its basal Rudraprayag Formation in the Alaknanda Valley show the original reflected Aravalli trend (NE-SW) and the NW-SE oriented poorly developed linear and planar structures are superimposed on the NE-SW trend. This is also supported by the isoclinally folded quartz veins on the S_0 surface of the massive penecontemporaneous lava beds and limestone with a plunge perpendicular to the NE-SW (or parallel to NW-SE) and the S_2 structures are parallel to the typical Himalayan trend (NW-SE) which is S_1 in the Naini Group. This indicates that the Himalayan trend has produced a cross foliation or foliation superimposed on the S_0 (NE-SW) of the Garhwal Group rocks in the Tertiary tectonic movements. The stromatolites and other primary sedimentary structures support that the rocks are in normal disposition in this area.

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SEPARATION OF AUXIN PROTECTORS FROM *ZIZYPHUS* GALL TISSUE BY SEPHADEX GEL FILTRATION

AUXIN protectors are considered to be the regulatory substances in controlling the levels of endogenous IAA. They prevent peroxidase-catalyzed oxidation of IAA by inducing a lag period, rather than changing the rate of oxidation. Auxin protectors are found in juvenile tissue, in wounded tissue, and in crown-gall tumor tissue[1]. The relationship between habituation and the appearance of auxin protectors in cultured tobacco cells has been reported[2]. Recently Stonier *et al.*[3], have described the masking of peroxidase-catalyzed oxidation of IAA in *Vigna* by auxin protectors and a rapid method for removing most of the auxin protectors. In the present communication the separation of auxin protectors from a mite induced gall is reported.

Fresh tissues from *Zizyphus jujuba* Lamk. normal stem and stem galls incited by a mite, *Eriophyes cernuus* Massee, and tissue cultures obtained from the same were used in the experiment. The tissues were isolated[4] and grown on modified Murashige and Skoog[5] medium and used in their third week of growth. Tissue extracts were obtained by grinding 5 g each of the tissues mentioned above in 20 ml of cold 20 mM potassium phosphate buffer at pH 6·1. These were filtered through cheese cloth, and centrifuged for 15 min at 12,000 r.p.m. at 4° C. The supernatants were stored at −15° C. The separation of auxin protectors was carried out basically by the method of Yoneda and Stonier[6]. Dextran gels Sephadex G-200, G-50, and G-15 were used to filter the extracts. In chromatographic columns (15 × 300 mm), the Sephadex was layered up to 250 mm and flushed with phosphate buffer at a rate of 0·12 ml/min in the case of Sephadex G-200 and 0·33 ml/min in the case of Sephadex G-50 column. One ml of the tissue extract was loaded on the top of the column. The column filtrate was collected serally as 5 ml fractions. Dextran Blue 2000 and Pyronin G, with a molecular weight of about 2,000,000 and 300, respectively, were used as dye markers to delimit molecular weights. One ml of Dextran Blue was added to the extract as a 0·15% solution in buffer. Pyronin G was added only on a Sephadex G-50 column as one ml of a 0·005% solution as soon as the extract had enetered the gel.

As shown in Fig. 2, two peaks of protector (Pr) activity in terms of lag in IAA oxidation, in extracts of both *in vivo and in vitro* gall tissues, were obtained with Se-hadex G-200 filtrates. PR-I, the larger peak (around fraction 7), was light brown in colour and its molecular weight exceeded 200,000 daltons, since it migrated as rapidly as the high molecular weight marker—Dextran Blue that appeared in fractions 5 and 6. Colourless PR-II, the smaller peak (around fraction 18), on the other hand, migrated more slowly in Sephadex G-200. In Sephadex G-50 that Pr-II, the larger peak (around fraction 8), moved as rapidly as the high molecular weight marker—Dextran Blue (fractions 5 and 6) is indicative of having a molecular weight approximately 10,000 daltons. In Sephadex G-50 eluates, the smaller peak Pr-III moved slowly and appeared around fraction 12 followed by low molecular weight marker-Pyronin G which appeared in fractions 14, 15 and 16. On the other hand, colourless Pr-III moved very rapidly through Sephadex G-15 column, suggesting its molecular weight to be approximately 2,000 daltons. With

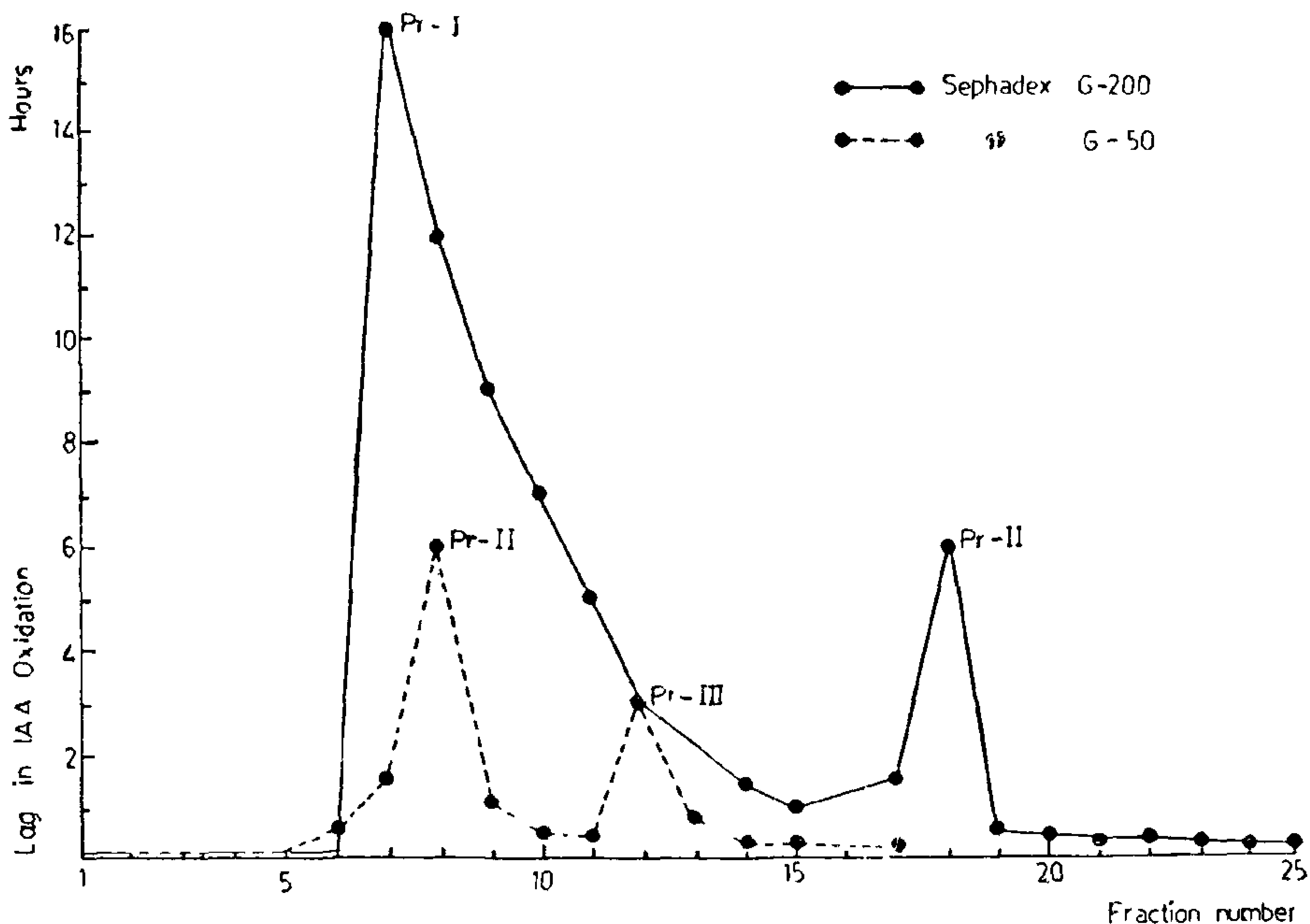

FIG. 1. Lag in IAA oxidation by Sephadex filtrates of gall tissue.

The protein content per gram fresh weight of the tissues were: *in vitro* normal (100·5 μg) and gall (162·6 μg); *in vivo* normal (1·94 mg) and gall (4·68 mg). The reaction mixture used to assay auxin protectors consisted of a mixture of 2,4-dichlorophenol, $MnCl_2$, and IAA each at a final concentration of 0·1 mM and horseradish peroxidase (HRP) at a final concentration of 0·25 μg/ml in 20 mM phosphate buffer, pH 6·1. To this reaction mixture 0·2 ml of Sephadex filtrate was added from each fraction before the addition of HRP. The final volume of the reaction mixture was made to 10 ml. Destruction of IAA was followed by removing 0·5 ml samples from the reaction mixture at various time intervals and assaying by mixing with 2 ml of Salkowski reagent. After one hour the absorbance of the mixture was measured at 530 nm.

Sephadex filtrates of normal tissue extracts, the oxidation of IAA proceeded in the normal way.

On the basis of the results presented, it can be concluded that *Zizyphus* gall tissues contained three substances or the polymers of the same substance which prevented IAA destruction by the enzymes normally found in the stem tissues. Preliminary studies suggest that the active site of auxin protectors is *o*-dihydroxyphenol. The presence of high levels of auxin protectors could lead to auxin autotrophy and hyperauxinity found in *Zizyphus* gall tissues.

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