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RESOURCE AVAILABILITY AND THE ABUNDANCE OF AN N-BASED DEFENSE IN AUSTRALIAN TROPICAL RAIN FORESTS

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Abstract. Plant defense theories predict that relatively resource-rich environments (those with more fertile soil) will support a greater abundance of plants with nitrogen-based chemical defense, but this has yet to be adequately tested. We tested this prediction by measuring the diversity and contribution to total biomass of cyanogenic plants (those that release hydrogen cyanide from endogenous cyanide-containing compounds) in the Australian tropical rain forest. We examined 401 species in thirty 200-m^2 plots, six at each of five sites, for cyanogenesis. In upland/highland rain forest, two pairs of sites similar in rainfall and altitude, but differing in soil nutrients, were selected, as well as one site in lowland rain forest. Sites differed markedly in species composition and foliar N was positively related to soil fertility. Holding altitude constant, we did not detect significant differences in the proportion of cyanogenic species with soil fertility, nor did we consistently detect significant increases in the contribution of cyanogenic species to total biomass on higher nutrient sites. Thus we found no clear evidence that soil fertility affects the distribution and prevalence of species investing in a constitutive N-based defense at the community level.

Key words: Australia; chemical defense; cyanogenesis; cyanogenic glycoside; herbivory; nitrogen; resource allocation; secondary metabolite; soil nutrient; tropical rain forest.

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INTRODUCTION

There are large differences among plant species in both the type and amount of chemical defense. Consequently, a number of hypotheses have been formulated concerning patterns of secondary chemistry within and between plant communities (Janzen 1974, Feeny 1976, Rhoades and Cates 1976, Bryant et al. 1983, Coley et al. 1985, Herms and Mattson 1992). Some theories emphasize herbivore pressure in selecting for plant defense characteristics (e.g., the apparency theory [Rhoades 1979]). Other theories emphasize the availability of resources in the environment in constraining the evolutionary response of plants to herbivory, thereby determining the type and amount of defense (Janzen 1974, Coley et al. 1985, Coley 1988, Herms and Mattson 1992). Resource-limited environments, where the replacement cost of tissue is assumed to be greater, are hypothesized to favor greater investment in chemical defense (see Janzen 1974, Grime 1977, Bryant et al. 1983, Coley et al. 1985, Coley 1988). The balance between resources in the environment (Bryant et al. 1983), and intrinsic growth rate and leaf lifetime are further predicted to influence the type of chemical defense (Coley et al. 1985, Coley 1988).

Importantly, systematic and directional pressures from both resource availability and herbivores may not only alter the defensive attributes of plant communities by changes within species, but may also determine which species remain components of the flora and which ones fail to persist (Gartlan et al. 1980). Nutrient availability or soil fertility (including soil N), for example, may constrain the distribution of species investing in a costly constitutive N-based defense. Thus, such species might be favored at sites of higher nutrient availability, while their persistence could be compromised at sites of lower nutrient availability where the large relative investment in N-based defense might incur a greater fitness cost.

Comparative community-level studies investigating patterns in the distribution of chemical defenses have focused on carbon-based "quantitative" defenses (sensu Feeny 1976; e.g., McKey et al. 1978, Davies et al. 1988, Waterman et al. 1988); relatively little is known about patterns in the distribution of N-defended species. With regard to alkaloids, one study compared the frequency of alkaloid-bearing plants between two forests on sites differing in soil fertility, and found a greater proportion of alkaloid-producing species in the forest on high nutrient soil (Gartlan et al. 1980). Alkaloids are rapidly turned over (Gershenzon 1994), relatively diverse, and frequently inducible (Baldwin 1994, Gershenzon 1994). They therefore represent a challenge, not only for detection and quantification, but more particularly for assessing costs associated with resource allocation to induced defense (Ohnmeiss and Baldwin 1994, Baldwin et al. 1998).

Here, we address the hypothesis that sites with greater resource availability, those with more fertile soils

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derived from high-nutrient substrates, support more N-defended plants, in terms of the relative number of species and their contribution to total biomass. We chose to examine cyanogenic plants because cyanogenesis is a constitutive defense that is relatively demanding of plant resources (Gleadow and Woodrow 2000*a*). The frequently high proportion of leaf N allocated to cyanogenic glycosides (up to 20% in leaves of tropical *Prunus turneriana* seedlings, for example; Miller et al. [2004]) represents a substantial potential diversion of N away from primary metabolism.

Methods

Field sites

Fieldwork was conducted from 1999 to 2002. Five sites were selected in lowland and upland rain forest in the tropics of northeastern Queensland, Australia, where there is a mosaic of high- and low-nutrient soils derived from contrasting substrates. On the Atherton Tablelands, where forest type varies with both substrate and altitude (Tracey 1982), two pairs of sites with similar altitude and rainfall were selected. The first pair in upland rain forest comprised a high-nutrient site on soil derived from basalt at Lamins Hill (17°22′24″ S, 145°42′30″ E; altitude 850 m above sea level [asl]; complex mesophyll vine forest [Type 1b; Tracey 1982]), and one on low-nutrient soil derived from granite at Mt. Nomico (17°13′18″ S, 145°40′24″ E, 900 m asl; complex notophyll vine forest [Type 6; Tracey 1982]). The second pair in highland rain forest comprised high- and low-nutrient sites on soils derived from basalt and rhyolite, respectively, at Longlands Gap (1100–1200 m asl), where there is a sharp boundary in forest type defined by basalt (17°27′42″ S; 145°28′30″ E; complex notophyll vine forest [Type 5a; Tracey 1982]) and rhyolite (17°27′18″ S, 145°28′36″ E; simple microphyll vine forest [Type 9; Tracey 1982]) parent substrates. A fifth site in lowland rain forest (complex mesophyll vine forest; Type 1a; Tracey 1982) near Cape Tribulation and Myall Creek (16°6′12″ S, 145°26′54″ E; altitude 40 m asl) on relatively nutrient-poor red clay–loam podsol derived from metamorphic substrate, was also selected. Due to the relative uniformity of substrate in the lowland area, no site with high-nutrient soil was found. Further information on the soils and climate is outlined in Miller et al. (2006).

To capture species diversity, six plots were established at each of the five sites (total 1200 m^2 per site). All individuals (palms, trees, vines) with a diameter at breast height (dbh) $\geq$ 5 cm were tagged and identified. All additional species (dbh $<$ 5 cm) present in lower strata, with the exception of herbaceous ground species, were also tagged and identified.

Chemical analyses

Quantification of cyanogenic glycosides.—In total, 401 woody plant species from 87 families were tested for cyanogenesis, and 18 cyanogenic species from 13 families were found (see Miller et al. 2006). The concentration of cyanogenic glycosides in leaves of all cyanogenic individuals was measured according to Miller et al. (2006).

Foliar and soil nitrogen.—Due to the large sample size, this part of the study focused on the high-nutrient basalt site at Lamins Hill, and the low-nutrient granite site at Mt. Nomico. A range of species ($n = 26$) that were exclusive to either basalt ($n = 14$ species) or granite ($n = 12$ species) soils were selected, as well as a range of key species ($n = 13$ species) common to both substrates. The species selected were well represented at each site, and the taxonomic group of species selected was considered important in order not to bias the data set; for example, species in the family Proteaceae, which typically have low foliar N, were selected in each category, while there were several species in each category from the dominant rain forest family, the Lauraceae. In addition, in order to analyze the proportion of leaf N allocated to cyanogenic glycosides, the foliar N content of the majority of cyanogenic individuals was determined. Total N concentration of 5–10 mg of freeze-dried tissue was determined according to Miller et al. (2004).

Soil samples (0–15 cm depth) were taken from five locations within each plot. These samples were pooled, oven-dried at 70°C, and put through a 2-mm sieve to remove root matter and stones. A subsample was finely ground using a Makla Mill (Crompton-Parkinson, Melbourne, Australia) and 1 mm sieve. Duplicates of samples (30–35 mg) were analyzed using a Carlos Erba NA 1500 Series 2 NCS Analyzer (Fisons Instruments, Milan, Italy), calibrated using the standard atropine.

Statistical analyses

The comparison of foliar N was conducted using both two-way ANOVA computed using a general linear model, and nested ANOVA using the Satterthwaite approximation for uneven subgroup sample size to compare species exclusive or common to basalt or granite (Sokal and Rholf 1995). Comparison of individual species data between sites was conducted using one-way ANOVA; post hoc analysis was performed using Dunnett's test for unequal sample sizes. A G test was used to compare the proportion of basal area in cyanogenic stems at each site. The normality of distributions of cyanogenic glycoside concentrations within populations was tested using the Ryan-Joiner (Shapiro-Wilks) normality tests computed using Minitab Release 14 (Minitab, Pasadena, California, USA), which was also used to compute general linear models and for regression analyses.

The species composition of sites was compared using ordination by nonmetric multi-dimensional scaling (NMDS), based on a matrix of Bray-Curtis similarity coefficients (Bray and Curtis 1957) for presence–absence data for all species present in each plot. Analyses were performed using Primer 5 for Windows (version 5.2.2, Primer-E Ltd., Plymouth, UK).

TABLE 1. Distribution, frequency, and proportional contribution of cyanogenic (CN) species to basal area in 1200 m^2 of forest at five sites differing in soil fertility in tropical rain forest in northeastern Australia.

Site	Substrate	Nutrient status	Total soil N (%)†	Total no. species	No. CN species	CN species (% of total)	Basal area (m^2)	CN species (% basal area)§
Lamins Hill	basalt	high	$0.60^{a} \pm 0.05$	169	10	5.9	13.39	7.1^{ab}
Mt. Nomico	granite	low	$0.25^{b} \pm 0.04$	169	8	4.7	5.74	3.3^{bc}
Longlands Gap	basalt	high	$0.75^{a} \pm 0.10$	116	6	5.2	9.70	13.4^{a}
Longlands Gap	rhyolite	low	$0.62^{a} \pm 0.14$‡	76	4	5.3	10.67	1.2^{c}
Lowland, Myall Creek	metamorphic	low	$0.33^{b} \pm 0.06$	139	9	6.5	7.42	11.6^{a}

Note: Total number of species and basal area (stems with dbh $\geq$ 5 cm) are also given.

† For soil N, significant differences ($F_{4,25} = 34.9$, $P < 0.0001$) are indicated by different letters. Data are means ± SE (% dry mass) based on pooled bulk samples, for each of six plots at five sites.

‡ The value for rhyolite soil was confounded due to difficulty sampling soil from the granular siliceous substrate, which, even after sieving, contained a substantial amount of organic matter.

§ Percentage of basal area in cyanogenic stems; significant differences at the 5% level determined by a G test are indicated by different letters.

RESULTS

Site characteristics and floristics

Species richness varied between the sites (Table 1). Upland rain forests at Mt. Nomico and Lamins Hill were the most species rich, with 169 species in the 1200 m^2 area. The lowland rain forest was also species rich (n = 139 species), while the highland rain forest on the nutrient-poor rhyolite at Longlands Gap was the least species rich ($n = 76$ species; Table 1). Sites also differed markedly in species composition as illustrated by NMDS ordination based on species presence–absence: upland and highland rain forest sites clustered separately from the lowland rain forest site, and among Atherton Tableland sites, high-nutrient sites clustered separately from low-nutrient sites (Fig. 1). On the Atherton Tableland, 66% of all species ($n = 336$) were restricted to either high-nutrient basalt, or low-nutrient granite/rhyolite substrate. There were also similarities between sites matched for altitude; the upland rain forest sites of Mt. Nomico and Lamins Hill shared 70 species, while highland rain forest sites at Longlands Gap shared 37 species. Lowland and upland rain forests differed substantially, with 57% of species in the lowland rain forest not found at the other sites. Thirty species were found at all sites. Species density and basal area are presented in Appendix A.

Soil nutrients and foliar nitrogen

This study did not include a comprehensive analysis of soil nutrients, because the soils in the study area are well characterized: basalt soils have higher concentrations of N, P, and available cations than soils derived from acid igneous (granite/rhyolite) or metamorphic substrates (e.g., Laffan 1988, Saker et al. 1999). We found that total N concentration determined for discrete soil samples at each site, with the exception of the rhyolite soil (which even after sieving contained a substantial amount of organic matter), was consistent with the known soil differences (Table 1; see Spain 1990, Kanowski 2001).

Overall, there was a positive relationship between soil N and foliar N, based on the comparison between Lamins Hill and Mt. Nomico (Appendix B and C). In addition to significant differences between species ($P <$ 0.001), foliar N differed significantly with soil type ($P <$ 0.05) across a range of species common to both sites ($n =$ 13) (Appendix B). For all species, the mean N concentration of individuals on basalt was higher than for conspecific individuals on granite. For the 13 species, the mean N content of individuals found on basalt (Lamins Hill) was 2.06% compared to 1.84% on granite (Mt. Nomico; Appendix B). In addition, the foliar N content of species restricted to basalt (n = 14) was significantly higher compared with those restricted to granite (n = 12) ($P < 0.01$; Appendix C). The relationship between soil nutrients and leaf N was also

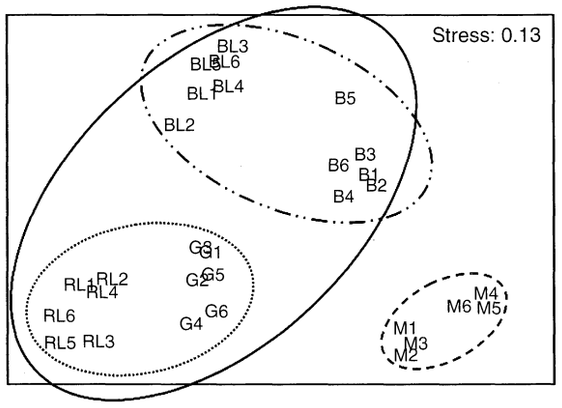

FIG. 1. Ordination by nonmetric multidimensional scaling (NMDS) using Bray-Curtis similarity coefficient based on presence–absence data for all species in six plots at five sites. One site is in lowland rain forest near Cape Tribulation (metamorphic substrate; dashed line; M) and four sites are in upland/highland rain forest on the Atherton Tableland (circled by solid line). On the Tableland, the higher nutrient basalt sites (dotted-dashed line) at Lamins Hill (B) and Longlands Gap (BL) were distinct from the lower nutrient sites (dotted line) on granite at Mt. Nomico (G) and on rhyolite at Longlands Gap (RL). The plots at each site are numbered 1–6.

TABLE 2. The percentage of N allocated to cyanogenic glycosides (CN-N/N%) in leaves of cyanogenic species.

Species	Family	CN-N/N%	*n*	Substrate
Beilschmiedia collina	Lauraceae	0.05–3.4	33	B1,G,R
Brombya platynema	Rutaceae	0.5–4.7	16	M
Cardwellia sublimis	Proteaceae	old: 0.06–0.44 young: 0.4–2.9 leaf tip: 0.5–4.2	12	B1,B2,G,R,M
Cleistanthus myrianthus	Euphorbiaceae	old: 0.03–0.06 young: 0.13–0.25	3	M
Clerodendrum grayi	Verbenaceae	1.9–10.0	7	B1,B2,G
Elaeocarpus sericopetalus	Elaeocarpaceae	3.5–14.6	9	G,R
Embelia grayi	Myrsinaceae	0.04–0.25	2	B1,B2
Flagellaria indica	Flagellariaceae	0.02–0.23	8	B1,G,M
Helicia australasica	Proteaceae	0.2–1.0	2	M
Helicia blakei	Proteaceae	0.02–0.06	3	B1
Mischocarpus exangulatus	Sapindaceae	0.7	1	B1
Mischocarpus grandissimus	Sapindaceae	0.02–0.07	2	G,M
Opisthiolepis heterophylla	Proteaceae	old: 0.03–0.8 young: 4.8	7 1	B1,B2
Parsonsia latifolia	Apocynaceae	0.5–8.2	2	B1,G,R
Passiflora sp. (Kuranda BH12896)	Passifloraceae	0.68–2.3	5	M
Polyscias australiana	Araliaceae	0.02–0.09	9	B1,B2,G,R,M
Prunus turneriana	Rosaceae	old: 6.4–13.5 young 10.5–11.4 leaf tips: 10.3–14.6	3	B1,M
Ryparosa kurrangii	Achariaceae	old: 3.5–9.8† young, range: 3.7–7.8	~2005	M

Notes: There was no significant site (soil nutrient) effect on CN-N/N% within or across species. The range of values for *n* individuals is given, as is the occurrence of species at different sites: basalt at Lamins Hill (B1) and Longlands Gap (B2), granite at Mt. Nomico (G), rhyolite at Longlands Gap (R), and metamorphic substrate in lowland rain forest (M). Values are for fully expanded (older) leaves, unless otherwise stated.

† Includes values determined by B. L. Webber (*unpublished data*).

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detected across all sites for the one species found commonly at all fives sites, *Polyscias australiana* (Appendix D).

Soil nutrients and the distribution of cyanogenic species

We found that the number of cyanogenic species tended to be greater at both the upland basalt sites than at the corresponding nutrient-poor (granite/rhyolite) sites, but that the percentage of cyanogenic species at each site did not differ significantly with substrate nutrient status (Table 1). In addition, we did not detect consistently significant differences in the contribution of cyanogenic species to total biomass (the percentage of basal area contributed by stems of cyanogenic species) within paired high- and low-nutrient sites (Table 1). Only at the adjacent sites at Longlands Gap, where the percentage of basal area in cyanogenic stems was 10 times higher on basalt (13.4%) than on low-nutrient rhyolite (1.2%), was this difference significant (Table 1). Furthermore, the contribution of cyanogenic species to basal area in the lowland rain forest on a low-nutrient soil type was comparatively high (Table 1).

Soil N effects on cyanogenic glycoside concentration and N allocation to defense

There was no detectable effect of soil N or of foliar N on concentrations of cyanogenic glycosides, either within species, or across species (data not shown), despite large variations in concentrations of cyanogenic glycosides between conspecific individuals (see Miller et al. 2006). Similarly, there was no detectable effect of soil N on the percentage of leaf N allocated to cyanogenic glycosides (CN-N/N%) either within or between species (data not shown). This percentage was frequently around 5%, and greater than 10% in the highly cyanogenic species; however, species with high values were not restricted to high-nutrient sites (Table 2). For example, while *Clerodendron grayi* (up to 10% CN-N/N%) and *Prunus turneriana* (up to 14% CN-N/N%) are more common on high-nutrient basalt soils, *Elaeocarpus sericopetalus* (up to 15% CN-N/N%) is restricted to nutrient-poor sites (Table 2).

DISCUSSION

Resource availability and the distribution of cyanogenic species

This is the first study to examine the prevalence of cyanogenic species in plant communities in relation to soil nutrients (N), and one of few community-based studies on N-based defenses. Overall, 4.5% of species were cyanogenic, and the frequency of cyanogenesis was similar at all sites (range 4.7–6.5%; Table 1). No effect of soil nutrients on the frequency of cyanogenesis was found. These values are similar to the only previous quantitative study in tropical rain forests which found

that 4.1% of 401 species were cyanogenic, and that 3.0% of total basal area was in cyanogenic stems (dbh > 10 cm); however, details of soil type were not provided (Thomsen and Brimer 1997). By contrast, the frequency of cyanogenic species in two nutrient-poor tropical communities (seasonal cloud forest in India and a woodland "restinga" in Brazil) differed markedly with 2.3% (Mali and Borges 2003) and 21% (Kaplan et al. 1983) of species reported to be cyanogenic, respectively. Though community-based, these studies did not report the size of plants tested, and consequently the frequency of cyanogenic species alone is uninformative in relation to the relative success of those species in different communities. In addition, in the latter study, some doubt with regard to the identification of cyanogenic species was expressed by the authors (Kaplan et al. 1983).

Information on the community-level distribution of other N-based defenses such as alkaloids is also relatively limited. Much of what is known about the distribution of alkaloids derives from phytochemical screening for novel bioactive compounds (e.g., Hartley et al. 1973); hence few of these studies have been community-based or defined with respect to forest area or plant size (see only Gartlan et al. 1980, Janzen and Waterman 1984, Mali and Borges 2003). In the comparative study by Gartlan et al. (1980), the proportion of alkaloid-producing species in forest on nutrient rich lateritic soils in Uganda was greater than in similar forest on nutrient-poor acid sand soils in Cameroon; however only 74 and 19 species, respectively, were investigated at each site. The Ugandan forest on fertile laterite was also characterized by higher foliar N and lower C-based defense (McKey et al. 1978, Gartlan et al. 1980), a pattern consistent with other comparative studies that together indicate that low leaf N and high levels of C-based metabolites are typical of forests on low-nutrient soils (Davies et al. 1988, Waterman et al. 1988, Mali and Borges 2003; but see Cunningham et al. 1999).

There are two key limitations in studies of the distribution of N-based defenses to date. One limitation in the large-scale comparisons between remote sites is that the forests differ not just in soil nutrients, but also in a range of other factors such as altitude, climate, herbivore community, human impact, and successional stage, as well as in the survey method applied to them. Thus interpretation in relation to soil nutrients is challenging and most likely confounded. The Atherton Tableland in northeastern Australia affords a powerful opportunity to draw community-level comparisons relating to soil type because of its mosaic of high- and low-nutrient substrates across a range of altitudes and rainfall zones. Thus, a strength of this study is that it was able to essentially control for altitude, climate, and successional stage. The second limitation is that the majority of studies report only the proportion of species deploying N-based defense, rather than a measure of biomass, which is a more meaningful indicator of relative success.

Contrary to our hypothesis, we did not detect a consistent effect of soil nutrient status on the proportional contribution of cyanogenic species to total biomass (Table 1). Specifically, no significant difference in percentage of basal area in cyanogenic stems was found between upland rain forest at Lamins Hill and Mt. Nomico, but a 10-fold difference between sites was detected at Longlands Gap (Table 1). These data therefore provide no clear evidence that soil N is a key determinant of the cost-effectiveness and distribution of a constitutive N-based defense, and indicate that the picture is more complex. Similarly, our data on the abundance of acyanogenic and cyanogenic individuals is not consistent with a nutrient effect on the cost-effectiveness of cyanogenesis. In contrast to studies that report an increase in the proportion of acyanogenic individuals at lower nutrient sites (Dickenmann 1982, Blaise et al. 1991, Louveaux et al. 1996, Schappert and Shore 2000), we found that all but one species in our study invested uniformly in cyanogenesis (i.e., acyanogenesis was absent; Miller et al. 2006), often with a sizeable allocation of total leaf N (e.g., up to 15% of leaf N in *Elaeocarpus sericopetalus*; Table 2), which was not correlated with soil nutrient content. An explanation for these contradictory findings is hard to identify. We have focused here on the differences in soil N, but limitation of other resources (e.g., phosphorus) may interact with N availability to modify the costs for a species of N-based defense at different sites (Gartlan et al. 1980). Moreover, the relative importance of abiotic factors (e.g., resource availability) and biotic factors (i.e., those related to herbivores) in determining patterns in defenses among communities remains much debated (Berenbaum 1995). It may also be that the effects of soil N are too subtle to consistently detect in a study of this size. Further systematic community-level studies of especially N-based plant defenses are required.

While differences in altitude and climate make direct comparison between lowland and upland/highland forests difficult, differences in the percentage of cyanogenic biomass between lowland and upland/highland rain forests were also inconsistent with the hypothesized soil N effect (Table 1). In the lowland rain forest, on low-nutrient soil, the percentage of cyanogenic forest biomass (11% basal area) was significantly higher than at all but one highland site (Table 1). In addition, the majority of cyanogenic species in the lowland were restricted to, or even endemic to, the lowland rain forest (Miller et al. 2006). The greater percentage of basal area in cyanogenic stems in the lowland may in part reflect differences in forest structure between the lowland and upland/highlands. The lowland rain forest is very patchy in composition (Tracey 1982), and two locally dominant lowland subcanopy trees, the cyanogenic species *Brombya platynema* and *Cleistanthus myrianthus,* contributed a large percentage of cyanogenic basal area. Interest-

ingly, in contiguous forest, a third cyanogenic subcanopy species, *Ryparosa kurrangii*, was dominant, comprising up to 30% of the subcanopy (stems > 10 cm dbh) (Webber 2005). It may also reflect that environmental factors other than soil nutrients influence the outcome of competition and predation and the persistence of a cyanogenic species at a particular site. The lowland site differed in more than just soil nutrients, most notably in climate, which is milder and warmer compared to the more subtropical climate at altitude. Studies among populations of species polymorphic for cyanogenesis have identified both direct effects (e.g., plant growth rates) and indirect effects (e.g., influence on herbivore populations) of climate on the viability of the cyanogenic phenotype (Kakes 1989). Herbivores are a potentially strong selection agent through the removal of biomass, compromising plant growth and reproduction, and even causing plant mortality (Marquis 1984). Indeed, along a transect following an altitudinal gradient, a greater proportion of cyanogenic plants were positively correlated with both temperature and insect density, independent of plant systematics (Kaplan et al. 1983).

Soil N, foliar N, and phenotypic variation in cyanogenesis

The absence of a detectable relationship between foliar N (soil N) and cyanogenic glycoside content, or between soil type and the allocation of N to cyanogenic glycosides both across and within species may in part reflect the small sample size for some of the cyanogenic species (Table 2); however, it is not an expected finding. While some studies report a positive relationship between N supply (soil and/or foliar N) and cyanogenic glycoside content (e.g., Forslund and Jonsson 1997, Gleadow and Woodrow 2000*b*, Graham 2002), soil or foliar N and cyanogenic glycoside content are not always correlated (e.g., Briggs and Schultz 1990, Goodger et al. 2002, Miller et al. 2004, Webber 2005, Simon et al. 2007). Notably, a glasshouse study of *Prunus turneriana*, a highly cyanogenic canopy tree species found in this study, found no effect of elevated N supply on cyanogenic glycoside content (Miller et al. 2004).

In summary, this study identifies substrate/soil fertility as a significant factor influencing species composition and foliar N in tropical rain forests of northeastern Australia (Fig. 1; Tracey 1982). Despite its significant effect on foliar N, however, we found no compelling evidence that soil nutrients affect the frequency and abundance of species investing in a constitutive N-based defense (viz. cyanogenesis).

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Reports

APPENDIX A

Summary of stem density and basal area (m^2/ha), based on six 200-m^2 plots (1200 m^2 total area) at five sites in tropical rain forest (*Ecological Archives* E-089-091-A1).

APPENDIX B

Foliar N concentrations (%) in fully expanded leaves from 13 species common to high-nutrient soil (basalt) at Lamins Hill and low-nutrient soil (granite) at Mt. Nomico (*Ecological Archives* E-089-091-A2).

APPENDIX C

Comparison of mean foliar N concentrations (% dry mass) between common species that were found exclusively on high-nutrient basalt (Lamins Hill) or the low-nutrient granite at Mt. Nomico (*Ecological Archives* E-089-091-A3).

APPENDIX D

Mean foliar N concentration in the tree species *Polyscias australiana* growing at each of five sites in upland/highland and lowland tropical rain forest (*Ecological Archives* E-089-091-A4).