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*REVIEW*

# EFFECT OF COMBINED NITROGEN ON NODULE FUNCTION OF LEGUMINOUS CROPS WITH SPECIAL REFERENCE TO TROPICAL SOILS

*BY*

C. K. Jayasinghe

**ABSTRACT**

The effect of combined nitrogen on nitrogen fixation has been discussed by several investigators and from their findings it could be concluded that inorganic nitrogen may have either stimulatory, neutral or negative effect on nitrogen fixation depending on the type of legume and soil condition.

It was reported in some instances that low levels of combined nitrogen especially when applied as a "Starter" fertilizer at germination, stimulates plant growth and enhances nodule mass and nitrogen fixation. On the other hand when most of the legumes are grown in soil high in available nitrogen, the nitrogen fixation rate is severely affected. This phenomenon may become more critical when legumes are grown as cover crops where non legume counterpart is fertilized with nitrogenous fertilizer.

The degree of inhibition appears to vary with several factors viz. concentration of combined nitrogen, host species, cultivar, strain of root nodule bacteria, rate of inoculation, light intensity and temperature. However, the most critical factor is the concentration of combined nitrogen and it has been shown that very low concentrations such as 240 ppm N reduced the nitrogen fixation of bradyrhizobia - legume symbioses whereas 80–100 kg of N $ha^{-1}$ inhibited the nodulation and nitrogen fixation of *Vigna* spp. totally. It was further pointed out that extra caution should be taken when recommending nitrogenous fertilizer to mixed cropping systems in tropical soils as nodulation and nitrogen fixation of legume counter parts such as *Pueraria*, *Desmodium* & *Mimosa* were reduced to neglegible levels when 140 ppm N was added to Sri Lankan rubber soils.

**Key Words**

Combined nitrogen, nitrogenase activity, nodulation, tropical oils, nitrogen fixation.

Inorganic (or available) nitrogen, applied as fertilizer or present in the soil itself through mineralization and organic nitrogen are referred to as combined nitrogen. The effect of combined nitrogen on nitrogen fixation has been discussed by several investigators (Franco, 1977; Gibson, 1976; Norris and Date, 1976; Kanehiro *et al.*, 1983), and from their findings it could be concluded that there may be three possible effects of the inorganic nitrogen supply on nitrogen fixation.:

(*a*) positive effects (stimulatory effects of low levels of combined nitrogen as starter doses).

(*b*) neutral effects

(*c*) negative effects (especially with high concentrations)

These effects have been further reviewed by Kanehiro *et al.*, (1983) who proposed a schematic diagram to explain the effect of different concentrations of combined nitrogen on nitrogen fixation. This model has been reproduced in Figure 1 with some modifications.

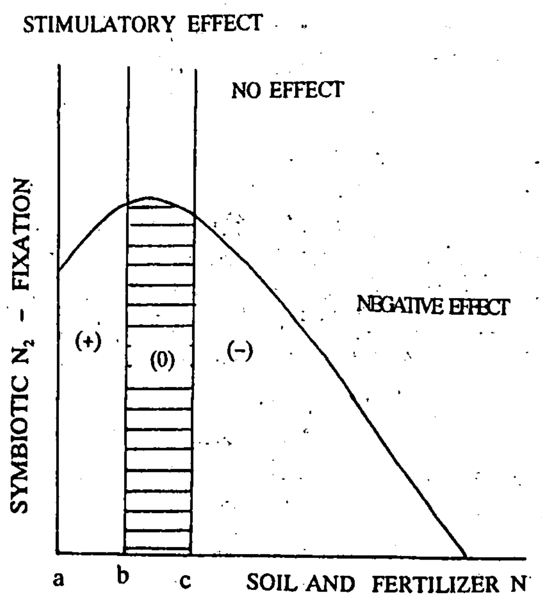


*Figure* 1: Schematic diagram showing the effect of combined nitrogen on symbiotically fixed nitrogen (after Kanehiro *et al.*, 1983).

## Stimulatory Effects

Low levels of combined nitrogen, especially when applied as a "starter" fertilizer at germination, frequently stimulates plant growth and enhances nodule mass and nitrogen fixation (Pate and Dart, 1961; Dart and Wildon, 1970; Dart *et al.*, 1976; Summerfield *et al.*, 1977; Agboola, 1978; Huxley, 1980; Minchin *et al.*, 1981; Eaglesham *et al.*, 1983).

For instance, Dart and Wildon (1970) found that the optimal level of nitrogen in various forms (ammonium nitrate, potassium nitrate, ammonium sulphate or urea) for nodulation, and nitrogen fixation in cowpea was in the range of 1 to 9 mg N $plant^{-1}$. With regard to *Vigna radiata*, total nitrogen in the plant was stimulated by addition of up to 30 kg N $ha^{-1}$ (Dart *et al.*, 1976). Eaglesham *et al.*, (1983) pointed out that nodulation and acetylene reduction activities could be stimulated significantly in cowpea by applying 36 to 72 mg N $plant^{-1}$. However, in most of these experiments existing levels of soil nitrogen were not stated; and hence it is difficult to asses the actual stimulatory concentrations of combined nitrogen.

Pate (1976) stated that many high yielding legumes possess a growth potential which connot be satisfied by even the most efficient type of symbiosis. Therefore, this might be the key factor which contributes to the stimulatory effects of combined nitrogen. On the other hand it is widely believed that application of "starter" nitrogen will satisfy the needs of the seedlings before commencing the nitrogen fixation (Kanehiro *et al.*, 1983). This results in a stronger plant with an extensive root system and more sites for root-nodule bacterial infections.

## Inhibitory effects

It is well established that when legumes are grown in soil high in available nitrogen, the nitrogen fixation rate is adversely affected. This has been demonstrated as far back as 1917 (Wilson, 1917 cited by Franco, 1977) and more recently by Munns (1968), Dart *et al.*, (1976), Eaglesham *et al.*, (1983), Miller *et al.*, (1982) and Graham and Scott (1984).

Inhibitory effects of high levels of combined nitrogen are significantly important in tropical soils as warm temperatures and moist conditions generally favour rapid nitrification if soil pH is favourable

(Kanehiro *et al.*, 1983). Some tropical soils with a high level of $NO_3$–N ranging from 18 to 400 ppm have a marked inhibitory effect on nodulation of legumes (Norris and Date, 1976).

The phenomenon of high background soil nitrogen may become more critical when legumes are grown in mixed or multiple cropping systems where non-legume counterparts are fertilized with nitrogenous fertilizers. For instance, approximately 100 kg of N $ha^{-1}$ $Y^{-1}$ is recommended in Sri Lankan rubber plantations during the immature period of tree growth (first 4 years) where legume cover crops are also grown extensively.

The decomposition of accumulated nitrogen-rich organic matter especially under pastures like *P. phaseoloides* can contribute significantly towards high soil nitrogen and high available nitrogen.

Two basic classes of inhibitory effects on nitrogen fixing legumes have been described, the first relating to the initiation and development of nodules and the second to the nitrogen fixation activity of already functioning nodules. Although several hypothesis have been postulated, the mechanism of inhibition in each case has not yet been adequately defined. For instance, it has been suggested that nitrates inhibit the initiation and development of the root-nodule bacterial infection thread inhibiting the production and curling of root hairs in the host (Munns, 1968, Kanehiro *et al.*, 1983). They have also suggested the possibility of changes in indole acetic acid around the root hair being involved and/or competing for the photosynthates within the host, in explaining the inhibitory effects of nitrates.

It has been pointed out that inhibition of *R. leguminosarum* bv *trifolii* infections on clover roots by nitrate nitrogen (15 mM $NO_3$–N) may be due to the effect of nitrates on Trifoliin A, a compound which is believed to be involved in the recognition of specific rhizobia by clover roots. Trifoliin A is thought to recognise similar saccharide residues on *R. leguminosarum* by *trifolii* and clover and to cross bridge them (Dazzo and Brill, 1978).

In pea roots it has been reported that nitrates affect the accumulation of pea lectin and it's receptors on roots, and this inhibits the infection process (Diaz *et al.*, 1981). However, changes in the chemistry of cell walls could have an important impact on the infection process as root-nodule bacteria must penetrate the cell wall to establish the infection.

The degree of inhibition appears to vary with several factors viz. concentration of combined nitrogen (Dart and Wildon, 1970; Miller *et al.*, 1982; Jayasinghe, 1986), form of nitrogen compound (Dart and Wildon, 1970; Eaglesham *et al.*, 1983) host species (Allos and Bartholomew, 1959; Eaglesham *et al.*, 1983; Danso and Eskew, 1984; Harper and Gibson,1984;) Gibson, 1984; Jayasinghe, 1983) cultivar (Gibson, 1974; Miller *et al.*,1982), strain of root-nodule bacteria (Pate and Dart, 1961; Franco, 1977) rate of inoculation (Herridge *et al.*, 1984), light intensity (Dart and Mercer, 1965) and temperature (Gibson, 1974).

However, the most critical factor is the concentration of combined nitrogen. For example, it has been shown with plants which were grown in agar, that 6 ppm nitrogen as nitrate (Norris and Date, 1976) and 15 mM nitrate (Dazzo and Truchet, 1984) inhibited the nodulation of clover and 20 mM nitrate inhibited the nodulation of pea (Diaz *et al.*, 1981). The nodulation of *Pueraria phaseoloides* was completely inhibited when they were grown in seedling agar incooperated with 44.8ppm N as $NH_4$ $NO_3$ (Jayasinghe, 1986).

Several investigations have been carried out on this aspect under glass house and field conditions and some findings relevant to bradyrhizobia are highlighted here. Dart and Wildon (1970) showed that nitrogen concentrations such as 240 ppm $pot^{-1}$ reduced the nitrogen fixation of *Bradyrhizobium* legume symbioses. Further it has been reported that there was no advantage in the application of nitrate in promoting nodule mass or acetylene reduction activity in three cultivars in cowpea in Texas (Miller *et al.*, 1982). With regard to *Vigna radiata* and *Vigna mungo*, nitrogen fixation of both crops was reduced with the addition of combined nitrogen, resulting in virtually no fixation at 80-100 kg of N $ha^{-1}$ as $NH_4NO_3$.

It is interesting to note that both nodulation and nitrogenase activity of *Pueraria phaseoloides*, a common cover crop of rubber plantations declined significantly ($P < 0.05$) beyond the addition of 70ppm N as $NH_4NO_3$ to rubber soils in Sri Lanka and completely inhibited at a concentration around 140 ppm N (Jayasinghe, 1986). Furthermore, Jayasinghe (1986) reported that nodulation and nitrogenase activity of legume cover crops, *Desmodium ovalifolium* and *Mimosa invisa* were also reduced to negligible levels when N was added to same soils at a concentration around 140 ppm. All these findings reaffirmed that it should be cautious when recommending nitrogen fertilizer mixtures to non-legume counter part in mixed cropping systems, if symbiotic nitrogen fixation to be utilized efficiently in tropical soils.

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