DOCTORAL THESIS

Mechanism Investigation and Efficiency Enhancement of Nucleic Acid Extraction by Using Plant Fiber-Based Adsorbents

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ABSTRACT

Nucleic acid extraction is an essential part during biological analysis. Some studies show DNA adsorbability of filter paper that is composed plant fiber-based adsorbent. However, the species of plant fiber-based adsorbents, the optimized extraction conditions, and the adsorbing principle of this specific adsorption technique remain to be studied. Taking DNA as the representative, the aims of this project were to offer a more complete scheme of the plant fiber-based adsorbent DNA extraction, identify a suitable plant fiber-based adsorbent for DNA adsorption, optimize this specific adsorption technique, explore the adsorption mechanism between DNA and plant fiber-based adsorbents. In order to achieve the goal, we investigated different approaches of current DNA extraction, screened different types of plant fiber-based adsorbents, modified and reconstituted DNA extraction methods, explored the feasibility of DNA extraction by novel means, optimized DNA extraction on the selected plant fiber-based adsorbents, analyzed the mechanism of the DNA to plant fiber-based adsorbents, validated the new approach using different samples, and further designed an automatic high throughput extraction device. Nanodrop spectrophotometer, electrophoresis and PCR were used to demonstrate the quantity, quality, integrity and utility of the extracted DNA. Furthermore, the crystallinity, hygroscopicity, pore size distribution, zeta potential and composition of the plant fiber-based adsorbents were studied to explore the principle of this specific method of adsorption. This project provided novel avenues for DNA extraction using plant fiber-based adsorbents. Moreover, it identified new efficient plant fiber-based

adsorbents for DNA extraction, including bamboo paper and degreased cotton; optimized the extraction conditions. This study showed that this novel method is 4.2 times more efficient than current approaches. Furthermore, it showed the strong correlation of crystallinity and hygroscopicity of plant fiber-based adsorbents in their DNA extraction capacity. The mechanism of adsorbed DNA on the plant fiber-based adsorbents is demonstrated for the first time. Eventually, the method could be used for nucleic acid extraction with an automatic high-throughput extraction device as depicted in the thesis. This is the first study to systematically explore the application of plant fiber-based adsorbents in DNA extraction. It lays a foundation for guiding relevant future studies on a multi-disciplinary basis. Further study can be enhanced by the manufacture of an 'ideal' plant fiber-based adsorbent and the optimization of the adsorption, washing and elution conditions for different kinds of nuclear acid extraction. Also, commercial kits could be developed using this technique for nucleic acid extraction and detection. Moreover, an all-in-one instrument for automatic extraction and detection could be developed.

Keywords: nuclear acid extraction; DNA extraction plant fiber-based adsorbents; bamboo paper; degreased cotton; crystallinity; hygroscopicity.