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DIPLOMARBEIT

Titel der Diplomarbeit

„Development in the cement apparatus of *Semibalanus balanoides* (L.) from subadult to adult

an ultrastructural study“

Verfasser

Michael Gruber

angestrebter akademischer Grad

Magister der Naturwissenschaften (Mag.rer.nat.)

Wien, 2011

Studienkennzahl lt. Studienblatt:	A 444
Studienrichtung lt. Studienblatt:	Diplomstudium Ökologie
Betreuerin:	Ao. Univ. – Prof. i.R. Dr. Waltraud Klepal

Development in the cement apparatus of *Semibalanus balanoides* (L.) from subadult to adult

An ultrastructural study

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Abstract

The cement glands of *Semibalanus balanoides* (L.) have been studied to find out possible changes in their number, location and organization from the subadult to the adult stage of the animal. Samples from Galway, Ireland revealed characteristics of the cyprid and the adult stages. The gland cells, already placed in the mantle tissue are still arranged in a cluster but begin to diverge. Their nuclei are round or oval, which is similar to the nuclei of the cyprid cement cells. At the storage pole of the gland cells in the subadult animals a great number of secretory vesicles appear. This could indicate an upcoming release of the proteinaceous content of the vesicles into the duct system. The observed duct lumen is almost empty except for the intracellular duct and some parts of the primary duct. Outside the cuticle on the base of the animal a layer of adhesive is detected showing a zonation of different electron density and also some external inclusions like algae. The observations confirm the subadult animal as transition stage between the cyprid and the reproductive adult barnacle.

Introduction

The balanomorph Thoracica *Semibalanus balanoides* belongs to the Cirripedia, a subclass of the crustaceans. Their planctotrophic nauplius larvae molt six times to the last non-feeding cyprid stage before attaching to a selected substrate. When exploring this substrate, a temporary adhesive is released through the attachment discs of the antennules. This temporary adhesive, termed “footprint” (Walker, 1984) keeps the cyprid in contact with the substrate against the forces of the surf but still allows some movement while testing the surroundings for even better settlement sites (Walker, 1992). It is assumed that the cement glands of the cyprid and the adult resemble each other closely but they are in different positions. During

metamorphosis from the cyprid to the subadult the glands move from behind the compound eyes of the cyprid to the perimeter of the base in subadult acorn barnacles (Khandeparker et al., 2007). Chemical and physical factors of the substrate and the water are analyzed by the cyprid prior to permanent settlement (Nott, 1969). In the cement glands of *Semibalanus balanoides*, proteins, phenols and polyphenol oxidase form complex tanned proteins (Walker, 1971) which provide the strength to hold onto the substrate. Membrane-based barnacles like *Semibalanus balanoides* have an intracellular duct within the cytoplasm of the cement glands, leading into collecting ducts placed in the mantle tissue and subsequently into the primary duct. The cement in the gland cells is stored in secretory granules (3-4µm in diameter in *Semibalanus balanoides* according to Walker, 1971). The content of these granules, collected by the intracellular duct is transported via the duct system to the base of the animal where it is extruded.

Many facts are known of the cement glands in the cyprid and in the adult barnacles (Fyhn & Costlow, 1976; Lacombe & Liguori, 1969; Lacombe, 1970; Walker, 1970, 1971, 1973; Walley, 1969), but so far nobody pointed out the differences of the cement apparatus in the two stages.

It is the aim of this investigation to show how these differences arise by studying the development of the cement apparatus from the subadult to the adult organism and by comparing the subadult with the cyprid and the adult stage. Emphasis is put on possible structural changes of the gland cells and the duct system. An attempt is made to find out how the cement gets from the gland cell into the duct and if there are structural differences of the cement within the duct compared with the released one.

Material and methods

Specimens of *Semibalanus balanoides* were collected in Galway, Ireland. Some of them were fixed immediately, others were kept in aquaria at a temperature of 21°C and a salinity of 25 ‰ for several weeks. These specimens were fed with *Artemia nauplii*. Eight individuals were investigated. Seven were fixed in Karnovsky-fixative for 3 hours and rinsed 3 x 10 minutes with Cacodylate buffer. One individual was fixed following the method by Eisenman & Alfert (1982), see appendix. To dissolve the calcareous parts, the samples were put into EDTA (10 %) for 24 to 48 hours. After fixation in 1 % Osmiumtetroxid for 2 hours, the samples were dehydrated in a graded series of Ethanol with Acetonitril as last step. Infiltration followed in three steps (3:1, 1:1, 1:3) with a mixture of Acetonitrile and Resin and embedding was done in Agar Low Viscosity Resin. Semithin sections (99 µm) were cut on a Leica Ultracut S microtome and stained with Toluidinblue for 10 seconds. These sections were analysed under a light-microscope. Ultrathin sections (70 nm) were also cut on a Leica Ultracut S microtome and stained with Uranyl acetate (15 minutes) and Lead citrate (7 minutes). These sections were viewed in a Zeiss EM 902 Transmission Electron Microscope.

Results

The subadult stage of *Semibalanus balanoides* shows cement gland cells placed laterally in the mantle tissue very close to the depressor muscles. These gland cells aggregate to clusters of up to 18 single gland cells (Fig.1). Most of them are ovoid, on average 20.58µm, at the most 38.31µm long and on average 11.13µm, at the most 20.84µm wide.

Within these clusters some dark cells are obvious (Fig.1). Apart from many Golgi bodies, mitochondria, free ribosomes, cisternae of rough endoplasmic reticulum, mostly vesicular, but also stacks of laminar forms, there are more ribosomes in the darker gland cells (Fig.2). This concerns both, free ribosomes and those in connection with the membranes of rough ER. In

comparison to the ordinary light gland cells the contour of the dark cells is wavy (Fig.3) and there is no significant difference in size between these two types.

Within the gland cell cluster arises the collecting duct, the connection to the intracellular duct. The intracellular duct (ICD) is formed by the gland cell itself. Its wall consists of microvilli placed at the storage pole of the gland cells where the secretory vesicles accumulate in the cytoplasm (Fig.4). These microvilli seem to be partly open-ended. Actin filaments are clearly seen within the microvilli (Fig.5). The lumen of the ICD is very narrow and filled with the electron dense content of the secretory vesicles (Fig.6).

The collecting duct is formed by a single cell layer and has no special differentiation of the inner lining. Inside the cytoplasm of these duct cells a great number of microtubules is obvious and the cells have a round nucleus (Fig.7). The collecting duct connects to the storage poles of a number of gland cells, where the ICD and the secretory vesicles are located (Fig.8a &b). This duct system shows no visible lumen. Within a gland cluster a various number of

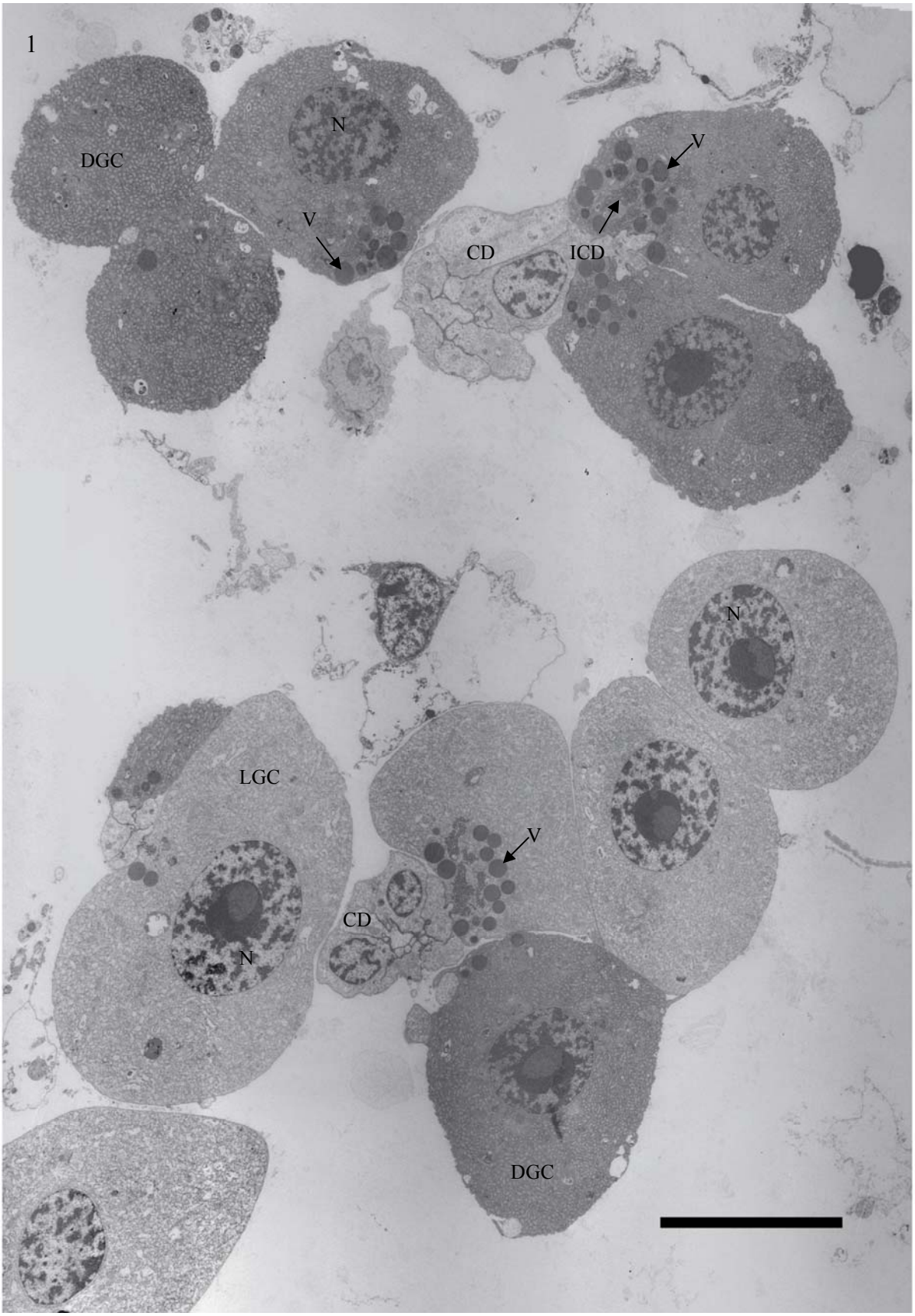


Fig.1: Cluster of round and oval cement gland cells showing with collecting duct (CD), the intracellular duct (ICD) within the glands and the secretory vesicles (V). Light gland cells (LGC) and dark gland cells (DGC) and also nuclei (N) with only one nucleolus are clearly to identify. Scale 10μ

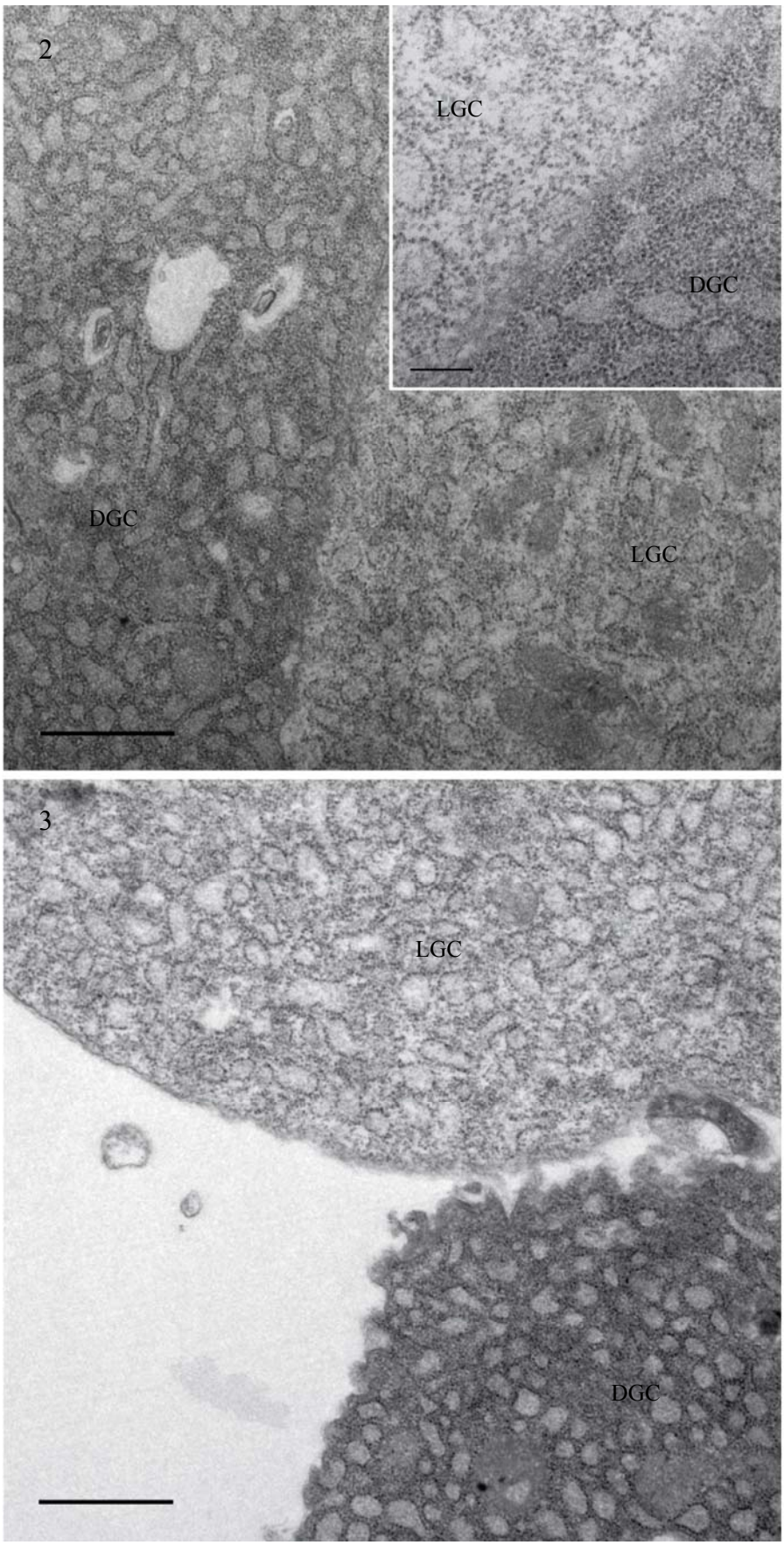


Fig.2: Light (LGC) and dark gland cell (DGC) with vesicular ER. Scale 1µ

Insert: Cytoplasm at higher magnification showing vesicular ER and a higher number of ribosomes in the dark cell. Scale 0.2µ

Fig.3: Wavy contour of dark gland cell (DGC) in comparison to the contour of the light gland cell (LGC). Scale 1µ

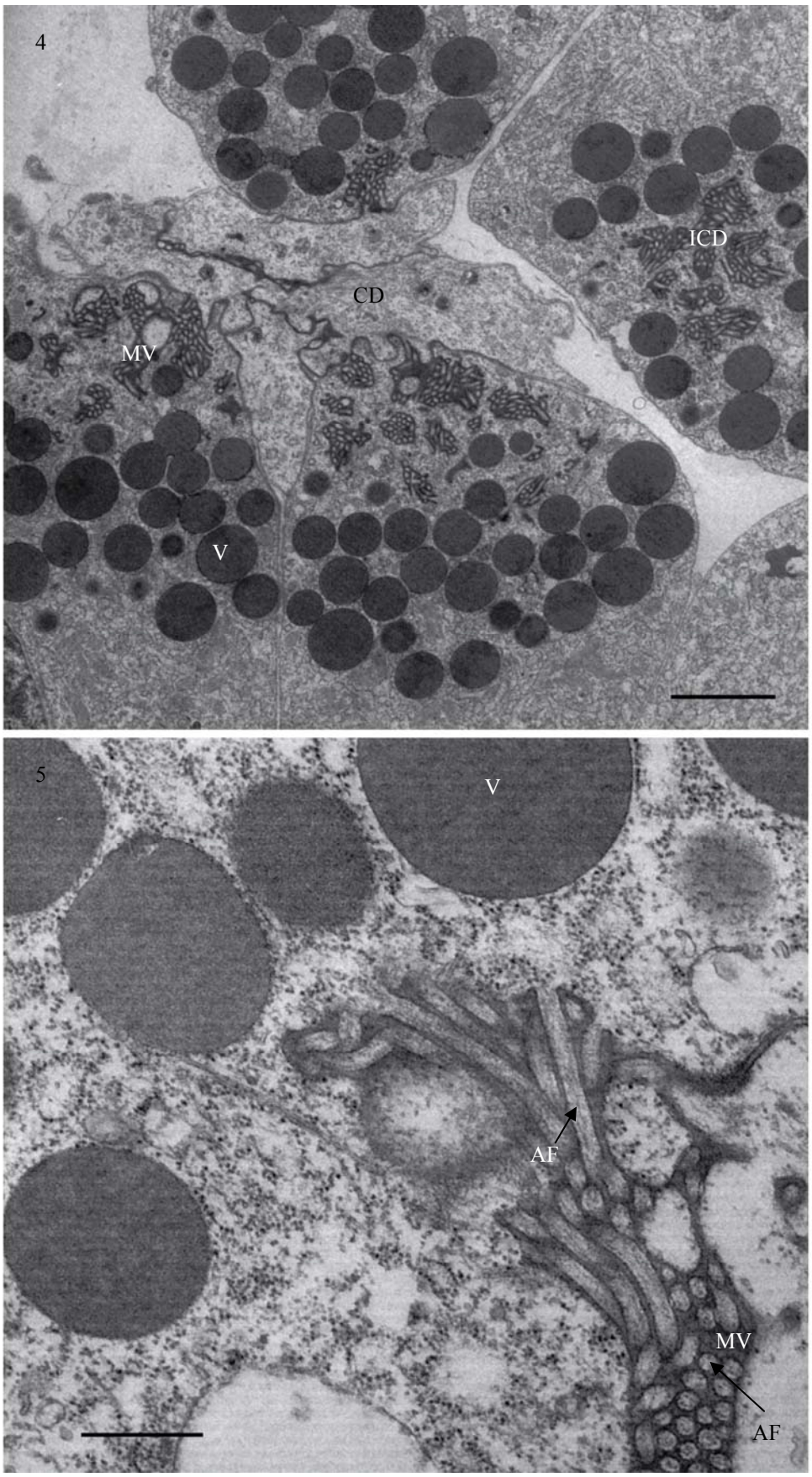


Fig.4: Accumulation of vesicles (V) and microvilli (MV) of the intracellular duct (ICD) at the storage pole. The collecting duct (CD) is in between the gland cells. Scale 2µ

Fig.5: Section of microvilli (MV) in the intracellular duct showing actin filaments (AF). Scale 0.5µ

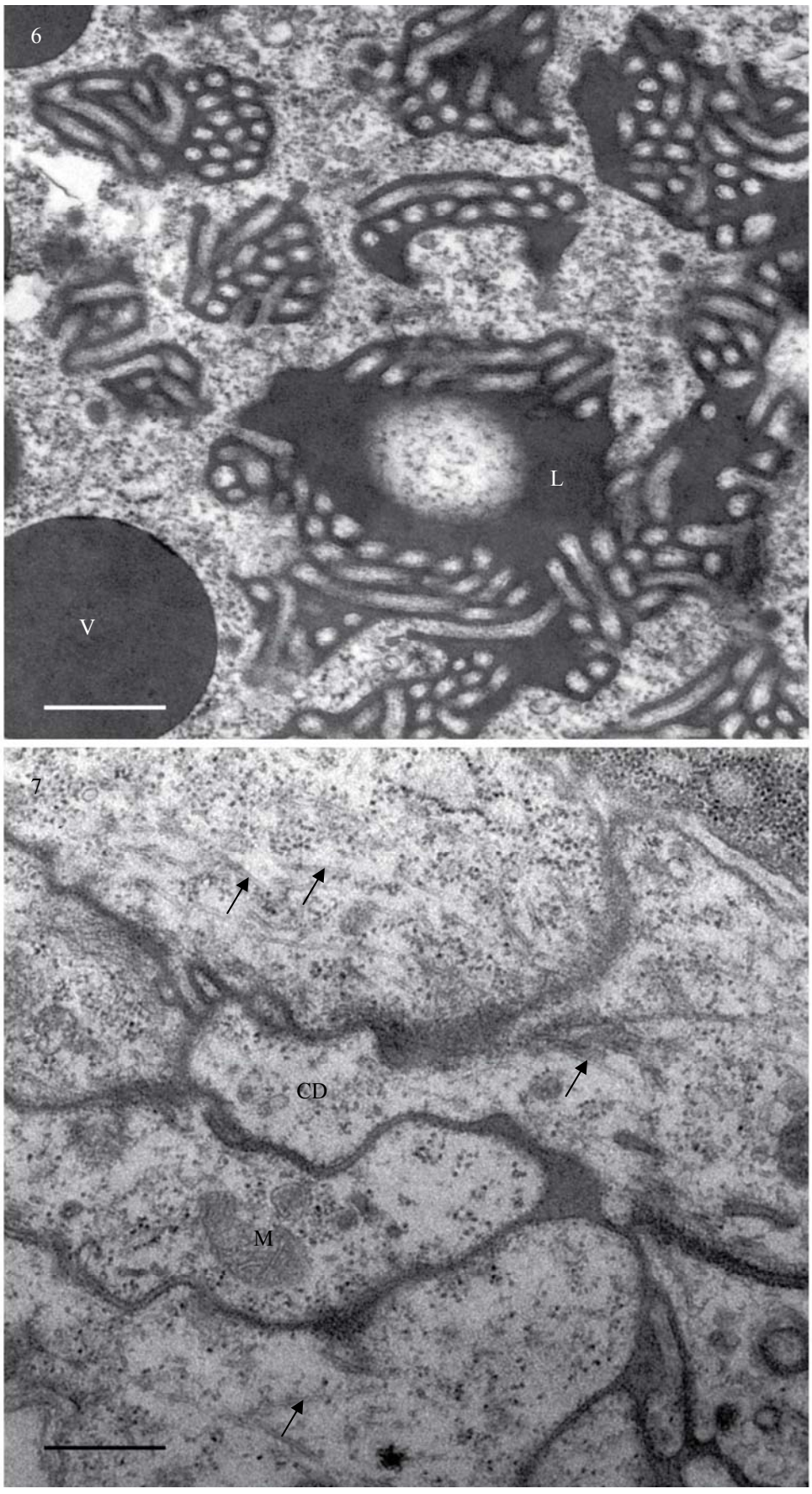


Fig.6: The lumen (L) of the intracellular duct filled with the same electron-dense content as the secretory vesicles (V). Scale 0.5µ

Fig.7: Cytoplasm of the collecting duct (CD) showing single microtubules (Arrows) and a mitochondrion (M). Scale 0.5µ

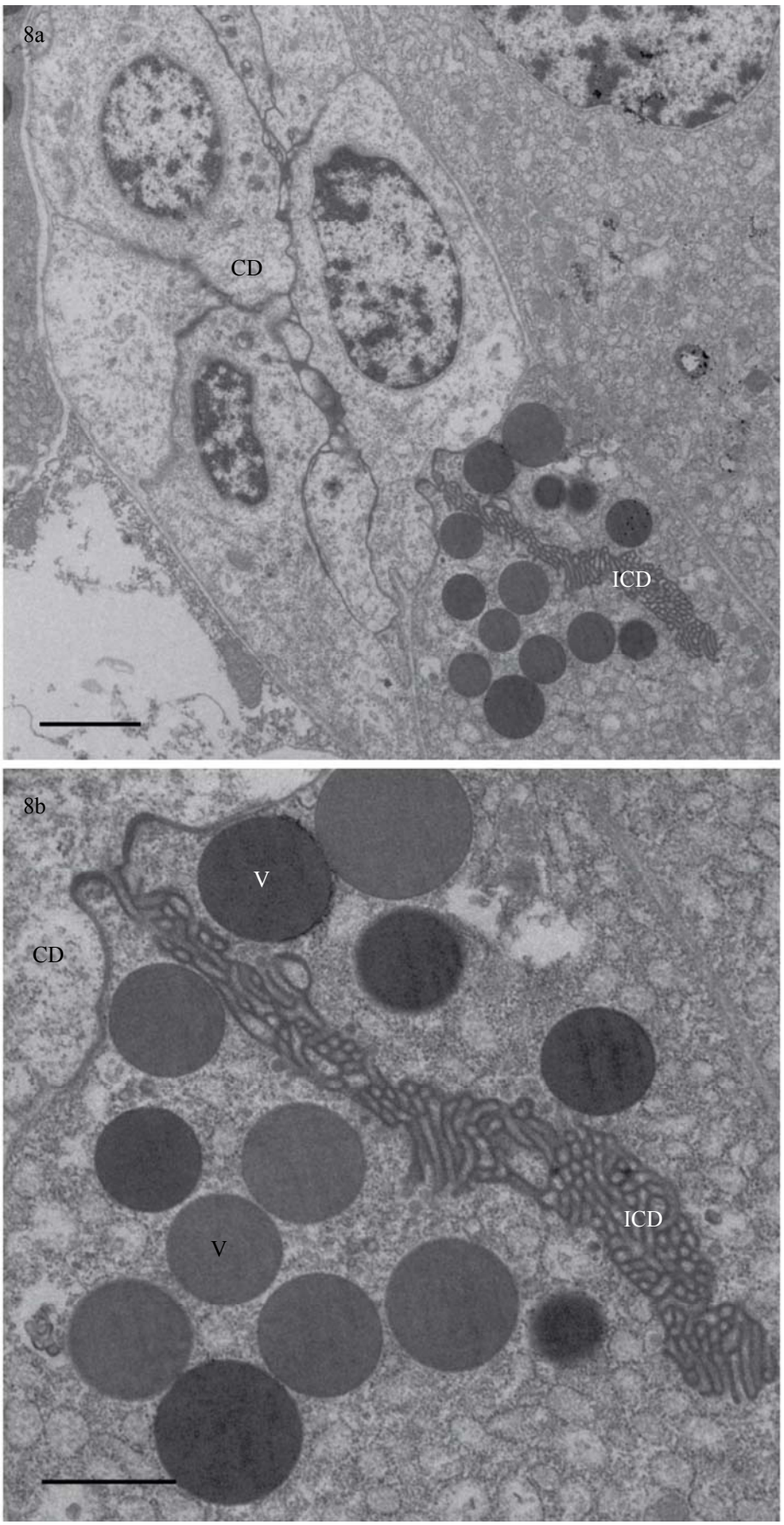


Fig.8:a) An overview of the connection between the intracellular duct (ICD) within the gland cell and the collecting duct (CD). b) Higher magnification of intracellular duct (ICD) and collecting duct (CD). Numerous vesicles (V) are significant at this transition. 8a: Scale 2μ; 8b: Scale 1μ

collecting ducts is detected. It could be one collecting duct sectioned at different sites but according to the literature there are many ducts. The same applies to the primary duct.
The primary duct is cuticle-lined and opens through the cuticle of the base. These openings occur singly or multiply (Fig.9 - 11) and only few granular components of the electron-dense content of the vesicles are visible inside their lumen. The progression of this duct is meandering through the mantle tissue and is identified as the continuation of the collecting duct (Fig. 12) showing a lumen that is mostly filled with the same electron-dense content as the secretory vesicles (Fig.13).

The average diameter of the secretory vesicles is between 0.5µm – 2.5µm which is smaller than in adults and their content is homogenous and highly electron dense. Around some vesicles appears an even higher electron-dense lining which is aligned at different regions of the vesicle (Fig.14). Increased density of this lining is combined with an increased disaggregation of the content. Vesicles with a homogeneous electron dense content never show this lining. At higher magnification (x20.000) it looks as if polysomes eventually accumulate here because the lining has patches that appear similar to the single granules (Fig.15). There are also some indications that the content of the secretory vesicles is released into the lumen of the ICD via exocytosis (Fig.16 & 17).
The nucleus of the cement glands in subadult *Semibalanus balanoides* has only one nucleolus (Fig.18 & 19). Most of the nuclei are round or oval with an average diameter between 5 and 12µm. This is in contrast to the bigger and mostly lobed nuclei which are usually found in the cement cells of adult barnacles. The ratio heterochromatin: euchromatin in the nuclei of the gland cells of the subadult animals is approximately 50: 50. On the inner cell membrane there is a higher accumulation of euchromatin and also some pores of the nucleus are visible.
The rough endoplasmic reticulum is vesicular or it forms laminar staples (Fig.20). At the secretion pole of the gland cells the staples are often arranged in loops (Fig.21). Ribosomes,

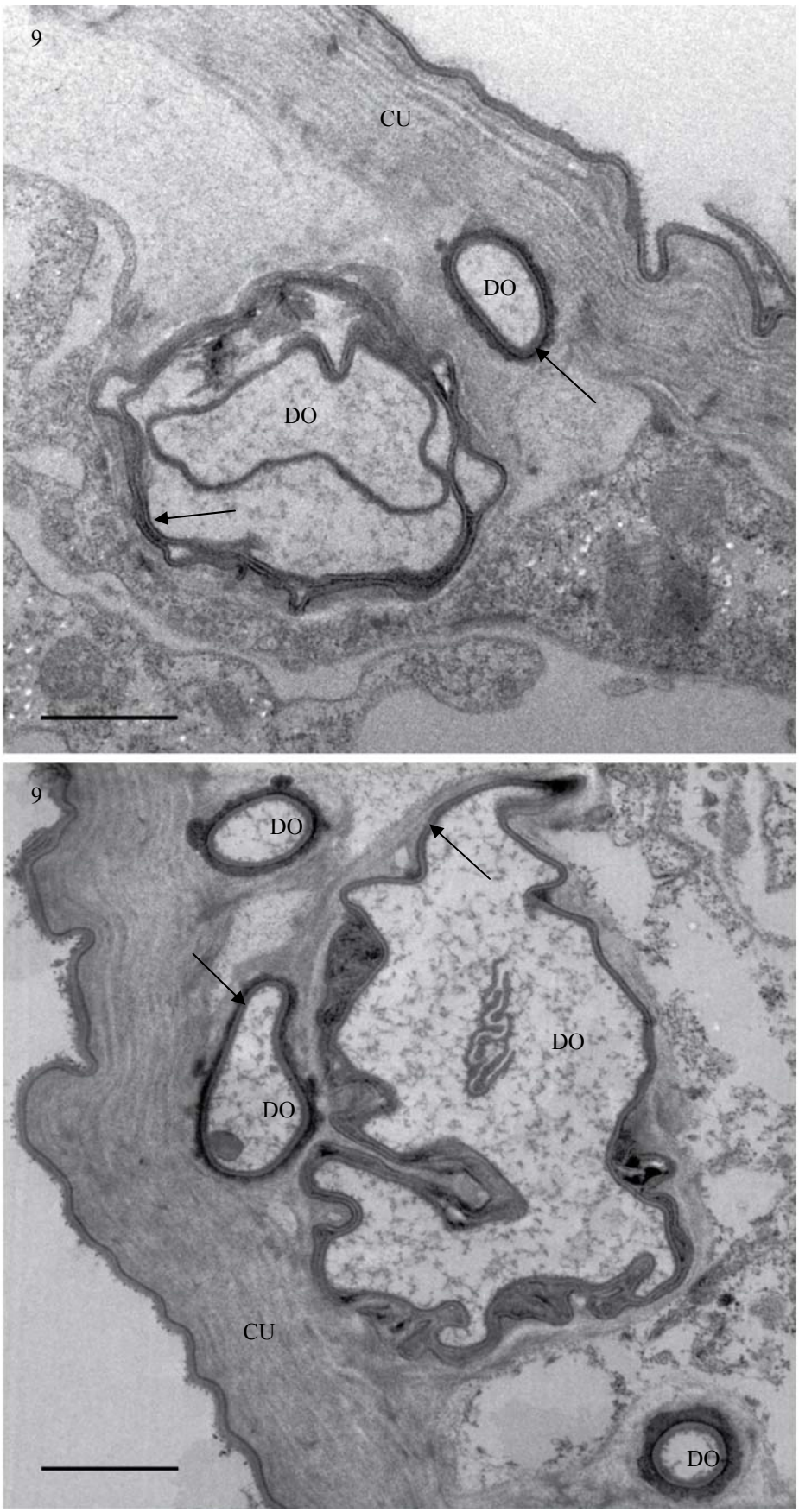
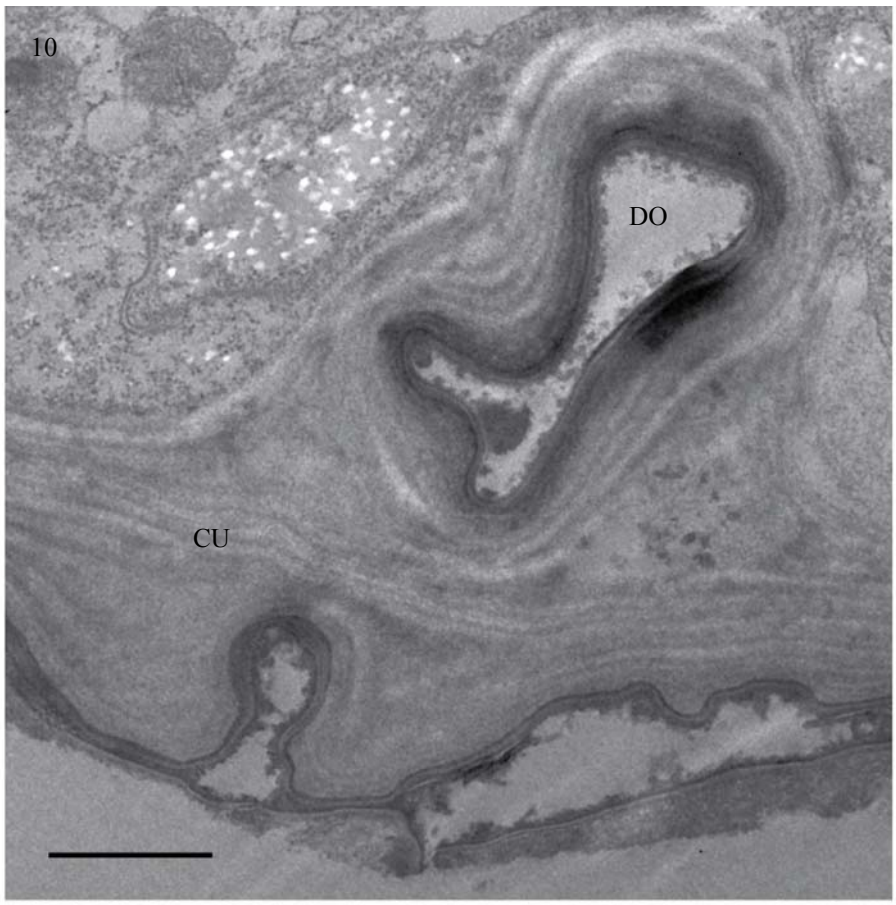


Fig.9: Multiple duct openings (DO) of the primary duct at the cuticle of the base (CU). The walls of the duct are clearly cuticle-lined (Arrows). Inside the lumen the content is granular with unidentified inclusions. Scale 1µ



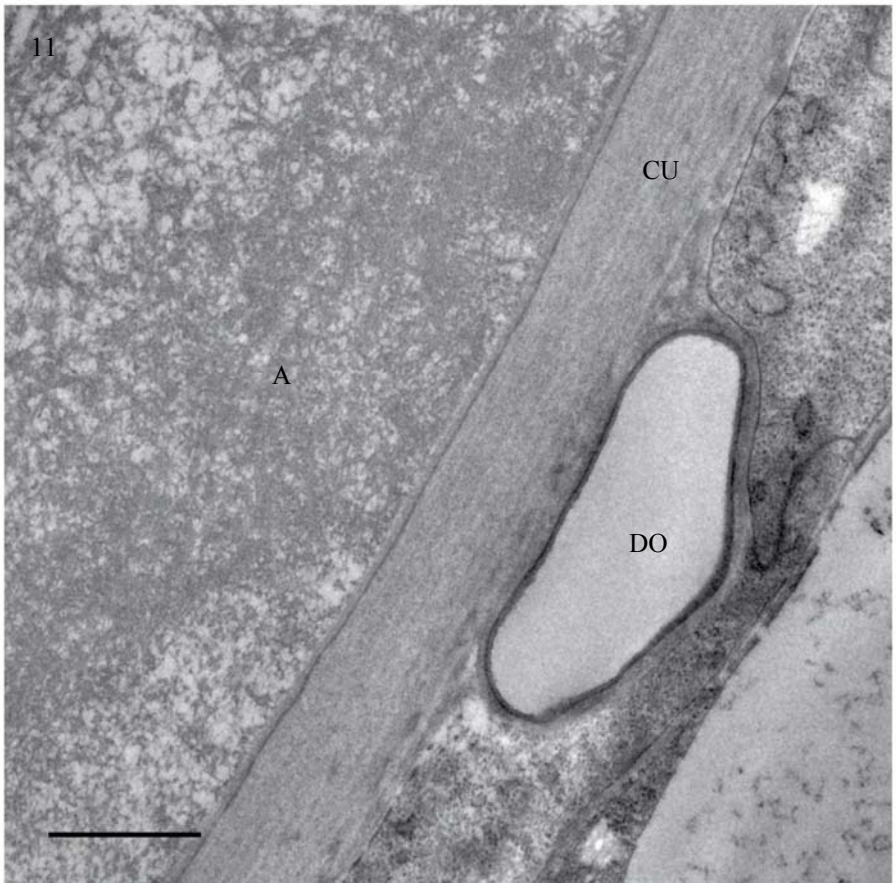


Fig.10: Single primary duct opening (DO) in connection with the cuticle of the base (CU). The lumen shows an apparently granular content and on the outside of the base some small amounts of adhesive are detected. Scale 1µ

Fig.11: Primary duct opening (DO) presenting an empty lumen. Outside the cuticle the already extruded adhesive (A) is visible. Scale 1µ

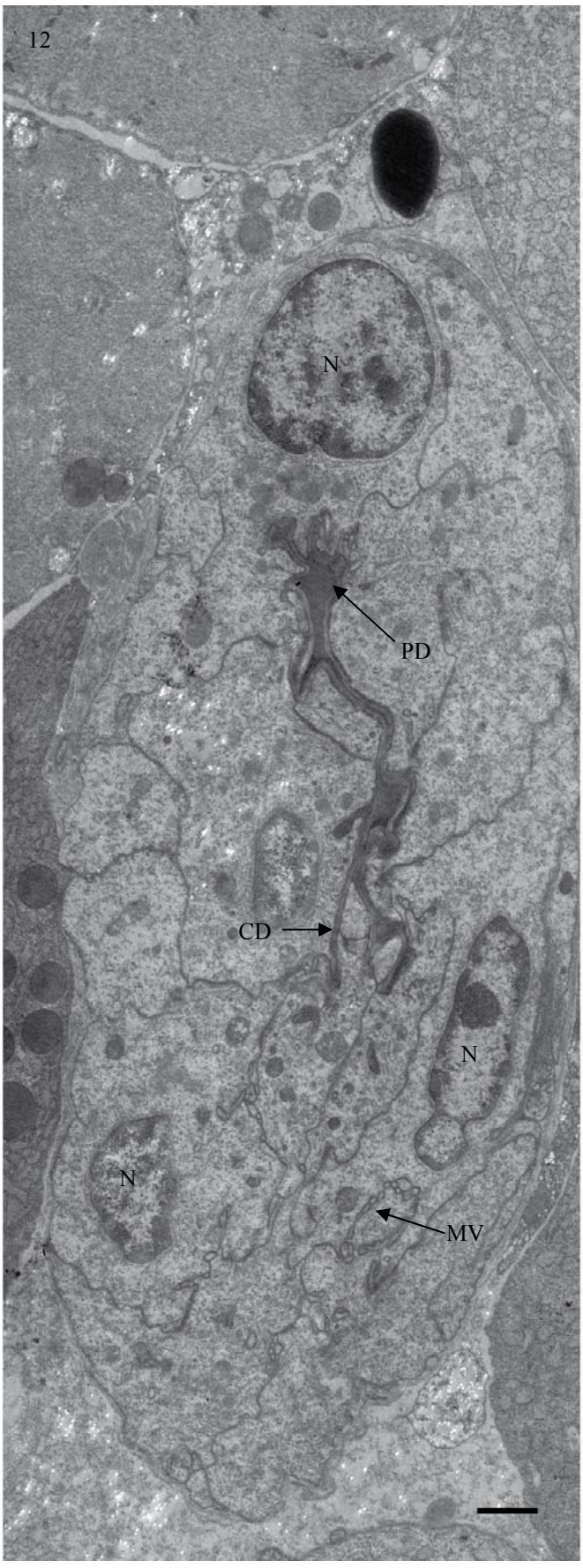


Fig.12: Overview of all three sections of the duct system. Transition between collecting duct (CD) and intracellular duct, identified by microvilli (MV), and the cuticle-lined primary duct (PD). Within the single cell layer of the collecting duct round or elongated nuclei (N) are clearly visible. Scale 1µ

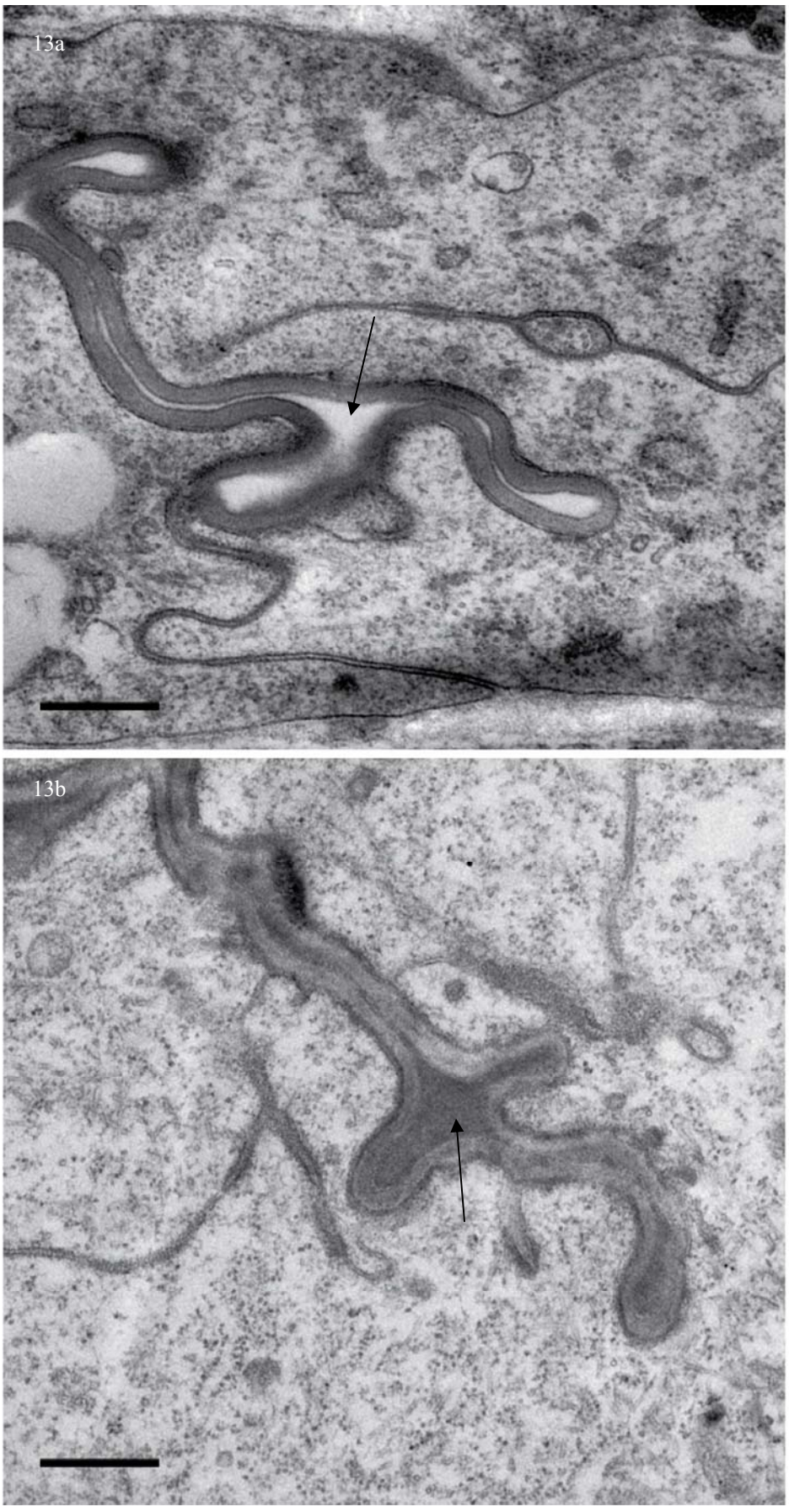


Fig.13: The cuticle-lined primary duct showing a) an empty lumen and b) a lumen that is filled with the same homogenous electron-dense material (Arrow) as in the secretory vesicles. Scale 0.5µ

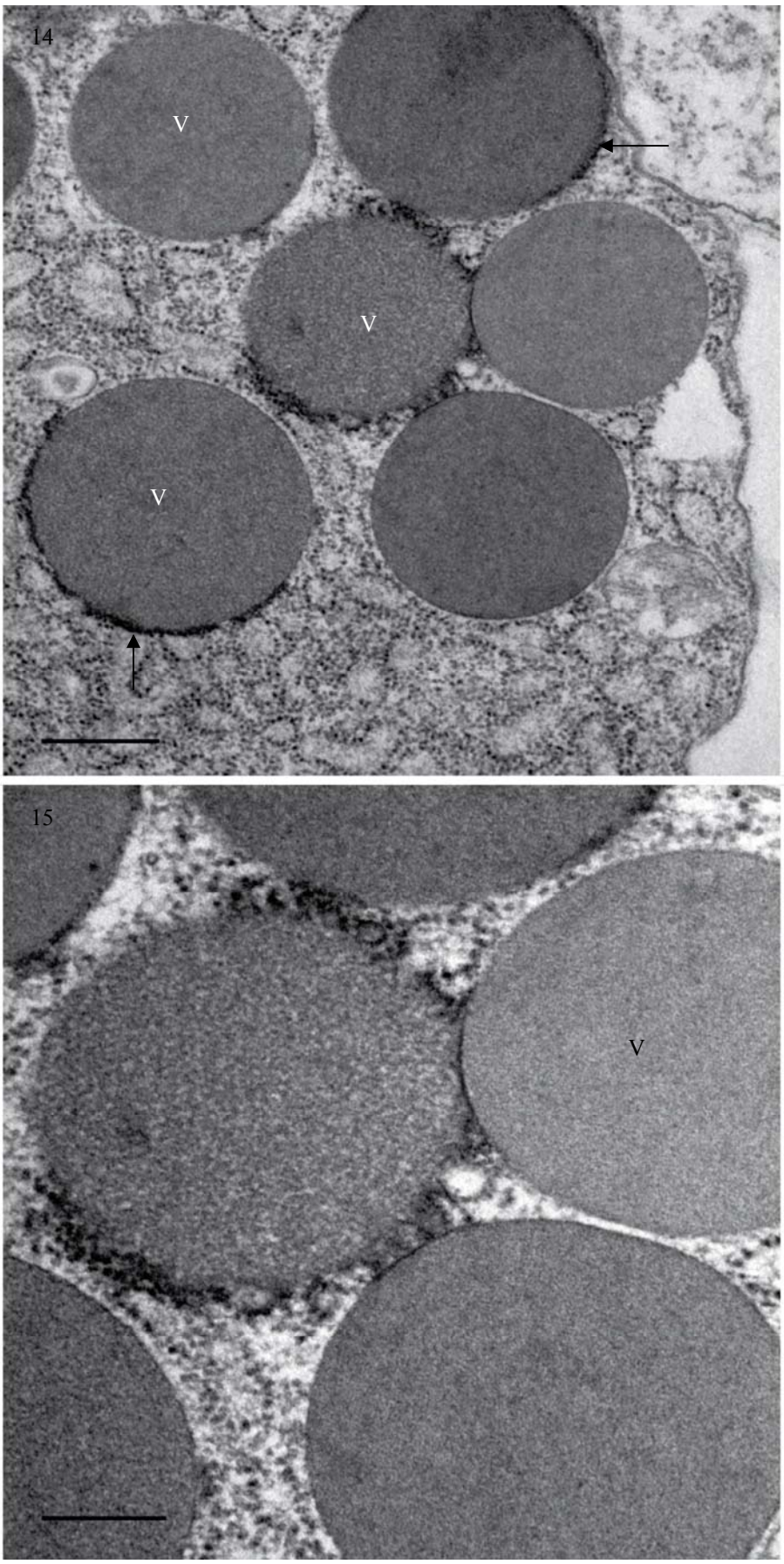
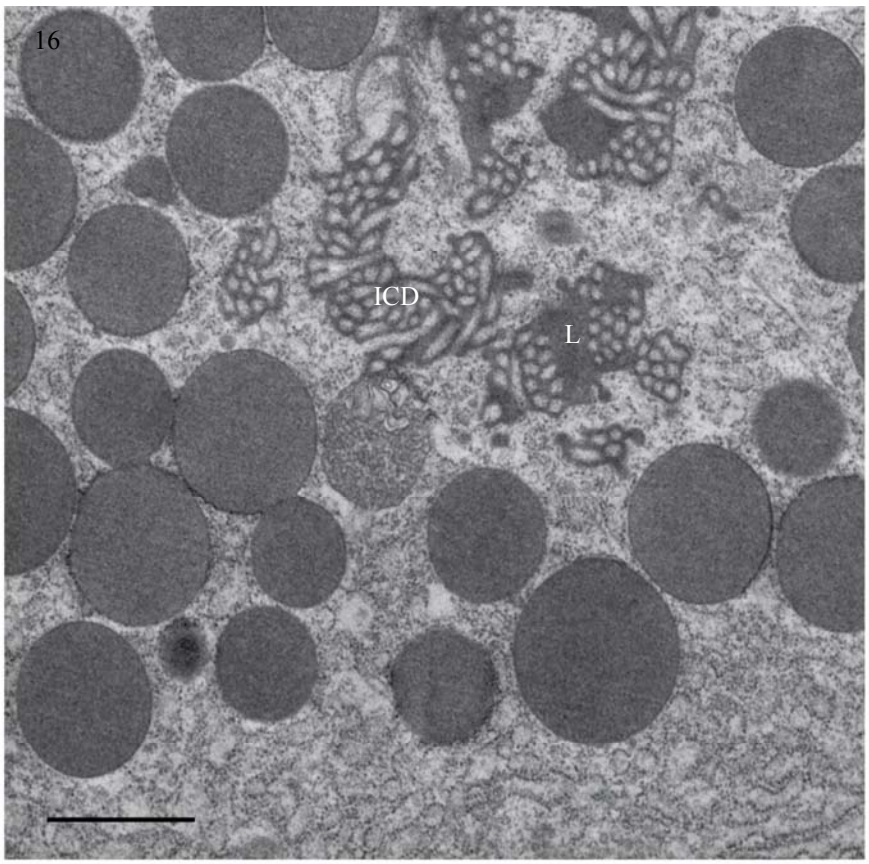


Fig.14: Secretory vesicles (V) with an electron-dense lining around their membrane. The lining is not continuous (Arrows). Scale 1µ

Fig.15: Higher magnification of Fig.14 showing the enhancement of the electron-dense lining. The lining seems to consist of polysomes. At the same time the vesicle content is granular and presumably in the stage of disaggregation. Scale 0.2µ



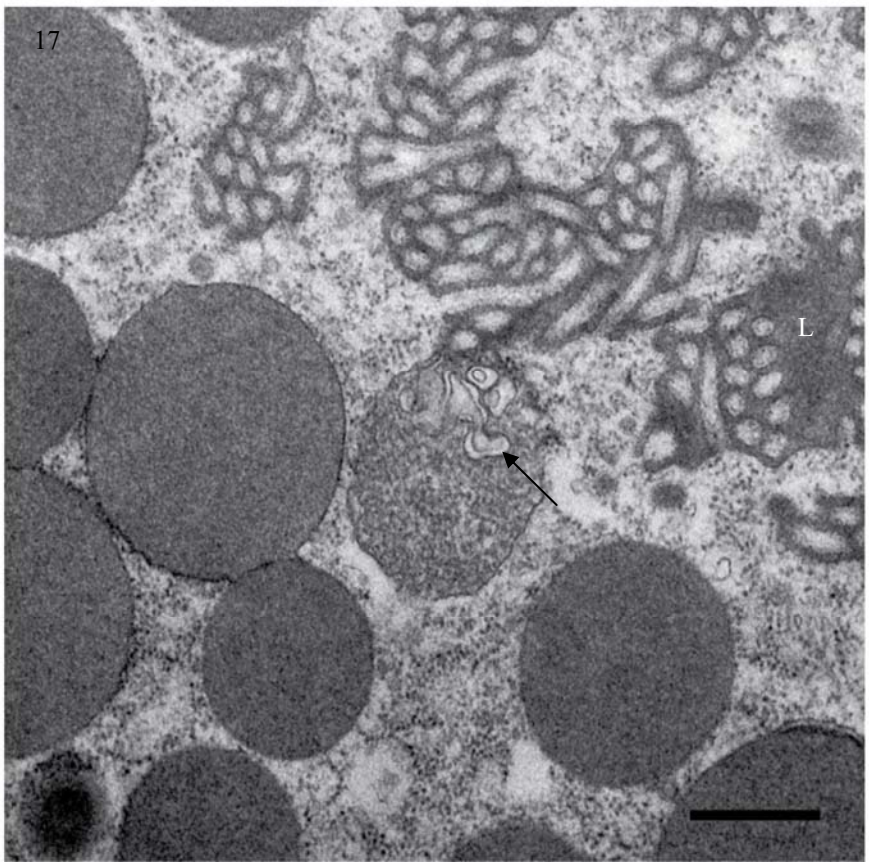
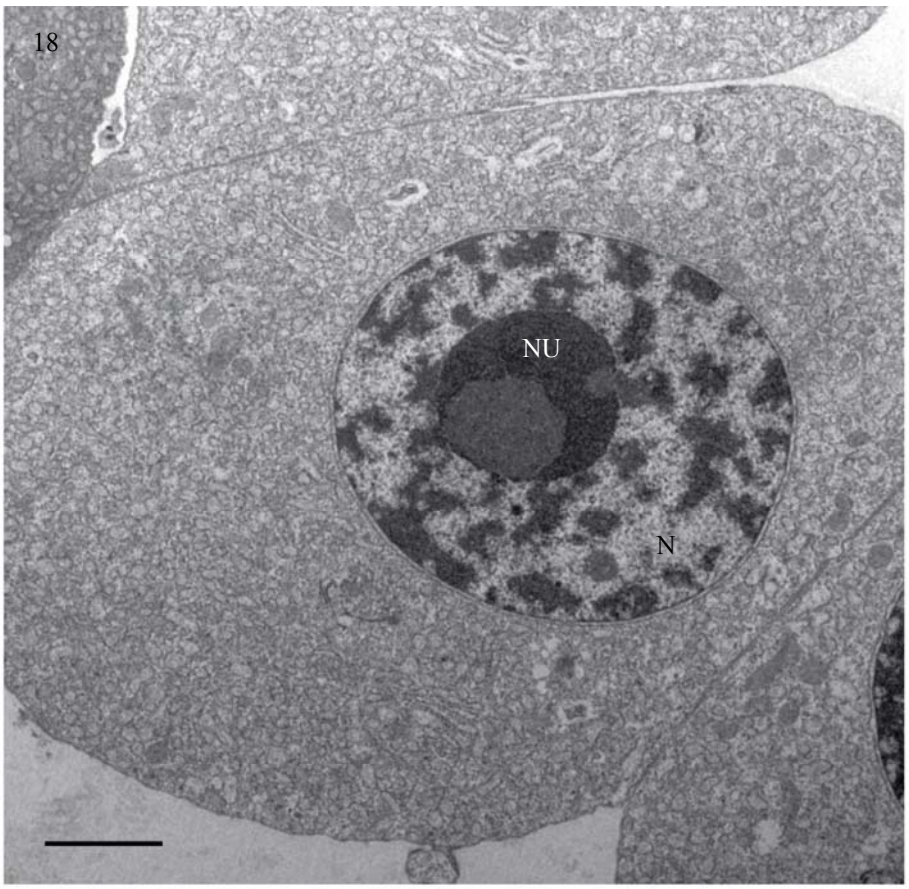


Fig.16: A potential uptake of the vesicle content by the intracellular duct (ICD). The content is already highly disaggregated and seems to be transported into the lumen (L) of the duct. Scale 1µ

Fig.17: Higher magnification of the potential uptake. Membranous structures are seen within the granular vesicle (Arrow). Scale 0.5µ



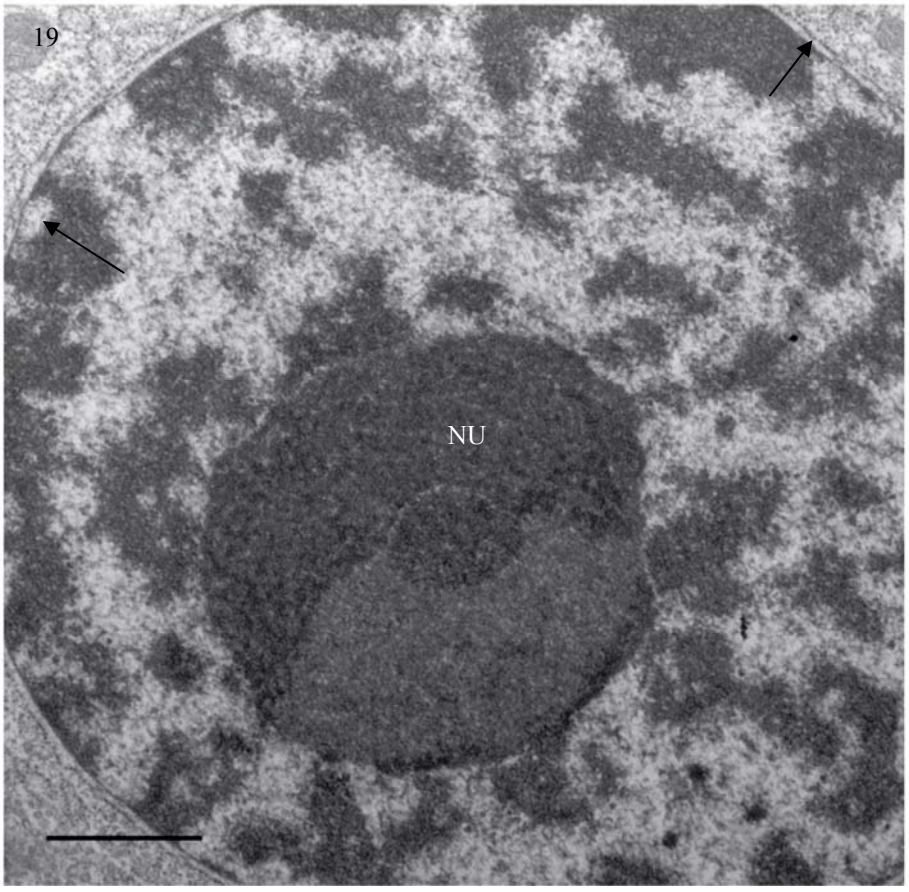
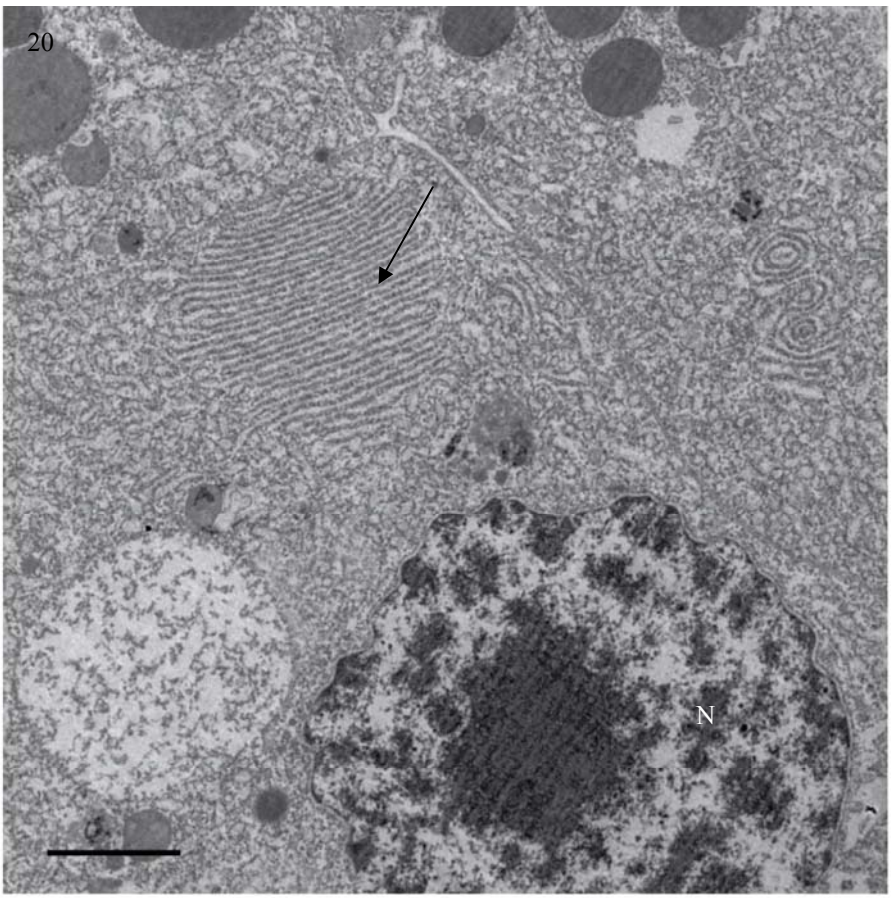


Fig.18: Gland cell with a round nucleus (N) placed centrally in the cytoplasm of the cell and the single nucleolus (NU). Scale 2µ

Fig19: Higher magnification of the nucleolus (NU). Note the double membrane and some pores of the nucleus (Arrows). Scale 1µ



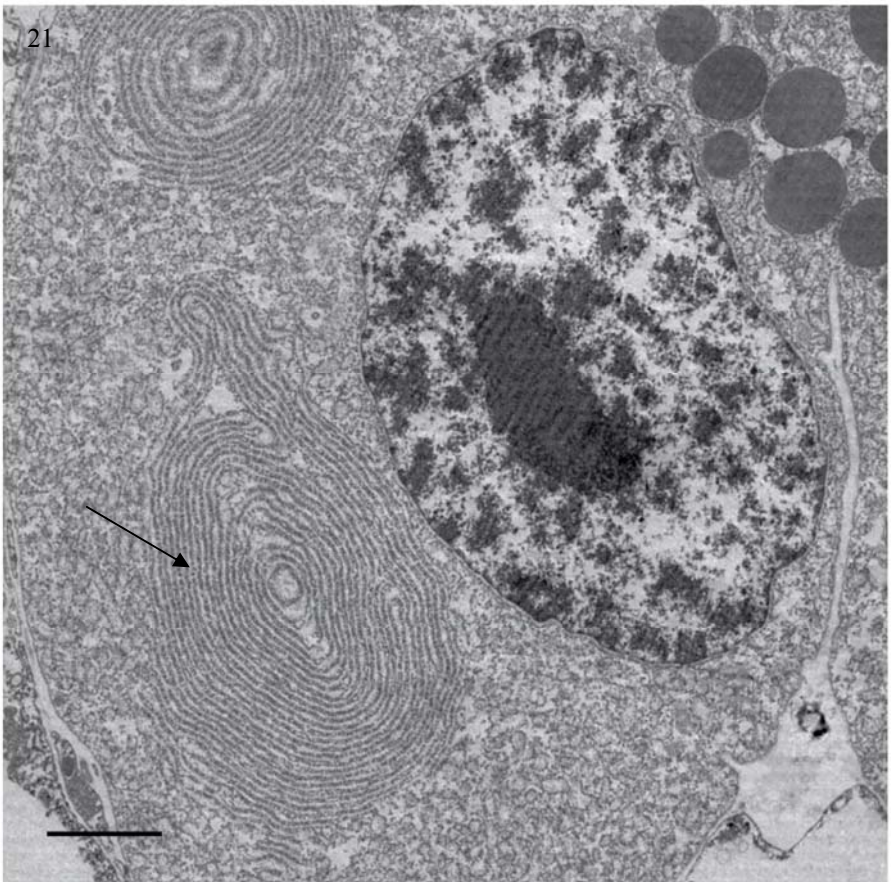


Fig.20: Stapled laminar rough endoplasmic reticulum (Arrow) placed inside the cytoplasm next to the nucleus (N). Scale 2µ

Fig.21: Rough endoplasmic reticulum arranged in stapled loops (Arrow) at the secretion pole of the gland cell. Scale 2µ

mitochondria, Golgi bodies and in association lysosomes of different size and shape are distributed all over the cytoplasm.

Cement is found outside the cuticle on the basis of the animal. This cement is not distributed over the whole base but forms patches. These individual patches consist of fibrillar or homogenous electron-dense material (Fig.22). When they are fibrillar, the fibers are arranged irregularly. Some patches show an obvious zonation. The zones have the same structural material and are separated by an electron-dense line. This line is orientated almost parallel to the base (Fig.24 - 26). The side of the cement facing the substrate sometimes includes algae or micro-organisms (Fig.23).

The cuticle of the base often forms foot-like elevations. The gaps between these elevations are mostly filled with adhesive material either of the same or of different structure. The cuticle may also show different electron density (Fig.27).

A summary of the results in comparison with the cyprid and the adult stage is given in table 1.

	Cypris larvae	Subadult	Adult
Gland cell organisation	2 clusters / α & β-cells	spreading clusters / no β-cells	single gland cells / no β-cells
Size of gland cells	120 x 120 x 60µm	up to 38µm in diameter	up to 250µm in diameter
Shape of gland cells	round or oval	round or oval	elongated and lobed
Supporting extrusion mechanism	Muscular sacs	depressor muscles	depressor muscles
Location	behind compound eye	lateral-ventral in mantle tissue	lateral-ventral in mantle tissue
Size of Vesicles	undescribed	0.5 - 2.5µm	3 - 4µm
Duct system	probably ICD, CD and PD	ICD, CD and PD	ICD, CD and PD
Adhesive	temporary / single layer	temporary - permanent / zonation	permanent / zonation
Movement	partially motile	sessile	sessile

Tab.1: Comparison of the cement apparatus in cypris larvae, subadult and adult animals. Data of the cypris larvae according to Walker (1971, 1973, 1992), Nott (1969) and Okano (1996), data of the adult taken from Walker (1970, 1992), Lacombe (1970) and Khandeparker (2007)

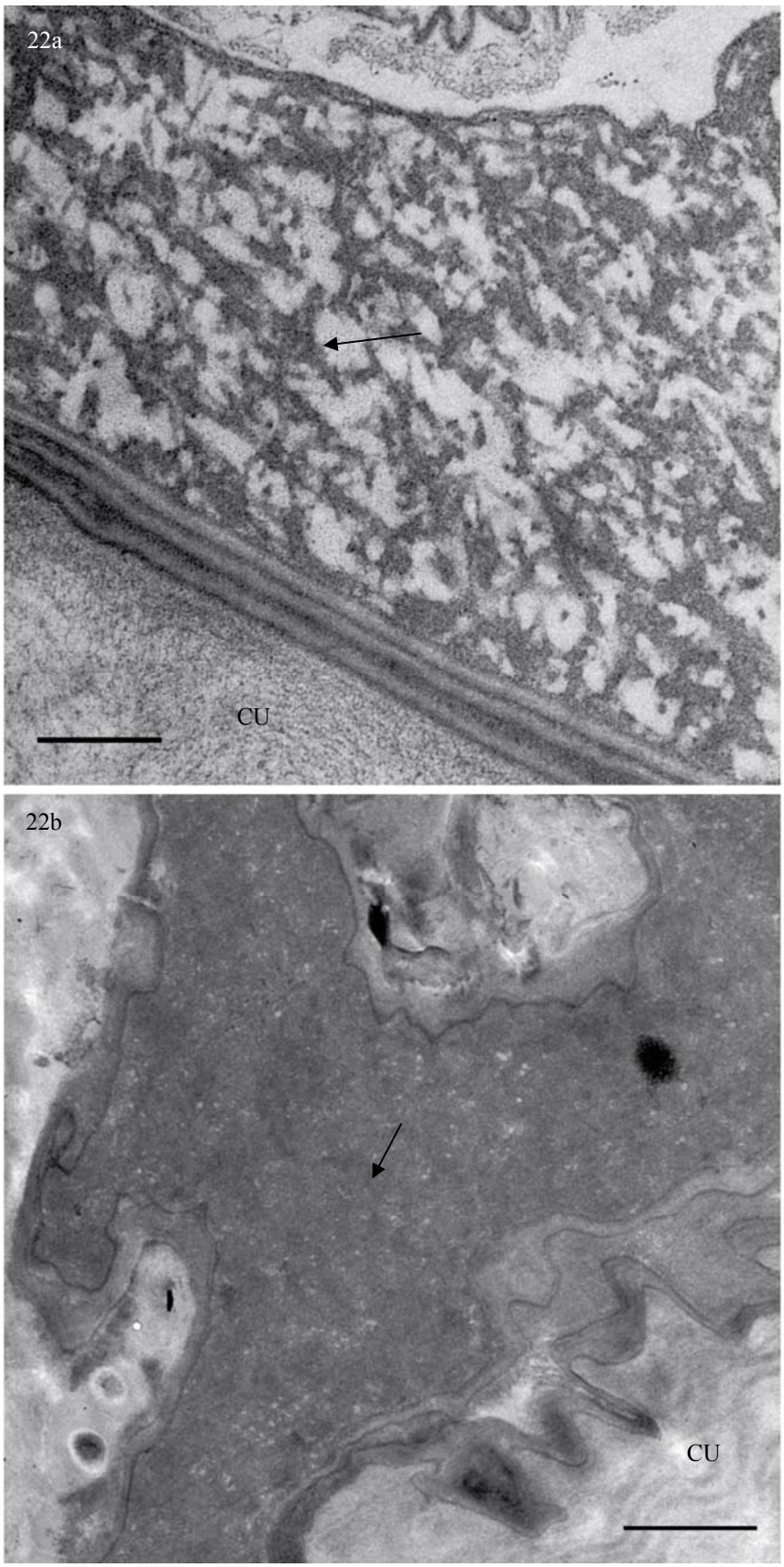
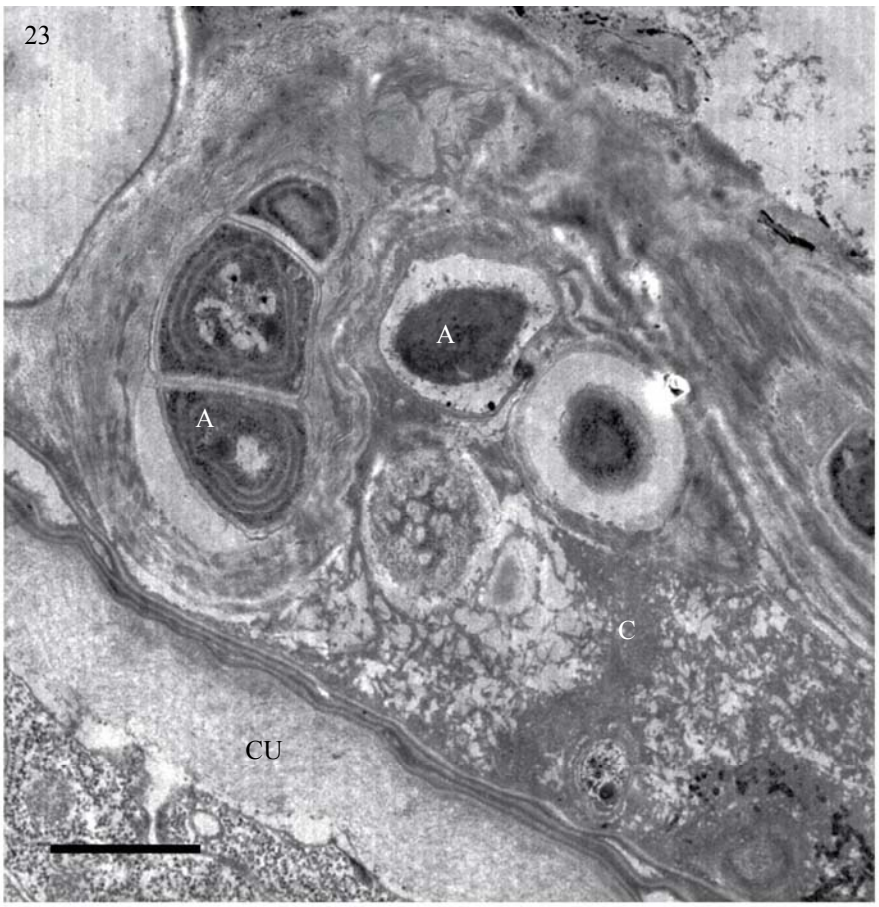


Fig.22: The material extruded by the duct system forms patches of a) fibrillar and b) homogenous electron-dense material (Arrows). Additionally the cuticle of the base (CU) is visible. Scale 22a: 0.2µ; Scale 22b: 1µ



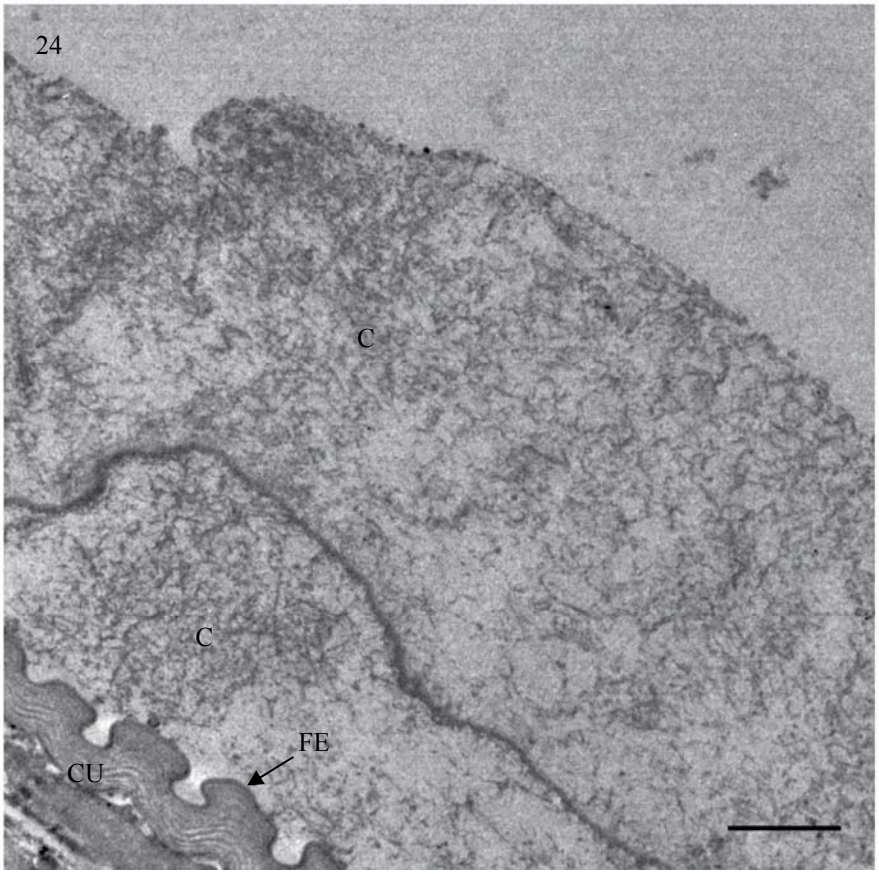


Fig.23: Incorporations of algae (A) inside the extruded cement (C). Around the algae the cement condenses. Scale 1µ

Fig.24: Electron-dense lining separating the patch into two zones of similar structural material. Cuticle (CU) forms foot-like elevations. (FE)

Scale 2µ

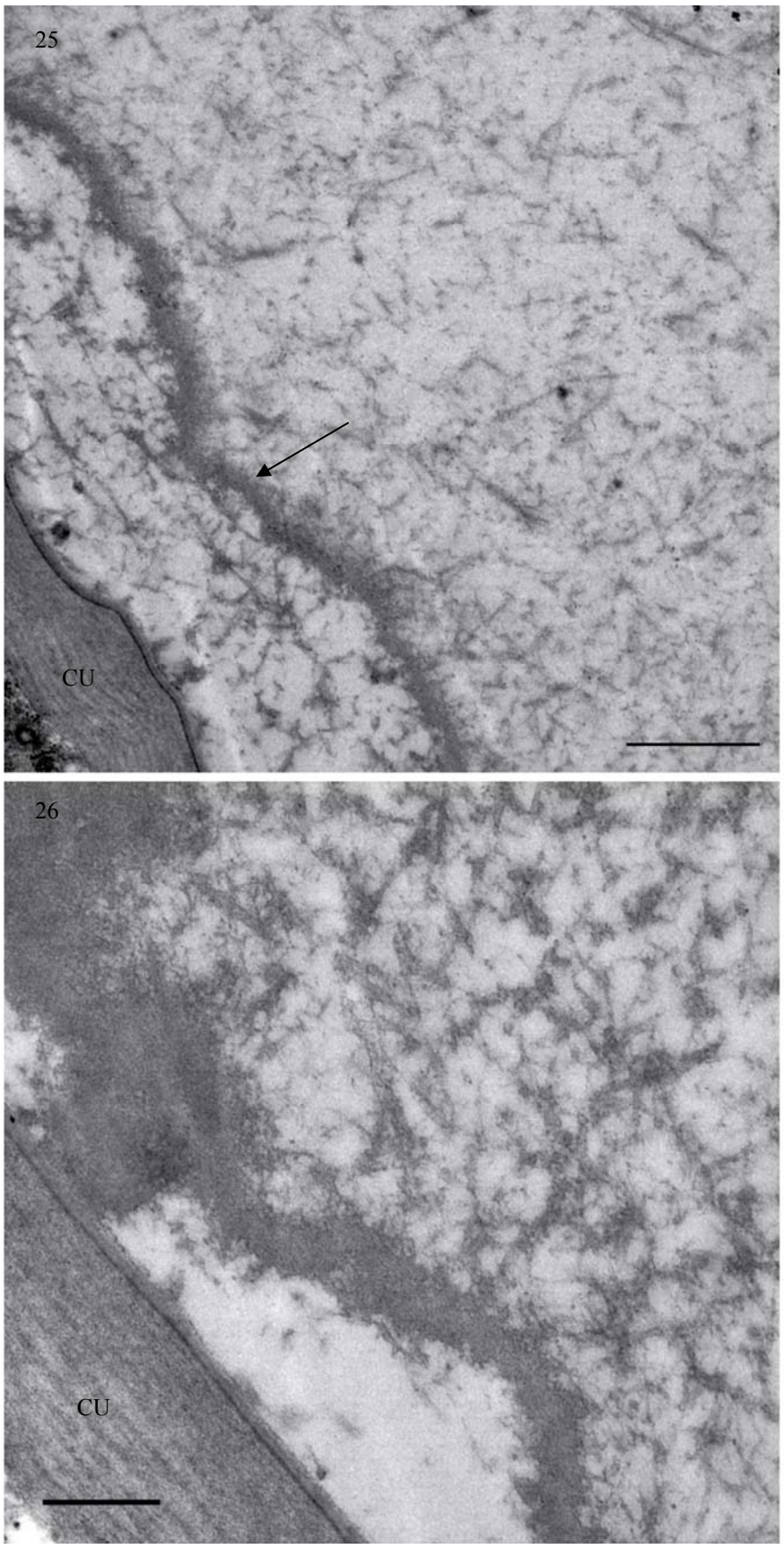


Fig.25: Electron-dense line (Arrow) running almost parallel to the cuticle of the base (CU). Scale 1µ

Fig.26: Note the difference in electron-density between the line and the extruded fibrillar material at higher magnification. Scale 0.5µ

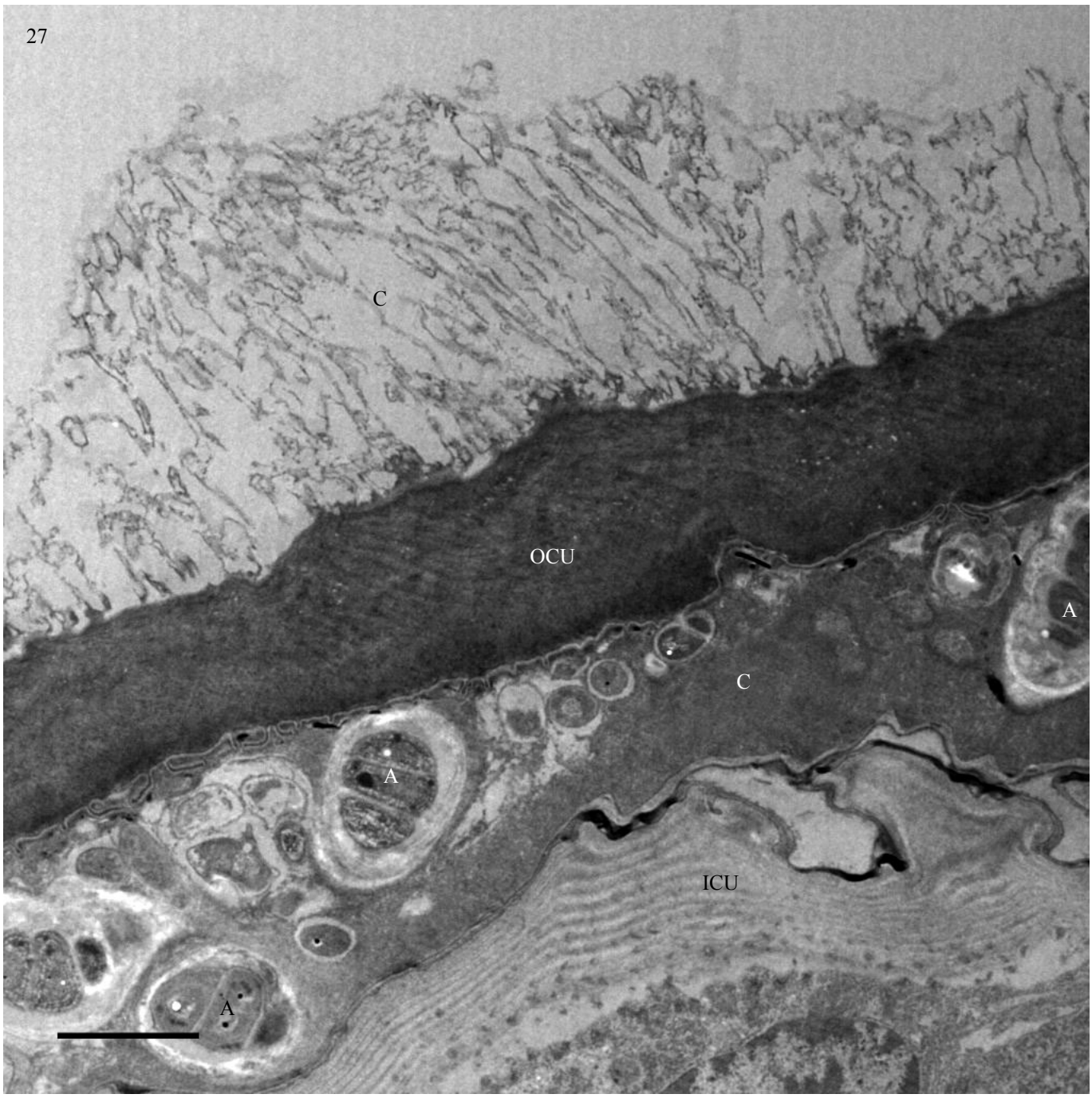


Fig.27: Two sections of cuticle distinguished in an outer and inner part. Between the outer (OCU) and inner section of the cuticle (ICU) there is a number of algae (A) included in the electron-dense cement. Outside the outer section of the cuticle the cement (C) is fibrillar and contains no inclusions. Scale 2µ

Discussion

The investigation of subadult Semibalanus balanoides reveals interesting transitions in the development of the gland cells between the larvae and the adults.
Lacombe (1970, p.169) postulated that the cement glands of adult *Semibalanus balanoides* „never appear as a distinct group of cells or as a rosette shaped cluster“. In this investigation, the cement glands of subadult organisms appear as clusters, as in newly settled cyprids. In addition to this the description of the cyprid gland cells by Walker (1971) indicates that there are α and β-cells. After settlement the β-cells disappear and only the α-cells remain as gland cells. In the observed samples no β-cells were identified which leads to the conclusion that *Semibalanus balanoides* had already decomposed this cell type, which occurs according to Walker (1973) five days after settlement. In the same paper it is stated that the occurrence of single nucleoli in the round nuclei, the orientation of gland cells around a collecting duct and the further dispersal of this cluster start around the 7^{th} day after attachment. In this study the determination of the exact attachment date and therefore the exact definition of age of each animal is not possible. But the still slightly clustered cement cells, the absence of β-cells and only one nucleolus in the nuclei of these cells are a clear indication of a young transitional developmental stage. Also the smaller size and shape of the nuclei, in contrast to the “relatively large lobulate nuclei with numerous nucleoli” as described by Walker (1992, p. 289) supports this theory.

The different electron-density of the darker gland cells is presumably caused by a higher number of ribosomes. This is just an assumption because counting of the ribosomes was not feasible and the statements only rely on visible differences in the electron microscopic photographs. But it could be an indication for an increased proteinsynthesis rate. Therefore a greater number of vesicles is expected. But this is not the case and the vesicles in the dark

gland cells are of about the same size as those in the light gland cells. There is also no difference in the number and appearance of mitochondria, Golgi-bodies or nuclei. Another assumption is that the darker gland cells are in a stage of beginning cell death because of their undulated contour. But the fact that the nuclei are active and that all the other cell organelles are present weakens this consideration. Ödling et al (2006, p.961) postulated that "it is possible that only some of the cement-secreting cells participate in secretion during a settling attempt, sparing other cells in order to provide the cyprid with the possibility to multiple attempts of settling." This argument could explain the different appearance of the glands caused by their secretion period. But further research needs to be done.

The duct system is special in a certain way because the lumen of the intracellular duct and the lumen of the primary duct are sometimes filled with electron-dense content. The collecting duct lying in between shows no visible lumen, neither filled nor empty. It is presumably collapsed.

Besides this, the lumen of the ICD shows the same homogenous content as the secretory vesicles and in certain regions around the microvilli membrane-remains were detected. This is eventually the beginning of the release of the adhesive because the proteinaceous content is stored in the vesicles of the gland cells right before extrusion through the duct system as it is described in the literature. This release of cement happens very quickly (Okano, 1996).

The investigation also shows that the primary duct is not widely ramified in subadults as it is in the adult specimen of *Semibalanus balanoides*. The openings of the primary duct are almost empty except for a few granular inclusions, presumably leftovers of cement which did not get extruded. To ensure a save transport of the presumably viscous proteinaceous content inside the duct system the biochemical reaction which hardens the cement must occur outside the duct when it gets in contact with seawater. Otherwise the content of the vesicles would harden inside the duct and block it.

Around some secretory vesicles at the storage pole near the ICD an electron-dense lining is visible. The content of these vesicles is granular in contrast to the homogenous electron-dense content of the non-lined vesicles. Reasons for this appearance could be a decomposing process or the start of the release of the proteinaceous content into the ICD by exocytosis. The fact that the lined vesicles are in close relation to the ICD seems to confirm the second theory. Unfortunately a clear uptake of the vesicle content is not detected.

Inside the cytoplasm Golgi bodies appear as small stapled cisterns with pinched off vesicles. The mitochondria occur in different but mostly elongated form, in most cases clearly showing transverse or oblique cristae. As previously described the rough ER is organized in vesicular and stapled lamellar form, distributed all over the cytoplasm, or in stapled looped form at the secretory pole. There is also the presence of lysosomes within the glands, which points to the process of decomposition. All these structures are not significantly different to that of the cytoplasm in adult *Semibalanus balanoides.* This strongly indicates another similarity between the subadult and the adult animal.

Outside the cuticle on the base extruded cement accumulates and sometimes forms a visible zonation. This could result from several different mechanisms. Walker (1971, p.211) believed that “sea water may have a direct effect on the outer layers”, or that “the tanning process may require some material from the environment, i.e. oxygen”. Also a stepwise attachment or reattachment after a disturbance, e.g. caused by tidal waves, predators or other biological/mechanical influences could form these zones. The observed layers show fibrillar and homogenous electron-dense structures what might be developmental stages of the cement hardening.

Another idea is that shortly before extruding the content it is modified within the duct by special add-ons, as it appears for example with sodium reabsorption in human perspiratory

glands. Unfortunately, because of their insolubility, the characterization of the containing proteins is still in process so these theories cannot be confirmed.

Although it is known that *Semibalanus balanoides* secretes adult cement some 40 days after settlement (Walker, 1973) the author also investigated adhesive forming zones of different densities and structures beneath the cuticle of the base of subadult animals. This adhesive could be the transition between the cyprids temporary adhesive and the permanent adhesive of the adult. A comparison of the extruded cement and the proteinaceous vesicle content inside the duct system was not possible because of the small content quantities. When the duct is filled with secretory material it appears homogenously electron-dense, similar to the cement. But more detailed observations are not possible.

As previously mentioned, the infoldings of the cuticle leading to the foot-like elevations are filled with adhesive material of fibrillar or homogenous electron-dense structure. The electron-dense structure is presumably the result of an early cement secretion. This assumption is supported by the fact that algae got incorporated already. The fibrillar cement is free of such inclusions and is therefore presumably younger. Likewise there is a difference in the electron density of the different slices of cuticle. The reason for this is unknown.

Conclusion

The investigation of subadult *Semibalanus balanoides* shows that in this transitional stage characteristics of both, cyprid and adults are visible. Gland cells are still arranged in clusters but start to spread out within the mantle tissue. Also the nuclei are round or oval, like in the cyprid and have only one nucleolus but there are certain exceptions with 2 or 3 nucleoli. The duct system with ICD, collecting duct and primary duct is already developed and seems to transport the proteinaceous content to the cuticle of the base where it is extruded. Vesicles are

visible in all gland cells with different diameters in relation to their developmental stage. The electron-dense lining around some of the vesicles could be the connection between vesicle and ICD at the beginning of exocytosis. Fibrillar and homogenous electron-dense adhesive is found outside the cuticle.

Acknowledgments

I thank Prof. Klepal for allowing me to write my diploma thesis at her department and for helping me understand these interesting but difficult animals. I also want to thank Daniela Gruber, Vanessa Zheden and Nikolaus Leisch for helping me out when problems appeared and for their calm nature when I was asking something again and again. Most of all I want to thank my girlfriend Lisi for supporting me all the time and helping me up when I was disheartened and of course my son Johannes and everyone in my family.

Appendix

Eisenman Fixation:

Buffer A: 0.2M cacodylate buffer, 0.1M NaCl; 0.35M sucrose; pH 7.2

Buffer B: 0.2M cacodylate buffer, 0.3M NaCl; ph 7.2

Fixative A: 4% (v/v) glutaraldehyde in 0.2M cacodylate buffer containing 0,1M NaCl; 0.35M sucrose; pH 7.2

Karnovsky Fixation:

Buffer: 0.1M cacodylate buffer; pH 7.3

Fixative: 2.5% glutaraldehyde and 4% Paraformaldehyde in 0.1M cacodylate buffer

Zusammenfassung

Der Zementapparat des balanomorphen Cirripediers *Semibalanus balanoides* (L.) wurde im Rahmen einer Diplomarbeit auf Basis von Semidünn- und Ultradünnschnitten untersucht. Das Interesse lag vor allem auf der Entwicklung von Drüsenzellen, Zementkanälen und Zementvesikeln der Subadulten, welche den Übergang zwischen letztem Larvenstadium, der Cypris und den geschlechtsreifen Adulten darstellen. Insgesamt wurden acht Tiere untersucht, welche aus Galway, Irland stammen. Einige wurden nach Karnovsky-Protokoll und andere nach Eisenman-Protokoll fixiert und in Low Viscosity Resin eingebettet. Die Untersuchung der Semidünnschnitte (1µm) erfolgte am Lichtmikroskop und die der Ultradünnschnitte (70nm) am Zeiss EM 902 Transmissionselektronenmikroskop.

Walker (1971) unterschied in den Zementdrüsen der Cyprislarven α und β-Zellen. Letztere degenerieren im Laufe der Entwicklung. In den untersuchten Tieren sind wie in den Adulten nur noch α-Zellen vorhanden, die Anordnung der Drüsen entspricht jedoch der in den Cyprislarven.

Innerhalb der Drüsenzellen von *Semibalanus balanoides* unterscheidet man zwischen Sekretpol, an dem es zur Bildung der sekretorischen Vesikel kommt und dem Speicherpol, von wo aus die Vesikel in das Drüsenkanallumen geschleust werden.

Am Speicherpol der Drüsenzellen von Subadulten sind auffallend viele Zementvesikel und dicht stehende Mikrovilli des Intrazellulärkanals zu finden. Von hier wird der proteinhaltigen

Inhalt der Vesikel an den Kollektorkanal und anschließend an den kutikulär-ausgekleideten Primärkanal weitergeleitet.

Das Sekret der Vesikel ist eine Mischung aus Phenolen, Phenoloxidasen und Proteinen und bildet beim Austritt durch die Kanalöffnungen entlang der Basis den Zement (Walker, 1971). Dieser wies in den durchgeführten Untersuchungen stellenweise eine deutliche Schichtung auf. Diese könnte durch eine Reaktion mit dem umgebenden Meerwasser, eine Härtung z.B durch Sauerstoff aus der Umgebung oder einer teilweisen Ablösung und wieder Anheftung an das Substrat zustande kommen. Ein auffälliges Kanal-Lumen wurde im Intrazellulärkanal und im Primärkanal, nicht jedoch im Kollektorkanal beobachtet. Das der ersten beiden war mit elektronendichtem, vermutlich proteinhaltigem Inhalt der Vesikel gefüllt und das des Kollektorkanals dürfte aufgrund des fehlenden Inhalts kollabiert sein.

Im Cytoplasma der Drüsenzellen sind vesikuläres, gestapelt laminares und spiralig angeordnetes Endoplasmatisches Reticulum, Golgi-Apparat, viele gebundene und freie Ribosomen und Mitochondrien. Die Zellkerne der Drüsenzellen sind oval bis rund und weisen ein relativ ausgeglichenes Verhältnis zwischen Hetero- und Euchromatin auf.

Die Drüsenzellen selbst liegen bereits lateral-ventral in der Mantelhöhle der subadulten Tiere wie dies bei geschlechtsreifen Adulten zu beobachten ist.

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