Table 1. Effect of oximes on AChE activity in DFP-treated mice.

Group	AChE activity*			Protection index
	Blood	Cerebral cortex	Corpus striatum	
1. Control	2.86 ± 0.40	5.96 ± 0.42	20.45 ± 0.88	—
2. DFP	0.47 ± 0.10[a]	2.01 ± 0.28[a]	4.85 ± 0.48[a]	—
(% inhibition)	84.0	66.0	76.0	—
3. DFP + atropine + 2-PAM	2.60 ± 0.24[b]	2.08 ± 0.26	4.76 ± 0.41	4.40
(% Reactivation)	89.0	1.80	00	
4. DFP + atropine + DEV-9	2.39 = 0.20[b]	2.48 ± 0.32[c]	4.80 ± 0.50	3.50
(% Reactivation)	80.0	12.0	00	
5. DFP + atropine + DEV-10	2.30 ± 0.22[b]	2.40 ± 0.30[c]	4.99 ± 0.56	4.50
(% Reactivation)	77.0	10.0	00	

Each value is mean ± SE of six animals.
*Activity expressed as μmoles acetylthiocholine hydrolysed per min per g tissue or ml blood.
[a]$P < 0.001$, vs group 1; [b]$P < 0.001$, vs group 2; [c]$P < 0.05$, vs group 3.

quaternary salts, these compounds do not readily penetrate into the central nervous system, causing poor reactivation in cerebral tissues[9]. Our results indicate that DEV-9 and DEV-10 partially crossed the blood–brain barrier and reactivated cortical AChE. Since, structurally the blood–brain barrier is different from area to area of the brain[10], reactivation of striatal (deeper area) AChE was not observed (Table 1). Protection studies indicated that both DEV-9 and DEV-10 gave appreciable protection, like 2-PAM, in DFP poisoning. We conclude that the protection afforded by the oximes was mainly due to peripheral reactivation of AChE. The efficacy of 2-PAM was more than that of the new oximes.

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Immobilization of α-amylase complex in detection of higher oligosaccharides on paper

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The method of identification of high-molecular-weight oligosaccharides on paper is limited in its sensitivity by the silver nitrate test. A method described here generates more reducing power of higher sugars *in situ*, facilitating easy identification of the spots. Chromatograms dipped in 0.1% of Sanzyme solution in 80% acetone adsorb α-amylase. After removal of acetone, enzyme was allowed to react on paper for 6 h in a moist chamber. This degrades high-molecular-weight sugars, generating higher reducing power. Consequently, they are easily detected on the paper by the silver nitrate test. It is a sensitive method of identification of higher oligosaccharides which could be routinely used in the study of natural products.

PAPER chromatography is a rapid, sensitive and inexpensive procedure in detection of low-molecular-weight biological molecules, especially carbohydrates. In conventional methods of separation of sugars, a sample mixture is applied at the origin of chromatographic paper, developed in a suitable solvent, and the spots are detected by silver nitrate test[1]. The reducing sugars appear as distinct spots, and the intensity of spots are dependent upon the amount of sample and reducing power of sugars. However, this procedure is limited to a few low-molecular-weight sugars. The high-molecular-weight compounds escape unnoticed as they possess less reducing power, although they contain considerable amounts of carbohydrate. In this communication, a procedure is described for identification of high-molecular-weight sugars on paper using an α-amylase complex. After separation of sugars, the reducing power of high-molecular-weight compounds is increased by adsorbing α-amylase complex on the paper and allowing the enzyme to act till the separated spots undergo further degradation *in situ*, generating more reducing power.

α-Amylase complex is commercially available in a powder form as Sanzyme, Uni-Sankyo Limited, Hyderabad. It is a crude fungal enzyme prepared by ethanol extraction of *Aspergillus oryzae*. It is reported to contain essentially α-amylase activity associated with another saccharogenic enzyme Taka-amylase B which converts starch into glucose.

Incomplete starch hydrolysates were prepared by reacting Sanzyme with 2% soluble starch solutions. One gram of starch was dispersed and gelatinized in 40 ml of distilled water. To it 40 mg of Sanzyme

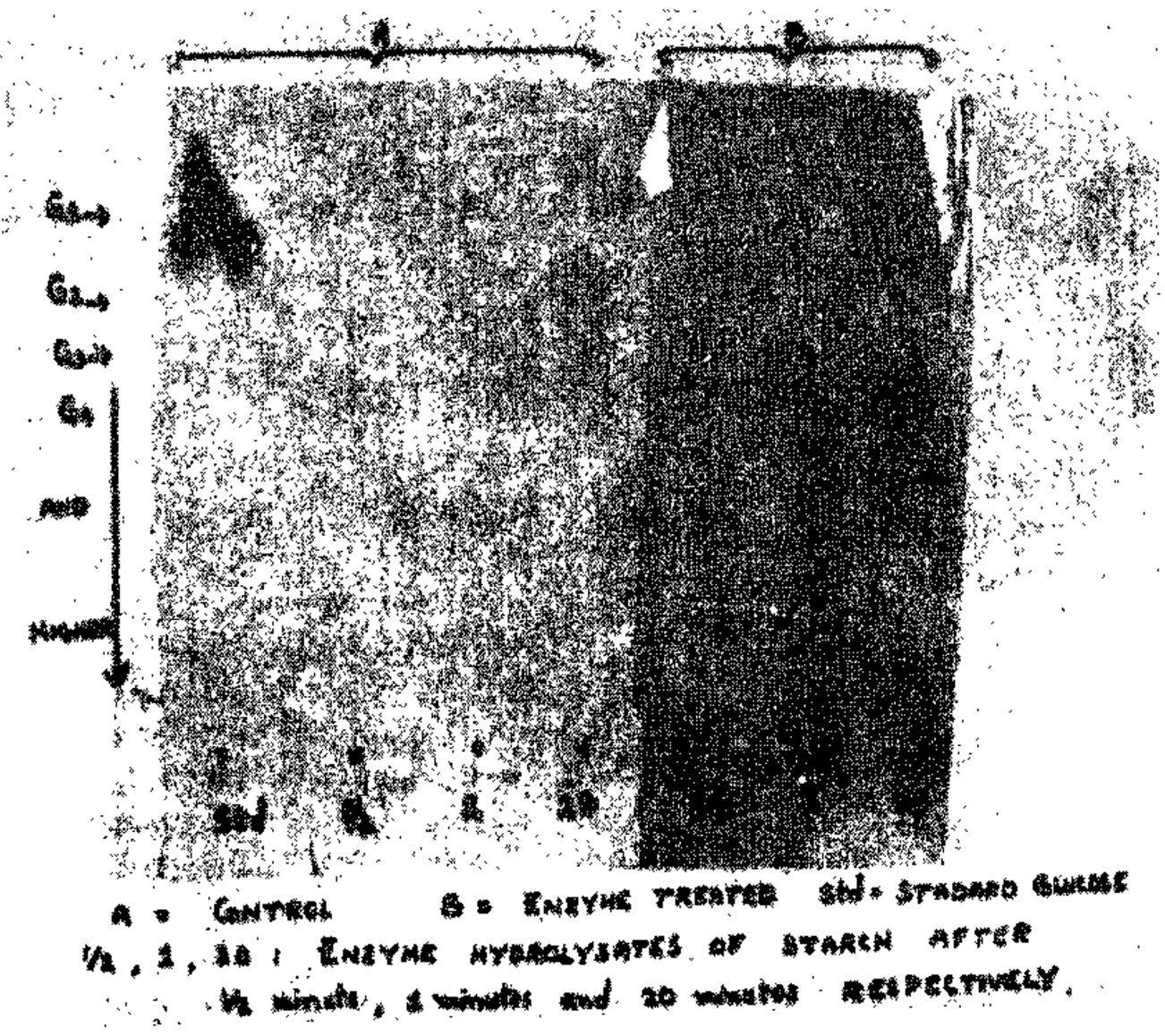

Figure 1. Detection of higher oligosaccharides on paper using amylase adsorption.

dissolved in 10 ml of distilled water was added and the reaction was carried out at room temperature in a total volume of 50 ml. Samples were withdrawn at various times for chromatography.

A chromatographic paper of about 21 cm × 23 cm was divided into two sections 'A' and 'B' (Figure 1). Section 'A' was used for control and section B for treatment with sanzyme. Samples of sugars were applied on a baseline 2.5 cm above the edge of the paper.

In both sections 'A' and 'B', about 10 µl each of 2% soluble starch and enzyme hydrolysates at 1 and 20 min were spotted on the paper. Standard glucose was also spotted as a reference compound. Ascending chromatography was carried out using 95% ethanol as a developing solvent. The chromatogram was dried and subjected to a second ascending chromatography to ensure finer resolution of higher oligosaccharides. The chromatogram was then dried and cut into 'A' and 'B' strips. Strip 'B' was briskly dipped in 0.1% Sanzyme solution dispersed in 80% acetone. (One hundred mg of Sanzyme was dissolved in 20 ml distilled water and mixed with 80 ml of distilled cold acetone.) The enzyme-treated paper was placed at room temperature for about 5 min to drive away the acetone vapours and then incubated in a moist chamber for about 6 h to allow the enzyme to act on the separated sugars. Finally, the high-molecular-weight sugars were identified by silver nitrate test. Control strip 'A' was also subjected to silver nitrate test to identify the reducing sugars.

Chromatogram 'B' shows the presence of higher oligosaccharides in both 1 and 20 min hydrolysates (Figure 1). These were not spotted in control 'A'. Chromatogram 'B' is slightly darker than 'A'. However, with slight purification of the enzyme, Sanzyme could be effectively used in identification of higher oligosaccharides on paper.

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Correction

Purification and some properties of a virus causing chlorotic mottle disease in carnations (*Dianthus caryophyllus* L.) S. K. Raj, M. Aslam and B. P. Singh, *Curr. Sci.*, 1990, **59**, 997.

Read 'potyvirus' for 'polyvirus' in lines 2, 6, 7 and 10 of last paragraph of paper (pages 998 and 999).