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Partitioning of water in binary biopolymer mixtures at low water content

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The dependence of the crystallinity of starch on water content has been used to monitor the amount of water associated with starch when it is mixed with egg white or xanthan gum. Measurements of the water sorption isotherms in the water activity range 0.2–0.9 predict that less water is associated with the starch in the mixture than would be obtained for starch alone at the same water activity. This is confirmed by the observed crystallinity. The effect can be quantified in terms of an interaction parameter ξ which is the ratio of the water content of starch in the mixture to that of starch alone at the same water activity. For starch /egg white mixture $\xi = 0.79$, whereas for starch/xanthan gum mixtures $\xi = 0.9$.

INTRODUCTION

In the food industry, it is of prime importance to understand the hydration behaviour of foodstuffs (Franks, 1991). However, the complexity of food products makes this difficult (Slade & Levine, 1991). A food product is normally composed of several compounds, often of unknown and polydisperse molecular weights and the interactions between them can give hydration behaviour which is not simply the sum of the hydration behaviour of the individual components (Chuang & Toledo, 1976; Iglesias *et al.*, 1980). The objective of the work described in this paper is to measure the distribution of water between two biopolymers in low moisture content mixture.

It is widely accepted in the literature (Lang & Steinberg, 1980; Leiras & Iglesias, 1991) that the shape of the sorption isotherms and hence the water partitioning within a mixture can be calculated from a weighted average of the sorption behaviour of the individual components. Thus the moisture content (g water/100 g dry solids) of a mixture of *i* components will be given by:

$$M_{\text{mix}} = \frac{\sum W_i M_i}{\sum W_i} \qquad (1)$$

where M_{mix} is the equilibrium moisture content of the mixture at a specific water activity, M_i the equilibrium moisture content of the *i*th component in the pure form at this water activity and W_i the dry weight of the *i*th component in the mixture.

From the sorption isotherms, the water partitioning can be predicted at any water activity if we assume that:

(a) the mixture is at equilibrium and so the water activity of all of the components are equal and equal to the overall water activity of the mixture;
(b) the sorption characteristics of each component in the mixture are unaltered by the presence of the other components.

The moisture content of a component *i* in the mixture at a particular water activity can be obtained from the sorption isotherm of the pure component. Several mathematical equations have been proposed in the literature to describe the sorption isotherm, one of the most widely used models being the G.A.B. model (equation (2)).

$$\frac{w}{w_m} = \frac{C.k.a_w}{(1 - k.a_w).(1 - k.a_w + C.k.a_w)} \qquad (2)$$

where w is the water content on a dry weight basis, w_m is the 'monolayer' moisture content, a_w the water activity and C and k are constants. This equation has been shown to successfully describe water sorption isotherms up to a water activity of 0.9 (Van der Berg, 1991; Jowitt & Mekenna, 1988).

It has been observed, however, that the weight average relationship does not accurately describe some mixtures, the observed sorption isotherms being closer to one component of the mixture than would be

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predicted by the ratio of their masses (Kaminski & Al-Bezweni, 1994). An interaction parameter, ξ, which is the factor by which the sorption isotherm of component i must be multiplied to give the observed value for M_{mix}, has been introduced to describe this non-ideal behaviour described by Kaminski and Al-Bezweni (1994). Equation (1) is thus modified to give:

$$M_{\text{mix}} = \frac{\sum W_i \xi_i M_i}{\sum W_i} \tag{3}$$

We have measured the water partitioning between the two components in a binary biopolymer mixture and the sorption isotherms of the mixture and the two separate components. This allowed us to determine whether the water partitioning could be predicted from the sorption isotherms of the two components.

To enable water partitioning to be measured experimentally, it is necessary to have a method determining the water content associated with one of the components in the mixture. In the case of native starch at low moisture content, it is possible to use X-ray powder diffraction to do this as starch is a semi-crystalline material and the percentage of crystallinity is dependent on its moisture content (Bear & French, 1941).

In this paper we have used this approach to compare the measured water associated with the starch with that predicted from sorption isotherms in mixtures of starch and the common, non-crystalline food hydrocolloids, egg white or xanthan gum.

MATERIALS AND METHODS

The egg white used was spray dried egg white powder, kindly supplied by Marlow Foods Ltd. Xanthan gum was supplied by Sanofi Bio-industries and the starch was an unmodified wheat starch supplied by Sigma Inc. The other chemicals used were of reagent grade and were supplied by Fisons Ltd.

All measurements were performed on samples which were dried (70°C under vacuum for 18 h) before mixing/rehydration to ensure that all measurements were performed following adsorption rather than desorption.

Mixed powder samples were prepared by shaking measured amounts (~100 g) of the pre-dried powders in a large glass bottle (3 l). Following mixing, the protein content of a 1 g sample was determined in various parts of the mixture using the Biuret method (Plumber, 1978). This demonstrated that the mixture was homogeneous on this sample scale.

Samples (*c.* 1 g) were prepared to known controlled moisture levels by isopiestic equilibration over saturated salt solutions (Table 1) at 25±0.5°C for 7 days (Wolf *et al.*, 1984). Samples were shown to approach equilibrium (weight change $\leqslant 0.001$ g/h) in approximately 48 h (Fig. 1).

Table 1. Saturated salt solutions used to prepare samples to controlled water activities

Salt solution	Formula	RH[a] (%)
Phosporus pentoxide	P_2O_5	1.5
Lithium bromide	LiBr	6
Lithium chloride	LiCl	11
Potassium acetate	KAc	22
Magnesium chloride	$MgCl_2$	30
Potassium carbonate	K_2CO_3	44
Sodium dichromate	$Na_2Cr_2O_7$	54
Sodium bromide	NaBr	59
Cupric chloride	$CuCl_2$	67
Sodium chloride	NaCl	75
Ammonium sulphate	$(NH_4)_2SO_4$	78
Potassium chloride	KCl	85
Potassium nitrate	KNO_4	94
Potassium sulphate	K_2SO_4	98
Water	H_2O	100

[a] RH, relative humidity.

Water activity measurements were carried out at 25°C on a Decogon CX-1 water activity meter following rehydration. Moisture contents for all samples were obtained gravimetrically by drying at 70°C under vacuum for 18 h.

X-ray powder scattering patterns were recorded on a Phillips APD 15 system using copper Kα radiation (0.16 nm) over an angular range from $2\Theta = 4°$ to $2\Theta = 38°$ in 0.05° steps. These conditions encompassed all the diffraction peaks from crystalline starch (Bear & French, 1941; Hizukuri & Nikuni, 1957). The pattern was analysed to give the percentage crystallinity from the ratio of the areas of the sharp 'crystalline' peaks and the broad 'amorphous' band (Fig. 2 and Clark, 1994; Komita & Nara, 1983). All patterns were re-integrated using slightly different parameters for baseline and peak onset, and the variance in calculated crystallinity (± 0.1%) was shown to be less than the variation in measurement (±0.5%).

RESULTS AND DISCUSSION

Figure 2 shows the X-ray powder diffraction patterns for starch, egg white and xanthan gum at *c.* 10 g water/100 g dry solid. Both xanthan and egg white show the broad scattering halo expected for amorphous polymers (Clark, 1994). The pattern for starch, however, clearly shows a combination of a broad halo with some sharper peaks overlaid. For the pure starch, the measured crystallinity by this method was shown to increase linearly over the moisture range 3–30 g water/100 g dry starch (Fig. 3). Above this range the crystallinity levels off at *c.* 32%. By performing a linear regression on this data the relationship between the moisture content of starch (M_s (g water/100 g dry solid)) and its crystallinity (C) was found to be:

$$C = 11.8 + 0.52 \times M_s \tag{4}$$

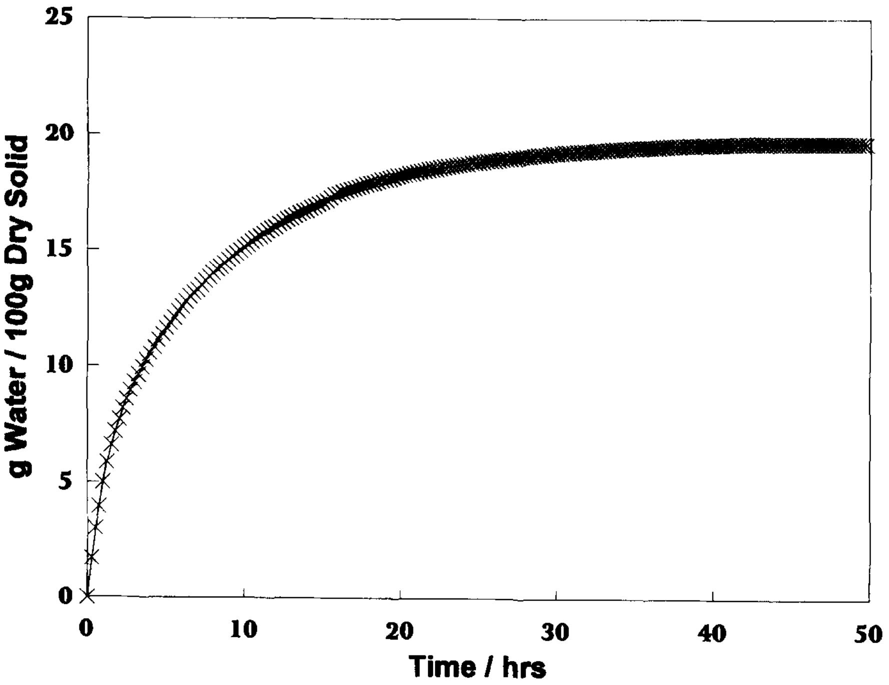

Fig. 1. Moisture content (dry weight basis) of a typical egg white sample (1 g) stored under an 65% relative humidity (RH) atmosphere plotted against time.

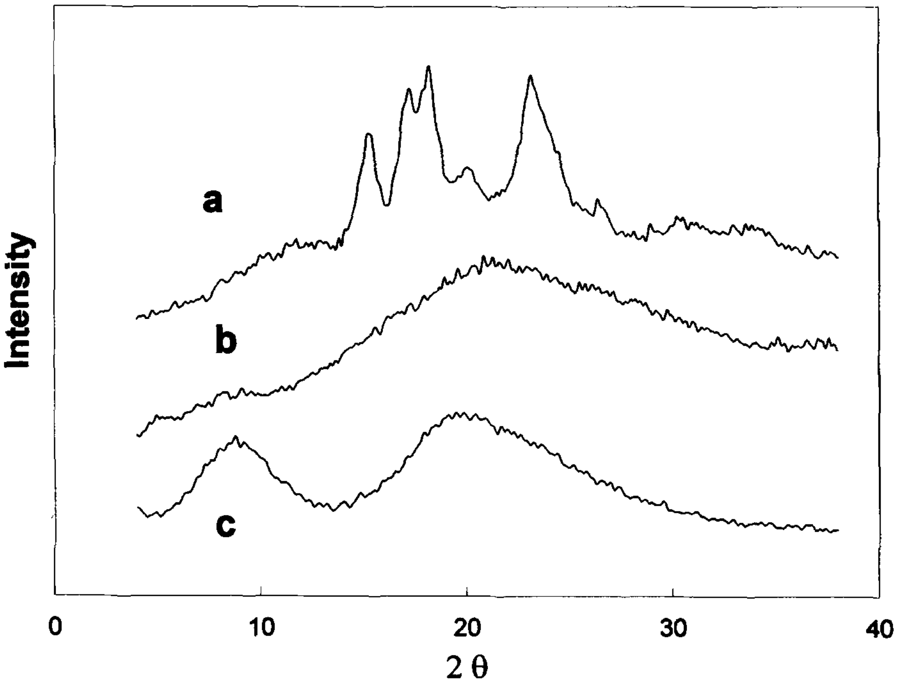

Fig. 2. X-ray powder diffraction patterns for: (a) unmodified wheat starch; (b) xanthan gum; and (c) egg white powder, all samples with a moisture content of 20 g water/100 g dry solid.

Since the other non-crystalline polymer present will only contribute to the amorphous component of the diffraction pattern then the moisture content of the starch in the mixture (M_{sm}) can be obtained from:

$$M_{sm} = \frac{1}{0.52}\left(\left(\frac{W_s + W_p}{W_s}\right)C - 11.8\right) \qquad (5)$$

where W_s is the dry weight of starch and W_p is the dry weight of the other polymer.

The sorption isotherms for egg white, starch and a 50/50 (dry weight) mixture of the two are shown in Fig. 4. Each point is an average of three samples equilibrated separately, and the moisture content and water activity for each of these samples was measured three times. The

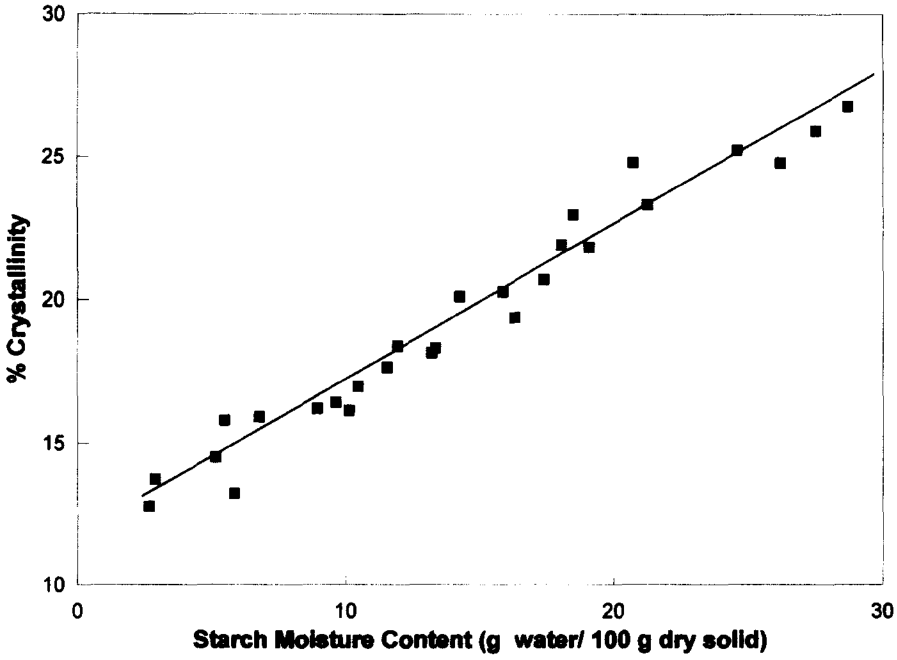

Fig. 3. Measured X-ray crystallinity of wheat starch as a function of moisture content. The line is obtained by linear regression.

base data were fitted to the G.A.B. equation using a non-linear optimisation programme. Good fits to the data ($r^2 \geqslant 0.98$) were obtained. This is displayed as the solid line in Fig. 4. On first examination, the sorption isotherm of the mixture appears to lie in the middle of the curves for the two components, however, on closer examination, the curve for the mixture can be seen to be nearer to the starch (lower) curve than the egg white (higher) curve.

This suggests that less water is associated with the biopolymers in the mixture than would be predicted from the isotherms of the individual biopolymers. This offset can be quantified by the parameter ξ defined by equation (3). Assuming ξ(starch) = ξ(egg white) then a value of 0.79 was obtained using the least squares method of Kaminski & Al-Bezweni, 1994. The calculation was carried out on data over the water activity range 0.2–0.9.

The water content associated with the starch predicted by this approach can be compared with that

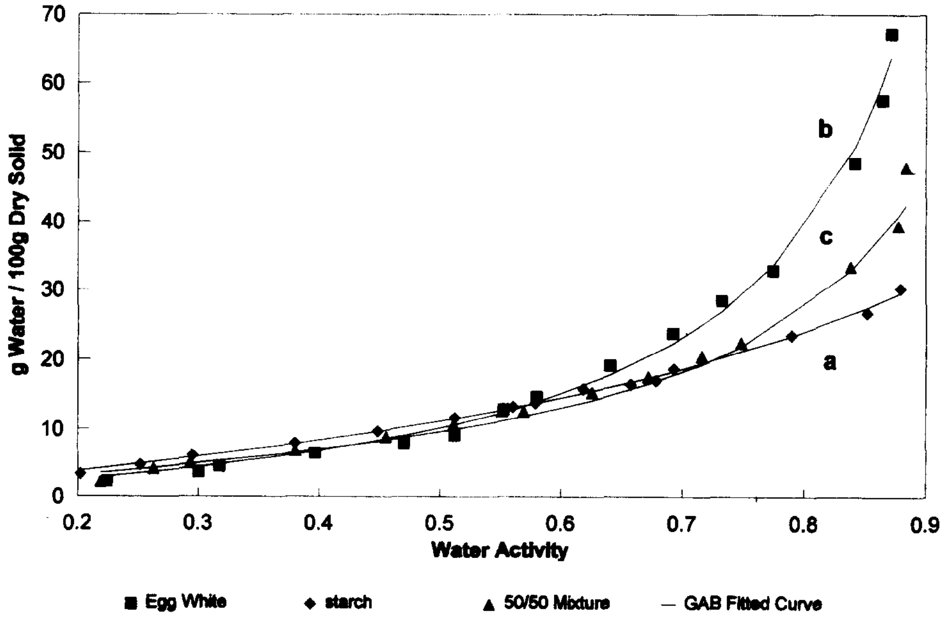

Fig. 4. Water sorption isotherm for: (◆) wheat starch; (■) egg white powder; and (▲) a 50/50 w/w mixture of the two. The solid lines represent the fitted G.A.B. curve for each sample.

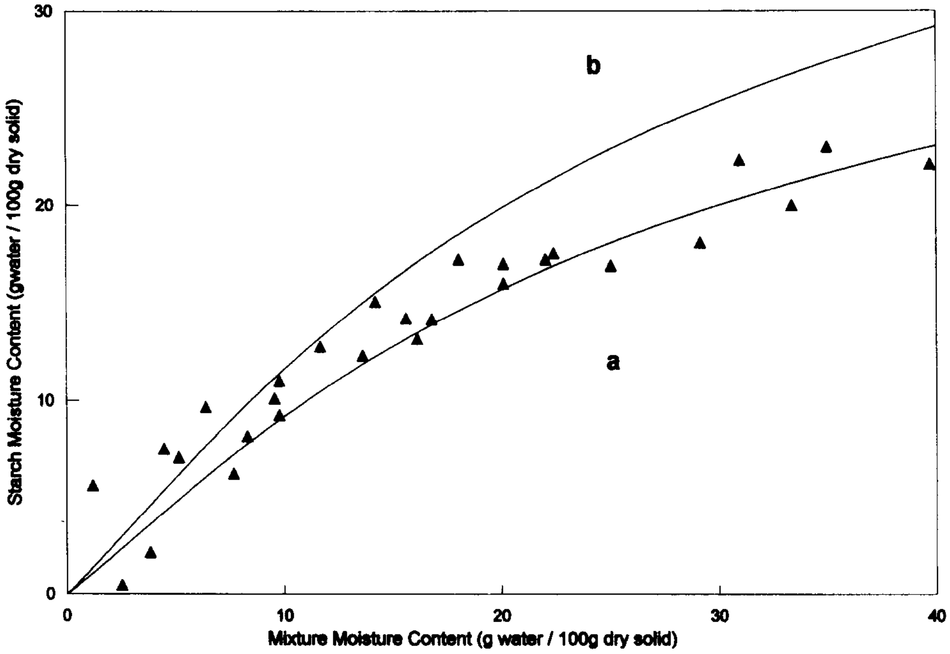

Fig. 5. Moisture content of the starch in a 50/50 w/w mixture of egg white/wheat starch as a function of the moisture content of the overall mixture. The solid lines are obtained from the G.A.B. equation which was fitted to the sorption isotherm measured for starch. For curve 'a' the moisture content is taken as 0.79 × M_s whereas for curve 'b' it is taken as M_s. The data points represent the moisture content of the starch within the mixture calculated from X-ray measurements. For further details see text.

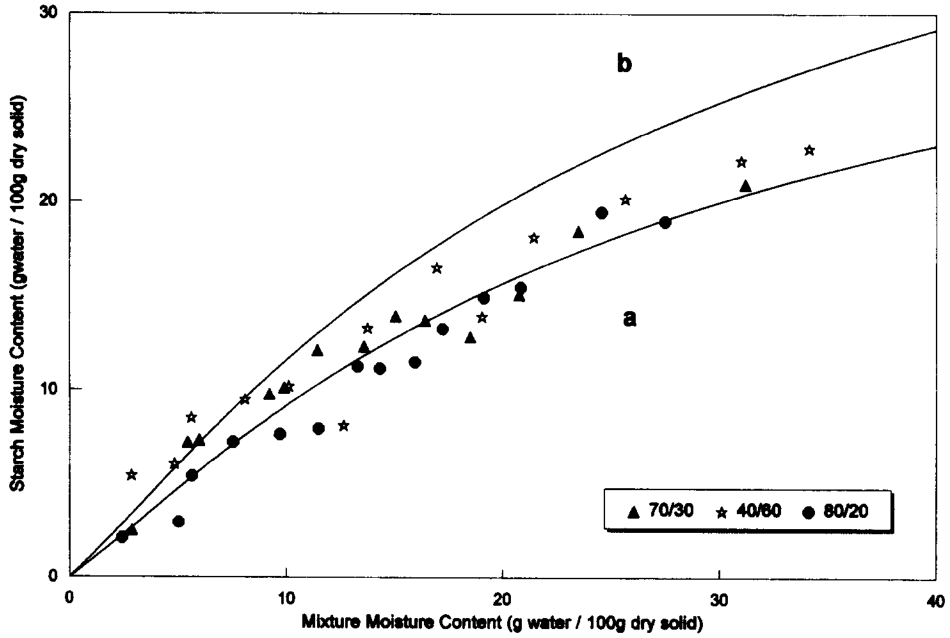

Fig. 6. Moisture content of the starch in mixture of egg white/wheat starch mixtures of differing ratios as a function of the moisture content of the overall mixture. Starch/egg white ratios: (☆) 40:60; (▲) 70:30; and (●) 80:20. For other details see legend to Fig. 5.

calculated from the measured crystallinities. In Fig. 5, curve 'b' is obtained assuming that the starch acquires a moisture content (M_S) at the water activity of the mixture predicted by the G.A.B. isotherm measured for starch alone. Curve 'a' assumes that the starch acquires a moisture content of ξM_s where $\xi = 0.79$.

It can be seen that at moisture contents lower than 12 g water/100 g dry solid there is little difference between the two curves, but at higher moisture levels the moisture content obtained from the crystallinities is clearly nearer to curve 'a' ($\xi = 0.79$).

Figure 6 compares the predicted and measured starch moisture contents in the mixture for different ratios of starch and egg white. It can be seen that at all the ratios

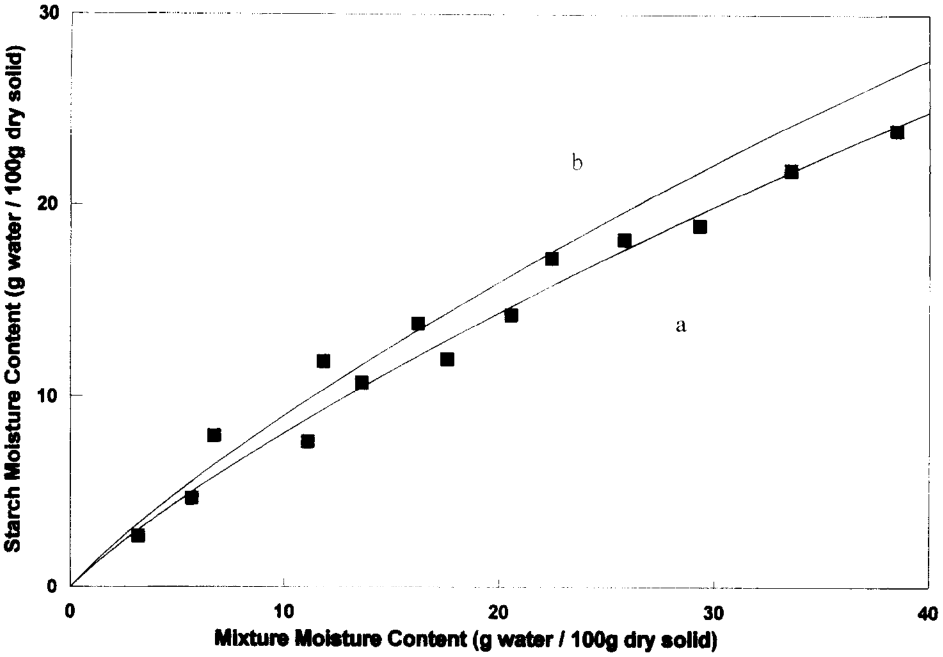

Fig. 7. Moisture content of starch in a 50/50 w/w mixture with xanthan gum as a function of the moisture content of the overall mixture. For curve 'a', the moisture content is taken as 0.9 × M_S. For other details see legend to Fig. 5.

investigated, the data are better explained assuming an interaction parameter ξ of 0.79 than assuming ideal behaviour ($\xi = 1$).

For mixtures of native wheat starch with xanthan gum, the measured sorption isotherm of the mixture is similar to the predicted isotherm from a weight average of the two component isotherms (data not shown). Following the same procedure as for egg white/starch mixtures, an interaction parameter $\xi = 0.9$ was calculated. As can be seen in Fig. 7, the predicted moisture content of starch within the xanthan/starch mixtures shows a good correlation with the measured values; the prediction based on the measured isotherm being slightly better than that based on the weight average hypothesis ($\xi = 1$).

The experimental approach used here suggests that in the mixed system the ratio of the amount of water taken up by the two biopolymers can be predicted from the sorption isotherms of the separate materials. Thus, at high water activities substantially more water is associated with the egg white than the starch. However, both biopolymers take up less water than would be expected from the individual sorption isotherms.

The exact nature of the interaction which gives rise to this non-ideal behaviour is not yet clear. Possibilities include hydrogen bonding between the two biopolymers or interpenetration of the biopolymers distorting the sorption behaviour of the sample. However, it is more probable that the 'interaction' really represents the non-equilibrium state of the samples due to restricted water diffusion within the sample (Slade & Levine, 1991).

CONCLUSION

From the sorption isotherm measurements of binary mixtures and the two components, it is possible to predict the water partitioning between the two components as a function of the moisture level. The moisture level of the starch in a binary mixture can be measured by X-ray powder diffraction. The predictions match the experimental data if an interaction parameter is taken into consideration. However, it is not yet possible to predict the water partitioning within a sample from a consideration of the sorption isotherms of the pure components alone.

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