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^{14}C-assimilate Distribution in *Phaseolus vulgaris* L. during the Reproductive Period[1]

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Abstract. **The distribution of ^{14}C-assimilates was examined in pot-grown 'Redkote' and 'Michelite-62' bean plants in which a lower or upper leaf was dosed with $^{14}CO_2$ at flowering, pod expansion, or pod maturation. Assimilates from the leaf at node 4 moved primarily to the roots at flowering, but were translocated to actively growing pods at later stages. Dosing of the terminal trifoliate of 'Redkote' resulted in radiocarbon transfer exclusively to the subtending pods during pod expansion and maturation. Distribution from leaves on branches of both varieties was restricted to pods on the branch. When the main-stem node-7 leaf of 'Michelite-62' was dosed, 51% of the activity was recovered from node-7 axillary pods, and less from pods at nearby nodes. Thus middle and lower main-stem leaves of beans generally supply assimilates to several centers of active growth, while distribution from upper mainstem and branch leaves is more restricted.**

Although the translocation of organic solutes in bean has been extensively studied (1, 2, 7, 9, 10, 12, 15, 16, 18, 21), only Wanner and Bachofen (18) investigated assimilate distribution after flowering. Such studies have been conducted in other species (3, 11, 17). The lack of detailed sampling in bean prompted us to reexamine the role of the leaves at different nodes in supplying carbohydrates to the pods of the determinate 'Redkote' and indeterminate 'Michelite-62' dry bean.

Materials and Methods

'Redkote' (RK) and 'Michelite-62' (M-62) bean were planted Dec. 31, 1971 in 15 cm styrofoam pots containing 40% #4 vermiculite, 40% peat moss, 10% loam and 10% sand to which 7 g of 18–9–9 Osmocote slow release fertilizer had been added. Plants were grown in a 21–23°C greenhouse and thinned to 1 per pot, 29 days after emergence. They were dosed with $^{14}CO_2$ in a walk-in growth chamber to which the plants were transferred 24 hr before dosing. Temp in the chamber was 24°C and light at plant level averaged 4.4×10^{-2} cals/cm^2/min (400–700 mμ) from fluorescent and incandescent lights. With RK, the central leaflet was enclosed in a cylindrical plexiglass leaf chamber of 500 ml vol (7) connected to a flowmeter, Beckman 315A infrared gas analyzer, diaphragm pump (Dynapump model 2) and a fine-control CO_2 injection system containing a mixture of $^{12}CO_2$ and $^{14}CO_2$ (13). The smaller size of M-62 leaflets allowed entire leaves to be enclosed. $^{14}CO_2$ was generated by adding excess 35% perchloric acid to Ba $^{14}CO_3$ (specific activity 55 mc/mM) in a 25 ml suction flask connected to the gas metering system. By adjusting inflow rate, CO_2 concn was maintained at 300 ± 10 ppm as measured by the infrared gas analyzer. Leaves of 4 plants were dosed simultaneously in 4 leaf chambers connected in parallel. Dosing time, adjusted for uptake of 200 μCi of $^{14}CO_2$ by the 4 leaves, was about 2 hr. After dosing, the plants were returned to a greenhouse and kept at 20–24°C and 4.95×10^{-2} cals/cm^2/min (400–700 mμ) light from warm white fluorescent fixtures for 24 hr from the start of dosing. The 24 hr photoperiod after dosing was necessary to prevent confounding translocation patterns with time-of-day effects (14), since dosing of the plants was done throughout the day. Plants were cut at soil level, subdivided into stem, leaf and axillary stem and reproductive tissue, dried at 70°C in a forced air drier, ground in a Wiley mill to pass a 60 mesh screen, and after thorough mixing, weighed out in triplicate samples of 5 mg. The samples were suspended in 10 ml of a thixotropic gel counting cocktail [40 gm cabosil (silicon dioxide thixotropic gel powder), 4 gm PPO (2,5 diphenyl oxazole), and 250 mg dimethyl POPOP (1, 4 bis (2-(4-methyl-5-phenyl oxizolyl)-benzene) in 1 liter toluene] and counted in a Packard Tricarb liquid scintillation counter. Sample counts were quench corrected by adding 270,000 dpm ^{14}C as K-acetate-^{14}C to increasing quantities of ground bean tissue in 10 ml of gel cocktail. Quenching averaged 30 to 70%, with stems having the least quenching. Hypocotyls were recovered and included in the stem samples.

The relative activity of the plant parts (cpm/5mg) was expressed as the % of the total recovered specific activity of all parts. Similarly, the % of total recovered activity for each part was determined by multiplying specific activity of each part by its dry wt. Plants were dosed at flowering (37 and 44 days after planting for RK and M-62 respectively), when pods were full length with growing seeds (46 and 65 days, respectively) and when seeds were full size and pod wall sensecence had started (67 and 79 days after planting, respectively). For RK, the trifoliate leaf at node 4 was dosed at all three maturity stages. The terminal leaf of the main stem at node 6, which subtends the first flowers to open, was also dosed at the 2 later maturities. A more rapid senescence of M-62 leaves prevented dosing of leaves at these same nodes. The trifoliate at node 4 was dosed at flowering, but senesced soon afterwards. Therefore, the trifoliate leaf at node 2 of the axillary branch at node 3 was substituted at the mid and late maturities. The leaf at node 7 of M-62, which subtends the first flowers to open on the main stem, was also dosed at these maturities. For comparison with the dosing of a leaf on an axillary branch of M-62, the leaf on the branch at node 4 of RK was dosed at 51 days after planting. Eight plants of each cultivar and leaf position were dosed at each maturity except the last, when only 4 plants of M-62 were used per leaf position.

Two plants from each maturity and leaf-dosed treatment were subdivided into 4 or 5 sections, including some roots, 24 hr after dosing, freeze dried in a Virtis tray freeze drier, pressed and autoradiographed for 48 hr on Kodak Blue Brand X-ray film.

Results

'Redkote' plants had 6 nodes at maturity, including the cotyledonary node. Under the low winter light intensities of the greenhouse, they developed no branches at node 2, where the unifoliate leaves arise. There was a branch bearing one trifoliate leaf and 1 to 3 pods at nodes 3 and 4, a very small branch or small, poorly developed pods at node 5 and 2 or 3 well-developed

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Table 1. Distribution of recovered activity in 'Redkote' and 'Michelite-62' plants dosed at node 4 with $^{14}CO_2$ at flowering, pod expansion and pod maturity.

Plant part	Recovered activity (%)			
	Redkote		Michelite-62	
	Flowering	Pod expansion	Pod expansion	Flowering
Treated leaf	54.3 ± 4.5[z]	28.0 ± 2.3	38.7 ± 3.9	73.3 ± 4.9
Untreated tissue at treated node	23.2 ± 1.9	38.4 ± 3.9	33.6 ± 9.3	6.3 ± 0.9
Tissue above treated node	1.3 ± 0.4	10.2 ± 2.6	11.2 ± 6.1	2.5 ± 0.7
Tissue below treated node	1.7 ± 0.8	14.1 ± 3.0	7.6 ± 1.6	2.6 ± 0.6
Hypocotyl	19.6 ± 3.7	9.4 ± 2.2	8.4 ± 1.5	15.2 ± 3.6

[z]SE.

Table 2. Distribution of recovered activity in 'Redkote' and 'Michelite-62' plants dosed at node 6 and node 7 respectively with $^{14}CO_2$ at pod expansion and pod maturity.

Plant part	Recovered activity (%)			
	Redkote		Michelite-62	
	Pod expansion	Pod maturity	Pod expansion	Pod maturity
Treated leaf	21.6 ± 3.0[z]	27.8 ± 5.7	24.1 ± 3.1	59.6
Untreated tissue at treated node	70.6 ± 3.0	56.0 ± 6.8	52.2 ± 5.7	7.0
Tissue above treated node	Top node treated		9.0 ± 6.5	2.3
Tissue below treated node	7.8 ± 1.2	16.2 ± 6.0	14.6 ± 5.2	31.1

[z]SE.

pods in the axil of the terminal node 6. The flowers at node 6 usually opened first, but flowering at the other main-stem and branch nodes soon followed so that most pods were at the same stage of development, as previously described by Jones (6).

When RK plants were dosed at the node-4 leaf during flowering more than half of the ^{14}C assimilate remained in the treated leaflet (Table 1). About 20% of the recovered activity was in tissues below the treated node, and less than 2% was above it. Autoradiographs indicated that the roots received considerable assimilate. Mature leaves other than those dosed seldom contained more than 0.5% of the total recovered activity. When the pods were full length (9 days after flowering), only 28% of the assimilate remained in the treated leaflet, and 23% went to lower tissues. The roots received considerably less assimilate than at flowering; 45% of the assimilate was translocated to the branches, with most of it going to the fruit. Specific activity was always highest for the branch in the axil of the treated node-4 leaf (Fig. 1). Pod development and specific activity at node 5 were uniformly small. If a plant had equal pod number at nodes 3 and 6, specific activity of the node-3 branch was higher (Fig. 1, plant 2). More pods at node 6 than 3 usually resulted in greater translocation to the upper nodes (Fig. 1, plants 1, 3, 6). Distribution of ^{14}C from node 4 during pod maturation was similar to that during pod enlargement (Table 1) except that the amount retained by the treated leaf was higher than at pod expansion.

M-62 plants typically had 10 to 12 main-stem nodes. Branches arose at nodes 2 to 5, being largest at node 3, where they had 3 to 5 nodes with pods at 1 or 2 axils. Branches at main-stem nodes 2 and 4 were usually 2 nodes long with small leaves and no pods. The branch at node 5 had 1 node and no pods, as sometimes occurred at node 6 although the latter would sometimes bear a large raceme with 1 or 2 well-developed pods. All dosed plants had pods in the axil of node 7, and in 1 or more upper main-stem axils.

When the node-4 leaf of M-62 was dosed at flowering, the translocated ^{14}C assimilates went primarily to the stem below node 4 and to the roots (Table 1). These tissues accounted for 18% of the recovered activity. The node-4 leaf was yellowing at the time of dosing, and retained 73% of the assimilate.

Dosing of the node-7 leaf of M-62 during pod expansion resulted in translocation of 52% of the assimilate to untreated tissues at that node (Table 2), primarily to the axillary pods (51.4%). Tissue above and below the treated node received

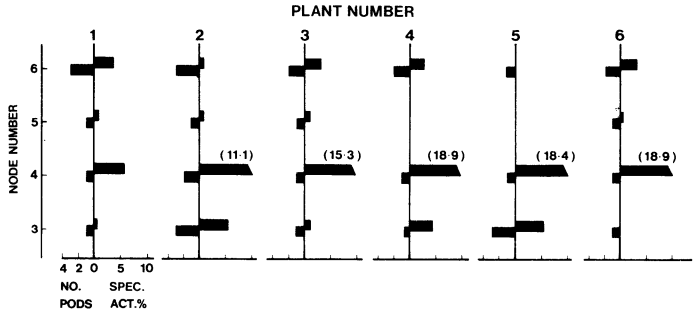

Fig. 1. Distribution of activity in main stem axils as related to pod numbers for individual nodes of 6 'Redkote' plants dosed at the node-4 leaf 46 days after planting.

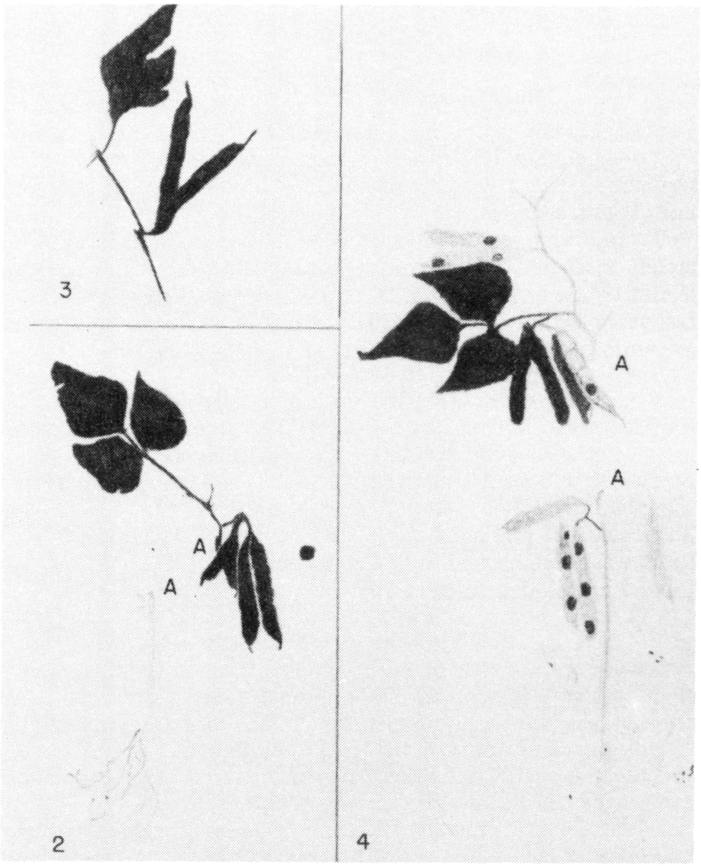

Fig. 2-4. Reproduction of composite autoradiographs showing ^{14}C distribution in 2 cultivars of beans 24 hr after dosing. Letters denote continuation of main stem. Fig. 2. Node 7 leaf of 'Mich. 62' dosed 65 days after planting. Fig. 3. Node 6 leaf of 'Redkote' dosed 46 days after planting. Fig. 4. Leaf 2 on branch 3 of 'Mich. 62' dosed 65 days after planting.

Table 3. Distribution of recovered activity in 'Michelite-62' plants dosed with $^{14}CO_2$ at the leaf node 2 of the branch at node 3, at pod expansion and pod maturity.

Node no.	Plant part	Recovered activity (%) Pod expansion	Pod maturity
8–11	axil	0.3	3.76
	stem	0.1	0.1
6,7	axil	0.5	–
	stem	–	0.18
4,5	axil	0.1	0.39
	stem	0.1	0.12
3, branch	dosed leaf	20.8	47.88
	leaves not dosed	0.1	0.67
	axil	75.0	23.80
	stem	0.8	2.06
1–3	axil	0.6	0.16
	stem and hypocotyl	1.4	17.14

almost equal amounts of ^{14}C assimilates, and 24% remained in the treated leaf. Very little ^{14}C was recovered in the vegetative apex of the indeterminate M-62 (Fig. 2). Leaves above node 7 may supply the apical meristem, but vegetative growth, as shown by production of new nodes, ceased completely during the pod expansion stage. As the pods approached maturity, translocation to pods at node 7 nearly ceased and considerable ^{14}C was translocated to axillary growth at nodes 2 and 3 and to the roots, and a much increased proportion (60%) of the activity remained in the dosed leaf.

Dosing of the node-6 leaf of RK at pod expansion resulted in transfer of ^{14}C exclusively to pods in the axil of that leaf (Table 2, Fig. 3); only traces of activity were present in the rest of the plant. Similar distribution occurred with dosing at pod maturation, except for slighly increased translocation to tissues below the treated node. Retention of activity by the treated leaflet was about 22% at pod expansion, and increased to 28% as the pods approached maturity.

To test whether translocation from leaves on branches is also largely restricted to the axillary pods of the fed leaf, the central leaflet of the branch leaf at node 4 was dosed. We found 63% of recovered activity in the axillary pods 24 hr later. Only trace amounts of activity were recovered from the rest of the plant and 24% remained in the treated leaflet.

A similar pattern occurred with M-62. Assimilate translocation from the node-2 leaf on the axillary branch at main stem node 3 was largely mobilized (75%) by pods on the same branch, with only 1% going to the roots, and 3% to the rest of the plant (Fig. 4, Table 3). The other 21% remained in the dosed leaf. At pod maturation, distribution of assimilates to the roots was reestablished (17%) and mobilization by pods on the treated branch decreased to 24%.

Discussion

Leaves on branches of both cultivars, and the terminal mainstem leaf of RK supplied carbohydrates only to pods on the same branch during pod expansion. If this pattern of assimilate distribution also occurs in the field, it implies that branch leaves and the terminal leaf on the main stem, which are in the upper part of the leaf canopy particularly at close spacing (19), are the main suppliers of assimilates to their subtending pods. Although lower leaves such as the one at node 4 also translocate assimilate to both the axillary branch at their own node and to the pods at node 6 (Table 1), shading would minimize their contribution at close spacings in the field. Since axillary branch and terminal main stem leaves contribute only traces of assimilates to the roots, a reduction in root growth and metabolism may occur during reproductive growth, especially at close spacings. Improved light penetration into the canopy brought about by more acute leaf orientation in relation to incident sunlight (20) could increase assimilate transport to the roots and improve growth at high plant populations.

The pattern of assimilate distribution in beans before flowering shows lower leaves supplying assimilates to the roots, upper leaves translocating mainly to the young, expanding leaves at the apex, and the middle leaves supplying both ends of the plant (2, 21). The predominantly downward transport held true at flowering for both cultivars used here. During pod development, however, assimilates were mobilized by the expanding pods and developing seedsk, although slight downward transport from the node-4 leaf still occurred. Appreciable upward translocation occurred only when "demand," i.e. number of developing pods relative to the rest of the plant, was high. The pods in the axils of different nodes thus seem to be competing for a limited supply of assimilates in a manner similar to the two shoots of the modified pea plants of Lovell (8).

The proportion of absorbed radioactivity retained by the fed leaf varied with leaf age, declining in RK from flowering to pod expansion and increasing again toward maturity (Tables 1, 2). Similar results have been reported for other crops (3, 8, 11, 17). Aside from leaf age effects, the changing mobilizing capability of the reproductive organs may also have been involved. The latter is thought to be mediated by growth hormones, although it is not clear whether the hormones act by stimulating protein synthesis at the point of action or by affecting the translocation pathway (4, 5, 10). The capacity of the node-4 leaf of RK to supply several sinks simultaneously, with relative mobilization depending on pod number, suggests a useful system for elucidating source-sink mechanisms of intact bean plants.

Literature Cited

1. Bachofen, R. and H. Wanner. 1962. Transport und Verteilung von markierten Assimilaten. IV. Direkte Messung der Transportgeschwindigkeit mit Isotopen. *Planta* 58:610-618.
2. Biddulph, O. and R. Cory. 1965. Translocation of ^{14}C metabolites in the phloem of the bean plant. *Plant Physiol.* 40:119-129.
3. Blomquist, R. V. and C. A. Kust. 1971. Translocation pattern of soybeans as affected by growth substances and maturity. *Crop Sci.* 11:390-393.
4. Davies, C. R. and P. F. Wareing. 1965. Auxin-directed transport of radiophosphorus in stems. *Planta* 65:139-156.
5. Hew, C. S., C. D. Nelson, and G. Krotkov. 1967. Hormonal control of translocation of photosynthetically assimilated ^{14}C in young soybean plants. *Amer. J. Bot.* 54:252-256.
6. Jones, L. H. 1967. Factors affecting the productivity of dwarf French beans. *Agr. Progress* 42:32-52.
7. Liu, P. S. 1971. The influence of translocation on photosynthetic efficiency of *Phaseolus vulgaris* L. PhD thesis, Cornell University, [*Diss. Abstr.* 32:118-B.]
8. Lovell, P. H. 1969. Interrelationships of cotyledonary shoots in *Pisum sativum*. *Physiol. Plant.* 22:506-515.
9. Mullins, M. G. 1970. Transport of ^{14}C-assimilates in seedlings of *Phaseolus vulgaris* L. in relation to vascular anatomy. *Ann. Bot.* 34:889-896.
10. _____. 1970. Hormone - directed transport of assimilates in decapitated internodes of *Phaseolus vulgaris* L. *Ann. Bot.* 34:897-909.
11. Palmer, A. F. E., G. H. Heichel, and R. B. Musgrave, 1973. Patterns of translocation, respiration loss, and redistribution of ^{14}C in maize labelled after flowering. *Crop Sci.* 13:371-376.
12. Plaut, Z. and L. Reinhold. 1969. Concomitant photosynthesis implicated in the light effect on translocation in bean plants. *Austral. J. Biol. Sci.* 22:1105-1111.
13. Redshaw, A. J. and H. Meidner. 1970. A fine-control carbon dioxide injection circuit for measurement of rates of net photosynthesis by infrared gas analysis. *J. Expt. Bot.* 21:410-413.
14. Russell, W. J. and D. R. Johnson. 1975. Translocation patterns in soybeans exposed to $^{14}CO_2$ at four different time periods of the day. *Crop Sci.* 15:75-77.
15. Seth A. K. and P. F. Wareing. 1967. Hormone-directed transport of metabolites and its possible role in plant senescence. *J. Expt. Bot.*

18:55-77.
16. Stark, Z. 1969. Pattern of ^{14}C-assimilate distribution in relation to their supply and demand in bean plants. *Act. Soc. Bot. Pol.* 38:583-600.
17. Pierre, J. C. and M. J. Wright. 1972. Distribution of ^{14}C photosynthates in timothy (*Phleum pratense* L.) during the vegetative stage of growth. *Crop Sci.* 12:191-194.
18. Wanner, H. and R. Bachofen. 1961. Transport and Verteilung von markierten Assimilaten. *Planta* 57:531-542.
19. Wien, H. C. 1971. The influence of spacing on light interception, growth and seed yield of *Phaseolus vulgaris* L. under temperate and tropical conditions. PhD Thesis. Cornell University. 183 p. University microfilms, Ann Arbor, Mich. [*Diss. Abstr.* 32:4965-B.]
20. _____and D. H. Wallace. 1973. Light-induced leaflet orientation in *Phaseolus vulgaris* L. *Crop Sci.* 13:721-724.
21. Willenbrink, J. 1966. Transport ^{14}C-markierter Assimilate in Phloem von *Pelargonium zonale* und *Phaseolus vulgaris*. *Planta* 71:171-183.

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Reduction of Peel Roughness of 'Shamouti' Orange with Growth Regulators[1]

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Abstract. **Excessive rough and thick peel, linked with large fruit sizes is found in 'Shamouti' oranges (*Citrus sinensis* (L.) Osbeck) grown under marginal soil and climatic conditions. This condition was overcome by early (April-May) sprays of succinic acid-2,2-dimethylhydrazide (SADH) and 2-chloroethyltrimethylammonium chloride (CCC, chlormequat), probably counteracting high endogenous promotors found in rough tissues. While SADH is too expensive to be profitable and it usually decreases fruit size, CCC can be used without this effect and is commercially rewarding.**

Some fruits of 'Shamouti' orange do not meet export standards when produced under conditions that cause them to be excessively large, thick-peeled and rough. Heavy soil, relatively arid climate and sour orange (*Citrus aurantium* L.) rootstock reportedly contribute to the development of substandard fruit.

The rough peel disorder begins to develop at a very early stage of fruitlet growth (13) and consists of excessive cell divisions and cell growth, mainly of external albedo layers (3). Peel thickness does not decrease after its early peak (1, 5), 2 months after petal fall, but remains the same, or further increases toward maturity. Clear differences in growth dynamics were detected between smooth and rough fruits, conducive to thicker peel and a smaller volume of pulp and juice in rough fruits, for fruits of equal size (3). Studies of the native growth regulators (2) showed higher cytokinin and gibberellin content in both albedo and flavedo of rough fruit at the time of maximum peel growth in June and also at early maturity in November. The higher levels of promotors were considered a decisive factor in the excessive peel growth producing rough fruits. It was therefore decided to test whether growth retardants could counteract them, as described in the following.

Materials and Methods

Experiments were carried out during 5 successive seasons at different places in the eastern portion of the coastal citrus belt of Israel where rough fruit is most common. This area is 12 to 25 km from the sea and has a semi-arid climate. It also has heavier alluvial soils than desirable for the 'Shamouti' orange. The following materials were screened in 1971 and 1972. SADH, CCC, and ammonium (5-hydroxycarvacryl) trimethyl chloride piperidine carboxylate (AMO-1618) were selected for their growth retarding effects. Benzothiazole-2-oxyacetate (BTOA), 2,4-dichlorophenoxy acetic acid (2,4-D) and gibberellin A_3 (GA) were applied to determine whether growth promotors would increase peel roughness and thickness. In subsequent years only SADH and CCC were used. In all cases 0.025% nonionic CIBA wetting agent (octylphenol-octaglycol ether) was added. Dates, doses and no. of trees per treatment are shown in tables. Fruits were randomly picked from experimental trees. Roughness was evaluated visually on a scale of 5 (1 = smooth to 5 = very rough, see Fig. 1); fruit

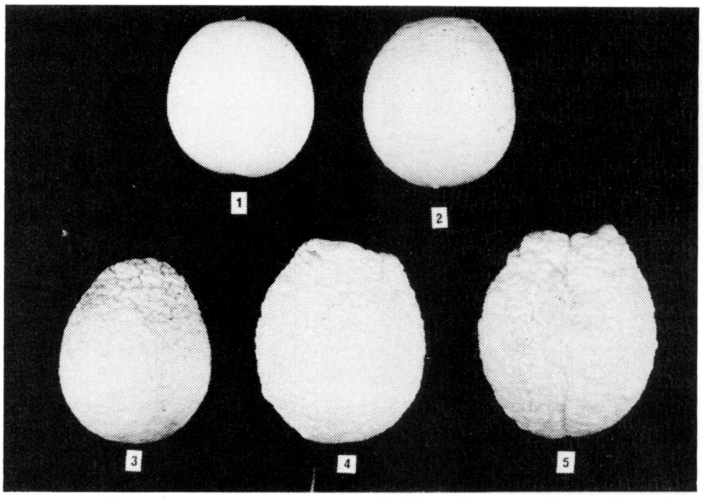

Fig. 1. Roughness ranks 1 to 5.

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