

Techniques of Seed and Seedling Production with Sugar Cane

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Historical

ALTHOUGH the history of sugar cane stretches far into the distant past, cane breeding is comparatively recent. The earliest mention of sugar cane is in legends concerning the Buddha about the fourth century B. C., and it is generally agreed that one of the centres of origin of cane is Northern India. From India it is believed to have been first carried to China probably via Assam. The Chinese then carried it to Formosa, Java and the Philipines. While in the earliest times the cultivation of cane was as a garden crop and its use restricted to the chewing of the stalks and drinking of the juice, the manufacture of sugar from cane was an established industry at the time of the advent of Europeans into this area. In the other direction, the Arabs introduced the cane into Southern Europe and North Africa. Columbus carried the cane thence to Santa Domingo on his second voyage in 1494. From Santa Domingo it spread to North and South America, Cuba and the West Indies.

There was then in the 19th Century a flourishing sugar industry spread generally along the equatorial belt. Naturally the number of varieties cultivated were few, and up to this time little need was felt for diversity since the existing varieties were apparently thriving and the manufactured sugar was fetching favourable prices in the market.

Danger signals began to appear about the middle of the 19th Century. About 1840, in Mauritius and Reunion, the predominant variety, Otaheite was affected by a disease which assumed epidemic proportions and threatened the entire industry there. The exact nature of the disease is unknown, but the epidemic was interpreted as a "degeneration" of the variety. Next, in 1860, disease in epidemic form threatened the industry in Brazil, and in 1872 disease appeared in Puerto Rico. In both these instances too, the variety affected was Otaheite. In every instance, the remedial measures adopted were the introduction of varieties from abroad and the testing of them for resistance to the prevalent disease.

Since the cane is normally propagated vegetatively, it was generally believed the plant had lost its capacity to produce viable seed. The degree of conviction with which this idea was held at the time is evident from the fact that a committee appointed by the Hawaiian Agricultural Society in 1851 to investigate the possibility of producing new varieties by hybridisation, reported in 1854 that seedling production was not possible. Further, a claim made by A. W. Parris of Barbados in 1858 to have raised some cane seedlings that appeared as volunteers in his plantation appears to have been ignored. Meantime, the inroads of disease and the threat of extinction of the industry continued.

About 1880, plantations in Java were affected by a disease later called Sereh, from the fact that affected plants became severely stunted and took the form of that grass. The immediate danger was averted by the adoption of expensive agronomic practices. However, as the danger of epidemic disease was very real and the potentialities of introducing new cane varieties were limited, the need for hybridisation was keenly felt and the study continued.

It is, therefore, not surprising that positive results were reported almost simultaneously a short time later from two quarters. In 1885, F. Soltwedel in Java after a detailed study of the cane flower reported that he saw no reason why the cane should not produce seed, and went on to produce seedlings in 1887.

In 1888, Harrison and Bowell also reported the production of seedlings from Barbados.

It should also be mentioned that although the fact that cane seedling production was possible was established *circa* 1888, the numerous problems of technique yet remained to be worked out and it was actually not until the turn of the century that systematic breeding and seedling testing got under way.

This marked the birth of a new era. Today, every major cane-growing country in the world spends considerable sums of money on breeding programmes.

In Ceylon, sugar cane has yet to attain the status of an industry. It is true that cane is cultivated in various parts of the Island. Three hundred acres are under cane in the Nagoda area on the banks of the Gin Ganga. There is a very small extent cultivated at Haldamulla and Ratnapura. There are also cane "gardens" (they cannot be called

anything else) in various places. These cultivations are mainly for the purpose of producing a crude jaggery, or for sale of the stalks for chewing at festivals.

That these cultivations are of negligible extent becomes apparent when it is realised that a modern sugar factory handles cane from 8,000-10,000 acres in one season.

Systematic investigations into the possibilities of cane cultivation were commenced at Polonnaruwa in 1951. As these preliminary investigations proved a success, in 1953, a study designed to explore the potentialities of cane breeding was commenced. Results of these studies are reviewed below.

In regard to problems of technique, the following questions need study :—

1. The principles of mating.
2. Flower dehiscence and pollen discharge.
3. The technique of controlled crossing and seed production.
4. The method of seed storage.
5. Technique of germination of the seed.
6. Seedling selection.

1. Principles of Mating

It is not proposed to deal with this point at length in this paper. Breeding was born out of the threat of extinction of the industry by disease. The canes in cultivation prior to this period (the so called Noble canes) while having a high sugar status were generally susceptible to disease, while the wild varieties have little sugar but a high degree of disease resistance. The practice, therefore, has been to cross Noble varieties with "Wild" relatives and back-cross these

progenies to the Noble varieties, the process being called Nobilisation. Two wild species *Saccharum spontaneum* (of which a number of strains exist) and *Saccharum robustum* have been used for this purpose.

In Ceylon, breeding has been limited due to (a) the need to perfect techniques initially, (b) the paucity of varieties (we are now importing a number of varieties) and (c) the small extent under cultivation.

It is of interest to note that a strain of *Saccharum spontaneum* has been found recently growing locally at Inginiyagalla and is now being propagated for study.

2. Flower Dehiscence and Pollen Discharge

The times of maximum flower dehiscence and the stage of pollen shedding are of fundamental importance. These points were studied over a season. All florets that had dehisced were plucked out with a fine forceps and counted in the first instance at four hour intervals. The results showed that maximum dehiscence occurred during the period 3.0 a.m. to 7.0 a.m. Following this, the number of florets which dehisced during the period 3.0 a.m.-7.0 a.m. were counted at one hour intervals. It was then found that maximum dehiscence took place during the period from 4.0-5.0 a.m. Pollen discharge was noted to follow closely on floret dehiscence.

3. Crossing Technique

Two problems arise. Firstly, the impracticability of emasculation of individual flowers and introduction of

desired pollen thereafter and as a consequence of this fact the need to centralise crossing as far as possible in one spot. Emasculation of the individual florets is not possible for four reasons—the minute proportions of the individual florets, the large number of florets on an inflorescence, the height at which the flowers occur (about 15-20 ft. above ground level) and the need for as large a number of seedlings as possible.

Numerous attempts have been made to induce pollen sterility as by dipping the inflorescence into warm water, &c., but the writer is not aware of any practical achievement in this field to date.

Therefore, recourse is made to the fortunate circumstance of the flower itself, that some varieties do not produce viable pollen (or produce very little) while others do so in variable profusion, female sterility being rare.

The method of determination of pollen viability is to tease out the anthers of mature flowers (those that are due to open the following day) into a dilute solution of iodine and make a count of the number of stained grains. Grains considered viable are smooth, round, firm and take a dark stain; non viable grains are generally shrunken and do not stain. The viability is expressed as a percentage of total pollen grains after counts on several microscopic fields have been made. These figures are inevitably approximate, and are generally grouped into the following classes:—0—2, 2—5, 5—15, 15—30, 30—50 and 50+. If the percentage of viable pollen is less than 5, that variety is suitable for use as a female and if more than 30 as a male. Those varieties producing more than 5 and less than 30 per cent. viable pollen are problematic and this factor tends to

set some limit on crosses. Actually they may be used as either male or female parents though with some what unsatisfactory results. This test has to be carried out regularly since variations (the reasons for which are still obscure) occur from season to season as well as during different periods in the same season. In all the seasons tested the varieties Co 453 and Co 617 produced abundant viable pollen while Co 527 and Co 655 proved females, Co 449 being intermediate. Co 421 showed wide fluctuations of pollen viability.

Other methods of determining viability (such as germination of the pollen grains on the styles of other flowers—*Hibiscus* sp.—and in nutrient solutions) have been tried but the iodine test is the simplest and easiest and is sufficiently reliable.

The technique of crossing thereafter consists simply of bringing a pollen fertile variety up to a pollen sterile one under conditions that will avoid contamination. Since florets on an inflorescence continue to open for about two weeks, it is necessary to bring pollen to the inflorescence during that entire period. Hence the desirability of centralisation.

There are two generally accepted methods of centralisation :

1. The Hawaiian method.
2. The Indian method.

The Hawaiian Method. The inflorescence together with much of the stalk is cut and stood in a dilute solution of sulphurous acid and phosphoric acid. When this solution is changed every other day and about one inch of the bottom of the stalk cut off at that time, the inflorescence remains fresh and

matures normally. It is hoped to test this method at Polonnaruwa during the coming season.

The Indian Method. The Indian method which has been successfully used at Polonnaruwa is based on the principle that the root primordia that occur at every node can be made to grow under certain conditions. Some weeks before flowering a potting mixture is wrapped round the upper part of the stalk so as to cover two or more nodes, and the mixture is kept moist. Roots develop in a very short time and before flowering the stalk is severed below these roots and the stalk transferred into a pot. The process does not interfere with emergence of the inflorescence or seed setting.

4. Seed Storage

At Polonnaruwa the cane flowers in late September-October and seed is ripe by late November-December. Planting does not commence until May of the following year. The seed, however, does not remain viable for more than a few weeks, so that a reliable means of storage becomes necessary. The standard method of seed storage has been used successfully. This consists of storing the seed in a dry atmosphere under refrigeration.

It takes 10-14 days for all the florets of an inflorescence to dehisce, and another 2-3 weeks for the seed to mature. Thereafter the entire arrows are cut, placed in cloth bags and sun dried for 2-4 days. At the end of this drying the seed (fuzz) comes freely off the stalks and can be easily collected free of stalk in the bag. For the tests at Polonnaruwa, the fuzz was now transferred

into empty bottles. A dry atmosphere being a prerequisite, about 10 gm. of calcium chloride tied in a calico square were placed in each bottle and about 20-25 grams of fuzz put in. The bottles were stoppered and the mouths sealed with paraffin wax. The bottles were then transferred to cold storage. At the end of 3 months, the bottles were opened and the seed sown in seed flats. A germination count of 500-600 seedlings was obtained per flat.

5. Seed Germination

The seed is sown on sterilised soil placed in shallow seed flats. No attempt is made to separate the seeds from the other flower parts and husks. When the flats are ready, the seed is spread evenly on the flat, settled with the hand and a fine spray of water and pressed firmly on to the soil surface. Thereafter the flats are placed in the open and moistened frequently to prevent the surface from drying out. Germination commences in about 48 hours. It is usual to apply a spray of a dilute fungicide at sowing, or in the next few days to guard against any fungal infection and subsequent death of seedlings.

6. Seedling Selection

When the seedlings are 3-4 in. tall they are transferred into nursery beds and from here, they are transplanted individually in the field.

It is not proposed to consider the details of seedling selection.

At the end of the first year the first selection of individual seedlings is done. Here all seedlings with undesirable agricultural habits are first discarded. The remaining seedlings are tested for sugar content with a hand refractometer and compared against standard varieties that are planted throughout the fields for this purpose. Those seedlings with high sugar are selected for further test.

The quantity of cane from a single plant is limited. The further process then is to multiply the selected seedlings so that sufficient planting material is available for replicated yield trials. In the meantime from the multiplication plots further study of the cane is made, now including germination, maturity testing, ratooning-capacity and disease resistance. Thus, further rejection is done until the final seedlings go into the fore-mentioned yield trials. From the germination of the seedlings, it therefore takes 7-8 years before a seedling is ready for release as a commercial variety.

In conclusion it must be stressed once again that the studies carried out at Polonnaruwa to date have been mainly in the nature of studying techniques more than anything else. Some seedlings have been produced incidentally and these have been transferred to the field for study, but the numbers so planted are comparatively small. It is hoped to embark on a more ambitious programme of seedling production in the coming year.