

Application of FTA-PCR technology combined with LIMS system in food safety inspection

1st Jianxin Li ^{*1,a}, 1st Jiaxing Li^{*2,b}, Anlang Ou^{3,c}, Jianlin Li⁴, Zhuosi Rong⁵, Yuhao Ma⁶, Jianhui Yang⁷

^{1,3}Jiangmen Vocational and Technical College, China

²Guangxi University of Traditional Chinese Medicine, China

⁴Guangzhou Institute of Technology, China

⁵Qingyuan Vocational and Technical College, China

⁶Guangdong Food and Drug Vocational College, China

⁷Dongsheng School, Longgang District, Shenzhen

Abstract: With the increasing awareness of public health in the world, traditional food safety testing technology can no longer meet the needs of increasingly complex food types and product structures. The combination of traditional testing and biotechnology is a new force in strengthening food protection testing technology. This article analyzes the FTA-PCR technology combined with the LIMS system, based on the traditional chemical food safety testing, combined with the effective application of biotechnology in food safety testing, and explained the FTA-PCR technology combined with the LIMS system for food safety testing. In order to provide some help for the better development of my country's new food safety testing technology.

1 Foreword

The macromolecular DNA coil is tightly combined with the FTA matrix, without DNA breaks, and the DNA binding rate is greater than 90%. DNA is adsorbed on the FTA card and can be stored for a long time after drying. Use FTA purification reagents to clean the contaminants that affect DNA analysis, such as heme and PCR inhibitors, to purify the bound DNA. The DNA remains on the filter paper during the purification process, and the purified DNA can be used for PCR detection. Gentle elution with FTA purification reagents can reduce the mechanical damage caused by conventional DNA extraction methods and maintain the form of DNA macromolecules. It can be used for molecular biology research and analysis, as well as nucleic acid purification, PCR, SNP, RT-PCR, RFLP analysis, gene cloning, nucleotide sequencing, etc.

2 Principle and practical application of FTA card

2.1 Principle

FTA (Flinders Technology Associates) (FTA Filter membrane, FTA Card). It's a special filter paper. It is soaked in strong denaturant and chelating agent with patent formula. The fiber matrix contains special chemical

substances. When cells are captured, it will be automatically cracked and combined with nucleic acid to maintain the integrity of DNA in the sample, protect nucleic acid from degradation, nuclease, oxidant and UV damage, and prevent the growth of bacteria and other microorganisms.

FTA card, as a medium for storing nucleic acids, can be used for direct PCR amplification or nucleic acid extraction for subsequent analysis.

2.2 FTA Technical advantages

1、 It only needs normal temperature environment, easy operation, fixed storage of nucleic acid without low temperature, just need to add blood or saliva to the card

2、 After removing the wafer with short punch or cleaning with pure water, FTA reagent or the buffer (< 30min), the downstream experiment can be carried out.

3、 The FTA card is safe and reliable without any other treatment. It has anti ultraviolet damage and antibacterial properties, and can inactivate harmful microorganisms / viruses

4、 Blood samples were stored at room temperature for more than 22 years, and oral cell samples were stored at room temperature for more than 12 years. The data is constantly updated ~

* Corresponding author: Jianxin Li
^{a,b,c}E-mail: kuangsf@gdyzy.edu.cn

5. Compatible with most molecular detection methods, such as PCR, RT-PCR, STR, RFLP, SNP, WGA, invader assay, HPLC-MS, etc

6. With large storage volume, the sample volume of circle sampling / storage can reach 125 μ 50. FTA gene card can reach 75 μ L

FTA card is impregnated with patented chemical formula, which can dissolve cell membrane and change protein properties. Nucleic acid is fixed and protected from UV light damage, microbial and fungal erosion. The infectious pathogens in the samples also lose their activity on the FTA card. The samples collected on the FTA card are very safe, and can be sent through the post office without the need to post danger labels. To recover the captured nucleic acid, a small wafer should be removed from the FTA card for elution, so that the clean nucleic acid can be left on the wafer, and then the wafer can be directly used for downstream applications, such as PCR (even after PCR reaction, the template DNA is still adsorbed on the FTA wafer, so PCR and other analysis can be repeated). It is recommended to use indicator FTA card for clear samples. The indicator FTA card will change color (pink turns white) after the sample is added, so as to identify the location of the sample.

The operation methods of FTA: sampling, cleaning, PCR amplification.

2.3 General use of FTA card

FTA membrane can collect, transport and archive all kinds of biological samples at room temperature. It was first used to store blood samples. In foreign countries, FTA card has been widely used in hospitals and the military. FTA cards are used in hospitals to keep blood samples of newborns for identification in case of missing children or other accidents. The project is called chip, or children identification program.

2.4 FTA Card Laboratory operation process

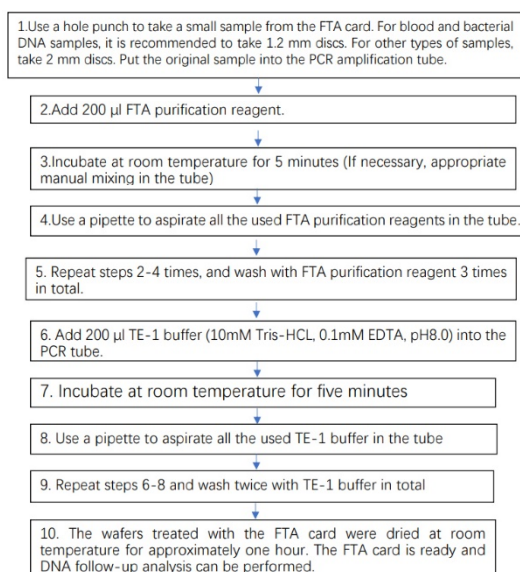


Figure 1. Schematic diagram of LIMS laboratory system flow

3 Polymerase Chain Reaction

PCR is a kind of molecular biology technology used to amplify specific DNA fragments, which can be regarded as a special DNA replication in vitro. The biggest feature of PCR is that it can greatly increase the trace DNA, increase the persuasiveness of the control group in food detection samples, and increase the fault tolerance of samples.

3.1 The key points to prevent contamination during PCR detection are as follows:

1、Positive control: In the establishment of PCR reaction laboratories and general inspection units, there should be a PCR positive control, which is an important reference mark for the success of the PCR reaction and whether the position and size of the product band meet the theoretical requirements. The positive control should be a sample with moderate amplification and good reproducibility, which is identified as the product by various identifications. If a recombinant plasmid is used as a positive control, its content should be low but not high (less than 100 copies).

2、Negative control: Every PCR experiment must be a negative control. It includes:

(1) Specimen control: If the tested specimen is serum, use the identified normal serum as a control; if the tested specimen is a tissue cell, use the corresponding tissue cell as a control.

(2) Reagent control: Do not add template DNA or RNA to the PCR reagents and perform PCR amplification to monitor whether the reagents are contaminated.

3、Repeatability test.

4、Select primers in different regions for PCR amplification. [1]

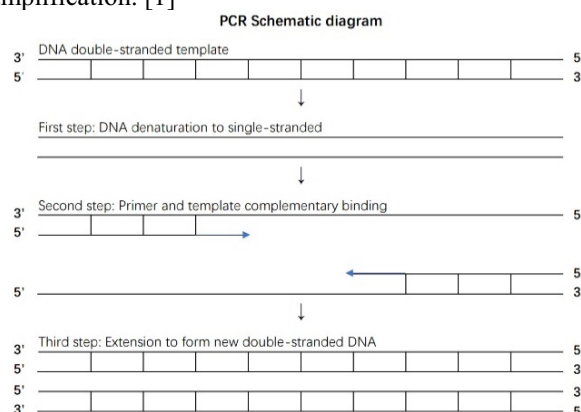


Figure 2. PCR Schematic diagram

4 LIMS system

LIMS is an information management system that combines database-centric information technology and laboratory management requirements. Laboratory Information Management System (LIMS), Laboratory Information Management System. LIMS (Laboratory Information Management System). One of the functions of LIMS is to provide statistical analysis of various

information in the entire laboratory, thereby providing a reliable basis for fair evaluation, so LIMS is conducive to the realization of quantitative management.

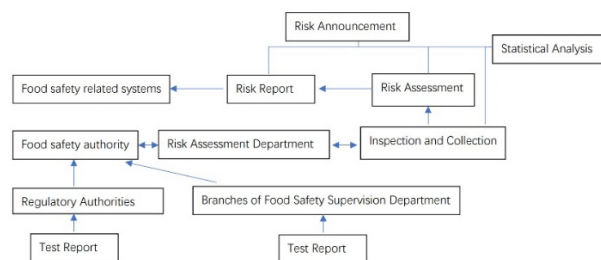


Figure 3. LIMS System Process diagram

4.1 Inspection process management:

The LIMS inspection process management can improve the standardization of the food inspection process, and at the same time can reduce the contingency of human operation, and make every inspection work and every inspection index more traceable as possible.

The main inspection process includes: acceptance, sample registration and sample management, inspection task assignment, inspection process management, experimental record management, result proofreading, consolidated report, report approval, report printing/sending, filing [2].

4.2 Sample Testing:

The core of the sample inspection of the LIMS system is to provide functions such as food inspection task management, food sample inventory management, initial data recording, inspection and inspection result import management, inspection and inspection report editing and review, and real-time query of food inspection and inspection progress.

4.3 Inspection quality control management:

Prepare plans for the quality of all samples, monitor the implementation process, and conduct risk assessments on the results of the implementation on a quarterly or yearly basis.

4.4 Report management

LIMS has a powerful reporting function. LIMS comes with a module dedicated to report design, allowing users to design reports that meet their own requirements. The types of reports can be regular inspection reports, analysis reports, quality reports, management reports, etc.

The combination of FTA card detection and PCR-DNA amplification technology has functional advantages: the PCR results of FTA card preserved samples and directly extracted samples are exactly the same. Efficient use: DNA combined with FTA card matrix can be amplified multiple times.

5 Application of FTA-PCR combined with LIMS system in actual detection

5.1 LIMS system

FTA-PCR combined with LIMS system can improve the detection accuracy in food safety production detection. According to the actual test results and the general requirements of experimental adjustment capabilities, including management and technical requirements.

As shown in Figure 3, after the FTA color card and PCR are used, the samples are stored in checkerboard sample locations. Each location has a unique identification code. For example, enter the number: A1-1 to quickly identify or Enter the specific information of this sample. 1-1 can be used to indicate the batch or location number, whether it is machine identification or manual search, when FTA-PCR detects samples, it not only has sample stability and detection convenience, but also increases the accuracy and accuracy of sample information.

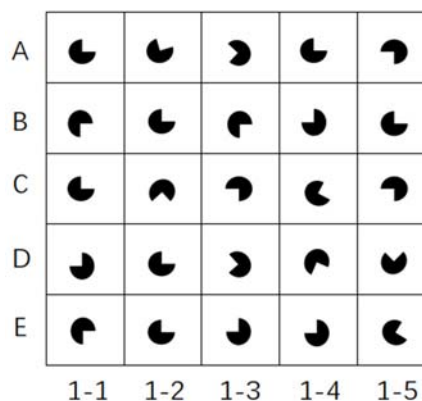


Figure 4. Example of the representation method of FTA color card samples based on PCR technology in the LIMS system

5.2 Simplicity

The operation process is more convenient and accurate to meet the needs of food safety testing, measurement and certification. The application of the LIMS system can realize the modern management of food inspection tasks and inspection processes, ensure the optimal configuration of food safety production inspection laboratories, and conduct real-time monitoring and traceability of the entire food safety inspection process [3].

PR-PCR Detection method:

FTA-PCR technology can be applied to the detection and analysis of food-borne microorganisms, viruses, drugs, mycotoxins and genetically modified foods in food safety testing [4].

Compared with traditional chemical methods, FTA-PCR technology has many advantages, and it is more and more widely used in food testing, but it also has certain shortcomings.

6 Conclusions

The combination of FTA-PCR biotechnology and the information technology with common databases as the core and the information management system that combines laboratory management needs. Although there is still a gap between the sensitivity and the chemical detection, the biotechnology is in the test methods and procedures. It is more convenient and intuitive, and the experiment period is shorter, which can make food safety more secure in different aspects.

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