Employing Botanical DNA to Forensically Tag and Authenticate Objects for Security Purposes

James A. Hayward and MeiLin Wan; Applied DNA Sciences, Inc.; Stony Brook, New York, USA

Abstract

Botanically derived, deoxyribonucleic acid (DNA) taggants can be used as a means to forensically tag and authenticate objects for security purposes. Typically, the sequence dependent, encrypted DNA tags are embedded into inks, varnishes, adhesives, as well as paper, laminates and a range of substrates. This paper will provide recent findings on 1) the DNA techniques utilized to authenticate the DNA in a botanically-marked printed materials, 2) methods used to DNA-tag components and products, 3) methods used to authenticate DNA-tagged materials, 4) the value of DNA toward enhancing quality control, and 5) the use of DNA tags to identify originals, interdict counterfeits and prevent diversion, as DNA is recognized as a form of forensic evidence trusted by law enforcement and admissible by courts around the world.

Keywords

DNA, forensic, authentication, security, covert, counterfeit, anti-counterfeit, diversion, fraud, taggant

Counterfeiting – A Global Threat

The evolution of counterfeiting as a trade nearly parallels the evolution of technology itself. The last two decades have witnessed explosive growth of technology, and the condensation of travel, communication and the massive impact of the internet ensured these new technologies were laterally propagated instantly across the planet. Now counterfeits emerge on the market nearly simultaneously with new product launches, in time for the counterfeits to benefit from the marketing efforts expended by the original. The World Customs Organization estimated that annual global trade in illegitimate goods was roughly \$600 billion in 2004, and was expected to double by 2014, representing between 5% to 7% of all world trade (Source: The International Anti-Counterfeiting Coalition). But this is more than a vexing nuisance for brand owners. Counterfeits threaten economies, destroy health and take lives, and destabilize the military.

The Defense Standardization Program Office (DSPO) Journal (Oct/Dec 2009) recognizes the definition of a counterfeit electronic part as "one whose identity or pedigree has been deliberately altered, misrepresented or offered as an authorized product." Early reports of counterfeit electronics emerged from industry. In May of 2006, the New York Times reported a massively coordinated effort of 18 factories in China to copy the entire product line of NEC. Nearly every aspect of the company's brand and product line had been replicated and sold along parallel paths. NEC even found itself honoring the warranties for the fakes.

In June of 2007, the US Department of the Navy suspected that an increasing number of counterfeit electronics was infiltrating the Department of Defense (DOD) supply chain. In collaboration with the Department of Commerce (DOC), a study was initiated to assess the defense industrial supply base and to determine the statistical frequency of counterfeit electronics penetrating DOD. The results of this study, finalized in January, 2010 (US DOC "Defense Industrial Base Assessment: Counterfeit Electronics") showed:

- All elements of the military supply chain have been directly impacted by counterfeit electronics;
- Stricter testing protocols and quality practices are required; and,
- The use of authentication technologies by parts manufacturers, distributors and integrators should be expanded.

Current Authentication Solutions are Inadequate

Efforts to secure the authenticity of electronics are first encountered at the primary and secondary packaging. Traditional security platforms to prevent counterfeits are now also part of the counterfeiter's target and consequently within the retinue of counterfeiter's resources. New advances in holograms, optical strips and RFIDs are often available as near-perfect copies within days of their initial launch.

Exacerbating the inadequacy of packaging security solutions, most distributors and integrators store microchips and semiconductors in high-volume bins, capable of matching the demands on their supply. This "bin approach" excludes the packaging to save space and time, so security must be implemented at the product level.

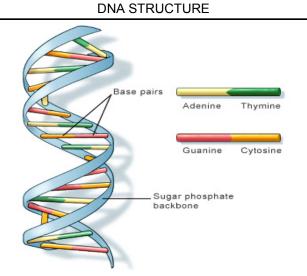
Product inspections offer limited value as a method of authentication. External visual inspections should not be used as a standalone authentication. Physicochemical characterizations are often destructive and rely on a degree of similarity to a bona fide original and the tolerance of the measurements.

Taggants can provide a unique code or fingerprint to authenticate originality. However, as evidentiary tools, the value of a taggant increases as a function of the density of its in formation content. Mineral taggants, which simply provide parameters of chemical identity and concentration, are only effective as rapid screening tools, often by hand-held detectors. Stochastic arrays of fibers or particles are difficult to incorporate in the media used to fabricate microchips and semi-conductors. Stochastic arrays of nanoparticulate ferrite can generate complex "fingerprint" patterns, but care must be exercised to ensure the magnetic field does not interfere with semiconductor function.

Forensic DNA as a High-Content, High-Resolution Taggant to Track Provenance and Ensure Authenticity

Evolved over eons, DNA (deoxyribonucleic acid) provides the blueprint for all of biology. The information content is massive, highly customized by organism, and capable of supercoiling and compaction into infinitesimal space. With a capacity for content that is often compared to computer machine code, DNA is a linear polymer of extraordinary molecular weight that stores its information as a sequence of infinitely variable organic bases. However, unlike binary machine code, DNA's code is quaternary, storing its content as a linear array of four organic options for each bit. With the uncanny variability of DNA (consider all the variable across organisms and the variation within species), matched by a stringent fidelity, and detection methods that readily identify single molecules of a unique sequence (a detectability that will never be matched by any chemical or physical assay), DNA has become the "Gold Standard of forensics." Challenged by courts around the globe since 1980, there is no better proof of identity, nor is any better proof of identity likely to evolve.

Used by forensic laboratories all around the world, including the FBI, **DNA authentication is absolute in character**. When used to identify individuals or to establish paternity, the error frequency for false positives is less than one in a trillion.



Source: the U.S. National Library of Medicine.

DNA Science

Structurally, the DNA of multi-cellular organisms consists of two long polymer strands of simple units called **nucleotides**. Each nucleotide is composed of a base, a sugar, and a phosphate. Nucleotides are arranged in two strands that form a spiral called a double helix. The structure of the double helix is somewhat like a ladder, with the base pairs forming the ladder's rungs and the sugar and phosphate forming the vertical sidepieces or backbone of the ladder, as depicted in Figure 1. There are four bases in DNA: adenine (A), guanine (G), cytosine (C), and thymine (T).

The order or sequence of these bases determines what biological instructions are contained in the gene, providing the information needed to build and maintain an organism. This sequence is normally represented by using the initial of the base (e.g., A for adenine, G for guanine, and so forth), generating a lineal code. For example, the sequence ATCGTT might instruct for blue eyes, while ATCGCT might instruct for brown eyes (Source: the National Human Genome Research Institute).

At the chemical level, the nuclei of all plants and all animals contain DNA in the same double helix shape and made out of the same four base compounds. The sequence of the base pairs, and the information they encode, is what determines the characteristics, shape, and function of the organism. As scientists continue to learn about the unique structure of DNA, they are exploring innovative techniques to harness the physiological power of the substance and apply it to commercial endeavors.

The Technologies of Applied DNA Sciences

Applied DNA Sciences (OTCBB: APDN), Inc., (based in the US on the campus of Stony Brook University in New York, and in Northern England Associated with the University of Leeds) provides botanically derived, deoxyribonucleic acid (DNA) taggants to provide forensic security and authentication solutions to protect products from counterfeiting and diversion. Patented and proprietary, **these complex botanical DNA sequences cannot be copied and are encrypted and stabilized to withstand the most aggressive chemical treatments.** Table 1, below, shows the robustness of the botanical DNA platform that will survive a wide range of UV, X-ray, gamma ray, pH, chemical and thermal exposures.

Trademarked as SigNature[®] DNA, APDN's botanical DNA has marked millions of commercial items. The Department of Energy (DOE) at the Idaho National Laboratories validated the platform. The platform is used by police forces throughout Europe to prove provenance of cash and logistic items.

APDN's Intellectual Properties (patents and trade secrets) provide the mechanisms for protecting DNA in harsh chemical and physical environments, the insertion of DNA into plastics, films, adhisves, inks, metal surfaces, and protects the methods used to enable DNA to function as a commercial authentication tool.

Table xx Applied DNA Sciences, Inc. DNA MARKERS' STABILITY

Test Specifics	Results
Equivalent to more than 350 years of UV energy accumulation in Denver	Stable
4 times the X-ray exposure by scanning machine in an airport	Stable
30 kGy (kilo-Gray) radiation exposure by γ-ray sterilization machine	Stable
Exposed to pH of 1 to 14 overnight, higher than 250 degrees celsius for 4 hours	Stable
	Equivalent to more than 350 years of UV energy accumulation in Denver 4 times the X-ray exposure by scanning machine in an airport 30 kGy (kilo-Gray) radiation exposure by γ-ray sterilization machine

Source: Applied DNA Sciences, Inc.

Applying SigNature DNA Taggants

The SigNature DNA process begins by taking the DNA from the cells of plants and arranging it to create a group of specific DNA codes or sequences called taggants. These taggants are then incorporated into a product as a unique and traceable identification system. As illustrated in the Figure (page 5), the SigNature DNA solution consists of three steps: (1) creating and encapsulating an encrypted DNA segment or taggant; (2) applying the taggant to a product or material; and (3) detecting the presence and authenticating the specific DNA taggant.

Botanical SigNature DNA

SigNature DNA security platform uses "green" botanical DNA to manufacture highly customized and encrypted DNA taggants that identify objects while addressing the issues of counterfeiting and fraud.

SigNature DNA:

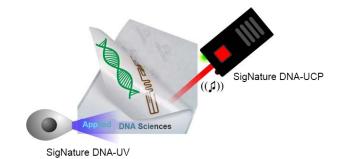
- is an environmentally friendly, "green" technology based on botanical DNA,
- is used as a taggant at extremely low concentrations,
- will not alter the quality of any carrier (such as ink, coatings, adhesives, plastics or commercial products)
- will not require major changes in manufacturing process,
- persists, in physicochemical extremes such as harsh outdoor environs and high temperatures,
- is chemically compatible in a wide range of security inks (overt and covert), varnishes, adhesives and substrates,
- can be layered with other security measures, including barcodes, holograms, RFIDs, etc.
- is detectable in the field, and finally,
- can be forensically authenticated when analyzed in the laboratory.

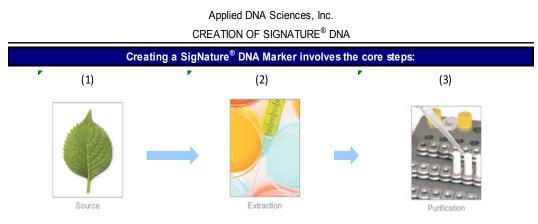
Key Attributes of SigNature DNA

Applied DNA has proved that its botanical DNA technology provides the following advantages over existing competitive security options.

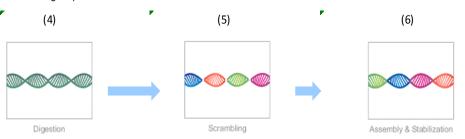
- Broad Applicability. Applied DNA's ability to integrate taggants in a variety of ways allows SigNature DNA technology to be embedded into almost any consumer product or item. SigNature DNA taggants do not alter the quality of the product and are stable and long-lasting. In addition, as SigNature DNA technology is safe to consume, it can be used in pharmaceutical drug tablets and capsules although it will require FDA approval.
- Easily Integrated with Printed Electronics and Other Anti-Counterfeit Technologies. In order to stay ahead of counterfeiters, multiple forms of technology should be used in printed electronics such as SigNature DNA taggants with RFID devices, labels, serial numbers, holograms, and other marking systems using inks, threads, and other media. The Company believes that combined with other traditional methods, the SigNature DNA solution provides a significant deterrent against counterfeiting, product diversion, piracy, fraud, and identity theft.

UV-DNA with RFID Label

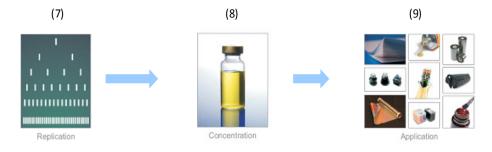




- (1) Source: Several selected plant species, apart from well studied plants, are used as source materials for Applied DNA Sciences' unique patented SigNature[®] DNA markers.
- (2) Extraction: Plant materials are then processed together for genomic DNA extraction. Plant cells are disrupted to release genomic DNA, which is then separated from insoluble plant debris.
- (3) Purification: Crude extracts of plant DNA is then processed to remove proteins and various contaminants to obtain DNA with higher purities.



- (4) Digestion: In order to rearrange plant genomic DNA for unique sequences, purified plant genomic DNA is digested with enzymes to obtain DNA segments with various lengths.
- (5) Scramble: DNA with various lengths is mixed together to create a soup of length polymorphic DNA.
- (6) Assembly: From that soup of length polymorphic DNA, assembly of unique DNA sequences can be accomplished by ligating digested DNA fragments together, which then undergo selection processes for DNA fragments with proper sizes.



- (7) Stabilization: Assembled unique sequence DNA fragments are stabilized through patented methodology to create stable DNA that can last over several hundred years.
- (8) Replication: Although the Company's DNA is unique and stabilized, it can be multiplied in huge quantities in a relative short period of time by using well known molecular biology techniques.
- (9) Application: The Company's SigNatue[®] DNA marker is unique and stable, and has forensic qualities that no other security markers can achieve. The application SigNature[®] DNA marker has multiple fronts, such as security printing, cash in transit, and identity/authenticity protection.

Source: Applied DNA Sciences, Inc.

Key Attributes of SigNature DNA ctd.

- Secure. Applied DNA maintains its records of DNA sequences on a secure server. Sequences are encrypted, available to individuals on only a partial basis.
- Low Cost and High Accuracy. SigNature DNA taggants are relatively inexpensive when compared to other anti-counterfeiting devices, such as RFIDs, integrated circuit chips, and holograms. The costs associated with the production of DNA taggants are not significant since the amount of DNA required for each taggant is small, and the cloning of the DNA segments is performed inside microorganisms such as yeast or bacteria, which are highly productive and inexpensive to grow. In addition, incorporating SigNature DNA into products does not require major changes to the manufacturing process or logistic chain. The relatively low cost of SigNature DNA does not affect its reliability. The probability of mistakenly identifying a SigNature DNA taggant is less than one in a trillion, making it virtually impossible to wrongly identify something marked with SigNature DNA.
- Resistant to reverse engineering or replication. The botanical SigNature DNA platform is virtually impossible to copy. Proprietary methods yield DNA taggants so complex that they are statistically impossible to duplicate. In addition, the DNA segment used in the taggants needs to be replicated billions of times in order for detection and identification to take place, a process that can only be achieved by applying matching strands of DNA. Thus, the sequence of the relevant DNA in a specific taggant must be known in order to manufacture the primer needed for the detection process. The inability of counterfeiters to duplicate SigNature DNA taggants has been proven in the marketplace. A European media manufacturer's production of 600 million optical digital video discs (DVDs) in China included 12 anticounterfeiting security platforms. Within nine months of the launch of the DVDs, 11 of those 12 anti-counterfeiting platforms were replicated in the market place, with SigNature DNA being the only exception. Moreover, SigNature DNA taggants on those DVDs remain effective to date, three years after launch.
- Scalable. DNA taggants can be produced in essentially infinite variety. Individual taggants of defined sequence can be manufactured in large scale. For example, in a single batch APDN recently marked 250,000 kgs of raw cotton fibers before ginning. DNA-tagged, individual fibers could be traced throughout the manufacturing process to the completion of retail garments and apparel.

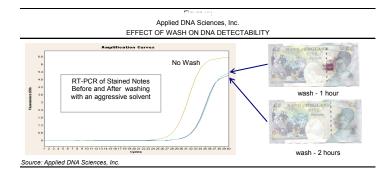
Tagging Cash, the Ultimate Arbiter of Brand Protection and Secure Logistics

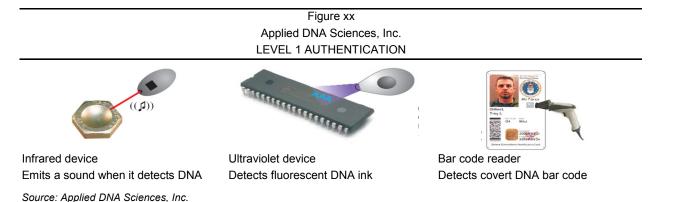
Since January 2008, Applied DNA has been working with Loomis UK, a cash-handling company that moves over £150 billion in cash annually. In July 2009, Applied DNA announced that it successfully authenticated stolen bank notes and other recovered evidence received from the UK Metropolitan Police, which is used to assist in the prosecution of the alleged criminals. The SigNature DNA markers present in the recovered evidence resisted removal even after vigorous washing and were also detected on personal items such as clothing and mobile phones belonging to the suspects in the investigations. To support this initiative, Applied DNA opened its first DNA Authentication Laboratory in Yorkshire, England.

SigNature DNA Protects Cash-in-Transit



UK Police departments have retained Applied DNA Sciences to assist in forensic authentication and the provision of expert witness statements. By May of 2010, over thirty criminal investigations in the UK have used SigNature DNA taggants on recovered, stolen currency. More than thirteen of these cases have progressed to conviction with cumulative sentences over 100 years. Police departments throughout Europe have begun using SigNature DNA in covert operation. In March of 2010, the head of the Swedish National Police issued a statement that it plans to use APDN's DNA taggants throughout its operations.





DNA Detection and Authentication

Applied DNA Sciences, offers three levels of detection for its SigNature DNA markers:

"Spot Test" DNA Detection

Instant detection of DNA markers typically embedded with up-converting phosphors, UV luminescent inks and other rapid optical reporters. Adding these non-forensic taggants to DNA affords the ability to quickly screen for the presence or absence of our DNA taggants using commercially available portable handheld detectors.

Forensic DNA Authentication

Applied DNA botanical SigNature DNA platform utilizes a method called **PCR**, or **Polymerase Chain Reaction**, to authenticate genuine items marked with its unique DNA sequence. PCR is a technique used in medical, biological, and forensic laboratories for a variety of applications. It is used to amplify specific regions of a DNA strand (or DNA target). This can be a single gene, a part of a gene, or a non-coding sequence. PCR can be used to amplify a single or multiple copies of a piece of DNA across several orders of magnitude, generating millions of copies of a particular DNA sequence. This authentication process provides absolute certainty about the presence or absence of specific types of DNA taggants, resulting in a forensic level of authentication which cannot be found with many other systems.

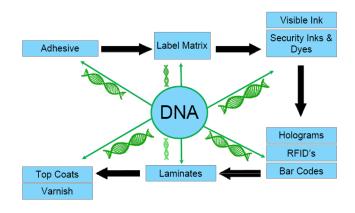
Applied DNA has developed proprietary methods comprised of multiplexing primers that are key components which allow for targeted and repeated amplification. With properly designed primers, PCR can amplify very specific DNA sequence fragments, even when there is only one copy of DNA. This is often critical for forensic analysis, when only a trace amount of DNA is available as evidence. In the case of Cash-in-Transit, PCR enabled the authentication of SigNature DNA-marked, ink-stained banknotes and evidence recovered by the UK Police even when the stained banknotes had been extensively washed.

Disclaimer

The assumptions, estimates, forecast and analyses made in this document are solely those of Applied DNA Sciences, Inc. No representation or warranties are made as to the fitness, accuracy, completeness, merchantability or use of the information presented for any purpose. In addition to PCR amplification, Applied DNA also utilizes **capillary electrophoresis (CE)** for forensic authentication. The size polymorphic results can be analyzed quantitatively by extrapolating DNA fragment size precisely using an internal size standard. The fluorescently labeled DNA fragments are separated by the gel within the capillary tube and excited by a powerful **Argon laser** for signal detection. The detection point of each DNA fragment is recorded and compared against an internal size standard for size determination. The measurement and analysis are primarily performed with specialized software. For DNA sequence analysis, DNA fragments typically go through a sequencing reaction and sequences can then be analyzed using the same CE machine.

DNA to Secure Printed Electronics

Botanical DNA taggants can be incorporated into electronic components in assorted ways, such as in-mold plastics, metal coatings, as well as security inks, varnishes, adhesives, threads etc. Using a combination of inks, raw material, and additional security measures in various stages of the manufacturing process, DNA provides highly specific and secure anti-counterfeit solutions for microchips and all electronic components.



For additional information, please contact:

James A. Hayward, Ph.D., Sc.D., President & CEO MeiLin Wan, Director, Project Management Tel: +1-631-444-6370 E-mail: info@adnas.com www.adnas.com