

RECENT MEALYBUG INVESTIGATIONS

By J. DICK

In a paper read to this Association in 1953,³ the results were given of a number of experiments on the effects of infestation by the sugarcane mealybug, *Saccharicoccus (Trionymus) sacchari* (Ckll.). It was shown that the presence of mealybugs on setts at the time of planting had a detrimental effect on germination and early growth, and that some part of this effect might persist even if the insects were removed before planting. This led to the hypothesis that the removal of juice during feeding was not the only mechanism through which mealybugs were able to affect the plant. The object of the present note is to discuss the results of more recent investigations in which this hypothesis has been further examined by studying the germination and early growth of cuttings inoculated with an extract prepared by grinding the bodies of mealybugs.

Preliminary trials, of which an example was quoted in the above-mentioned paper, showed that, when cuttings were inoculated with this extract, germination was significantly reduced. Of three varieties tested, Co.301 was the most severely affected and this variety was therefore used in subsequent trials.

In the next experiment, setts were inoculated with untreated extract; with extract prepared from mealybugs which had been dipped in formaldehyde solution in order to destroy any fungi or bacteria which might be present on their surface; and with extract autoclaved after preparation in order to destroy all organisms which might be present. Four replications, each consisting of ten single-budded cuttings, were planted for each treatment and for the control, in which the cuttings were inoculated with distilled water. Figures, indicating the numbers of buds which had germinated after fourteen days, are shown in Table I.

TABLE I

Numbers of Buds Germinating

Control	Un-treated Extract	Auto-claved Extract	Formal-dehyde Extract
9	2	9	9
7	2	9	7
7	1	8	4
10	4	7	9
33	9	33	29

Significant differences between totals: 10 at 19 : 1, 14 at 99 : 1.

This result suggested that the depression in germination might be due to some type of infection, presumably fungal or bacterial, introduced in the mealybug extract. Setts of cane were therefore surface-sterilised with alcohol and were then inoculated by dipping the cut ends in the extract. A control series was surface-sterilised and dipped in distilled water. Both series were then kept in glass jars for four days at 30°C. At the end of this period, both buds and root primordia in the control series had sprouted, while those in the inoculated series, after starting to develop, had apparently died. The cut ends in this series were covered with a luxuriant growth of fungi, apparently mainly saprophytic. It was considered that, if infection had caused failure to germinate, the causal organism should have grown into the stick and should have been present at the base of the bud. Pieces of tissue from this region, where some discoloration was evident, were removed, surface-sterilised, planted in maltose-peptone liquid medium and incubated for two days at 30°C. At the end of this period, the only organism growing in the culture medium was apparently a species of yeast.

To discover whether this yeast might be responsible for depression in germination, setts were planted after inoculation with yeast, mealybug extract or distilled water. Results for one such test are given in Table II.

TABLE II

Numbers of Buds Germinating

Water	Yeast	Mealybug
10	8	3
10	9	5
9	8	2
10	8	7
39	33	17

Significant differences between totals: 11 at 19 : 1, 17 at 99 : 1.

These figures would appear to indicate that the yeast was of no significant importance as a cause of poor germination. However, two factors might have influenced the result, namely concentration, which might have been lower in the yeast culture than in the extract, and the length of the cuttings, which might have had some bearing on the time required for infection to travel from the cut ends of the sticks to the base of the buds.

Consequently, single-budded cuttings two, four or six inches in length were inoculated by dipping the ends in a culture of the yeast which had been allowed to grow for a week at 30°C., or in the mealybug extract, undiluted or diluted with three or fifteen parts of distilled water. For completeness, a control, dipped in distilled water, was inserted for each length and dilution. Ten cuttings were planted for each treatment and two repetitions of the entire experiment were carried out. In Table III, which shows the numbers of buds which had germinated after a fortnight, the sum of the two repetitions for each treatment is given.

TABLE III

Germination of Setts 6", 4" or 2" long, After Dipping in Water (W), Yeast (Y) or Mealybug Extract (M), Undiluted (1), or Diluted 1/4 (2) or 1/16 (3)

6" W	19	6" Y 3	17	6" M 3	19
6" W	19	6" Y 2	18	6" M 2	18
6" W	19	6" Y 1	18	6" M 1	13
4" W	17	4" Y 3	15	4" M 3	16
4" W	17	4" Y 2	14	4" M 2	12
4" W	18	4" Y 1	14	4" M 1	9
2" W	16	2" Y 3	16	2" M 3	7
2" W	14	2" Y 2	9	2" M 2	3
2" W	14	2" Y 1	8	2" M 1	1

Analysis of these figures indicated significant effects for all three factors: treatment, length and dilution. There was a significant interaction between length and treatment, particularly in the mealybug series. The effect of dilution was less than that of treatment or length. Mealybug extract had, moreover, produced a significantly greater effect than yeast. It was estimated that, in the yeast culture used, there could not be fewer spores than in the mealybug extract. It was therefore concluded that, although the yeast might under certain conditions cause some reduction in germination, it could not be responsible for the whole effect observed after inoculation with the mealybug extract.

In attempting to explain the mechanism by which mealybugs affect the germination and early growth of cane, some four possibilities are apparent: the direct effect of removal of juice during feeding; the existence of some unknown disease of a virus nature; infection by organisms, such as fungi or bacteria, carried on or within the bodies of the insects; or the presence of some chemical toxin produced either by the insects themselves or by organisms associated with them.

Although the direct effect of feeding undoubtedly has some bearing on observed results, it is presumably not the only factor involved since inoculation with mealybug extract affects germination and early growth. Against the existence of some unknown virus it might be argued that, since practically every stick of cane is attacked by mealybugs at some period of its life, all cane in Natal should be infected with this hypothetical virus and it should not be possible to produce further symptoms by inoculation. As far as infection is concerned, the only organism found at the base of the buds in inoculated sticks was a yeast which, although it could be shown to occasion some reduction in germination, was not as potent in this respect as the mealybug extract itself. The possibility therefore still exists that some form of chemical toxin, produced by the mealybug or by some organism associated with it, is inoculated into the cane when the insect feeds and is, at least in part, responsible for the symptoms observed. The fact that the extract is inactivated by heat or by treatment with formaldehyde does not preclude the existence of such a toxin, since many chemical substances of this nature are precipitated by either heat or formaldehyde. The existence in mealybugs of toxins capable of affecting the growth of plants is not without precedent, since Carter,^{1,2} in a number of papers, has demonstrated that wilt of pineapples is caused by just such a material. As far as sugarcane mealybug is concerned, it is therefore proposed to investigate the matter further.

Summary

A description is given of some experiments which demonstrated that inoculation with the body fluid of mealybugs adversely affected germination and early growth of sugarcane. Investigation of this phenomenon showed the presence of a yeast, associated with the insects, which had some effect but was not as potent as the body fluid itself. The existence of some toxin associated with the insects is suggested as a possible cause of observed symptoms.

REFERENCES

- ¹ Carter, W. (1933): The Pineapple Mealybug, *Pseudococcus brevipes*, and Wilt of Pineapples. *Phytopath.* **23**, 207-242.
- ² Carter, W. (1937): The Toxic Dose of Mealybug Wilt of Pineapple. *Phytopath.* **27**, 971-981.
- ³ Dick, J. (1953): Mealybug and Its Effect on Sugarcane. *Proc. 27th. Congr. S.A.S.T.A.*, 113-118.

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South African Sugar Association,
Mount Edgecombe.
March, 1956.

The Chairman said that we had heard with much interest Dr. Dick's paper on the effects of the mealybug, and he hoped that chemists in the audience would be able to supply further data on this subject.

Dr. Dick stated that from his latest experiments it would appear that the effect on the cane sett might be due to an organism associated with the mealybug and not to the mealybug itself.

Dr. Dodds mentioned that Australians were surprised that so much time was spent in South Africa on mosaic disease which in Australia was regarded as being of no importance while in this country it is a serious threat. **Dr. Dick**, in reply to a question from Mr. W. J. Hempson, explained that the mealybug was a small soft-bodied insect found under the leaf sheaths of the cane.

Mr. Rault stated that two new varieties, i.e. N:Co.339, and N:Co.293 planted on the eighteen different sections constituting the Natal Estates had yielded the excellent returns of 8.8 and 9.9 tons of sucrose per acre respectively. The acreage cut represented a total of 48 and 79 acres.

As one of them was a carrier of mosaic and the other of "smut," the field management had decided as a precautionary measure, to restrict their propagation on account of the possible damage to the not so resistant already established varieties.

This anomalous position indicated that as in the case of Australia, carriers of mosaic under certain forms, could still be prolific yielders, very profitable to the industry for the time being.

Dr. Dick said that it had been hoped to grow only varieties which were immune to mosaic, but now all that could be hoped for was that we might be able to propagate resistant varieties. The danger was that if tolerant varieties were allowed to be planted they might act as a source of infection to less tolerant varieties.

The Chairman, Mr. du Toit, asked Mr. King to comment upon the remarks by Mr. Rault that N:Co.339 had been condemned although its yield was very high, simply because it contracted mosaic disease.

Mr. King said that there was a great danger of strain mutations taking place when there was a large number of mosaic infected plants about. These strains may be more or less virulent than the common strain now present in the industry. If a more virulent strain arose there was the possibility that N:Co. 339 would no longer be a tolerant variety.

Mr. Pearson commented on Mr. Buzzacott's remark that mosaic was not important whereas Mr. Henzel from Jamaica had stressed the importance of it in the West Indies.

Mr. Main enquired what the effect of mosaic on Co:339 would be over a certain period.

Mr. King said that experiments had shown, when carried up to the 3rd ratoon, that there was no particular loss in N:Co.339. In N:Co.310 it had been found that after about the 2nd or 3rd ratoon the variety recovered from the disease.

Dr. Brett asked Dr. Dick if he did not think that the effect of wilt in pineapple could not be entirely accounted for by a simple toxin.

Dr. Dick said that Mr. Carter was apparently satisfied that it was the effect of a toxin.

Dr. Dodds asked if there were not more than one species of mealybug in this country. Apparently the one in Louisiana that attacked cane was very different from the principal one here.

Dr. Dick replied that for a long time it was thought that there was only one species of mealybug but he himself had some time ago found another species. This however was more susceptible to the attacks of parasites, and would not develop to the same extent as the one mentioned in the paper.

The Chairman said that we had long known of the existence of mealybug in this country but no great importance was attached to it. Dr. Dick had now opened up a wide field of investigation, and had draw attention to the harmful effects of the mealybug. This field of investigation would embrace the work not only of the entomologist but also that of the botanist and chemist.