ESRF	**Experiment title: Untangling the early evolution of Lepidosauria (lizards, snakes, tuatara) using the transitional fossil *Megachirella***	**Experiment number**: ES-1391
Beamline: BM18	**Date of experiment**: from: 20.02.2024 to: 21.02.2024	**Date of report**: 31.01.25
Shifts: 3	**Local contact(s)**: **Vincent Fernandez**	*Received at ESRF:*

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Report:

This report describes the phase-contrast synchrotron X-ray micro-computed tomography experiment carried out between 20th and 21st of February 2024 at the Beamline BM18 of the ESRF. There was a single object that was analysed. It was a block of limestone measuring 160 mm x 120 mm x 30 mm that preserves a fossil reptile skeleton. The animal was named *Megachirella watchleri* and held in the Museo di Scienze Naturali dell'Alto Adige in Bolzano, Italy under a number PZO 628. It is dated Middle Triassic (Anisian), so it is more than 240 million years old. The skeleton is composed of partial skull, neck, shoulders, forelimbs, and part of the trunk. The anatomy of this animal is a key to understand the early evolution of lizards, snakes, and tuatara because it could be the oldest known lizard in the world. Therefore, *Megachirella* offers a unique snapshot from time when lizards and their kin were originating (diverging from each other). Previous studies on its anatomy and relationships faced significant limitations because the skeleton is partially entombed in the rock matrix and previously applied lab-based X-ray micro-CT provided very noisy pictures of the bones and rock interior with serious beam-hardening artifacts. That resulted in uncertainty and incompleteness of interpretations.

The experiment at ESRF was successful. We obtained 3.2 Tb of data in total in the form of tiff files. The resolution of the images is 6 μm. The contrast is good, permitting precise separation of each bone from the surrounding matrix and other elements (Fig. 1). The contrast and resolution not only allows identification of particular bones but also we can observe very fine anatomical details like vascular canals and palatal dentition.

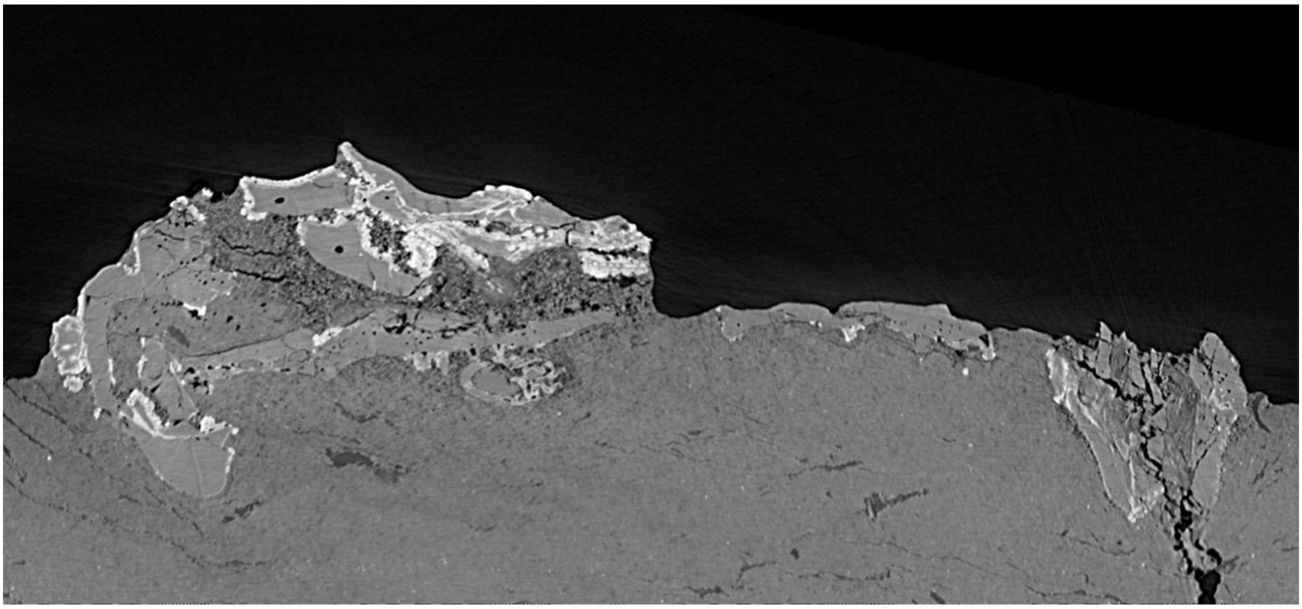

Fig. 1. Image (cropped) of the transverse cross-section of the mid-portion of the skull of *Megachirella*. Some bones are fractured, especially on the right but can be easily distinguished from the surrounding rock matrix. White surroundings of many bones probably represent a pyritic crust. Without scale.

Currently we are in the process of segmentation of obtained data. Each bone on each slice is marked using a specialized software. Analyzed slices are combined to form a 3D renderings of each bone. We have processed almost all elements of the skull so far but the postcranial skeleton awaits further work. However, we can already identify many new informations about this animal that were unavailable when analyzed with other methods. We can now revize the previous interpretations and, for example, clearly demonstrate that the palatine bone bears teeth, and the postfrontal and postorbital are unfused. Moreover, the squamosal is reduced to a rod-like element which is a feature unique for lizards while in previous studies it seems to be more complex and thus primitive. These new data offer an opportunity to revize the phylogenetic position of *Megachirella* and its significance to the early evolution of lizards and its relatives.

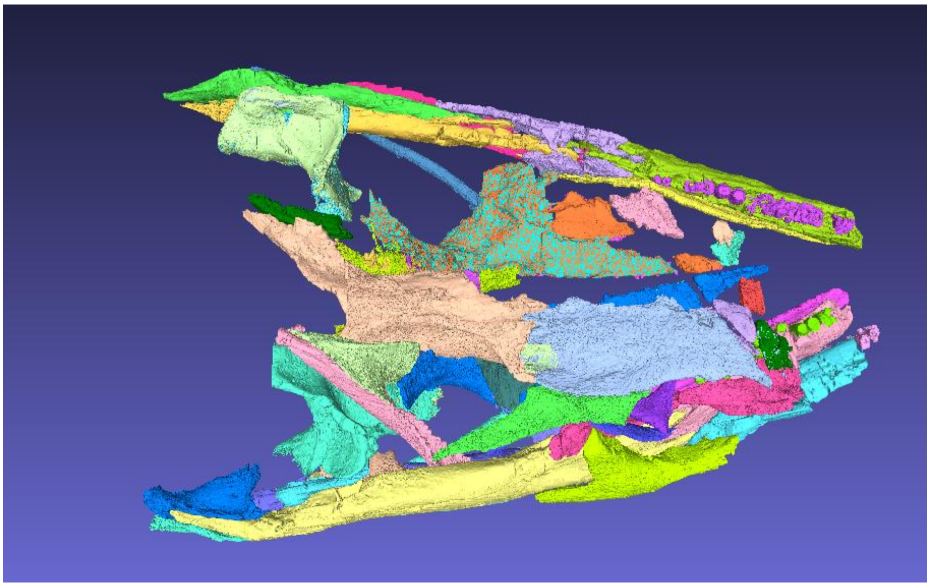

Fig. 2. 3D renderings of the skull elements of *Megachirella* in dorsal view. Without scale.