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Gelatinisation Properties of Different Cereals and Pseudocereals

During the mashing process for beer production, intact starch granules can not be hydrolysed below the gelatinisation temperature in an appropriate time. This is an important fact, when adjunct is used. The inactivation temperatures of amylolytic enzymes of barley malt are about 70 °C for β-amylase and 80 °C for α-amylase. To guarantee sufficient starch degradation to fermentable sugars, gelatinisation has to take place prior to amylolysis. For an optimised mashing process the gelatinisation temperature is an important specification. Still it is not a recommended method in the brewing industry. In this work we used a method that is established in other industries. We analysed different cereals and pseudocereals that could be used as adjunct for the brewing process. Further mashing trials were done on different rice samples to demonstrate the importance of the gelatinisation temperature in terms of brew house yield and wort quality.

Descriptors: Gelantinisation temperature, rotation viscosimeter, adjuncts, cereal, pseudocereal

1 Introduction

Worldwide unmalted cereals and pseudocereals are used as malt surrogates for wort and beer production as extract source. Most common cereals are maize, rice and sorghum. In most European countries wheat, maize, barley and rice are allowed for beer brewing purposes.[10] In common literature different definitions exist for the term adjunct.[3, 4] In this publication, adjunct is a synonym for unmalted cereal or pseudocereal.

There are different reasons for using adjuncts in the brewery. On the one hand there are quality aspects and on the other hand there are economical reasons. Beer brewed with adjuncts as an ingredient has low amounts of nitrogen as a consequence. That beer is supposed to be lighter and more stable than beer brewed with 100 % barley malt.[3, 4] The economical reasons are based on the inexpensiveness of adjuncts. Up to 40 % of the throw can be substituted by adjunct without the necessity of adding enzymes. [1, 11]

The main purpose of mashing is to degrade starch to fermentable sugars. This is done by amylolytic enzymes. Starch granules from malted cereals are already attacked by enzymes during germination process. Therefore they can be hydrolysed below gelatinisation temperature.[2, 15] Native starch granules as they occur in unmalted cereals and pseudocereals have to be gelatinised before they are degraded by amylases.[2] It has been mentioned before that unmalted cereals like maize, rice and wheat have to be cooked prior to amylolysis [3] and that a low gelatinisation temperature of the adjunct is of benefit.

Neither ASBC [17], EBC [5] nor MEBAK [12] recommend a method to determine gelatinisation temperature, even though Coors already published in 1976 the gelatinisation temperature as a quality aspect. To demonstrate the importance of such a method, mashes with 80 % rice were done. Different temperatures for the rice "cooker" were chosen. Two rice samples with different gelatinisation temperatures were used, and the wort was analysed for extract yield and apparent attenuation limit (AAL).

The aim of this paper is to introduce a method which has been mentioned elsewhere [7, 9] to determine the gelatinisation temperature, as well as to demonstrate by two rice samples that a low gelatinisation temperature allows infusion mashing. To give an overview several cereals and pseudocereals were investigated for their gelatinisation temperature.

2 Materials and methods

2.1 Micro scale mashing

Three different mashing procedures were carried out using 40 g of rice and 10 g of barley malt and 200 g distilled water. The time temperature program is presented in figure 1. For mashing procedure 1, rice and malt were mashed in together at 65 °C. For procedure 2 (mashing-in temperature (t_{mi}) = 70 °C) and procedure 3 (t_{mi} = 85 °C), rice was mashed in alone and malt was added after cooling the mash down to 65 °C (figure 1). At the end of the mashing process mashes were cooled down, filled up with distilled water to 450 g and filtered. The worts were analyzed for extract, viscosity, AAL and fermentable sugars.

Rice was grounded using the Laboratory Mill 3100 (0.8 mm sieve, 16,000 rpm) from Perten Instruments GmbH, Hamburg, Germany. Two rice samples were used. Rice A was a kind gift of Euryza GmbH, Hamburg, Germany, and rice B was broken rice from Brazil. The barley malt was of standard quality. All mashing procedures were carried out in duplicate.

2.2 Determination of sugars

Fermentable sugars were quantified using high-performance anion-exchange chromatography (HPAEC) with pulsed amperometric detection (PAD). The apparatus used was supplied by Dionex Cooperation, Sunnyvale, USA, and consisted of the following parts:

- Autosampler (including thermal compartment and eluent organiser): AS 50
- Gradientpump: GP40

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Tables and Figures see Appendix.

- Detector: ED 40 with a gold working electrode

The water used for the method or for sample preparation was deionised (resistivity = 18.2 MΩ-cm). Samples were cooked for 5 s to inactivate all amylolytic activity. To the sample 2-deoxy-d-glucose (Sigma, St. Louis, USA) and cellobiose (Sigma, St. Louis, USA) were added and deionised water was added to have a final dilution of the sample of 1:1000. 2-deoxy-d-glucose was used as internal standard for glucose, fructose and sucrose. Cellobiose was used as internal standard for maltose and maltotriose.

2.5 µl of sample were injected and separated using a CarboPac PA 10 guard column (25 × 2) and a CarboPac PA 10 analytical column (100 × 2). The oven compartment was set to 30 °C and the flow was set to 0.25 ml/min. The gradient used is shown to table 1. Eluent A consisted of 250 mM NaOH prepared using 50 % (w/w) sodium hydroxide (Baker, Netherlands). Eluent B was deionised water. Both eluents were kept under pressure using helium.

Table 1 Gradient for sugar separation

Time [min]	Eluent A [%]	Eluent B [%]
-10.0	9	91
0.0	9	91
17.9	9	91
18.5	80	20
55.0	80	20
55.1	9	91
75.0	9	91

The potentials for the PAD are presented in table 2.

Table 2 Settings of the PAD

Time [s]	Potential [V]	Integration
0.00	0.1	
0.20	0.1	begin
0.40	0.1	end
0.41	-2.0	
0.42	-2.0	
0.43	0.6	
0.44	-0.1	
0.50	-0.1	

2.3 Wort Analysis

All wort analysis were carried out according to MEBAK methods.[12, 14]

2.4 Determination of the gelatinisation temperature

The gelatinisation temperature was determined measuring the rheological behaviour of a flour water suspension using the rapidviscoanalyser RVA-4 from Newport Scientific, Australia. The method is based on the change of viscosity caused by the hydration, swelling and disintegration of starch granules and a possible realignment of starch molecules.[9] The viscosity is influenced by the rate of temperature increase and decrease as well as by the shear rate caused by a rotor.[16] Therefore the temperature-time profile and the rotor speed are important parameters. The gelatinisation temperature is the temperature, where an abrupt increase of viscosity happens.[16] Since only that specification is in the focus of our investigations, the pasting behaviour was only considered to that point. Analyses were carried out, according to ICC standard No. 162 [7], using the manufactures profile STD 1 (fig. 2). The high speed rotation in the beginning of this method is used for homogenisation of the suspension. Data collection was done in 0.5 s intervals instead of 4.0 s. Samples were grounded using the Laboratory Mill 3100 (0.8 mm sieve, 16,000 rpm) from Perten Instruments GmbH, Hamburg, Germany.

The weighted sample, according to the standard methods, was based on 14 % moisture. Variations were corrected by formula 1 and 2 (fig. 3).

$$\textit{formula}\ (1)\ E_k = \frac{86 \times E}{100 - W_P}$$

$$\textit{formula}\ (2)\ W_k = 25 + E - E_k$$

E = Sample weight [g]

E_k = corrected sample weight [g]

W_p = moisture content of sample [%]

W_k = corrected water weight [g]

Fig 3 Formula for sample and water weights

3 Results and discussion

3.1 Micro scale mashes

In figure 4 the extract yields of the mashing procedures are shown. Rice B has a significant increase in extract yield, when the mashing-in temperature is raised from 70 to 85 °C. For rice A the yield is not effected that much. There are no relevant changes in AAL for both rice samples. This indicates that the soluble starch is degraded to fermentable sugars and that the starch of rice B needs higher temperatures for gelatinisation until it can be hydrolysed. The viscosity (fig. 5) of the wort based on 8.6 % extract is increasing with the mashing-in temperature. For procedures 1 and 2 rice A has a significantly higher viscosity.

3.2 Fermentable sugars

Table 4 gives an overview of the fermentable sugars. According to the higher gravity of the wort, rice A has higher yields of glucose, maltose and maltotriose for the procedures 1 and 2. The glucose yield is the highest one for procedure 1 and is decreasing with increasing mashing-in temperature. Maltase, an α-glucosidase [E.C. 3.2.1.20], catalysis the hydrolytic cleavage of maltose to glucose. Maltase from barley has a temperature optimum of 35–40 °C and is inactivated by temperatures above 50 °C very rapidly.[13] Therefore α-glucosidase from rice, which has an optimum at 55 °C [8], must be responsible for the high glucose yield.

Table 5 Statistical analysis of the gelatinisation temperature

Run	Gelatinisation temperature [°C] automatic	manual
1	81.5	78.3
2	82.3	77.4
3	52.9	76.6
4	64.5	78.3
5	53.8	78.3
6	78.2	78.2
7	80.7	78.2
8	84.8	78.3
9	84.0	76.7
10	85.5	77.4
11	83.1	77.4
mean	75.5	77.7
SD	12.4	0.7
CV [%]	16.4	0.9

3.3 Gelantinisation temperature of rice samples

Figure 6 shows the curves of the RVA of rice A and B. Rice A has a gelatinisation temperature of 67 °C and rice B of 81 °C. These results explain the low extract yield for rice B using mashing procedures 1 and 2. Mashing procedure 3 has a mashing-in temperature of 85 °C. This temperature is high enough for rice B to gelatinise.

3.4 Statistical evaluation of the RVA

Table 5 shows the data of eleven runs that were done with the same sample. The automatic determination of the pasting temperature has a mean value of 75.5 °C and a relative standard deviation of 16.4 %. The results of the manual determination are 77.7 °C and 0.9 %. The manual evaluation is to be preferred.

4 Conclusion

The mashing trials with different mashing-in temperatures have demonstrated that the extract yield is strongly affected by that parameter. Mashing-in temperatures far below the gelatinisation temperatures lead to poor extract yields. Knowing that temperature is of benefit since the mashing-in temperature can be adjusted either upwards or downwards. In both cases it is an economical advantage. Lowering the temperature saves energy and lifting it avoids loss of extract.

Determination of the gelatinisation temperature should be a routine analytic for the quality control. Even though it is time consuming, it is advisable to do a manual evaluation of the gelatinisation temperature instead of the software controlled one. The relative thermostability of rice alpha-glucosidase could be an interesting tool to produce beer with an ester flavour.[6]

5 Appendix

Since rice is not the only cereal used as adjunct in the brewery, the authors present in table 6 a list of cereals and pseudocereals we investigated for their gelatinisation temperature at our chair in the past year. The table shows a randomised collection. A meaningful weight sample was investigated by trials.

In table 6 the cereals and pseudocereals and the number of each investigated are shown also the number of samples analysed are mentioned. For wheat, barley, buckwheat, oat, sorghum, emmer, einkorn, proso millet, durum and kamut the amount of samples presented in the column "Samples investigated" is equivalent to the number of varieties. The rice samples, including broken rice as it is delivered to breweries, as well as samples donated by Euryza GmbH, Germany, were of food quality. The maize samples were grits as they are used for brewing.

Table 6 shows the average gelatinisation temperatures and the minimum and maximum values of the investigated cereals and pseudocereals. The widest range between maximum and minimum can be found in the rice samples. As shown before, a lower gelatinisation temperature can result in a higher extract yield at relative low temperatures in the adjunct "cooker". Triticum has the lowest average gelatinisation temperature, whereas and oat has the highest. Rice shows the widest spread between minimum and maximum of 24 °C.

6 Literature

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Appendix

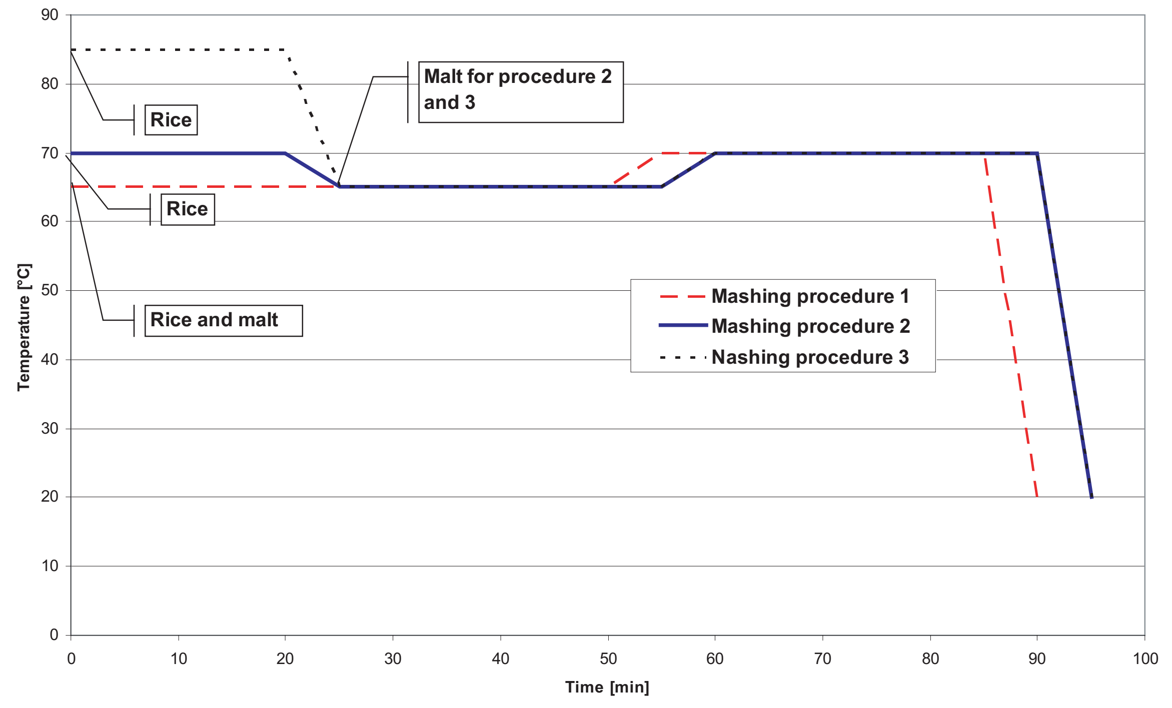

Fig. 1 **Mashing profile**

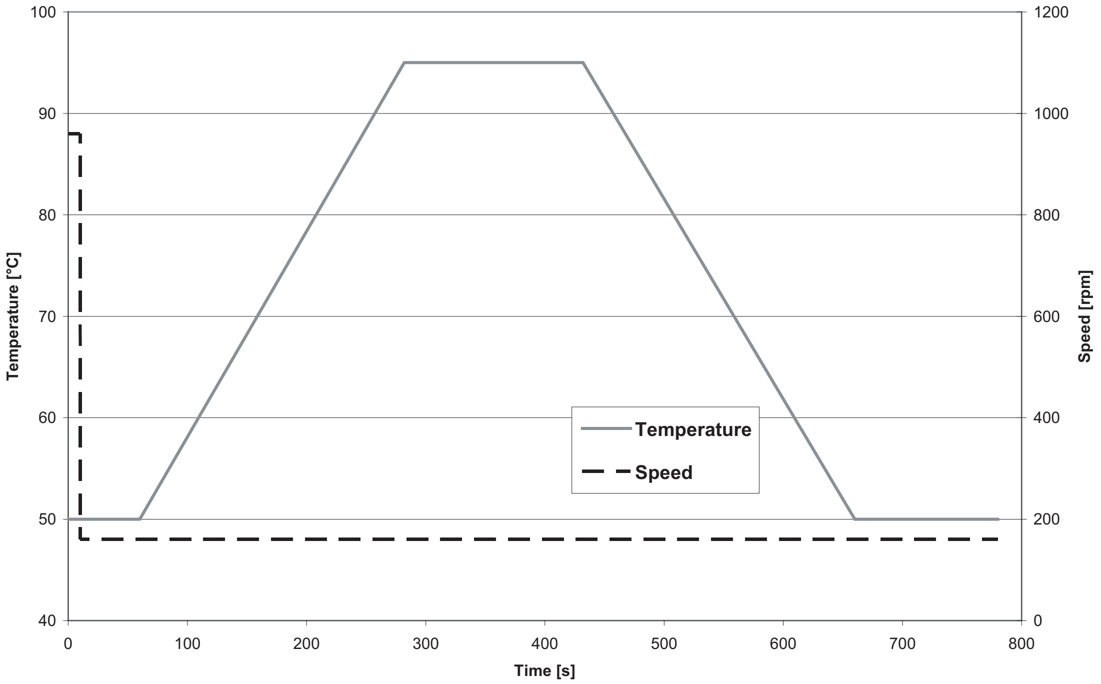

Fig. 2 Temperature time profile of the RVA

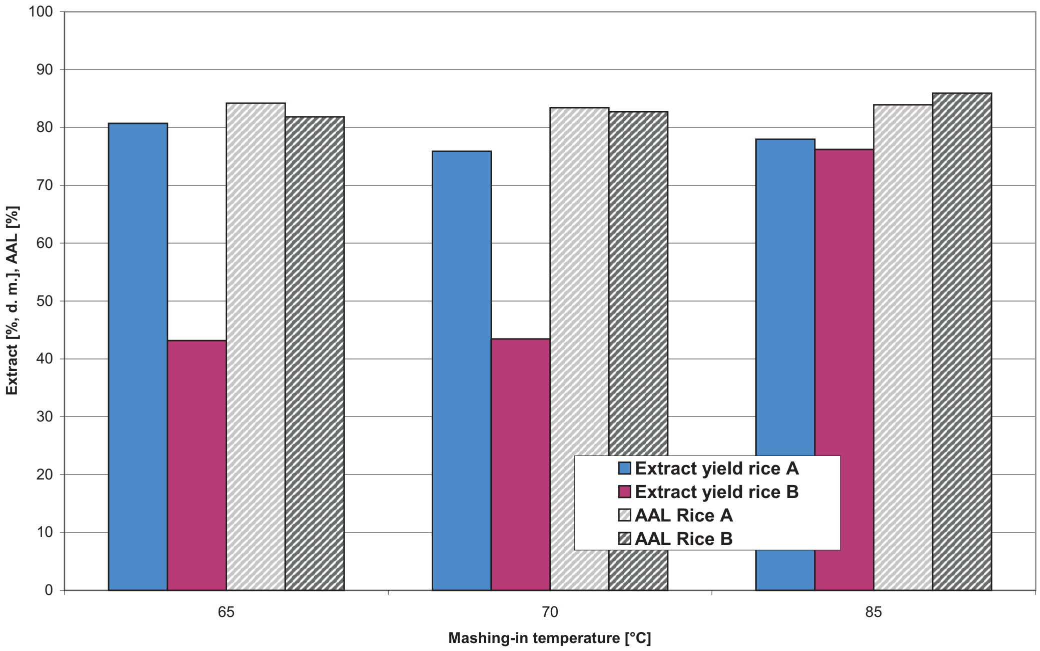

Fig. 4 Extract yield and AAL

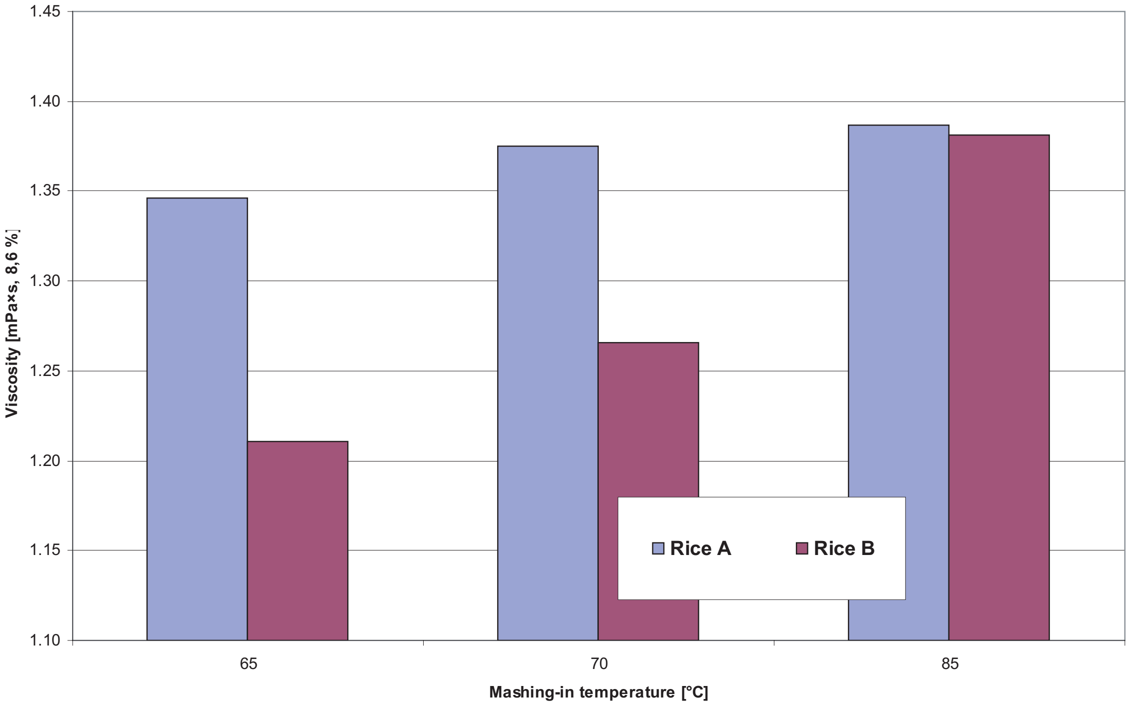

Fig. 5 Changes in viscosity

Table 3 Analyses of micro scale mashing

Mashing-in temperature [°C] Rice sample	65 A	 B	70 A	 B	85 A	 B
Gelatinisation temperature [°C]	67	81	67	81	67	81
Moisture malt [%]	4.5	4.5	4.5	4.5	4.5	4.5
Moisture rice [%]	12.7	10.6	12.7	10.6	12.7	10.6
Extract [% w/w]	9.05	5.07	8.56	5.10	8.77	8.61
Extract [% w/v]	9.36	5.16	8.83	5.19	9.06	8.89
Extract [%, d. m.]	80.7	43.2	75.9	43.5	78.0	76.2
Viscosity (8.6%) [mPa×s]	1.346	1.211	1.375	1.266	1.387	1.381
AAL [%]	84.2	81.8	83.4	82.7	83.9	85.9

Table 4 Fermentable sugars

Mashing-in temperature [°C] Rice	65 A	 B	70 A	 B	85 A	 B	CV [%]
Fructose [g/l]	0.05	0.92	0.07	0.03	0.08	0.01	4.0
Glucose [g/l]	16.40	14.55	8.09	2.85	3.63	2.58	1.1
Sucrose [g/l]	0.73	n. d.	0.89	1.16	0.89	1.01	1.0
Maltose [g/l]	37.61	13.08	44.77	25.61	52.37	52.52	2.6
Maltotriose [g/l]	10.46	2.87	9.99	5.31	10.77	12.47	1.3

n. d. = not detectable

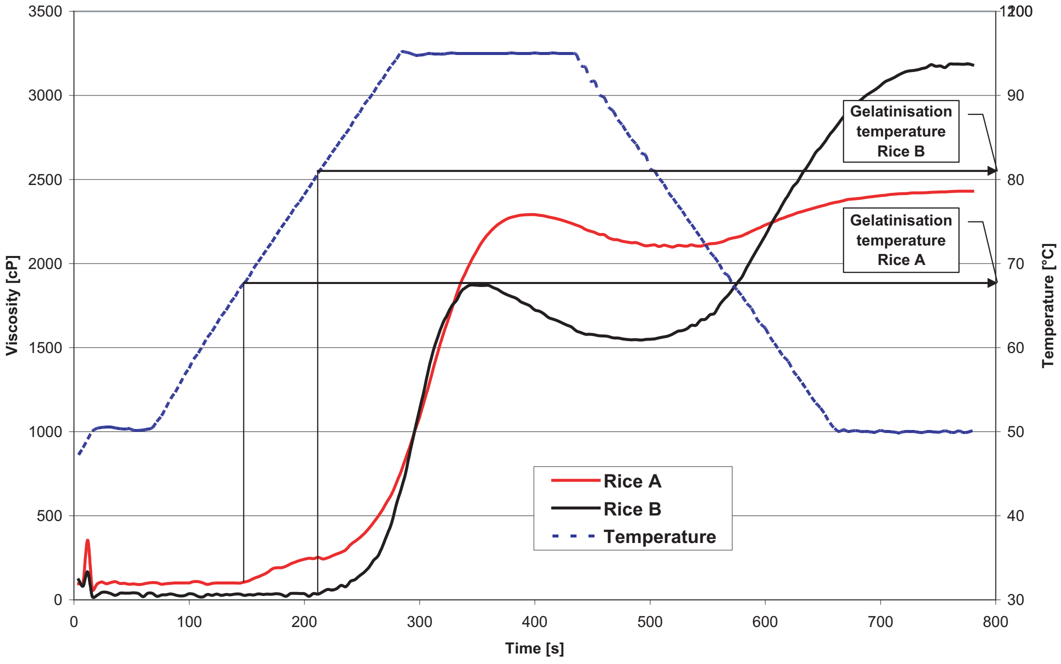

Fig. 6 RVA curves of rice A and B

Table 6 Gelatinisation temperatures of different cereals and pseudocereals

Cereal/ Pseudocereal	Samples investigated	Origin	Gelatinisation temperature [°C]			Spread
			Minimum	Maximum	Average	
Amaranth	1	Bolivia	–	–	64	–
Barley	16	Germany	65	69	67	4
Buckwheat	2	Germany	–	–	70	–
Durum	4	Germany	62	64	63	2
Einkorn	1	Germany	–	–	65	–
Emmer	1	Germany	–	–	64	–
Kamut	1	Germany	–	–	66	–
Maize	9	Europe	73	79	76	7
Oat	6	Germany	89	91	90	2
Proso millet	8	Germany	73	77	75	4
Quinoa	1	Peru	–	–	64	–
Rice	25	Europe/ South America	67	91	81	24
Sorghum	1	Germany	–	–	69	–
Triticale	10	Germany	56	64	61	8
Wheat	6	Germany	63	66	64	4