## SOME FACTORS INFLUENCING STARCH IN SUGARCANE

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### **Summary**

When cane is milled, a linear relationship exists between the percentage starch and percentage sucrose extracted, although the slope and intercepts of the line vary from sample to sample, the slope apparently decreasing with increasing starch concentration in the cane. The concentration of starch in the juice increases with increasing extraction, but is not very strongly dependent at high extractions within the range considered.

When cane is left standing before milling the starch content decreases gradually at first, but rapidly after about five days.

Starch is largely concentrated in the top 2 or 3 feet of cane and, hence, cutting off this portion would reduce the starch in the juice appreciably. The portion of lowest starch content is between 5 and 6 feet from the top. The starch per cent sucrose in the top 6 in. is very high, generally decreasing rapidly to a minimum at 5-6 feet. Nodes were found to contain 3.5-4.0 times the concentration of the internodes on a dry weight basis, but no significant difference was found between nodes and buds.

Of the three major plant-foods, potash application is by far the most efficacious in the reduction of the starch content of the cane, a large reduction of starch generally accompanying large increases in yield on potash-deficient soils. Nitrogen application, while generally causing a reduction of starch where there is a significant yield response, does not seem to have nearly such a marked effect as potash. Phosphate application has no apparent effect on the starch content.

Although the reduction in the number of nodes per unit length of cane with increased growth undoubtedly accounts for some of the decrease in starch due to potash application, it is unlikely that it is the primary cause. It seems that accumulation of starch is a condition peculiar to cane grown on potash-deficient soils, since even varieties which are notoriously high in starch contain only very small amounts when grown on soils containing ample available potash.

Irrigation seems to decrease the starch in cane, probably because of increased growth and plant-food uptake. This effect was especially pronounced in young cane, but gradually became less pronounced towards maturity. There was a marked decrease of starch with age up to harvesting for dryland and moderately irrigated cane, but no change for heavily irrigated cane. Although starch in approximately 3 year-old cane was considerably lower than in approximately 2 year-old cane in three instances, in the 4th the reverse was true, and therefore, no definite conclusions regarding the effect of age can be drawn.

Of the commercial varieties, N:Co.310 under normal potash-deficient conditions is considerably higher in starch than the other varieties, N:Co.292

being intermediate and Co.331 tending to be the lowest. The two P.O.J. varieties, which contain no S. Barberi "blood", were lower in starch than even Co.331.

Of the other agronomic factors considered, only the application of E.D.B. in the control of eelworm effected a reduction of starch.

#### Introduction

In 1926, E. Haddon<sup>6</sup> drew attention to the presence of starch in Uba Cane grown in Natal, and considered that it might be the cause of massecuite from this cane having an abnormally high viscosity, and being difficult to boil.

Feuilherade<sup>5</sup> in 1929, confirmed that juices from Uba cane were difficult to clarify and that the after-products were very viscous. He also found difficulty in crystallizing sucrose from the massecuite, and attributed these phenomena to high starch content.

Balch<sup>2</sup> working in Louisiana, mentioned the difficulty due to starch that the South African Sugar Industry was experiencing in the processing of juice from Uba cane. He pointed out that only small amounts of starch in raw sugars are required to reduce filtration rates during the refining process, in addition to the fact that it probably reduced the crystallisation rate.

Much more recently, Alexander<sup>1</sup> stated that the amount of starch found in South African Sugar products was quite considerable, appearing to rank among the highest in the world. This was true in spite of the fact that new varieties such as Co.301, N:Co.310 and Co.331 had completely replaced Uba.

Boyes<sup>3</sup> also regarded starch as one of the chief offenders in the poor filtrability of South African and Australian raw sugars and investigated enzyme hydrolysis as a method of removing it from the mixed juice.

It seems, however, highly desirable to reduce the starch content of the cane juice as much as possible even before the cane enters the factory, and consequently we have been investigating the effect of certain agricultural practices and phenomena on the starch content of the cane juice.

#### The Methods of Determination of Starch

(a) In Cane Juice: The method used was that due to Balch<sup>2</sup> which is based on the more involved method of Pucher and Vickery.<sup>7</sup> The simplification was made possible by the absence of interfering substances in the raw cane juice. The method is both sensitive and accurate<sup>2</sup> and was employed by Alexander<sup>1</sup> and Boyes<sup>3</sup>, but it only measures granular starch and not soluble starch.

100 ml. of the juice is measured into a 400 ml. beaker, 0.5 ml. glacial acetic acid added to cause

flocculation of colloids and improve filterability and 2 g. kieselguhr. This mixture is filtered through a 7 cm. Buchner funnel using a filter-pad of Whatman No. 41 filter paper and 1 g. kieselguhr which has the property of quantitatively retaining the starch granules. The filter-pad is washed with about 100 ml. cold water, 80 ml. 70 per cent alcohol solution, and finally with 80 ml. hot denatured alcohol. (The benzene washing<sup>1,2</sup> was found to be unnecessary.)

The filter-pad, including the paper, is transferred quantitatively to a 250 ml. beaker and 40 ml. neutralized calcium chloride solution (S.G. 1.3) is added. The beaker is covered with a watch-glass and the mixture boiled on a hot-plate for 18 minutes. After cooling, the mixture is transferred quantitatively to a 100 ml. volumetric flask, and made up to the mark with water. A further 1.3 ml. water are added to compensate for the volume of the solid matter (kieselguhr, etc.). After thorough shaking the mixture is centrifuged for 15 minutes and the clear liquid decanted.

An aliquot (usually 5 or 10 ml. depending on the starch concentration) is pipetted into a 100 ml. volumetric flask and about 50 ml. water, 5 ml. 2N acetic acid solution, 1 ml. 10 per cent potassium iodide solution and 10 ml. 0.01N potassium iodate solution are added in that order. The volume is made up to 100 ml. with water and, after shaking, the optical density is measured at a wavelength of 600 m $\mu$ . in a Coleman "Junior" Spectrophotometer using a blank containing the same reagents.

The optical density of the solution containing the starch-iodine complex (blue) is proportional to the concentration of starch. If absolute results are required, pure cane starch must be used in making up the standard solution. This is because starch granules are made up of two fractions, amylose and amylopectin, the proportion of which varies according to the plant source. It is the amylose which gives the deep blue colour with iodine, the amylopectin giving a red-violet colour, and hence equally concentrated solutions of starch from different plant sources give different colour intensities with iodine.

Relative results were obtained by using as a standard a stable solution of potato starch. In order to determine the factor required to convert these results to absolute values, a small amount of reasonably pure cane starch was isolated and the colour ratio determined between its iodine complex and that of potato starch. The procedure was as follows:

N:Co.310 juice was screened through a 325 mesh sieve, centrifuged and the solid matter suspended in a large volume of water. After leaving to stand about 1½ days, the supernatant liquid was carefully decanted and the precipitate resuspended in water. The bulk of the impurities was removed by allowing them to settle out and pouring off the water containing the suspended starch frequently. The resulting suspension of starch was centrifuged, washed with methyl alcohol and ether and dried in a vacuum over calcium chloride.

The starch sample so obtained was 90.65 per cent pure, based on dry weight. The purity was determined

by the conventional method of hydrolyzing with hydrochloric acid to glucose and determining this by the Luff-Schoorl Method. About 97 per cent of the granules were less than 10 microns diameter, the majority being 2-4 microns.

The colour ratio between iodized solutions of equal concentrations of pure potato and cane starch was 1.56. The potato starch originally weighed out to make up the standard solution was found to be 90.75 per cent pure. Since the original weight was used to construct the calibration curve, the factor by which to multiply all quoted results is 1.56 x 0.9075=1.42, in order to obtain the absolute values.

(b) In Complete Cane Stalks and Bagasse: The stalks are chopped up in a chaff-cutter, shredded in a Weddell Shredder, dried at 110°C. and ground as fine as possible in either our Wiley Cutter Mill or our C. & N. Junior Beater-Cross Grinding Mill.

Two methods can be used to determine the starch content of this ground material:

(i) About 10 g. dry sample is weighed into a 400 ml. beaker, 100 ml. calcium chloride solution (pH 7.0, S.G. 1.3) added and the mixture boiled gently for 18 minutes, after which it is filtered through a 7 cm. Buchner funnel using a similar filter-pad to that used for juice. After draining, the material is returned to the beaker and boiled with about 100 ml. water to extract any remaining solubilized starch, and the mixture refiltered. This is repeated and the combined filtrate made up to 500 ml. with water.

An aliquot, usually 25 ml., is transferred to a 100 ml. volumetric flask and the colour-developing reagents added as for juice samples. Since the filtrate has a yellow to brown colour, a blank must be determined on each unknown using only the 5 ml. 2N-acetic acid (since this destroys much of the colour) and omitting the potassium iodine and iodate solutions. The colour intensity is again measured at a wavelength of 600 m $\mu$ .

- (ii) About 4 g. dry sample is boiled with 80 ml. calcium chloride solution for 18 minutes, cooled, and the mixture transferred quantitatively to a 200 ml. volumetric flask and made up to the mark, after shaking to expel air bubbles. The volume of the sample (density about 1.3 g./ml.) is allowed for. The development and measurement of colour is the same as under (i), including the blank correction.
- (c) In Leaves and Sheaths: The leaves or sheaths are cut into small pieces, air or oven dried and ground as finely as possible in the C. & N. or Wiley Mill. The methods of starch determination are the same as for stalks and bagasse.

# The Degree of Starch-Cell Rupture Attained with our Wiley and C. & N. Mills

It was possible to obtain a finer product with the C. & N. Mill than with the Wiley Mill. To ascertain whether all the starch containing cells had been ruptured, thus allowing entry of boiling calcium chloride solution and resulting dispersion of the starch granules, samples of ground stalks and leaves were further ground using quartz and a pestle and mortar.

The results showed that a considerable portion (at least 34 per cent in one case and 45 per cent in another) of starch-containing cells in leaves had not been ruptured, but that apparently all of the corresponding cells in stalks had been ruptured.

Starch-containing cells in stalks and bagasse are incompletely ruptured in the Weddell Shredder, as grinding of the shredded material in one of the above mills gives higher results for starch, e.g.:

•	A	B	C
	(bagasse)	(stalks)	(bagasse)
Shredder sample Ground sample	210 ppm	344 ppm	514 ppm
	362 ppm	485 ppm	780 ppm

## The Relationship Between Starch and Sucrose Extraction

The fraction of juice expressed by a mill varies with the pressure of the rollers. In the case of our laboratory mill, the normal setting gave approximately 65 per cent sucrose extraction, but the pressure could readily be varied by slackening or tightening the nuts.

To obtain juice at different roller pressures, the sample of stalks was milled four or five times, each time increasing the pressure and collecting the juice fractions separately. Each fraction was analysed for its granular and total starch concentration, as well as its sucrose concentration. The resulting bagasse was shredded, and sucrose and starch determined.

In order to determine total starch (i.e. granular+soluble), twice the volume of alcohol is added to the juice sample, which is allowed to stand for at least an hour to precipitate all the soluble starch. The mixture is filtered through a Buchner funnel as for juice omitting the washing with cold water, which would re-dissolve the alcohol-precipitated starch. The subsequent procedure is exactly as for granular starch in juice.

The results obtained are shown in Tables 1 to 3.

Table 1

SAMPLE A Variety N:Co.376		-		Extraction of sucrose (%)	Starch in ju	uice (ppm) Total	Total starch in juice % cane	Extraction of starch (%)
1st Fraction	 			 49.3	51.4	63.4	0.00263	31.9
2nd Fraction	 			 	79.4	88.8	0.00154	
1st and 2nd Combined	 • •			 68.5	59.5	70.9	0.00417	50.7
3rd Fraction	 				64.5	73.8	0.00044	
1st, 2nd and 3rd Combined	 		٠.	 75.0	60.0	71.2	0.00461	56.0
4th Fraction	 		٠.	 1	64.5	78.1	0.00045	
1st—4th Combined	 			 81.2	60.4	71.9	0.00506	61.5

In computing the extraction of starch, the following results were also used:

Starch in bagasse=107 ppm; starch in bagasse per cent. cane=0.00317.

Starch per cent cane = 0.00506 + 0.00317 = 0.00823. The average ratio of granular to total starch is 0.83.

Table 2

SAMPLE B				Extraction	Starch in ju	uice (ppm)	Total starch	Extraction of
Variety N:Co.310				sucrose (%)	Granular	Total	% cane	starch (%)
1st Fraction	 		 	48.4	490	526	0.0207	18.3
2nd Fraction	 		 		762	778	0.0090	
1st and 2nd Combined	 		 	62.0	551	582	0.0297	26.2
3rd Fraction	 		 		727	751	0.0082	
1st, 2nd and 3rd Combined	 	• •	 	74.6	582	612	0.0379	33.5
4th Fraction	 		 	1 1	686	753	0.0045	
1st—4th Combined	 		 	81.4	591	624	0.0424	37.4

Starch in bagasse=2,207 ppm.; starch in bagasse % cane=0.0708; Starch % cane=0.0424+0.0708=0.1132.

The average ratio of granular to total starch is 0.94.

Table 3

SAMPLE C					Extraction	Starch in ju	iice (ppm)	Total starch	Extraction of
Variety N:Co.310			÷		sucrose (%)	Granular	Total	% cane	starch (%)
1st Fraction	• • • • • • • • • • • • • • • • • • • •	••		•••	 25.2	280	328	0.00708	13.0
2nd Fraction						363	431	0.00737	
1st and 2nd Combined					 44.7	317	373	0.01445	26.5
3rd Fraction					 ļ	356	421	0.00674	
1st, 2nd and 3rd Combined					 62.8	328	387	0.02119	- 39.8
4th Fraction						355	405	0.00401	
1st—4th Combined					 73.8	332	390	0.02520	46.2
5th Fraction					 1	398	434	0.00182	
1st—5th Combined					 78.5	336	393	0.02702	49.6

Starch in bagasse=880 ppm.; Starch in bagasse % cane=0.02746; Starch % cane=0.02702+0.02746=0.05448. The average ratio of granular to total starch is 0.85.

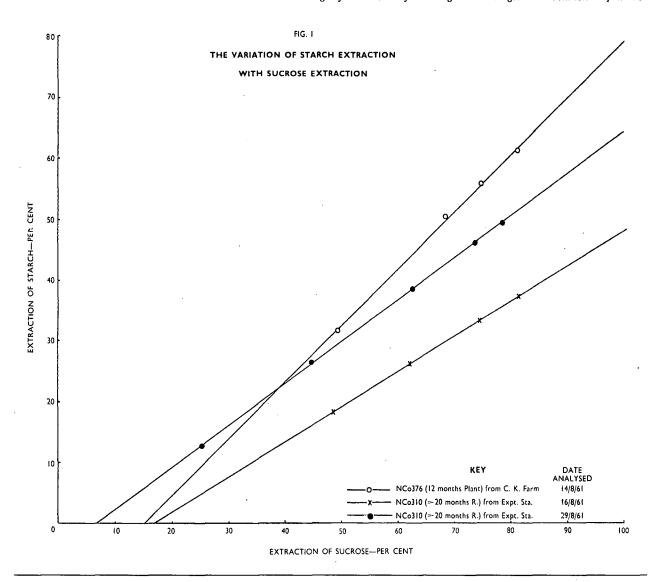


Figure 1 shows clearly that the extraction of starch is linearly dependent on sucrose extraction. It appears that the slope of the lines is dependent on the total starch per cent cane, the greater the latter the less the former, although more results are required to confirm this. Assuming that the linear relationship holds from 0 per cent starch extraction up to 100 per cent sucrose extraction, it can be seen that (i) starch extraction does not commence until a certain amount of sucrose has been extracted, (ii) by no means all the starch is extracted at 100 per cent sucrose extraction, especially if the starch content of the cane is high.

An examination of the tables shows that the starch concentration of the juice is only slightly dependent on sucrose extraction at high extractions (say greater than 70 per cent), becoming more strongly dependent at lower extractions. The sucrose extraction generally varied between 60 and 70 per cent using our laboratory mill, but, except where widely different varieties were milled, it seldom varied by more than 3 or 4 units in any one lot of samples. On an average, the measured

starch concentration increased by 0.43 per cent per unit sucrose extraction increment between 60 and 70 per cent, and hence the error introduced in ignoring the effect of small extraction variations is probably considerably smaller than other errors (e.g. sampling). It is generally necessary to apply a correction for extraction only when results from lots of samples milled at different times or from widely different varieties are being compared, and it can only be very rough, as an accurate one would entail determining a correction curve for each sample.

# The Effect of Time-Lapse Between Cutting and Milling on the Starch Content of Cane Juice

Four samples, as nearly identical as possible, were obtained by cutting each of, say, 20 sticks into four equal lengths, and making four composite heaps.

The first sample was milled and the juice analysed immediately. The other samples were milled after leaving 3, 5 and 10 days respectively. The results are recorded in Table 4.

Variety N:Co.310	able 4			
Time (days)	o	3	5	10
Granular starch (ppm) Total starch (ppm) Brix Granular starch % Brix	234 — 18.6 0.126	203 217 19.0 0.107	195 206 19.0 0.103	56 58 19.3 0.029

The results of a repeat experiment are given in Table 5.

Table 5 Variety N.Co 210

variety N.Co.310	, ==-		. —	
Time (days)	0	2	4	8
Granular starch (ppm) Total starch (ppm) Brix Granular starch % Brix	292 331 19.1 0.153	256 281 19.8 0.129	208 217 19.7 0.106	67 69 19.7 0.034

Inspection of the above tables shows a gradual decrease in the starch concentration of the expressed juice up to 5 or 6 days, and thereafter a rapid decrease. On the contrary, the brix remains constant or increases slightly. Enzymatic breakdown to glucose could be responsible for the starch decrease.

### The Distribution of Starch in the Mature Cane Stalk

Preliminary results indicated that the top half of the cane stalk contained more starch than the lower half and also that the nodes had a considerably higher starch concentration than the internodes.

To determine the variation of starch concentration along the length of the stalk, each stick in a sample of, say, 10-20 sticks was cut into 6 in. or 1 ft. lengths starting at the top, the corresponding portions of each stick being mixed, ground and analysed for starch. The results are recorded in Table 6.

Table 6

•	ADIC	TV N	O. 11			STA	ARCH % CA	NE	STAR	ROSE	
v	AKIE	TY N:	(Co. 3)	10		Sample A 19.10.61	Sample B 23.10.61	Sample C 29.12.61	Sample A 19.10.61	Sample B 23.10.61	Sample C 29.12.61
1st 6 inches						0.175	0.117	0.057	2.30	1.54	0.87
2nd 6 inches	• •				• •	0.247	0.162	0.117	1.52	1.00	0.86
2nd foot				• •		0.194	0.141	0.128	1.06	0.77	0.77
3rd foot	• •					)	0.097	0.102	1	0.52	0.59
4th foot							0.062	0.061	] }	0.33	0.35
5th foot						<b>▶</b> 0.079	0.047	0.054	> 0.42	0.26	0.31
6th foot							0.034	0.049		0.18	0.28
7th foot			• •		••		0.043	0.056*	11	0.22	0.31*
8th foot			• •		• •	0.102*	0.052*		0.54*	0.27*	

<sup>\*</sup>The remainder of the stalk (of varying length).

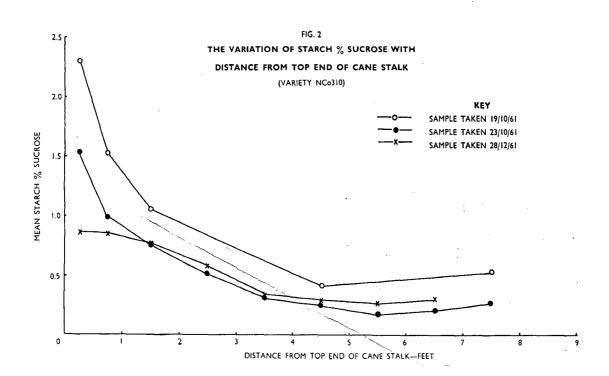


Figure 2 shows clearly that starch per cent sucrose decreases rapidly in the first 2 or 3 ft. of cane. The starch concentration, however, increases to a maximum between 9 and 15 inches and then decreases rapidly to a minimum between 5 and 6 feet.

From these results, it appears that the top two feet would have to be cut off in order to reduce the starch content of the expressed juice to any appreciable extent. This would have reduced the overall starch concentration of samples "A" and "B" by 27 per cent and of sample "C" by 16 per cent. Cutting off the top foot would have caused a reduction of 12 and 2 per cent respectively.

To determine the ratio between the starch concentration in the nodes and internodes, the nodes were cut out carefully with a hacksaw, and half an inch of cane between the nodes and internodes was discarded. In one sample the buds were also separated from the nodes to ascertain whether they differed in starch concentration from the nodes. The different portions were ground and analysed for starch.

It was found that on a dry weight basis the concentration of starch in the nodes is about 3.5-4.0 times that of the internodes, and the concentration in the buds and nodes is approximately the same.

## The Influence of Fertiliser Application on the Starch Concentration of Cane Juice

Since a large number of field experiments (Regional Fertiliser Trials etc.) was in existence, it was thought expedient to use these trials to determine the effect of fertiliser application on starch in cane juice. Accordingly, starch analyses were made on the juice from all such samples which were sent in for sucrose analysis during 1961. In most instances, composite samples of juice were taken and only main effects considered.

# The Effect of Potash Application on the Starch Concentration in the Juice

Although in most trials the starch content of the juice showed a definite decrease with increasing potash application, some showed no decrease or even a small increase. The growth data pointed to a correlation between the decrease in starch and the increase in growth due to potash application. The starch results have, therefore, been divided into:

- (a) those where there is a significant growth response
- (b) those where there is no significant growth response

Note.—Wherever "significance" is mentioned, statistical significance at the 5 per cent level is implied.

Table 7 shows the starch results where the increase in yield is *significant* for one or both increments.

Table 7

			Starc	n Concer	tration (	(ppm)	Per	cent Increas	e in
Code Number	Date Anal- ysed	Variety	K1 0 lb /a. K <sub>2</sub> 0	K2 150 lb /a. K <sub>2</sub> 0	K3 300 lb./a. K <sub>2</sub> 0	Av. of K2 and K3	Starch content of juice	Tons cane per acre	K <sub>2</sub> 0 content of juice
RFT 29/57/1R1 RFT 28/57/1R1 RFT 14/56/1R2 RFT 40/57/1R1 RFT 39/57/1R1 RFT 39/57/1R1 RFT 44/58/1R1 RFT 13/56/1R2 EX 16/58/2R1 RFT 36/57/1R1 RFT 42/57/1R1 RFT 43/57/1R1	8/6 13/6 15/6 23/6 29/6 27/7 11/8 11/9 9/10 8/11 17/11	N:Co.376 N:Co.376 N:Co.376 N:Co.292 N:Co.310 N:Co.376 N:Co.376 N:Co.376 N:Co.292 N:Co.376	215 288 120 336 513 204 328 296 269 295 303	191 214 121 267 416 131 257 179 299 321 228	159 203 82 309 433 146 193 140 265 314 185	175 208 102 288 424 139 225 159 282 318 206	-18.6 -27.8 -15.0 -14.3 -17.2 -32.2 -31.4 -46.1 + 4.8 + 7.5 -32.0	+55.7 +14.3 + 9.8 +53.5 +61.6 +45.0 +33.4 + 9.6 +35.6 +67.8 +51.5	+100.0 + 69.8 + 66.5 +213.5 +161.0 + 49.7 + 96.8 +305.6
Averages			288	239	221	230	-20.1		
*EX 4/51/1R4	17/8 18/8	N:Co.310 N:Co.310	459 1335	409 714	356 572	382 643	-16.7 -51.8	+30.0 +176.4	+ 62.7 +183.3
Averages			897	564	464	513	-42.8		

<sup>\*</sup>For these trials, the levels of Potash were 0, 100 and 200 lb. K<sub>2</sub>O/acre respectively.

Another interesting fertiliser trial (Nitrogen and Potash only), which employed variety N:Co.310 and received five levels of potash (0-400 lb. K<sub>2</sub>O per acré) in 100 lb. increments, gave analyses of 524, 411, 381, 331 and 308 ppm starch respectively in the juice. Since the average of the last four starch results was

358 ppm, the percentage increase in the starch content of the juice was -31.3, as against the percentage increase in yield of +88.3, which was highly significant.

Table 8 records the starch results where the yield response was not significant.

Table 8

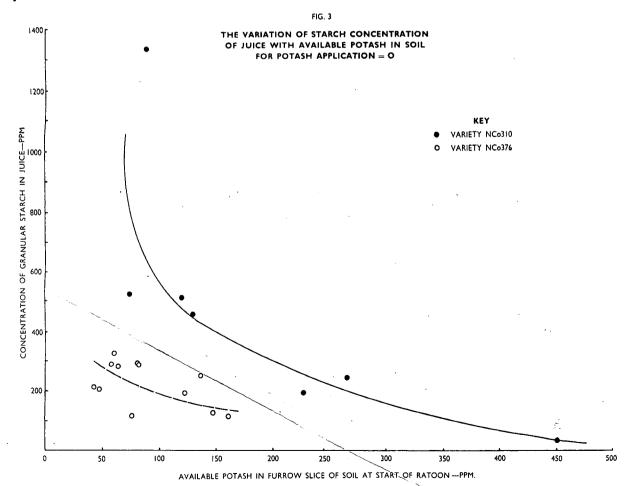
			Starcl	1 Concer	itration (	(ppm)	Per	cent Increas	se in
Code Number	Date Anal- ysed	Variety	K1 0 lb /a. K <sub>2</sub> 0	K2 150 lb./a. K <sub>2</sub> 0	K3 300 lb./a. K <sub>2</sub> 0	Av. of K2 and K3	Starch content of juice	Tons cane per acre	K₂0 content of juice
RFT 33/57/1R1 RFT 35/57/IR1 RFT 50/58/IR1 RFT 26/57/1R1 RFT 32/57/1R1 RFT 34/57/1R1 EX 2/51-57/2R1	13/6 13/7 3/8 4/8 10/8 24/8 16/11	N:Co.310 N:Co.310 N:Co.310 N:Co.376 N:Co.376 N:Co.310 N:Co.376	31 120 199 195 284 246 257	32 127 228 157 232 231 235	30 101 252 217 227 206 211	31 114 240 187 229 219 223	0 - 5.4 +21.0 - 4.1 -19.2 -11.2 -13.2	-0.9 -2.7 +0.8 -3.6 +1.8 -2.3 +2.9	+ 26.9 + 40.4 + 18.1 + 224.2 - 1.6 + 15.1
Averages			190	177	178	178	- 6.3		

An inspection of the above tables reveals that, where there is a significant increase in yield due to potash, there is in all cases except two a considerable decrease in the starch content of the juice. When the increase in yield is not significant, however, there is also on an average a slight, though possibly not significant, decrease in starch.

The above observations are exceptionally well illustrated in the case of the trial EX 5/51/1R4, which showed an extremely large increase in yield due to potash application. Starch analyses of samples from all the individual plots were made, and responses were statistically evaluated. The results are given in the 3-way table 9.

Table 9
Starch Concentration in Juice in ppm.

	P0	P1	P2	K0	K1	K2	Mean
 N0	 1112	816	871	1398	805	597	933
NI	 886	885	873	1336	707	601	881
N2	 783	821	816	1273	629	518	807
K0	 1422	1281	1304	1336	714	572	874
K1	 760	680	702				!
K2	 600	562	555		ndard E		
Mean	 927	841	853	土4	3.2 ppr	n.	



In this extreme case of potash deficiency, the average decrease of 382±30.5 ppm starch per increment of 100 lb. K<sub>2</sub>O per acre is significant, but it is to be noted that this decrease is far greater in the first increment of potash than in the second. Thus KO-K1=622± 61.1 ppm starch which is highly significant, but K1- $K2=142\pm61.1$  ppm. starch, which is not significant.

In passing, it is of interest to note that the average decrease of 63±30.5 ppm starch per increment of 100 lb. nitrogen (N) per acre is not significant, but could indicate a trend. Also, the decrease in starch due to phosphate application is not significant, in spite of the significant increase in yield of  $3.80\pm1.75$ tons cane per acre per increment of 100 lb. P<sub>2</sub>O<sub>5</sub> per

The relationship between the starch concentration in the juice and the potash concentration in the soil on which the cane was grown is illustrated in graphical form in Fig. 3.

This graph not only provides strong additional evidence for the efficacy of potash in the reduction of the starch content of the cane, but also brings out very well the difference in starch content between varieties N:Co.310 and N:Co.376, the concentration of starch in N:Co.310 at any particular soil-potash level being considerably higher than that in N:Co.376. These varietal differences will, however, be discussed later. It is noteworthy that even a variety (N:Co.310) which is normally found to be very high in starch at

ordinary soil-potash levels (50-250 ppm) had a very low starch content when grown on soil having a soilpotash level of 450 ppm. In fact, in all instances where cane was grown on very high potash-soils, the starch concentration of the juice was found to be very low, e.g. (i) N:Co.376 from the potash-rich Umhlatuzi alluvial soils, gave juice with only 5-15 ppm starch, (ii) the Illovo Sugar Estates' lysimeter tanks which were heavily enriched with potash produced N:Co.376 with very low starch (15-25 ppm in juice), (iii) only 25-30 ppm starch was found in N:Co.310 and N:Co. 376 grown on Umfolozi alluvial soil.

It is known that nodes contain 3.5 to 4.0 times the concentration of starch of internodes and that wellgrown cane generally has less nodes per unit length than poorly-grown cane. This undoubtedly is one factor responsible for the reduction of starch due to potash application, but it by no means accounts completely for the very large decrease in starch usually encountered.

## The Effect of Nitrogen Application on the Starch Concentration of the Juice

Since in many cases there were large yield responses to Nitrogen application, it was thought that this plantfood might also have a reducing effect on starch. The results obtained for the different field trials are summarised in Tables 10 and 11.

Table 10 Increase in yield significant for one or both increments of N.

			St	arch Cor	centratio	on (ppm)	١	Per ce	ent Increase	in
Code Number	Date Anal- ysed	Variety	NI 0 lb./a. x N	N2 100 lb./a. x N	N3 200 lb./a. x N	N4 300 lb./a. x N	Av. of N2, N3 etc.	Starch content of juice	Tons cane per acre	K₂0 content of juice
RFT 28/57/1R1	13/6 13/7 27/7 3/8 4/8 10/8 24/8 17/11	N:Co.376 N:Co.310 N:Co.376 N:Co.310 N:Co.376 N:Co.376 N:Co.310 N:Co.293	212 188 167 242 205 240 279 276	227 120 184 223 182 240 263 264	211 122 147 209 184 217 170 196	241 86 126 276 195 287 195 170	226 109 152 236 187 248 209 210	+ 6.6 -42.0 - 9.0 - 2.5 - 8.8 + 3.3 -25.1 -23.9	+18.8 + 7.0 +53.0 + 6.0 +63.4 +96.1 +35.2 +22.3	-11.1 -40.8 -18.6 0 + 8.9 - 8.7 -24.4
Averages			226	213	182	197	197	-12.8		
*EX 16/58/2R1 *EX 2/51-57/2R1	11/9 16/11	N:Co.376 N:Co.376	202 225	210 275	157 232	_	183 253	- 9.3 +12.5	+51.6 +44.6	-43.6 -29.1
Averages			213	242	194	_	218	+ 2.3		
EX 4/51/1R4	17/8	N:Co.310	438	407	424	_	415	- 5.2	+ 7.1	-11.9

It is interesting also to include the results of the Nitrogen-potash trial, mentioned earlier under "the effects of potash", in which N:Co.310 was used and five levels of Nitrogen (0-400 lb. per acre in 100 lb. increments) were applied. The starch results obtained

were 486, 422, 461, 369 and 335 ppm respectively. Averaging the last four, the percentage increase in starch was -18.3, and in yield was +89.8, which was again highly significant.

Levels of N: 0, 150 and 300 lb. N per acre respectively. Levels of N: 100, 200, 300 and 400 lb. N per acre respectively. x R.F.T. trials—urea only; EX trials—sulphate of ammonia only.

	Table 11	l
Increase i	n yield not	significant.

			Starch Concentration (ppm)					Per cent Increase in		
Code Number	Date Anal- ysed	Variety	N1 100 lb./a. x N	N2 200 lb./a. x N	N3 300 lb./a. x N	N4 400 lb./a. x N	Av. of N2, N3, etc.	Starch content of juice	Tons cane per acre	K₂0 content of juice
*RFT 29/57/1R1	8/6 13/6 15/6 23/6 29/6 11/8 9/10 8/11	N:Co.376 N:Co.310 N:Co.376 N:Co.292 N:Co.310 N:Co.376 N:Co.292 N:Co.376	194 33 95 281 463 310 350 287	206 37 144 256 451 322 331 350	186 29 85 367 434 270 241 385	202 32 79 325 418 230 236 392	198 33 103 316 434 274 269 376	+ 2.1 0 + 8.4 +12.5 - 6.3 -11.6 -23.1 +31.0	+ 7.3 + 0.4 + 4.0 -10.7 - 3.1 + 4.8 - 0.2 + 4.5	- - - + 3.2 -31.9 -25.8 +17.8
Averages			252	262	250	239	250	- 0.8		
‡EX 5/51/1R4	18/8	N:Co.310	933	881	807	_	844	- 9.5	+29.4	-29.8

- \* Levels of N: 0, 100, 200 and 300 lb. N per acre respectively.
- ‡ Levels of N: 0, 100 and 200 lb. N per acre respectively.
- x R.F.T. trials—urea only; EX trial—sulphate of ammonia only.

The majority of trials (9 out of 12) having significant yield responses to Nitrogen show a corresponding decrease in starch concentration of the juice, although generally not to the same extent as with potash. The decrease in starch may, here, be simply due to the increase in growth causing fewer nodes per unit length.

It appears from Table 11 that nitrogen application not accompanied by increase in yield does not on the average show a tendency to decrease the starch. The result for EX 5/51/1R4 (discussed previously) is interesting, however, in that a large increase in yield was accompanied by a decrease in starch, although both these effects were statistically not significant.

On the whole, the results indicate that application of nitrogen contributes to a certain extent towards the

reduction of starch, although the effect is completely overshadowed by that of potash.

# The Effect of Phosphate Application on the Starch Concentration of the Juice

In general the application of superphosphate did not cause a significant response in yield, probably due to the already high level of phosphate in most soils on which cane has been grown as a result of repeated fertiliser application. There was, however, a possibility that phosphate might have some influence on starch, especially as it plays a vital part in starch synthesis in the plant.

The results are summarised in Tables 12 and 13.

Table 12

Increase in yield significant for one or more increments.

		_	Starch Concentration (ppm)			Per cent Increase in			
Code Number	Date Anal- ysed	Variety	P1 0 lb./a. P <sub>2</sub> O <sub>5</sub>	P2 100 lb./a. P <sub>2</sub> O <sub>5</sub>	P3 200 lb./a. P <sub>2</sub> O <sub>5</sub>	Av. of P2, P3	Starch content of juice	Tons cane per acre	K₂O content of juice
RFT 29/57/1R1	8/6 27/7 9/10 8/11	N:Co.376 N:Co.376 N:Co.292 N:Co.376	183 166 277 328		206 165 288 303		+12.6 - 0.6 + 4.0 - 7.6	+15.0 +18.0 + 9.2 +44.5	+ 7.6 + 1.7 -14.6
Averages			238		241		+ 1.3		
EX 4/51/1R4 EX 5/51/1R4	17/8 18/8	N:Co.310 N:Co.310	416 927	438 841	447 853	442 847	+ 6.4 - 8.6	+ 8.6 +26.9	$-10.2 \\ -5.2$
Averages			671	639	650	644	- 4.0		

	Table 1	3
Increase in	n yield no	t significant.

and the organical in th									
			Starch	Concen	tration (	ppm)	Per c	ent Increase	in
Code Number	Date Anal- ysed	Variety	P1 0 lb./a. P <sub>2</sub> O <sub>5</sub>	P2 100 lb./a. P <sub>2</sub> O <sub>5</sub>	P3 200 lb./a. P <sub>2</sub> O <sub>5</sub>	Av. of P2 P3	Starch content of juice	Tons cane per acre	K <sub>2</sub> O content of juice
RFT 28/57/1R1  RFT 33/57/1R1  RFT 14/56/1R2  RFT 40/57/1R1  RFT 39/57/1R1  RFT 35/57/1R1  RFT 35/57/1R1  RFT 32/57/1R1  RFT 32/57/1R1  RFT 32/57/1R1  RFT 34/57/1R1  RFT 26/57/1R1  RFT 43/57/1R1	13/6 13/6 15/6 23/6 29/6 13/7 3/8 10/8 11/8 24/8 4/8 17/11	N:Co.376 N:Co.310 N:Co.376 N:Co.292 N:Co.310 N:Co.310 N:Co.376 N:Co.376 N:Co.376 N:Co.376 N:Co.376 N:Co.376	210 33 116 280 464 120 260 256 297 218 162 224		238 28 106 309 429 92 201 222 262 235 199 222		+13.3 -15.2 - 8.6 +10.4 - 7.5 -19.2 -22.7 -13.3 -11.8 + 7.8 + 22.8 - 0.9	+ 7.9 + 5.1 + 2.7 +20.1 + 1.8 + 3.3 + 0.7 + 5.3 + 1.0 + 9.0 +13.3 + 2.1	
Averages			220		212	_	- 3.6		
EX 16/58/2R1 EX 2/51-57/2R1	11/9 16/11	N:Co.376 N:Co.376	193 258	194 230	207 223	201 226	+ 3.9 -12.2	+ 4.9 +12.4	- 9.5 + 8.0
Averages			225	212	215	213	- 5.3		

No conclusion can be drawn from these figures. There seems to be no correlation between the percentage decrease in starch and the percentage increase in growth due to phosphate application. Similarly, there seems to be no proof for the contention that phosphate applications increase the starch content of the juice.

### The Effect of Irrigation on the Starch Content of Juice

Advantage was taken of an irrigation trial laid down by the Illovo Sugar Estates to ascertain the effect of irrigation on the starch content of cane juice. The juice of samples which were sent in periodically for sucrose analysis, was compounded for the main effects WO (no irrigation), W1 (irrigation to 50 per cent of difference between wilting point and field capacity) and W2 (irrigation to field capacity) and analysed for starch. The results obtained were as follows:

Table 14

Variety: N:Co.310		*Starch Con	*Starch Concentration in Juice (ppm				
Date A	nalysed	W0	WI	W2			
21/3/61		542	449	259			
1/5/61		400	· 385	249			
29/5/61		288	321	281			
4/7/61		191	161	123			
8/8/61		281	258	238			

<sup>\*</sup>The sucrose extraction varied considerably from one analysis to the next and, hence, all results have been corrected (as nearly as possible) to 60 per cent sucrose extraction.

Inspection of the table shows that the decrease in starch due to irrigation is very large in young cane, becoming progressively smaller as the cane matures. It is interesting to note that there was a marked decrease in starch concentration with age for dryland cane, and also for treatment W1, although to a lesser extent. This decrease was, however, absent when the cane was irrigated to field capacity.

The response in yield to treatment W1 at harvesting was highly significant, but no further significant response was caused by additional irrigation. It is probable that the reduction in starch at each stage is due to more luxuriant growth as a result of irrigation, with consequent fewer nodes per unit length of cane and increased uptake of plant-foods resulting from their greater availability.

#### The Variation of Starch in Cane Juice with Variety

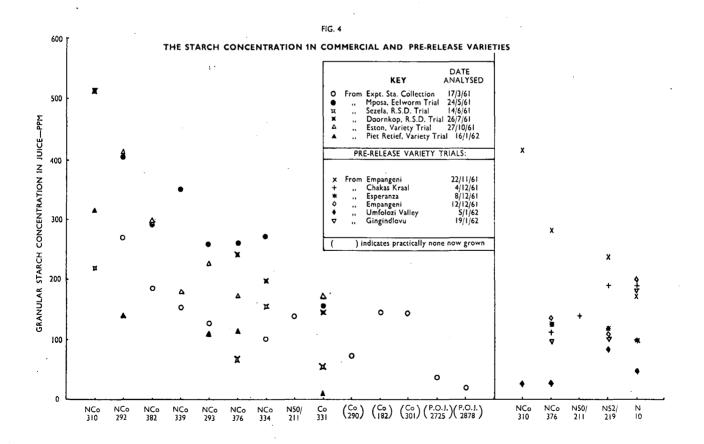
A number of workers (e.g. (1), (2), (4) and (8)) have investigated varietal differences in the starch content of cane and cane juice and have found that, in some cases, considerable differences occur. Preliminary work in this laboratory also indicated varietal differences.

Dutt and Narasimhan<sup>4</sup> surveyed semi-qualitatively the occurrence of starch in a number of species and varieties of Saccharum. They found that in Saccharum Officinarum and S. Robustum no starch is stored in the cells, whereas the S. Sinense varieties show fair quantitites of starch and in S. Barberi and S. Spontaneum quite large accumulations occur, although some varieties of the latter contain very little.

In our laboratory juice from several varieties of the different species quoted above was analysed quantitatively for starch and the results obtained are recorded in Table 15.

Table 15

			Starch Co	oncentration in Juic	ntration in Juice (ppm)		
Species		Variety	Collection E3, analysed 15/3/1961	Collection D2, analysed 13/12/1961	Collection D2 analysed 12/1/1962		
S. Officinarum	••	Badila	58 25 25 21 29	114 52 57 59 96			
S. Robustum		Yellow Caledonia	34 47 61	112 54 38 19			
S. Sinense	••	Agaul Yoh-tan-san Kavangire	21 325 203 191	736 907 720			
S. Barberi		Uba Sin Nombre Cape selection Townsend's selection	590	1,259 697 907	606 — —		
S. Spontaneum .	· · · · · · · · · · · · · · · · · · ·	Pasoeroean		907 427 54 — — —	262 163 535 2,117		



It can be seen from the table that the S. Officinarum and S. Robustum varieties all have a low but definite starch content. S. Sinense varieties are high and S. Barberi varieties very high in starch, confirming Dutt and Narasimhans' observations. S. Spontaneum varieties are very variable, their starch contents ranging from low to extremely high values.

The commercial varieties now being grown in South Africa are all derived from three of the above species viz. S. Officinarum, S. Barberi and S. Spontaneum, and therefore, it is of interest to note the proportion of "blood" from these three species in each. This is given in Table 16.

Table 16

	% S.	% S.	% S.
	Officinarum	Barberi	Spontaneum
N:Co.292, 293, 310, 334, 339, 376 N:Co.382 *Co.301 Co.331 *P.O.J. 2725, 2878	59.4 68.7 50.0 50.0 87.5	12.5 18.8 37.5 25.0	28.1 12.5 12.5 25.0 12.5

\*Practically none now grown.

Figure 4 shows the results of starch analyses on the juice from the varieties in Table 16 and two pre-release varieties, taken from samples sent in for sucrose analyses from variety and other trials.

Inspection of the figure reveals that N:Co.310 generally has a considerably higher starch concentration than other varieties, confirming the observations from Figure 3.

It is noteworthy, however, that in the one case where its starch concentration was low and comparable with that of other varieties, the cane was grown on Umfolozi alluvial soil which is high in available potash.

Although there is considerable variation from one trial to another, the varieties N:Co.293, 334, 339, 376 and N50/211, Co.281, Co.301, and the pre-release varieties N52/219 and N10 all seem on the whole to have approximately the same level of starch (about half that of N:Co.310). N:Co.292 appears to be intermediate between these and N:Co.310 and the remaining Co. varieties tend to be lower. The low level of starch in the P.O.J. varieties is interesting in that they have no S. Barberi "blood" which would here appear to be the predisposing factor for high starch.

Additional evidence that N:Co.310 has a considerably higher starch content than N:Co.376 is provided by the average figures for starch analyses on ground stalk samples—3,500 and 1,900 ppm (on dry weight) respectively. These samples were taken from the Experiment Station collection during late January and early February this year.

It may be concluded that all the widely-grown commercial varieties contain considerable amounts of starch on normal sugar-belt soils.

## The Influence of some other Factors on Starch in Cane Juice

It has been claimed by some workers that the pH of the soil influences the starch content of the cane. This claim could not be substantiated. Also, there appears to be no correlation between the available  $P_2O_5$  in the soil and starch in the cane, which is as would be expected considering the inconclusive results of phosphate application quoted earlier.

The effect of age on starch in cane juice is shown by the following analyses on 2 year, 3 year and in one case 4 year-old cane, which in each case was growing together in the same field:

- (a) Cane from U.V.S. (Co.331): 2 years—165 ppm, 3 years—45 ppm.
- (b) Cane from Inglemere Estate (N:Co.310): 22 months—320 ppm, 32 months—120 ppm.
- (c) Cane from Frasers (N:Co.310): 19 months—358 ppm, 29 months—284 ppm.
- (d) Cane from Compensation (N:Co.310): 2 years—144 ppm, 3 years—212 ppm, 4 years—219 ppm.

Although there is a considerable decrease in starch with age in the first three cases, the last result is in sharp contradiction to the others and, therefore, no definite conclusion can be drawn.

Ratoon Stunting Disease showed no tendency to change the starch concentration of the juice.

Application of E.D.B. in the chemical control of eelworm reduced the starch in the juice considerably in two eelworm trials investigated. Here again, this reduction is probably due to better growth and uptake of potash etc., as a result of the reduction of eelworm activity.

No significant change in starch due to lodging was observed in the one case investigated.

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Mr. W. J. G. Barnes, in the chair, said the question of starch in sugarcane was most important and the author had set out clearly the field of investigation he had explored.

Dr. Douwes Dekker stated that starch was one of the main impurities in our sugar which adversely affected filtration in the refinery. Phosphate content, wax, probably gums and silica content were also responsible for poor filtration, but starch was one of the most important impurities.

He was happy to see from the paper that the better the cane was treated in the field the lower was the starch content. As it was in the interest of growers to produce the best possible cane the starch content would tend to be reduced automatically.

A conclusion drawn in the paper was that delay between cutting and milling led to a reduction in starch content but other factors became involved, and the gum content increasing, would make the recovery of sugar more difficult. It was not good practice to keep cane for a period before milling.

He pointed out that the sucrose extraction by the laboratory mill reached 60 to 70 per cent and while the curves shown in Figure 1 were linear in this range, at an extraction of 92 to 95 per cent this no longer applied, and the curve would take a steep rise upwards. It was to be hoped that further tests at high extractions could be made.

The effect of nitrogen, phosphate and potash were very important. A correlation between phosphate and starch content in sugar had been found in Queensland but in Natal the correlation was not so evident.

Mr. du Toit said he was pleased the S.M.R.I. found this work of great interest. It was in fact an attempt to find the factors responsible for high starch content of sugar so that those factors might be eliminated. We knew that one of our most popular varieties was high in starch content, as were other varieties such as Uba. The author had demonstrated big differences between varieties in starch but big differences were apparent within a variety. It was very important that he had found low starch content in N:Co.310 in some cases, as this variety was usually high in starch. These low starch figures were shown in the graph to be associated with high potash content of soil, but the author was careful not to claim that high potash content was the only reason for this.

Dr. Douwes Dekker had drawn attention to the fact that starch rapidly decreased a few days after the cane was cut, but he had correctly stated that a delay in milling was not a practice to be recommended, and the author had not advocated this. He thought this reduction in starch content might be due to enzymatic action, and at the refinery it had been observed that sugars on long storage filtered well. This should be further investigated.

**Dr. Douwes Dekker** considered that the increased filtrability of sugar on long storage could not be due to enzymatic action on starch because the enzymes present in the juice would be destroyed in the processing of raw sugar.

The author had shown that a large proportion of the starch present in the juice was in granular form. This fact was being made use of in a method now being studied to remove starch from mixed juice by using centrifugal force. So far these experiments had not been successful as the large scale continuous centrifuge was not so effective as the small laboratory type, which could remove some 85 to 90 per cent of the starch, as against the large machine's 30 to 50 per cent. It was hoped that the efficiency of the machine could be improved considerably.

Mr. Main related that in India, using the de Haan process, complete failure was experienced in carbonatation factories owing to the starch present. After abandoning the process and going back to dry liming, improvement was enormous, the starch content in factory liquors and sugars being greatly improved. It was found that the same applied in sulphitation factories, provided bluish, magnesium-containing lime, with a lower than usual Ca0 content, was used.

**Dr. Brett** asked if there was any seasonal variation in starch content.

Mr. du Toit said it was difficult to separate season and age, but if starch content of juice at the factories was determined, this could be done throughout each season and seasonal contents be thus compared. A seasonal difference had been found by the S.M.R.I.

**Dr. Douwes Dekker** said it was of interest that Felixton sugar was known as a low starch content sugar and he noted that this was probably due to the fact that it was produced from cane grown on alluvial flats with a high potash content. This was true of Umfolosi and Pongola sugars but they were not so low as Felixton.

Mr. Main stated that relative to magnesium content of lime the only lime he could find some years ago with this desirable property, was Port Shepstone lime which could not be obtained in quantity.

Mr. Dicks thought that in Figure 1 of the paper the straight line approximation was appropriate only with the limits of the present investigation and that, as pointed out by Dr. Douwes Dekker, linear interpolation in the upper region as well as in the lower region presented perhaps a false picture of the true relationship over the whole range. Furthermore would it not be better to combine the two lines showing the trend for N:Co.310? The N:Co.310 was ratoon cane and the N:Co.376 plant cane and one might thus confuse varietal differences with plant and ratoon differences.

Mr. Wood said the straightness of the line within the limits explored seemed to point to its being straight outside those limits. But he would not assume this to be the case. In reply to Mr. Dicks' query about the two lines shown for N:Co.310 and also ratoon and plant cane figures being shown, he felt more could be obtained by showing the result of each experiment seperately.

Mr. du Toit pointed out that the author had given the graph only to show the liniarity in the range studied and no real conclusion was drawn by interpolating the lines further. No claim was made from the graph that one variety was higher in starch than another.