproductive organs are discernible except the fertilization pocket (=vesicula seminalis, =talon). Lack of a visible fertilization pocket is diagnostic in distinguishing this stage from the next one. Albumen gland, 0.24–0.36 mm in diameter, forms 3–6 folds, the lobular structure appearing in older individuals. Oviducal and prostatic parts of spermoviduct distinguishable. Bursa copulatrix slightly wider than its duct. Vagina much thicker than before. At this stage it is the biggest of all reproductive organs. Neither its lumen, nor any inner structures are visible yet. Penis just a little longer, but much thicker, the width-length ratio increasing almost twofold in comparison with the preceding stage.

Juvenile stage III. Individuals of shell diameter 2.8–3.7 mm and total length of genitalia 3.9–5.1 mm. Albumen gland with a typical lobular structure, much bigger, 0.48–0.56 mm in diameter. Its relative dimensions increase also, the ratio of diameter to the length of vagina being about 0.80 as compared with 0.55 at the second juvenile stage and 0.30 at the first one. Fertilization pocket elongated, protruding beyond the contour of albumen gland. Spermoviduct bent in loops, similarly as in mature animals. Prostatic part with characteristic numerous small folds. Bursa copulatrix grows slowly and regularly, at this stage being 2.2–2.8 times wider than duct. Vagina somewhat longer and again thicker, with lumen showing and the inner structures appearing in older animals. Simultaneously appears the papilla, with which vagina opens into the proximal, thin-walled part of female duct. Penis, similarly as vagina, is a little longer and much thicker, width-length ratio again increasing almost twofold in comparison with the earlier stage. It is close to the final shape, thickest in the middle, narrower at both ends, but still without the glandular cap on distal end.

[1] The thick-walled part of female genital duct is often named "vaginal gland", "perivaginal gland" or "vaginal papilla". But there is no glandular tissue in this organ. It is built solely of epithelia and muscles. The term "papilla" applies to any conical protrusion. But the "vaginal papilla" in question is a whole, separate organ, not a protrusion of any kind, and is elliptic in shape. It is simply a vagina, though of a rather complex inner structure, and I prefer to call it this.

Mature stage I. Individuals of shell diameter 3.5–4.5 mm and total length of genitalia 4.9–6.2 mm. Albumen gland increases absolutely (diameter 0.62–0.81 mm) as well as relatively, its ratio to length of vagina being 0.97–1.4. Fertilization pocket large, extending considerably outside the contour of albumen gland. Spermoviduct about twice wider than in the last juvenile stage. Fully developed prostatic part consists of many little folds. Oviducal part starts folding. Width ratio of these parts is about 1:3. Bursa copulatrix attains a diameter 2.8–6 times that of duct. Size and proportions of vagina remain virtually unchanged, but its inner structures acquire final form. Thick, muscular walls converge proximally so as to form a big muscular papilla with a narrow orifice. On the inside these walls are covered with many sharp processuses which in the distal part point distally, i.e. towards the bursa copulatrix and in the proximal part point proximally, towards the orifice of papilla. Vagina is coaxial with the duct of bursa copulatrix, while the free oviduct enters through a narrow, constricted aperture at the junction of these two ducts, at right angles to their axis. It is at this stage that penis reaches its top size (length 1.0–1.4 mm). Tip of penis is covered by a big cap of glandular tissue, which adds a lot to such a marked step-up in length.

Mature stage II. This stage is reached by animals of shell diameter not less than 4.4 mm and total length of reproductive system over 6.0 mm. Albumen gland 1.1–2.4 mm in diameter, about 3–4 times as big as in the first mature stage. It does completely overgrow the fertilization pocket, so that the latter is no longer visible. Spermoviduct is also growing, and finally is about twice wider than in the preceeding stage, but due solely to the growth and folding of the oviducal part. The prostatic part declines and is narrower than in mature stage I. Width ratio of prostatic to oviducal parts is 1:6, and may reach even 1:8. Oviducal contents of swelling mucus similar as in the other species. Bursa copulatrix with a diameter of 0.50–0.76 mm is 7–11 times wider than its duct, which is the easiest to notice diagnostic trait of this stage. There is some well-staining contents, usually in the form of a spherical concretion.

Vagina is profoundly changed in comparison with the earlier stage, the changes being irregular, and range of individual variation extremely wide. It may be very small, may be very wide, may be unrecognizably out of shape. The inner processuses may disappear partially or completely, or become smaller and more numerous. If they persist, they do not form any consistent pattern. Papilla is gaping as if torn widely open. All these ravages are presumably due to copulation and to the eggs, making their way through, though some independent atrophic changes can not be excluded.

Penis is shrunken and disfigured. Glandular cap, covering its tip, usually atrophies.

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To sum up, in postembryonic ontogenesis the proximal reproductive organs, i.e. penis, vagina and genital atrium develop earlier, getting almost their final length as early as the juvenile stage II. In juvenile stages these organs predominate in size over the distal ones, seemingly underdeveloped. The proximal organs grow wider and thicker until they reach maximum size at mature stage I, to serve in copulation. After copulation, during mature stage II these organs deteriorate visibly. The distal organs on the other hand, i.e. albumen gland and spermoviduct start later and it is only at the first mature stage that some balance of size is reached. But then the distal organs are growing on, and in fully mature animals it is these organs, functioning in production of eggs, that form the bulk of reproductive system. Only bursa copulatrix does not fit in this pattern, growing regularly up to the moment of copulation, when the receipt of foreign sperm causes it to expand strikingly.

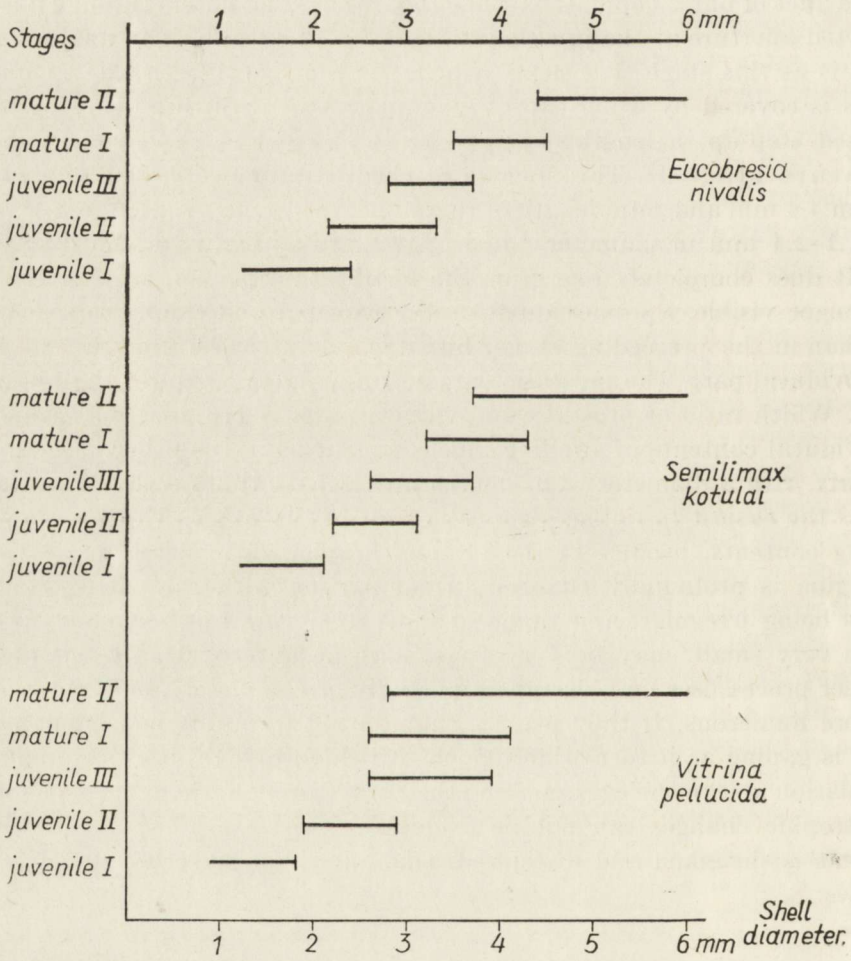


Fig. 1 Shell size and reproductive maturity.

## DISCUSSION

Comparison of shell size of animals at different stages of reproductive maturity (fig. 1) makes evident that correlation between these two phenomena is not particularly strong. To put it in other words, shell size is of relatively little value in judging the degree of maturity in these species. This is especially true of *Vitrina pellucida*, in which animals of shell diameter 2.8 mm can represent the early juvenile stage II as well as be fully mature, ready to deposit eggs, representing mature stage II. On the other hand, individuals as big as 3.7 mm in shell diameter can still be juveniles. Such discrepancies between size and maturity are, to some extent at least, due to the animals' life span, which can be either about 8 months or about 20 months (one-year or two-years breeding cycle), depending upon local climate. In populations of different life span there is a different correlation between growth rate and maturation rate. These matters will be dealt with at length in a subsequent paper.

## SUMMARY

In postembryonic ontogenesis of reproductive system of *Vitrina pellucida* (MÜLL.), *Semilimax kotulai* (WESTLD.), and *Eucobresia nivalis* (DUM. et MORT.) three successive juvenile stages and two mature ones are described. Distinction of juvenile stages is arbitrary, that of mature ones is not, mature stage I comprising animals which are ready to copulate, mature stage II those which have already copulated and are about to lay eggs. Shell size is not sufficient to estimate the degree of maturity, due mainly to differences between populations with a one-year or two-years breeding cycle.

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Acknowledgements

Appropriate permissions to enter the stricts reservation Hala Pyszna and to collect material there, granted by the Tatra National Park authorities, are hereby gratefully acknowledged.

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## STRESZCZENIE

[Tytuł: Dojrzewanie płciowe niektórych krajowych *Vitrinidae* (*Mollusca, Gastropoda*)]

W rozwoju osobniczym układu rozrodczego *Vitrina pellucida* (MÜLL.), *Semilimax kotulai* (WESTLD.) i *Eucobresia nivalis* (DUM. et MORT.) autor wyróżnia i opisuje trzy kolejne stadia młodociane i dwa dojrzałe. Rozróżnienie stadiów młodocianych jest arbitralne, natomiast granicę między dwoma stadiami dojrzałymi wyznacza moment kopulacji. Wielkość muszli nie wystarcza do oceny stopnia dojrzałości zwierzęcia, głównie z powodu różnic między populacjami jednorocznymi i dwuletnimi, co zostanie rozwinięte bardziej szczegółowo w następnej publikacji.

## РЕЗЮМЕ

[Заглавие: Половое созревание некоторых польских видов из семейства *Vitrinidae* (*Mollusca, Gastropoda*)]

Автор выделил и описал в процессе индивидуального развития *Vitrina pellucida* (MÜLL.), *Semilimax kotulai* (WESTLD.) и *Eucobresia nivalis* (DUM, et MORT.) три последовательные ювенальные стадии и две зрелые. Ювенальные стадии выделены произвольно на основании морфологических признаков; граница между двумя зрелыми стадиями определяется моментом копуляции. Величина раковины не может служить критерием степени зрелости животного главным образом вследствие различий, которые имеются между однолетними и двулетными популяциями. Этот вопрос будет более подробно разработан в следующей публикации.

## PLATE I.

*Vitrina pellucida* (MÜLL.)
Valley Dolina Kościeliska, Tatra Mts, Poland.
Genitalia.

Fig. 1. Juvenile stage I. Shell diameter 1.7 mm. Slide 199. Elevation 1240 m. 16 Aug. 1965.

Fig. 2. Juvenile stage IIa. Shell diameter 2.3 mm. Slide 171. Elevation 1240 m. 30 Oct. 1964.

Fig. 3. Juvenile stage II b. Shell diameter 2.1 mm. Slide 196. Elevation 980 m. 16 Oct. 1965.

Fig. 4. Juvenile stage III a. Shell diameter 3,8 mm. Slide 162. Elevation 1240 m. 19 July 1965.

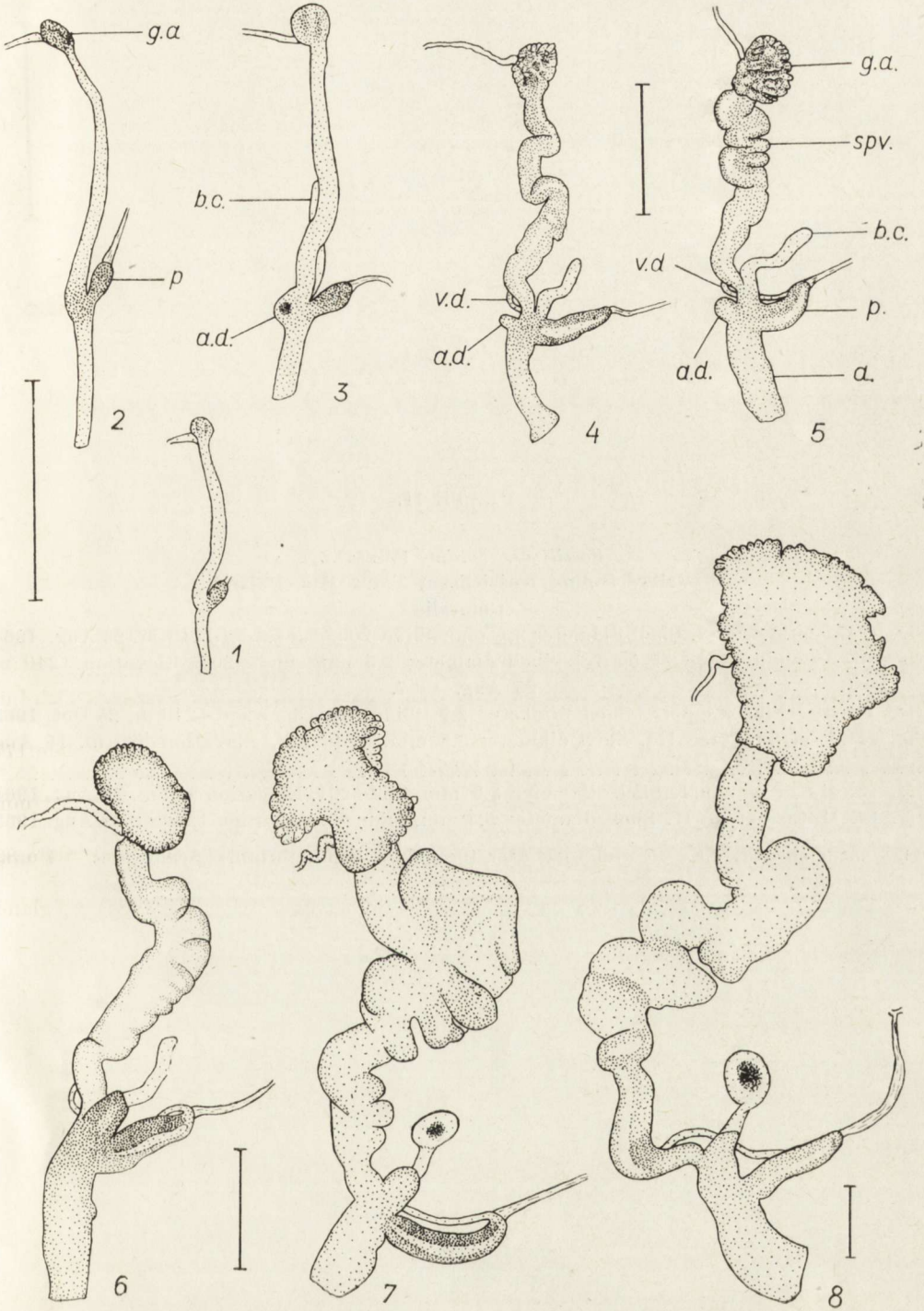
Fig. 5. Juvenile stage III b. Shell diameter 3.5 mm. Slide 163. Elevation 1240 m. 19 July 1965.

Fig. 6. Mature stage I. Shell diameter 4.1 mm. Total length of genitalia 4.7 mm. Slide 177. Elevation 980 m. 25 Aug. 1964.

Fig. 7. Mature stage II. Shell diameter 2.8 mm (sic!). Total length of genitalia 5.3 mm. Slide 194. Elevation 980 m. 29 Oct. 1964.

Fig. 8. Mature stage II. Shell diameter 6.0 mm. Total length of genitalia 11.9 mm. Slide 82. Elevation 1240. 25 Oct. 1963.

a. – atrium, a.d. – atrial diverticulum, b.c. – bursa copulatrix, g.a. – albumen gland, p. – penis, spv. – spermoviduct, v.d. – vas deferens. Scale lines – 1 mm.

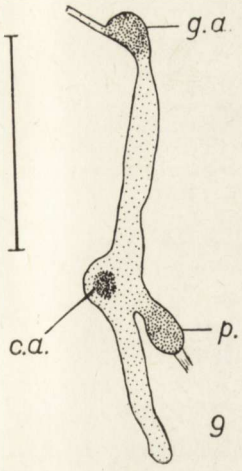


## PLATE II.

*Semilimax kotulai* (Westld.)
Valley Dolina Kościeliska, Tatra Mts, Poland.
Genitalia.

Fig. 9. Juvenile stage I. Shell diameter 2.1 mm. Slide 206. Elevation 1240 m. 26 Aug. 1964.
Fig. 10. Juvenile stage II (early). Shell diameter 2.5 mm. Slide 207. Elevation 1240 m. 25 Aug. 1967.
Fig. 11. Juvenile stage II. Shell diameter 2.9. Slide 108. Elevation 1240 m. 25 Oct. 1963.
Fig. 12. Juvenile stage III. Shell diameter 3.3 mm. Slide 218. Elevation 980 m. 15 Aug. 1965.
Fig. 13. Mature stage I. Shell diameter 3.9 mm. Slide 201. Elevation 980 m. 25 Aug. 1964.
Fig. 14. Mature stage II. Shell diameter 5.3 mm. Slide 63. Elevation 1240 m. 28 Aug. 1963.

a.s. – arm sheath, c.a. – copulatory arm (brachium copulatorium). Scale lines – 1 mm.

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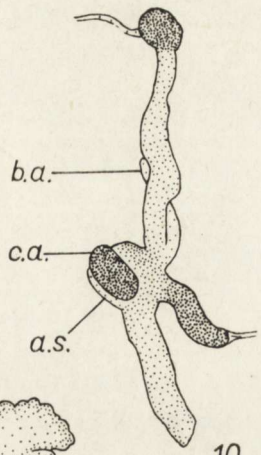


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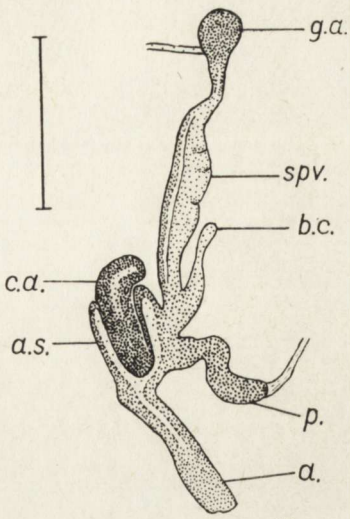


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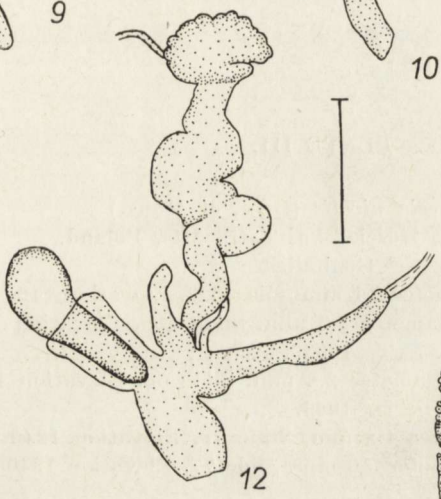

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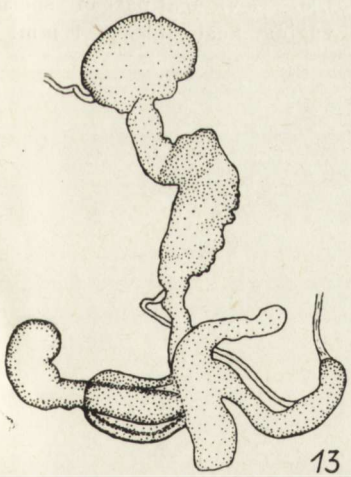

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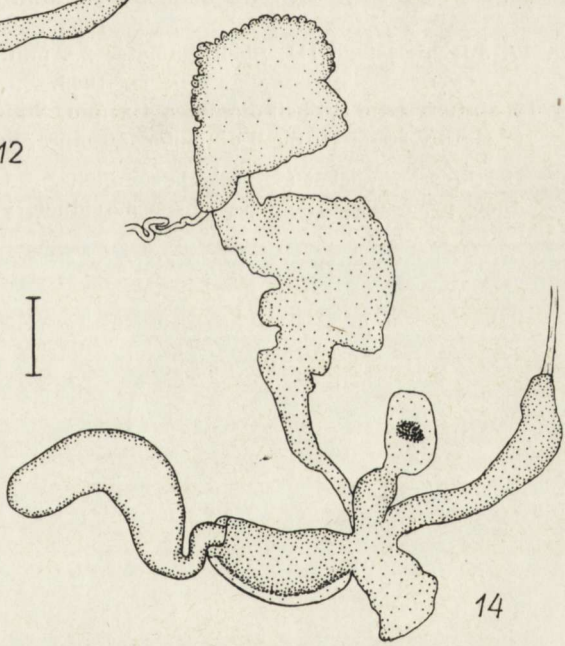

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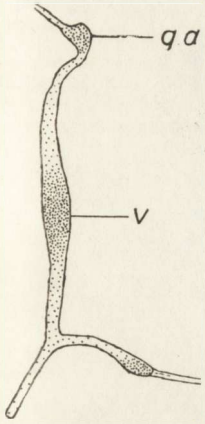
## PLATE III.

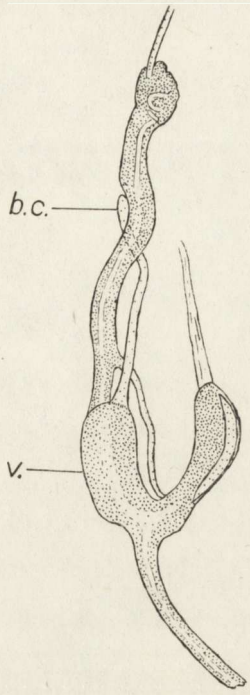
*Eucobresia nivalis* (Dum. et Mort.)
Valley Dolina Kościeliska, Tatra Mts, Poland.
Genitalia

Fig. 15. Juvenile stage I. Shell diameter 2.1 mm. Slide 229. Elevation 1420 m. 25 Oct. 1963.
Fig. 16. Juvenile stage II. Shell diameter 2.2 mm. Slide 61b. Elevation 1240 m. 28 Aug. 1963.
Fig. 17. Juvenile stage III. Shell diameter 2.8 mm. Slide 66. Elevation 1240 m. 24 Sept. 1963.
Fig. 18. Mature stage I. Shell diameter 4.2 mm. Slide 78. Elevation 1240 m. 25 Oct. 1963.
Fig. 19. Mature stage II. Shell diameter 5.6 mm. Slide 77. Elevation 1240 m. 25 Oct. 1963.

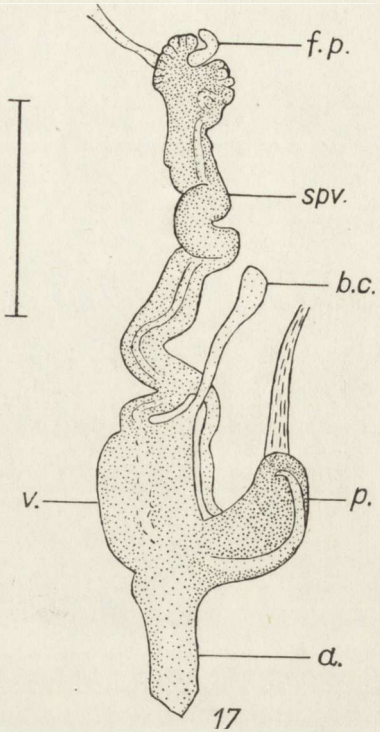
f.p. – fertilization pocket (= vesicula seminalis, = talon), o. – oviducal part of spermoviduct, pr. – prostatic part of spermoviduct, v. – vagina. Scale lines – 1 mm.



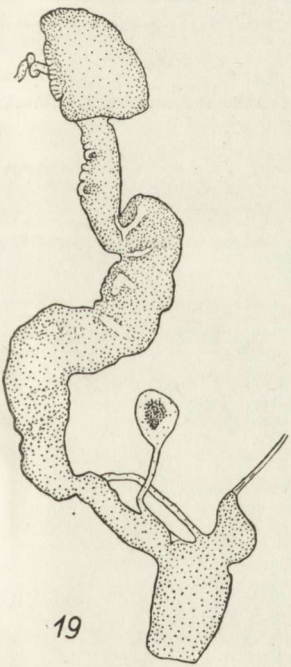

15




16




17



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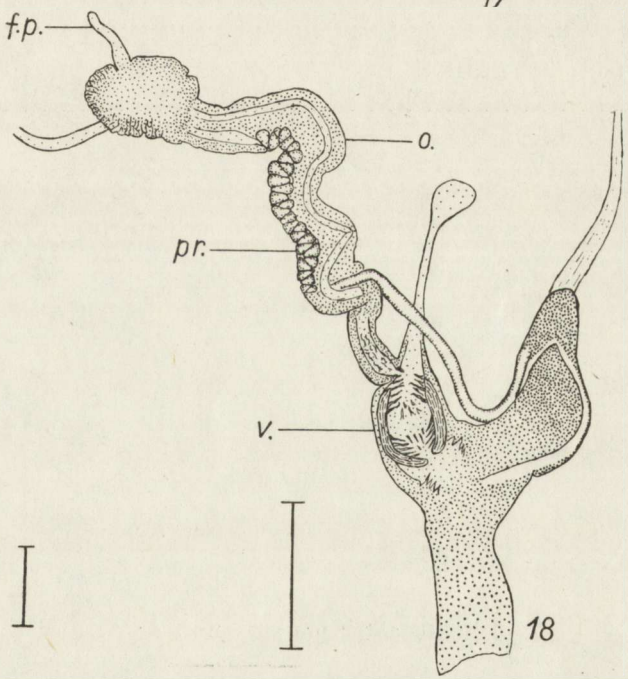


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Redaktor pracy — prof. dr A. Riedel

Państwowe Wydawnictwo Naukowe — Warszawa 1975
Nakład 1050+90egz. Ark. wyd. 1,25 druk. 1 1/8. Papier druk. sat. kl. III 80 g B. Cena zł 10,—
Zam. 2108/75 — A-14 — Wrocławska Drukarnia Naukowa