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Some Effects of Ringing, Defoliation, Sorbitol-spraying, and Bending of Shoots on Rate of Translocation, on Accumulation, and on Growth in Apple Trees

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Abstract

Young apple trees *(Malus × domestica)* were ringed, defoliated, or sprayed with sorbitol. The distribution of ^{14}C after exposure to $^{14}CO_2$, the concentration of sugars and the subsequent growth was measured. Ringing increased accumulation of ^{14}C and sugars in the leaves or the top of the tree, but reduced sugar concentration in the lower part of the trunk. The sugar concentration of the top of the tree was reduced following defoliation in September or October. Ringing and defoliation in September reduced root growth and to a smaller extent growth of trunk and branches. The total spring growth of new shoots and leaves showed no clear effect of differences in accumulation and growth in the previous autumn. Sorbitol-spraying in the autumn did not affect either sugar content or the following year's new growth. Bending of current year's shoots in the summer had only a slight effect on the distribution of ^{14}C and the accumulation of sugar in the leaves.

Introduction

In the autumn, apple trees accumulate reserves which disappear again in the following spring (Hansen 1967 b). However, the importance of the reserves for the new growth, and in particular for the extent of it, is not very clear. It is occasionally claimed to be considerable (Kozlowski & Keller 1966). However, attempts at establishing different levels of reserves by ringing in the autumn produced no differences in new shoot growth in the following year (Priestley 1964). The present study attempts to elucidate this problem, by seeking to vary the amounts of reserves not only by ringing, but also by defoliation and sorbitol-spraying in the autumn. Sorbitol was chosen because it appears to play a central part in the carbohydrate metabolism of apple trees (Hansen 1970). Sorbitol and sugar analyses were made, as these substances appear to be chiefly responsible for the fluctuations in the reserves (Hansen 1967 b) The distribution of assimilates within the tree after ringing is illustrated by exposure to $^{14}CO_2$. This method was also used in an attempt to illustrate the effects of ringing and of bending of shoots on the rate of translocation out of the leaves and on the accumulation in them. Ringing and bending have long been known to be measures affecting growth and development in trees (Priestley 1962, Jonkers 1967).

Material and methods

Experiment 1

Six two-year-old specimens *Malus × domestica* 'Spartan' growing well were ringed on July 13, 1967 by removal of an approximately 2 mm wide bark ring from the lower part of the trunk. The growing current year's shoots of 6 similar specimens were bent near the shoot base by means of special hooks into a horizontal position, while 6 non-treated specimens were used as controls.

Six different vigorous current year's shoots from each treatment were exposed to 5 μCi $^{14}CO_2$ per shoot on July 16, July 28, or August 16. The leaves were exposed for 4–6 hours inside plastic bags tied around the shoots (Hansen 1967 a). The exposed shoots were harvested 3 days and 1 month, respectively, after exposure, and the leaves were dried at 80 ° C.

Experiment 2

Two-year-old specimens of 'Cortland', growing well, were divided into 3 batches each containing 13 trees. On June 27 1969 the trees of one batch were ringed, while the current year's shoots of another were bent horizontally as above. On July 7 and 24, respectively, each of ten vigorous shoots per batch were exposed to 5 μCi of ^{14}C applied to no.s 7–9 of the large fully developed leaves as counted from the base of the shoot. For both dates the shoots were harvested 3 days after exposure to ^{14}C, individual shoots being cut into sections and dried. On September 4 samples of bark were taken from the upper parts of the trunk of the trees used for the different treatments.

Experiment 3

One-year-old specimens were used of rootstock type E M II, planted in pots in the spring of 1969. Throughout the summer the trees were watered with nutrient solution and grew well. In September they were divided into 6 uniform batches each of 18 specimens which were treated according to the following plan.

R_1	early ringing	(September 7)
R_2	late ringing	(October 4)
D_1	early defoliation	(September 7)
D_2	late defoliation	(October 4)
S	sorbitol-spraying, 4 times during October with an 8 % aqueous solution, begun October 6	
C	control (untreated)	

The ringing procedure consisted of the removal of an approximately 1 mm wide ring of bark from the lower part of the trunk. Defoliated specimens had all their leaves removed.

The trees wintered in a ventilated greenhouse. On 4–18 December 9 specimens from each batch were harvested, the top was separated from the lower part of the trunk, at a place corresponding to the position of the ringing. Each part was then divided into bark and wood, and the roots were also kept separate. Representative samples of 5–7 grammes of the fresh bark and root material were taken, and combined for each 3 trees for sugar analysis. The remainder was dried.

The remaining trees were harvested 2–9 July in a manner similar to the December harvesting.

The leaves of some of the ringed and untreated trees were exposed to $^{14}CO_2$ either on 11 and 23 September, or on 6 and 20 October. After sampling in December the ^{14}C-activity was measured in the extracts.

Analyses

The ^{14}C-activity in the dried and ground samples was determined by measuring bricks in a Frieseke and Hoepfner window-less methane-flow counter. A relative measure of the total amount of ^{14}C in the sample may be obtained by multiplying the result of the count, after correcting for background, with the dry weight of the sample in grammes (tc = total counts, Hansen 1967 a). For the method used here the tc value for total ^{14}C taken up will be around 15000 per 5 μCi of ^{14}C applied.

Representative samples of the fresh materiael were extracted with boiling 80 % aqueous methanol, and the amounts of glucose, sucrose, and sorbitol were determined following separation by paper chromatography (Hansen 1967 a).

The ^{14}C-activity in extracts after methanol extraction was determined by measurement of up to 1.5 ml of extract in 10 ml of toluene + triton X-100 (2:1) containing Beckman fluoralloy TLA in a Beckman liquid scintillation counter. Correction was made for »quenching«, and the results expressed in disintegrations per minute (dpm).

Table 1. ^{14}C-activity (total counts) in different materials of $^{14}CO_2$-exposed extension shoots following different treatments

*deviates significantly from control (95% level).

Exper.	Date for			Material	Treatment		
	treatment	^{14}C-expos.	sampling		control	bent	ringed
1	13. VII	16. VII	19. VII	exp. leaves a)	8710	9150	10160
		»	19. VIII	» »	4930	6920*	7100*
		28. VII	31. VII	» »	6390	7840	11940*
		»	28. VIII	» »	4000	5300	5590
		16. VIII	19. VIII	» »	6530	6180	12240*
		»	16. IX	» »	3960	4120	8720
2	27. VI	10. VII	13. VII	exp. leaves a)	3954	3635	7024*
		24. VII	27. VII	» »	4280	5673*	8012*
		10. VII	13. VII	low. shoot b)	1437	1685	202*
		24. VII	27. VII	» »	1286	1555*	331*
		10. VII	13. VII	mid. shoot c)	1544	1162	2003
		24. VII	27. VII	» »	1550	1590	3264*
		10. VII	13. VII	up. shoot d)	2574	2424	2999
		24. VII	27. VII	» »	90	70	1182*

a) exp. 1: all shoot leaves; exp. 2: leaf no. s. 7-9 from shoot base. b) 1-6 internode. c) 7-11 internode. d) remaining internodes.

Results

I. Effect on translocation of ^{14}C.

The transport out of the leaves of the photosynthates is considerably reduced by ringing (Table 1), although not very obviously for the first week following ringing. Further, ^{14}C is accumulated in the middle and upper parts of shoots, whereas the content in the lower parts is low. Only in a couple of cases was there an increased ^{14}C-content in the exposed leaves following bending of the shoots, also there was a tendency to accumulation of ^{14}C in the lower parts of shoots.

The distribution of ^{14}C supplied in the autumn as $^{14}CO_2$ to the upper leaves of ringed and control trees in Experiment 3 shows clearly that the ringing causes an almost complete suspension of the translocation of photosynthates to the lower regions of the tree (Table 2).

II. Effect on accumulation of carbohydrates

A. Leaves and bark (summer)

The reduction in transport of photosynthates out of the leaves following ringing shows itself also by an accumulation of certain constituents of the sugar fraction in leaves, and probably also in the bark of the upper

Table 2. Methanol soluble ^{14}C-activity (total dpm) in different tissues of ringed and untreated trees after exposure to $^{14}CO_2$

Experiment 3, December sampling of 3 trees exposed to 30 μCi $^{14}CO_2$

Ringing	Sept.		Oct.	Control		
^{14}C-exposure	Sept.	Oct.	Oct.	Sept.	Sept. +Oct.	Oct.
Upper bark	422	985	996	124	424	479
Lower bark	10	4	15	162	228	210
Root	6	3	15	143	214	282

parts of the trunk (Table 3). However, the accumulation occurred in varying degrees, and to some extent different compounds were accumulated in the two exeperiments. The percentage of leaf dry matter of fresh weight increased after ringing (46.3 and 45.3 per cent as against 39.8 and 38.4 in remaining trees in Experiments 1 and 2, respectively). The leaves of the ringed trees acquired a strong crimson-purple hue.

No accumulation effect was observed in the leaves following bending, on the contrary, almost the opposite appeared to be the case (Table 3).

trunk. The sucrose concentration of the lower bark is similarly reduced after defoliation in September. Dry matter as a percentage of fresh weight is reduced after defoliation (42.8 and 23.1 for bark and root, respectively, as against 48.5 and 26.3 as average values for remaining trees).

The total sugar contents of bark and root (Fig. 2 a), based on concentration and dry matter quantity, are lowest after defoliation and ringing in September, chiefly due to the reduction in root volume following these two types of treatment (see following section). The sugar content following sorbitol-spraying does not deviate significantly from that of untreated control specimens.

Table 3. The concentrations of sugars in different materials following different treatments

Expressed in per cent of methanol (80%) - insoluble residue.

Exper.	Material	Sugar	Treatment control	 bent	 ringed
		glucose	3.00	2.36*	3.65*
1	Leaves[a])	sucrose	2.15	2.03	2.42
		sorbitol	14.4	14.7	16.9*
		glucose	2.05	1.77	3.82*
2	Leaves[b])	sucrose	3.05	3.05	5.31*
		sorbitol	15.6	11.7	12.2
		glucose.....	3.08	2.30	4.22
2	Bark[c])	sucrose.....	1.94	1.58	3.32
		sorbitol	4.42	3.58	7.30

[a]) leaves from current year's shoots sampled August 19 and 21.
[b]) » » » » » » July 13 and 27.
[c]) bark from upper trunk September 4.

B. Trunk and root after various autumn treatments (Experiment 3). December

After ringing the sugar concentrations tend to increase in the bark above the ring, whereas the bark below the ring shows a reduction of its sucrose-concentration. In the root the over-all concentration is increased after ringing in October (Figure 1). For the roots defoliation presents a somewhat similar case. Otherwise defoliation reduces the concentration in the sugar fraction of the upper bark of the trunk.

July. There were no significant differences in the sugar content of the analyzed bark (average 585 mmol sugar + sorbitol/kg of insoluble residue, standard deviation per tree 50).

III. Effect on growth (Experiment 3).

The total dry matter content of the tree is reduced following ringing and defoliation, in

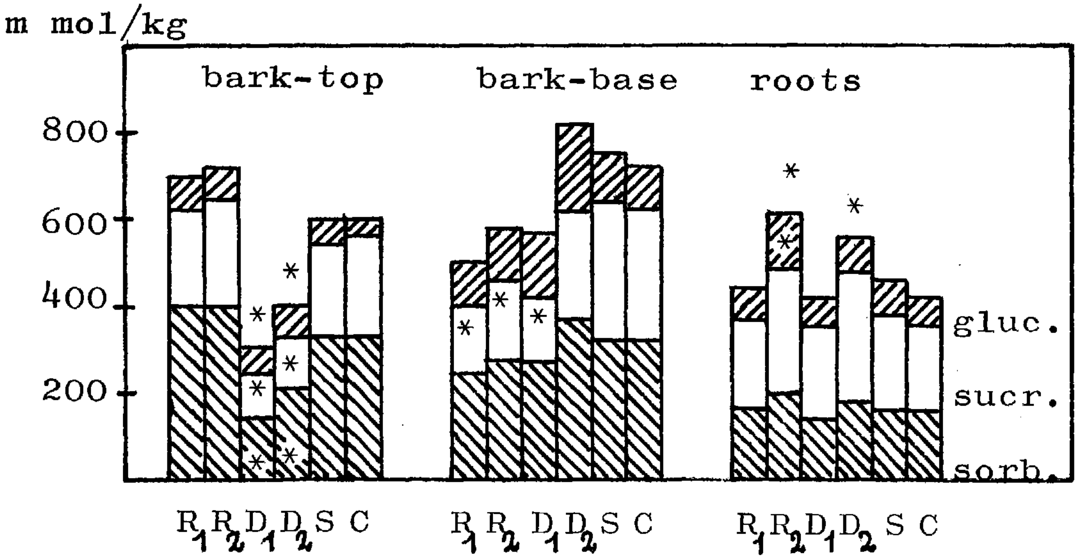

Figure 1. *The concentrations of sorbitol, sucrose and glucose (mmol per kg. methanol - insoluble residue) in different tissues in December after different treatments.* R_1 and R_2 = ringing in Sept. and Oct., respectively, D_1 and D_2 = defoliation in Sept. and Oct., respectively, S = sorbitol-spraying in Oct., C = control. Experiment 3, average of 9 trees. *indicates a significant deviation from the control (95% level).

particular after treatment in September (Fig. 2 b). The reduction is relatively greatest in the root. Of a total of 18 ringed specimens 5 were found to be dead in the following spring, whereas no trees were lost from the other treatments.

When sampling in July there is still a distinct effect of some of the autumn treatments on certain organs of the tree (Fig. 2 c). However, there appears to have been some levelling of the differences. Thus the root volume which was already reduced after early ringing and early defoliation decreased less during the winter than did the root volume in the other treatment groups (cf. Quinlan 1969). The differences in the amount of new growth are also slight compared to the preceding effects on the trees in the previous autumn. Only the woody parts of the new shoots in the upper parts of the trees are less after ringing than after the other treatments. On the other hand there is here, contrary to the other treatments, a clear additional shoot growth on the lower parts of the trunk below the ringing.

Autumn sorbitol-spraying had no apparent effect on the subsequent growth reactions.

Discussion

The present results are in agreement with experiments that show, that ringing or other means of suspending or reducing the activity of the phloem, 1) reduce the rate of translocation from the leaves, 2) cause accumulation of carbohydrates above the position of the ringing), 3) affect the sieve tube concentration, and 4) change the distribution of assimilates (Wiggans 1918, Greene 1937, Bolsunov 1955, Nekrasova 1958, Zimmermann 1960, Kato & Ito 1962, Hartt 1963, Neales & Incoll 1968, Noel 1970). In the present study bending of shoots or branches, which is a far gentler type of treatment than ringing, caused only a tendency to a reduction of the transport out of the leaves. Mika (1969) exposed a single leaf on a shoot to $^{14}CO_2$ immi-

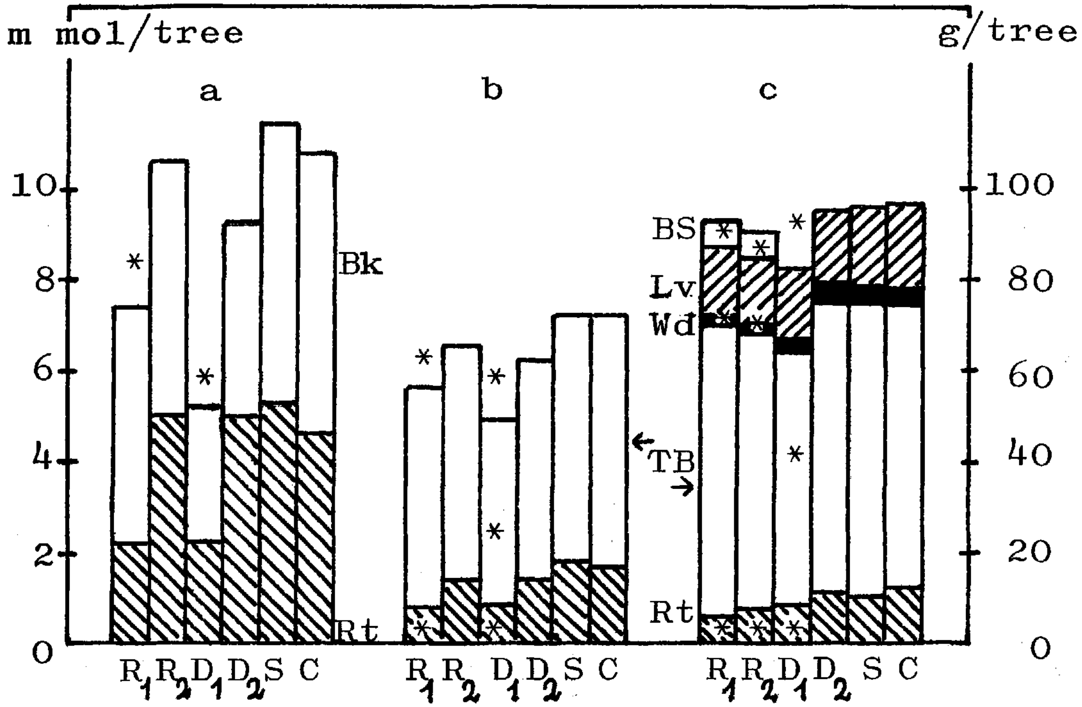

Figure 2. *Sorbitol and sugar contents (mmol/tree) in December (a) and dry weight (g/tree) in December (b) and July (c), after different autumn treatments.* For symbols see Fig. 1. Experiment 3, average of 9 trees. Rt = root, Bk = bark of trunk + branches, TB = trunk + branches, Wd = woody parts of new top shoots, and Lv = leaves of new top shoots, BS = new base shoots.

diately following bending, and found a higher ^{14}C-content in the apex of the shoot so long as there was terminal growth, but later there was an increase in the amount of ^{14}C in the bent part. The latter is in agreement with the tendency found on July 27 in Experiment 2. The low ^{14}C-content in the shoot apex at this time suggests that terminal growth was slight (Table 1).

Defoliation removes the most important source of assimilates and both defoliation and ringing may, irrespective of the date of execution, have a considerable impact on subsequent growth (Davies 1959, Maggs 1964, Özerol & Titus 1968). Root growth may be considerable during the later part of the growth season, and hence was particularly affected by the treatment in Experiment 3. Similar results were obtained by Head (1969) and Priestley (1964). In most cases ringing causes an increase in the content and accumulation of carbohydrates (Noel 1970), whereas there is a decrease after defoliation (May 1960, Zimmermann 1960, Kato & Ito 1962, Staesche 1967, Splittstoesser & Meyer 1971). In the present experiment ringing tends to cause an increased accumulation of sugar and sorbitol in the upper parts of the tree, and a reduction of the concentration in the lower trunk, whereas defoliation reduces the concentration in the bark. Priestley (1964) and Hennerty (1971) found no significant effects on the carbohydrate concentration in the spring after ringing, defoliation, and other treatments in the preceding autumn or summer. However, it is possible that a certain levelling of possible concentration differences may already have taken place. The concentration in the

Table 4. Contents of sorbitol + sucrose + glucose (mmol/tree) in bark + roots in December and dry matter (g/tree) in new shoots in the following July.

Symbols, see Fig. 1

	R_1	R_2	D_1	D_2	S	C
mmol/tree ..	7.4	10.6	5.1	9.3	11.4	10,8
g/tree	22.7	23.9	18.2	20,0	21.1	21.5

roots is increased after late ringing and defoliation (Fig. 1). This may be due to a faster inhibition of growth than of accumulation (cf. accumulation after cessation of growth, Hansen 1970). Davidson & Milthorpe (1966) found a rapid suspension of root growth following defoliation.

During the spring period of new growth, reserves accumulated during the previous year are used (Hansen 1967 b, Quinlan 1969). The amount of new growth taking place until the beginning of July bears no clear relation to the previous considerable differences in quantities of the various organs or in their sugar contents (Fig. 2, Table 4). A major part of the seasonal variation in the reserves of the tree appears to take place in the sugar fraction (Hansen 1967 b). It is impossible to determine whether special forms of reserves may have been mobilized in ringed or defoliated plants (Davidson & Milthorpe 1966). Nor did Priestley (1964) find any effect of ringing in the autumn on the amount of new growth in the following summer. The photosynthetic conditions appear to play a greater rôle in the absolute production of shoots (Priestley 1963, Tepper 1967). With the exception of the very earliest phases of development in the spring, the leaves soon appear to take over supplying other parts also (Hansen 1971). The removal of parts of shoots in young fruit trees at times when it might interfere with the later supply from reserves, showed no, or only a slight, effect on the new growth (Christensen & Hansen 1969). Hence it is doubtful whether reserves in young apple trees play as great a part quantitatively for the new growth as they may do in certain other plant species (Gäumann 1935, Kozlowski & Keller 1966).

Oversigt

Nogle virkninger af ringning, afbladning, sukkersprøjtning og nedbøjning af skud på translokationshastighed, ophobning og vækst hos æbletræer

Unge æbletræer blev ringet på stammen, fik alle blade fjernet eller blev sprøjtet med sorbitol efterår eller sommer. Fordelingen af ^{14}C efter tilførsel af ^{14}CO$_2$ til bladene, koncentrationen af sukker samt den efterfølgende vækst blev målt. Efter ringning øgedes ophobningen af ^{14}C og sukker i bladene eller i træets øvre dele, mens sukkerkoncentrationen i stammens nederste del blev nedsat. Efter fjernelse af alle blade i september eller oktober blev sukkerkoncentrationen i træets top reduceret i forhold til ubehandlede træer. Ringning eller afbladning i september reducerede væksten i rødder og i mindre omfang i stamme og grene.

Tilvæksten i nye skud og blade næste forår var ikke tydeligt påvirket af de etablerede forskelle i ophobning eller vækst fra det foregående efterår. Sprøjtning med en sorbitolopløsning flere gange i oktober gav ingen sikker påvirkning af sukkerindholdet eller væksten det følgende forår. Nedbøjning af årsskuddene om sommeren havde kun en ringe virkning på fordelingen af tilført ^{14}C eller på ophobning af sukker i bladene.

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