Countering Counterfeiting of Drugs: Unique Fluorescent Inks for Direct Printing onto Pharmaceuticals

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Abstract

Counterfeiting of pharmaceuticals is a growing and extremely concerning problem as supported by the increased prevalence, revenues (in the tens of billions), and sophistication of counterfeit medications. Current anti-counterfeit technologies for pharmaceuticals are generally easy for counterfeiters to imitate and typically rely on printing on packages, engraving or standard printing on pills, and pedigrees (only adopted by a few governmental organizations). Considering the dire impacts of counterfeit pharmaceuticals, including loss of life, an extra layer of difficult to replicate anti-counterfeit technology could alert medical personnel and consumers of potential counterfeit pharmaceuticals prior to consumption. In this contribution, two "non-toxic" fluorescent compounds (fluorescein and tryptophan) where formulated into inks for direct printing on medications. Fluorescein is a compound currently used in eye surgeries and tryptophan is a non-toxic amino acid. These inks were successfully printed on a number of medications and evaluated for authentication purposes. The success of these inks may ultimately lead to an easy way for medical personnel and consumers to identify counterfeit pharmaceuticals.

Introduction

Counterfeiting of pharmaceuticals is a growing and extremely concerning problem as supported by the increased prevalence, revenues (in the tens of billions), and sophistication of counterfeit medications and the dire consequences, including death, of consuming counterfeit medications [1]. For example, 2000 to 2010, the number of reported incidents of counterfeit medicines increased 10-fold (from just under 200 to over 2000) [2]. Multiple examples of counterfeit pharmaceutical consumption taking human life have been reported including the staggering number of deaths attributed to counterfeit anti-malarial drugs, cough syrup, and heparin [3-5]. Current anti-counterfeit technologies for pharmaceuticals are generally easy for counterfeiters to imitate and typically rely on printing on packages, engraving or standard printing on pills, and pedigrees (only adopted by a few governmental organizations) [6, 7]. Considering the dire impacts of counterfeit pharmaceuticals, an extra layer of difficult to replicate anti-counterfeit technology could alert medical personnel and consumers of potential counterfeit pharmaceuticals prior to consumption.

A semi-covert fluorescent security label that can be printed directly onto medications may provide a simple and efficient next level security measure by which an authentic pill may be distinguished from a counterfeit. The goal of this project was to synthesize a number of "non-toxic" fluorescent compounds and formulate them into inks for printing directly on medications. In this contribution, we will present recent findings in the use of non-toxic inks for direct printing on medications.

Materials and Methods

Materials

Sodium hydroxide (reagent grade), concentrated sulfuric acid and methanol (HPLC grade) were obtained from Fisher Scientific. Resorcinol and pthalic anhydride were obtained from Acros Organic. Great Value brand blue food coloring was used.

Synthesis and Ink Formulation

Fluorescein for printing on pharmaceutics was of interest both for its relatively covert and relatively non-toxic nature. Fluorescein synthesis reported was based on a condensation reaction reported by Adolf Von Baeyer (1871) [8, 9] according to Figure 1. In a reaction vessel, 1 g of resorcinol, 1 g of phthalic anhydride and 4 mL of concentrated sulfuric acid were carefully heated to 200° C until the solid dissolved in acid and became a dark red mixture. This reaction mixture was then allowed to cool to room temperature.

Deionized water (60 mL) was added to the cooled reaction mixture and it was stirred for five minutes to dissolve the product. Then, NaOH (100 mL of a 1 M aqueous solution) was added to the solution and it was stirred to quench the sulfuric acid and to form the sodium salt of fluorescein. Subsequently, concentrated HCl was added dropwise to the solution until an orange precipitate was formed. The mixture was then filtered using a Buchner funnel apparatus and the obtained solid was allowed to dry overnight.

Fluorescein ink comprised of 1.5 mM solution of fluorescein in 1:1 (v/v) methanol:water was prepared [10]. Some inks were prepared using blue food coloring dyes to mask the fluorescein's yellow color in order to increase the security. A solution comprised of 20% (w/v) blue food coloring in water was prepared. Two drops of the blue food coloring were added per mL of the 1.5 mM fluorescein solution [10].

Tryptophan was of interest for printing on pharmaceutics because of it is non-toxic, cost-efficient, and covert due to UV to UV fluorescence. A tryptophan based ink was formulated using the following procedure.

A 0.1 M potassium phosphate buffer was prepared in a 10 mL volumetric flask. The buffer was prepared by adding 0.62 mL of K_2HPO_4 and 0.38 mL of KH_2PO_4 and diluting to 10 mL with water. The final pH of the buffer was 7.0.

A tryptophan ink was then formulated by placing 30.6 mg of L-Tryptophan into a 5 mL volumetric flask. Potassium phosphate buffer (2 mL) was then added in order to stabilize the pH of the tryptophan and prolong the shelf-life of the solution. Water was

then added up to 5 mL. The final concentration of the tryptophan solution was 30 mM.

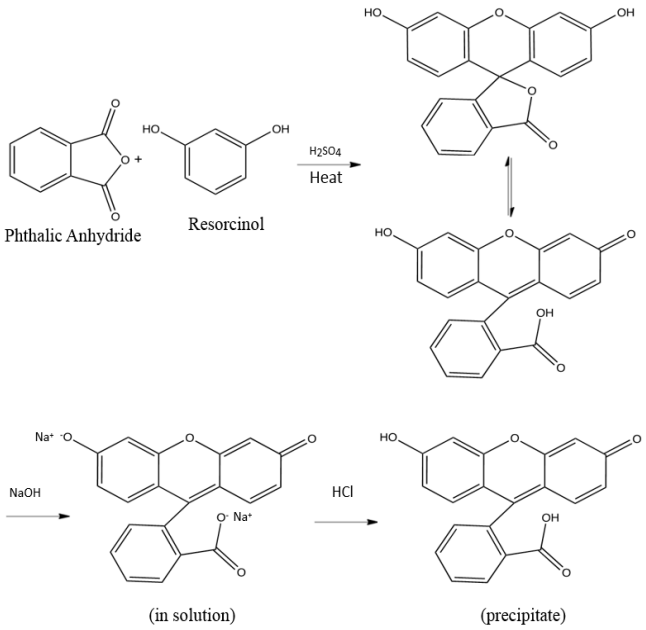

Figure 1. Schematic representation of fluorescein synthesis.

Equipment

All down-converting samples were viewed under long-wavelength (365 nm, 115 V, 60 Hz) ultraviolet light using a UVP lamp. All UC samples were viewed under Opto Engine LLC MDL-H-980 nm infrared diode laser system. The visible spectra of the OV patterns were obtained using a VSC® 6000/HS desktop video spectral comparator. The images were captured photographically using a Nikon D3000 digital SLR with a AF-S DX Micro-NIKKOR 40 mm f/2.8G Macro Lens with a UV/IR filter.

Tryptophan detection was viewed using a modified Vivitar Vivicam F126 digital camera. The UV/IR filter was removed and replaced with an Edmund Optics 360 nm bandpass filter. Images were collected upon irradiation with the long-wavelength UV light.

The fluorescein inks, masked fluorescein inks, and tryptophan inks were printed on a variety of substrates using the direct-write, aerosol jet printer. For all the inks, different AutoCAD patterns that act as tool paths for the printer were designed. Ink (1-2 mL) was added to a glass vial, atomized, delivered to the nozzle of the printer, and deposited on the substrate. Various parameters of the printer were controlled using the software interface to produce line widths as small as 25 µm. High resolution patterns were achieved by optimizing the printing parameters of the printer. The substrates used include a) Equate® 200 mg ibuprofen tablet, b) Motrin® PM tablet, c) Bayer tablet, and d) Advil tablet.

Results and Discussion

Fluorescene

Characterization of the down-converting ink: Figure 2 shows the red fluorescein powder produced from the synthetic process mentioned above. When mixed in a 1:1 (v/v) methanol:water solvent, the dye appeared yellow-green, as shown in Figure 2b, and fluoresces bright green under long-wavelength ultraviolet excitation as shown in Figure 2c. The fluorescein excitation and emission spectra were recorded to determine the excitation and emission maxima at 448 nm and 519 nm, respectively (See Figure 3). The excitation-emission maxima are similar to the values reported by Bennet (2001) (excitation at 480 nm and emission peak at 525 nm) [11], with the deviation likely due to variations in the solvent nature.

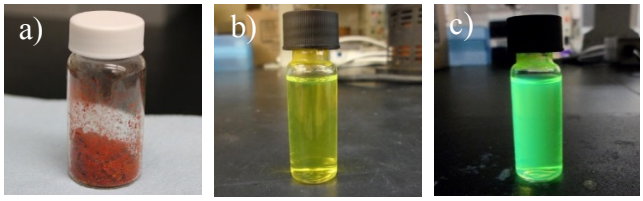

Figure 2. a) Red fluorescein precipitate. b) Fluorescein dissolved in methanol under ambient light. c) Fluorescein dissolved in methanol under long- wavelength ultraviolet light.

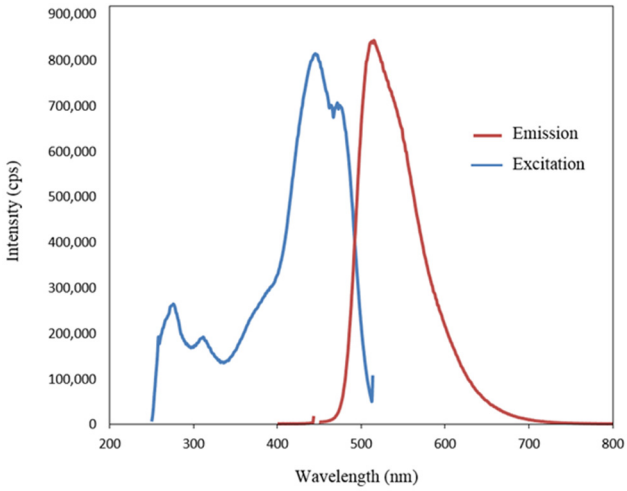

Figure 3. Excitation-emission spectra of fluorescein in water.

A QR code (size 4×4 mm and thickness ~1 µm) was printed on an ibuprofen tablet with the fluorescein ink using the aerosol jet printer. The printed QR code was covert under the ambient conditions, as shown in Figure 4a, but luminesced green when excited with a long wavelength UV light. The luminescent QR code was readable with a smart phone QR reader application used to directly open a desired webpage URL. Approximately 0.1 mL of ink is consumed to print a QR code of 4x4 mm size.

Another approach to produce covert/semi-covert printing on pharmaceutics was tested by printing a visible design on a pharmaceutical tablet using a colored ink embedded with down-converting ink. For demonstration purposes, we used food grade blue color. The blue food coloring does not fluoresce under long-wavelength UV irradiation, but the addition of fluorescein to the ink produces a fluorescent signature under the UV light. To evaluate this approach, blue food coloring with embedded fluorescein ink was printed onto a Motrin tablet as shown in Figure 5a. A QR code with the same dimensions mentioned earlier was printed on the tablet using an aerosol jet printer. These inks are highly fluorescent on Motrin tablets and this could be because of the tablet coating. The print on the tablet shows no sign of

fluorescent properties under ambient light (see Figure 5a), but is highly fluorescent under long-wave ultraviolet light (see Figure 5b). Indeed, the mixed blue food coloring/fluorescene ink printed on the Motrin tablet is more fluorescent relative to the fluorescene ink printed on the Ibuprofen tablet. We attribute the difference in brightness to the background scattering of the tablet.

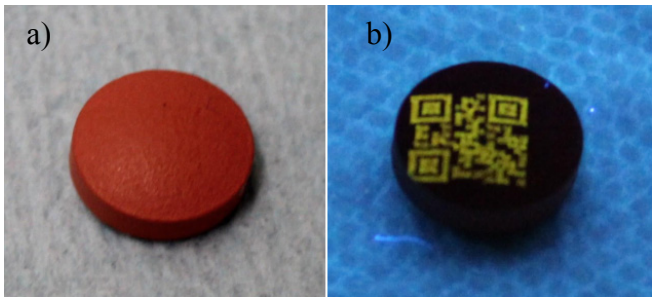

Figure 4. a) Ibuprofen tablet with covert fluorescein printing under ambient light. b) Ibuprofen with fluorescein ink printed when exposed to a long wavelength ultraviolet light.

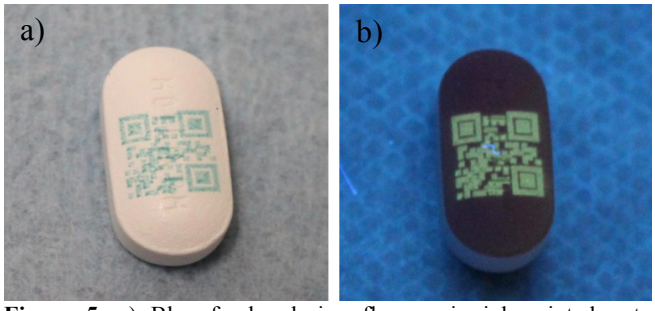

Figure 5. a) Blue food coloring fluorescein ink printed onto Motrin under ambient light. b) Blue food coloring fluorescein ink printed onto Motrin under long wavelength ultraviolet light.

Tryptophan

The emission and excitation spectra of tryptophan (in aqueous solution) are shown in Figure 6. The excitation spectrum was fairly narrow, with a maximum of about 297 nm. The emission spectrum of tryptophan was relatively broad and the bulk of the emitted light was still in the ultraviolet region of the electromagnetic spectrum. Although some of the light emitted can be considered in the visible spectrum (>400 nm), it was not visually apparent when exciting the tryptophan at 297 nm. Figure 7 shows the formulated tryptophan ink dropped on a microscope slide, dried, irradiated with 297 nm light, and imaged through a long-pass filter. Figure 7a shows the original image with the tryptophan fluorescence barely visible and Figure 7b shows a digitally enhanced image of the tryptophan on the microscope slide. The digitally enhanced image clearly reveals the UV fluorescence of the tryptophan spot. This ink formulation is currently being used to print onto coated pharmaceuticals. It was necessary to print on coated pharmaceuticals because compressed "chalky" pills generally fluoresced under the UV irradiation used. This created a large background fluorescence and masked the fluorescence of the tryptophan. Currently, bandpass filters for the irradiation source are being investigated to selectively excite tryptophan.

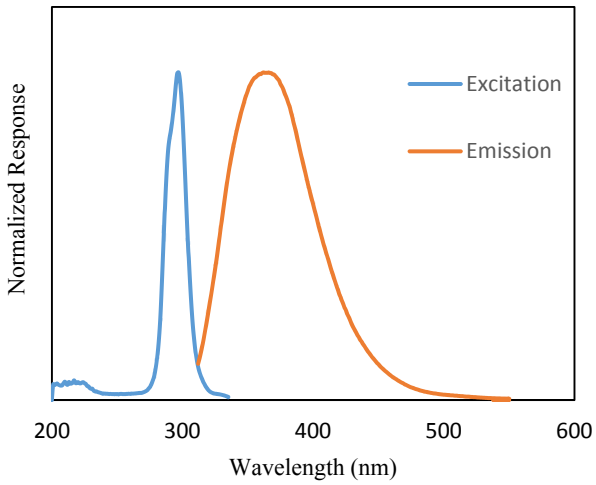

Figure 6. Excitation – emission spectra of tryptophan in water.

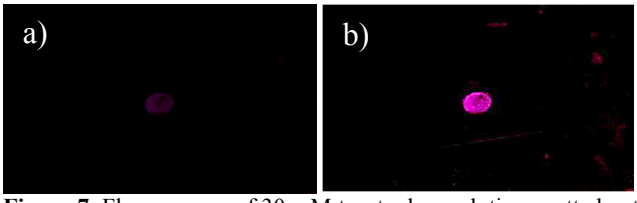

Figure 7. Fluorescence of 30 mM tryptophan solution spotted onto glass microscope slide. Image captured with longwave UV wand and modified Vivitar camera. a) Original photograph. b) Digitally enhanced image.

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Author Biography

Dr. Brian A. Logue received his PhD from Oregon State University in 2000. He then spent four years with the U.S. Army where he was deployed for Operation Enduring Freedom and subsequently directed a research group for the U.S. Army Medical Research Institute of Chemical Defense. He joined South Dakota State University in 2004 and is currently an Associate Professor working extensively on the analysis of cyanide, its metabolites, and cyanide antidotes with multiple publications in the area. Dr. Logue also studies Materials and Organic Chemistry in the area of organic modification of inorganic surfaces and development of security inks