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Anatomy and morphology

Oral presentation

What is hiding in the *Bufo* skin? Revealing of the structures in the skin of European toads using standard histological and micro-CT techniques

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In the skin of many terrestrial anuran taxa, a specific acellular mineralized tissue layer (the Eberth-Katschenko layer) has been documented. This layer is generally positioned in between the stratum spongiosum and the stratum compactum of the dermis and has a role in reducing water loss. Here, we document and compare the amount of calcium deposition in the skin of the head and the parotoids (the external skin glands) in males and females of the common toad *Bufo bufo* and the spined toad *B. spinosus*. *Bufo bufo* and *B. spinosus* are morphologically similar and both show a conspicuous sexual dimorphism. Using standard histological techniques, we detected calcium as an amorphous ‘ground substance’ located in the stratum spongiosum, just above the Eberth-Katschenko layer. We observed large variability in the number of calcium deposits between the species and the sexes. Using micro-computed tomography (micro-CT) we were able to quantify the level of the toad skin calcification. *Bufo spinosus* females stand out compared to conspecific males and *B. bufo* on account of a strong calcification of the dorsal and ventral skin and the parotoid glands. Species and sexes significantly differed in the amount of calcium deposits (Fisher’s exact test, $p < 0.001$) and pairwise comparisons showed that groups differed from one another ($p < 0.05$), suggesting size and sexual dimorphism in these traits. We conclude that micro-CT scanning is useful for the quantification of calcified structures in the anuran skin, and keeps a promise for further studies on taxonomic and geographic variation.