REFERENCES

- Atkin, L., Schultz, A. S., Williams, W. L. & Frey, C. N. (1943). *J. Amer. chem. Soc.* **65**, 922.
- Axelrod, A. E. & Elvehjem, C. A. (1939). *J. biol. Chem.* **131**, 77.
- Barton-Wright, E. C. (1946). *Practical Methods for the Microbiological Assay of the B Complex and Essential Amino Acids*. London: Ashe Laboratories, Ltd.
- Bolliger, A. (1939). *Analyst*, **64**, 416.
- Dorfmann, A. & Koser, S. A. (1942). *J. infect. Dis.* **71**, 241.
- Euler, H. von (1936). *Ergebn. Physiol.* **38**, 1.
- Euler, H. von & Adler, E. (1938). *Hoppe-Seyl. Z.* **252**, 41.
- Euler, H. von, Adler, E. & Eriksen, T. S. (1937). *Hoppe-Seyl. Z.* **248**, 227.
- Gulland, J. M. & Jackson, E. M. (1938). *J. chem. Soc.* p. 1492.
- Handler, P. & Klein, J. R. (1942). *J. biol. Chem.* **143**, 49.
- Handler, P. & Kohn, H. I. (1943). *J. biol. Chem.* **150**, 447.
- Hoagland, C. L. & Ward, S. M. (1942). *J. biol. Chem.* **146**, 115.
- Jandorf, B. J., Klemperer, P. N. & Hastings, A. V. (1941). *J. biol. Chem.* **138**, 311.
- Krebs, H. A. & Henseleit, K. (1932). *Hoppe-Seyl. Z.* **210**, 33.
- Lennerstrand, A. (1941). *Ark. Kemi Min. Geol.* **A14**, No. 16.
- Lwoff, A. & Lwoff, M. (1937). *Proc. roy. Soc. B*, **122**, 360.
- McIlwain, H. (1946a). *Biochem. J.* **40**, 269.
- McIlwain, H. (1946b). *Nature, Lond.*, **158**, 898.
- McIlwain, H. (1947). *Advanc. Enzymol.* **7**, 419.
- McIlwain, H., Roper, J. A. & Hughes, D. E. (1948). *Biochem. J.* **42**, 492.
- McIlwain, H. & Stanley, D. A. (1948). *J. gen. Microbiol.* **2**, xii.
- Mann, P. J. G. & Quastel, J. H. (1941). *Biochem. J.* **35**, 502.
- Myrback, K. (1933). *Ergebn. Enzymforsch.* **2**, 139.
- Neuberg, C. (1942). *J. Amer. chem. Soc.* **64**, 2722.
- Schlenk, F. (1943). *Arch. Biochem.* **3**, 93.
- Schlenk, F. (1945). *Advanc. Enzymol.* **5**, 207.
- Schlenk, F., Hellström, H. & Euler, H. von (1938). *Ber. dtsh. chem. Ges.* **71**, 1471.
- Schlenk, F. & Schlenk, T. (1947). *Arch. Biochem.* **14**, 131.
- Sumner, J. B., Krishnan, P. S. & Sisler, E. B. (1946). *Arch. Biochem.* **12**, 19.
- Warburg, O. & Christian, W. (1936). *Biochem. Z.* **288**, 291.

The Amylose Content of the Starch Present in the Growing Potato Tuber

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During the last few years it has been shown that starch consists of two components: amylose, which is a linear polymer and comparable with cellulose, and amylopectin, which is a branched chain polymer comparable with glycogen. The advances in this field have been greatly assisted by the development of the potentiometric iodine titration method for estimating the percentage of amylose in starch (Bates, French & Rundle, 1943; Hudson, Schoch & Wilson, 1943). By use of this procedure the amylose content of many starches has been determined, and starches have been examined in which the amylose content varies from less than 1% in the waxy starches to over 50% in certain varieties of pea starch (Bates *et al.* 1943; Hilbert & MacMasters, 1946). The published values of amylose contents for starches of similar botanical origin, e.g. maize, sometimes differ and this suggests that the amylose content is not necessarily a constant property of the starch from any given type of botanical species. Furthermore, by use of the newer methods for end-

group determination, and with a knowledge of the amylose content of a starch, it is now possible without separation of the components to determine the proportion of glucose residues present in the amylopectin fraction of the starch as terminal groups (Brown, Halsall, Hirst & Jones, 1948). For example, the sample of rice starch which was examined by Hirst & Young (1939), who used the methylation technique, has now been shown to contain 12% of amylose, and hence we have present in the amylopectin fraction one non-reducing terminal group/26 glucose residues. In other samples of rice starch, however, an amylose content of 14–15% (Brown *et al.* 1948) has been found, and the amylopectin fraction has been shown to contain one non-reducing terminal residue/20 glucose residues. This indicates that different varieties of rice starch do not all contain the same type of amylopectin. It will be recalled that Meyer & Heinrich (1942) have claimed that the starches extracted respectively from potato tubers, leaves and shoots, contain different pro-

portions of amylose and amylopectin, the shoots in particular having a high proportion (approx. 46 %) of the amylose component.

In view of this evidence it was considered desirable to investigate the amylose content and the ratio of terminal residues to non-terminal residues present in the starch of two different varieties of the same botanical species throughout their period of growth. We therefore decided to examine at frequent intervals during the growing season the starch present in the tubers of two varieties of potatoes, one an early and the other a main crop variety.

EXPERIMENTAL

Growing of potatoes. The potatoes were grown from virus-free Golden Wonder and King Edward 'seed', which was very kindly given to us by the Seale Hayne Agricultural College.

Isolation of the starch. The potatoes (c. 600 g.) were washed to remove soil, scraped to remove the skin and cut into small pieces, which were broken up in an ordinary mincing machine. The coarse mash thus obtained was filtered and then ground up with a little water in a mill. The resulting fine slush was filtered, and allowed to stand for some hours in 1 % sodium sulphite solution with intermittent shaking. The solid matter was then allowed to settle out and the supernatant liquid, which was usually dark in colour, was decanted. This process was repeated two or three times until the sulphite solution no longer developed any colour. The solid matter was then suspended in water and filtered through coarse muslin, when fibrous material remained on the cloth whilst the starch granules passed through. They were allowed to settle and then washed by decantation with distilled water, the water being poured off before complete sedimentation had occurred. This procedure ensured that any of the less dense fibrous

material which had come through the filter was removed from the starch. Finally, the starch was filtered off on a hardened smooth filter paper to avoid contaminating it with cellulose fibres, washed first with water and then with methanol, and dried under diminished pressure. The yield of starch was of the order of 40–60 g. More starch of a poorer quality (e.g. slightly discoloured) could be obtained by repeating the above extraction process with the fibrous material left on the muslin filter.

Investigation of the starches. All starch samples were first defatted by treatment in a Soxhlet apparatus for 18 hr. with a constant boiling mixture of dioxan (80 %) and water (20 %) (Schoch, 1942, 1946). The sulphated ash content of the fat-free starch was determined.

The amylose contents of the starch samples were determined by means of the iodine potentiometric method of Bates *et al.* (1943) as modified by Hudson *et al.* (1943). In our determinations a Cambridge potentiometer reading to 0.1 mV. was used in conjunction with a mirror galvanometer. In the calculation of the amylose content the amylose was assumed to take up 21.5 g. iodine/100 g. amylose (Higginbotham & Morrison, 1947). In many of the previous publications on this subject a lower figure (c. 19.2 g. iodine/100 g. amylose) has been used for the calculation of amylose content, and the percentages of amylose quoted will therefore be a little high. It is probable that some of the work which has been reported as having been carried out with pure amylose refers in fact to samples of amylose containing 5–10 % amylopectin.

In addition to determining the amylose content certain other properties were investigated with certain of the starch samples. With these samples nitrogen estimations (micro-Dumas) were carried out to see whether any appreciable amount of protein was being isolated along with the starch. The optical rotations of these samples in *n*-NaOH were also determined, and finally the samples were oxidized by KIO₄, and from the amounts of formic acid produced the number of glucose residues containing one non-reducing terminal

Table 1. *Properties of starches from two varieties of potatoes*

Date of collecting potatoes	Ash content (%)	N (%)	$[\alpha]_D^{20}$ in <i>n</i> -NaOH	Amylose* (%)	A†	B†
Golden Wonder potatoes						
10. vii. 46	0.8	< 0.1	+160°	18	32	26
16. vii. 46	1.2	—	—	18	—	—
24. vii. 46	0.8	—	—	19	—	—
31. vii. 46	0.9	—	—	19	—	—
7. viii. 46	1.2	< 0.1	+169°	18	33	27
14. viii. 46	0.5	—	—	16	—	—
20. viii. 46	0.4	—	—	16	—	—
5. ix. 46	0.7	—	—	16	—	—
10. ix. 46	0.9	< 0.1	+160°	16	32	27
King Edward potatoes						
31. vii. 46	0.6	< 0.1	+159°	16	32	27
7. viii. 46	1.1	—	—	18	—	—
14. viii. 46	0.7	—	—	15	—	—
20. viii. 46	0.6	< 0.1	+159°	15	33	28
5. ix. 46	0.8	—	—	16	—	—
10. ix. 46	0.5	—	—	16	—	—
24. ix. 46	0.8	0.4	+161°	17	33	27

* The estimated error in the figures of amylose contents is $\pm 1\%$, e.g. $18 \pm 1\%$ amylose.

† Col. A gives the number of glucose residues/one non-reducing terminal residue in the whole starch, and col. B the corresponding figure for the amylopectin fraction. The estimated error in the end-group determinations is ± 2 in 30 glucose residues, e.g. 32 ± 2 glucose residues/one non-reducing terminal residue.

residue was calculated for the various samples. From these figures and the percentages of amylose present the number of glucose residues containing one non-reducing terminal residue in the amylopectin fraction was estimated. The results of the analyses and calculations are given in Table 1.

DISCUSSION

The figures for the amylose content of the starch for King Edward potatoes varied between 15 and 18 %, but showed no definite upward or downward trend. In the case of the starch for Golden Wonder potatoes the upper and lower figures were 19 and 16 %, respectively, and the lower values were observed at the later period of growth. The variation is, however, only just outside the estimated range of experimental error, and it is not possible at this stage to decide whether or not it is significant. For neither of these starches can it be said, on the basis of the present data, that any noticeable change in the proportion of amylose and amylopectin occurs during the period of growth. Furthermore, the

results indicate that the amylopectins present in the two varieties of potato are identical, and belong to that group of amylopectins which contain some 24–26 glucose residues/terminal group. This proportion of end-groups is the same as that found also in other samples of potato starch (Brown *et al.* 1948).

SUMMARY

1. Samples of the starches present in the tubers of two varieties of potato, Golden Wonder and King Edward, have been isolated at intervals throughout the period of growth of the plants.

2. No significant variation in the properties of amylose (c. 17 %) and amylopectin (c. 83 %) was observed during the period of growth.

3. In the amylopectin portion of the starches there was one terminal glucose residue for every 24–26 glucose residues, and the amylopectin for the two varieties of potatoes appeared to be identical.

REFERENCES

- Bates, F. L., French, D. & Rundle, R. E. (1943). *J. Amer. chem. Soc.* **65**, 142.
 Brown, F., Halsall, T. G., Hirst, E. L. & Jones, J. K. N. (1948). *J. chem. Soc.* p. 27.
 Higginbotham, R. S. & Morrison, G. A. (1947). *Chem. Ind.* p. 45.
 Hilbert, G. E. & MacMasters, M. M. (1946). *J. biol. Chem.* **162**, 229.
 Hirst, E. L. & Young, G. T. (1939). *J. chem. Soc.* p. 1471.
 Hudson, C. S., Schoch, T. J. & Wilson, E. J. (1943). *J. Amer. chem. Soc.* **65**, 1380.
 Meyer, K. H. & Heinrich, P. (1942). *Helv. Chim. Acta*, **25**, 1038, 1639.
 Schoch, T. J. (1942). *J. Amer. chem. Soc.* **64**, 2954.
 Schoch, T. J. (1946). The fractionation of starch. In *Advances in Carbohydrate Chemistry*, **1**, 274. New York: Academic Press.

The Sorption of DDT and its Analogues by Chitin

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On the basis of a general correlation between the possession of a chitinous cuticle and sensitivity to DDT poisoning, and the fact that chitin will sorb DDT from colloidal suspension, Richards & Cutkomp (1946) put forward the hypothesis that the chitin of the cuticle serves to concentrate DDT selectively from the media with which the insects are treated, and so to give a higher concentration inside the animal.* If this is actually the case then it is reasonable to expect that the toxicity of compounds

similar to DDT will be affected in some degree by the extent to which they are sorbed by chitin, and that, in making comparisons of their potencies account must be taken of the degree of concentration of these substances in the insect by the sorptive action of the chitinous cuticle.

Experiments were therefore carried out to observe the sorption by chitin of a number of DDT analogues from colloidal suspensions. In order to do this it was necessary to devise a method for estimating the DDT or its analogues sorbed from colloidal suspension, and also to obtain a general picture of the effect of time, concentration and amount of chitin present on the amount of DDT sorbed.

* These authors say that DDT is adsorbed by chitin. This implies a surface action and since no proof is offered as to the mechanism of the process the term 'sorption' has been used throughout this paper since this word does not imply any particular mechanism.