

NITROGEN NUTRITION OF THE 'HASS' AVOCADO: *WHERE DOES ALL THE N GO?*

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Abstract

The avocado fruit is not only rich in fat and oil but also contains a high concentration of protein relative to other fruit. Thus, the avocado fruit is a strong sink for both carbon and nitrogen. When fruit development and vegetative growth are concurrent and, thus, in competition, the distribution, transport and allocation of nitrogen within the mature bearing tree is of importance. Is nitrogen fertilizer allocated according to sink activity existing at the time of fertilizer uptake? To what extent does application of nitrogen fertilizer stimulate sink activity? Is it necessary to time the application of nitrogen to the phenology of the tree?

1. Introduction

A review of the literature regarding the nitrogen economy of the avocado provided two well-documented facts relevant to this topic:

1. In most avocado producing nations, growers fertilize their trees to maintain leaf N concentrations between 2.0 to 2.6%.
2. Not only is the oil-rich avocado fruit a major sink for carbon, it is also a major sink for nitrogen. Avocados have the greatest concentration of protein of any commercially produced deciduous, subtropical or tropical fruit tree crop (Hall et al., 1980). Whereas other fruit average 0.8% protein on a fresh weight basis (FAO, 1970), avocados routinely exceed 2.3% protein per unit fresh weight (Pearson, 1975; Slater et al., 1975; Hall et al., 1980).

In this overview, I have used the California avocado fruit, tree and industry as models. The industry comprises approximately 26 000 hectares, which yield 230 000 metric tonnes annually (Affleck, 1992). Over the last 8 years, yield has averaged 8.8 tonnes per hectare (Affleck, 1992). With the world average being 4 to 8 metric tonnes per hectare, California is typical of other production areas (Wolstenholme, 1987; Barros and Sanchez, 1992; Diaz-Robledo, 1992; Illsley, 1992).

California avocado growers use 84 to 168 kg N per hectare, on average, with many growers far exceeding this rate. At these high rates of nitrogen fertilization, "*Where does all the N go?*" becomes a critical issue because of the potential for excess nitrogen to enter into the groundwater. Although an extremely important topic from the viewpoint of protecting the environment and human health, as well as the wasted dollars to the grower through the loss of fertilizer not used by the tree to produce the crop, nitrate leaching will not be addressed herein. However, the issue is raised in order to remind us of its increasing seriousness. The focus of the overview is on identifying where all the nitrogen goes within the tree.

One of the major nitrogen-containing components of living tissues is protein. The 'Hass' avocado in California averages 2.4 g protein per 100 g fresh weight (Slater et al., 1975). A typical California avocado weighs 200 to 300 g fresh weight (Slater et al., 1975). Thus, there is 5.0 to 7.5 g of protein per avocado fruit, which represents more than 1 g of nitrogen per fresh fruit. (This calculation was based on two factors commonly used for calculating g protein per 100 g tissue by multiplying Kjeldahl N by 6.25 or 5.7; Hall et al., 1980). In contrast to avocado fruit, avocado leaves from common scion varieties average only 4 mg protein per g fresh weight (table 1) (Lovatt and Cheng, 1990). This level of leaf protein is 7.5-fold lower than the protein concentration of citrus leaves and 5-fold lower than that of squash leaves (table 1) (Lovatt and Cheng, 1990). It is of interest that avocado leaves had the lowest percent water (60%) (greatest dry matter content) of the leaves in this comparison. The dry matter content of avocado leaves was confirmed in the present study (table 4). With the exception of G 755 and Toro Canyon, the protein level of avocado rootstocks commonly used in California was more than 2-fold lower than that of two common citrus rootstocks (table 1) (Lovatt and Cheng, 1990).

It is also of interest that the activity of key nitrogen-metabolizing enzymes in avocado scion leaves is typically lower than in the roots of avocado rootstocks (tables 2 and 3). Nitrogen can only be assimilated as ammonia. Thus, nitrate fertilizer taken up by the roots of the avocado must be reduced to ammonia before assimilation can take place. Plants can accumulate large amounts of nitrate in their tissues without reducing it to ammonia. Nitrate accumulates in the vacuoles as a reserve pool, whereas cytoplasmic nitrate is in the "inducing" pool. This pool regulates the activity of nitrate reductase by inducing its synthesis and through substrate regulation. Although nitrate reductase is present in both leaves and roots, in some plants nitrate is reduced preferentially in one organ in comparison to the other. Nitrate can accumulate to concentrations as high as 10% of the plant's dry weight, but on average, plant nitrate concentrations range from 0 to 0.2% dry weight (Fernandes and Rosiello, 1995). For 'Hass' avocado leaves and young actively growing roots, nitrate concentrations were $0.21 \pm 0.02\%$ and $0.18 \pm 0.02\%$, respectively (table 4). Nitrate reductase activity of 'Hass' avocado leaves was significantly lower than that found in the leaves of most other scion varieties, with the exception of 'Bacon', or in the roots of most avocado rootstocks (table 2) (Lovatt and Cheng, 1990). Similarly, the activity of glutamine synthetase, the primary enzyme of ammonia assimilation, was typically lower in leaves of avocado scion varieties, especially 'Hass' and 'Bacon', than in the roots of avocado rootstock varieties (table 3) (Lovatt and Cheng, 1990). It is interesting to note that although nitrate reductase activity tended to decrease in both leaves and roots from the early summer to early fall sampling date, glutamine synthetase activity generally increased in both tissues over the same period. The relatively higher concentration of nitrogen metabolism in the roots than in the leaves of the avocado suggests that the rootstock may be a more important factor in nitrogen nutrition than the scion and, thus, emphasizes the importance of good root health to avocado production.

In order to determine the relative importance of different organs of the avocado tree as sinks for nitrogen, we took apart an avocado tree and had each of the components analyzed for nitrogen so that a model could be constructed illustrating the allocation of nitrogen to each component expressed as kg N per hectare.

The results are not intended to be definitive but instead to be instructive and heuristic. Hopefully, they will stimulate researchers in other avocado-growing areas to examine nitrogen allocation in their orchards. Future research in our laboratory will include an in-depth

investigation of nitrogen allocation in 'Hass' avocado trees during both the " on" and " off " cycles of alternate bearing.

2. Material and methods

An 8-year-old 'Hass' avocado tree on Duke 7 rootstock located at the University of California South Coast Research and Extension Center, Irvine, CA, was extracted from the ground and dissected in September 1995 (September is the standard time for determining tree nitrogen status by leaf analysis in California). The total fresh weight of each component was determined. A weighed sub sample was dried in a forced air oven at 60°C until completely dry and the final weight recorded. Oven-dried samples were ground in a Wiley mill to pass through a 40-mesh screen and analyzed for nitrogen using the standard Kjeldahl method. The results were used to calculate kg N per hectare by the following equation:

$$\begin{array}{c} \text{Kg N} \\ \text{-----} = \\ \text{Ha} \end{array} \quad \begin{array}{c} \text{Calculation of} \\ \\ \\ \end{array}$$

$$\frac{\text{g N}}{\text{g dry wt tissue}} \times \frac{\text{g dry wt tissue}}{\text{g fr wt tissue}} \times \frac{\text{total fr wt tissue}}{\text{tree}} \times \frac{100 \text{ trees}}{\text{ha}}$$

Because of the difficulty of completely recovering all the roots from the soil, the data underestimate root nitrogen costs. Likewise, on a whole tree basis, the data do not include nitrogen costs associated with the loss of pollen, flowers, fruit, or leaves that abscised prior to September 1995.

3. Results

At the time we took the tree apart, it was bearing 67 kg fruit. New shoots represented 1.2 kg fresh weight, leaves 24 kg, small branches less than 2.5 cm in diameter and green in color weighed 41.35 kg, whereas larger branches between 2.5 to 5.0 cm in diameter with brown phelloderm totalled 24.25 kg fresh weight, and scaffolding branches, 70.25 kg. The scion component of the tree trunk weighed 12.1 kg and the rootstock portion of the trunk weighed 17.35 kg fresh weight. Scaffolding roots contributed 11.0 kg fresh weight, small roots 3.3 kg, and fine actively growing roots 0.8 kg. The dry matter content as a percentage of the fresh weight of each tissue is given in table 4. With exception of actively growing root tips, the dry matter content of avocado tissues was greater than 30% (dry weight/fresh weight).

Total nitrogen content was greater in the younger (current year) tissues and in those that were actively growing (table 4). The greater concentrations of nitrate were also observed in these tissues, but in addition scaffolding roots had a significant concentration of nitrate (table 4). It is interesting to note that for both the scion trunk and rootstock trunk, the bark had an approximately 2-fold greater concentration of total nitrogen than the wood and that this ratio was the same with regard to the nitrate content of these two tissues.

Using the equation given in the *Material and methods* section above, the total nitrogen content of each component of the tree was calculated on a fresh weight basis and multiplied by the total biomass of the component to give total N (fresh weight) per tree which was converted to kg N (fresh weight) per hectare (table 5). The 'Hass' avocado stores a significant proportion of its nitrogen in the scion half of the tree. With a 10 to 20% loss in leaves each spring, there is a considerable loss in nitrogen to the individual tree and to the orchard (1.8 to 3.5 kg N/ha), some of which may be reutilized by the tree as the leaf litter decomposes. With a harvest of 10 tonnes of fruit per hectare in a given year, approximately 28 kg N per hectare is removed. If yield is increased from 10 tonnes to 20 tonnes per hectare per year, there will be a total cost of 56 kg N per hectare in the fruit. At 30 tonnes of fruit per hectare per year, the cost is 84 kg N per hectare per year. An annual 20 to 30% increase in vegetative growth costs 14 to 21 kg N per hectare per year.

The time(s) at which nitrogen is in critical demand by the avocado tree is not known. The period of fruit set, which is characterized by competition between young developing fruit and the developing vegetative flush, may be a time when nitrogen is in critical demand. If soil reserves of nitrogen are readily available or if the nitrogen observed to accumulate in small branches is available and singly or in combination can satisfy the tree's requirement for nitrogen at those times that are critical, the timing of nitrogen fertilizer application is not important. However, on the sandy well-drained soils found in some avocado-growing areas, yield might be enhanced by applying nitrogen to the tree at some times but not at others. We have examined this possibility by determining the effect of supplying an extra dose of nitrogen to the tree at key times in its phenology to identify nitrogen fertilization strategies that increase yield. The results thus far from the on-going field experiment suggest that in an "off" year, but not in an "on" year, 'Hass' avocado trees benefit from receiving extra nitrogen in April (table 6). In an "on" year, it appears that extra nitrogen might be more effective if applied in February. The cumulative yields thus far suggest that the November application of additional nitrogen may also be of benefit. While preliminary in nature, the results of this experiment taken together with the results of the within-tree nitrogen-allocation study above indicate that the nitrogen demand of 'Hass' avocado trees is different in "on" and "off" years and suggest that they should be fertilized accordingly.

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Table 1 - Protein content

Plant (tissue)	X ± STD DEV mg protein/g fr wt ² (n = number of experiments)
Avocado scions (leaves)	
Hass	4.0 ± 0.5 (n = 5)
Bacon	4.2 ± 0.3 (n = 5)
Fuerte	4.7 ± 0.3 (n = 5)
Pinkerton	3.5 ± 0.4 (n = 5)
Gwen	4.8 ± 0.9 (n = 5)
Avocado rootstocks (roots)	
Duke 7	0.6 ± 0.5 (n = 4)
Topa Topa	0.7 ± 0.2 (n = 5)
G 755	1.7 ± 0.2 (n = 5)
Borchard	0.6 ± 0.2 (n = 5)
Thomas	0.6 ± 0.2 (n = 5)
Toro Canyon	1.6 ± 0.2 (n = 5)
Citrus scions (leaves)	
Washington navel orange	33.0 ± 4.0 (n = 4)
Valencia sweet orange	31.4 ± 1.2 (n = 4)
Lemon	30.4 ± 2.2 (n = 4)
Grapefruit	27.8 ± 3.5 (n = 4)
Citrus rootstocks (roots)	
Rough lemon	2.9 ± 0.3 (n = 3)
Carrizo citrange	1.7 ± 0.3 (n = 3)
Squash	
Leaves	21.3
Roots	2.1

²Protein determined by the method of Bradford (1976).

Table 2 - Nitrate reductase activity in leaves and roots of avocado scion and rootstock varieties, respectively

nmol nitrate reduced/mg protein/hr ^z		
Variety (tissue)	EARLY SUMMER (June/July)	EARLY FALL (Sept/Oct)
Scions		
Hass	7	3
Bacon	7	2
Fuerte	12	4
Pinkerton	10	27
Gwen	20	<1
Rootstocks		
Duke 7	95	17
Topa Topa	33	30
G 755	27	17
Borchard	36	30
Thomas	32	13
Toro Canyon	15	64

^zNitrate reductase activity was assessed according to the method of Scholl et al.(1994).

Table 3 - Glutamine synthetase activity in leaves and roots of avocado scion and rootstock varieties, respectively

nmol glutamyl hydroxamate synthesized/mg protein/hr ^z		
Variety (tissue)	EARLY SUMMER (June/July)	EARLY FALL (Sept/Oct)
Scions		
Hass	338	5867
Bacon	<1	<1
Fuerte	5773	2130
Pinkerton	2008	9476
Gwen	932	6374
Rootstocks		
Duke 7	10435	17533
Topa Topa	4766	6926
G 755	747	2086
Borchard	7022	7318
Thomas	14757	45603
Toro Canyon	2396	12503

^zGlutamine synthetase activity was assessed according to the method of McCormick et al.(1982).

Table 4 - Dry matter content of 'Hass' avocado tissues expressed as a percentage of fresh weight, leaf total nitrogen and nitrate content as percent dry weight

Tissue	% dry wt	% N	% NO ₃
New shoots	36	2.50 ± 0.14	0.10 ± 0.02
Leaves	40	1.85 ± 0.07	0.21 ± 0.03
Fruit	33		
Seed		0.68 ± 0.07	0.07 ± 0.02
Flesh		0.85 ± 0.07	—
Small branches (≤ 2.5 cm)	38	1.53 ± 0.15	0.11 ± 0.02
Small branches (2.5-5.0 cm)	55	0.78 ± 0.16	0.07 ± 0.01
Scaffolding branches	47	0.46 ± 0.13	0.05 ± 0.01
Scion trunk	48		
Bark		0.77 ± 0.06	0.14 ± 0.02
Wood		0.38 ± 0.03	0.06 ± 0.02
Rootstock trunk	41		
Bark		0.73 ± 0.06	0.12 ± 0.02
Wood		0.31 ± 0.02	0.06 ± 0.01
Scaffolding roots	37	0.77 ± 0.04	0.25 ± 0.04
Small roots	35	0.73 ± 0.14	0.14 ± 0.05
New roots	4.6	1.35 ± 0.20	0.18 ± 0.02

Table 5 - Distribution of N in the 'Hass' avocado tree on a fresh weight basis times 100 trees per hectare

Tissue	kg N/ha
New shoots	1.8
Leaves	17.5
Fruit (100 kg fruit/tree)	28.0
Small branches (ú 5.0 cm)	34.4
Scaffolding branches	15.1
Scion trunk	2.2
Rootstock trunk	2.2
Scaffolding roots	3.1
Small roots	0.8
New roots	0.5
Total	105.6

Table 6 - Effect of applying 56 kg N (1/3 of the total N fertilizer applied) at key times in the phenology of the 'Hass' avocado tree

Double N treatment date	Yr 1	Yr 2	Yr 3	Cumulative yield
	kg fruit/ha			
Control ^z	4.7	13.2	2.1	19.0
	Percent increase (+) or decrease (-) in yield			
Mid-Jan	-20	+5	+22	+5
Mid-Feb	-43	+12	-52	-4
Mid-Apr	+87	-13	+94	+26
Mid-June	-15	-4	-33	-5
Mid-Nov	+66	+20	-23	+27

^zControl trees received 28 kg N/ha as ammonium nitrate in late Jan to early Feb, mid-Apr, mid-June, mid-July, late-Aug to early Sept and late Oct to early Nov.