A MULTIPLEX PCR SYSTEM OF INSERTION-DELETION POLYMORPHISM WHICH APPLY TO PERSONAL IDENTIFICATION OF CHINESE POPULATION

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InDel indicates insertions or deletions (insertion-deletion) of nucleotide fragments of different sizes at the same site in the genome sequence between the same or closely related species and is a gap in sequence derived from alignment of the homologous sequence. InDel is widely distributed across the genome and occurs in a high density and large numbers in a genome. The InDel polymorphic molecular marker is a PCR-amplified marker that is based on specific primers designed from both sides of the site of sequence of insertion / deletion. It is essentially a length polymorphic marker still, and one can use the convenient electrophoresis platform for genotyping. InDel molecular markers have the advantage of high accuracy and good stability, which help to avoid confusion in subsequent analysis due to marker specificity and complexity, as is often seen in other length polymorphic markers. Furthermore, mixed or highly degraded DNA samples can be successfully amplified with InDel markers, and effectively typed. Because of its abundance, convenient typing platform and other advantages, InDel molecular markers have been applied to genetic analyses of animal and plant populations, molecular assisted crops and farmed animal breeding, human forensic genetics, medical diagnostics and other research areas. The development of the InDel molecular marker located on functional genes, combined with chromosome walking and fine gene mapping, has enabled the application of these molecular markers in the screening of genes related to important economic traits, which is conducive to the further development and utilization of these valuable genes. In this review, on the basis of an overview of the InDel marker development and applications, we discuss some of the technical limitations of the development and limited efficiency of genetic analysis, as well as potential future applications in the fine mapping and genetic structure of large numbers of individuals.

Using NCBI, combined with the existing literature reports, 83 loci selected in accordance with the standards to meet the requirements, choose 600 samples of different ethnic groups in China, the method of using Sequenom to test the 83 loci, the classification Information, and calculate the allele frequency and expected heterozygosity, Hardy Weinberg genetic Information chain, balance, etc, to evaluate 83 loci and selection, finally confirmed 30 InDel site for composite amplification. The 30 sites is used only for identification of forensic science personal recognition, did not see that is associated with health sensitive Information of literature reports. 30 highly polymorphic of InDel makers were selected by using the dbSNP database for population of China. The multiplex PCR system was developed by

using a five fluorescence dye labeling system. To investigate the polymorphism of Han, Kazak, Dai, Miao and Yao, and find the application and characters of InDel markers.establish a multiplex PCR system, using InDel genetic markers which apply to forensic DNA identification of Chinese population.

This study successfully established a set of 31 loci multiplex PCR system,that contains 30 highly polymorphic InDel markers and an Amelogenin gender marker. The cumulative discrimination power（CDP）of the 30 InDel markers was 0.999999999957,0.99999999999,0.999999999974,0.999999999875 and 0.999999999966 for the Han, Kazak, Dai, Miao and Yao, the pairwise population F_{ST} estimates are less than 0.0448。Genetic Polymorphisms survey showed that the 30 InDel markers with highly polymorphic and small differences between groups.

In this study, through the screening of no chain 30 InDel site and a gender identification of Amelogenin gene, constitute 31 locus of complex multiple PCR amplification system, and through the capillary electrophoresis technology, established a set can be used for forensic DNA laboratory of the supplementary tool for the identification of DNA. The han people of CDP, the comparison of CPE and RMP, including the research and the absence of Qiagen company ® DIPplex kit system efficiency, the finished product basic equivalent to 9 STR system efficiency, and 42 IISNPs loci and 18 STR loci system efficiency difference is large, the scholar thinks, like InDel second-class gene genetic marker, if you want to achieve with the STR kit as the commercialization of the commonly used in forensic personal identification ability need 60 such sites, consistent with the investigation of this system. In this amplification system, 31 site amplification of size is less than 220 bp, while limiting the increase the possibility of a site, but increase the success rate of degradation of DNA for examination, inspection, can be used as a powerful supplement of STR testing system.This multiplex PCR system is a forensic DNA identification tool that applied for Chinese populations.

Fig1 DNA Type of 9947A

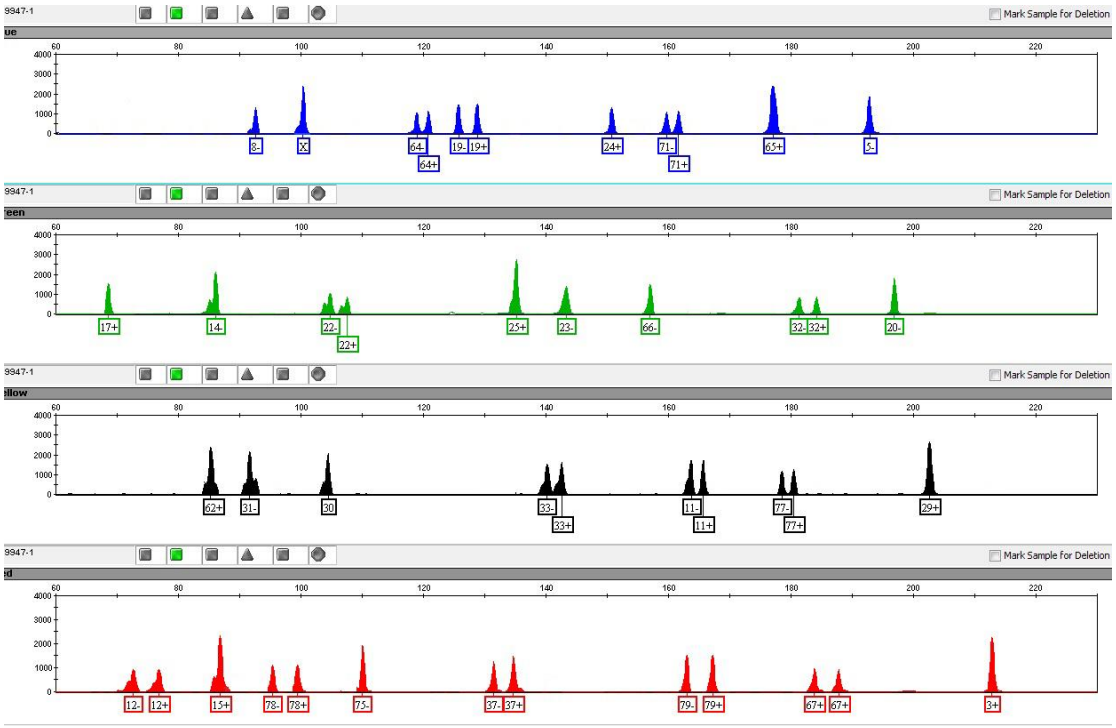

Table1 The comparison of CDP CPE and RMP in different systems

	CDP	CPE	RMP
This system	0.999 999 999 957	0.985 806 21	4.29E-11
DIPplex	0.999 999 999 985	0.987 710 49	1.42E-11
9 STR Loci	>0.999 999 99	0.999 9	—
42 IISNP	—	0.999 82	9.5E－18
18 STR Loci	0.999 999 999 999 999 999 999 973 339	0.999 999 973 339	5.74E-22