The ability of several high arctic plant species to utilize nitrate nitrogen under field conditions

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Abstract. The ability to utilize NO₃⁻ in seven high arctic plant species from Truelove Lowland, Devon Island, Canada was investigated, using an in vivo assay of maximum potential nitrate reductase (NR) activity and applications of ¹⁵N. Plant species were selected on the basis of being characteristic of nutrient-poor and nutrient-rich habitats. In all species leaves were the dominant site of NR activity. Root NR activity was negligible in all species except Saxifraga cernua. NO₃⁻ availability per se did not appear to limit NR activity of the species typically found on nutrient-poor sites (Dryas integrifolia, Saxifraga oppositifolia, and Salix arctica), or in Cerastium alpinum, as leaf NR activities remained low, even after NO₃⁻ addition. ¹⁵NO₃⁻ uptake was limited in D. integrifolia and Salix arctica. However, the lack of field induction of NR activity in C. alpinum and Saxifraga oppositifolia was not due to restricted nitrate uptake, as ¹⁵NO₃⁻ labelled NO₃⁻ entered the roots and shoots of both species. Leaf NR activity rates were low in three of the species typical of nutrient-rich habitats (O. digyna, P. radicatum and Saxifraga cernua). Although two species (D. integrifolia and Salix arctica) showed little utilization of NO₃⁻, we concluded that five of the seven selected high arctic plant species (C. alpinum, O. digyna, P. radicatum and Saxifraga cernua and Saxifraga oppositifolia) do have the potential to utilize NO₃⁻ as a nitrogen source under field conditions, with the highest potential to utilize NO₃⁻ occurring in three of the species typically found on fertile habitats.

Key words: Arctic plants – Ammonium – Nitrate – Nitrate reductase – ¹⁵N

Materials and methods

Study site and plant material

The study was conducted at Truelove Lowland, Devon Island, Northwest Territories, Canada (75° 33' N, 84° 40' W) during July 1989 and July-August 1990. The habitats examined included a sparsely vegetated raised beach ridge that has low soil fertility (Walker and Peters 1977), and two fertile sites: a eutrophic region...
1–3 m below the camp outdoor toilet, and an area surrounding a lemming burrow on another raised beach ridge. Air temperatures on the beach ridges are usually below 7°C during the summer, with soil temperatures normally being much lower (Courtin and Labine 1977). The ecology of the lowland is comprehensively described in an International Biological Program study of the Truelove Lowland Tundra Biome (Bliss 1977).

Two groups of plant species were selected for study on the basis of being representative of habitats containing low or high levels of soil nutrients. Dryas integrifolia (M.), Saxifraga oppositifolia L., and Salix arctica Pall. were chosen as being characteristic of nutrient-poor, exposed, xeric environments (Porsild and Cody 1980). For species representative of fertile, mesic sites, we chose Cerastium alpinum L., Vahl, Oxyria digyna (L.) Hill, Papaver radicatum Rottb. and Saxifraga cernua L. (Porsild and Cody 1980). Despite differing in their characteristic habitats, all species could be sampled at the infertile Beach Ridge site. In addition, all the selected species could be sampled from the more fertile Camp Toilet site, with the exception of P. radicatum, which was sampled from the nutrient-rich Lemming Burrow site.

Soil sampling and analyses

As an index of NH4+ and NO3− available to the selected species, soil samples were collected and analyzed for exchangeable NH4+ and NO3−. Eight soil samples from 2–15 cm depth were collected from the crest of the Beach Ridge site at 1 m intervals. Soil was also collected around the upper 10 cm of roots in five species growing at the Beach Ridge site, from Saxifraga cernua growing at the Camp Toilet site, and from the roots of P. radicatum growing at the Lemming Hole site. The effect of NO3− application (see below) on pool sizes of NH4+ and NO3− in soils collected from close to the roots of three species growing at the Beach Ridge site (D. integrifolia, Salix arctica, and Saxifraga oppositifolia) was also assessed.

Samples were sieved through a 4 mm wire mesh, extracted for 1 h using a 1 M KCl solution, allowed to settle for 10 min, filtered through Whatman No. 42 paper, and the concentration of NH4+ and NO3− determined using an Orion model 95–10 NH4+ specific electrode (Orion Research Inc. Boston, USA; Myers and Paul 1968). Following the initial determination of NH4+ concentration in each sample, TiCl3 reductant (Orion Research Inc.) was added; converting all NO3− to NO2− and subsequently to NH3 (Orion Application Procedure Number 115). The concentration of NO3− was then calculated.

Plant sampling for NR activity measurements

When both root and leaf NR activity measurements were made in the NO3− application experiment, intact plants were excavated with the soil material retained. The plants were sealed in plastic bags, and transported to the field laboratory, where the soil was carefully removed by rinsing in distilled water. In earlier preliminary optimization studies, measurements of potential leaf NR activity were made using individual leaves sampled from non-excavated plants. In each case, leaves were detached from the plants using a new blade, and transported to the field laboratory for immediate assay.

Leaf and root in vivo NR activity measurements

NR activity was determined using an in vivo assay (Jaworski 1971 as modified by Havill et al. 1974), an assay that gave estimates of the maximum potential rate of NR activity, rather than the actual in situ rate of NO3− reduction. Fully expanded leaves and non-woody roots were sliced into 2 mm sections to facilitate greater penetration of incubation medium. Because of their small size, entire leaves were used from Saxifraga oppositifolia. Incubation of plant material in 5 mL of medium (100 mM phosphate buffer, pH 7.2) took place in the dark at 20°C, with 0.4 mL aliquots being removed after 24 and 54 min.

We determined the optimal concentrations of n-1-propanol and KNO3 in the incubation medium (Havill et al. 1974) in leaves of each species, and roots of P. radicatum and D. integrifolia. As a result, the following concentrations of propanol and KNO3 were used in subsequent assays of leaf NR activity: P. radicatum, 3% propanol, 10 mM KNO3; O. digyna, 2% prop., 200 mM KNO3; C. alpinum, 3% prop., 200 mM KNO3; Saxifraga cernua, 3% prop., 50 mM KNO3; D. integrifolia, 2% prop., 50 mM KNO3; Saxifraga oppositifolia, 2% prop., 10 mM KNO3; and Salix arctica, 2% prop., 20 mM KNO3. 2% propanol and 50 mM KNO3 were used in subsequent assays of root NR activity for all species.

Two minutes of vacuum infiltration were found to significantly increase NO3− production in the leaves of species that exhibited measurable NR activity and was, therefore, used in subsequent assays. NO3− concentration of each 24 and 54 minute aliquot was determined colorimetrically (Jaworski 1971). Production of NO2− was found to be linear between 24 and 54 min in all species. Checks for NO3− loss were periodically made by replacing the incubation medium's NO3− source (Havill et al. 1974) with 28 mM NaNO3, NO3− loss, due to NO2− reductase activity, typically, represented less than 2% of the total NO3− liberated into the incubation medium.

The plants chosen for the optimization and assay temperature experiments were sampled from the Camp Toilet site, or from around the Lemming Burrow site in the case of P. radicatum. Both sites had significant amounts of exchangeable NO3−. In this way, NO3− would have been naturally available to all species used in the optimization studies, increasing the potential for NR to be active.

NO3− applications

The effects of NO3− application on exchangeable soil NO3− and NH4+ concentrations, and on leaf and root NR activity, were assessed at the nutrient-poor Beach Ridge site. Individual plants at this site were sparsely distributed, often being separated from other plants by several metres. A 20 x 20 cm area surrounding each isolated plant was carefully watered with 40 mL of 0.24 M NaNO3 solution (20 g(N) m−2), and then immediately rinsed with 80 mL of distilled H2O. To allow the solutions to penetrate the soil below each plant, and to allow sufficient time for NO3− uptake and induction of NR, plants were sampled 10 days after initial fertilization. In each case, four treated and four non-treated plants of each species were measured. Subsequent analyses of extractable soil nitrogen demonstrated that the resultant pool sizes of NO3− at the level of the roots were within the range experienced at the nutrient-rich sites (e.g. Camp Toilet, see results section).

Labelled 15N uptake study

To assess the ability of the Arctic species to take up NO3− and NH4+, NaNO3 and NH4Cl solutions were labelled with 5% atom excess 15N (Cambridge Isotope Laboratories, USA) and spread at the rate of 20 g (N) m−2 around individual plants growing at the Beach Ridge site. To reduce surficial contamination of the fertilized plants, we rinsed each plant with 80 mL of distilled water immediately after the nutrient addition. At harvest, the excised roots and shoots were rinsed in distilled water. As it rained on several days before and during the 15N treatment experiment, 15N uptake would not have been affected by a lack of soil moisture. Small plant size necessitated pooling of the replicate samples for subsequent analysis. Non-treated plants were used as controls. Following excavation and rinsing, the plants were oven dried at 60°C for 2 days.

Chemical analyses of 15N-labelled tissue

Upon return to the University of Toronto, the samples were redried (70°C for 1 d) and ground. Total nitrogen of Kjeldahl destruction