A possible interpretation of the profiles for the production rates is that the onset (or the cessation) of flow and its subsequent continuation stimulate the cells by two independent mechanisms. This is best seen qualitatively in the expression for the production rate (Eq. 2). The first term represents a transient decay in production after the initial perturbation, with the rapidity of decrease determined by the time constant C^{-1}. The step change in shear stress at the onset of flow may represent an acute stimulation in which cell response attenuates within several minutes. The second term (D) represents a constant production rate that dominates at long times (large values of CT). The continuation of flow provides a continuous stimulus, the degree of which is a function of the qualitative nature and possibly the magnitude of the flow. This may explain why the peak production rates for the two flow profiles with identical mean shear stresses were the same yet led to different steady-state rates.

Using a nonrecirculating flow system, Grabowski *et al.* (*5*) found that cultured bovine aortic endothelial cells subjected to a step change in steady shear stress responded with a burst in the PGI_2 production rate that decayed within minutes. Furthermore, the peak production rate was a function of the magnitude of the step change in the shear stress (*5*). These findings are consistent with our data.

Our results suggest that production of PGI_2 by the vascular endothelium under physiologic flow conditions may be significantly higher than that reported in studies based on cells cultured under stationary conditions (*9*). Furthermore, the lower concentrations of PGI_2 reported for veins relative to arteries may be due to the relative lack of pulsatile flow and the lower shear stress in veins. Our findings provide further evidence that the qualitative nature of blood flow may have a controlling role in endothelial cell function.

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12. For each experiment, the flow circuit was assembled and was autoclave sterilized. Under a laminar flow hood, the circuit was primed with 20 ml of culture medium, and a slide with a monolayer of cultured cells was positioned to form one of the plates of the parallel-plate chamber. Care was taken to avoid entrapment of air bubbles in the flow channel. The flow apparatus was then taken to the 37°C air curtain, where the gassing and roller pump connections were made.
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14. Samples were drawn into ibuprofen (Sigma) solution to give a final concentration of 1 m*M* to inhibit PGI_2 production by any suspended cells. Sample and replenishing volumes were both 1 ml. Attached cells were suspended by trypsinization and counted with a hemocytometer to determine the total viable count, as determined by trypan-blue exclusion staining.
15. PGI_2 was assayed by radioimmunoassay of the nonenzymatic breakdown product 6-keto-$PGF_{1\alpha}$. Before assaying, the samples were incubated at 37°C for 1/2 to 1 hour. Antibody to 6-keto-$PGF_{1\alpha}$ was obtained from Seragen. The assay sensitivity was 5 pg per 0.1 ml, and the cross-reactivity of the antibody with other culture media components was negligible. Sample concentrations were computed from scintillation counter data with a computer program modified from D. Rodbard *et al.* [*Radioimmunoassay* **1**, 469 (1978)]. Cumulative production of PGI_2 was determined by performing a mass balance over the flow loop, taking into account samples withdrawn and medium replenished.
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Xylem-Tapping Mistletoes: Water or Nutrient Parasites?

Abstract. *Most mistletoes parasitize higher plants by tapping the xylem (a conduction tissue) of their hosts. Field observations of diurnal gas exchange parameters and carbon isotope ratios in xylem-tapping mistletoes from three continents support the hypotheses that water use efficiency and carbon isotope composition are related and that mistletoes which are parasitic for water are also nutrient parasites, differing in their water use efficiency relative to that of their hosts on the basis of host nitrogen supply in the transpiration stream.*

Mistletoes are obligate epiphytic parasites of higher plant species (*1*). While a few mistletoes derive substantial nutritional benefits by connecting directly to the host's phloem tissues, most mistletoes tap only the xylem tissues and derive no nutritional benefit from their hosts other than the small amounts of organic carbon and nutrients carried in the transpiration stream (*1–3*).

While much is known about the anatomy and systematics of xylem-tapping mistletoes (*1, 3, 4*), less is known about their carbon, water, and nutrient relations (*3*), and several unusual physiological features of mistletoes have not been adequately explained (*5*). One such feature is that mistletoe leaves tend to have transpiration rates several times higher than those of their hosts (*1, 3, 5, 6*). Another is that they accumulate large amounts of calcium, potassium, and phosphorus and smaller amounts of nitrogen (*1, 3, 5, 7*). This holds true for autoparasitic mistletoes as well (*8*). Because xylem-tapping mistletoes exhibit high species diversity in arid and semiarid regions of the world (*1*), it is possible that this type of parasitism evolved principally as a means of water acquisition in water-limited habitats. However, many of these regions are also extremely nutrient-poor, and xylem-tapping parasitism may have evolved principally as a means of nutrient acquisition.

In this report we provide evidence, from many xylem-tapping mistletoe species at different global locations, that supports the hypothesis of Schulze *et al.* (*5*), namely that the unusually high transpiration rates of these mistletoes are in part a mechanism to acquire nitrogen, the nutrient potentially most limiting to their growth. To do this we compared

Table 1. Carbon isotope discrimination values ($\delta^{13}C$) in parts per thousand for mistletoe-host pairs from different arid regions in the United States (*15*), central Australia (*16*), and South Africa (*17*). The data are means ± standard errors. A more detailed analysis shows that the difference in carbon isotope ratio between mistletoe-host pairs is dependent only on host nitrogen content.

Region	Number of pairs	Host	Mistletoe	$\delta^{13}C$ (mistletoe) − $\delta^{13}C$ (host)
		Nitrogen-fixing hosts		
United States	7	−26.29 ± 0.50	−26.51 ± 0.23	−0.23 ± 0.41
Central Australia	28	−26.87 ± 0.21	−28.28 ± 0.30	−1.41 ± 0.33
South Africa	4	−24.67 ± 0.33	−25.73 ± 0.96	−1.06 ± 0.81
		Nonfixing hosts		
United States	8	−23.43 ± 0.10	−26.60 ± 0.14	−3.18 ± 0.19*
Central Australia	19	−26.54 ± 0.29	−28.83 ± 0.21	−2.30 ± 0.31*
South Africa	11	−24.70 ± 0.41	−26.91 ± 0.56	−2.21 ± 0.52

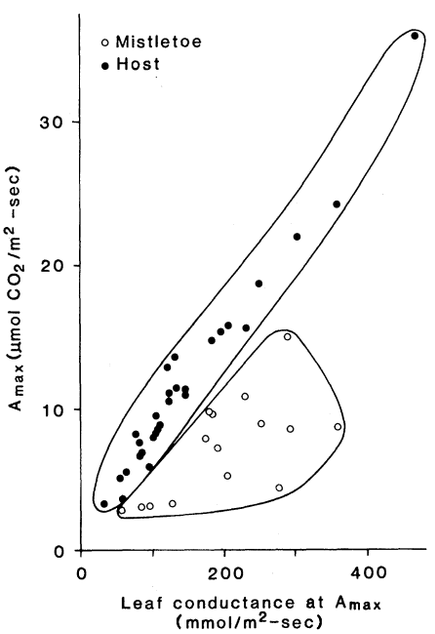
*Significantly different from value for nitrogen-fixing hosts in same region ($P \leq 0.01$, *t*-test).

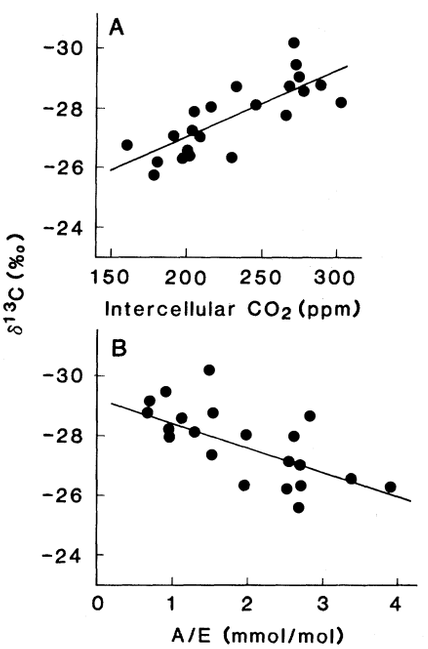
the performance of mistletoes growing on nitrogen-fixing hosts to that of mistletoes growing on hosts that do not fix nitrogen, since the former tend to have higher nitrogen contents in their xylem sap (*3, 7, 9*).

Surveys of the diurnal course of leaf performance under natural conditions and different water regimes in the spring were done to determine the relations between maximum photosynthetic rate during the day and leaf conductance (*10*) for mistletoe and host species in central Australia (Fig. 1). In all cases, for any photosynthetic rate, leaf conductance was higher in the mistletoe leaf than in the host leaf. Consequently, under these conditions, water use efficiency (molar ratio of CO_2 uptake as photosynthesis to water loss as transpiration) was lower in the mistletoe leaves. The slope of the photosynthesis to leaf conductance relation (Fig. 1) describes the decrease in CO_2 concentration between the atmosphere and intercellular air spaces; therefore, these results also indicated that intercellular CO_2 concentrations were always higher in the mistletoes than in the hosts.

Carbon isotope discrimination values ($\delta^{13}C$) were measured (*11, 12*) to further evaluate the relative differences in water use efficiency between mistletoes growing on hosts that fix nitrogen and those that do not. Farquhar *et al.* (*13*) proposed that the carbon isotope discrimination ratio is a measure of the average daytime intercellular CO_2 concentration of a leaf, and supporting evidence has been provided by several recent studies (*12, 14*). The carbon isotope discrimination value of host or mistletoe leaf tissue (an integrated long-term estimate) was highly correlated with the measured average intercellular CO_2 concentration during the day for that same tissue (based on single-day observations) (Fig. 2A). Water use efficiency was negatively related to intercellular CO_2, and there was also a significant relation between the carbon isotope ratio and the measured daily water use efficiency of the leaf (Fig. 2B). To further examine these parasitic plants, we extended our observations and used measurements of carbon isotope discrimination ratios from plants at different locations to evaluate the relative differences in water use efficiency between mistletoes and their hosts.

In this analysis we emphasize the difference in water use efficiencies of the mistletoe and its host (differences in carbon isotope ratio between parasite and host) rather than absolute water use effi-





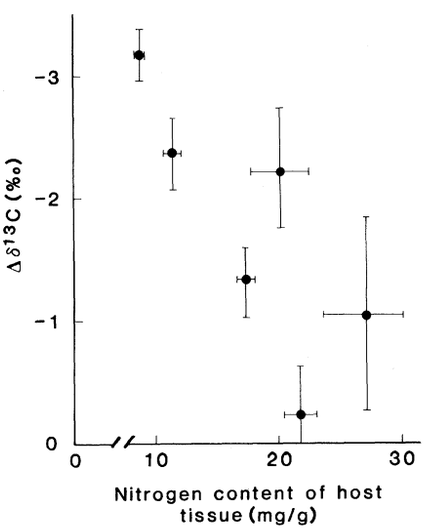


Fig. 1 (left). Relation between daily maximum rate of photosynthesis (A_{max}) and the concomitant leaf conductance to water vapor under field conditions for mistletoes and host plants in central Australia, September 1981. The data were collected by the method of Schulze *et al.* (*19*). Fig. 2 (middle). (A) Relation between carbon isotope ratio and average daytime intercellular CO_2 concentration $[CO_2]_i$ for mistletoes and host plants in central Australia, September 1981. The regression line is $\delta^{13}C = -22.54 - 0.0226[CO_2]_i$ ($P \leq 0.01$). Standard error of the slope is 0.0042. (B) Relation between carbon isotope ratio and daily water use efficiency [molar ratio of photosynthesis (A) to transpiration (E)] for mistletoe and host plants in central Australia. The regression line is $\delta^{13}C = -29.46 + 0.863(A/E)$ ($P \leq 0.05$). Standard error of the slope is 0.227. Fig. 3 (right). Relation of the mean difference in water use efficiency between mistletoes and host plants (as estimated by carbon isotope ratios) and mean nutritional status (as estimated by leaf tissue nitrogen content) for nitrogen-fixing and nonfixing hosts in Australia, South Africa, and the United States. The data are means ± standard errors ($r = 0.783$ and $P \leq 0.05$, one-tailed test).

ciency. This is because plants were sampled from many different locations; the absolute growth rate or water use efficiency may be influenced by site differences in soil water availability and evaporative demand. Negative differences in the carbon isotope ratio values between a mistletoe and its host indicate a negative difference in water use efficiency. A change in water use efficiency may be caused by a lower photosynthetic rate or a greater transpirational rate or both. Measurements were derived from mistletoe-host pairs in arid and semiarid habitats in the western United States (*15*), central Australia (*16*), and South Africa (*17*). For all three continents the difference in carbon isotope ratio between mistletoe and host was smaller if the host was a nitrogen fixer than if the host was not a nitrogen fixer (Table 1). This implies that, in each habitat, water use efficiency of the parasite was closer to that of its host if it grew on a host that had a greater nitrogen supply in the transpiration stream. Included in these comparisons were individuals of the same species growing on both nitrogen-fixing and nonfixing hosts. Hosts differed in carbon isotope ratio by 5.4 per mil depending on habitat. The carbon isotope ratios for the mistletoes differed by 3.1 per mil depending on habitat and host quality.

The leaf-tissue nitrogen contents from mistletoe-host pairs in the United States, Australia, and South Africa were greater when the host was a nitrogen fixer. The differences in water use efficiency between a mistletoe and its host on all three continents appear related to the nutritional quality of the host, since the differences in carbon isotope ratio between parasite and host were strongly correlated with the nitrogen content of the host (Fig. 3) (*18*).

If xylem-tapping mistletoes were provided with a higher rate of nitrogen supply (through a change in hosts or in host quality), we might observe a faster growth rate but not any change in the difference in water use efficiency between host and parasite. The mistletoe could thus be regarded as primarily a water parasite, gathering nutrients passively for growth. We would also expect little or no control of the rate of water loss by mistletoe stomates. A second possibility is that the mistletoe could reduce its transpiration rate through stomatal control or increase its photosynthetic rate (utilizing the increased nitrogen supply to increase photosynthetic protein levels), either of which would result in an improved water use efficiency (closer to that of its host) under conditions of higher nitrogen supply. In such a situation, differential transpiration rates between mistletoe and host could be regarded as a means of nutrient acquisition that was under mistletoe control. Mistletoe stomates do close at night and in response to environmental factors, suggesting a strong control over rates of water loss.

Schulze and Ehleringer (*7*) recently showed that growth rates of *Phoradendron californicum*, a xylem-tapping mistletoe in North America, were higher on nitrogen-fixing host species than on adjacent nonfixing species and that the differences in water use efficiency between mistletoe and host were smallest when the mistletoe grew on nitrogen-fixing hosts. The results of our study greatly extend those initial water-use efficiency observations to a much larger data set from three continents and indicate the importance of nutrient parasitism by xylem-tapping mistletoes (*5*), which are inherently dependent on their hosts for water as well.

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16. The following mistletoe-host pairs were sampled in the central Australian desert: *Amyema cambaquei* on *Casuarina cunninghamiana*, *Amyema gibberulum* on *Grevillea wickhamii*, *A. gibberulum* on *Hakea eyreana*, *Amyema linophyllum* on *Casuarina cristata*, *Amyema mackayense* on *Avicennia marina*, *Amyema maidenii* on *Acacia coriacea*, *A. maidenii* on *Acacia cowleana*, *A. maidenii* on *Acacia kempeana*, *A. maidenii* on *Acacia monticola*, *Amyema miquelii* on *Angophora costata*, *A. miquelii* on *Eucalyptus largiflorens*, *A. miquelii* on *Eucalyptus sideroxylon*, *Amyema quandang* on *Acacia aneura*, *A. quandang* on *Amyema brachystachya*, *A. quandang* on *Acacia papyrocarpa*, *Amyema priessii* on *A. brachystachya*, *Amyema sanquineum* on *Eucalyptus* sp., *Dendrophthoe vitallina* on *Casuarina glauca*, *Diplatia grandibracteata* on *Eucalyptus leucophloia*, *Lysiana casuarinae* on *Gossypium robinsonii*, *L. casuarinae* on *Acacia acradenia*, *Lysiana exocarpi* on *A. aneura*, *L. exocarpi* on *A. brachystachya*, *L. exocarpi* on *Acacia tetragonophylla*, *L. exocarpi* on *Acacia victoriae*, *L. exocarpi* on *Heterodendron oleaefolium*, *L. exocarpi* on *Myoporum platycarpum*, *L. exocarpi* on *Pittosporum phylliroides*, *L. exocarpi* on *Templetonia egena*, *Lysiana murrayi* on *A. aneura*, *L. murrayi* on *A. brachystachya*, *L. murrayi* on *A. kempeana*, *Lysiana spathulata* on *A. aneura*, *L. spathulata* on *A. cowleana*, *L. spathulata* on *A. coriacea*, *L. spathulata* on *Acacia farnesiana*, *L. spathulata* on *A. monticola*, *L. spathulata* on *Callitris columellaris*, *Lysiana subfalcata* on *Atalaya hemiglauca*, *L. subfalcata* on *Cassia oligophylla*, and *L. subfalcata* on *Ceriops tagal*.
17. The following mistletoe-host pairs were sampled in the deserts of South Africa: *Agelanthus discolor* on *Acacia reficiens*, *A. discolor* on *Rhigozum trichotomum*, *Odontella welwitschii* on *Boscia polyantha*, *Phragmanthera glaucocarpa* on *Lannea discolor*, *Septulina glauca* on *Tamarix* sp., *Tapinanthus oleifolius* on *Acacia erubescens*, *T. oleifolius* on *Acacia hereroensis*, *T. oleifolius* on *Euclea pseudobenus*, *T. oleifolius* on *Parkinsonia africana*, *Viscum capense* on *Antizoma miersiana*, *V. capense* on *Boscia foetida*, *V. capense* on *Zygophyllum divaricata*, *Viscum rotundifolium* on *B. foetida*, *V. rotundifolium* on *Ehretia rigida* and *V. rotundifolium* on *Mearua schinzii*.
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