

Scientists overcome inaccessibility of caves through molecular genetic approach

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An international group of scientists has used a novel highly sensitive method for detection of environmental DNA in groundwater to extend the poorly known range of the rare subterranean amphibian from the Dinaric Karst. With this highly sensitive non-invasive method they discovered 12 new localities of the olm (*Proteus anguinus*). Their findings were published on 27th March 2017 in the journal *Scientific Reports*.

Puzzling the minds of many famous naturalists, from Linnaeus, Cuvier and Humboldt, to Lamarck and Darwin, *Proteus* is one of the world's prime symbols of natural heritage and study. This blind salamander is by far the largest cave animal in the world. In over 250 years of research, it has only been sighted at 300 subterranean sites along the Dinaric Karst. *Proteus* is a globally threatened species, and is vulnerable due to groundwater pollution.

So far, the hurdle to studying and protecting *Proteus* has been its subterranean habitat inaccessibility. The authors, Špela Gorički, David Stankovi?, and others developed an indirect method to search for *Proteus*, detecting its DNA released in water (environmental DNA or eDNA). Traces of *Proteus* eDNA were searched for in water samples collected from karst springs, wells or caves by a quantitative real time polymerase chain reaction-based approach.

The authors conducted the most extensive survey of *Proteus* distribution in Slovenia, Bosnia & Herzegovina and Montenegro, established its



likely presence at seven new sites along the southern limit of its known range, and newly documented its presence in Montenegro.

The study also developed a more specific method to detect the eDNA of the black *Proteus* morph, a rarely spotted animal that is confined to an area of 30 km2 in Southeastern Slovenia. This sampling doubled the known black *Proteus* sites - a new basis for an efficient conservation management - and documented that both black and white *Proteus* populations live side by side.

"Our results show that the eDNA approach is suitable not only in biogeography and conservation of rare and endangered species, but it is efficient also in addressing questions in evolution and taxonomy of the cryptic subterranean fauna." said Dr. Špela Gorički, the first author on the study. "All countries, both within the previously known and the newly established range of *Proteus*, should confront the challenge to preserve it. Not only would we lose such an extraordinary animal, but the people would lose their only source of drinking water", said Gregor Aljančič of the Tular Cave Laboratory, the senior author.

More information: Špela Gorički et al. Environmental DNA in subterranean biology: range extension and taxonomic implications for Proteus, *Scientific Reports* (2017). DOI: 10.1038/srep45054

Provided by Tular Cave Laboratory

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