B38 Probing Single Strand DNA Using Bioconjugated Quantum Dots

Shannon Gross, BS, and Jorn Chi Chung Yu, PhD, Sam Houston State University, Department of Chemistry 1003 Bowers Boulevard, Box 2117, Huntsville, TX 77341*

After attending this presentation, attendees will have the opportunity to discuss a method for detecting small amounts of DNA with a novel nano-material which has unique advantages over current dyes. The ease of adaptation of this technique to forensic labs from other chemistry-focused areas will be shown. Discussion of similar efforts towards advances in science being applied to forensics will be encouraged.

This presentation will impact the forensic community by demonstrating a new method that will impact the determination of profiles of those DNA samples that couldn't be determined by conventional DNA analysis procedures. Controlling high variations of small quantity, and degraded DNA samples have been a challenging problem in forensic DNA profiling. The investigation of novel nano- materials and single strand DNA is potentially applicable to single cell DNA analysis.

DNA analysis of small quantity, and degraded samples has been a challenging problem in forensic DNA profiling. Our investigation of novel nano-materials in single strand DNA is potentially applicable to single cell DNA analysis. The new development will have a great impact on the determination of profiles of those DNA samples which are too degraded or small to be determined by conventional DNA analysis procedures. Attendees will have the opportunity to discuss our method for detecting small amounts of DNA with quantum dots and the differences between quantum dots and typical fluorophores which have influenced the method development.

Quantum Dots (QDs) are semiconductive nanocrystals with fluorescent properties dependent on the particle size which can be synthesized but are also commercially available from a number of sources. They are more ideal for trace analysis than current fluorophores because they are more strongly fluorescent and typically have a slower decay rate. The nanocrystals are soluble in organic solvents unless they are coated with a polar group. It is this group which typically determines the chemistry to be employed to conjugate it to a molecule in aqueous solution. After the adaptation of QDs to aqueous solution, attempts to supplement or replace current fluorophores and dyes in biological and chemical fields have gained great attention in the scientific community. Some successful replacements of fluorescent cellular dyes, even in vivo, have demonstrated the versatility of this novel material.

Generally, strategies on bio-conjugation of QDs have been mainly based on the covalent linkage, electrostatic attraction and biotin–avidin interactions of QD surface ligands with target molecules. Determination of functionalities of QD bioconjugates are usually characterized by microscopic, spectroscopic techniques, immunoassay, and gel electrophoresis. In our previous work, we have adapted the established carbodiimide-mediated coupling reaction (such as N-(3-dimethylaminopropyl)-N'-ethylcarbodiimide (EDC) - N- hydroxysuccinimide (NHS) chemistry) to allow conjugation of antibodies to water soluble QDs. Biological activity of antibody conjugated QDs was investigated by capillary electrophoresis immunoassay (CEIA). Separation of un-reacted QD bio-conjugates and immunocomplex was visualized in the electropherogram.

To improve the limit of detection of forensically valuable DNA while limiting additional costs in adopting new techniques is desirable within the forensic community. In this work, single strand DNA analysis was investigated by using a commercially available biotin conjugated QDs (biotin-QD). Strapavidin was selected as a bridging molecule between biotin-QD and a biotinylated DNA strand. Detection of different sizes of DNA was achieved by a genetic analyzer (Applied Biosystems, ABI 310). Optimization of the method, including injection time, buffer selection, buffer additives, detection wavelengths, will be reported and discussed. The goal is to use this novel nano-material combined with the separation power of CE to develop a rapid detection assay for single strand DNAs. This new technique offers the potential to analyze smaller sample sizes, thus minimizing evidence destruction and allowing analysis of samples expected to be too small to detect by current methods. It is also expected that the technique is highly transformative, because, other than the bio-conjugation chemistry, the technique utilizes equipments and materials currently available in a forensic DNA lab.

Single Strand DNA Profile, Quantum Dot, Capillary Electrophoresis

 ** Presenting Author*