Seasonal patterns of growth and nitrogen fixation in field-grown pea

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Abstract
The seasonal patterns of growth and symbiotic \(N_2\) fixation under field conditions were studied by growth analysis and use of \(^{15}\)N-labelled fertilizer in a determinate pea cultivar (\(Pisum sativum\) L.) grown for harvest at the dry seed stage.

The patterns of fertilizer N-uptake were almost identical in pea and barley (the non-fixing reference crop), but more fertilizer-N was recovered in barley than in pea. The estimated rate of \(N_2\) fixation in pea gradually increased during the pre-flowering and flowering growth stages and reached a maximum of 10 kg N fixed per ha per day nine to ten weeks after seedling emergence. This was the time of early pod-development (flat pod growth stage) and also the time for maximum crop growth rate and maximum green leaf area index. A steep drop in \(N_2\) fixation rate occurred during the following week. This drop was simultaneous with lodging of the crop, pod-filling (round pod growth stage) and the initiation of mobilization of nitrogen from vegetative organs. The application of fertilizer-N inhibited the rate of \(N_2\) fixation only during that period of growth, when the main part of fertilizer-N was taken up and shortly after. Total accumulation of fixed nitrogen was estimated to be 244, 238 and 213 kg N ha\(^{-1}\) in pea supplied with nil, 25 or 50 kg NO\(_3\) -N ha\(^{-1}\), respectively. About one-fourth of total \(N_2\) fixation was carried out during preflowering, one fourth during the two weeks of flowering and the remainder during post-flowering. About 55% of the amount of N present in pods at maturity was estimated to be derived from mobilization of N from vegetative organs. “Starter” N (25 or 50 kg NO\(_3\) -N ha\(^{-1}\)) did not significantly influence either dry matter and nitrogen accumulation or the development of leaf area. Neither root length and root biomass determined 8 weeks after seedling emergence nor the yield of seed dry matter and nitrogen at maturity were influenced by fertilizer application.

Introduction

Symbiotic \(N_2\) fixation in field-grown legumes is influenced by many environmental factors (Sprent and Minchin, 1983). Combined N is known to inhibit \(N_2\) fixation when present at high levels in the soil (Jensen, 1986b, Oghoghorie and Pate, 1971). On the other hand, a certain level of combined N may be needed during early crop development in order to overcome N-deficiency during the period from when the cotyledon N-sources are exhausted until nodules are formed and capable of supplying the plant with symbiotically fixed nitrogen (Mahon and Child, 1979). However, it has been observed that temperate legumes, like pea, in contrast to tropical legume species, do not suffer from a period of nitrogen hunger following the exhaustion of cotyledons (Pate, 1958; Sprent and Minchin, 1983).

During later growth stages in pea and other legumes \(N_2\) fixation may be the only N-source for growth. However, studies using the “standard” acetylene reduction (AR) assay (incubation in closed vessels), have demonstrated that AR-activity reaches a maximum around flowering and is reduced during fruit/seed formation (Bethlenfalvay and Phillips, 1977; Bethlenfalvay et al., 1977; Dean and Clark, 1980; LaRue and Kurz, 1973; Lawrie and Wheeler, 1973; Young, 1982). It has
also been observed that more than 50% of the pea root nodules may be destroyed before flowering (Pate, 1958). This indicates that a high proportion of the N requirement of seeds is derived from mobilization of nitrogen from vegetative organs. Pate (1985) recently reported that in a determinate cultivar only about 35% of total pod nitrogen is assimilated post-flowering.

The "standard" AR-technique, which has been used for establishing profiles of N₂-(C₂H₂)-reduction during growth (e.g. LaRue and Kurz, 1973), cannot be used for quantification of N₂ fixation, because it is extremely difficult to get a sound estimate of the amount of nodule tissue in field-grown plants and because the ratio of C₂H₂-reduction to N₂ fixation often deviates from the theoretical ratio of three. Furthermore, it has recently been demonstrated that the "standard" AR-assay may be erroneous, due to the effect of acetylene on bacteroid respiration (Minchin et al., 1983). The ¹⁵N isotope dilution technique, even though it is not without problems, is the most reliable method for quantifying N₂ fixation. However, in most studies, when this technique has been used, only a single determination of N₂ fixation was made at maturity (Witty, 1983).

The aims of the present study were 1) to determine the course of N₂ fixation in field-grown pea, and 2) to evaluate the effects of "starter" N on growth and N₂ fixation in pea. Growth analysis and the ¹⁵N dilution technique with barley as reference crop were used.

Methods

Site

The experiment was carried out in 1984 on a loamy sand soil in a Rise experimental field. The field was cropped with spring barley in 1983 and 30 kg P and 50 kg K ha⁻¹ were applied before sowing in 1984. The soil contained 30 kg NO₃⁻ + NH₄⁺-N ha⁻¹ at sowing in the 0–25 cm layer; pH was 6.9 and the cation exchange capacity 15 meq 100 (g soil)⁻¹.

Experimental design

Field pea (Pisum sativum L.) 'Bodil', an early, white-flowered, dwarf and determinated cultivar was used. Nil, 25 or 50 kg NO₃-N ha⁻¹ was added at the time of seedling emergence. Spring barley (Hordeum vulgare L., cv. 'Nery') was grown as the non-fixing reference crop at the same N-levels as the pea crop. The experimental layout was nine randomized split-plot designs with crops as main plots and N-fertilizer levels as subplot and four replicates. Each subplot consisted of ten rows of length 4.40 m which were spaced 15 cm apart.

The crops were sown on 16 April with a ten-rowed Øyjord drill and seedling rates corresponded to 80 and 350 emerged pea and barley plants per m², respectively. On 30 April 88 ± 5 (± SE) pea seedlings m⁻² had emerged. Pea and barley seeds were treated with 'Thiram 80' and 'Baytan', respectively, before sowing.

The ¹⁵N-labelled N-fertilizer was applied at rates corresponding to 25 or 50 kg N ha⁻¹ two days after seedling emergence. The N-fertilizer consisted of a mixture of KNO₃ and Ca(NO₃)₂ with ¹⁵N enrichments of 0.97 to 3.83 atom % ¹⁵N excess. The labelled fertilizer was added to a microplot (6 rows of 1.2 m within the mainplot) as an aqueous solution distributed by a spray. Unlabelled Ca(NO₃)₂ was applied to the remaining part of the plot. The plots were sprayed with pesticides to control weeds and leaf eating weavills.

Sampling procedures

Nine harvests were taken during the growth season. From 40 cm of the central two rows in the ¹⁵N-microplot plants were cut 2 cm above ground and collected for ¹⁵N-analysis. Plants in the guard rows were discarded before the plants from the remaining area were collected and weighed. At the first four harvests only a combined above-ground biomass yield was determined. In harvest 5 to 8 plants were separated in vegetative organs and pods. At the last harvest, at physiological maturity in pea, pods were further separated in seeds and pod walls.

The leaf area was determined on plants from harvest 1 to 7. Three to five plants were randomly selected from each plot and the area of stipules and leaflets were measured by a Delta-T Device Area-meter. The leaf area indexes (LAI) of green and yellow leaves were calculated assuming a plant stand of 88 plants m⁻².

Root length of pea was determined at the full