DETECTION AND ENHANCEMENT OF LATENT FINGERPRINTS USING DEXTRAN-BASED BIOPOLYMER POWDERS OBTAINED FROM LIQUID ANTHOCYANIN EXTRACT

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Abstract: Dextran is extensively exploited in medicine and pharmacy, but, currently no studies using this biopolymer as a powder system for enhancement of latent fingerprints were published. In this paper four different formulations of dextran-based biopolymer powders, obtained by simple precipitation of dextran within anthocyanin solution, were synthetized and characterized in order to determine potential of these biopowders in forensic application. Since detrimental effect on humans is often present while routinely employing commercial dusting methods, the main advantage of prepared dextran-based biopowders are their non-toxic properties, contributing to the safer/healthier operating conditions. The interactions between components of the systems were confirmed by FT-IR analysis. Optical microscopy was used to determine the size of the prepared biopowders, while simultaneously confirmed the interaction between powders and the sweat/lipid residues present in the latent trace. The results demonstrated the potential of novel dextran-based biopowders to supplement routinely employed physical systems in visualizing latent fingerprints.

Keywords: Latent Fingerprints, (Bio)polymers, Dextran, Brassica oleracea var. capitata f. rubra, Anthocyanins, Forensic Science.

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INTRODUCTION

Fingerprints possess unique features, specific for each person and, thus they are one of the most valuable forensic evidence that could be found at the crime scene (Bumbrah, Sharma, & Jasuja, 2016). They often remain as random prints on surfaces of different objects, when the fingertip comes in contact with the surface. Fingerprint features are classified as loops, arches and whorls. On the other hand, fingermarks contain some tiny, specific and distinguishing characteristics called minutiae points, which were necessary for reliable identification of persons (Mitrović, 1998). The papillary line traces are transferred by the sweat (eccrine) glands, which secrete sweat and other components through the sweat pores, leaving a trace characteristic for each person. Other compounds, such as blood, oil, ink, dye, etc. can often be found and transferred from finger surface to the substrate along with the fingerprint (Champod, Lennard, Margot, & Stoilovic, 2004). Therefore, three particular types of fingerprint could be found in forensic practice: patent, plastic and latent. From the forensic point of view, latent fingermarks are of particular interests. These traces are imperceptible and (when freshly deposited) consist of sweat and lipid secretions, where sweat contains, approximately, water (98%), minerals (0.5%), organic compounds (0.5%) and the 1.0% of other residuals (Färber, Seul, Weisser, & Bohnert, 2010). Therefore, these traces must be visualized first, which is why different optical, physical and chemical methods have to be employed. After visualization process is completed, these fingerprints can be processed in the same manner as visible ones (Färber, Seul, Weisser, & Bohnert, 2010; Trapecar & Balazic, 2007).

Chemical methods imply chemical reactions between chemical species and fingerprint residues, followed by the formation of steady complexes (Datta, Lee, Ramotowski, & Gaensslen, 2001; Milašinović & Koturević, 2016), and many are routinely employed on porous surfaces (ninhydrin, silver nitrate, iodine fuming), and non-porous surfaces (cyanoacrylate fuming, iodine fuming) (Wanga, Yang, Wanga, Shi, & Liu, 2009). Nevertheless, these methods have disadvantages commonly related to their toxicity and potential formation of complexes (thus disabling further examination of fingermarks). On the other hand, physical methods include physical interactions or binding of certain powders or dyes to some particular residues from the (latent) prints. Commonly used powder formulations are regular, metallic, luminescent and thermoplastic, while their choice depends on the surface and its characteristics: illumination, texture, color, porosity, etc. (Bumbrah, Sharma, & Jasuja, 2016; Datta, Lee, Ramotowski, & Gaensslen, 2001; Milašinović & Koturević, 2016).

By marking toxicity and detrimental effect on human health as the biggest drawbacks of current approaches, scientists are aiming at developing some novel systems (or even methods) that will overcome the mentioned problem and, additionally, satisfy cost-benefit demands. This paper deals with dextran-based biopolymer powders, obtained by the precipitating method, using potassium periodate (KIO_4) as an initiator, *N*,*N*'-methylenebisacrylamide (MBA) as a crosslinking agent and methanol as a precipitation solvent. Dextran is a complex (yet cheap and non-toxic water soluble), branched and hydrophilic polysaccharide composed of anhydroglucose rings, obtained from bacteria (particularly from *Lactobacillus*, *Leuconostoc* and *Streptococcus* species), widely used in medicine and pharmacy, as a component of drug-delivery (nanoparticle) systems, material that reduces blood viscosity and prevents the formation of blood clots, etc. (Wang, Dijkstra, & Karperien, 2016; Wasiak, et al., 2016). KIO_4 is an oxidizing agent and it was used to obtain aldehyde functionalities of dextran chains, in order to improve interactions with fingerprints trace residues (Maia, Carvalho, Coelho, Simões, & Gil, 2011). In this paper, four dextran-based biopolymer powder formulations were prepared, and their potential in developing latent fingerprints was tested.

MATERIALS AND METHODS

Materials

Dextran powder was purchased from Sigma-Aldrich (USA), KIO_4 from Merck (Germany), MBA from Acros Organics (USA) and methanol from Centrohem (Serbia). Distilled water was used for all buffer solutions preparation. Acetate buffers of various pHs were prepared by dissolving sodium acetate and acetic acid in distilled water, in order to obtain buffer solution of desired pH value. Buffer solutions were used to extract anthocyanins from *Brassica oleracea* (var. *capitata*, f. *rubra*) and afterwards, the obtained liquid anthocyanin extract was used to dissolve dextran powder, the initiator and the crosslinking agent. Besides *B. Oleracea* (var. *capitata*, f. *rubra*), all materials were used without further treatment or purification.

Preparation of Dextran-based Biopowders

Acetate buffer solution (pH ~ 3.52) (dissolving medium) was prepared by modifying experimental procedures described by Chandrasekhar et al (2012). This medium was used to extract anthocyanins from *B. Oleracea* (var. *capitata*, f. *rubra*). Briefly, 50 g of *B. Oleracea* (var. *capitata*, f. *rubra*), ground with blender *Bosch* (180W power, Germany), were added to 200 mL of acetate buffer solution, then mixed using a magnetic stirrer *Velp Scientifica* (~600 rpm) and heated (~30 min) until boiling of the mixture was achieved. After cooling, the obtained mixture containing anthocyanins' extract was filtered using a metal sieve and the filtrate was kept in a refrigerator at 4 °C until further use. The obtained anthocyanin mixture was used to obtain different color of desired powders, as well for better enhancement through complexing with fingerprint sweat and lipid residues, since it was demonstrated that anthocyanins have indicator chemical properties (i.e. color change in accordance with change in pH value) (Chandrasekhar, Madhusudhan, & Raghavarao, 2012).

Furthermore, four different formulations of dextran-based biopowders were prepared in order to determine their capability in visualizing latent fingermarks. Briefly, 2.0000 g of dextran powder was dissolved in 200 mL of prepared mixture containing anthocyanins' extract. Afterwards, the obtained mixture was divided into 4 equal parts (volume of each was 50 mL). The first mixture was left as blank; in the second mixture the initiator in ratio 10:1 (dextran:KIO_4) was added; in the third mixture the crosslinking agent MBA (8 w/w.% by mass of biopolymer) was added; in the fourth mixture both KIO_4 and MBA were added taking the same ratios as was already stated. The mixtures were stirred at low speed (~200 rpm) and at room temperature using a magnetic stirrer. After homogenization, methanol in 1:3 v/v ratio (mixture:methanol) was added, in order to precipitate polymer from the mixtures. When the precipitate was formed, the mixtures were filtered using a filter paper. After air-drying at room temperature for ~24h, dry precipitate was kept in the drying oven at 37 °C for additional few hours. Finally, the obtained dry formulations were ground with pestle and mortar to fine powders and kept in desiccator until further application.

CHARACTERIZATION OF THE PREPARED POWDER FORMULATIONS

FT-IR Analyses

The formulations of synthetized biopowders were recorded in dry and solid state, using the *Bomem MB* 100 FT-IR spectrophotometer. Powders in amount of 1.5 mg were mixed and ground with 75 mg

of potassium bromide and then compressed into pallets at pressure of 11 t for about a minute, using the *Graseby Specac* model: 15.011. The spectra were obtained in the wavenumber range between 4000 to 400 cm^{-1}, at 25 °C and at 4 cm^{-1} resolution spectra.

Optical microscopy

The obtained powder formulations were recorded with optical microscope Leica FS C Comparison Macroscope, equipped with The Leica IM Matrox Meteor II Driver Software Module. Powders were tested in dry state, with and without backlight. Prior to imaging under the microscope, latent fingerprints left on the microscope glass slides were developed using prepared powder formulations and pure dextran powder (control powder).

Development of latent fingerprints

In order to estimate the ability and performances of the prepared powders, three male donors deposited sebaceous and dry fingerprints onto a paper (porous), rubber (semi-porous) and glass (non-porous) surface using only a thumb of their right hand. Following the guidelines proposed by the International Fingerprint Research Group (IFRG), sebaceous (oily) and dry fingerprints were deposited on the mentioned surfaces using a technical scale, in order to simulate real manipulating procedures and determine the pressure on surfaces (force applied to accommodate 100-150 g, per fingerprint), and the prints were then left under laboratory (humid) conditions for a short period of time. That period allowed the traces to dry and reduce the residues, by the time the latent fingerprints were developed with synthetized powders and two control powder formulations, using BVDA Squirrel hair brush (BVDA, The Netherlands).

Optical microscopy was used in order to approximate the size and uniformity of prepared powders, as well to assess their performances in visualizing latent fingermarks on glass surface (on which the best results were obtained). Therefore, according to the already described procedure, sebaceous and dry fingerprints randomly deposited onto the glass microscopic slides (properly labeled), were left for a few minutes and then 4 prepared powder formulations and pure dextran powder (control powder) were used for their visualization. After the short period of time, fingerprints were halved with the thick slide barrier and 2 different powders were applied on the same fingerprint – synthetized powders were applied to the left and pure dextran powder was applied to the right barrier side, using BVDA Squirrel hair brush. Such type of visualization enabled direct comparison between applied powder formulations with the aim at evaluating their size range, uniformity and efficiency in developing latent fingerprints.

Additionally, the prints were left under dry (desiccator) and humid (laboratory) conditions (relative humidity around 60%) and afterwards, fingerprints were developed in different time intervals – after 24 hours, 7 days and 1 month. The exception is the first series with 60 fingerprints (explained above), developed immediately after deposition used as a pretest to determine the direction of subsequent experiments. That resulted in total of 204 fingerprints, with 72 fingerprints per storage condition (and 60 additional fingerprints used as a pretest), where 180 were enhanced from a glass,12 from a rubber and 12 from a paper surface.

The IFRG recommends the implementation of 4 phases in order to estimate new detection and visualization techniques and systems. By conducting the experimental procedures as stated above, Phase 1 (*Pilot Studies*) was completed, in order to determine the functionality of the prepared powders (International Fingerprint Research Group (IFRG), 2014).

RESULTS AND DISCUSSION

The synthetized powders were labeled as S(Dex/KIO_4/MBA), where "Dex" refers to dextran, "KIO_4" to the initiator and "MBA" to the crosslinking agent content used to prepare the desired powders. The obtained powder formulations were tested on paper, rubber and glass surface. The powders were applied at least on 20 different fingerprints on each of the surfaces, in order to determine the possibility and reproducibility of their application, i.e. to achieve the desired visualization results. The best results were obtained with the sebaceous fingerprints deposited onto the glass surface. On the other hand, when applied onto the white paper surface with latent prints, it was not possible to achieve the satisfying contrast between the fingermark and the background, due to the white powder color appearance. Additionally, the development of fingerprints on rubber surface was poor, since that surface contains many bulges and indentations, thus disabling the binding of the prepared powders to the fingerprint residues. Therefore, the prepared powders did not provide satisfactory results on paper and rubber surface and additional research in order to improve this system should be conducted.

Figure 1 shows sebaceous and dry fingerprints of one donor, developed with four prepared powder formulations and two control powders (BVDA Magnetic silver and pure dextran powder). The prints were then photographed under visible light with a 12 MP camera (aperture f/2.2, pixel size 1.22 μm) using black background surface in order to achieve adequate contrast. When observing dry fingermarks, it was evident that their visualization was poor and the satisfactory results were not obtained with none of the applied powders. It is already well known that dry prints pose a great challenge in forensic investigation of fingerprints (Lennard, 2007). On the other hand, the development of sebaceous fingermarks was far better, since all powders have developed full fingerprint images, while at the same time visualizing the basic pattern type (double loop), the papillary lines (without disruption in their flow) and some minutiae points, as well.

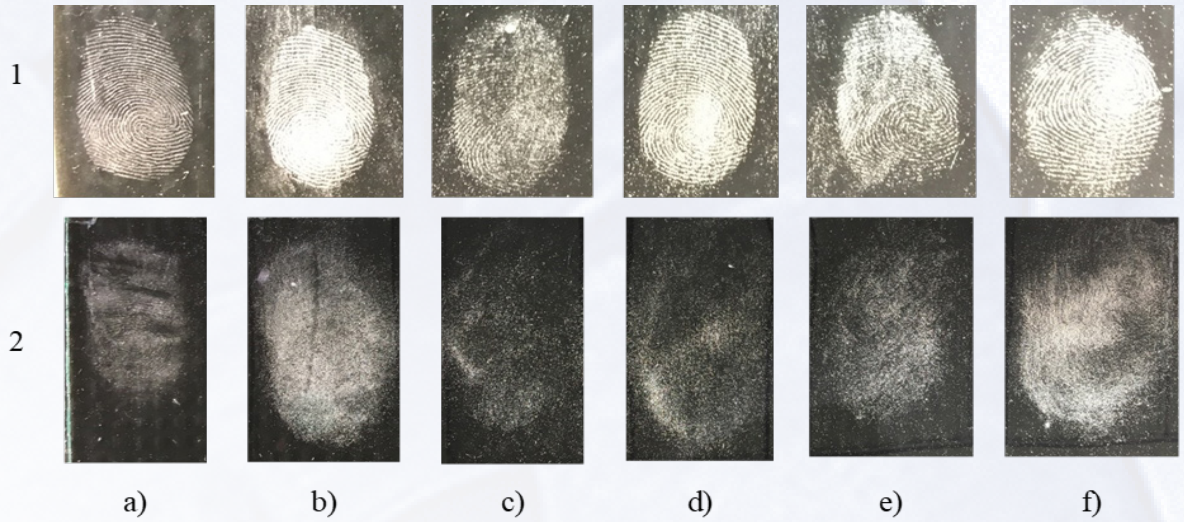

Figure 1. Sebaceous (1) and dry (2) fingerprints developed on glass surface, using the following powder formulations: a) BVDA Magnetic silver powder; b) Pure dextran powder; c) S(1/0/0); d) S(1/1/0); e) S(1/0/1) and f) S(1/1/1); and recorded under visible light using black background surface for adequate contrast.

When compared with control powders (Figure 1, 1-a) and 1-b)), the best results are obtained with formulation S(1/1/0) (Figure 1, 1-d)), while other tested formulations showed relatively satisfying results as well, which may be associated with diameter size of powders' particles. As confirmed by optical

microscopy, smaller particles better adhere and bind to sweat and lipid fingerprint residues, which was noticeable with synthetized powder formulations. On the contrary, pure dextran powder somewhat "over powdered" a fingerprint due to larger and non-uniform distribution of particles' size (Gürbüz, Özmen Monkul, İpeksaç, Gürtekin Seden, & Erol, 2015).

FT-IR Analyses

FT-IR analyses were performed in order to evaluate interactions between the components of prepared systems. Figure 2 shows the spectra of pure dextran and the prepared powder formulations S(1/0/0), S(1/1/0), S(1/0/1) and S(1/1/1). All spectra in Figure 2 contain some characteristic bands: 3385 cm^{-1} due to O–H stretching and 2360 cm^{-1} due to the stretching of C–H (Carp, et al., 2010; Mehta, Rucha, Bhatt, & Upadhyay, 2006; Mitić, Cakić, & Nikolić, 2010). The band at 1154 cm^{-1} can been assigned to stretching vibrations of the C–O–C bond and glycosides bridge, while band at 1017 cm^{-1} can be associated with stretching of C–O–H (Chiu, Hsiue, & Chen, 2004; Mehta, Rucha, Bhatt, & Upadhyay, 2006; Mitić, Cakić, & Nikolić, 2010). The weak band at 1110 cm^{-1} can be ascribed to the vibration of the C–O bond at the C4 position of the glucopyranose units (Mitić, Cakić, & Nikolić, 2010). Peaks at 905, 841, and 758 cm^{-1} can be assigned to α-glucopyranose ring deformation modes (Cakić, Nikolić, Ilić, & Stanković, 2005; Carp, et al., 2010). Additionally, weak shoulder peak at 1077 cm^{-1} may be due to complex vibrations involving the stretching of the C6–O6 bond with participation of deformational vibrations of the C4–C5 bond (Guerrero, Kerry, & de la Caba, 2014; Nikolić, Cakić, Mitić, & Ilić, 2008). However, according to Mitić, et al. (Mitić, Cakić, & Nikolić, 2010), peaks at 1041 and 1017 cm^{-1} present in all spectra are related to the crystalline and amorphous phases respectively, and can be responsible for more or less ordered structures of dextran chains. Finally, the band at 3830 cm^{-1}, slightly more intense in spectra 4 and 5 (Figure 2) when compared with others, can be related to N–H stretching frequency of the acrylamide (in MBA) (Fan, Zhang, & Feng, 2005).

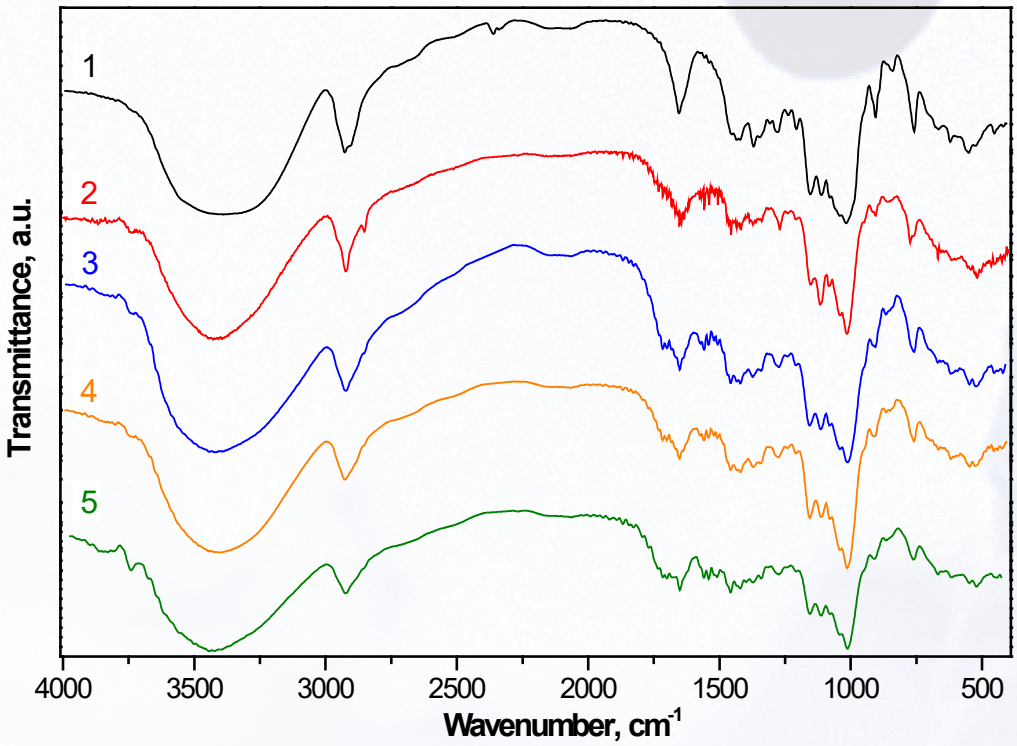

Figure 2. FT-IR spectra: 1) pure dextran; 2) S(1/0/0); 3) S(1/1/0); 4) S(1/0/1) and 5) S(1/1/1).

Optical microscopy

Figure 3 shows the images of prepared powders taken by Leica FS C Comparison Macroscope, equipped with the Leica IM Matrox Meteor II Driver Software Module, using magnification ×30 and dark-field contrast technique (backlighting). Since the best results were obtained on non-porous (glass) surface, the same surface was used for further analyses. The powder formulations were deposited onto microscopic glass slides in the form of a fine (thinner) and amassed (thicker) layer, in order to compare the uniformity and size of the particles.

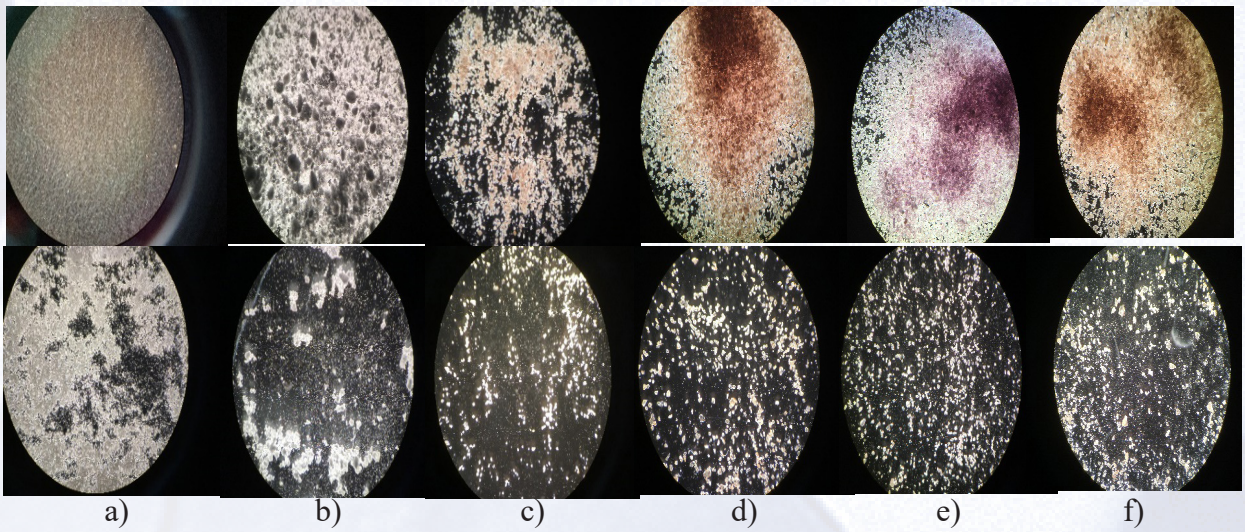

Figure 3. Microscopic images of prepared powders, deposited onto microscopic slides and recorded with optical microscope (magnification ×30, using dark-field contrast technique): a) BVDA Magnetic silver powder; b) Pure dextran powder; c) S(1/0/0); d) S(1/1/0); e) S(1/0/1) and f) S(1/1/1) Numbers 1 and 2 denote the images with powders in the form of amassed (thicker) and fine (thinner) layer respectively.

When comparing both thick and thin layers, it is evident that BVDA Magnetic silver powder (Figure 3, a) 1 and 2) contains more uniform and smaller particles than all other powder formulations. However, thicker layer of pure dextran powder (Figure 3, b) 1) possesses many irregular and non-uniform particles when compared to the prepared powder formulations (Figure 3, c)-f) 1), which can be related to "over powdering" of fingerprints when pure dextran powder is being applied. When observing thinner layers, these characteristics are even more obvious (Figure 3, b)-f) 2).

Subsequently, in order to confirm the previous presumptions, pure dextran and prepared powder formulations were used to develop latent fingerprints on glass surface. Therefore, sebaceous and dry fingerprints randomly deposited onto the labeled glass microscopic slides using technical scale were left for a few minutes and then 4 prepared powder formulations and pure dextran powder (control powder) were used for their visualization. After a short period of time, the fingerprints were halved with a thick slide barrier and 2 different powders were applied on the same fingerprint – synthetized powders were applied to the left and pure dextran powder was applied to the right barrier side, using BVDA Squirrel hair brush. Afterwards, the samples of enhanced fingerprints were recorded under the optical microscope (magnification ×15), using dark-field (Figure 4, 1) and bright-field (Figure 4, 2) contrast techniques. Three male donors have left fingerprints and their visualization and scanning were repeated several times (only sebaceous fingerprints were shown within the manuscript for practical reasons).

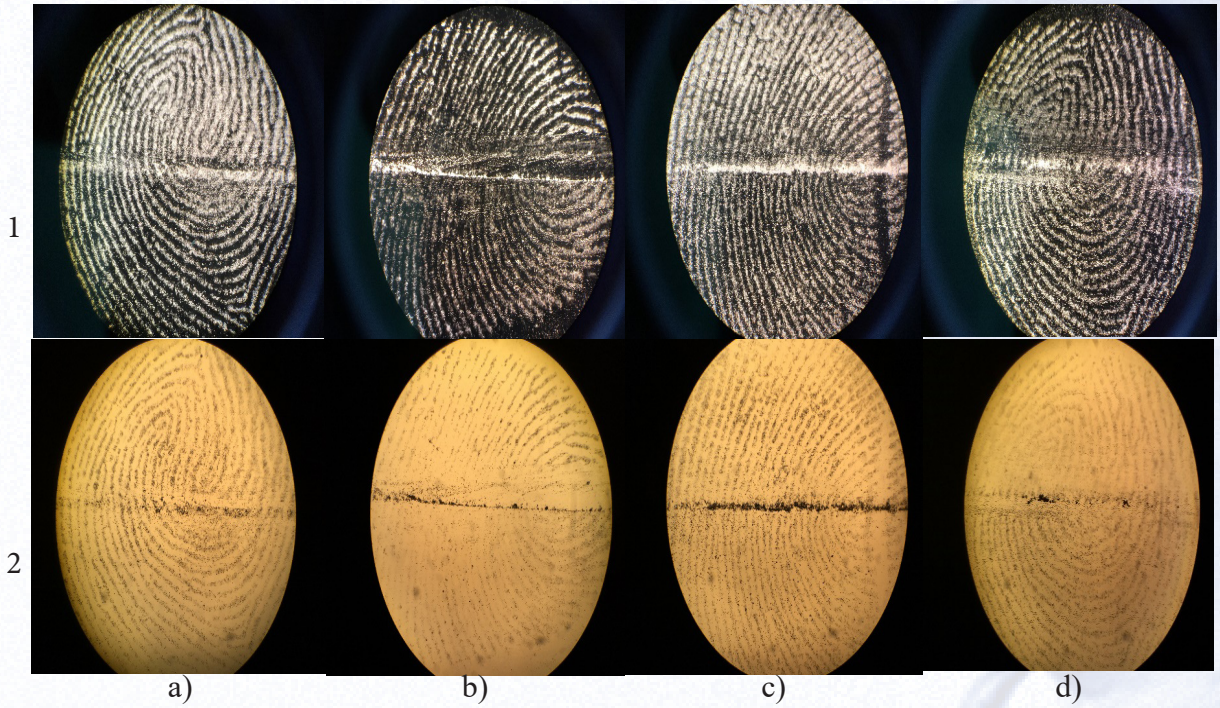

Figure 4. Sebaceous fingerprints deposited onto glass microscopic slides, left for a few minutes and developed using the prepared powders: a) S(1/0/0); b) S(1/1/0); c) S(1/0/1) and d) S(1/1/1) (left-half side of the images) and pure dextran powder as a control powder (right-half side of the images), recorded with optical microscope (magnification ×15), using: 1) dark-field and 2) bright-field contrast techniques.

When compared to the pure dextran powder (control powder), all prepared powder formulations showed better results in terms of developing latent fingermarks, which was reflected in visualizing the papillary lines with their continuous flow and making perceptible some minutiae as well. As we hypothesized previously, this may be associated with smaller diameter size of the prepared powders' particles, when compared to pure dextran powder, which enabled their better adhesion and binding to fingerprint sweat and lipid residues (Gürbüz, Özmen Monkul, İpeksaç, Gürtekin Seden, & Erol, 2015). Additionally, when applied with a brush, the prepared powdered formulations bonded with fingerprint residues and did not remain in the interpapillary space, when compared with the control powder. On the other hand, pure dextran powder has also developed fingerprints with persuasive results, but with noticeable "over powdering" of traces. Formulation S(1/0/0) (Figure 4, a), left-half side of the images) showed as good results as S(1/1/1) (Figure 4, d), left-half side of the images) and even better results than formulations S(1/1/0) and S(1/0/1) (Figure 4, b) and c), left-half side of the images), which was very promising, since that formulation contains only dissolved dextran powder precipitated with methanol, without initiator and/or crosslinker (KIO_4 and MBA show toxic, detrimental and irritating effect, while MBA is also potentially carcinogenic, as already explained and confirmed by Lent et al. (Lent, Crouse, & Eck, 2017; George, et al., 1998)). Therefore, very satisfying visualization of sebaceous fingerprints was achieved using glass surface as substrate, and with readily available, cheap and innoxious dextran-based powdered system.

However, it was evident that dry fingerprints could not be developed with applied powders, due to the lack of lipid and sweat residues deposition onto different substrates. Those prints must still be continuously and thoroughly investigated in order to overcome one of the main problems in forensic examination of fingerprints (Lennard, 2007).

CONCLUSIONS

In this paper, four different dextran-based powder formulations were obtained by simple precipitating method and were characterized in order to determine their potential application in development of latent fingerprints. Dextran was used due to its availability and low price, water solubility and non-toxic properties. The initiator and the crosslinking agent were used in order to obtain aldehyde functionalities of dextran chains and their crosslinking respectively, with the aim at enhancing the interactions with fingerprint residues. However, KIO_4 and MBA showed toxic and detrimental effect, which is unfavorable for the desired bio-based powder system. Based on the obtained results, formulation S(1/0/0) showed the best properties, with small and uniform particles, good binding to the fingerprint residues and their clear visualization, and the system is less harmful and satisfies the cost-benefit requirements. Additionally, all prepared powders showed better results in terms of developing latent fingermarks when compared to the pure dextran powder (control powder). However, the obtained results did not meet the expectations regarding color appearance, since the color of applied powders was not appropriate to visualize fingerprints on (white) paper surface and the hypothesized complexing of anthocyanins has not contributed to the enhancement of fingerprints on a rubber surface. On the other hand, as many commercial dusting formulations, these powders are easily handled and applicable, requiring no prior knowledge and the method itself is non-destructive, avoiding irreversible loss of traces. Finally, the additional research should include other (bio)polymers or addition of bio-based dyes and indicators, in order to expand the application of these systems on other surfaces and under different conditions, with the aim at supplementing some of the routinely employed physical methods in visualizing and enhancing latent fingerprints.

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