# THE PRODUCTION OF AMYLOSE AND AMYLOPECTIN IN CORN ENDOSPERMS AND IN POTATO TUBERS[1]

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## ABSTRACT

Experimental evidence indicates that amylose and amylopectin are produced simultaneously. Consequently, any proposed mechanism for the synthesis of starch based on the assumption that the branching enzyme is inactivated at some time during the day, in order to enable the synthesis of amylose, would appear to be invalid. Amylose (3.1%) was produced in very immature waxy corn endosperm by covering the ears with cellophane bags. One can postulate that the production of starch by plants occurs via glycogen. That is, plant glycogen is attacked by a theoretical debranching enzyme which (a) removes the outer or available branches of the glycogen to form amylopectin and then (b) connects these removed branches end-to-end to form amylose. In normal waxy endosperm the absence of amylose can be explained by assuming the presence of an inhibitor of the proposed debranching enzyme. The production of amylose in very immature waxy corn endosperm indicates that the activity of this inhibitor may be diminished by retarding the growth of certain factors in the very immature waxy corn endosperm. The average chain length of amylose appears to increase with an increase in the average chain length of the corresponding amylopectin. These results can be explained by assuming that the degree of polymerization of the unit chains removed by the proposed debranching enzyme remains constant. Consequently, the degree of polymerization of an amylose appears to be a function of the chain length of its parent glycogen.

In most plants, starch-synthesizing cells produce both the linear and the branched components amylose and amylopectin. At present only two types of enzymes have been found to be directly connected with the synthesis of either starch or glycogen from glucose-1-phosphate: phosphorylase and the branching enzymes (2,10,16). The mechanisms of these enzymes are well known. Recently the role of phosphorylase in the synthesis of animal glycogen has been questioned (4). According to Niemeyer (see ref. 4), phosphorylase may only be used *in vivo* to degrade glycogen to glucose-1-phosphate instead of to aid in its synthesis. Glycogen is synthesized in *Neisseria perflava* by means of amylosucrase (11). Also, sucrose may be directly converted into starch, as suggested by Ewart *et al.* (8) and supported by Badenhuizen (1). It would appear, therefore, that the entire mechanism for the synthesis of starch is open to criticism. The common notion (2,10,12,16,18) that amylose is the precursor of amylopectin may be entirely false.

The most difficult problem in proposing a mechanism for the

[1] Manuscript received October 14, 1957. Journal Paper No. J-3227 of the Iowa Agricultural Experiment Station, Ames, Iowa, Project No. 1116. This work was taken in part from a thesis submitted by S. R. Erlander to Iowa State College in partial fulfillment of requirements for the Ph.D degree, December, 1956. This research was supported in part by a grant from Corn Industries Research Foundation.

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synthesis of starch is how to account for the presence of the linear component amylose in starch and its absence in animal glycogen. Whelan and Walker (20) have recently proposed a compartment method for the separate synthesis of amylose and amylopectin. The author (5) has proposed that plant glycogen is synthesized first and this glycogen is then converted into amylose and amylopectin by an unknown debranching enzyme.

To obtain information on the mechanism employed by plants to synthesize starch, experimental investigations were carried out along three lines: 1) the effect of light and dark on the total and relative amounts of amylose synthesized in the corn plant; 2) the effect of light on the synthesis of amylose in the potato starch; and 3) the effect of water evaporation from the kernel on the synthesis of amylose in the corn plant. Data are also included on the relationship between the degree of branching of the amylopectin and the molecular weight of the corresponding amylose in corn starches. The results of these investigations are discussed in the light of the author's (5) proposed mechanism for synthesis of starch from glycogen.

## Experimental Work

*Corn Varieties and Method of Sampling.* The varieties of corn plants used, including their endosperm genealogy, were Seneca Chief sweet corn $su_1$, $su_1$, $su_1$, *Wx*, *Wx*, *Wx*; Iowa 4297 dent corn $Su_1$, $Su_1$, $Su_1$, *Wx*, *Wx*, *Wx*; and Iowax 5 hybrid waxy corn $Su_1$, $Su_1$, $Su_1$, *wx*, *wx*, *wx*. The sweet, dent, and waxy varieties were hand-pollinated, respectively, on July 20, 25, and 24 in 1955. All samples of a given variety were pollinated at the same time. Three samples were collected from each variety at 12-hour intervals on the 13th and 20th day after pollination: 13th morning, 13th evening, 14th morning, 20th morning, 20th evening, and 21st morning. A sample of each variety was also collected at maturity. The immature kernels were collected between 5:45 and 6:30 in the morning and between 5:45 and 6:30 in the evening. Each ear was first husked while on the plant, then picked, and then immediately shelled with a knife and frozen on solid carbon dioxide in order to keep enzyme degradation to a minimum. All immature samples were kept frozen on dry ice until they were processed for starch (no longer than 4 days after picking).

In order to study the effect of water evaporation from the kernel during plant growth, some samples that were picked on the 13th morning and 20th morning had been covered with cellophane bags and husked about the third or fourth day after pollination. Two cellophane bags, tied on by an elastic band, covered the ear. The outer bag

contained a small amount of water to retard evaporation. These samples were collected as above.

*Isolation of Corn Starch Samples.* The starch was isolated from the kernels in a room held at approximately 4°C. The frozen kernels were ground with a small amount of iced distilled water for 3 minutes in a Waring Blendor (16,21). The slurry was screened through No. 17 nylon bolting cloth. The magma was then squeezed as dry as possible. The press cake was ground for another 3 minutes with fresh water, then screened as before. The combined extracts were centrifuged in an International centrifuge. Microscopic examination of the separated granules indicated no detectable granule damage. The gluten in the starch was removed according to the method of Maywald, Christiansen, and Schoch (17). The above starch-gluten mixture was shaken with 20% Pentasol mixture for three or four 15-minute periods. Each period was followed by centrifuging, discarding the supernatant, and adding fresh 20% Pentasol solution. The final starch was washed with 95% ethanol and Soxhlet-extracted for 48 hours with 95% ethanol to remove the associated fatty acids. The starches were then air-dried and

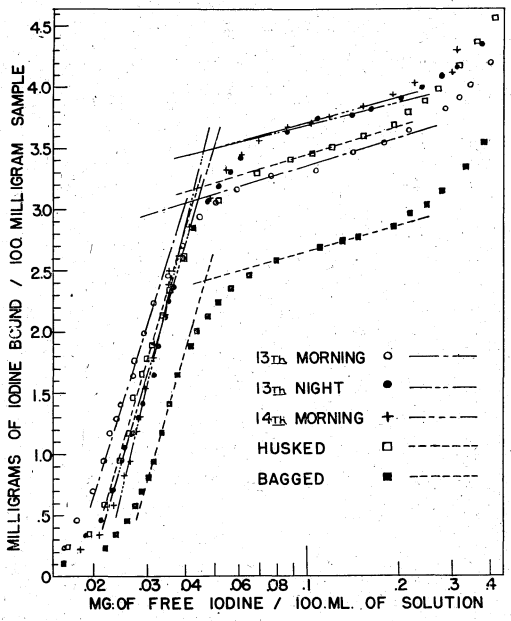


Fig. 1. Immature sweet corn starch samples. Iodine titration curves for the 13th morning, 13th night, 14th morning, husked ears, bagged ears (see legend and see text for the explanation of these starch samples). The intersection of the two lines for each titration curve gives mg. of iodine bound per 100 mg. of sample (iodine-binding capacity). The iodine titration curves for the other corn starch samples were similar to the above curves.

kept in closed containers. The mature samples were isolated in the same way without sulfur dioxide treatment, since it is believed that hydrolysis of the starch occurs with sulfur dioxide treatment.

*Growth and Isolation of Potato Starch Samples.* Certified Irish Cobbler potatoes were planted June 1, 1955, in three plots which had been equally fertilized and were located 3 to 5 feet apart. Four plants were planted in each plot. In the first plot the plants were exposed to natural sunlight throughout the day and to a fluorescent lamp at night. The adjustable light was held a few inches above the plants and was turned off for 2 hours each night. The second plot was exposed to the regular diurnal variations in light. The third plot was shaded so that the plants were exposed to sunlight for approximately 2 hours every afternoon.

The mature potato tubers were ground in a meat grinder. Water was added and the starch slurry was passed through No. 17 nylon bolting cloth. The same procedure as described above for separation of protein and fat was then followed.

*Iodine Titration of Starch Samples.* The percent amylose in all of the samples was determined according to the method of Lansky, Kooi, and Schoch (15), which is a modification of the method of Bates, French, and Rundle (3). The data, however, were graphed in a different manner as suggested by Dexter French[3] in order to increase the

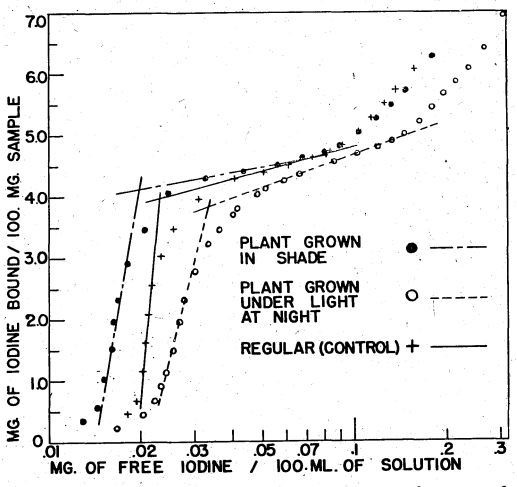


Fig. 2. Potato starch samples. Iodine titration curves for samples of mature potato starch produced under various conditions. The potato tuber starch samples were obtained from potato plants grown in the shade, grown under a fluorescent light at night and sunlight during the day, and grown under normal conditions as a control.

[3] D. French; private communication.

precision. A plot was made of mg. of iodine bound per 100 mg. sample versus the logarithm of mg. of free iodine per 100 ml. of solution. The intercept of the two lines gives the mg. of iodine bound per 100 mg. of sample. Typical plots for the corn and potato starch samples are given in Figs. 1 and 2. All of the samples were corrected for moisture content by drying representative samples in a vacuum oven at approximately 60°C. until a constant weight had been obtained (4 or 5 days).

*Ferricyanide Number Determination.* The sweet corn amylose was separated from the amylopectin by the method of Lansky, Kooi, and Schoch (15) after refluxing under a helium atmosphere in a buffered *n*-amyl alcohol solution for 24 hours (7). The adhering phosphate was removed by dialysis. The ferricyanide number was determined on the amylose solution according to the method described by Kerr (13). To determine the concentration, a weighed portion of the amylose solution was predried under a heat lamp and finally in a vacuum oven at 95°C. until constant weight was obtained.

## Calculations

Let us assume that amylose is produced only during the daytime and amylopectin only during the night. Then from the percentages of amylose and the 24-hour yields given in Table I, we can calculate the percentage of amylose for the 13th night according to the equation:

$$A_{13N} = \frac{(Y_{14M} - Y_{13M})A_{14M} + Y_{13M}A_{13M}}{Y_{13M} + (Y_{14M} - Y_{13M})A_{14M}} \quad (1),$$

where

$A_{13M}$ = predicted fraction of amylose for the 13th night, assuming that amylose is produced from 6 a.m. to 6 p.m and amylopectin from 6 p.m to 6 a.m.;

$A_{13M}$ = fraction of amylose in the 13th morning sample;

$A_{14M}$ = fraction of amylose in the 14th morning sample;

$Y_{13M}$ = yield of starch in g. per ear for the 13th morning sample; and

$Y_{14M}$ = yield of starch in g. per ear for the 14th morning sample.

Yields were expressed as g. per ear since this gives roughly the amount of starch per plant (or per kernel). To reduce the error in the yields, the 13th night yield was not used in the calculation. It was assumed in the above calculation that the increase in the percentage of amylose from the 13th morning sample to the 14th morning sample is negligible. From Table I we see that this assumption is valid.

Similar calculations were made for the 20th night samples by substituting the numbers 20 and 21 for 13 and 14, respectively, into equa-

tion 1, since these numbers designate the number of days after pollination.

Because of the possibility of a large error in the daily yields, calculations were also made from weekly yields. Various rates of starch production were assumed, since the exact rate of increase in starch per day for an average ear or kernel was not known. The rate constants were obtained by averaging in each case the three rate constants obtained from the 7-day increases: 13th morning to 20th morning, 13th night to 20th night, and 14th morning to 21st morning. The daily increases in starch yield ($Y_{14M} - Y_{13M}$ and $Y_{21M} - Y_{20M}$) were obtained from the calculated rate constants using the experimental values for $Y_{13M}$ and $Y_{20M}$. In all of these calculations the experimental values for $A_{13M}$ and $A_{20M}$ were used, but the values for $A_{14M}$ and $A_{21M}$ were obtained from the observed weekly increase in percent amylose, assuming zero-order kinetics. Only a small error is involved in assuming a zero-

TABLE I

TWELVE-HOUR-PERIOD YIELDS FOR DENT, SWEET, AND WAXY CORN STARCH, AND THEIR IODINE-BINDING CAPACITIES

| SAMPLE [a] | No. OF EARS | TOTAL STARCH YIELD [b] | PERCENT STARCH PRODUCED DURING DAYTIME [c] | IODINE BOUND PER 100 MG. OF SAMPLE | PERCENTAGE OF AMYLOSE [d] |
|---|---|---|---|---|---|
| | | *g/ear* | | *mg* | |
| Dent corn | | | | | |
| 13th morning | 10 | 6.67 | ... | 3.40 | 17.90 |
| 13th night | 10 | 6.27 | −19.6 | 3.40 | 17.90 |
| 14th morning | 10 | 8.31 | ... | 3.55 | 18.70 |
| 20th morning | 3 | 21.0 | ... | 4.25 | 22.35 |
| 20th night | 3 | 21.9 | 17.6 | 4.40 | 23.15 |
| 21st morning | 3 | 26.1 | ... | 4.48 | 23.60 |
| Sweet corn | | | | | |
| 13th morning | 11 | 1.16 | ... | 3.05 | 16.05 |
| 13th night | 11 | 1.33 | 29.8 | 3.50 | 18.40 |
| 14th morning | 11 | 1.73 | ... | 3.48 | 18.30 |
| 20th morning | 3 | 5.76 | ... | 4.38 | 23.05 |
| 20th night | 3 | 5.98 | 5.59 | 4.28 | 22.50 |
| 21st morning | 4 | 9.70 | ... | 4.35 | 22.90 |
| Waxy corn | | | | | |
| 13th morning | 8 | 5.27 | ... | ... | .... |
| 13th night | 8 | 8.6 | 34.2 | ... | .... |
| 14th morning | 8 | 15.0 | ... | ... | .... |
| 20th morning | 2 | 26.7 | ... | ... | .... |
| 20th night | 2 | 30.8 | 36.3 | ... | .... |
| 21st morning | 2 | 38.0 | ... | ... | .... |

[a] Numbers designate the number of days after pollination.

[b] After defatting and correcting for moisture content. The daily or 24-hour increases in starch yield were calculated by using only the morning samples (see text).

[c] These values were obtained directly from all of the experimental morning and night samples, for example, $100(1.33 - 1.16)/1.73 = 29.8\%$ for the 13th-night sweet corn starch yield. The values were not used to calculate the amount of amylose produced during the daytime because of the possibility of a large error, i.e., the difference between the 13th or 20th morning starch sample yields and their corresponding night samples is small (see text).

[d] Assuming that pure amylose binds 19 mg. of iodine per 100 mg. of sample.

order increase in amylose because of the small weekly increase in percent amylose during this period.

The percentage of amylose in all samples was calculated, assuming that pure amylose has an iodine-binding capacity of 19 mg. of iodine per 100 mg. of sample. This assumes that any change in the molecular weight of the amylose does not change the iodine-binding capacity (13). It was also assumed that any protein impurities present in the starch bound iodine to the same extent in all of the starches.

## Results and Discussion

*Production of Starch in Corn Endosperms.* A comparison of the amount of experimentally determined amylose with the amount predicted, assuming that amylose is produced during the daytime and amylopectin during the night, is made in Table II. The theoretical values were calculated as discussed above. The precision of the iodine affinity determination according to Lansky *et al.* (15) should be $\pm 0.08\%$. The estimated error in the theoretical values in Table II is approximately 10% in all cases. Therefore the differences in the predicted and experimental values are real.

TABLE II

COMPARISON OF EXPERIMENTAL AND PREDICTED PERCENT AMYLOSE, ASSUMING DAYTIME PRODUCTION OF AMYLOSE

| CORN STARCH SAMPLES | EXPERIMENTAL VALUES | THEORETICAL VALUES | | | |
|---|---|---|---|---|---|
| | | From Daily Yields | From Weekly Yields | | |
| | | | Zero-Order Production | 1st-Order Production | 2nd-Order Production |
| 13th-night dent | 17.90 | 22.1 | 23.3 | 21.2 | 20.1 |
| 13th-night sweet | 18.40 | 24.5 | 26.5 | 20.1 | 18.1 |
| 20th-night dent | 23.15 | 27.4 | 24.8 | 26.1 | 30.0 |
| 20th-night sweet | 22.50 | 33.4 | 26.1 | 28.0 | 37.4 |

The above calculations can also be made, assuming that amylopectin is produced during the daytime and amylose during the night. When this is done, the theoretical value for the percent amylose for the 13th or 20th night sample is lowered by approximately the same amount as it is raised under the previous assumption. That is, the 13th-night sample of sweet corn starch would have a theoretical value from daily yields of approximately 12.3% amylose as compared to the experimental value of 18.4%, assuming that only amylopectin is produced during the daytime.

The results of these calculations indicate that amylose and amylopectin are produced at the same time. Thus amylose cannot be produced during the daytime or night by inactivating the branching

enzyme. In other words, any assumption that the branching enzyme is inactivated during the daytime or night because of changes in environment would appear to be invalid. Therefore one must either rely on Whelan and Walker's compartment theory (20) or on theories such as the author's (5) which introduce different enzymatic mechanisms.

The data of Table I show clearly that most of the starch (both amylose and amylopectin) is synthesized during the night. This agrees with the results of Puhr and Hume (19), who found that the maximum production of starch in leaves occurs between 7 p.m. and 1 a.m.

The average chain lengths (6) of the 13th-day waxy, 13th-day dent, and 13th-day sweet corn amylopectins are, respectively, 17.2, 14.7, and 12.5 glucose units. Although these are average chain lengths, it is evident that those amylopectins or glycogens having a large average chain length will possess longer and less sterically hindered linear chains. Consequently, the waxy amylopectin should be able to crystallize more readily to form starch granules. This may account for the apparently large amount of starch produced in the waxy corn endosperms during the daytime (see Table I). The amylopectin (or glycogen) in sweet and dent corn endosperms would begin to crystallize in larger amounts during the evening after partial debranching by the proposed debranching enzyme has occurred.

*Production of Starch in Potato Tubers.* The potato tubers obtained from plants grown in the shade were fewer in number but comparable in size to the normal potatoes. The potato tubers obtained from plants grown under constant light (except for 2 hours) were quite small in size, indicating that large diurnal variations in light (rest periods) are necessary for the production of the tubers.

The samples from plants which were exposed to light at night had 20.25% amylose; those in the shade had 21.65% amylose; and the control samples had 20.95% amylose (see Fig. 2). Thus the variation in both cases was 0.70% amylose from the control. The amounts of potato starch (0.179 g.) used in the iodine titrations were within 0.4% of each other. The moisture in all of these samples was determined under the same conditions and at the same time as described under "Experimental Work." Therefore any large error in the observed percent amylose will be an absolute error and not a relative error. The observed differences in the percentage of amylose in the potato starch samples may therefore be real. At present they cannot be fully explained, but they may be due to a slight change in the activity of the proposed debranching enzyme caused by a change in the growing conditions. If this is true, then the proposed debranching enzyme may be most active (less inhibitor present?) when the plant is exposed to

less light.

The small differences in percentages of potato amylose would appear, however, to rule out the possibility that amylose is synthesized during the daytime and amylopectin during the night (or vice-versa) in potato tubers. The results on the production of amylose in potato tubers are therefore in agreement with the above conclusions concerning the production of corn starch.

*Production of Starch in the Bagged and Husked Corn Ears.* The effect of water evaporation on the production of starch in the corn endosperm was studied by bagging and husking the ears after pollination (see "Experimental Work"). The results are listed in Table III. The husked samples showed no marked variation in amylose content from the control. They were, however, exposed to the sunlight and their pericarps became extremely hard. This may have prevented a rapid loss of moisture. The percentages of amylose in the bagged samples for dent and sweet corn are much lower than those in the control samples. Wolf *et al.* (21) have shown by both alcohol fractionation and iodine titrations that the percentage of amylose increases with maturity. The iodine titration results listed in Table I and those of Maywald *et al.* (17) are in agreement with the results of Wolf *et al.* (21). Therefore the percentage of amylose in the starch appears to be an

TABLE III

COMPARISON OF STARCH YIELD AND PERCENT AMYLOSE OF CONTROL WITH THAT OF BAGGED AND HUSKED SAMPLES

| SAMPLE[a] | TOTAL STARCH YIELD[b] | PERCENTAGE OF AMYLOSE[c] |
|---|---|---|
| | CONTROL | CONTROL |
| Dent corn starch | | |
| 13th, Husked | 1.50/ 6.67 | 17.7 /17.9 |
| 13th, Bagged | 0.13/ 6.67 | 12.5 /17.9 |
| 20th, Husked | 19.0 /21.0 | 24.1 /22.4 |
| 20th, Bagged | 3.9 /21.0 | 19.6 /22.4 |
| Sweet corn starch | | |
| 13th, Husked | 0.56/ 1.16 | 16.7 /16.1 |
| 13th, Bagged | 0.18/ 1.16 | 12.7 /16.1 |
| 20th, Husked | 5.2 / 5.76 | 21.9 /23.1 |
| 20th, Bagged | 0.11/ 5.76 | 15.2 /23.1 |
| Waxy corn starch | | |
| 13th, Husked | 2.0 / 5.27 | 1.1 / 0.92 |
| 13th, Bagged | 0.58/ 5.27 | 3.1 / 0.92 |
| 20th, Husked | 17.0 /26.7 | 0.87/ 0.68 |
| 20th, Bagged | 3.4/26.7 | 0.84/ 0.68 |

a Numbers designate the number of days after pollination. See "Experimental Work" for definition of the terms "husked" and "bagged."

b Total starch yield in g. per ear of defatted and dried corn starch samples divided by that of the respective control starch samples.

c Percentage of amylose in sample and control obtained by assuming that pure amylose binds 19 mg. of iodine per 100 mg. of sample.

indication of the maturity of the particular corn endosperm.[4] Consequently, the low amylose content of the bagged samples indicates that the growth of their endosperms has been retarded. Also, the immature appearance of the bagged kernels (small and white) and their very small starch yield (see Table III), gives further evidence that bagging the ears retards the growth of the corn endosperm. This may have been caused by either 1) a decrease in the amount of water evaporating from the kernel, or 2) a change in atmospheric conditions surrounding the corn ear. If the former is true, then one might conclude that water evaporation is necessary for rapid crystallization of starch in the corn endosperm and hence for the maturation of the endosperm. From this experiment one can conclude that the absence of amylose in Badenhuizen's (1) incubated young leaves of *Scilla ovatifolia* Bak. was due to a retarding of some of the growth factors in the starch-synthesizing cells.

From the results on samples of sweet and dent corn starch one would expect that bagging the ears of waxy corn plants would also retard the growth of the endosperm. Assuming that this is true, then the results in Table III for the bagged sample of the 13th-day waxy corn starch indicates that amylose is produced by retarding the growth of waxy corn endosperm. This sample stains blue with iodine whereas the control stains brown, indicating that the iodine affinity is not due to some unknown anomaly. There is a possibility that the amylose is due to impurities from the ovary or pericarp. However, such impurities should have manifested themselves in the husked and control waxy corn starch samples. The production of amylose in the more immature cells of waxy corn endosperm was also noted by Lampe (14). The production of amylose in incubated waxy maize endosperm cells as found by Badenhuizen (1) and Fuwa (9) may also be due to retarding the growth of certain factors in the starch-synthesizing cells. These factors may be correlated with the synthesis of an inhibitor to the proposed (5) debranching enzyme. It is postulated that the production of amylose in very immature waxy starch-synthesizing cells is due to less activity or to a destruction of this inhibitor.

*Chain Length of Amylose as a Function of the Chain Length of the Corresponding Amylopectin.* The ferricyanide numbers of the 20th-day and mature sweet corn starch samples and their approximate number-average degrees of polymerization ($\overline{X}_n$) estimated from other studies (13) are compared with the corresponding chain length (6) of the amylopectin in Table IV. The ferricyanide number from Kerr (13) for

[4] This experimental observation indicates that a starch having 100% amylose content can never be obtained, no matter what the genetic background of the endosperm may be.

mature dent corn amylose and the corresponding $\overline{X}_n$ is also listed in Table IV. The smaller degrees of polymerization of the sweet corn amyloses as compared to the dent corn amylose may be due in part to hydrolysis or oxidation during the isolation. The degradation may however, be slight, since buffered solutions were used (7). Nevertheless,

TABLE IV

NUMBER-AVERAGE DEGREES OF POLYMERIZATION OF THE AMYLOSES COMPARED TO THE AVERAGE CHAIN LENGTH OF THE CORRESPONDING AMYLOPECTIN

| CORN STARCH SAMPLE | AMYLOSE FERRICYANIDE NUMBER | $\overline{X}_N$ [a] | CHAIN LENGTH [b] OF AMYLOPECTIN |
|---|---|---|---|
| 20th Day sweet | 4.17 | 140 | 12.2 |
| Mature sweet | 2.79 | 270 | 16.4 |
| Mature dent | 1.43 [c] | 455 [c] | 25.0 |

[a] Approximate.
[b] See ref. 6.
[c] See ref. 13.

since both sweet corn amyloses were isolated under the same conditions, the degree of degradation should be approximately the same in both cases. The results, therefore, indicate that the molecular weight of sweet corn amylose increases with an increase in the chain length of the amylopectin.

According to the author's proposed mechanism (5), the synthesis of amylose can be thought of as connecting end-to-end the branches which are removed from the parent glycogen. The average number of branches connected consecutively by the debranching enzyme can be called the "degree of polymerization" of these removed branches. This average "degree of polymerization" would most likely depend on the enzymatic properties of the particular debranching enzyme and the effective number (activity) of receptor groups available to the debranching enzyme-chain complex. If we assume for the sweet and dent corn endosperms that the proposed debranching enzymes have the same properties in all the corn endosperms and that the average "degree of polymerization" is constant, then the molecular weight of the amylose will be a function only of the average length of the removed chain. This would account for the above observed increase in molecular weight of the amyloses with an increase in the average chain length of the corn amylopectins. However, other explanations for this phenomenon could also be used without employing the author's (5) proposed mechanism. For example, assuming that the amylose is the precursor of amylopectin, then the optimum chain length for phosphorylase (or whatever enzyme system is involved in the synthesis of straight chains) could increase with maturity. Nevertheless, it is pointed

out that the results do not contradict the author's (5) proposed mechanism for the synthesis of starch from glycogen.

**Acknowledgment**

The author would like to take this opportunity to thank Dexter French for his interest and enlightening criticisms. The author also wishes to thank G. F. Sprague and W. A. Russell of the Agronomy Department, Iowa State College, for planting and hand-pollinating the corn.

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