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Article 1-3. The Application of Spectral Informatics on the Genetic Marker/光谱信息及其在遗传标记技术的应用

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Abstract

This article designs two new kinds of application methods of spectral informatics on the analysis of the DNA/RNA molecules.

Key words: Spectral Informatics, Genetic Marker, Fluorescence Binding Technology, Structural Biology, FISH Probe.

Method 1

The common methods used as chromosome binding technology mainly include Centromere Binding Technology (C-Binding), Guinacrine Binding Technology (G-Binding), Giemsa Binding Technology (G-Binding), Reverse Binding Technology (R-Binding), etc. These chromosome binding technologies are designed on the basis of the various staining chemicals [1], which reflect different regions or functions in chromosome. In my article, DAPI fluorescence binding technology, which results in the appearance of AT rich region on chromosome, is selected as an example of designing a new method in observing the structure of DNA molecules at three dimensions:

If the slide glass is the horizontal plane, and the vertical line is the eyesight line of microscope for DNA molecule observation, then the angle between the planes of AT DNA sequences and the eyesight line observed by microscope is ± α ($0° \leq \alpha \leq 90°$), and α is generally uniform in the DNA molecules of a species, but varies among different species. If this angle tends to be zero, then the DAPI binding tends to be not observed; If this angle tends to be 90°, then the DAPI binding tends to be more clearly observed by florescence microscope. The structure of DNA molecules can be consequently deduced by the clearness of fluorescence binding. Despite of fluorescence polarization, it is expected to obtain the reliable and re-occurring results when the other experiment conditions remains constant.

Consequently, it is to classify the DNA samples by the molecular cytogenetic

karyotype using FISH technology firstly, based on which the DNA molecular structure is further deduced according to the clearness of fluorescence binding. Then this structural biology is correlated to the functional groups of biological traits (such as pathological characters or immunological characters) as bio-markers.

Step 1. The molecular cytogenetic karyotype of chromosomes is analyzed by fluorescence in situ hybridization (FISH) technique;

Step 2. These chromosomes examined in experiment are classified by multivariate cluster analysis and genetic distance analysis on the basis of molecular cytogenetic karyotype, preliminarily leading to different families of chromosomes.

Step 3. Within each preliminary family of chromosomes, the DNA molecular structure of each chromosome is deduced according to the clearness of fluorescence binding described above. Then it is further to classify different sub-families of chromosome by this structural biology marker.

Step 4. Each sub-family of chromosome is correlated with the functional groups of biological traits (such as pathological characters or immunological characters) as bio-markers.

In order to quantify the clearness of fluorescence binding, the spectrometer is used to measure the brightness of each spectral line on fluorescence binding (or high-resolution images are taken and then the brightness of each spectral line is measured by image processing software). More chromosome binding showing other base pairs (such as GC base pair) are selected to compare with AT-rich base pair for further analysis.

Methods 2

There is a novel Tech designed for future Research & Development in Air quality Monitoring:

After a strain of pathogenic virus (or bacteria) has been identified in Lab, the unique DNA or RNA sequences specifically for this pathogen strain, which can not lead to PCR bands in the other microbial strains but result in clear PCR bands in this pathogenic strain only, are screened and synthesized into FISH probes for FISH step again. The methods of FISH probe preparation is listed [1]. Then the concentration of this pathogen strain in water solution can be measured by ultraviolet spectrophotometer, yielding a feasible and affordable method for routine air quality monitoring. The steps are listed below, taking virus as an example. Please note, the specific and unique DNA or RNA sequences (or the specific locus), which are indicated by the unique PCR bands in gene mutation strain of pathogen only, is the key for this selection of FISH probe preparation. It is expected that the gene mutation

strain of pathogen results in unusual and sharp increase of airborne density, as compared to its parental strain of pathogen, because the gene mutation significantly increases the genome replication rate. After airborne pathogen is collected by air quality samplers, the pathogen samples are transferred into the water solution:

Step 1. SDS-PAGE method for protein separation is conducted exactly, to separate the virus DNA or RNA molecules from its protein 'coat' around DNA or RNA molecules [2].

Step 2. In total five different densities of a virus strain (such as the gene mutation one) are prepared in Lab. The different densities of a virus strain can be obtained by adjusting the water solution concentrations proportionally.

Step 3. Specific FISH probe is prepared for this virus strain, and FISH procedure is conducted on five densities of this virus sample without the last drying process, leading to five different water solution concentrations (Sample 1, Sample 2 ..., Sample 5) of virus genomes binding FISH probe.

Step 4. The same volume of virus water solution are abstracted from Sample 1, Sample 2, ..., Sample 5, respectively, and the density of each virus water solution is counted by transmission electron microscopy after drying process.

Step 5. The fluorescence intensity in five different water solution concentrations (Sample 1, Sample 2 ..., Sample 5) of virus genomes binding FISH probe is measured by ultraviolet spectrophotometer. Then the regression equation for ultraviolet spectrophotometer is consequently worked out by correlating the fluorescence intensity in five different water solution concentrations (Sample 1, Sample 2 ..., Sample 5), to the correspondingly counted density of each virus water solution in step 4.

Step 6. After the regression equation for ultraviolet spectrophotometer is calculated specifically against the virus strain, air quality monitoring of this airborne virus family can be implemented routinely. Of course, this measurement method of virus density can be also applicable on other virus of different transmission patterns, which are not only applicable on the airborne ones.

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