LXXI. NOTE ON THE STORAGE OF CARBOHYDRATE AND FAT BY *SACCHAROMYCES FROHBERG* WHEN INCUBATED IN SUGAR SOLUTIONS.

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THE storage of carbohydrate and fat by yeast on incubation in various sugar solutions (glucose, fructose, sucrose, maltose, lactose) was studied by Smedley-MacLean and Hoffert [1923; 1924]; the investigation has now been extended to include galactose.

The brewery yeast used in the previous experiments, a mild ale yeast which did not ferment galactose, produced on incubation in a 4 % solution of this sugar no increase in fat and a fall in carbohydrate. *Saccharomyces Frohberg*, a yeast known to ferment galactose after acclimatisation, was therefore tried. This yeast was acclimatised to galactose by culturing on a hydrolysed lactose and yeast extract medium, and grown, at laboratory temperature, on a 5 % sucrose and yeast extract medium, 4 litres of which furnished about 20 g. of yeast after a week's growth.

6.25 g. of the *Frohberg* yeast were analysed as a control, and two portions of 6.25 g. incubated in 500 ml. of each of the two solutions to be compared, for 44 hours, with oxygenation, at room temperature (*ca.* 15°).

The methods of estimation of carbohydrate and fat were those described in the previous communication.

Possibly owing to the different media on which they had been grown, the carbohydrate content of the *Frohberg* was somewhat lower than that of the brewery yeast, while the fat content of the former was nearly twice as great.

The storage in 2, 4 and 5 % glucose and galactose solutions was studied, equal portions of the same sample of yeast being incubated, under the same conditions, in the two solutions to be compared. An increase of carbohydrate storage and of fat storage with increasing concentration of sugar was shown both in glucose and galactose solutions. There was no significant difference between the amount of carbohydrate stored on incubation in the same concentration of glucose or galactose; in glucose, however, more fat was stored at all concentrations than in galactose (see Figs. 1 and 2).

The effect of the presence of phosphate (0.49 g. $\text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O} + 0.143 \text{ g. KH}_2\text{PO}_4$ per 500 ml. of solution, p_{H} about 7.8) in 5 % solutions of glucose and galactose was then observed. It was shown previously that addition of phosphate to solutions of glucose, fructose, sucrose and maltose, brought about an increased storage of fat and also of phosphorus in brewery yeast incubated therein. This effect was marked and led to the suggestion that fat formation in yeast proceeded *via* hexosephosphate. The *Frohberg* yeast behaved differently. When it was incubated in a 5 % glucose solution to which phosphate had been

added there was no increase in fat storage, the variation was rather a decrease but not of significant amount; the presence of phosphate in a 5 % galactose solution caused a slight increase in fat, but this was not of the same order as the increases referred to above.

Fig. 1.

Fig. 2.

Estimation of phosphorus content, by Neumann's method, showed no significant variations.

A somewhat more marked effect seems to be shown on carbohydrate storage. The addition of phosphate to a glucose solution caused a diminution, to a galactose solution an increase, in carbohydrate content.

Estimation of the residual sugar in a solution in which yeast had been incubated, showed that in parallel experiments, nearly twice as much glucose was used up as galactose. Tests were performed to ascertain whether the *Frohberg* yeast lost its power of fermenting galactose when grown on sucrose media and the results indicated that this was so; since the amount of galactose (up to 10 g.) used up during a 44-hour incubation could not, however, have been accounted for by increased storage within the cell and some fermentation of galactose must have taken place though this was probably retarded. There was no

Table I.

The numbers represent g. per 12.5 g.* original yeast.

% sugar in medium	Original yeast	After incubation in glucose solution		After incubation in glucose + phosphate solution		After incubation in galactose solution		After incubation in galactose + phosphate solution		After incubation in maltose solution		After incubation in fructose solution		After incubation in sucrose solution	
		Carbo-hydrate g.	Fat g.	Carbo-hydrate g.	Fat g.	Carbo-hydrate g.	Fat g.	Carbo-hydrate g.	Fat g.	Carbo-hydrate g.	Fat g.	Carbo-hydrate g.	Fat g.	Carbo-hydrate g.	Fat g.
2	0.72	0.172	1.00	0.406	—	0.88	0.288	—	—	—	—	—	—	—	—
	0.82	0.166	0.76	0.414	—	0.88	0.314	—	—	—	—	—	—	—	—
	1.00	0.144	0.84	0.374	—	0.96	0.330	—	—	—	—	—	—	—	—
	0.98	0.128	0.92	0.376	—	—	—	—	—	—	—	—	—	—	—
4	Mean	0.88	0.152	0.88	0.392	—	0.90	0.310	—	—	—	—	—	—	—
	0.70	0.178	1.10	0.496	—	1.03	0.364	—	—	—	—	—	—	—	—
	0.63	0.156	1.22	0.410	—	1.07	0.310	—	—	—	—	—	—	—	—
	—	—	1.13	0.450	—	—	—	—	—	—	—	—	—	—	—
5	Mean	0.66	0.166	1.16	0.454	—	1.04	0.336	—	—	—	—	—	—	—
	0.96	0.238	1.41	0.552	0.96	0.386	1.46	0.406	1.74	0.508	0.90	1.16	0.380	1.02	0.370
	1.06	0.150	1.00	0.492	0.78	0.482	1.14	0.366	0.88	0.338	1.33	0.98	0.346	1.24	0.348
	0.93	0.182	1.12	0.482	0.90	0.366	1.34	0.370	1.20	0.404	—	—	—	—	—
	0.84	0.166	1.27	0.366	0.82	0.368	1.08	0.338	1.50	0.348	—	—	—	—	—
	0.82	0.158	—	—	—	—	1.05	0.316	—	—	—	—	—	—	—
	0.74	0.158	—	—	—	—	—	—	—	—	—	—	—	—	—
	0.94	0.180	—	—	—	—	—	—	—	—	—	—	—	—	—
	0.96	0.194	—	—	—	—	—	—	—	—	—	—	—	—	—
	0.94	0.180	—	—	—	—	—	—	—	—	—	—	—	—	—
	1.02	0.152	—	—	—	—	—	—	—	—	—	—	—	—	—
	0.67	0.158	—	—	—	—	—	—	—	—	—	—	—	—	—
	0.71	0.138	—	—	—	—	—	—	—	—	—	—	—	—	—
	Mean	0.84	0.160	1.20	0.464	0.86	0.400	1.22	0.360	1.32	0.400	1.11	0.272	1.07	0.363

Milligrams phosphorus per 12.5 g. of original yeast in g.

Mean

Original yeast

After incubation in 5 % glucose solution

After incubation in 5 % glucose + phosphate solution

After incubation in 5 % galactose solution

After incubation in 5 % galactose + phosphate solution

72, 66, 68, 76, 98
74, 44 — — —
64, 98, 88, 106 —
80, 56, 84 — —
84, 70, 66, 54 —

76

60

88

74

68

* Results are given per 12.5 g. of yeast for comparison with previous work on brewery yeast.

difference between the amount of sugar used up from a sugar solution and from a similar solution with phosphate.

The *Frohberg* yeast was also incubated in 5 % solutions of maltose, fructose, and sucrose. The carbohydrate storage, in these sugars, was of the same order as in glucose and galactose at the same concentration; particularly remarkable was the total absence of the extra carbohydrate storage shown by brewery yeast in maltose, attributed then to the possible direct formation of glycogen from maltose. Fat storage was poorer in maltose solution than in galactose and in fructose and sucrose of the same order as in galactose; it was in no case so good as in glucose.

SUMMARY.

1. *Carbohydrate storage.* The carbohydrate storage of *Saccharomyces Frohberg* on incubation in solutions of glucose, galactose, fructose, sucrose and maltose of the same strength is similar in all cases. Glucose and galactose were investigated in varying concentration whereby it was shown that there was an increased storage of carbohydrate with increased concentration of either sugar in the medium.

Frohberg yeast behaved similarly to brewery yeast when incubated in solutions of glucose, galactose, fructose and sucrose but showed a marked difference on incubation in maltose. While *Frohberg* yeast stored normal amounts of carbohydrate in the case of this sugar, in brewery yeast an abnormally high carbohydrate storage occurred.

2. *Fat storage.* *Frohberg* yeast showed a greater power of forming fat when incubated in solutions of glucose than in those of galactose, fructose, sucrose, or maltose; the last seemed the least effective. In the cases of glucose and galactose the amount of fat stored increased with increase of sugar in the surrounding medium. The addition of phosphate to the medium did not appear to cause any such increase of fat storage as was previously noted in the case of brewery yeast.

REFERENCES.

- Smedley-MacLean and Hoffert (1923). *Biochem. J.* **17**, 720.
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