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Modulating rheo-kinetics of native starch films towards improved wet-strength

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ABSTRACT

Starch directly functionalised in the plant by modulation of its biosynthesis by mutagenesis and transgene technology was exploited for its extended functionality beyond the normal variation. In this study we investigated the rheological and mechanical properties of films from such structurally highly different starch types derived from potato and cereal sources of normal and mutant and transgenic backgrounds. A new improved technique was developed to permit the dynamic mechanical analysis of films in the presence of water. It was found that the amylose content was decisive for the mechanical properties of the films – an increase in the amylose content resulted in both a higher stress and strain at break. Interestingly, there was no correlation between the speed of hydration and mechanical water resistance of the films. Generally, the films were clear and transparent, even after wetting. Transgenic potato starch with a low content of phosphate displayed an extraordinary combination of high robustness, transparency, mechanical strength and extensibility even in a wet condition. The combination of optimal phosphate and amylose concentrations in this sample probably favoured hydration and amorphisation without compromising the inter-chain interactions of the polysaccharide network.

1. Introduction

Large amounts of plastic materials are used for disposable materials, such as food packages. For environmental reasons, there is a desire to replace petroleum-based plastic materials with materials based on renewable resources. For example, in 2008, containers and packaging amounted to 31% (w/w) of the municipal solid waste in the USA (United States Environmental Protection Agency, 2009). These large quantities result in large costs for the municipal waste management. Besides, plastic disposables that are not biodegradable but end up outside the waste management system cause tremendous harm to e.g. marine wild life (Derraik, 2002). Materials from plant-derived polysaccharides are not only materials originating from renewable resources, but also materials which are biodegradable, which can possibly facilitate the waste management and reduce littering. Starch is an important plant-derived polysaccharide, which is attractive since it is pure, abundant and inexpensive, has good film-forming properties and provides an excellent gas barrier (Miller & Krochta, 1997; Rindlav-Westling, Stading, Hermansson, & Gatenholm, 1998; Stading, Rindlav-Westling, & Gatenholm, 2001).

Typically, starch is composed of the two distinct polysaccharides, amylose and amylopectin, where amylose is a linear chain of α-1,4-glucans with only occasional α-1,6 branches, while amylopectin is a much larger polyglucan with more frequent branch points (approx. 5%) stabilising double helical segments generating lamellar, semi-crystalline granules in the plant (Damager et al., 2010). Most starches are also phosphorylated in the amylopectin fraction either at C-3 (approx. 30%) or at C-6 (approx. 70%) of the glucosyl unit (Bay-Smidt, Wischmann, Olsen, & Nielsen, 1994; Hizukuri, Tabata, & Nikuni, 1970). Potato starch has a relatively high content of phosphate esters resulting in approximately 0.5% (one of 200–300 glucose units) being phosphorylated, whereas cereal endosperm starches contain much less covalently linked phosphate (<0.01%) (Blennow, Bay-Smidt, Wischmann, Olsen, & Møller, 1998; Tabata, Nagata, & Hizukuri, 1975).

In a materials manufacturing context, starch has the disadvantage of being highly water-sensitive – at least compared to synthetic plastics. Such drawbacks limit the use of raw starch in food packaging applications. Chemical modification can solve

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Table 1
Starch samples investigated with different methods (+). Amylose and phosphate concentrations are indicated.

Origin	Dynamic swelling/dissolution	Mechanical properties	Photo	Amylose (%)	Phosphate (nmol G6P/mg starch)
Normal maize	+	+	+	28[a]	0.1[a]
Waxy maize			+	0.0	0.1
Normal wheat	+			28	0.0
Normal wheat	+			32[a]	0.0[a]
Deglutenised wheat	+			19	0.4
Normal pea		+	+	39[a]	0.5[a]
Normal rice	+			12	0.5
High amylose barley (HAB)	+			32	0.2
Barley	+			28	0.1
Normal potato	+		+	17[a]	23[a]
Potato low phosphate, transgenic (PLP)	+	+	+	23[b]	2.2[b]
Potato high amylose, high phosphate, transgenic (PHAP)	+	+	+	37[b]	54[b]
Amylose	+	+		100	0.0

[a] Hansen, Blennow, Pedersen, Nørgaard, and Engelsen (2008).
[b] Wickramasinghea, Blennow, and Noda (2009).

some of these problems (Miller & Krochta, 1997). Interestingly, the content of phosphate in a starch system has a tremendous effect on its hydration, swelling, rheology and crystalline packing (Kozlov, Blennow, Krivandin, & Yuryev, 2007; Viksø-Nielsen, Blennow, Kristensen, Jensen, & Møller, 2001; Wiesenborn, Orr, Casper, & Tacke, 1994). Especially, phosphorylated starches display a high viscosity and clear transparent gels (Viksø-Nielsen et al., 2001; Wiesenborn et al., 1994). The mechanism behind these phenomena include solubilisation of crystalline segments of the starch (Hejazi et al., 2008), and recent molecular models (Hansen et al., 2008) suggest that a phosphate-induced helix-breaking mechanism leads to local plasticisation of the starch (Blennow & Engelsen, 2010). Amylose is also extremely important for starch paste and gel formation. At some chain lengths, amylose tends to crystallise and retrograde (Miles, Morris, Orford, & Ring, 1985), but amylose chains longer than approximately 1000 glucose units form a gel consisting of cross-linked amylose chains (Clark, Gidley, Richardson, & Ross-Murphy, 1989; Gidley & Bulpin, 1989).

Using mutants and transgene technology, starch structures can be modulated directly in the crop to extend their functionality (e.g. Blennow et al., 2005; Kozlov et al., 2007; Viksø-Nielsen et al., 2001). Complex and extreme rheological behaviour appears when phosphate and amylose are simultaneously engineered in the starch (Blennow et al., 2005), changing the phosphate and amylose contents in starch provides a general means of modulating the starch functionality in general eliminating or decreasing the need for subsequent chemical modification. Of special interest is the potential for utilising these modulations to develop new materials with enhanced functionality, for example for advanced packaging applications. Sitohy and Ramadan (2001) suggest, for example, that phosphorylation may be a useful tool to control the biodegradability of packaging materials.

In this study, structurally highly different starch types derived from normal starch crops, transgenically modified potato and mutant maize, were selected and used to prepare films and the rheological and mechanical properties of these were investigated. We introduce a new improved rheological technique using dynamic mechanical analysis (DMA) to make it possible to study the kinetics of the hydration of film segments. Transgenically modified potato starches displayed interesting structural combinations and were shown to exhibit a unique combination of high wet-strength and clarity, with a potential for the generation of new types of bulk bio-based materials.

2. Materials and methods

2.1. Starches

Potato and maize starch were obtained from Cerestar-AKV I/S, Cargill (http://www.cerestar.com). Rice starch was provided by KMC, Denmark (http://www.kmc.dk). The Pajbjerg Foundation, Svalöf Weibull AB and Bente Wischmann DTU (Technical University of Denmark) kindly provided wheat and barley starch. Transgenic potato tubers were generated as described (Blennow et al., 2005; Kozlov et al., 2007) and starch from transgenic potato tubers was prepared according to Blennow et al. (1998). Amylose and starch phosphate concentrations in the starch samples were analysed as described by Kozlov et al. (2007), and can be seen in Table 1.

2.2. Preparation of films

Homogeneous starch pastes from native starches were generated using a Brabender Amylograph (Brabender, Duisburg, Germany). 12 g of dry native starch was placed in the mixing bowl and deionised water was added up to a total of 400 g. The sample was mixed in the amylograph and the temperature was ramped from 20 to 95 °C at 1.5 °C/min. When a temperature of 95 °C was reached, mixing was continued for 30 min, after which water was added to compensate for evaporated water. At this point, glycerol (3.6 g glycerol generating 30% (w/w) glycerol/dry starch) was added to plasticise some samples. The mixture was vigorously stirred by hand with a spatula and then mixed in the amylograph for another 5 min at 95 °C. Portions of approximately 25 g of solution were cast in Petri dishes with a diameter of 90 mm using disposable syringes. The solution was allowed to dry to form a film at a constant temperature of 23 °C and a relative humidity (RH) of 50%. After drying, the films were peeled from the Petri dishes and cut into test pieces.

Amylose films were prepared by mixing 2.5% (w/w) amylose type III from potato (Sigma Chemical Co., St. Louise, MO) with deionised water. After degassing, the amylose water mixture was heated in a pressure chamber to 135 °C where it was held for 30 min, and then allowed to cool at room temperature to <100 °C. During heating and cooling, the mixture was continuously stirred with a magnetic stirrer. For films with added plasticisers, glycerol (>99.5% purity, BDH Laboratory Supplies, Poole, England) was then added, followed by thorough mixing. The amount of glycerol plasticiser was 30% (w/w) by dry weight of amylose. Portions of approximately 20 g of solution

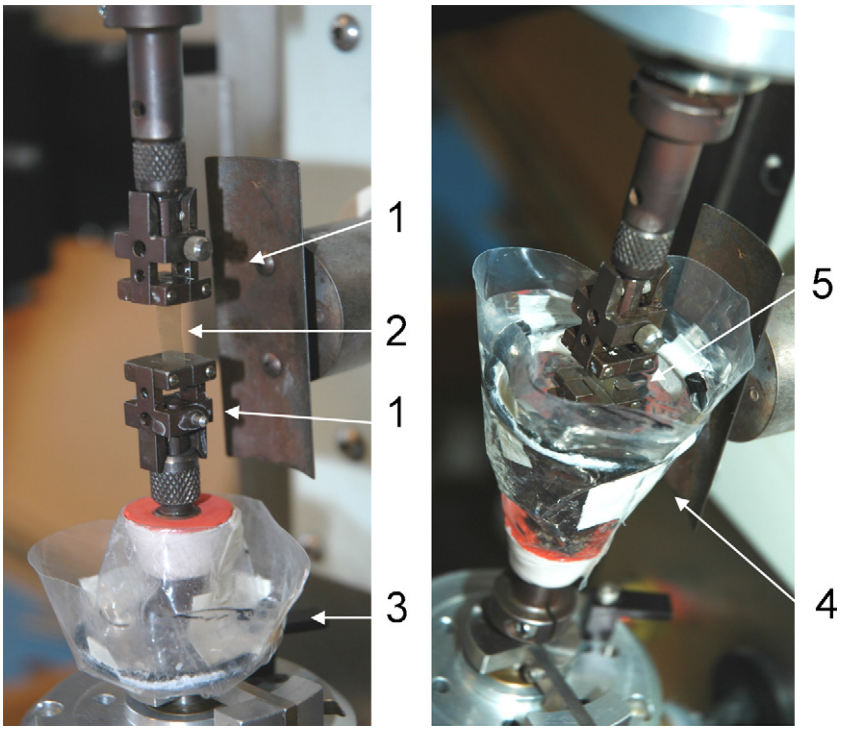

Fig. 1. Film clamps (1) hold a sample film (2). The water-tight container (3) was lowered to enable the sample to be clamped in position. Water was subsequently poured into the raised water-tight container (4). The sample film (5) was then submerged in water.

were cast in Petri dishes with a diameter of 90 mm using disposable syringes. The solution was allowed to dry to form a film at a constant temperature of 23 °C and RH of 50%. After drying, the films were peeled from the Petri dishes and cut into test pieces.

2.3. Rheological kinetics

To permit a dynamic rheological analysis (DMA) of wetting/swelling/dissolution kinetics at constant saturated equilibrium humidity, a new method developed from Edrud, Petersson, and Stading (2003) was used. The instrument, a Rheometrics Solids Analyser, RSA-II (Rheometric Scientific, Piscataway, NJ, USA) used a vertical oscillating movement to measure the storage modulus (E') and loss modulus (E'') of the sample. The phase angle, i.e. the phase difference between E' and E'' in the oscillatory test, was calculated and recorded by the instrument software. To be able to analyse the kinetics of the samples, it had to be possible to completely submerge the sample in water during the measurements in a manner which did not affect the measurements in other ways. For this purpose, a small flexible water-tight plastic container was designed. The container (Fig. 1) could be lowered to enable the sample to be clamped in position. The container could then be raised so that water could later be added to completely cover the sample (Fig. 1).

The container was as light as possible to reduce the load on the instrument transducer. The design was optimised to avoid contact with the uppermost geometry potentially leading to erroneous measurements. A maximum oscillation frequency of 0.5 Hz was used. Film clamps (Fig. 1) were used to grip the sample. When the sample was in position, the clamp separation was adjusted to give zero load on the film at the start of the measurement. The measurement was then started and the instrument was programmed to apply a small minimum force to keep the sample stretched in tension. Superposed on the stretching load, the instrument applied an oscillating movement. The dynamic force had to be lower than the stretching force to avoid sample buckling. If the sample became softer, the instrument would adjust the clamp position to maintain sufficient stretching force/signal ratio for the instrument. These functions were essential to ensure that the films swelling/dissolution could be measured. When the measurement gave stable values, water was poured into the container, submerging the sample fully in approximately 1–3 s. Since the samples were very hydrophilic they quickly lost their rigidity. The instrument had to then quickly separate the film clamps and increase the amplitude to maintain the forces. The original clamping distance was ca. 25 mm and the greatest increase in sample length was 4.5 mm.

Fig. 2 shows an example of a graph of the elasticity modulus E' as a function of time during dissolution.

A non-linear exponential curve was fitted to the data for the slope of E':

$$y = A \cdot e^{(-n \cdot t)} \quad (1)$$

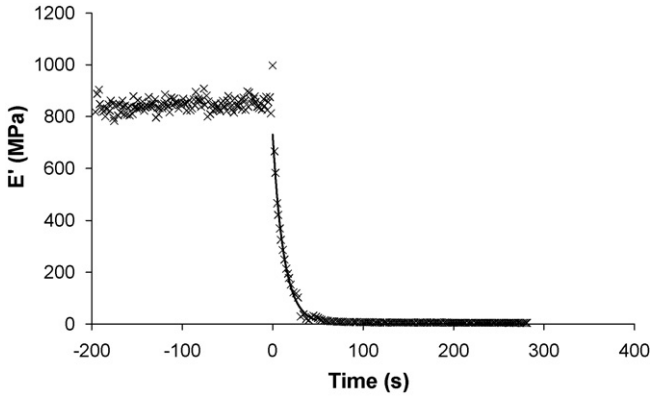

Fig. 2. Typical time course of E' developed as a function of hydration of a starch film. At time zero, water is added and an abrupt drop in E' is observed. The solid line is the fitted non-linear exponential curve.

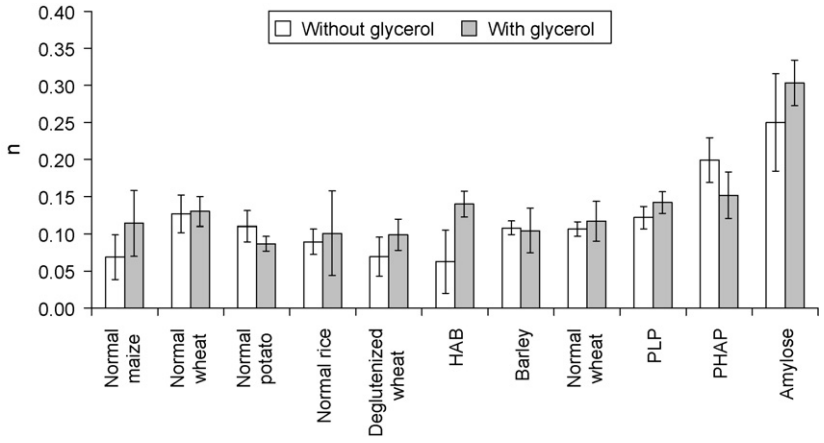

Fig. 3. Dynamic measurement of swelling/dissolution speed for films with and without plasticiser. The n parameter has been extracted from the fit of the data to the exponential $y = A \cdot e^{(-n \cdot t)}$. The error bars show the standard deviation based on three replicates.

From this fit, parameters A and n were obtained, where n represents the reaction constant for the loss of rigidity. The A parameter was not used further in this method. All the samples were tested in triplicate. All samples were conditioned at 23 °C and 50% RH before testing. Samples were approximately 12 mm long and 6 mm wide. Before each measurement, the sample thickness was measured using a digital micrometer (model IDC-112CB, Mitutoyo Corp., Tokyo, Japan). The sample thickness varied between 0.04 and 0.15 mm.

2.4. Mechanical properties

All the samples were conditioned at 23 °C and 50% RH for at least 48 h before testing. For wet testing, the strips were immersed in deionised water for 3 min, wiped with a paper tissue to remove excess water, and thereafter mounted in the testing machine. After swelling, the thickness of the samples varied between 0.15 and 0.29 mm. Other samples were tested dry. Mechanical properties were measured using an Instron 5542 single-column universal materials testing machine (Instron, Norwood, MA, USA). Mechanical testing was carried out in accordance with the ASTM D882-91 standard, using 2 mm wide samples, a crosshead speed of 0.24 mm/s, and an original clamping distance of 23–47 mm; 10 samples were evaluated for each type of starch, and for each wet or dry test. Rubber padded clamps were used. The instrument recorded force and strain. Young's modulus, stress at break, and strain at break were calculated from the original clamping distance and the cross-sectional area. The waxy maize and normal potato starches displayed no measurable wet-strength and could not be analysed. The wet mechanical properties of normal maize, normal pea, PLP, PHAP and amylose were characterised.

2.5. Photography

Film transparency was documented by digital photography recorded with a Nikon D70 camera (Nikon Nordic AB, Solna, Sweden). The film samples were placed on a laminated sheet of paper with a printed text. This background was chosen to provide contrast in the pictures, to emphasise the turbidity of the samples. Normal potato and PLP were also photographed in free vertical position, to demonstrate the curling of the samples. One representative sample from each of the native starch films (all starches except for the pure amylose) and from the amylose film was chosen for digital documentation. Sample films were photographed before and after being immersed in deionised water for 3 min at 25 °C.

3. Results

3.1. Wetting/swelling/dissolution kinetics

To quantify the kinetics of dissolution/swelling the parameter n in Eq. (1) was used as a measure of the swelling. A higher n-value indicates faster swelling. All the samples showed a similar swelling behaviour (Fig. 3). The method was satisfactory for most of the sam-

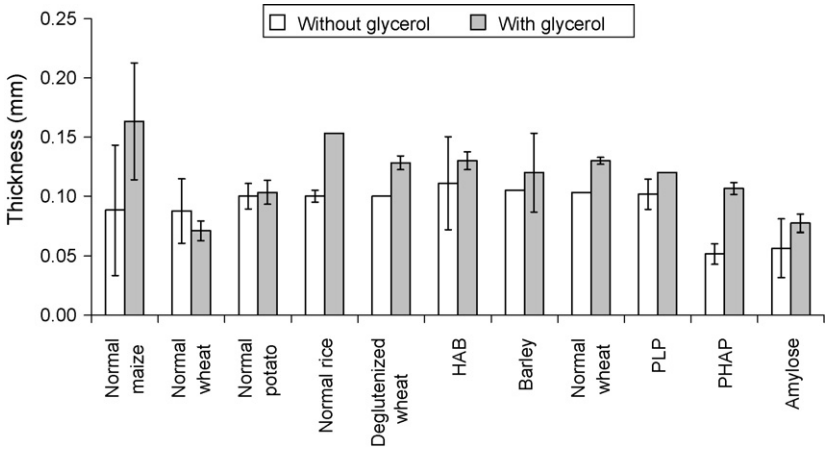

Fig. 4. Sample thickness for films with and without plasticiser. The error bars show the standard deviation based on three replicates.

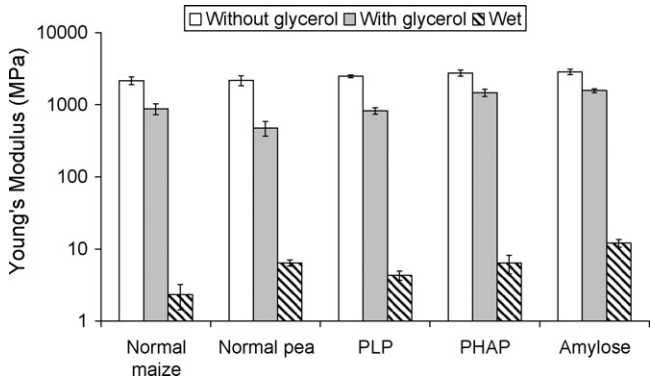

Fig. 5. Young's modulus of starch films without glycerol (white) and with glycerol (grey), and of wet films without glycerol (striped). The error bars show the standard deviation based on nine replicates.

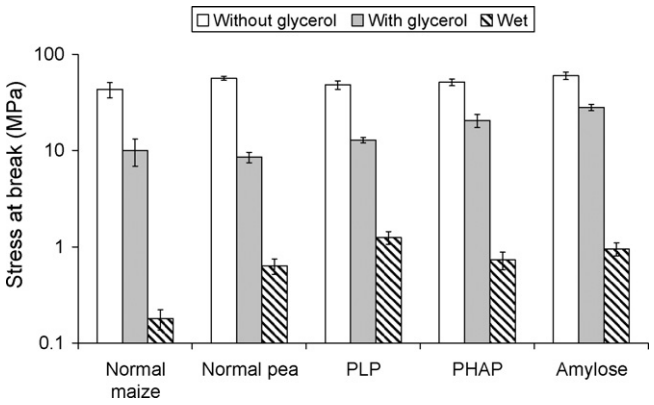

Fig. 6. Stress at break of starch films without glycerol (white) and with glycerol (grey), and of wet films without glycerol (striped). The error bars show the standard deviation based on nine replicates.

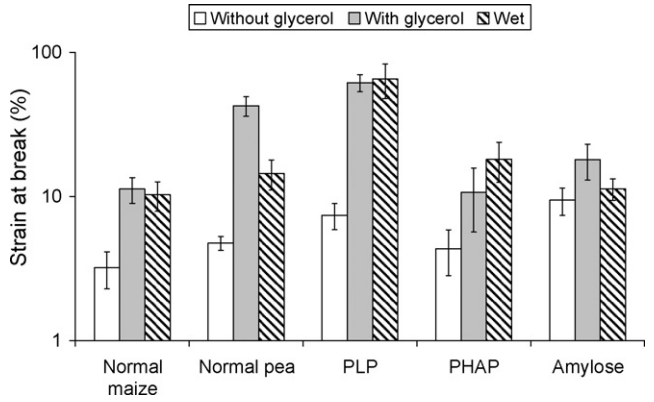

Fig. 7. Strain at break of starch films without glycerol (white) and with glycerol (grey), and of wet films without glycerol (striped). The error bars show the standard deviation based on nine replicates.

ples. However, the waxy maize dissolved when being immersed, and could not be studied further due to the rapid dissolution.

Since sample thickness could have an effect on the swelling speed, the thickness was determined (Fig. 4). Most of the films showed approximately the same thickness, although the plasticised samples tended to be slightly thicker. Comparing the sample thickness and the *n*-values indicated a possible correlation for samples with extreme thicknesses, where thicker films showed a slower swelling – for example, normal maize and normal rice were thicker than average and also showed lower *n*-values. No overall significant correlation between thickness and *n*-value was however found.

Some films that showed good wet resistance in the following mechanical measurements nevertheless showed a very high *n*-value, as for example the amylose films, demonstrating the genuine swelling and not molecular dissolution. These films seemed to swell quickly but to only a limited extent. Before submersion in water, a low phase angle (the phase difference between E' and E'') was observed. When water was added, the phase angle initially rose (indicating a more viscous material), passing through the transition from glassy to rubbery, but then dropped to a low level again, indicating the formation of solid (crystalline) material. The unplasticised films tended to stabilise at values similar to those before submersion. For the plasticised films, the initial phase angle was similar. After submersion, the plasticised films stabilised more slowly and an indication of lower values could be seen. This in turn indicated that the unplasticised materials were more solid. The plasticised samples also tended to show higher *n*-values, although no significant effect could be seen in the measurements (Fig. 3). The tendency matches the observation that plasticised starch films absorbed moisture more quickly than unplasticised films reported by Mali, Sakanaka, Yamashita, and Grossman (2005). All the samples had *n*-values in the range of 0.07–0.20, except for the amylose films which showed a very high *n*-value of ca. 0.3; indicating a very rapid swelling. In general, films with higher amylose content displayed faster swelling.

3.2. Film mechanical properties

3.2.1. Dry film characteristics

Due to the well documented plasticising effect of glycerol (e.g. Forssell, Mikkilä, Moates, & Parker, 1997), it was expected that the Young's modulus and the stress at break would decrease and the strain at break would increase with increasing glycerol content. This effect was clearly demonstrated in our data (Figs. 5–7). For the unplasticised films, both stress at break and strain at break increased with increasing amylose content (Fig. 8), in agreement with previous data (Lourdin, Della Valle, & Colonna, 1995). However, an interesting exception was the transgenic PLP which had a normal amylose content (23%) but still showed a higher stress at break and strain at break than for example normal maize with a similar amylose content (28%). In the PLP starch, it can be speculated that the presence of starch phosphate acted as an internal plasticiser, even though the phosphate content was low (2.2 nmol G6P/mg starch) and that this had a positive influence on the strain at break. However, plasticisation generally leads to a decrease in the stress at break, whereas the PLP showed no

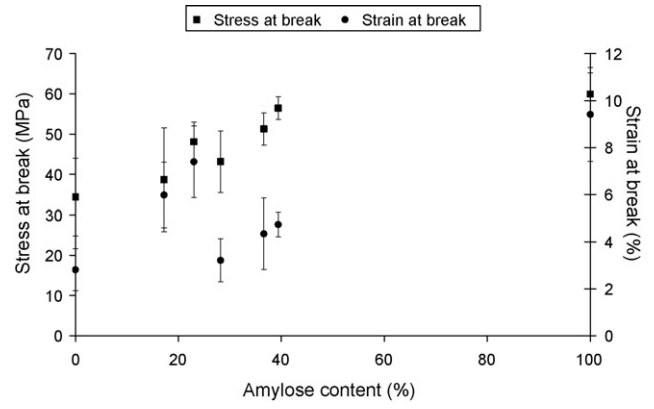

Fig. 8. Stress at break (squares) and strain at break (circles) as a function of amylose content of dry films from waxy maize (0%), normal potato (17%), PLP (23%), normal maize (28%), PHAP (37%), normal pea (39%) and amylose (100%) without glycerol. The error bars show the standard deviation based on nine replicates.

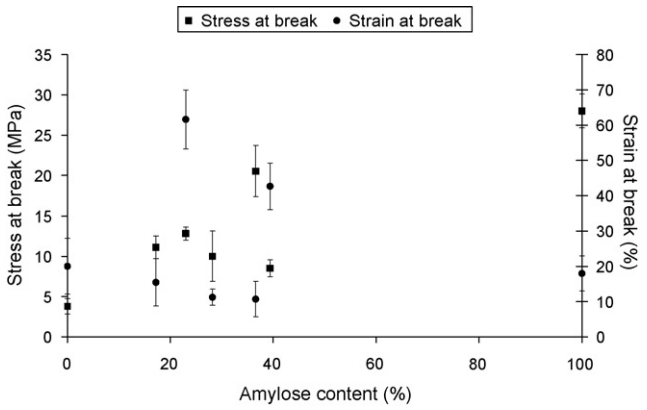

Fig. 9. Stress at break (squares) and strain at break (circles) as a function of amylose content of dry films from waxy maize (0%), normal potato (17%), PLP (23%), normal maize (28%), PHAP (37%), normal pea (39%) and amylose (100%) with glycerol. The error bars show the standard deviation based on nine replicates.

such behaviour. Hence, another factor is probably contributing to the mechanical properties of PLP; Rindlav-Westling, Stading, and Gatenholm (2002) argue in favour of co-crystallisation between amylose and amylopectin in films with an amylose content of approximately 25%. It is possible that the network in the PLP films behaves in a similar way, and that this is responsible for the combination of high stress and strain at break values, and that such interactions are favoured by the presence of low concentrations of starch phosphate. In contrast to the increased stress at break and strain at break of PLP, the further increase in phosphate content characteristic of PHAP (54 nmol G6P/mg starch) had no apparent effect on the mechanical properties of unplasticised films. Films prepared with this starch type, having a higher amylose content (37%), exhibited a high stress and strain at break, which could possibly overrule the effect of the phosphate, apparent only at lower amylose concentrations.

In contrast to the trend for a higher amylose content to give a higher strain at break in unplasticised films, the strain at break was fairly constant (disregarding amylose content) in films plasticised by glycerol (Fig. 9). This behaviour was also reported by Lourdin et al. (1995). However, in the present investigation, the pea and the PLP film showed a much higher strain at break than for example normal maize or PHAP when plasticised by glycerol. Glycerol-plasticised films prepared from PHAP displayed a higher stress at break than all the other samples prepared from native starches, even normal pea which had an amylose content similar to that of PHAP. However, the phosphate content in PHAP was far higher that that of normal pea. Perhaps a co-crystallised network was also present in PHAP and contributed to the superior strength compared to that of normal pea.

3.2.2. *Wet film characteristics*

The mechanical properties of the films were significantly affected by wetting and hydration. In general, the Young's modulus and the stress at break both decreased, even compared to the glycerol-containing films, and the strain at break was approximately equal to that of the glycerol-containing films. The wet samples showed the same trends with respect to stress and strain at break as did the glycerol-plasticised samples, i.e. both stress at break and strain at break of the films with amylose levels of 30% and above were fairly constant (Fig. 10). The only film that deviated was PLP that had stress at break and strain at break values significantly higher than the other samples. The mechanical properties induced by hydration of this film indicate the presence of some molecular combinations providing unique hydration properties. Moreover, PLP showed a remarkable robustness when wetted, in contrast to normal potato starch which was insensitive to deformation upon wetting.

It was interesting to observe the wet performance of the PLP films. PLP contained a smaller amount of amylose than most of the other starches (similar to the normal potato starch), but the presence of starch phosphate in this sample perhaps made the film more durable during extension compared to the other samples, both when wetted and when plasticised. It is possible that there is an optimal combination of starch phosphate at approximately 2 nmol G6P/mg starch and a fairly low amylose concentration. Starch phosphate increases hydration (Damager et al., 2010) and, since water has a plasticising effect on starch (Forssell et al., 1997), it seems plausible that starch phosphate has an indirect plasticising effect on starch. The moisture content of the native starches (data not shown) was approximately 11% for samples with very low starch phosphate contents, and approximately 20% for samples with a starch phosphate content $\geq$2 nmol G6P/mg. Not even PHAP (54 nmol G6P/mg) displayed a higher moisture content. This indicates that there was a maximum level of hydration, and thereby plasticisation, due to starch phosphate, and that this was reached already at 2 nmol G6P/mg starch. At higher starch phosphate concentrations there is a risk for breakage of molecular junctions between starch molecules followed by dissolution of the starch network. Hence a decrease in the phosphate in potato starch to approximately 2 nmol G6P/mg starch can support hydration and amorphisation without jeopardising interchain interactions. Rindlav-Westling et al. (2002) argue in favour of a network including co-crystallised amylose and amylopectin at an amylose content of 25%. It is possible that optimal amylose and starch phosphate contents of PLP promoted such a network formation, and that such a network is responsible for the wet performance of that sample. Films of waxy maize and normal potato starches were also tested with respect to their wet mechanical properties, but they were too weak to be tested. Waxy maize contained no amylose and normal potato starch had an amylose content lower than most other starches in this study, combined with a relatively high starch phosphate content, and it seemed probable that these structural combinations were decisive for the poor stability of the hydrated films.

Among the samples that were mechanically tested, normal maize was the one that was most affected by wetting, in the sense that its mechanical properties were decreased most (with the exception of waxy maize starch and normal potato starch). We found no indication of any extraordinary *n*-values (Fig. 3) for the normal maize and PLP. Hence, we conclude that the rate of swelling upon hydration was not

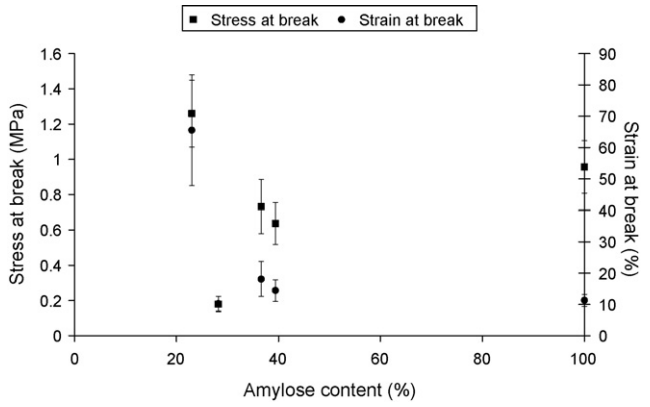

Fig. 10. Stress at break (squares) and strain at break (circles) as a function of amylose content of wet films from PLP (23%), normal maize (28%), PHAP (37%), normal pea (39%) and amylose (100%) without glycerol. The error bars show the standard deviation based on nine replicates.

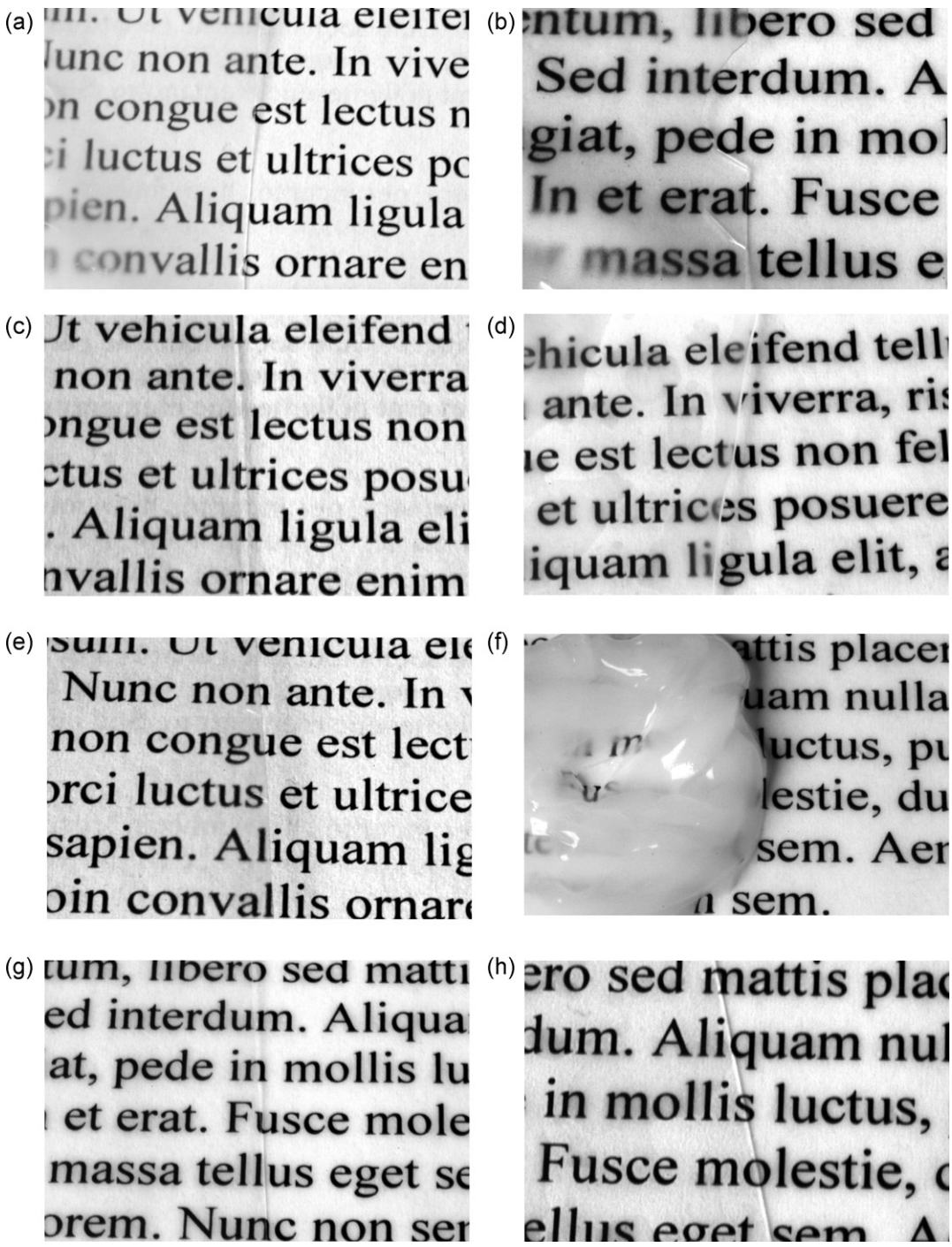

Fig. 11. Photographs where the left halves are covered by starch films of (a) dry normal maize, (b) dry PHAP, (c) dry normal potato, (d) wet normal potato, (e) dry waxy maize, (f) wet waxy maize, (g) dry PLP and (h) wet PLP.

directly related to the mechanical properties of the swollen starch films.

3.3. Visual appearance

Due to phase separation and re-crystallisation, concentrated aqueous starch dispersions are expected to form an opaque gel upon cooling (Leloup, Colonna, & Buleon, 1991). Normal maize and, despite its high content of plasticising starch phosphate, PHAP films showed tendencies to develop turbidity (Fig. 11a and b). The reason for this effect can be the fairly high amylose content and the long amylopectin chains present in this type of transgenic starch (Blennow et al., 2005). The visual appearances of the samples in our study were not however greatly affected by immersing them in water, except for normal potato starch that became slightly more opaque (Fig. 11c and d) and waxy maize which swelled dramatically and lost its shape (Fig. 11e and f). This behaviour was in agreement with the mechanical data which showed that these two samples did not display any water resistance. Normal maize starch contained very low starch phosphate levels, which can explain the turbidity according to Viksø-Nielsen et al. (2001), since this starch type lacks plasticising starch phosphate groups.

The rest of the films did not develop any turbidity, either before or after wetting. Apparently, only minor phase separation

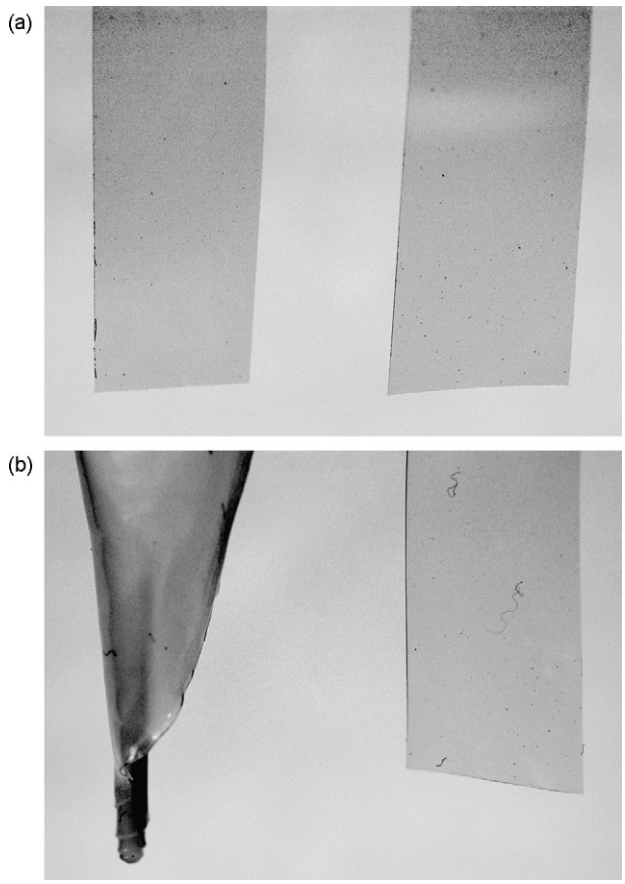

Fig. 12. Starch films prepared from normal potato starch (left) and PLP starch (right) (a) before and (b) after wetting.

takes place in these systems. However, our structural data cannot explain the difference in turbidity between e.g. the maize and the wheat films. Remarkably, the PLP films retained their transparency (Fig. 11g and h) and robustness (Fig. 12) combined with their high mechanical strength upon wetting demonstrated above. Such a behaviour is seemingly unique and supposedly an effect of optimal conditions for starch phosphate to plasticise the hydrated matrix and amylose providing a cross-linked gel network to strengthen the film.

4. Conclusion

This investigation demonstrated that native starch having no post-harvest chemical modification can be used to produce high quality films using starch derived from mutants and transgenic plants. The amylose and starch phosphate content of the starch are extremely important for the mechanical properties as well as for the visual appearance of the starch films under both dry and wet conditions. A minor starch phosphate plasticisation combined with a moderate inter-amylose–amylopectin based network provides an optimal structural combination for robust and transparent films. Further knowledge of the impact of amylose and starch phosphate promotes the design of new improved materials and food ingredients. An improved method allowed defined dynamic swelling/dissolution data to be obtained also for hydrated films.

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