Botany. — *The respiration of the stem of ripening sugar cane.* By J. W. HES.

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Introduction.

The production of the photosynthetic activity of a sugar cane plant provides the greater part of the material for the building up of the plant body and is also a source of energy for both construction (growth) and maintenance. The excess of the carbondioxide assimilation can be stored as a reserve material, in the case of sugar cane sucrose, in such an abundance, that it can be extracted from the stalks on an economic basis and thus cane is grown for this reason.

The difference between the total amounts of carbondioxide assimilation products and the part thereof incorporated in the plant tissues (dry matter) or serving as the source of energy consumed in respiration, is roughly speaking the amount of sugar in the cane at harvest time.

We are fairly well informed about the dry matter after years and years of cropping cane. Concerning assimilation there are some data available from preliminary investigations, for instance those carried out in Java by KAMERLING (4), KUYPER (5, 6) and COELINGH and KONINGSBERGER (2). But as far as we know there is no quantative information about respiration processes, at least not in the Java-literature. Hence we started a series of experiments to obtain some preliminary information on this matter.

Material and Methods.

In working out these experiments cane was chosen of one variety, twelve months old and grown under conditions representing approximately the average Java conditions in the cane fields. The material involved in the experiments was selected from stems of about the same size, length and habitus. After cutting the internodes were cleaned with soap and water, rinsed and dried. To prevent desiccation both ends were dipped in melted paraffine.

The respiration was determined by measuring the carbondioxide production. A current of humid air deprived of carbondioxide was passed through a respiration vessel, having a volume only slightly exceeding that of the cuttings, and then in succession two Pettenkofer-tubes containing barytawater. The carbondioxide liberated in two hours by the internodes was absorbed and determined in the usual manner by titration. All experiments have been carried out in a dark room at 28° C.

In order to find the amount of dry matter (total solids), sucrose and reducing sugars, the internodes used for measuring the carbondioxide pro-

duction were weighed (fresh weight) and then chopped to a finely divided condition and subsequently crushed in a small hydraulic press to disintegrate the tissues. The pulp along with the juice already expressed by this treatment was made up to a known weight by the addition of water. After adjusting the pH to 7.5 and adding one gram of calciumcarbonate the mixture was digested for one hour at boiling temperature, this proved to be sufficient for a complete extraction of all sugars. After cooling the acidity was checked and found to be neutral in all determinations.

In the fiber determination the pulp was separated from the extract and washed in running water until the washings were free from dissolved sugar as disclosed by the α-naphtol reaction. The sugar-free fiber was dried to constant weight in a drying oven at 105 °C (fiber-weight) and then immersed in a 10 % sucrose solution for 12 hours in a refrigerator. Water was imbibed by the fiber, thus leaving the sucrose solution more concentrated. The increase in polarization is a means for computing the amount of "colloid" or "imbibition" water attached to the fiber. We assumed this amount equal to that originally present in the cane tissues (in Dutch: brixvrij rietwater, translated: brix-less cane-water).

The separated extract was analysed by methods usually employed in the Java sugar industry (3). The amount of dissolved substance (gravity solids) was calculated from the pycnometric specific gravity, whereas after clarification reducing sugars were determined by the Schoorl's iodide method with Luff's solution (3). Sucrose determination was carried out ter clarification by double polarization using STEUERWALD's method of inversion at room temperature (7). The results obtained in this way were checked by a chemical sucrose determination calculated from the difference between the invert sugar contents before and after inverting, cf. BROWNE and ZERBAN (1). Both methods gave equal quantities of sucrose.

In order to refer the percentages of substances found in the extract to a fresh weight of cane basis, the following formula was used:

$$\frac{a\left\{b-\left(c+\frac{c\times d}{100}\right)\right\}}{e} = \%\ \text{originally present in the cane, where:}$$

a = percentage of substance (sucrose, reducing sugars or gravity solids) in the extract

b = total of fresh cane weight and added water

c = fiber weight

d = "colloid" or "imbibition" water in percentage of the fiber

e = fresh weight

The total dry weight (total solids) was calculated by adding the fiber weight to the amount of gravity solids reduced to cane basis by the formula given above.

Nitrogen determinations were carried out by the usual Kjeldahlmethod.

Experiments.

After cutting the carbondioxide production turned out to be markedly higher than normal being a reaction to injury. To avoid this effect the cuttings were always prepared 24 hours in advance for all experiments. The carbondioxide production was determined the next day and the day after, generally for two periods of two hours daily. The two daily values agreed very well, the differences never exceeding 10 %. After these two days the cuttings were analysed, thus determining the fresh weight, dry weight and percentage of sucrose and reducing sugars of the stem part involved in an experiment. These data are plotted in the graphs of figure 1 and 2. The horizontal axis, laid off with a centimeter scale, represents the heights of the internodes above the surface of the soil; the schematic drawing of a cane stalk underneath shows the decreasing length of the internodes from foot to top. On the vertical axis the percentage of the constituent parts related to the fresh weight is indicated, with the exception of the nitrogen, which is expressed as a percentage of the dry matter. The total dry weight is indicated by dots, sucrose by circles, reducing sugars by triangles and nitrogen by crosses. Smoothed curves have been drawn through the series of points determined. The number of points indicating the chemical composition is much greater than those in figure 1 denoting the respiration, as in most experiments more than one cutting was used and each was analysed separately. The nitrogen was estimated in internodes chosen along the length of one stalk, the dry matter of these internodes is also introduced in figure 2.

The bottom scale in figure 1 has the same meaning as that in figure 2. On the lefthand scale the production of carbondioxide in the two-hour periods of the first day is indicated in grams per 24 hours per kilogram of dry weight.

Discussion.

The distribution of sugars along the cane stalk as indicated in fig. 1 is perfectly similar to the figures in WENT's publication on this subject (8). WENT also examined the part of the stalk below the soil surface, which has lower sucrose contents, this part was however not involved in our experiments. The remarkable fact that the reducing sugars reach a maximum percentage some internodes below the top is also clearly demonstrated in our graph.

To make sure that neither appreciable gas production nor consumption occurred even during a whole day, we examined internodes of various ages in a one liter flask, closed with a manometer and placed in a bath of constant temperature. Therefore we can assume that sugars, anyhow carbohydrates, are the substrate of the respiration of the cane stalk tissues.

The respiration in the lower part of the stalk up to about one meter, in which the amount of dry matter, sucrose, reducing sugars and nitrogen

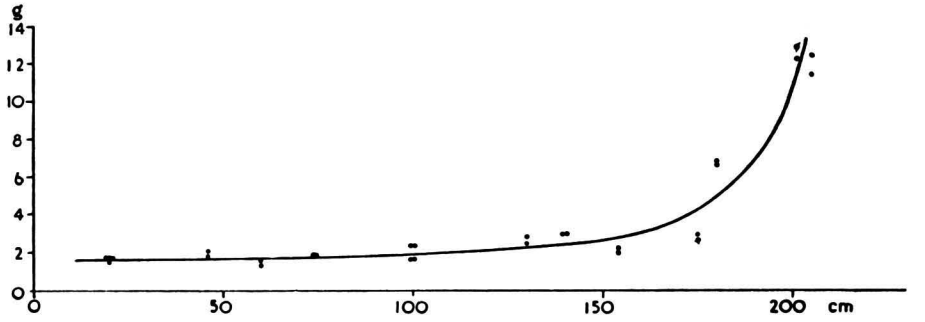

Fig. 1. The march of the respiration level along the stem of ripening sugar cane. The ordinate shows the respiration level expressed as grams of carbondioxide daily produced by 1 kilogram of dry matter, the abscisse the distance from the soil surface.

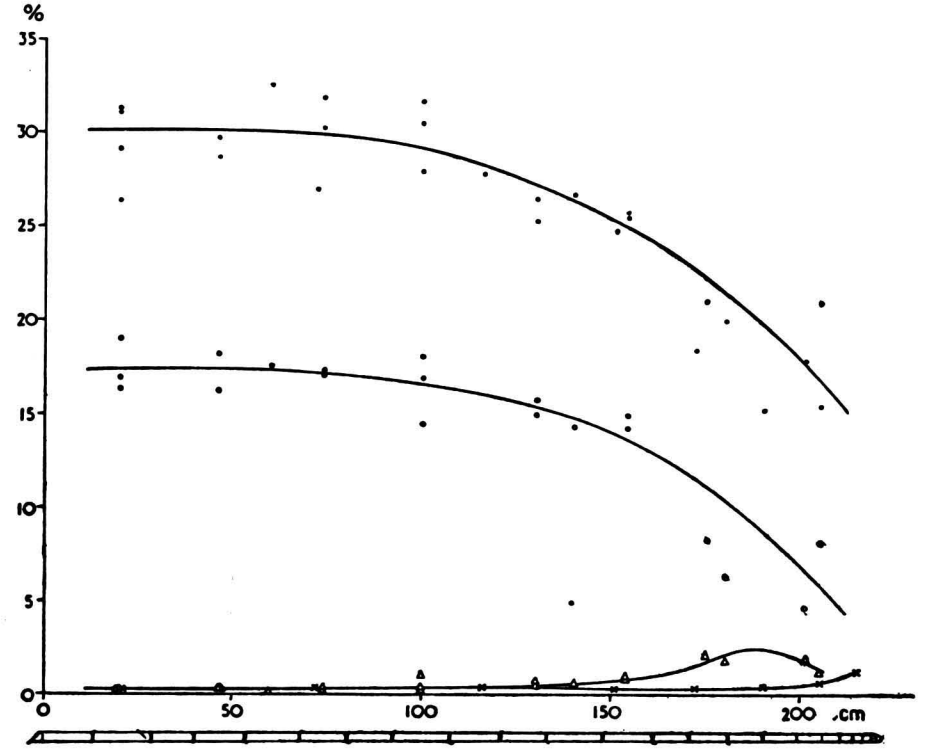

Fig. 2. The chemical composition of a ripening cane stem indicated on the ordinate in % of the fresh weight, with exception of the nitrogen, calculated as % of the dry weight. Sucrose indicated by circles, reducing sugars by triangles, dry matter by dots and nitrogen by crosses. Abscisse as in figure 1.

remains on the same level, has a value of about 2 grams of carbondioxide per 24 hours per kilogram dry matter, corresponding to the dissimilation of about 1.36 grams of hexoses. This part of the stem can be considered as mature and its sucrose contents as at the maximum percentage for the cane under examination. In the next part of the stem the accumulation of sucrose is continuing and the respiration value increases slowly in apical direction with the decreasing sucrose concentration. Still nearer to the top where the growth is gradually becoming more important, the respiration increases rather rapidly to a value of 13 grams of carbondioxide or expressed in terms of the dissimilation of hexoses about 9 grams per kg dry weight per day.

In the younger internodes the percentages of reducing sugars and nitrogen are higher than in the older ones. There is however no direct relation between these quantities and the respiration level. Regarding as dry weight the value obtained by substracting the total sugars from the total dry weight, there is no important fluctuation in this value along the stalk. Hence there is no obvious relation between either this dry weight value or the respiration level. Among the data considered in this investigation the percentage of sucrose is perhaps the best expression for the physiological activity of the internodes along the stem.

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Summary.

The respiration intensity of ripening sugar cane was examined and measured as carbondioxide production. The lower part of the stalk, which has already attained its maximum sucrose contents up to where sucrose accumulation is still continuing, shows a respiration level of approximately 2 grams of carbondioxide per kilogram daily. From this point upwards the production increases gradually, but near the top there is a sudden increase to approximately 13 grams of carbondioxide. Obviously there is no clear and simple relation between the carbondioxide elimination and the chemical composition as far as sucrose, reducing sugars, nitrogen or dry substance are concerned.

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