polarized light. The hexagonal crystals dissolved first when the cells having both hexagonal and needle-like crystals were treated with 50 per cent alcohol or dilute acid. The needles were more stable. A remarkable feature was the presence of abundant inclusion bodies in some cells (Fig. 5), while others had none.

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SOIL SCIENCE

Reversal of Nitrate Inhibition of Nodulation by Indolyl-3-acetic Acid

NITRATE appears to have an influence on the action of indolyl-3-acetic acid (IAA) in the initiation and development of the symbiosis between legumes and bacteria of the genus *Rhizobium*. For example, it is often suggested that IAA is responsible for the curling of root hairs that is taken to be a prelude to the bacterial invasion of the legume root[1], and Thornton[2] noted that the percentage of lucerne root hairs that were deformed in the presence of *Rhizobium meliloti* culture filtrates was reduced if the plants were simultaneously supplied with $NaNO_3$. IAA is likewise proposed to be involved in the nodulation process itself[3], the nodule biogenesis likewise being repressed by nitrate. Recently, Tanner and Anderson[4] suggested that the inhibition may be attributable to a destruction of auxin by the nitrite produced from the bacterial reduction of nitrate. In this communication, it is demonstrated that not only may nitrate influence the IAA-induced changes but also that IAA may, conversely, mitigate the inhibition of nodulation by nitrate.

Lucerne seedlings inoculated with an effective strain of *R. meliloti* were grown aseptically in Hoagland's nitrogen-free nutrient solution[5] supplemented with various concentrations of KNO_3 and 0·8 per cent agar. Each treatment group consisted of 12–15 plants. The data of Table 1 not only confirm the repressive influence of nitrate, but also show an effect of nitrogen supply on nodule distribution. With increasing nitrate concentration, the absolute and relative abundance of nodules found on the surface of the rooting medium increased. The nitrate influence on the site of nodule genesis is particularly pronounced with young lucerne plants, and the percentage of surface nodules decreased with time as additional sub-surface nodules appeared. These observations support the proposal[4] that the nitrate effect in

Table 1. EFFECT OF NITRATE ON NODULATION OF LUCERNE

KNO_3-N (p.p.m.)	% Plants nodulated 10*	20*	30*	% Surface nodules 10*	20*	30*	Av. No. nodules/ plant
0	71·4	100	100	7·1	12·2	36·9	4·6
17	6·7	46·7	93·3	100	41·7	43·9	6·1
35	7·7	61·5	84·6	100	77·8	44·2	3·3
70	8·3	50·0	58·3	100	75·0	50·0	1·8
140	0	23·1	53·8	0	66·7	64·3	1·7

* Days after inoculation with *Rhizobium meliloti*.

suppressing nodulation results from a local and possibly an external change induced by the inorganic nitrogen. Further, the fact that excised bean roots become infected and develop nodules when nitrate is supplied to the base of the root, as described by Raggio, Raggio and Torrey[6], argues for an external, local effect of the anion.

The reversal of the inhibition by IAA is demonstrated by the data of Table 2. Each treatment was replicated four or five times. Nitrate in the rooting medium did not abolish nodule formation, but rather increased the time for the appearance of the first nodule and decreased nodule abundance. Whereas no enhancement in nodulation rate was noted in the presence of 10^{-8} M IAA, plants provided with both nitrate and 10^{-8} M IAA nodulated at about the same rate as nitrate-free lucerne. At this concentration, the auxin likewise reversed the nitrogen-induced inhibition of nodule frequency. A hundred-fold lower IAA level exerted only a partial reversal, whereas roots grew abnormally at 10^{-6} M concentrations. Although the percentage of plants that become nodulated and the abundance of the root structures varied from experiment to experiment, the same pattern of reversal was observed in each instance.

Table 2. REVERSAL OF NITRATE INHIBITION BY INDOLYL-3-ACETIC ACID

Treatment KNO_3-N (p.p.m.)	IAA (M)	% Plants nodulated 10*	20*	30*	Av. No. nodules/plan 30*
0	0	80	100	100	7·4
0	10^{-8}	100	100	100	8·0
14	0	20	80	100	6·6
140	0	20	20	100	2·2
140	10^{-10}	0	60	100	4·8
140	10^{-8}	100	100	100	5·4
140	10^{-6}	0	40	60	2·2

* Days after inoculation with *Rhizobium meliloti*.

Should IAA production be a necessary prelude to or an indispensable part of the bacterial invasion, then the effect of exogenous IAA in reversing the repression may result from a replenishment of the endogenous auxin destroyed by the nitrite generated in the reduction of the nitrate[4]. Such an explanation seems unlikely, however, in view of the vast excess of nitrate relative to auxin. Alternatively, the auxin may enhance plant growth and nitrogen assimilation so that the nitrate concentration falls below the level required to exert an inhibition. This hypothesis seems unlikely inasmuch as rooting media initially supplied with 140 p.p.m. nitrate-nitrogen and 10^{-7} M IAA still contained an average of 95 p.p.m. nitrate-nitrogen after 29 days of plant growth, a level distinctly deleterious to nodulation. Nevertheless, the possibility of microenvironmental interactions at the specific locus of infection cannot yet be excluded.

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