D16 Detection and Identification of Rhinoceros Species by Specific Primers

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After attending this presentation, attendees will understand the use of the primers quote from the paper to identify the parts and products of Rhino.

The community may be encouraged to design species specific primers to detect parts and products of wildlife.

Rhinoceros are the first animals listed in the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES), which prohibits the hunting, trading and use of endangered species. In Chinese traditional medicine, powder of Rhinoceros has long been acclaimed by herbalists as a cure for fever and used as an ingredient in a range of products, most often in the form of tablets and herbal teas, although blocks or raw horn have also been found. Owing to the over exaggerating curing effects and huge profit, it was smuggled continuously, although there is also a strict wildlife conservation law here in Taiwan to prohibit the trading of the parts or products of Rhinoceros.

There are currently a series of tests that can indicate the possible presence of Rhinoceros horn powder, including immunology test of keratin, electrophoresis of horn protein extraction and microscopic observation of horn surface structures. More definitive tests include FINS system with cytochrome-b sequencing and the comparison to the national database.

But these kinds of identification methods were not going to work if the specimens were a mixture of more than two different species or there were more than two species of Rhinoceros horn powder put together, the peaks of ATGC spelling would be mixed up, resulting in much more difficulty for sequencing the species differentiating segments of cytochrom-b DNA. A more efficient and confirmative method should be developed to resolve the problem and enhance the law enforcement.

There are five endangered species of Rhinoceros, Ceratotherium simum, Diceros bicornis, Rhinoceros unicornis, Rhinoceros sondaicus and Rhinoceros sumatrenis. A pair of primers for amplifying consensus DNA sequence of 12S rRNA of mtDNA was designed to confirm the existence of Rhinoceros DNA. The primers have been tested on a range of different animal parts and products. DNA extracts only give a PCR product in the presence of Rhinoceros horn powder.

For the purposes of determining how many species are in a powder, Rhinoceros species specific primers were also selected and then compare the sequence to the DNA database using the FastA method. After electrophoresis of the multiplex PCR products by using ABI 310, peaks of different dyes for each specific species of Rhinoceros could be detected without ambiguity, for example for Ceratotherium simum blue dye peak 351bp long would be found, for Diceros bicornis yellow dye peak 351bp long would be found, for Rhinoceros unicornis green dye peak 376 bp long would be found, etc.

The Rhino-specific and Rhinoceros species specific primers could be an efficient method for detecting parts and products of Rhinoceros.

Rhinoceros, Products of Wildlife, DNA Detection

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