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# Changes in the midgut diverticula in the harvestmen *Amilenus aurantiacus* (Phalangiidae, Opiliones) during winter diapause



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### ABSTRACT

The harvestmen *Amilenus aurantiacus* overwinter in diapause in hypogean habitats. The midgut diverticula have been studied microscopically (light microscopy, TEM) and biochemically (energy-storing compounds: lipids and glycogen) to analyze changes during this programmed starvation period. Throughout the investigated period, the epithelium of the midgut diverticula is composed of secretory cells, digestive cells and adipocytes. Additionally, after the middle of overwintering, the excretory cells appear, and two assemblages of secretory cells are present: the SC1 secretory cells are characterized by electron-dense cytoplasm with numerous protein granules, and the SC2 cells by an electron-lucent cytoplasm with fewer protein granules. The autophagic activity is observed from the middle of overwintering, indicating its vital role in providing nutrients during this non-feeding period. Lipids and glycogen are present in the midgut diverticula cells, except in the excretory cells. Measurements of the lipid droplet diameters and the lipid quantities yielded quite comparable information on their consumption. Lipids are gradually spent in both sexes, more rapidly in females, owing to ripening of the ovaries. Glycogen rates decrease towards the middle, and increase just before the end of overwintering, indicating that individuals are preparing for the epigean active ecophase.

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#### 1. Introduction

The midgut is the largest organ in harvestmen and performs many vital functions, including digestion, nutrient absorption, nutrient and waste storage and water uptake (Becker and Peters, 1985; Shultz and Pinto-da-Rocha, 2007). It is divided into an anterior ventriculus and a posterior postventriculus by a dorsal transverse fold. Numerous prosomal and opisthosomal tubular lateral branches or diverticules arise from the lateral walls of the ventriculus, sometimes referred to collectively as the midgut gland (Shultz and Pinto-da-Rocha, 2007). The luminal side of the midgut diverticula is composed of a single-layer epithelium of columnar cells. Their apical plasma membrane is differentiated into microvilli projecting into the midgut lumen (Shultz and Pinto-da-Rocha, 2007).

*Amilenus aurantiacus* hatch in late summer (Martens, 1978). After the first autumn frost in November or December, they enter hypogean habitats and overwinter there (Martens, 1969, 1978). During overwintering in caves, they moult twice and undergo a diapause (Bourne, 1978b). In March or April, adults or subadults (own unpublished data) return to the epigean habitats. They mature in late spring and die in the late summer or early autumn (Martens, 1969, 1978; Bourne, 1978a). During winter, in some caves individuals occur in mass; they do not feed in that period of their life cycle (Jequier, 1964; Martens, 1969; Bourne, 1978a; Novak et al., 1984, 2012, 2013). In central Europe, *Amilenus aurantiacus* is the most abundant non-troglobiotic terrestrial macroinvertebrate species overwintering in caves (Novak et al., 2013).

Bourne (1978b) provided the first information on the digestion of *A. aurantiacus* during its winter diapause. The liquid part of the consumed food passes into the midgut diverticula, while solid particles with a little liquid are retained in the midgut. In the



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**Fig. 1.** Semithin transverse section of the opisthosoma in *Amilenus aurantiacus* at the beginning and at the end of overwintering, LM. a. November. The epithelium of the midgut diverticula consists of adipocytes (AC) and secretory cells (SC). b. November. The midgut diverticula epithelium is composed of adipocytes (AC) and secretory cells (SC). c. April. The epithelium is composed of adipocytes (AC), secretory cells (SC) and excretory cells (SC) and excretory cells (SC) and excretory cells (EC). FB, fat body; HS, hemolymph space; LU, lumen of the midgut diverticula; MT, Malpighian tubule; T, trachea.

midgut diverticula lumen, the food is fused with exocytotic vesicles deriving from the apical part of the abundant digestive cells. Scarce digestive cells are present in the midgut epithelium, as well. In the hindgut, the remnants of digestion are enveloped by a membrane



**Fig. 2.** Semithin section of *A. aurantiacus*. LM. a., b. January; c. April. The adipocytes (AC) are characterized by numerous lipid droplets (L) and the secretory cells (SC) by numerous protein granules (P) and individual lipid droplets (L). In b, c two types of secretory cells are seen. The SC1 secretory cell contains numerous protein granules (P) and cytoplasm of dense appearance, while the SC2 secretory cell contains fewer protein granules and a lucent cytoplasm. N, nucleus. Bar: 10 µm.

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