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## **Nucleolar structure across evolution: the transition between bi- and tricompartimentalized nucleoli lies within the class Reptilia**

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Two types of nucleolus can be distinguished among eukaryotic cells: a tricompartimentalized nucleolus in amniotes and a bicompartimentalized nucleolus in all the others. However, though the nucleolus' ultrastructure is well characterized in mammals and birds, it has been so far much less studied in reptiles.

In this work, we examined the fine structure of the nucleolus in various tissues from different reptilian species belonging to 4 groups of living reptiles (3 turtles, 5 lizards, 2 crocodiles, and 3 snakes). For this purpose, we applied the acetylation method on different reptilian tissues in order to distinguish clearly the different nucleolar components. We also used different cytochemical (AgNOR proteins) and immunocytochemical (DNA, fibrillarin, nucleolin) markers as well as an inhibitor of rRNA synthesis (actinomycin D) to investigate the structural organization of the reptilian nucleolus. Our results clearly indicate that turtles aside, all the living reptiles have a tripartite compartmentalization of the nucleolus. Furthermore, all the tissues from a same species have the same type of nucleolus, although the importance and the repartition of those components could vary from one tissue to another. We also reveal that, contrary to the mammalian nucleolus, the reptilian fibrillar centers contain small clumps of condensed chromatin and that their surrounding dense fibrillar component is thicker. Finally, we also report that Cajal bodies are detected in reptiles.

Altogether, we believe that these results have profound evolutionarily implications since they indicate that the point of transition between bipartite and tripartite nucleoli lies at the emergence of the amniotes within the class Reptilia.