Trends in Nitrate Reduction and Nitrogen Fractions in Young Corn (Zea mays L.) Plants During Heat and Moisture Stress

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Drought is a problem almost every year in many areas of the world. Drought reduces both quality and quantity of plant material produced and is a major factor in preventing stabilization of agriculture.

Nitrate is the principal form of nitrogen absorbed by higher plants. However, before it can be used in amino acid and protein synthesis, it must be reduced to ammonia. The first step in this reduction process is conversion of nitrate to nitrite by the nitrate reductase enzyme. With available nitrate nitrogen, reduced activity of the enzyme may result in nitrate accumulation. Reduced nitrate reductase activity has been attributed to decreased light, low moisture, low fertility, genotype differences, and possible other factors.

In some cases, nitrate has accumulated in sufficient quantities to be toxic to livestock. As early as 1895, Mayo (7) reported losses of cattle that had eaten corn fodder containing 25% potassium nitrate.

Molybdenum has been reported to be a metal constituent of the nitrate reductase enzyme. Thus, moisture stress may influence nitrate reduction by limiting molybdenum uptake or availability.

Decreased nitrate reduction in plants under stress may influence the quantity and/or quality of other nitrogen fractions and of total nitrogen. West (8) found that water stress quantitatively reduced protein in corn seedlings germinated from 1 to 6 days in darkness, apparently by slowing protein degradation in the endosperm-scutellum and protein formation in the seedling. Chen et al. (2) reported protein levels in citrus seedlings responded differentially to intensifying water deficits. A 3-phase system of protein fluctuation was detected; protein increased at the beginning of dehydration, decreased at medium dehydration, and increased again slightly at extreme dehydration.

This study was conducted to determine the influence of varying degrees of heat and moisture stress on relative nitrate reductase activity and the contents of nitrate, molybdenum, various nitrogen fractions, and total nitrogen in young corn plants.

**MATERIALS AND METHODS**

Experiments were conducted in growth chambers with a light intensity of approximately 2000 f.-c. K-189® corn was grown in 6-inch plastic pots filled with vermiculite, 12 plants per pot. Vermiculite was maintained near field capacity with double strength Hoagland No. 1 nutrient solution (4) and temperature was maintained at 20° C. A 16-hour photoperiod (4 a.m. to 8 p.m.) was used both before and during stress. Entire plants above vermiculite were always sampled at 9 a.m.

Plants were exposed to stress conditions at the 4-leaf stage (about 10 days old). The vermiculite was saturated with the Hoagland No. 1 nutrient solution and allowed to drain to field capacity. Plants received no further moisture during the experiment. Temperature in the chamber was increased to a constant 38° C. and relative humidity was decreased by means of a dehumidifier operating continuously in the chamber. Relative humidity gradually decreased from above 90% to 40% after 7 days when the experiments were terminated. First wilting of leaves was noted after 3 days. In preliminary experiments, the permanent wilting point was reached after 9 days of stress.

Moisture status of plants was determined in entire plants by changes in moisture content and in relative turgidity. Moisture content was determined by weighing entire plants before and after drying for 22 to 24 hours at 70° C. Relative turgidity was determined by modification of the method outlined by May and Milthorpe (6). Plants were weighed immediately after being harvested, placed in a saturated atmosphere in the dark at 25° C. for 6 hours to regain turgidity, and reweighed. Percent relative turgidity was expressed as a ratio of weight initially present to that at full turgidity.

Nitrate reductase was extracted from duplicate samples of fresh plant material. Samples were ground in an Omnimixer at 16,000 rpm for two 1-minute intervals, using media described by Hageman and Flesher (3). The extract was filtered through a fine mesh sieve and centrifuged at 2° C. for 15 minutes at 20,000 g. The supernatant was assayed. Extracts were kept at 2—3° C. throughout the entire analysis.

Nitrate reductase activity was measured by the Evans and Nason method as reported by Hageman and Flesher (3), using 0.2 ml of extract and a 30-minute incubation.

Extraction of nitrate was the same as for nitrate reductase except that plant material was blended in distilled water at the ratio of 1:5 by weight and the filtrate centrifuged 5 minutes at 6,000 g.

Determinations of nitrate were made by further modification of the Nelson, Kurtz, and Bray method as reported by Woolley.