TECHNICAL NOTE

C. Anthony Pounds,¹ M. Phil.; Ronald Grigg,² Ph.D.; and Theeravat Mongkolaussavaratana,² Ph.D.

The Use of 1,8-Diazafluoren-9-one (DFO) for the Fluorescent Detection of Latent Fingerprints on Paper. A Preliminary Evaluation

REFERENCE: Pounds, C. A., Grigg, R., and Mongkolaussavaratana, T., "The Use of 1,8-Diazafluoren-9-one (DFO) for the Fluorescent Detection of Latent Fingerprints on Paper. A Preliminary Evaluation," *Journal of Forensic Sciences*, JFSCA, Vol 35, No. 1, Jan. 1990, pp. 169–175.

ABSTRACT: The use of a new amino acid reagent 1,8-diazafluoren-9-one (DFO), which produces a highly fluorescent species with latent fingerprints on paper, is described. Spectral characteristics of the fluorescent fingerprint show excitation (λ_{ex} approximately 470 nm) and emission (λ_{ex} approximately 570 nm) maxima in the visible part of the spectrum. Some printing inks fluoresce under these conditions and would therefore hinder fingerprint detection, but optical brighteners present in paper do not interfere. Fluorescent fingerprints visualized by DFO revealed more fingerprint detail than ninhydrin, the standard method for such surfaces.

KEYWORDS: questioned documents, 1,8-diazafluoren-9-one (DFO), fingerprints, papers

The visualization of latent fingerprints using high-intensity light sources has become widely accepted since 1977 when Dalrymple et al. [I] first reported the use of an argon ion laser to induce fluorescence of fingerprints deposited on paper. Creer [2] in particular has reported considerable success in visualizing fingerprints on a wide range of casework samples using this technique.

The use of high-intensity light sources is thus well established, and fingerprints can be visualized either by virtue of their natural fluorescence or when fluorescent fingerprints are obtained by derivatization or dye staining. Derivatization of fingerprint components, such as amino acids to produce a fluorescent species, is a particularly attractive technique as the increase in sensitivity can be from one to four orders of magnitude over nonfluorescent methods [3].

There have been several reagents reported that produce fluorescent species with the amino acid fraction of latent fingerprints. The main application has been for visualizing latent fingerprints on paper. Such reagents include dansyl chloride [4], fluorescamine [5], 4-chloro-7-nitrobenzofurazan (NBD-C1) and 7-nitrobenzofurazan ethers [6], o-phthal-

Received for publication 23 Feb. 1989; accepted for publication 31 March 1989.

¹Central Research and Support Establishment, Home Office Forensic Science Service, Aldermaston, Reading, Berkshire, United Kingdom.

²Professor and post-doctoral research fellow, respectively, Department of Organic Chemistry, The Queens University, Belfast, Northern Ireland.

170 JOURNAL OF FORENSIC SCIENCES

aldehyde [7], naphthalene-2,3-dicarboxaldehyde [8], the amino acid products of ninhydrin [9], and it analogs benzoninhydrin and 5-methoxyninhydrin when treated with zinc or cadmium salts [10].

All these reagents have disadvantages. The amino acid products with dansyl chloride, fluorescamine, and o-phthalaldehyde are all excited by ultraviolet (UV) light which also induces fluorescence of optical brighteners present in paper $\{11\}$. The product with naphthalene-2,3-dicarboxaldehyde, although excited at longer wavelengths, has only a short lifetime. A longer wavelength (465 nm) is also used to excite NBD-Cl, but this reagent reacts nonspecifically with water, optical brighteners, and some colored papers [12]. Benzoninhydrin, 5-methoxyninhydrin, and ninhydrin, the latter being the standard amino acid reagent for revealing latent fingerprints on paper, all produce colored fingerprints which, on treatment with zinc or cadmium salts, fluoresce. This process requires strict control of humidity, and maximum fluorescence is obtained at $-196^{\circ}C$ [13]. At this temperature background fluorescence may also increase creating detection problems.

This report describes a new amino acid reagent, 1,8-diazafluoren-9-one (DFO)³ (Fig. 1), which reacts with latent fingerprints to give a red-colored product which is highly fluorescent.

Experimental Procedure

Synthesis of DFO

The reagent was synthesized according to Druey and Schmidt [14]. (Although they prepared the compound in 1950, it was not realized till now that it produced a highly fluorescent product with amino acids.)

Reagent Formulations and Usage

DFO--DFO (50 mg) was dissolved in a mixture of 4 mL of methanol and 2 mL of acetic acid and then diluted to 100 mL with 1,1,2-trichlorotrifluoroethane (Fluorisol). The sample was dipped in the freshly prepared reagent for 5 s and, after allowing it to dry for 30 s, was dipped a second time. Fingerprints were then revealed by heating the paper at 100°C for 10 min.

Ninhydrin—The ninhydrin reagent used was the nonflammable formulation (NFN) of Goode and Morris [15]. After treatment, fingerprints were revealed by placing samples in a humidity chamber set at 80° C and 80° relative humidity (RH).

Ninhydrin/Zinc Complex—The zinc chloride solution used to treat ninhydrin-developed fingerprints was that reported by Goode and Morris [15]. Samples were placed in a humidity chamber set at 80°C and 80% RH until the change in fingerprint color was complete.

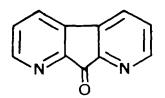


FIG. 1-Structure of 1,8-diazafluoren-9-one.

³British Patent Application 8826237.3.

Fingerprints

Latent fingerprints were deposited on several types of paper including reproduction paper, cardboard, checks, cards, buff envelopes, newspaper, and three types of writing paper. The latent prints were cut into two, one half being treated with DFO, the other half with ninhydrin followed by zinc treatment.

Visualization

The fluorescent fingerprints were visualized with:

1. A video spectral comparator (VSC-1, Foster & Freeman Limited, 25 Swan Lane, Evesham, Worcestershire, U.K.) using blue-green light excitation. Fluorescence was observed through a 610-nm filter.

2. A 12-W argon ion laser operated at 514 nm. Fluorescence was observed through 610-or 550-nm filters. When required, cooling to -196° C was achieved by immersion of the sample in liquid nitrogen.

3. A mercury vapor lamp filtered to give light of 546-nm wavelength (Model UV 202/11A, Ultrafine Technology, 16 Foster Rd., Chiswick, London).

Spectra

Glycine was selected to represent the amino acid fraction of latent fingerprints. Excitation and emission spectra of the glycine/DFO complex were determined using a UMSP80 fluorescence spectrometer to assess optimum conditions for fluorescence detection.

Results

Spectra for the reaction of DFO with glycine shows (Fig. 2) that the fluorescent species has a maximum excitation wavelength of approximately 470 nm (λ_{ex}) and a maximum emission wavelength of approximately 570 nm (λ_{ex}).

Figure 3*a* shows a latent fingerprint cut into two, one half having been developed with DFO and the other half with ninhydrin. The halves were observed under white light.

Figure 3b shows the same fingerprint excited with blue-green light on the VSC-1 (the ninhydrin-developed half being further treated with a zinc salt to render it fluorescent).

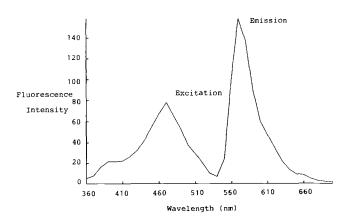


FIG. 2— Excitation and emission spectra of glycine on paper after treatment with 1,8-diazafluoren-9-one.

172 JOURNAL OF FORENSIC SCIENCES

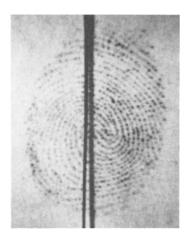


FIG. 3a—Fingerprint developed with (left) 1,8-diazafluoren-9-one and (right) ninhydrin. Photographed using normal procedures.



FIG. 3b—As for Fig. 3a, but ninhydrin half (right) treated with zinc. Photographed from VSC-1 using blue-green light and 610-nm filter at room temperature.

Figure 4a depicts a latent fingerprint cut into two halves, one half being treated with DFO and the other half with ninhydrin. Figure 4b shows the same fingerprint illuminated using a 12-W argon ion laser and Fig. 4c when illuminated with light from a mercury vapor lamp. Figure 4d shows both halves illuminated with a 12-W argon ion laser. The ninhydrin-developed half has been rendered fluorescent by zinc treatment and cooled to -196° C with liquid nitrogen.

Discussion

The results show that latent fingerprints developed by DFO using normal white light illumination do not appear to be developed quite as well as those treated with ninhydrin, the standard method for such surfaces. This may be due to the contrast of the red fingerprint being less than that for the purple fingerprints developed with ninhydrin.

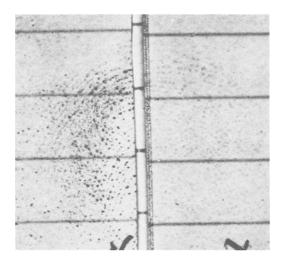


FIG. 4a—Fingerprint developed with (left) ninhydrin and (right) 1,8-diazafluoren-9-one. Photographed using normal procedures.

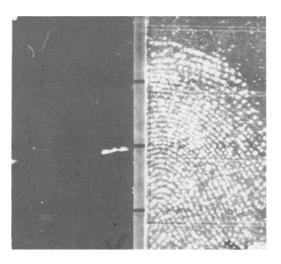


FIG. 4b—Same as for Fig. 4a but illuminated using 514-nm wavelength light from 12-W argon ion laser and photographed through a 610-nm filter at room temperature.

When fingerprints treated with DFO are illuminated using a suitable light source and viewed through an appropriate filter, considerable enhancement is achieved compared to ninhydrin/zinc-developed fingerprints even when the latter are cooled to -196° C with liquid nitrogen.

DFO is highly successful in producing fluorescent latent fingerprints, but there is a problem with interference from colored printing inks that fluoresce; this was particularly noticeable for checks and paper currency. However, a brief test on a paper sample before treatment will indicate if this is likely to cause difficulties. Illumination with light of longer wavelengths may overcome this problem to some extent. Accordingly, when fingerprints treated with DFO were excited with light of wavelength 546 nm from a mercury vapor

174 JOURNAL OF FORENSIC SCIENCES

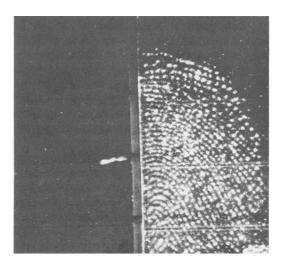


FIG. 4c---Same as for Fig. 4b, but illuminated at 547-nm wavelength from a mercury vapor lamp.

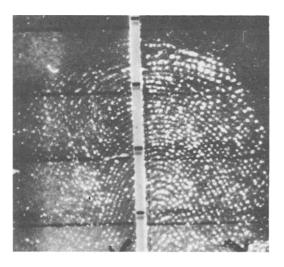


FIG. 4d—Same as for Fig. 4b, but ninhydrin half (left) treated with zinc. Photographed through a 550-nm filter at $-196^{\circ}C$.

lamp (compared to the 514-nm argon ion laser line or blue-green light of 400 to 600 nm), interference from some printing inks was found to be reduced. For the future, analogs of DFO will be synthesized in an attempt to effect a shift in excitation to longer wave-lengths. The greater sensitivity of DFO compared with ninhydrin means that the reagent will also have other applications involving the detection of amino acids, for example, using thin-layer chromatography and gas chromatography.

Conclusions

The new amino acid reagent, 1,8-diazafluoren-9-one (DFO), produces highly fluorescent fingerprints when illuminated with an appropriate light source. Sensitivity was found to be greater than the ninhydrin methods for revealing latent fingerprints. The reagent is easy to prepare and use, and there is no need to cool to -196° C for maximum fluorescence. Its use on colored printing that fluoresces is a limiting factor.

The toxic and carcinogenic properties of the reagent have yet to be determined.

Acknowledgments

The authors express their gratitude to Miss A. Thurston for helping with the experimental work, to Carl Zeiss (Oberkochen Limited, West Germany) for producing the emission and excitation spectra of glycine/DFO, and to Mr. K. C. Creer of the Metropolitan Police Forensic Science Laboratory for photographing fingerprints illuminated with a 12-W argon ion laser.

References

- [1] Dalrymple, B. E., Duff, J. M., and Menzel, E. R., "Inherent Fingerprint Fluorescence— Detection by Laser," *Journal of Forensic Sciences*, Vol. 22, No. 1, Jan. 1977, pp. 106–115.
- [2] Creer, K. C., "Operational Experience in the Detection and Photography of Latent Fingerprints by Argon Ion Laser," *Forensic Science International*, Vol. 23, 1983, pp. 149–160.
- [3] Seitz, W., "Fluorescence Derivatisation," Critical Reviews of Analytical Chemistry, Vol. 9, 1980, pp. 367-405.
- [4] Burt, J. A. and Menzel, E. R., "Laser Detection of Latent Fingerprints. Difficult Surfaces," Journal of Forensic Sciences, Vol. 13, No. 2, April 1985, pp. 364–370.
- [5] Menzel, E. R., Fingerprint Detection with Lasers, Marcel Dekker, Inc., New York, 1980, pp. 65-66.
- [6] Almog, J., Zeichner, A., Shifrina, S., and Scharf, G., "Nitro-Benzofurazanyl Ethers—A New Series of Fluorigenic Fingerprint Reagents," *Journal of Forensic Sciences*, Vol. 32, No. 3, May 1987, pp. 585–596.
- [7] Mayer, S. W., Meilleur, C. P., and Jones P. F., "The Use of Ortho-Phthalaldehyde for Superior Visualisation of Latent Fingerprints," *Journal of the Forensic Science Society*, Vol. 18, Nos. 3 and 4, July/Oct. 1978, pp. 233–235.
- [8] Almog, J., Rozen, S., and Scharf, G., "Reaction of Naphthalene-2,3-Dicarboxaldehyde with Amino Acids and with Latent Fingerprints on Paper," Israel Police Research and Development Report DM/2037, Sept. 1981.
- [9] Herod, D. W. and Menzel, E. R., "Laser Detection of Latent Fingerprints: Ninhydrin Followed by Zinc Chloride," *Journal of Forensic Sciences*, Vol. 27, No. 3, July 1982, pp. 513–518.
- [10] Lennard, C. J., Margot, P. A., Stoilovic, M., and Warrener, R. N., "Synthesis of Ninhydrin Analogues and Their Application to Fingerprint Development: Preliminary Results," *Journal* of the Forensic Science Society, Vol. 26, No. 5, Sept./Oct. 1986, pp. 323–328.
- [11] Zahradnik, M., The Production and Applications of Fluorescent Brightening Agents, Wiley and Sons, Chichester, 1982.
- [12] Warrener, R. N., Kobus, H. J., and Stoilovic, M., "An Evaluation of the Reagent NBD-C1 for the Production of Luminescent Fingerprints on Paper: 1. Support for a Xenon Arc Lamp Being a Cheaper and Valuable Alternative to an Argon Ion Laser as an Excitation Source," *Forensic Science International*, Vol. 23, 1983, pp. 179–188.
- [13] Stoilovic, M., Kobus, H. J., Margot, P. A. J.-L., and Warrener, R. N., "Improved Enhancement of Ninhydrin Developed Fingerprints Using Low Temperature Photoluminescence Techniques," *Journal of Forensic Sciences*, Vol. 31, No. 2, April 1986, pp. 432–445.
- [14] Druey, J. and Schmidt, P., "Phenanthrolinchinone und Diazafluorene," *Helvetica Chimica Act*, Vol. 50, 1950, pp. 1080–1087.
- [15] Goode, G. C. and Morris, J. R., "Latent Fingerprints: A Review of Their Origin, Composition and Methods for Detection," AWRE Report 022/83, AWRE, MOD(PE) Aldermaston, Berkshire, UK.

Address requests for reprints or additional information to C. A. Pounds Central Research and Support Establishment Home Office Forensic Science Service Aldermaston, Reading, Berkshire RG7 4PN United Kingdom