

# HYPER BRANCHED STARCHES - A NEW CARBOHYDRATE CLASS: RESULTS OF AN *IN VITRO* DIGESTION STUDY

O. Häusler, L. Deremaux, C. Petitjean

Roquette Frères, 62080- Lestrem, France

Email [olaf.haeusler@roquette.com](mailto:olaf.haeusler@roquette.com), Tel +33 (321) 63-3753

## INTRODUCTION

Naturally occurring starches are consisting of two polymers, the linear amylose and the branched amylopectine. The amylopectine content is important for basic properties of these starches such as the physical stability in aqueous solution (retrogradation) or the digestibility by human amylases. Due to the fact that amylopectine contains both  $\alpha$  1,4 and  $\alpha$  1,6 linkages (in the branching), it is possible to use the ratio of both linkage types for characterization of the branching rate. The common starches such as potato or maize starch do not exceed 4% of  $\alpha$  1,6 bonds, whereas pure vegetable amylopectine contains 5% of these bonds.

A new enzymatic process [1, 2] opens the production route for starches having higher branching rates than usually found in plants. Branching enzymes cut the 1,4 linkages, typically in linear amylose, and create new 1,6 bonds by fixing these starch chains on the substrate structure. No other new bonds, such as 1,2 or 1,3 (as occurring in pyrodextrins) are formed due to this specific enzymatic reaction pattern. These hyper branched starches (HBS) offer various new applications due to their new physicochemical properties as well as to a higher resistance to digestion by human amylases or amylo-glucosidases.

## EXPERIMENTAL METHODS

### Materials

Experimental products from Roquette Frères were used for these trials. They are obtained by treating starch paste (obtained from commercial starch such as maize starch) with specific branching enzymes. Optional enzymatic treatment could be done to increase the branching rate of the obtained product. The reaction products are purified and decolorized using black carbon treatment.

Last step is spray drying to obtaining stable and cold water soluble powders.

### Branching rate

Samples are dissolved in D<sub>2</sub>O and measured with <sup>1</sup>H-NMR- spectrometry. The signals for the glycosidic linkages  $\alpha$  1,4 and  $\alpha$  1,6 are quantified. The “branching rate” is calculated from the percentage of  $\alpha$  1,6 glycosidic bonds, related to the sum of all bonds.

### Molecular weight

The sample is dissolved in pure water and analyzed with size exclusion chromatography, equipped with a MALLS- Detector. The M<sub>w</sub> – distribution is recorded, the mean value of the complete weight distribution is calculated.

### *In vitro* digestibility

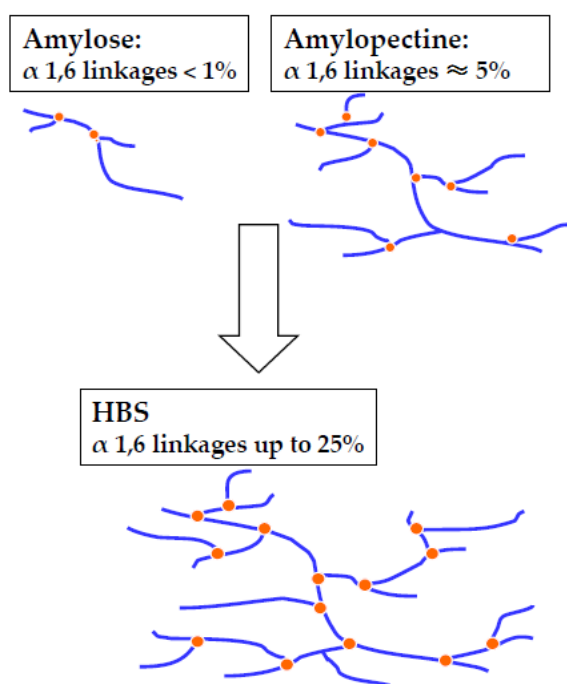
The chosen model simulates the human digestion of a carbohydrate under standardized experimental conditions. It is a part of an internal serial screening of the digestibility of carbohydrate based products. The test sample is dissolved in maleate buffer (pH 7). Pancreatin from porcine pancreas (Sigma-Aldrich, Ref. P 7545) is added. The blend is incubated for 30min @ 37°C. After this time, an intestinal acetone powder from rat intestine (Sigma-Aldrich, Ref. I 1630) is added and the incubation continues for 3hours 30min further.

Samples are taken at different times. The free glucose content is measured by colorimetry, using Roche Hitachi 704 equipment (Roche Diagnostics AG). The digestion process is evaluated by calculation and expressed as a percentage of released glucose, related on the dry substance of the test material. Maltose serves as a standard for comparison, water as control.

## RESULTS AND DISCUSSION

All tested samples were freely soluble in cold water. The solutions were physically stable and had a lower viscosity, compared with starch products at the same concentration. The NMR analysis indicates the presence of 1,4 and 1,6 bonds only, but with variable ratios. No other glycosidic linkages were detectable. HBS with a branching rate of 25% have been obtained. Increasing the branching rate resulted in products with a lower molecular weight but with a sugar content of less than 1%. All products had a very narrow molecular weight distribution. Figure 1 illustrates in a schematic way the structure of common starches and of the HBS.

Figure 1: Structure of hyper branched starches



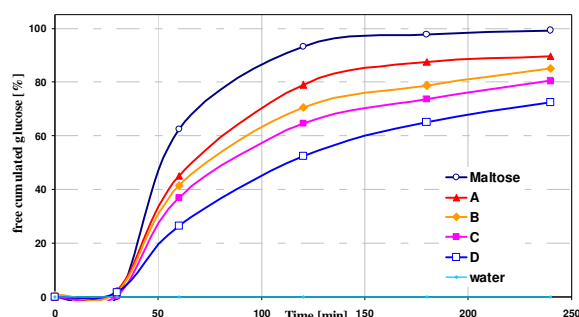
Some selected products have been tested using the established *in vitro* model. Their main properties are given in Table 1.

Table 1: Characterization of selected experimental products.

Sample	[%] 1,6 bonds	Molecular weight
Maltose	0	342
A	7.6	127 000
B	10.3	116 000
C	13.3	76 000
D	20.6	40 000

The *in vitro* digestion studies indicate a resistance to rapid digestion. Increasing the branching rate reduces the enzymatic digestion speed (see Figure 2).

Figure 2: Results of *in vitro* digestion studies



Due to the adaptable digestibility (mainly correlated with the branching rate), HBS could be valuable for applications where reduced metabolism speed is needed. These new starches are “slow carbohydrates” as requested e.g. in sport foods. It could also serve as a regulator of the digestion speed and of the intestinal absorption, inducing longer lasting diffusion toward the plasmatic compartment. These new polymers are a source of slowly available glucose, such as classified by Englyst [3]. HBS may also have prolonged plasma half time when injected, so that they are valuable candidates for all parenteral applications such as plasma volume expander or osmotic agents for peritoneal dialysis. The absence of any other bond than 1,4 and 1,6 should guarantee a high clinical tolerance and the absence of immunological reactions, such as those known for dextrans.

## REFERENCES

- [1] EP 1.177.216 B1
- [2] EP 1.369.432 B1
- [3] Englyst *et al*, Am. J. Clin. Nutrition, (69), 1999, 448-454