# The Influence of Formulation on Yield Response and Bark Damage Following the Application of Yield Stimulants Above the Tapping Cut

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When Hevea yield stimulant formulations containing 2,4-D or 2,4,5-T are applied to renewing bark above the tapping cut, the nature of the carrier greatly influences the magnitude of the effect on bark renewal and, to a lesser extent, yield. Differences between commercial 2,4-D and 2,4,5-T preparations are shown to be largely due to differences in carrier viscosity and composition. None of the mixtures tested can be said to be completely safe for use on valuable renewing bark and it is considered that application to scraped bark below the tapping cut is generally to be preferred.

The application of yield stimulant formulations containing 2,4-dichlorophenoxyacetic acid (2,4-D) and 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) to scraped bark below the tapping cut at six-monthly intervals is now a wellestablished estate practice (BAPTIST AND DE JONGE, 1955; RUBBER RESEARCH INSTITUTE OF MALAYA, 1959 and 1960; DE JONGE, 1961; BLACKMAN, 1961) which normally gives highly satisfactory results when used according to There are, however, a recommendations. few disadvantages in the technique, as at present employed. Thus much of the yield increase is concentrated in the first few weeks following each application and during this time the length of flow is greatly prolonged, often necessitating an afternoon collection if much of the yield increase is not to be harvested as cup lump. The cost of scraping the bark, prior to application, may also be comparatively high, forming the major part of the cost of application (RUBBER RESEARCH INSTI-TUTE OF MALAYA, 1960).

These reasons have led to investigations into the possibility of applying yield stimulants, at intervals of one or two months, to the newlytapped bark above the cut. This technique was first reported by BAPTIST AND DE JONGE (1955) and DE JONGE (1957), who utilised the technique previously employed by BEELY AND BAPTIST (1939) for the application of palm oil to promote bark renewal. PUDDY AND WARRIAR (1961) have claimed considerable advantages for the application of the 2,4-D-containing proprietory product Stimulex above, as opposed to below, the cut; these may be summarised as follows:

- 1. A comparatively slow yield increase, usually reaching a maximum after two or three months and thereafter remaining steady
- 2. Over long periods, the response is more persistent
- 3. Bark renewal is accelerated and yield from  $3\frac{1}{2}$ -year-old treated bark was similar to that of 10-year-old untreated bark.

In addition to the above, it may be noted that, because no scraping is required, application above the cut, despite the increased frequency, is the cheapest method of application (RUBBER RESEARCH INSTITUTE OF MALAYA, 1960).

DE JONGE (1957) has discussed the anatomy of renewing bark treated with 2,4-D and 2,4, 5-T. He found that the increased bark thickness consisted of non-laticiferous outer tissues and that there was no increase in the number or size of latex vessels. Irregular and unsatisfactory bark renewal was noted after \* With Appendix by J. B. GOMEZ. treatment in several experiments and the conclusion was reached that such applications could not be recommended if the bark was to be tapped again. Similar evidence led BOATMAN (1959) to come to the same conclusion.

It was hoped that it might be possible to find a compound with satisfactory yield-stimulatory activity but lacking the ability to initiate the uncontrolled meristematic activity which leads to bark damage. But the work summarised by BLACKMAN (1961) showed that, amongst the wide range of plant growth regulators and related compounds tested, there was a close correlation between yield response and increase in bark thickness. Thus reduced meristematic activity could only be obtained by the use of a compound of lower yield-stimulating power.

It was interesting to note, however, that, in this series of experiments, 2,4-D and 2,4,5-T produced rather similar effects on bark renewal when formulated in the same way; whereas it had been noticed in many previous field trials that Stimulex was less damaging than the R.R.I.M. formulation of 2,4,5-T (RUBBER RESEARCH INSTITUTE OF MALAYA, 1959). It thus seemed possible that the manner of formulation of the yield stimulant might play a part in controlling the extent of bark damage. It was with this in mind that the present work was initiated.

# **EXPERIMENT** 1

Stimulex contains  $1\frac{1}{2}$ % of 2,4-D acid in a carrier of seven parts palm oil and three parts coconut oil (PUDDY AND WARRIAR, 1961), whereas the R.R.I.M. formulation consists of the butyl ester of 2,4,5-T in a 5:3 mixture of palm oil and petrolatum grease (BAPTIST AND UDE JONGE, 1955). It was decided to test Stimulex together with a number of other formulations containing  $1\frac{1}{2}$ % 2,4-D, either as the acid or the butyl ester, in either thin (palm oil or palm oil/coconut oil) or highly viscous (palm oil/petrolatum or hydrous Ianoline) carriers. The carriers selected had all previously been used successfully in yield stimulation experiments.

#### METHODS

### Layout of Experiments

The experiment was set up in May 1960 on illegitimate seedling trees planted in 1931 in Field 25(v), R.R.I.M. Experiment Station. A randomised-block single-tree-plot design with forty-five replications was used. The trees were tapped S/2.d/2.100% in good renewed bark. The mixtures were painted on a strip of bark  $\frac{3}{4}$  to 1 in. in width immediately above the tapping cut, at monthly intervals. Care was taken to avoid overlapping successive applications.

#### Recording

Yields. Individual tree yields were recorded by coagulating the latex obtained at each tapping; the cup coagula so obtained being air-dried for a month before weighing. Records of yield taken before treatment were used as a basis for the correction of post-treatment yields by co-variance analysis.

Bark renewal. The thickness of the treated bark was measured with a Schlieper bark gauge six months after the first application. Such measurements cannot be relied upon to give accurate absolute values for bark thickness, since the sharp edge of the gauge frequently penetrates some distance into the outer wood; but the results provide useful comparative results, provided errors due to different operators are avoided.

In an attempt to assess the quality of the renewing bark and its suitability for future tapping, a visual scoring technique was adopted. The scores given ranged from 0 (corresponding to perfectly smooth renewal, free from all wounds and blemishes) to 6 (for the highest degree of damage encountered, in which large areas of bare wood were exposed). It was considered that renewed bark with a score of 4 or higher was definitely unsatisfactory and unlikely to be suitable for future tapping. Some eight months after treatment, samples of treated bark in selected treatments and corresponding samples in the control trees were removed for anatomical observations.

## RESULTS

Yields

Mean yields over a period of six months after treatment are given in *Table 1*. All the treatments gave satisfactory yield increases. It will be seen that there are no significant differences between any of the five pairs of acid and ester formulations. There is a tendency for the thicker carriers to give slightly higher yields, but this is not established. There is little difference between the 2,4-D and 2,4,5-T formulations in palm oil/petrolatum. Bark Renewal

The effects on bark thickness and quality are set out in Table 2. All the treatments significantly increased thickness and had a deleterious effect on bark renewal. There are, again, no significant differences between the effects of the acid and ester formulations on bark thickness and although, in the quality scores, the differences do reach significance in two cases, they are not consistent with each other. There are large differences between the thicker carriers, lanolin and palm oil/petrolatum, which induced intense meristematic activity and caused very severe damage, and the thinner carriers, such as 'Stimulex'. palm oil and palm oil/coconut oil, which were comparatively moderate in their effects. It will be noted, however, that even the latter group gave mean scores ranging from 3.3 to 4.1, in contrast to the 2.2 of the control trees. The 2,4,5-T treatments caused severe damage, but not significantly greater than the corresponding 2,4-D treatments.

## Correlations between Bark Thickness, Quality Score and Yield

As shown in Figure 1a, there is an excellent correlation between the mean bark thickness and quality scores of the various treatments, indicating a close relationship between the induction of meristematic activity and the occurrence of damage. There are also moderately strong correlations between the mean yields and bark thickness (Figure 1b) and mean yields and quality score (Figure 1c), reflecting the tendency for the thicker mixtures to give slightly higher yield increases, thicker bark and higher scores.

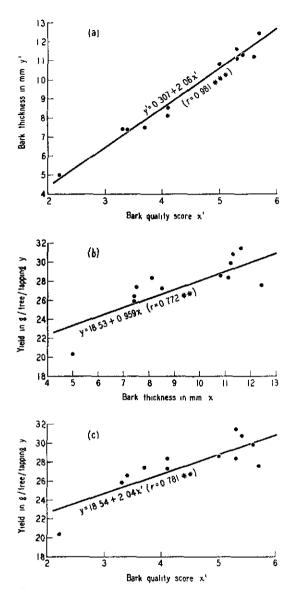


Figure 1. Experiment 1. The relationship between treatment means for: (a) thickness of treated bark and bark renewal quality score; (b) yield per tree per tapping and the thickness of treated bark; (c) yield per tree per tapping and bark renewal quality score.

Anatomical observations. These are reported in the Appendix.

Treatment	Grams/tree /tapping	% control	
A. Control, no treatment	20.3	100	
B. 'Stimulex'	27.3	134	
C. 1.5% 2,4-D acid in palm oil/coconut oil	25.9	128	
D. 1.5% 2,4-D ester in palm oil/coconut oil	27.4	135	
E. 1.5% 2,4-D acid in palm oil	28.3	139	
F. 1.5% 2,4-D ester in palm oil	26.4	130	
G. 1.5% 2,4-D acid in hydrous lanolin	28.6	141	
H. 1.5% 2,4-D ester in hydrous lanolin	31.5	155	
I. 1.5% 2,4-D acid in palm oil/petrolatum	28.4	140	
J. 1.5% 2,4-D ester in palm oil/petrolatum	30.8	152	
K. 1.5% 2,4,5-T acid in palm oil/petrolatum	29.9	147	
L. 1.5% 2,4,5-T ester in palm oil/petrolatum	27.6	136	
Min. sig. diff. (P=0.05)	3.6	18	

#### TABLE 1. EXPERIMENT 1. MEAN YIELDS FOR SIX MONTHS AFTER TREATMENT

# TABLE 2. EXPERIMENT 1. THICKNESS OF TREATED BARK AND BARK RENEWAL QUALITY SCORES

(The quality scores ranged from 0 for perfectly smooth blemish-free renewal to 6 for the worst condition encountered. Bark with a score of 4 or above was judged to be unsuitable for future tapping)

	Treatment	Thickness in mm	Quality score
A.	Control — no treatment	5.0	2.2
B,	Stimulex	8.5	4.1
C.	1.5% 2,4-D acid in palm oil/coconut oil	7.4	3.3
D,	1.5% 2,4-D ester in palm oil/coconut oil	7.5	3.7
E.	1.5% 2,4-D acid in palm oil	8.1	4.1
F.	1.5% 2,4-D ester in palm oil	7.4	3.4
G.	1.5% 2,4-D acid in hydrous lanolin	10.8	5.0
H.	1.5% 2,4-D ester in hydrous lanolin	11.6	5.3
I.	1.5% 2,4-D acid in palm oil/petrolatum	11.1	5.3
J.	1.5% 2,4-D ester in palm oil/petrolatum	11.3	5.4
К.	1.5% 2,4,5-T acid in palm oil/petrolatum	11.2	5.6
L.	1.5% 2,4,5-T ester in palm oil/petrolatum	12.4	5.7
	Min. sig. diff. (P=0.05)	1.3	0.4

## **EXPERIMENT 2**

In considering the results obtained in the first experiment, the question arose as to whether the effect of a carrier material on yield and bark renewal could be predicted on the basis of its viscosity. In order to test this point, a range of carriers were made up based on 'Carnea' and 'Limea' oils, which are hydrocarbon oils available in a wide range of viscosities,\* together with petrolatum grease\*\* as a thickening agent. Palm oil and palm oil/ petrolatum were included for comparative purposes. One per cent acid equivalent of the n-butyl ester of 2,4,5-T was used as the active agent in all the experimental mixtures, but Stimulex (1.5% 2,4-D acid) was also included for comparison.

Viscosities were measured at 25°C using a Model LVF Brookfield viscometer. The rheological behaviour of such mixtures is complex and it is not claimed that much significance can be attached to the absolute values obtained. Nevertheless, they made possible the preparation of a number of carriers with a range of viscosities and compositions, as listed in *Table 3*.

#### METHODS

### Layout of Experiment

The experiment was started in April 1961 on twinned seedling trees planted in 1932 in Field 22B, R.R.I.M. Experiment Station. A randomised single-tree plot design with 45 replications was employed. The trees were tapped S/2.d/2.100% in renewed bark, which was of fairly good quality.

#### Recording

This was carried out as for Experiment 1.

- \* Supplied by the Shell Company of the Federation of Malaya, Ltd.
- \*\* Shell Otina C.

TABLE 3. EXPERIMENT 2. VISCOSITY OF CARRIERS EMPLOYED (Measurements in centripoises, using a Brookfield viscometer, Model LVF, at 25° C)

Composition of carrier †	Viscosity
Carnea 15	21
Carnea 41	145
Carnea 15/petrolatum (5:1)	150
Palm oil	190
Stimulex (palm oil/coconut oil, 7:3)	260
Carnea 72	1050
Carnea 15/petrolatum (5:2)	1000
Limea 75	2000
Limea 79	4650
Carnea 72/petrolatum (7:1)	4800
Carnea 15/petrolatum (7:4)	4700
(Palm oil/petrolatum (5:3)	40,000
Carnea 72/petrolatum (2:1)	39,000
Carnea 15/petrolatum (4:5)	40,000

<sup>†</sup> All mixtures, except Stimulex, contained 1% acid equivalent of *n*-butyl ester of 2,4,5-T. Listed in order of viscosity, those having similar viscosities being bracketed together.

# RESULTS

Yields

The mean yields for six months after the first application of treatments are shown in *Table 4*. Stimulex gave the highest yield increase. It has been shown previously that 2,4-D gives a significantly higher response than 2,4,5-T in some experiments and that, in others, the reverse is the case (BLACKMAN, 1961). The differences between the various 2,4,5-T treatments mostly fail to reach significance at the five per cent level. If the mean yields are plotted against the viscosity of the carrier (expressed as  $log_{10}$ ) for the treatments receiving 1% 2,4,5-T, as in Figure 2a,

it will be seen that no significant correlation emerges. The same is also true if only the unmixed Carnea-Limea series of oils are considered (*Figure 2b*), but a good correlation is obtained for the series formed by the addition of petrolatum to Carnea 15 (*Figure 2c*). A suggestion of a similar effect for the incorporation of petrolatum into Carnea 72 cannot be confirmed because of the small number of mixtures tested.

### Bark Thickness

These measurements are summarised in *Table 5*. All the experimental treatments led to highly significant increases in bark thickness, these being particularly high for the

TABLE 4. EXPERIMENT 2. MEAN YIELDS FOR SIX MONTHS AFTER TREATMENT

Treatment *	Grams/tree /tapping	control
A. Control-no treatment	31.6	10 <b>0</b>
B. Stimulex	46.5	147
C. Carnea 15	39.1	124
∫D. Carnea 41	39.5	125
L. Carnea 15/petrolatum (5:1)	40.8	129
F. Palm oil	40.6	129
G. Carnea 72	38.7	122
H. Carnea 15/petrolatum (5:2)	41.2	130
J. Limea 75	37.7	119
K. Limea 79	42.7	135
L. Carnea 72/petrolatum (7:1)	41.7	132
M. Carnea 15/petrolatum (7:4)	44.7	141
N. Palm oil/petrolatum (5:3)	38.6	122
O. Carnea 72/petrolatum (2:1)	41.2	130
P. Carnea 15/petrolatum (4:5)	44.6	141
Min. sig. diff. (P=0.05)	5.7	18

<sup>•</sup> All mixtures, except Stimulex, contained 1% acid equivalent of *n*-butyl ester of 2,4,5-T. Formulations bracketed together are of similar viscosity. With the exception of Stimulex, they are arranged in order of increasing viscosity.

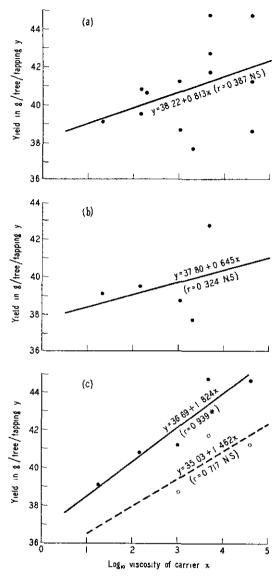


Figure 2. Experiment 2. The relationship between mean yield per tree per tapping and  $log_{10}$  viscosity of yield stimulant carrier for: (a) all treatments receiving 1% 2,4,5-T; (b) the Carnea-Limea series of oils; (c) mixtures of Carnea 15 and petrolatum ( $\bullet$ ——••) and Carnea 72 and petrolatum ( $\circ$ ——••).

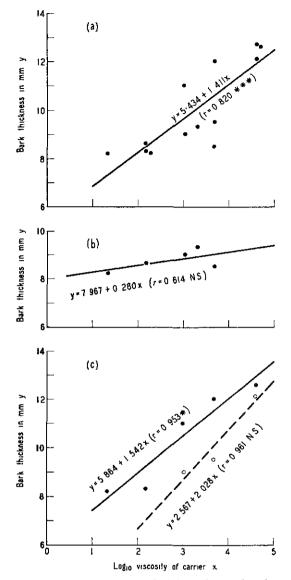


Figure 3. Experiment 2. The relationship between mean thickness of treated bark and  $log_{10}$  viscosity of yield stimulant carrier for: (a) all treatments receiving 1% 2,4,5-T; (b) the Carnea-Limea of oils; (c) mixtures of Carnea 15 and petrolatum ( $\bullet$ ——••) and Carnea 72 and petrolatum ( $\circ$ —••••).

	Treatment *	Thickness in mm	Quality score	
А.	Control - no treatment	5.7	2.5	
B.	Stimulex	9.9	4.6	
С.	Carnea 15	8.2	3.4	
ſD.	Carnea 41	8.6	3.8	
ĹΕ.	Carnea 15/petrolatum (5:1)	8.3	4.2	
F.	Palm oil	8.2	4.3	
ſG.	Carnea 72	9.0	4.2	
<u></u> ۱н.	Carnea 15/petrolatum (5:2)	11.0	5,2	
J.	Limea 75	9.3	4.5	
ſK.	Limea 79	8.5	4.5	
{ L.	Carnea 72/petrolatum (7:1)	9,5	4.9	
M.	Carnea 15/petrolatum (7:4)	12.0	5.7	
∫N.	Palm oil/petrolatum (5:3)	12.7	5.6	
{ <b>o</b> .	Carnea 72/petrolatum (2:1)	12.1	5.8	
P.	Carnea 15/petrolatum (4:5)	12.6	5.9	
 Min	, sig. diff. (P=0.05)	1.1	0.4	

# TABLE 5. EXPERIMENT 2. THICKNESS OF TREATED BARK AND BARK RENEWAL QUALITY SCORES

\* All mixtures, eccept Stimulex, contained 1% acid equivalent of *n*-butyl ester of 2,4,5-T. Formulations bracketed together are of similar viscosity. With the exception of Stimulex, they are arranged in order of increasing viscosity.

more viscous mixtures. The Carnea 15/petrolatum mixtures of similar viscosity caused significantly greater increases than did Carnea 72 or Limea 79, suggesting a difference in behaviour not accounted for by the viscosity measurements. The mean bark thickness for each of the 2,4,5-T treatments has been plotted against log<sub>10</sub> carrier viscosity in *Figure 3a*. A very highly significant overall correlation is obtained. If, however, only the unmixed Carnea-Limea series is considered, as in *Figure 3b*, the correlation coefficient is only 0.614. The Carnea 15/petrolatum and Carnea 72/petrolatum series (*Figure 3c*) have correlation coefficients of 0.9530 and 0.9607 respectively, although the latter, depending on only three points, is not significant at the P=0.05level. The difference in slope between *Figures* 3b and 3c is also noteworthy.

# Bark Renewal Quality

The mean scores of the different treatments are also listed in *Table 5*. The renewal of the control trees was satisfactory but all the treatments resulted in highly significant reductions in quality. All but two of the mixtures tested produced renewed bark with mean scores of four or over (judged to be unsatisfactory for future tapping). When the relationship with carrier viscosity is considered,

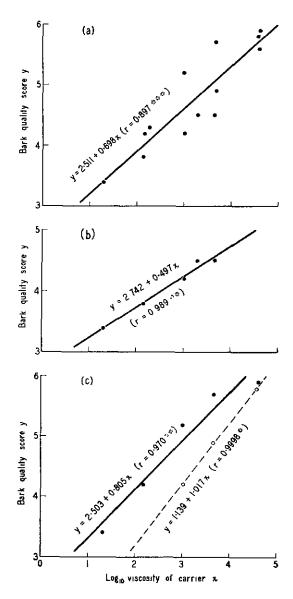


Figure 4. Experiment 2. The relationship between mean bark renewal quality score and  $log_{10}$  viscosity of yield stimulant carrier for: (a) all treatments receiving 1% 2,4,5-T; (b) the Carnea-Limea series of oils; (c) mixtures of Carnea 15 and petrolatum ( $\bullet$ ——••) and Carnea 72 and petrolatum ( $\circ$ ——••).

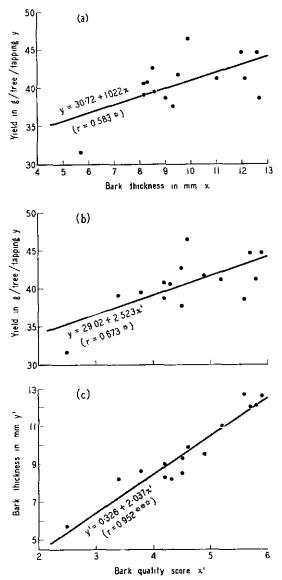


Figure 5. Experiment 2. The relationship between treatment means for: (a) yield per tree per tapping and thickness of treated bark; (b) yield per tree per tapping and bark renewal quality score; (c) thickness of treated bark and bark renewal quality score.

a significant correlation is shown for all the 2,4,5-T treatments combined (Figure 4a), for the Carnea-Limea series (Figure 4b), for the addition of petrolatum to Carnea 15 and even for the three Carnea 72/petrolatum mixtures (Figure 4c). As in the case of the bark thickness measurements, the slope of the regression lines is greater for the petrolatum mixtures than for the unmixed oils.

# Correlations between Bark Thickness, Quality Score and Yield

It will be seen from Figures 5a and b that there is not a strong correlation between mean yield and bark thickness nor between yield and renewal quality score although both reach significance at the 5% level. There is, however, a clear relationship (Figure 5c) between bark renewal score and bark thickness, as noted in Experiment 1; increased bark thickness being accompanied by poorer renewal.

# EXPERIMENTS 3 AND 4

While Experiment 2 was in progress, it became possible to include additional treatments in two experiments being set up for other purposes. A comparison was, therefore, made between:

- A. Control, untreated
- B. Stimulex
- and C. 1% acid equivalent of *n*-butyl ester of 2,4,5-T in Carnea 15/ petrolatum (5:1, by weight)

The 2,4,5-T preparation was identical with Treatment E in Experiment 2 and was selected because it was more adherent to the bark than preparations made with Carnea 15 or Carnea 41 without petrolatum and showed little or no tendency to flow down after application. It also gave a good yield response and appeared to be slightly less damaging than Stimulex and much less so than palm oil/ petrolatum.

# METHODS

# Layout of Experiments

Experiment 3 was carried out on plots of the unselected clones of the RRIM 500 series large-scale clone trial in Field 30 at the R.R.I.M. Experiment Station. The trees were tapped V/2.d/2.100% on a high panel of virgin bark. Experiment 4 was laid out on the small-scale clone trial in Field 33, R.R.I.M. Experiment Station, the trees being tapped S/2.d/2.100% in bark of first renewal. In both experiments tree plot designs were employed, there being 46 trees per treatment in Experiment 3 and 50 in Experiment 4.

# Recording

Yield recording was carried out as for the earlier work, except that these experiments were continued for twelve months instead of six. An attempt was made to gain a more detailed picture of the effect of the stimulants on bark renewal. Rather than make repeated measurements on the same strip of bark (with the likelihood of causing serious wounding and interfering with the assessment of renewal quality), it was decided to take measurements only at the conclusion of the experiment and to record the thickness of the renewing bark of various ages on each tree. The sampling positions are set out in Table 6. Bark renewal quality scores were assessed as before, at the conclusion of the experiment.

# RESULTS

# Yield

Mean yields in the two experiments are given in *Table 7*. Both stimulants gave large increases in yield and no significant difference was established between them.

# Bark Thickness

The measurements are presented in Figures 6 and 7. The general trends are very similar in the two experiments, despite the fact that one is dealing with first renewal and the other with second renewal bark. There were no significant differences in thickness between the untapped bark immediately below the cut in any of the treatments. The one-monthold renewing bark, tapped since the last application of stimulant, also shows little or no effect of treatment. A relatively small increase in thickness is seen in two-monthold bark, which received the final stimulant application. Very large increases in thickness due to stimulation are seen in the older treated bark, but there is somewhat less effect

TABLE 6.	SAMPLING	POSITIONS	FOR	BARK	THICKNESS	MEASUREMENTS	IN
		EXPER	IMEN	TS 3 A	ND 4	÷	

Position	Description	Months since tapping
1	Untapped bark below cut (untreated)	-
2	Untreated renewing bark	1
3	Treated renewing bark	2
4	2 <b>3 93 2</b> 9	4
5	۰٫ ۲٫ ۲٫	7
6	ı) <u>1</u> 9 <u>2</u> 1	10
7	yy yy I3	13
8	Untreated renewing bark	14
9	ž3 ž3 žj	24

#### TABLE 7. EXPERIMENTS 3 AND 4. MEAN YIELDS FOR TWELVE MONTHS AFTER TREATMENT

<b>T</b>	Experin	nent 3	Experiment 4		
Treatment	Grams/tree /tapping	% control	Grams/tree /tapping	control	
A. Control no treatment	44.9	100	35.1	100	
B. Stimulex	68.4	152	47.5	135	
C. 1% 2,4,5-T in Carnea 15/ petrolatum (5:1)	66.8	149	46.0	131	
Min. sig. diff. (P=0.05)	9.7	22	5.3	15	

in the case of the thirteen-month-old bark which received the first application. A small treatment effect is seen in the 14-month-old bark which was not itself treated. This is presumably due to upward translocation of the growth regulators. There is no detectable effect in the case of the 24-month-old bark. It would appear that the effect of the yield stimulant on bark thickness is virtually complete within six months of application. There are no significant differences between the two mixtures.

# Bark Renewal Quality

The quality scores are summarised in *Table* 8. It is evident that both treatments induced some degree of damage. The 2,4,5-T mixture was slightly less damaging than Stimulex in Experiment 3, the difference in score just reaching significance at the 5% level, but the figures were very similar in Experiment 4.

### DISCUSSION

The present work has shown that the increase in bark thickness which results from appli-

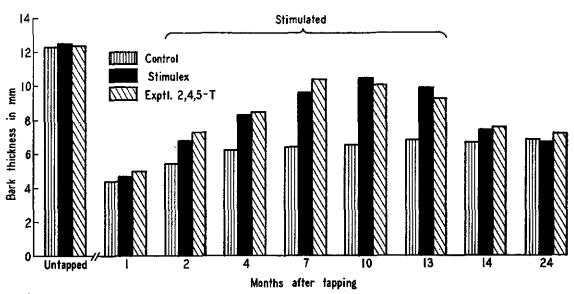


Figure 6. Experiment 3. The effect on bark renewal of applications of Stimulex and an experimental 2,4,5-T mixture. Comparison of thickness of virgin bark below the tapping cut with renewing bark of various ages. As indicated, the renewing bark between two and thirteen months old had received applications of the stimulants during the first month after tapping.

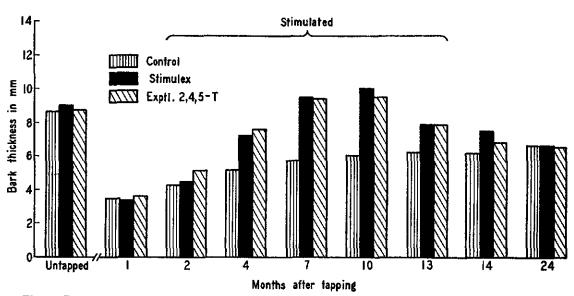


Figure 7. Experiment 4. The effect on bark renewal of applications of Stimulex and an experimental 2,4,5-T mixture. Comparison of thickness of bark of first renewal below the tapping cut with bark of second renewal of various ages above the cut. As indicated, the renewing bark between two and thirteen months old had received applications of the stimulants during the first month after tapping.

Treatment	Experiment 3	Experiment 4	
A. Control — no treatment	2.2	2.5	
B. Stimulex	3.9	3.5	
C. 1% 2,4,5-T in Carnea 15/petrolatum (5:1)	3.5	3.6	
Min. sig. diff. (P=0.05)	0.4	0.5	

# TABLE 8. EXPERIMENTS 3 AND 4. BARK RENEWAL QUALITY SCORES

cation of yield stimulant mixtures to renewing bark above the tapping cut is invariably accompanied by some degree of deterioration in the standard of renewal, with uneven growth, cracking and exaggration of minor tapping wounds. A close correlation was established in both Experiments 1 and 2 between the magnitude of the increase in thickness and the decrease in standard of renewal. Therefore, to be suitable for application to valuable renewing bark, the tendency of a yield stimulant mixture to induce meristematic activity should be minimal.

The carrier employed in the formulation can greatly influence the extent to which such meristematic activity is induced. As far as the Carnea-Limea series of hydrocarbon oils is concerned, there was a slight, but definite. tendency for damage to increase with rising viscosity. At a given measured viscosity. mixtures containing a large proportion of petrolatum had very much greater effects on renewal. From this point of view, at least, use of a large proportion of petrolatum in a mixture should be avoided. No information is at present available as to the reason for this effect of petrolatum. In the absence of a growth regulator, its effects on renewal are relatively slight (DE JONGE, 1957). Other greasy substances, for example lanoline in Experiment 1, may be equally deleterious.

It is evident that the large differences in effect on renewal between the R.R.I.M. formulation of 2,4,5-T and thinner mixtures such as Stimulex are largely attributable to the high proportion of petrolatum in the former. The present experiments confirm that there is little difference between the effects of 2,4-D and 2,4,5-T on bark renewal when applied in similar carriers. There also appears to be little difference between formulations containing the free acids or the *n*-butyl esters of 2,4-D or 2,4,5-T, although significant differences in both yield and bark renewal were recorded in an earlier experiment (BLACKMAN 1961; *Tables 1* and 2) in which 2,4,5-T acid and *n*-butyl ester were compared in a carrier of white petrolatum jelly. Possibly the slight solubility of the free acid in this carrier was responsible for its lower activity.

The effects of carrier on yield response are clearly less pronounced than on renewal. A significant correlation was established between yield and viscosity for the addition of petrolatum to Carnea 15, but not for the unmixed Carnea-Limea oils, while a correlation was established between the magnitude of yield increase and the extent of meristematic activity and damage induced in both Experiments 1 and 2. From this it would appear that a reduction in damaging potential towards the bark is likely to be accompanied by at least a small loss in yield.

The thin hydrocarbon oils such as Carnea 15 and 41 do not adhere well to the bark and tend to run down from the point of application, particularly if applied to scraped bark below the cut. Thus in a general-purpose yield stimulant, the incorporation of a small proportion of petrolatum as a 'sticker', as in

the 2,4,5-T mixture tested in Experiments 3 and 4, may be desirable. The experimental 2,4,5-T mixture tested in these two experiments has shown itself to be very similar to Stimulex in its effects. The authors would, however, hesitate to recommend it, or any of the mixtures tested, for application to renewing bark which is to be tapped again. The four experiments described here, together with earlier observations, strongly suggest that such applications always lead to some deterioration in the quality of renewed bark. How serious such a deterioration would be. depends on several factors, such as the standard of tapping and renewal prior to treatment, the degree of susceptibility of the planting material and, probably, environmental conditions. Certainly the risk involved appears to be sufficient to prohibit any recommendation for general use.

In the case of applications to scraped bark below the tapping cut, the treated bark is tapped away before any serious damage can develop and subsequent renewal is usually scarcely affected. Extensive experience has confirmed the safety of the technique, provided reasonable care is taken. Fortunately progress is also being made to overcome those minor difficulties which led to an interest in applications above the tapping cut. Thus lighter scraping (RUBBER RESEARCH INSTITUTE OF MALAYA, 1961) offers promise of a reduction in scraping costs on thick virgin bark. The stimulation of one-third of each task at two-monthly intervals helps to even out the overall yield trend and, if necessary, the stimulated trees can be tapped first and collected last to ensure that as much of the increased yield as possible is harvested as The effect of smaller but more frequent latex. applications below the cut is also being investigated, although the economics of such a method would need careful evaluation.

Evidence has also accumulated (RUBBER RESEARCH INSTITUTE OF MALAYA, 1963) that, in trees tapped on virgin bark or good renewed bark, the yield increases obtained by monthly or two-monthly applications above the tapping cut are frequently somewhat lower than those from six-monthly treatment below the cut. Only in the case of trees tapped on rather poor renewed bark, which is difficult to scrape successfully, have consistently better yield responses been obtained from abovethe-cut applications. Such trees will normally be on their last tapping cycle when there is no objection to applications of 2,4-D or 2,4,5-T to the renewing bark.

If it may be concluded from the above that application to scraped bark below the tapping cut is generally to be preferred, then the composition of the yield stimulant carrier becomes somewhat less critical, since damage is normally avoided, even with a carrier so potentially damaging as the present palm oil/ petrolatum mixture. Nevertheless, it is felt that it is desirable to minimise the danger involved in any accidental misuse, especially as comparatively young high-yielding trees are now beginning to be treated. The present work is, therefore, being extended to test alternative 2,4,5-T formulations for use below the tapping cut.

# ACKNOWLEDGEMENTS

The authors wish to express their appreciation of the work on the statistical analyses by the Head of Statistics Division, Mr A. Fong Chu Chai, Mr G. C. Iyer and Mr R. Narayanan. The co-operation of the Manager and Staff of the R.R.I.M. Experiment Station is gratefully acknowledged, as is the technical assistance of Mr B. Manikam, Mr K. Vasudevan and Mr S. Tharmalingam.

Botanical Division

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J. B. GOMEZ

STRUCTURE OF NORMAL RENEWING BARK

# Mechanism of Renewal

Renewal starts primarily by the formation of a secondary cork cambium (phellogen) from peripheral cells of the exposed parenchyma of the approximately one millimetre of phloem tissue left untapped during the tapping operation. These cork cambium cells divide, forming phelloderm tissue on the interior, and phellem (cork) on the exterior. Thus, the activity of the phellogen creates a tissue called 'periderm' which comprises the phellem, phellogen and phelloderm. Meanwhile, the vascular cambium divides, forming the phloem of the renewing inner bark-a normal activity associated with secondary thickening, in this instance accelerated by the wound reaction after tapping. The formation of the phellogen and subsequent formation of periderm occurs within a few days in bark exposed after tapping, as evidenced by the presence of chlorenchyma which is observed in renewing bark of 1-2 weeks growth.

Young laticifers have their origin in renewing bark from tissue produced by the vascular cambium. They originate from initials situated close to the cambium. No differentiation into laticifers occurs in the more mature parts of the phloem. Latex vessels retained in the untapped part of the bark are gradually displaced outwards as new phloem tissue is produced from the vascular cambium.

# General Organisation of Tissues in Control Trees

There is a zone of clear tissue adjacent to the vascular cambium containing rows of latex vessels. This has been produced by the vascular cambium since tapping (a growth period of 8 months). This is designated as 'new phloem tissue' in the discussion below. Immediately outside the 'new phloem tissue' is a zone of bark with a deep coloration said to be due to the presence of anthocyanin (BOBILIOFF, 1923). The tissues outside the vascular cambium embracing the 'new phloem

The effect of stimulants containing 2,4-D and 2,4,5-T on renewing bark has been reported briefly (DE JONGE 1955; 1957); Significant increases in bark thickness were recorded for stimulated bark. No significant difference could be established in the number of latex vessel rows in treated bark. The present investigation was carried out to examine in further detail the effect of 2,4-D formulations on the laticiferous system of renewing bark and on general bark anatomy.

The materials for this investigation were obtained from Experiment No. 1 in Field 25(v) of R.R.I.M. Experiment Station already reported. A band of renewing bark one inch in width immediately above the tapping cut received stimulant application monthly for six consecutive months. Fifty samples were taken from each of three treatments in the experiment. The treatments were control (A). Stimulex (B), and 2,4-D acid in a palm oil/ petrolatum base (I). Bark samples were taken from the strip of bark first stimulated, which at the time of sampling was of 8 months' renewal. Control trees were sampled from a similar position approximately 8 in. above the cut as shown in Figure 1. Samples were fixed in formalin acetic alcohol. Transverse sections were prepared and stained in Sudan blue.

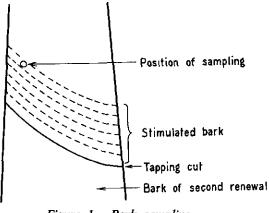


Figure 1. Bark sampling.

tissue' and the pigmented zone is described as 'normal tissue'. Thus, the term 'normal tissue' covers residual phloem left after tapping and the newly produced phloem. Periderm, situated outside this 'normal tissue', accounts for the rest of the bark. However, in view of the anatomical changes occurring in stimulated trees (see later) the tissue exterior to the normal tissue has been termed 'disturbed tissue'.

# STRUCTURE OF BARK TREATED WITH 2,4-D STIMULANT

#### General Organisation

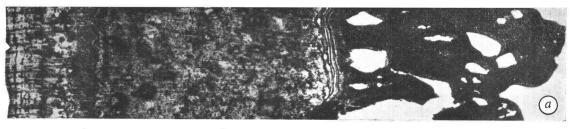
The various elements constituting the 'normal tissue' described above for the control trees are clearly identified in stimulated trees. The periderm, however, shows marked effects of the stimulants. The inner periderm cells are in an active meristematic state, clearly evidenced by their large nuclei, thin walls, and orientation pattern. The tissues outside the 'normal tissue' in stimulated trees have been described here as 'disturbed tissue'. The 'disturbed tissue' in stimulated trees in fact includes a few outer layers of residual phloem in addition to the newly formed periderm. *Figure 2* shows a comparison of the control and Treatment I.

# Measurements

Bark thickness was measured using vernier callipers before samples were sectioned. Thickness of 'normal tissue' and new phloem tissue were measured by microprojection of transverse sections. The number of latex vessel rows was counted from transverse sections stained in Sudan blue. The extent of 'disturbed tissue' was computed from data of total bark thickness and 'normal tissue' measurements. Corky tissues were measured from the cut end of bark samples.

#### **Observations**

The 'normal tissue' in stimulated bark, with its inner zone of 'new phloem tissue' composed of sieve tubes, laticifers and comple-



--->'New phloem tissue` ----->'Normal tissue`

'Disturbed tissue'

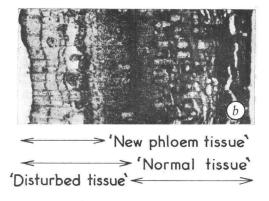


Figure 2. Tissues in stimulated and unstimulated bark: (a) Treatment I; (b) Control.  $(\times 10)$ .

mentary parenchyma does not show any abnormal anatomical features.

In the 'disturbed tissue' the most striking phenomenon is the active meristematic state of the phelloderm cells and few outer layers of residual phloem left at the time of stimulation. The outer periderm consisted mostly of deeply fissured, thick, hard, corky tissue which showed macroscopic malformations. Because of its meristematic nature, the outer phelloderm frequently tore away with the hard outer tissues during sectioning. It is possible that similar difficulties might be encountered in future tapping of such bark.

At the time of application of the stimulant, the bark tissue treated was in a primary state of renewal. It is assumed from observations on normal bark renewal that the phellogen would have been formed in bark exposed during earlier tappings. It would then seem that the stimulant rendered the phellogen more active.

Frequently one or two rings of latex vessels at the junction of 'normal tissue' and 'disturbed tissue' showed signs of disruption in stimulated bark, a phenomenon which is absent in normal renewing bark. The intense meristematic activity of cells adjacent to such latex vessel rings evidently caused disruption and displacement.

A further effect of the meristematic nature in 'disturbed tissue' is the reduction of stone cell density. This is clearly noticeable in *Figure 2*. Quantitative studies to determine the total amount of stone cells have not been attempted.

A summary of observations made is given in *Table I*. It is evident that:

 There is a significant increase in total bark thickness in bark treated with stimulants.
 The thickness of 'normal tissue' and 'new phloem tissue' is unchanged by stimulant applications.

3. The thickness of 'disturbed tissue' increases from control to treatments B and I. The mean values are significantly different between treatments. Stimulex shows a 97% increase and 2,4-D in palm oil/petrolatum a 210% increase from control.

4. The total number of latex vessels is significantly less for Treatment I than for the control. For Treatment B the number was less than in the control but the difference was not significant.

5. The mean number of latex vessel rings in 'new phloem tissue' is significantly lower in Treatment I. Stimulex treatment did not show a significant difference from the control. As the thickness of the 'new phloem tissue' is unchanged there is in fact a lower production of latex vessel rings in new phloem produced after Treatment I.

6. Corky tissues are increased in stimulated trees. The mean values are 200% of the control in Treatment B and 500% in Treatment I.

# DISCUSSION AND CONCLUSIONS

Significant increases in bark thickness due to stimulant application have been reported twice (DE JONGE, 1955 and 1957). However this increase was only in the non-latex bearing part of bark (DE JONGE, 1957). The present investigation clearly brings out the fact that 'new phloem tissue' development is unchanged by stimulant application. Nevertheless the frequency of latex vessel production has been affected by Treatment I. The stimulant based on palm oil/petrolatum has undoubtedly caused more damage and has upset the rhythm of latex vessel production. Stimulex has not shown this effect conclusively but Table I shows a suggestion of a decrease in the number of latex vessel rings.

The significant overall effect on bark anatomy due to stimulants has been one of active regeneration of tissue in the unproductive bark; virtually a significant increase in periderm formation. The amount of corky tissues and the amount of 'disturbed tissue' produced by phellogen confirm earlier findings that increase of bark thickness is due to increase of the unproductive region of bark.

# ACKNOWLEDGEMENTS

The material for this investigation has been kindly supplied by Dr S. G. Boatman and Mr P. D. Abraham from their experiments.

			Thickness in centimetres of					No. of latex vessel rings in	
		Total bark	'Normal tissue'	New phloem tissue	'Disturbed tissue'	Corky tissue	Whole bark	New pholem tissue	
Control	Mean	0.480	0.182	0.114	0.296	0.056	10.6	6.2	
(Treatment A)	s.e.	±0.0169	±0.0070	±0.0054	±0.0143	±0.0024	±0.58	±0.43	
Stimulex	Mean	0.776	0.180	0.119	0.592	0.138	9.9	5.5	
(Treatment B)	s.¢.	$\pm 0.0356$	±0.0101	±0.0071	$\pm 0.0347$	±0.0192	±0.47	±0.31	
	Mean	1.104	0.159	0.100	0.943	0.313	7.4	3.3	
2, 4-D in palm oil/petr (Treatment I)	s.e.	±0.0567	±0.0114	±0.0066	$\pm 0.0524$	$\pm 0.0351$	±0.53	±0.39	

# TABLE I. SUMMARY OF OBSERVATIONS ON BARK OF CONTROL AND TREATMENTS B AND I. (Forty-five trees each)

I thank Ir P. de Jonge and Dr S. G. Boatman for valuable discussions, Mr Chen Khyun Thai for technical assistance and Mr R. Narayanan for the statistical analysis.

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