Development of a New Ecosystem Research Method Using DNA Analysis

Background

The Environmental Impact Assessment Law requires prediction and assessment of the impact of business activities on an ecosystem. In ecosystem research and impact prediction, elucidating predator-prey relationship and herbivore-plant relationship, among creatures that compose an ecosystem, is important, but a method to easily and highly accurately research this relation has not been established.

Objectives

To develop a highly efficient and accurate new research method using DNA analysis, to estimate populations of wild animals and to learn about their feeding habits, which are important in elucidating predator-prey relationship and herbivore-plant relationship.

Principal Results

The hare is a target species that typically represents the characteristics of a regional ecosystem, as a representative herbivore, and it is also regarded as an important food source for rare birds of prey. In this study, we attempted development of the above-mentioned method, using hares.

1. Estimation of populations using individual identification through DNA analysis

Hares frequently leave feces, so they can easily be found in their habitat. If the individual animal that excreted a given specimen of feces can be identified from it, it would be possible to estimate the population by researching many specimens of feces. Through individual identification by using DNA of intestinal cells attached to feces, we could identify individual animals, from feces found on snow on the day after a snowfall, because such feces is well preserved. As such, we conducted research seeking to estimate the population in an 18.5-ha area of Japanese cedar forest, at the base of Mt. Komagatake, in Akita prefecture. As a result, we could identify an individual animal from every feces specimen collected, and confirmed that 15 individual animals were in this research area on the night before the research. This method is much more reliable compared with the conventional fecal pellet count, etc., which estimates habitat density based on feces specimen numbers, and it can be applied to a wide area, so this is an effective method to estimate a population of hares with high accuracy.

2. Identification of food source plants through DNA analysis of feces content

As wild animal feeding habit survey, analysis by observation of feces and stomach contents is common, but it is often difficult to determine food species when its form has changed due to crushing / digestion. As such, we studied a food habit survey using DNA analysis, to enable identification of food plant species even from residue of feces that has changed in form. To use DNA sequences of chloroplast genes, which are specific to plant species, for identification of food plants, we determined DNA sequences of 700 plants species, collected mainly at the base of Mt. Akita-Komagatake, which is the research area, and built a DNA database. As a result of extracting and analyzing DNA from undigested residue of hare feces specimens collected from a cut-over area and a natural beech forest, DNA sequences of nine types, and seven types, respectively, of plants were detected, and the plants used as food sources could be identified on a species level, through comparison with the DNA database. In addition, we conducted similar analysis on feces specimens from Japanese serows and copper pheasants, and confirmed that we can identify their food plants. In this way, this method can be expected to be widely usable for food habit survey of herbivore.

The research method combining feces specimens and DNA is effective, without giving unnecessary stress to the target animals, even when catching them or visual confirmation is difficult. This method, which also has possibility of use for behavioral research and population genetics, and other, in addition to population estimation and food habit survey, is considered to be very effective for study of wild animals.

Future Developments

To highly accurately and efficiently implement research and evaluation of wildlife and the ecosystem in Ecosystem assessment, we will study applicability of the research method using DNA analysis.

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Reference

Matsuki R. et. al., 2003, “Study on the ecosystem sustaining a pair of Golden Eagles -Identification of food plants by DNA analysis from animal feces-”, CRIEPI Rep. U03008, (in Japanese)
Matsuki R. et. al., 2004, “Study on the ecosystem sustaining a pair of Golden Eagles -Population size estimation of Japanese hare by fecal DNA typing-”, CRIEPI Rep. U03066, (in Japanese)

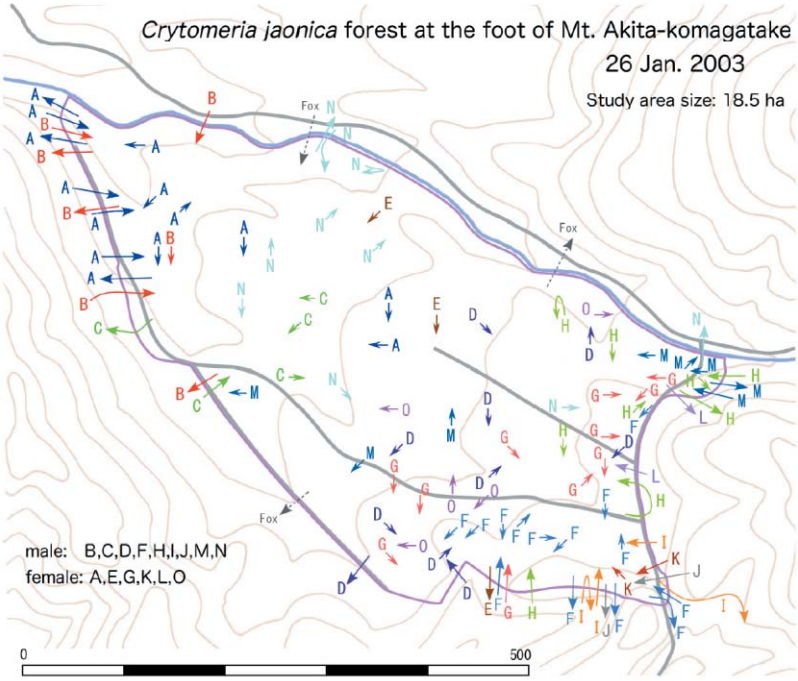


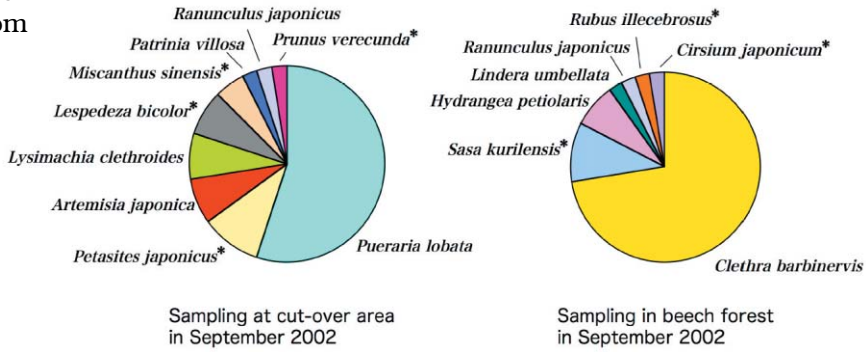
Fig.1 Individual Identification and Estimation of the Population of Hares

We recorded entry and exit of every track of footprints that crossed the research route, which was the circumference of the research area, and collected applicable feces. Subsequently, we collected feces randomly from inside the research area, and identified the individual animals that excreted the specimens, from DNA of each feces. We collected a total of 106 pellets, and confirmed the existence of 15 hares, consisting of nine males and six females. In this research, we could also grasp entry into and exit from the research area, of each confirmed animal.

Fig.2 Frequency of Appearance of Food Plants Detected from Hare Feces

We analyzed DNA contained in each hare pellet. We determined plant species on 40 detected DNA sequences, and identified food plants.

* Representative species most frequently seen in the research area, having the same DNA sequence as multiple related species, were identified.



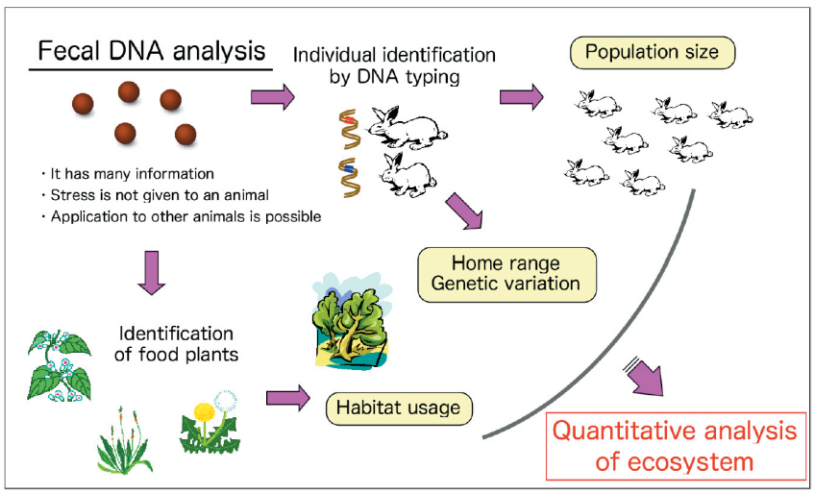


Fig.3 New Ecosystem Research Method Using DNA Analysis

We can grasp habitat home range, population fluctuation, etc., by fecal DNA analysis. Further, we can also grasp habitat usage by identifying food plants. Such an ecological research method on wild animals using DNA analysis is effective for quantitative analysis and assessment of an ecosystem.