Ethylene Evolution by Tomato Plants Receiving Nitrogen Nutrition from Urea

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Abstract. Urea fertilization of 'Heinz 1350' tomato (*Lycopersicon esculentum* Mill.) in sand or soil culture did not enhance ethylene evolution or restrict growth relative to plants receiving NO_3^- , whereas NH_4^+ nutrition doubled the relative rates of ethylene evolution and restricted relative growth. Inhibitors of N transformations in media (nitrapyrin, Np; hydroquinone, HQ; and phenylphosphorodiamidate, PPD) had no apparent stimulator effects on ethylene evolution of plants grown on urea or NO_3^- nutrition in sand or soil. Ethylene evolution was enhanced by PPD relative to that by Np or HQ for plants receiving NH_4^+ nutrition. Each inhibitor had toxic effects on plant growth. Increasing K⁺ supply from 0 to 8 mM in nutrient solutions decreased ethylene evolution and increased plant growth with urea fertilization. Urea had low phytotoxicity if its hydrolysis to NH_4^+ was prevented in the media. Chemical names used: p-dihydroxybenzene (hydroquinone); benzenephosphorodiamide (phenylphosphorodiamidate); 2-chloro-6-(trichloromethyl)pyridine (nitrapyrin).

Tomato plants grown with NH_4^+ -N nutrition develop a syndrome of disorders identified as NH_4^+ toxicity (Maynard et al., 1966; Uljee, 1964; Wilcox et al., 1973). A characteristic of physiological stress from NH_4^+ toxicity in tomato is evolution of ethylene at rates exceeding those of unstressed plants (Corey et al., 1987). In soil culture, the stress can be lessened by providing NH_4^+ -nourished plants with K⁺ at molar equivalency to the NH_4^+ supply (Maynard et al., 1968; Barker and Lachman, 1986; Corey et al., 1987). In sand culture, in addition to adequate K⁺, buffering of the medium to a neutral pH is required to prevent NH_4^+ toxicity and elevation of ethylene evolution (Maynard and Barker, 1969; Corey and Barker, 1989).

Since urease activity by soil microorganisms releases urea-N as NH_4^+ , urea is an ammoniacal fertilizer, giving the possibility that its use may lead to NH_4^+ toxicity. It is not clear whether urea as such is phytotoxic. However, if the urea molecule is less toxic to plants than NH_4^+ , inhibition (Bremner and Douglas, 1971; Martens and Bremner, 1984) of the hydrolysis of urea may prevent or lessen its phytotoxicity.

In the present greenhouse study, ethylene evolution, as a measurement of plant stress, was determined for tomato receiving urea, NH_4^+ , or NO_3^- nutrition. The effects of inhibitors of urease and nitrification and the

effects of K⁺ supply on ethylene evolution by plants on urea nutrition also were assessed.

In one experiment, 'Heinz 1350' plants established in soil culture were given 100 ml 0.02 M urea, $(\text{NH}_4)_2\text{SO}_4$, or $\text{Ca}(\text{NO}_3)_2$ daily for 3 weeks. Concurrently with this experiment, 'Heinz 1350' was grown with 0.02 M urea in soil culture with two urease inhibitors (hydroquinone and phenylphosphorodiamidate) and a nitrification inhibitor (nitrapyrin) added individually to the urea solution at 10 mg·liter⁻¹. In a second experiment, 'Heinz 1350' was grown for 4 weeks in sand culture using complete nutrient solutions (Hoagland and Arnon, 1950) with 210 mg N/liter from NO_3^- , NH_4^+ , or urea in sand culture. Also in this experiment, 'Heinz 1350' was grown in sand culture using the same nutrient solutions with the two urease inhibitors and the nitrification inhibitor added individually at 10 mg·liter⁻¹. In a third experiment, 'Heinz 1350' was cultured in sand for 5 weeks, with 210 mg N/liter from urea and with K⁺ progressively increasing from 0 to 8 mM (0 to 312 mg K/liter) in the nutrient solutions. In the experiments in sand culture, 100 ml or more of nutrient solutions were added daily to displace solution held by the sand.

All experiments were conducted in a glasshouse with natural irradiance in Spring 1988 and 1989. Ethylene evolution was measured by sealing plants in bell jars and subsequently collecting samples of the atmosphere under the bell jars for analysis by gas chromatography (Corey et al., 1987). Acidity of the sand culture was determined electrometrically on aqueous eluates from the media at the end of the experiments. Ammonium-N and nitrate-N in eluates or soil samples were determined volumetrically

(Brenner, 1965) and electrometrically (Barker, 1971), respectively. Analyses of Ca, Mg, and K were by atomic absorption spectrophotometry (Barker and Corey, 1988). All experiments were factorial with all treatment combinations and were conducted in randomized, complete-block designs of four or five replications.

In sand culture, plants receiving urea nutrition had growth, expressed as dry weight of shoots, equal to that of plants on NO_3^- , and exceeding that of plants on NH_4^+ (Table 1). Lower leaves of plants grown on urea were slightly chlorotic, and plants grown on NO_3^- had no symptoms of nutritional stresses. However, plants receiving NH_4^+ had chlorosis, necrosis, and cupping of laminae and produced ethylene at a rate exceeding that of the plants receiving urea or NO_3^- . Amounts of NH_4^+ detected in the urea-based solution were not significantly different from those of the NO_3^- -based solution. Plants fertilized with urea in soil-based media showed some indications of nutritional stresses. These plants had moderate leaf epinasty but no stem lesions associated with NH_4^+ toxicity (Barker et al., 1967). Urea nutrition produced slight but nonsignificant restrictions in shoot growth and a small, nonsignificant elevation in ethylene evolution relative to NO_3^- -grown plants, whereas the effects due to NH_4^+ nutrition were significant (Table 2).

In sand culture, phenylphosphorodiamidate (PPD) inhibited plant growth relative to that obtained with nitrapyrin or hydroquinone (Table 3). The inhibition of growth by these chemicals was greater with urea or NH_4^+ than with NO_3^- . The symptomatology for the plants receiving NH_4^+ could not be distinguished among the three inhibitors due to the dominance of NH_4^+ -toxicity symp-

Table 1. Growth and ethylene evolution by 'Heinz 1350' tomato grown on ammonium, nitrate, or urea in sand culture.

Form of N	Measurement ^a		
	NH_4^+ -N in eluate (mg·liter ⁻¹)	Shoot dry wt (g/plant)	Ethylene evolution (nl·g ⁻¹ ·hr ⁻¹)
NH_4^+	89 a	13 b	35 a
NO_3^-	0 b	21 a	13 b
Urea	8 b	20 a	16 b

^aMeans followed by different letters within columns are significantly different at $P \leq 0.05$, using Duncan's multiple range test.

Table 2. Growth and ethylene evolution by 'Heinz 1350' tomato grown on ammonium, nitrate, or urea in soil culture.

Form of N	Measurement ^a		
	Stem lesion rating	Shoot dry wt (g/plant)	Ethylene evolution (nl·g ⁻¹ ·hr ⁻¹)
NH_4^+	3 a	6 b	35 a
NO_3^-	0 b	10 a	13 b
Urea	0 b	8 ab	16 b

^aMeans followed by different letters within columns are significantly different at $P \leq 0.05$, using Duncan's multiple range test. Stem lesion rating: 0 = no lesions; 3 = severe lesions.

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Table 3. Interactions and main effects of N form and chemical inhibitors of N transformation on growth and ethylene evolution by 'Heinz 1350' tomato grown in sand culture.

Form of N	Measurement ^a											
	NH ₄ ⁺ in eluate (mg·liter ⁻¹)				Shoot dry wt (g/plant)				C ₂ H ₄ evolution (nl·g ⁻¹ ·hr ⁻¹)			
	Np	HQ	PPD	Mean	Np	HQ	PPD	Mean	Np	HQ	PPD	Mean
NH ₄ ⁺	102 b	128 a	112 b	114 a	9 a	10 a	6 b	8 c	37 b	49 b	93 a	60 a
NO ₃ ⁻	1 a	6 a	2 b	2 b	15 a	15 a	12 b	14 a	18 a	26 a	18 a	21 b
Urea	14 a	6 a	4 a	8 b	15 a	17 a	5 b	12 b	20 a	13 a	20 a	19 b
Mean	39 a	46 a	41 a		13 b	14 a	8 c		25 a	30 a	45 a	

^aMeans for N form × inhibitor interaction (rows) and marginal means (main effects) are significantly different ($P \leq 0.05$) if followed by different letters by Duncan's multiple range test. Abbreviations: Np, nitrapyrin; HQ, hydroquinone; PPD, phenylphosphorodiamidate.

Table 4. Ethylene evolution by tomato plants grown in soil culture with urea and nitrification or urease inhibitors.

Inhibitor ^a (g/plant)	Measurement ^a			
	Shoot dry wt	N in soil ^a		Ethylene evolution
	(g/plant)	NH ₄ ⁺ (mg·g ⁻¹)	NO ₃ ⁻ (mg·g ⁻¹)	(nl·g ⁻¹ ·hr ⁻¹)
None	8.0 b	0.15 b	0.16 a	7.6

soil.

Table 5. Concentration of K, dry weight, and ethylene evolution by shoots of 'Heinz 1350' tomato grown on urea nutrition with variable K levels in sand culture.

Measurement	K (mmol/L)	Shoot dry wt (g/plant)	Ethylene evolution (nl·g ⁻¹ ·hr ⁻¹)
Regression of measurement against K supply; L, linear; Q, quadratic.			
*** $P \leq 0.05$ or 0.01, respectively.			

toms. With urea nutrition, plants receiving nitrapyrin had symptoms resembling incipient NH₄⁺ toxicity, although little NH₄⁺ (≈ 1 mM) accumulated in this solution, and the quantities accumulating were not significantly different from those of the other treatments with urea. Lower leaves of the plants receiving hydroquinone were chlorotic. Marginal necrosis or chlorosis occurred on the lower leaves of the plants receiving

NO₃⁻ with nitrapyrin, hydroquinone, or PPD. Ethylene evolution was stimulated by PPD only for the plants receiving NH₄⁺. Urease activity in the nutrient solutions was apparently negligible. The lack of a significant difference in NH₄⁺ concentration in the medium between nitrapyrin and hydroquinone or PPD supports this conclusion.

In soil culture, dry weights of shoots of plants receiving urea were affected only slightly by the inhibitors. Plants receiving PPD had severe foliar burn but no stem lesions. Plants grown with nitrapyrin developed severe stem lesions and twisting of the young foliage. Addition of hydroquinone had no effect on the appearance of the foliage, although severe stem lesions developed. Only the addition of PPD enhanced ethylene evolution relative to that of the plants receiving urea and no chemical inhibitor (Table 4).

Concentrations of K⁺ in the shoots increased linearly as K⁺ increased in the nutrient solution on sand culture (Table 5). Increases in dry weights of shoots accompanied increases in K⁺. Although already at relatively low levels, ethylene evolution by the shoots declined substantially when the concentration of K⁺ reached 2 mM in the solution and 2% in the shoots. Shoot concentrations of Ca and Mg averaged 1.47% and 0.39%, respectively, on a dry-weight basis, and were unaffected by K⁺ supply. The acidity of the nutrient solution increased as K⁺ increased in the solution. The pH decrease may have been due to plant absorption of K⁺ and release of H⁺ to maintain electrical neutrality in the solution. The increase in acidity did not promote phytotoxicity from urea. With NH₄⁺ nutrition, declines in pH associated with absorption of NH₄⁺ intensify the phytotoxicity of NH₄⁺ (Barker et al., 1967). Enhanced phytotoxicity of NH₄⁺ in acid media appears to be a unique interaction between form of N and H⁺ concentration. Only traces of NH₄⁺ were detected in the media in this experiment.

This research shows that urea has low phytotoxicity. Toxic effects of urea seem related to formation of NH₄⁺ in the medium.

Because of low microbiological activity in sand culture, conversion of urea to NH₄⁺ was negligible. In soil-based media, urease activity may be sufficient to lead to NH₄⁺ toxicity, although the toxicity was suppressed relative to that noted in cultures with NH₄⁺ alone. Chemical inhibition of urease stops NH₄⁺ accumulation in media; however, chemicals used for this action should be evaluated for their phytotoxicity.

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