

Application of Molecular Biology Techniques in Forensic Medicine

Yue Xu

Nanjing Medical University, Nanjing, Jiangsu, 210029, China

ABSTRACT

With the development of the times, the research content of molecular biology is gradually deepened, and more theories and techniques are gradually known or mastered by people, as a traditional discipline, forensic medicine should be able to take this opportunity to make greater progress and breakthroughs. The main content of this paper focuses on the application of some commonly used molecular biology techniques in the field of forensic science, and shows the practical results of the combination of molecular biology and forensic science, it provides a new perspective or idea for the combination of disciplines.

KEYWORDS

Forensic molecular identification; MicroRNA detection; Next-generation sequencing(NGS); Forensic genetics; Forensic entomological DNA barcoding

1 Introduction

Molecular biology itself is a discipline that studies the structure, function and related role of biological macromolecules at the molecular level. Forensic Medicine is the theory and technology of applied medicine, biology and other natural sciences, to study and solve medical problems in legal practice.

Accordingly, molecular biology has a strong application value in the field of forensic medicine. It can not only solve the problems of identity identification and parentage judgment in criminal investigation, but also play an important role in many sub-disciplines such as forensic pathology and forensic toxicology, and promote the development of forensic science in a more accurate and efficient direction.

At present, the application of molecular biology in the field of forensic science mainly includes PCR technology, the application of micrnas, DNA barcoding technology, HRM technology and DNA sequencing technology. This article will quote specific cases, the application of the above techniques in the field of forensic medicine is described to provide a train of thought or perspective for the effective combination of the two disciplines.

2 The Application of PCR Technology in Forensic Science

PCR technology, namely polymerase chain reaction, is a molecular biology technology that simulates the natural process of DNA replication in vivo and amplifies DNA molecules in vitro according to the principle of DNA semi-retention replication.

In the study of Yang Rui et al. , it was pointed out that many existing flora analysis techniques are difficult to meet the needs of forensic science, and DPCR technology itself has a wide range of applications in the field of forensic science. The combination of dPCR and HRM can combine traditional PCR techniques and reduce the number of samples requiring STR typing or next-generation sequencing. This is undoubtedly effective in reducing the cost of the test and has a higher test efficiency. In addition, it is also pointed out that the DPCR-HRM technique does have good discrimination ability and stability.

In addition, we mention an additional technique developed on the basis of PCR, namely the ARMS (amplification of tissue mutation system) technique. This technique uses specific primers to amplify mutant target sequences with high precision and probes to detect the amplified products. Tian Huan and Dong Yingchun pointed out in their paper "ARMS-PCR technique for detection of DNA methylation in forensic body fluid samples" that ARMS-PCR technology can detect the level of DNA methylation with only one amplification, it has the potential to identify forensic body fluid samples.

3 Application of Micrnas in Forensic Science

Micrnas are a class of non-coding single-stranded RNA molecules, about 20 to 24 nucleotides in length, encoded by endogenous genes. Because of its tissue-specific and non-degradable characteristics, it has been widely concerned in the field of forensic medicine. Li Ranran pointed out that the detection system of microRNA has good stability and sensitivity in the detection and discrimination of peripheral blood, semen, saliva and menstrual blood samples.

Micrnas are small non-coding RNA molecules (ncrnas) that are involved in the regulation of many life processes. They

are more stable than mrnas, this allows forensic scientists to extract micrnas from paraffin-fixed tissue sections

4 Molecular Identification of Necrophagous Insects (DNA Barcoding)

In the field of forensic science, necrophagous insects are of great significance for scene investigation, especially for the estimation of time of Death (PMI) .

There are many techniques in molecular biology to identify necrophagous insects. For example, DNA barcoding is based on the genetic material, or DNA sequence, that is unique to each species. After extracting biological DNA, a specific DNA fragment with sufficient variability is amplified and then compared with other fragments in international databases to complete rapid identification of the species, and that particular piece of DNA with enough variability is the DNA barcode.

In the study of Xu et al. , DNA barcoding technique was proved to be effective in identifying common necrophagous flies in the Yangtze River Delta Economic Zone.

5 HRM Technology and Forensic Medicine

HRM (high-resolution melting curve) was also mentioned in addition to PCR. This is a technique of gene analysis in which different melting curves are formed due to the difference of melting temperature of single nucleotide. Because of its high sensitivity, HRM can detect differences in individual bases.

In forensic science, HRM also has a wide range of applications. Because of its ability to detect small differences in DNA sequences, it can have a good application in forensic identification. In a forensic study on the identification of hallucinogenic poisons by HRM technique, Zhang Xiaochun et al. , on this basis, we screened the nuclear gene fragments of the common hallucinogenic mushrooms such as Cuba's pleurocarpus capillaris, and established a rapid and effective method for the identification of hallucinogenic poisons. In addition, HRM technology can also be applied to genotyping. This makes it useful in paternity testing and family relationship analysis.

Al ípio Dos Santos Rocha and Isis Salviano Soares de Amorim et Al. 7 extracted 8 different mtDNA from mucosal swabs using DNAzol reagent and then analyzed by HRM after PCR amplification, after which they concluded that, that is, DNA from different samples can be distinguished by different HRM curves, especially with accurate results for the screening of small differences in the hypervariable (HV) regions I and II, which clearly shows that the HRM curves are useful for the identification of DNA from different samples, hRM technology can improve the efficiency of research in forensic genetics laboratories.

On a larger scale, HRM can even be used to identify specific species. In 2021, Wannapimol Kriangwanich with Korakot Nganvongpanit et al. 8 noted that a method combining simple inter-sequence repeat markers (issrs) with HRM techniques has been used for species identification of 38 mammalian species.

6 DNA Sequencing Technology and Forensic Medicine

DNA sequencing technology, that is, the technology of determining DNA sequence, is to determine the direction and structure of recombinant DNA by analyzing the base sequence of DNA fragments, and to locate, identify and compare mutations. The common DNA sequencing technologies include Sanger sequencing, next-generation sequencing (NGS) , third-generation sequencing and so on.

Gao Nana et al amplified the V1-V3 region of the 16S rna gene of bacteria and ITS1 and ITS2 regions of the ITS gene of fungi. The amplified product was tagged and sequenced with ISEQ100, and identified with a matching microbial identification system. The identification results showed that the identity rate with the control bacteria at the genus level was as high as 100% , even if it was accurate to the species, the results also had 81.1% inhibition rate with the control bacteria. Such accurate results also prove that the next-generation gene sequencing technology in microbial identification has a good application effect.

In addition, the identification of blood type is particularly important in the daily work of forensic medicine. In clinical work, we sometimes encounter difficult specimens that can not be determined, in the Chinese Journal of blood transfusion in 2020, it was recorded that Zhao Yuan, Bo Yan, Feng Qiang et al. , the end result, while certainly limited, is clearly usable on the whole.

Zhao congying, Xu Hui et al. screened 111 highly polymorphic SNP genetic markers from the 1000genomes Project Phase III database and sequenced them using the MGISEQ-2000 sequencing platform, in other words, gene sequencing technology has high sequencing performance in ethnic identification, especially in the identification of East Asian ethnic groups, which can meet the needs of identification and has good development prospects and space.

In contrast to conventional DNA sequencing, or Sanger sequencing, NGS refers to high-throughput DNA sequencing technologies that are not benchmarked against it, it can greatly improve throughput and greatly reduce the need for fragment cloning methods commonly used in Sanger sequencing. Furthermore, Yaran Yang et al. point out that short tandem repeat (STR-RRB- analysis will remain the most commonly used genetic technique in forensic science for a long time to come.

7 Conclusion

Based on the foregoing, it is not difficult to find that molecular biology technologies, such as PCR, HRM, microRNA, DNA barcoding and sequencing, are gradually deeply intertwined with applied sciences such as forensic science, the techniques of molecular biology will change, and even reshape, the way forensic scientists work, and will provide more robust and objective evidence for the administration of Justice, forensic science can provide better insights into molecular biology, or provide new research directions and topics based on practical needs.

Due to the limitation of space and the author's limited ability, we will not introduce the application of molecular biology techniques in forensic science. However, we know that both disciplines will have a longer-term development in the future. Although molecular biology started much later than traditional disciplines such as forensic science, it is the "Leader" in modern science, its rapid development and innovation will inevitably bring more and more effective, fast and precise technologies, which will also promote the development and application of traditional disciplines, including forensic science.

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