Construction of cost-effective workflows enabling development of simplified identification tools and designation of endangered species of amphibians and reptiles

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[Abstract]

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Accurate species recognition and identification are among the most important issues in biodiversity conservation and related activities. In this research project we aimed to develop tools and systems to facilitate easy and accurate identification of Japanese amphibians and reptiles. We also conducted taxonomic revisions and evaluation of vulnerability for several potentially endangered species and for species groups that contain such species. In our taxonomic revisions we examined 18 species or species groups, and found at least eight potentially undescribed species, of which two have since been described. For evaluation of the vulnerability of species or populations, we carried out genome-wide SNP analyses for seven groups and clarified evolutionarily significant units for conservation. We estimated their heterozygosity from the SNP data as a simple measure of population vulnerability, and based on those scores, we suggested relative priorities among populations/species for the application of conservation measures. We also conducted whole-genome shotgun analysis for 48 species, to establish references for future genomic analyses of Japanese amphibians and reptiles. In developing species identification tools, we first determined nearly complete mitochondrial DNA sequences of over 350 individuals of 199 species of the Japanese ectothermal tetrapods (>90% of total) and complied the sequences as reference data for future DNA barcoding analyses. Based on the data, we designed new primers for urodelan (salamander) DNA barcoding that can identify all the species known from Japan. Second, we developed a method to identify a particular protected salamander, which is sometimes sold in the pet market illegally, by use of the loop-mediated isothermal amplification (LAMP) technique. This method can identify the species within one hour without any special equipment, and thus it is useful as a control measure to monitor the illegal trade. Third, we developed a species identification tool based on photographic images, using artificial intelligence (AI) learning, to discriminate several protected species from highly similar species. As a result, we successfully developed an AI learning model that can discriminate the Japanese giant salamander from introduced Chinese congeners using simple photographic images. These species are hybridizing in the wild, which poses a serious problem for conservation of the Japanese giant salamander. The new tool developed here will help prevent further introduction of the Chinese species to other river systems.

[References]

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