

GIS and eDNA analysis system successfully used to discover new habitats of rare salamander

September 6 2019



A Yamato salamander (*Hynobius vandenburghi*) and its egg sac (lower right). Credit: Koki Tsunekawa (Nature and Science Club Bioscience team leader, Gifu Senior High School 3rd grade)



A research team has successfully identified an unknown population of the endangered Yamato salamander (*Hynobius vandenburghi*) in Gifu Prefecture using a methodology combining GIS and eDNA analysis. This method could be applied to other critically endangered species, and could also be used to locate small organisms that are difficult to find using conventional methods.

The study was conducted by students from the Bioscience team in Gifu Senior High School's Nature and Science Club (which has been conducting research into the species for 13 years). They were supervised by teachers and aided by university researchers, including Professor Toshifumi Minamoto from Kobe University's Graduate School of Human Development and Environment.

It has been reported that there are approximately 50 *Hynobius* species of salamander worldwide, around 30 of which are endemic to Japan. *Hynobius vandenburghi* (until recently known by its previous classification of *H. nebulosus*), is only found in central and western Japan, with Gifu Prefecture marking the northeast limit of the species' distribution (Figure 2). However, like approximately 60 percent of amphibian species in Japan, it falls under the ranking of critically endangered and vulnerable species, mainly due to habitat decline. Only three sites providing habitats for Yamato salamanders had been discovered in Gifu Prefecture up until recently.





A distribution map of Hynobius vandenburghi. Credit: Kobe University

The research team used a combined methodology of GIS and eDNA analysis with the aim of discovering more Yamato salamander habitats. GIS (Geographic Information System) is a spatial analysis tool that allows data and geographic information to be collected, displayed and analyzed. *Environmental DNA* analysis involves locating DNA of the species in the environment (in this case, in water samples) to understand what kind of organisms live in that habitat.

First of all, <u>environmental factors</u> (such as vegetation, elevation, and gradient inclination and direction) that prevail near the known habitats in Gifu Prefecture were identified, and this information was entered into the GIS to locate new potential habitats. This resulted in a total of five new potential sites being discovered—three in Gifu city, and one site each in Kaizu and Seki cities.

Next, each site was visited and water samples were taken. Yamato salamander often lay their egg sacs in shallow water near rice paddies and wooded areas, so the <u>water samples</u> were taken from these environments. The samples were then analyzed for Yamato salamander



eDNA; eDNA was discovered in the water from the Kaizu City site, the Seki City site and one of the Gifu City sites.



Conducting field surveys at the Kaizu city site (left) and the pair of egg sacs found at the site. Credit: Kobe University

Field surveys were also conducted to find eggs or adult <u>salamanders</u> at each of the sites where eDNA was discovered. A single pair of egg sacs were found at the Kaizu city <u>site</u> (Figure 3). This lends support to the idea that the combined methodology of GIS and eDNA analysis can be successfully used to find new habitats of rare and elusive <u>species</u> like the Yamato salamander.

As this research was carried out by supervised <u>high school students</u>, it is anticipated that this combined methodology can be used not only by experts but also as a useful tool for citizen-led conservation efforts. Another advantage of the GIS and eDNA analysis method is that it requires less time, energy and funds compared to conventional field capture (locating animal specimens). This could prove invaluable for identifying and protecting the habitats of <u>endangered species</u> in the face



of rapidly declining biodiversity worldwide.

More information: Yusuke Sakai et al, Discovery of an unrecorded population of Yamato salamander (*Hynobius vandenburghi*) by GIS and eDNA analysis, *Environmental DNA* (2019). DOI: 10.1002/edn3.31

Japanese Ministry of the Environment (2019). Red List 2019. Electronic version available at: <u>www.env.go.jp/press/files/jp/111318.pdf</u> (accessed August 28, 2019) (in Japanese)

Provided by Kobe University

Citation: GIS and eDNA analysis system successfully used to discover new habitats of rare salamander (2019, September 6) retrieved 27 April 2024 from <u>https://phys.org/news/2019-09-gis-edna-analysis-successfully-habitats.html</u>

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