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## Note

**4,4'-Bis(dimethylamino)diphenylcarbinol, a sensitive spray reagent for the detection of thiols and carboxylic acids**

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In the presence of acid 4,4'-bis(dimethylamino)diphenylcarbinol (BDC-OH) gives rise to an intense blue "carbonium-immonium" ion<sup>1</sup>, the colour of which is bleached on addition of thiols<sup>2</sup>. This forms the basis of a sensitive method for the quantitative spectrophotometric determination of thiols<sup>2</sup>.

We wish to report that an acidic solution of the carbinol can be used as a convenient and sensitive spray reagent for the detection of thiols on thin-layer and paper chromatograms. The spray reagent consists of BDC-OH, obtained from N.B.C. (Cleveland, Ohio, U.S.A.) and used without further purification, in acetone containing aqueous acetic acid. Thiol compounds are revealed on silica gel G and GF<sub>254</sub> chromatoplates by the almost immediate appearance of white spots on an intense bright blue background; on cellulose plates or paper chromatograms the

TABLE I

SENSITIVITY OF VARIOUS COMPOUNDS FOLLOWING APPLICATION OF 4,4'-BIS-(DIMETHYLAMINO)DIPHENYLCARBINOL SPRAY REAGENTS

<i>Spray solution</i>	<i>Compound*</i>	<i>Amount (μg)</i>	<i>Sensitivity** (μl of solution of the stated concentration)</i>
BDC-OH in acetone (0.1%)— aqueous acetic acid (5%)— water (1:1:6)	Cysteamine hydrochloride	0.1	0.5 (1.76 mM)
	Cysteine (free base)	0.1	0.5 (1.65 mM)
	Ac-Gly <sub>2</sub> -(Cys-Gly <sub>2</sub> -NH <sub>2</sub> )	0.2	1.0 (0.24 mM)***
	Ac-Gly <sub>2</sub> -(Cys-Gly <sub>2</sub> ) <sub>2</sub> -NH <sub>2</sub>	1.0	1.0 (0.82 mM)***
	Acetyl hydrazide	1.0	0.5 (26.9 mM)
	Arginine	10	5.0 (11.4 mM)
	Histidine	10	5.0 (9.5 mM)
BDC-OH in acetone (0.1%)	Glutamic acid	1.0	0.5 (13.5 mM)
	Ac-Gly <sub>2</sub> -OH	1.0	0.5 (11.4 mM)
	Z-Cys(Bzl)-Gly <sub>3</sub> -OH	4.0	2.0 (3.8 mM)***

\* Abbreviations recommended by the IUPAC-IUB.

\*\* Smallest amount easily detectable determined by serial dilution using silica gel G chromatoplates (spot diameter, 2-4 mm). The sensitivity is greater using cellulose plates or chromatography paper.

\*\*\* In ethanol.

background is pale blue. The reagent is specific for thiols and does not react with disulphides and S-protected cysteine derivatives. Nucleophiles such as amines, hydrazides, and imidazoles react with the reagent, albeit more slowly, giving rise to pale blue or white spots. The lack of sensitivity of the reagent to these nucleophiles\*, compared with its high sensitivity to thiols, enables it to be used for the specific detection of thiols. The sensitivity of the reagent to various thiols is summarised in Table I. It is of comparable sensitivity to the Ellman reagent<sup>3</sup> [5,5'-dithiobis(2-nitrobenzoic acid)] and the reagent of Grassetti and Murray<sup>4</sup> [2,2'-dithiobis(5-nitropyridine)], that are capable of detecting 0.2  $\mu$ g of cysteine, and superior to the nitroprusside reagent<sup>5</sup>, which often gives transient spots.

Conversely, a neutral solution of the reagent in acetone can be used for the detection of carboxylic acids, which are revealed as bright blue spots on a pale blue background. The sensitivity of this form of the reagent is superior to that of the common "indicator" methods for the detection of carboxylic acids<sup>6,7</sup> (see Table I).

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\* The reagent is sensitive to acetyl hydrazide (see Table I) but the white spot appears slowly.