Effect of pulsed electric field, mild thermal pretreatment and calcium on texture changes of potato (*Solanum tuberosum* L.) during subsequent cooking

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Submitted: April 28, 2021

Revision: July 27, 2021

Abstract

The effect of pulsed electric field (PEF) and/or mild thermal processing at 60 °C (T60) on the cooking behaviour of potato tissue in media with different Ca^{2+} concentrations (0-0.5%) was investigated. The rate constant for texture degradation during cooking seemed to decrease after the different pretreatments in the order Untreated > T60 > PEF > PEF-T60, but only a significant effect (α=0.05) could be found in the case of the combination pretreatment in 0.5% Ca^{2+} medium. These texture changes were linked with changes in pectin degree of methylesterification (DM) and Ca^{2+} crosslinking obtained after the pretreatments. The mild thermal pretreatment has little effect on the pectin DM but promotes ionic crosslinking. Both the PEF and combination pretreatment reduce the pectin DM and increase ionic crosslinking significantly. An exponential correlation was found between the texture degradation rate constant and the pectin properties studied.

Keywords: Pulsed electric field; Thermal processing; Potato; Texture; Pectin

1. Introduction

A pulsed electric field (PEF) treatment involves the submission of biological cellular material to short intensive electric pulses, while positioned between two electrodes and immersed in a conductive medium. If the critical electric field strength is exceeded, the treatment results in the electroporation of the cell membranes. In the food industry, PEF technology is currently used to improve diffusion associated processes (e.g. drying and extraction) in plant tissues, to reduce cutting forces of plant tissues (0.7-3 kV/cm), and to inactivate micro-organisms in liquid foods (15-40 kV/cm) (Botero-Uribe et al., 2017; Leong et al., 2014; Puértolas et al., 2012; Toepfl et al., 2006; Vorobiev & Lebovka, 2008). This research paper investigates the effect of PEF technology on the texture evolution of potato tissue during subsequent cooking.

The sensory appreciation of fruit and vegetable tissue based food products is largely determined by texture perception, which is related to the food structure (Wilkinson et al., 2000). The main texture determining factors in low-starch plant tissues are the turgor pressure, maintained by an intact cell membrane, and the integrity of the cell wall, composed of a pectin, hemicellulose and cellulose network. Neighbouring cells are glued together by a middle lamella, which mainly consists of pectin (Christiaens et al., 2016; Gonzalez & Barrett, 2010; Jackman & Stanley, 1995; Waldron et al., 2003).

In general, the texture softens during thermal processing. At temperatures from 60 °C, the semipermeable character of the cell membrane is lost, resulting in a reduced turgor pressure (Gonzalez et al., 2010; Gonzalez & Barrett, 2010). In contrast to cellulose and hemicellulose, the structure of pectin changes during thermal processing, making pectin an important texture determining factor during thermal processing (Ranganathan et al., 2016; Sila et al., 2008; Williams & Besler, 1996).

Pectin consists of three building blocks: linear subdomains of homogalacturonan (HG) and branched subdomains of rhamnogalacturonan I (RG-I) and II (R-II), usually accounting for 60%, 20-35% and 10% of pectin, respectively (Mohnen, 2008). HG is composed of linked galacturonic acid (GalA) residues and can be depolymerised during thermal treatments at temperatures higher than 80 °C by a beta-eliminative reaction. With pectin playing an important role in the cell wall strength and cell adhesion, the depolymerisation of pectin results in a softer texture. However, the structure of HG determines the ease of depolymerisation (Christiaens et al., 2016; Sila et al., 2006).

HG with a low degree of methylesterification (DM) is less susceptible to the beta-elimination reaction (Christiaens et al., 2016). The DM can be lowered by the action of pectin methylesterase (PME), whose activity can be promoted by mild temperatures. Therefore, mild thermal pretreatments (15-60 min at 60-75 °C) are currently used to preserve texture during subsequent cooking (Lemmens et al., 2009; Moens et al., 2020; Sila et al., 2006; Sila et al., 2005; Smout et al., 2005). Also cations may promote PME activity: as plant PME is positively charged and tightly associated with the cell wall by ionic interactions, the cations may compete with PME to bind the negatively charged GalA residues, liberating the enzyme and preventing enzyme entrapment (Christiaens et al., 2016; Nari et al., 1991). The altered membrane integrity after the mild thermal pretreatment may also promote the transport of cations to the cell wall (Ranganathan et al., 2016).

Furthermore, blocks of negatively charged demethylesterified GalA residues (with the pK_a of polyGalA being 3.38) (Kyomugasho et al., 2015) allow the formation of ionic crosslinks between GalA residues and divalent cations such as Ca^{2+}, within and between different HG chains. Ionically crosslinked pectin is less susceptible to depolymerisation and solubilisation at elevated temperatures and the denser pectin network results in a firmer texture (Celus et al., 2018; Christiaens et al., 2016; Sila et al., 2009). Therefore, mild thermal pretreatments are often performed in the presence of Ca^{2+} ions that may promote the formation of such crosslinks (Lemmens et al., 2009; Sila et al., 2005; Smout et al., 2005). Too high concentrations of divalent cations, on the other hand, may also inhibit PME activity (Christiaens et al., 2016).

In the case of starch containing fruits and vegetables, such as potatoes, also the presence of starch affects the texture, especially during thermal processing (Ranganathan et al., 2016). At temperatures from 50 °C, starch absorbs water and starts to swell (Lovegrove et al., 2017). The gelatinised starch inside the cell drives the cells towards a spherical shape and creates a force that separates neighbouring cells. Due to the promotion of cell separation, the presence of starch is associated with a mealy texture (Binner et al., 2000; Laborde & Padella-Zakour, 2003; Ranganathan et al., 2016; Van Dijk, Fischer, Holm, et al., 2002). Pectin in the middle lamella, on the other hand, is responsible for cell adhesion and counteracts cell separation. While starch gelatinisation and cell membrane integrity loss occur at an early stage of cooking, from 50 °C and 60 °C, respectively, the depolymerisation and solubilisation of pectin becomes the rate determining step at the later phase of cooking at temperatures higher than 80 °C. Therefore, the structure and crosslinking of pectin are important factors controlling the cooking behaviour of starch containing tissues (Kaaber et al., 2007; Ormerod et al., 2002; Ross et al., 2011; Van Dijk, Fischer, Beekhuizen, et al., 2002).

The PEF treatment softens the texture of fresh plant tissues: the turgor pressure is lost due to electroporation of the cell membranes. Therefore, PEF technology can be used to reduce the cutting force of fruit and vegetable tissues (Fincan & Dejmek, 2003; Leong et al., 2014; Ranganathan et al., 2016). However, little information is available on the effect of PEF technology on pectin structure and the consequences for texture during subsequent cooking. Recently, it was shown that a PEF pretreatment may help to preserve the texture of carrot vascular tissue during subsequent cooking, and that a PEF pretreatment prior to a mild thermal pretreatment enhances the texture preserving effect of the mild thermal pretreatment in both carrot cortex and vascular tissue. Although a complete understanding of the molecular mechanisms involved may need further investigation, it was shown that PEF enables the demethylesterification of pectin in vascular carrot tissue, especially if combined with a mild thermal pretreatment. The authors hypothesise that the electroporation of cell membranes facilitates the transport of intracellular cations to the cell wall, the site of PME action, promoting PME activity (Moens et al., 2020). However, as carrots have a relatively high pectin content and PME activity (Alonso et al., 2003; Houben et al., 2011; Moens et al., 2020), the effect of PEF on pectin structure, especially the DM, and on texture during subsequent cooking may be less pronounced in other fruit and vegetable tissues containing less pectin, a smaller proportion of HG, and a lower PME activity.

Potato pectin has a relatively high RG-I content, and only consists for 25% of HG. The presence of RG-II in potato cell wall has not been proven yet (Oomen et al., 2003; Ralet et al., 2016; Vincken et al., 2000). The larger amount and the longer pectic side chains of RG-I in potatoes may explain the smaller texture preserving effect of mild thermal pretreatments during subsequent thermal processing (Ranganathan et al., 2016). Moreover, PME extracts from potatoes show a lower activity than extracts from carrots (Moens et al., 2020; Moens, De Laet, et al., 2021). The activity of an extract, on the other hand, may not be representative for the activity *in situ*. Additionally, potatoes have a relatively high starch content (about 60-70% of dry matter) (Kolbe & Stephan-Beckmann, 1997; Moens, De Laet, et al., 2021), which may also affect the texture during thermal processing.

For this reason, this paper investigates the effect of PEF and/or mild thermal processing on potato texture during subsequent cooking in the presence of different levels of Ca^{2+} ions and on associated pectin changes. The cooking behaviour, DM and calcium crosslinking of pectin was studied after submission of potato tissue to no pretreatment, a PEF pretreatment, a mild thermal pretreatment at 60 °C, and a combination pretreatment. Finally, the pectin DM and ionic crosslinking were correlated with the cooking behaviour.

2. Materials and methods

2.1 Plant materials

Potatoes (*Solanum tuberosum* L.) of the variety Fontane were grown and harvested by a local farmer in Belgium (Carolushoeve, Hoegaarden). Potatoes without cracks or bruises were retrieved and stored for maximum two weeks before use in a fridge at 8 °C.

2.2 Pretreatments of potato tissue

The potatoes were submitted to pretreatments similar to the ones in the study of Moens et al. (2020) on carrot tissues. Therefore, the potatoes were submitted to no pretreatment (untreated), a pulsed electric field (PEF) pretreatment, a mild thermal pretreatment of 20 min at 60 °C (T60), and a combination of a PEF and mild thermal (PEF-T60) pretreatment (**Figure 1**). The mild thermal pretreatment was performed in demineralised water (0% Ca^{2+}) or demineralised water containing 0.05% Ca^{2+} (1.8341 g $CaCl_2.2H_2O$/l) or 0.5% Ca^{2+} (18.3411 g $CaCl_2.2H_2O$/l). The tissues that underwent no mild thermal pretreatment (untreated and PEF) were soaked in the same media for 20 min at 4 °C.

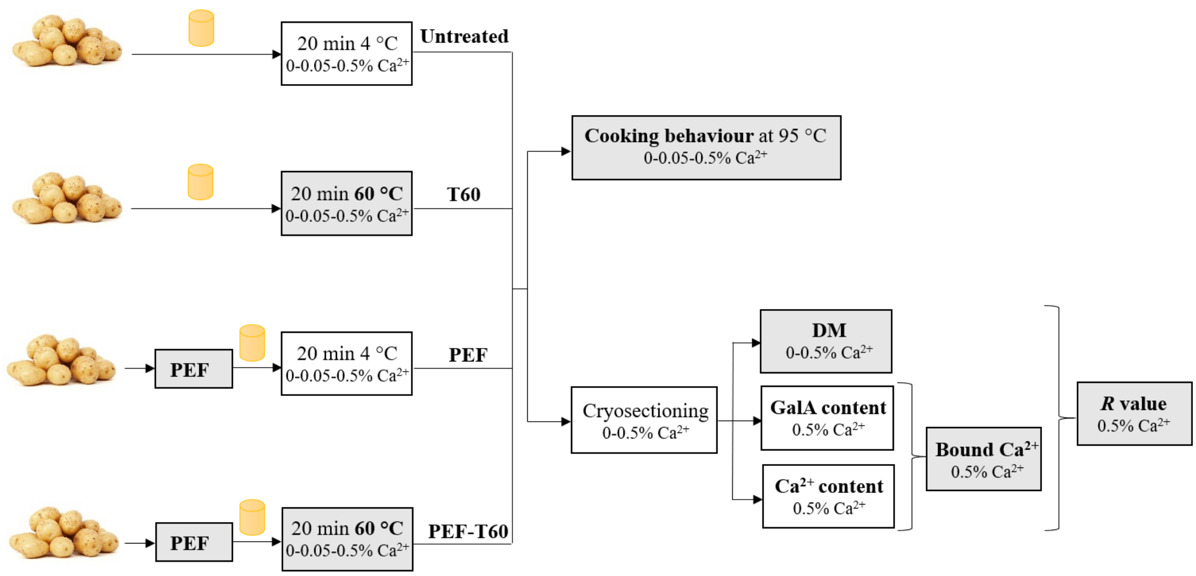

Figure 1 Research strategy to study the effect of pulsed electric field (PEF) and mild thermal processing (T60) on the cooking behaviour of potato tissue, pectin demethylesterification and Ca^{2+}-crosslinking. DM = degree of methylesterification, GalA = galacturonic acid.

2.2.1 Pulsed electric field pretreatment

The PEF treatment protocol was based on the one used by Moens et al. (2020). Potatoes were manually peeled, rinsed in tap water, blotted dry with tissue paper and exposed to electric pulses using the batch Cellcrack III of Elea-DIL (German Institute for Food Technologies, Quackenbrück, Germany). The PEF unit comprised a treatment chamber with stainless steel parallel electrodes (24 x 22.5 x 0.5 cm, w x h x t) with an interelectrode distance of 29.7 cm, insulator thickness of 2 cm, and total volume of 12 l. Five potatoes were positioned randomly in the treatment chamber and immersed completely with standardised water (0.6156 g NaCl/l and 0.0923 g $CaCl_2.H_2O$/l, conductivity 1400 µS/cm at 25 °C) (Ben Ammar et al., 2011; Liu et al., 2017; Willemsen et al., 2017) to obtain a total weight of 4.5 kg (with 1:3 approximate weight ratio of potatoes:medium). The exponential monopolar pulses were characterised by an amplitude of 30 kV, electric field strength of 1.01 kV/cm, pulse energy of 450 J/pulse, frequency of 2 Hz, pulse width of 95 ± 5 µs (TBS 1102B-EDU digital oscilloscope, Tektronix, Köln, Germany), and specific energy input of 100 J/kg per pulse (based on total mass in the treatment chamber, being 4.5 kg). In order to select an appropriate PEF treatment intensity, an approach similar to the one in the study of Moens et al. (2020) was used. The total specific energy input was varied by increasing the number of pulses from 0 to 40 pulses, and the hardness of potato tissue was measured after each treatment intensity and modelled to a fractional conversion model. An amount of pulses (20) and total specific energy input (2.000 kJ/kg) at the plateau value of hardness was chosen as treatment intensity, as this intensity was assumed to result in maximal cell electroporation at the used electric field strength (data included in separate paper under review). The initial medium and potato temperature was approximately 20 °C. The temperature increase of the medium during the PEF treatment was measured to be less than 0.5 °C. Taking into account the total specific energy input and the specific heat capacity of both treatment medium and potato tissue, the estimated temperature increase of the potato tissue was less than 1 °C. Within one minute after the PEF treatment, the potatoes were removed from the treatment chamber and medium, and within 5 minutes they were vacuum packed (A300/16, Multivac, Wolfertschwenden, Germany) and placed in a fridge at 4 °C, where they were stored for maximum 16 h before use.

2.2.2 Mild thermal pretreatment

Potato tissue was thermally pretreated at 60 °C to promote PME activity, applying the treatment protocol of Moens et al. (2020) with some adjustments. From one untreated or PEF pretreated potato, eight cylinders (1 cm diameter, 1 cm height) were excised from the outer medulla using a stainless steel bore. The cylinder axis was parallel with the longitudinal axis of the potatoes. The cylinders were rinsed with 0.05% ascorbic acid to prevent browning, after which they were encapsulated per eight in stainless steel tubes (110 mm length, 13 mm internal diameter, and 1 mm thickness). Subsequently, the tubes were filled with demineralised water (0% Ca^{2+}), or demineralised water containing 0.05% Ca^{2+} (1.8341 g $CaCl_2.2H_2O$/l) or 0.5% Ca^{2+} (18.3411 g $CaCl_2.2H_2O$/l). The potato cylinders encapsulated in the stainless steel tubes were heated 20 min (including 3 min come up time) at 60 °C in a temperature-controlled water bath (MP-5, Julabo GmbH, Seelbach, Germany) (Smout et al., 2005).

2.2.3 Calcium infusion

To make sure all samples were exposed to Ca^{2+} to the same extent, potato cylinders were excised from untreated and PEF pretreated potatoes using the method described in Section 2.2.2. The potato cylinders were encapsulated per eight in stainless steel tubes (110 mm length, 13 mm internal diameter, and 1 mm thickness), after which the tubes were filled with 0% Ca^{2+}, 0.05% Ca^{2+} or 0.5% Ca^{2+}. Subsequently, the cylinders were incubated for 20 min at 4 °C.

2.3 Cooking behaviour of potato tissue

The cooking behaviour of the untreated and PEF and/or mild thermally pretreated potato tissue was determined (**Figure 1**) and was defined as the decrease of the relative hardness as a function of holding time at 95 °C.

2.3.1 Thermal treatment

The untreated and PEF and/or mild thermally pretreated potato tissues, encapsulated in stainless steel tubes with different media, were transferred from the 60 °C water bath (Section 2.2.2) or fridge (Section 2.2.3) to a temperature-controlled water bath at 95 °C (Julabo MP-5, Julabo GmbH, Seelbach, Germany). Three stainless steel tubes, containing 24 potato cylinders in total, derived from three different potatoes, were withdrawn at different holding times (t) at 95 °C (0-240 min). Samples with holding time 0 min were withdrawn after 5 min come up time (De Roeck et al., 2010; Moens et al., 2020). The stainless steel tubes were immediately cooled for 5 min in an ice bath and conditioned for 20 min in a 20 °C water bath (DC30 and W13, Haake, Karlsruhe, Germany) before texture measurement.

2.3.2 Texture measurement

The texture of the potato cylinders was measured using the TA.XT2i Texture Analyser (Stable Micro Systems, Godalming, United Kingdom) equipped with a 25 kg load cell, heavy duty platform and Texture expert exceed software (version 2.64, Stable Micro Systems, Godalming, United Kingdom). The hardness of the potato cylinders was defined as the peak force required to compress one cylinder to a 30% strain level, using an aluminum cylindrical probe with 25 mm diameter (P25, Stable Micro Systems, Godalming, United Kingdom) and test speed 1 mm/s (De Roeck et al., 2010; Moens et al., 2020). The relative hardness of potato tissue after a certain holding time in a specific medium was calculated as the average hardness at that holding time in the specific medium divided by the average hardness of untreated raw potato tissue (t = -5 min) soaked in the corresponding medium. The raw relative hardness was the relative hardness of untreated or pretreated potato tissue at t = -5 min. The cooking behaviour was defined as the decrease of relative hardness as a function of holding time ($t \geq 0$ min).

2.4 Characterisation of pectin after PEF and/or mild thermal processing

To investigate the effect of the different pretreatments and Ca^{2+} ions on PME activity, the DM of pectin from raw untreated and pretreated tissue, soaked or pretreated in 0% and 0.5% Ca^{2+}, was determined (**Figure 1**). The effect of the different pretreatments on calcium crosslinking of pectin was investigated in the case of the highest calcium concentration (0.5% Ca^{2+}).

2.4.1 Cryosectioning of potato tissue

The raw untreated and pretreated potato cylinders (Section 2.2.2 and Section 2.2.3) were incubated for 5 min in an ice bath, transferred from the tubes with medium (0% Ca^{2+} and 0.5% Ca^{2+}) to new tubes without medium, and stored in an ice bath until cutting. Thin sections of outer medulla of 90 µm thickness (approx. 7 x 7 mm) were cut transversally from the potato cylinders using a cryomicrotome (Reichert, Austria). To obtain cell wall material, they were washed in 70% ethanol to remove intracellular compounds such as starch and proteins, and stored at 4 °C in 70% ethanol until analysis (Christiaens et al., 2011; Moens et al., 2020). Each type of pretreatment was performed twice with five potatoes. Hence, sections were cut from ten different potatoes after two pretreatment repetitions.

2.4.2 Pectin degree of methylesterification

The sections were washed in phosphate buffered saline solution (40 mM NaCl, 2.7 mM KCl, 8.0 mM Na_2HPO_4 and 1.5 mM KH_2PO_4) at pH 7.4 and in demineralised water, after which the sections were transferred to a parafilm and dried by applying a drop of 70% ethanol (Christiaens et al., 2011; Moens et al., 2020). The pectin DM was measured using Fourier-transformation infrared (FT-IR) spectroscopy (FTIR-8400S, Shimadzu, Kyoto, Japan), according to the method of Kyomugasho et al. (2015). For each repetition of pretreatment, the DM was measured in fivefold (one measurement per potato).

2.4.3 Galacturonic acid content

Per repetition of pretreatment, two cryosections cut from five different potatoes were put together (5 x 2 sections, approx. 0.8 mg) and washed overnight at 4 °C by end-over-end rotation in demineralised water. Next, the sections were washed for 1 h in technical ethanol at 4 °C and dried overnight at 40 °C. The mass of the dried sections was determined and their GalA content was determined using a procedure based on the method of Ahmed & Labavitch (1977). Therefore, the sections were hydrated overnight in 1 ml demineralised water, 2 ml 98% sulphuric acid was added and the suspension was stirred for 2 h in an ice-bath to hydrolyse pectin. The hydrolysate was diluted to obtain a volume of 5 ml, after which the GalA content of the hydrolysate was quantified by the method of Blumenkrantz & Asboe-Hansen (1973). The hydrolysis step was performed twice per pretreatment repetition, and the GalA content of each hydrolysate was measured in triplicate.

2.4.4 Calcium content

Per repetition of pretreatment, five cryosections cut from five different potatoes were put together (5 x 5 sections, approx. 2 mg) and washed and dried as mentioned in Section 2.4.3. The dried sections were weighted in a porcelain crucible, hydrated with 1 ml ultrapure water (organic free, 18 MΩ·cm resistance), dried overnight at 100 °C, and ashed in a muffle furnace (Controller P330, Nabertherm, Lilienthal, Germany) at 550 °C for 20 h. The ashes were cooled to room temperature and dissolved in 9.9 ml ultrapure water, after which the solution was acidified with 0.1 ml 65% nitric acid and left overnight at 4 °C. The solution was filtered (0.45 µm membrane filter, Chromaphil® A-45/25, Machery-Nagel, Düren, Germany) and its calcium content was measured radially at 318 nm using inductively coupled plasma optical emission spectroscopy (iCAP 7400 ICP-OES Duo spectrometer, Thermo Scientific, MA, USA). An SPS-SW2 certified standard was used for calibration (Gwala et al., 2020; Moens et al., 2020). The calcium content was determined once per pretreatment repetition.

2.4.5 Pectin-calcium crosslinking

The bound calcium content was defined as the proportion of GalA units that is bound to calcium. It was calculated as the calcium content of the sections (mol/g) divided by the GalA content of the sections (mol/g). The bound calcium content was determined once per pretreatment repetition with 0.5% Ca^{2+} medium, using the corresponding GalA and Ca^{2+} measurements. The standard deviation on this parameter was calculated using error propagation taking into account the standard deviation on the repeated GalA measurements.

The proportion of demethylesterified GalA units that is bound to calcium was defined as the *R* value. Therefore, the calcium content of the sections (mol/g) was divided by the content of demethylesterified GalA units of the sections (mol/g) (Celus et al., 2018; Fang et al., 2008), with the content of demethylesterified GalA units being calculated using the DM and the GalA content of the particular sections. The *R* value was determined once per pretreatment repetition with 0.5% Ca^{2+} medium, using the corresponding DM, GalA and Ca^{2+} measurements. The standard deviation on this parameter was calculated using error propagation taking into account the error on the repeated GalA and DM measurements.

2.5 Statistical data analysis

The raw relative hardness (t = -5 min) of potato tissue after the different pretreatments was compared per calcium concentration using the Tukey HSD test with a significance level of 5% in JMP Pro (version 13.1.0, SAS Institute, Cary, NC, USA).

The cooking behaviour was modelled (proc nlin, SAS software, version 9.4, SAS Institute, Cary, NC, USA) using the fractional conversion model: $H = H_r + (H_0 - H_r)*exp(-k*t)$ with H = relative hardness of potato tissue at holding time t (% raw untreated), H_0 = initial relative hardness at t = 0 min (% raw untreated), H_r = residual relative hardness (% raw untreated), k = rate constant of texture degradation at 95 °C (min^{-1}), and t = holding time at 95 °C (min). For this, the relative hardness values were used, together with a weight factor, defined as 1/(standard error/relative hardness)². Consequently, relative hardness values with a large standard error were downweighted in the modelling. The model parameters H_0, k and H_r were estimated and compared between types of pretreatment and between calcium concentrations using their 95% confidence intervals. The adjusted coefficient of determination of each model was calculated as $R^2_{adj} = 1 - (m-1)(1 - SSR/SST)/(m-j)$ with m = number of observations, j = number of model parameters, SSR = sum of squares due to regression and SST = total sum of squares (De Roeck et al., 2010; Moens et al., 2020).

The initial relative hardness, rate constant and residual relative hardness were (separately) modelled as a function of the variables pretreatment (Untreated, PEF, T60 and PEF-T60) and calcium concentration using a linear model (proc mixed, SAS software, version 9.4, SAS Institute, Cary, NC, USA) to investigate which variable has a significant effect on the model parameters that describe the cooking behaviour. A weight factor, defined as 1/(standard error/estimate)², was used to take into account the unequal errors on the estimated initial relative hardness, rate constant and residual relative hardness.

The DM of pectin was compared between pretreatments and calcium concentrations (0 and 0.5 % Ca^{2+}) using the Tukey HSD test with a significance level of 5% in JMP Pro (version 13.1.0, SAS Institute, Cary, NC, USA).

The DM values were correlated with the estimated rate constants of the fractional conversion models per calcium concentration (proc nlin, SAS software, version 9.4, SAS Institute, Cary, NC, USA) using the exponential model $k = k_{DM0}*exp(a_{DM}*DM)$, with k = the rate constant of texture degradation at 95 °C (min^{-1}), DM = the degree of methylesterification (%), k_{DM0} = the rate constant of texture degradation at 95 °C if DM = 0%, and a_{DM} = exponential factor expressing the sensitivity of k for the DM. The adjusted coefficient of determination of each model was calculated.

The bound calcium content and the R values were compared between the different pretreatmens by means of their 95% confidence intervals. The middle of the interval was equal to the average value of the parameter of both pretreatment repetitions, and the interval width was determined by the sample size (n=2), a significance level of 5%, and the standard deviation of the average parameter value of both pretreatment repetitions, obtained using error propagation taking into account the standard deviation of the parameter value per pretreatment repetition. Samples with overlapping 95% confidence intervals were assumed to have a similar bound calcium content or R value.

The bound calcium content and R values were correlated with the estimated rate constants of the fractional conversion models in the case of 0.5% Ca^{2+} (proc nlin, SAS software, version 9.4, SAS Institute, Cary, NC, USA) using the exponential model $k = k_{i0}*exp(a_i*i)$, with k = the rate constant of texture degradation at 95 °C (min^{-1}), i = bound calcium content (BCC) or R value (R), k_{i0} = the rate constant of texture degradation at 95 °C if i = 0, and a_i = exponential factor expressing the sensitivity of k for i. The adjusted coefficient of determination of each model was calculated.

3. Results and discussion

3.1 Effect of PEF and mild thermal pretreatment on raw potato texture

Potato tissue was submitted to a PEF pretreatment, a mild thermal pretreatment at 60 °C and a combination pretreatment in media with different Ca^{2+} concentrations. The relative hardness of the raw pretreated tissues is shown in **Figure 2**. As the raw relative hardness of untreated tissue was 100% (not shown), all pretreatments reduced the relative hardness of the raw tissue. The PEF pretreatment promotes membrane electroporation (Boussetta et al., 2013; Fincan & Dejmek, 2003; Leong et al., 2014), while the mild thermal pretreatment at 60 °C affects the physical organisation of membrane lipids (Gonzalez et al., 2010; Gonzalez & Barrett, 2010). Both phenomena alter the integrity and permeability of the cell membrane and result in a lower turgor pressure and a softer texture. However, it seems that the PEF pretreatment has a larger impact on the texture than the mild thermal pretreatment, for all tested Ca^{2+} concentrations. Possibly, the PEF pretreatment has a larger impact on the membrane integrity than the mild thermal pretreatment. This difference in texture was also observed for both carrot cortex and vascular tissue (Moens et al., 2020). However, the harder texture after the mild thermal pretreatment can also be explained by the presence of swollen starch, resulting in a harder texture. The texture of tissue submitted to the combination pretreatment was similar to the texture of PEF pretreated tissue in the case of 0% Ca^{2+}, but harder than the texture of PEF pretreated tissue in the case of 0.05% and 0.5% Ca^{2+}. Probably, this is related with differences in starch swelling and/or Ca^{2+} crosslinking.

In general, the raw relative hardness of potato tissue increases if higher Ca^{2+} concentrations are used. The infusion of Ca^{2+} ions, which may be facilitated by cell membrane disintegration by the PEF (Leong et al., 2018) and the mild thermal pretreatment (Sila et al., 2005; Smout et al., 2005), may promote the ionic crosslinking of pectin and may result in a firmer cell wall, stronger middle lamella and harder texture. Therefore, the data suggest that Ca^{2+} binding is promoted with increasing amounts of Ca^{2+} (0.05% and 0.5% Ca^{2+}), for each of the pretreatments. As the formation of Ca^{2+} crosslinks requires the presence of blocks demethylesterified GalA residues, both the changes in pectin DM and Ca^{2+} crosslinking are investigated in Section 3.3. and Section 3.4, respectively.

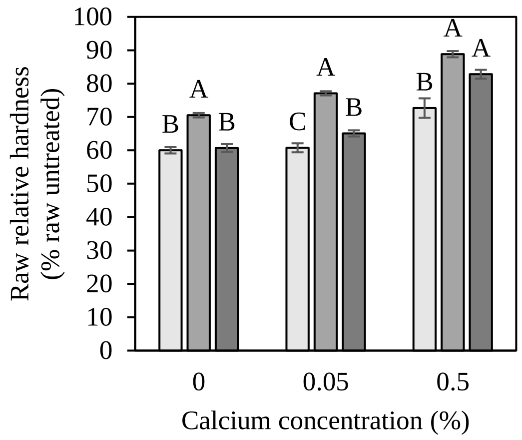

Figure 2 Relative hardness of raw potato tissue after a pulsed electric field pretreatment ( ), a mild thermal pretreatment at 60 °C ( ), and a combination of a pulsed electric field and mild thermal pretreatment ( ) in media with different Ca^{2+} concentrations. Significant differences in texture between pretreatments were investigated per Ca^{2+} concentration using the Tukey HSD test (α=0.05), and are indicated with different letters.

3.2 Cooking behaviour of potato tissue after PEF and mild thermal pretreatment

The cooking behaviour of potato tissue at 95 °C after no and a PEF and/or mild thermal pretreatment in media with different Ca^{2+} concentrations is shown in **Figure 3**. It is clear that the different pretreatments have a distinct effect on the texture of potato tissue during subsequent cooking and that the effect of the pretreatments also depends on the Ca^{2+} concentration of the surrounding medium. To be able to compare the cooking behaviour of potato tissue after the different pretreatments in the different media, the data points were modelled to a fractional conversion model. This isothermal curve describes the relative hardness as a function of holding time at 95 °C (**Figure 3)**. As some large deviations between texture measurements were found for some samples, the model was fitted using a weighted nonlinear regression procedure (Section 2.5). The estimates of the initial relative hardness, the rate constant, and the residual relative hardness, and the corresponding adjusted coefficient of determination (R^2_{adj}) are shown in **Table 1**. The high values of R^2_{adj} and the randomly distributed residuals (differences between predicted and experimental values, results not shown) indicate that the fractional conversion model is a good model to describe the texture degradation during the thermal treatments at 95 °C. R^2_{adj} was somewhat smaller for the PEF pretreated tissue in 0.05% Ca^{2+}, meaning the corresponding estimates are less precise.

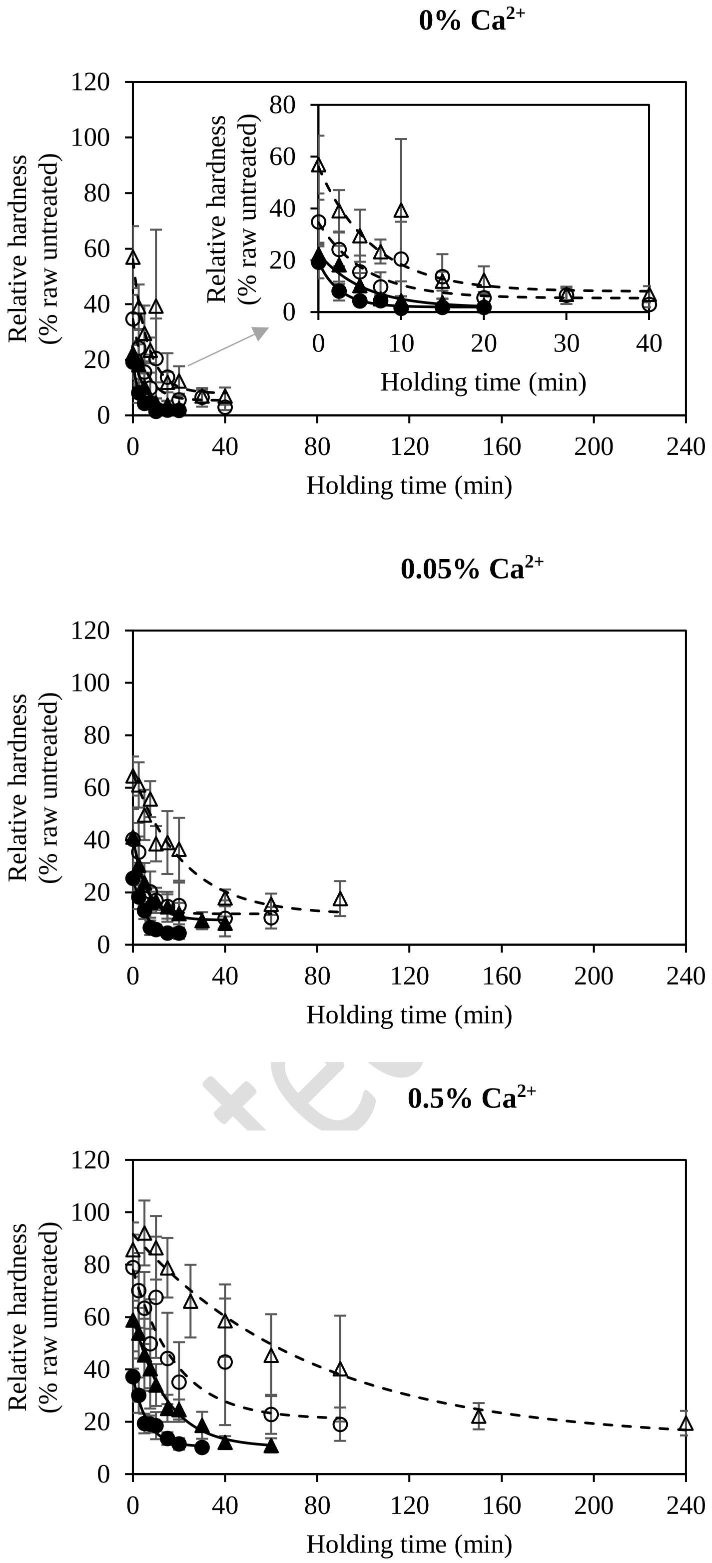

Figure 3 Cooking behaviour of potato tissue after no pretreatment (● Untreated), a pulsed electric field pretreatment (○ PEF), a mild thermal pretreatment at 60 °C (▲ T60), and a combination pretreatment (△ PEF-T60) in media with different calcium concentrations. The data were modelled to a fractional conversion model using weighted nonlinear regression (Untreated and T60: full line, PEF and PEF-T60 dashed line). Error bars represent standard deviations on the texture measurements.

Table 1 Parameters estimates and their standard error of the fractional conversion model, describing the cooking behaviour of untreated, pulsed electric field pretreated (PEF), mild thermally pretreated (T60), and PEF and mild thermally pretreated (PEF-T60) potato tissue at 95 °C in media with different calcium concentrations. H_0 = initial relative hardness, k = rate constant of texture degradation at 95 °C, H_r = residual relative hardness, R^2_{adj} = adjusted coefficient of determination. Significant differences in parameter estimates between pretreatments and calcium concentrations are indicated with different letters in superscript, based on the overlap of their 95% confidence intervals.

Calcium concentration (%)	**Pretreatment**	H_0 (% raw untreated)	k (10^{-2} min^{-1})	H_r (% raw untreated)	R^2_{adj}
0	Untreated	19.15 ± 0.73^{F}	38.10 ± 5.72^{A}	1.94 ± 0.51^{B}	0.985
	PEF	34.50 ± 1.99^{D}	17.37 ± 4.42^{ABC}	5.37 ± 2.19^{AB}	0.932
	T60	22.63 ± 1.02^{EF}	18.40 ± 4.29^{ABC}	1.74 ± 1.36^{B}	0.973
	PEF-T60	56.08 ± 2.44^{C}	15.23 ± 2.84^{ABC}	7.94 ± 3.18^{AB}	0.959
0.05	Untreated	25.61 ± 0.90^{E}	16.74 ± 2.92^{ABC}	2.28 ± 1.63^{B}	0.981
	PEF	41.39 ± 3.24^{D}	19.36 ± 6.10^{ABC}	11.80 ± 2.86^{A}	0.872
	T60	40.83 ± 1.19^{D}	15.62 ± 1.70^{B}	9.41 ± 1.04^{A}	0.983
	PEF-T60	65.49 ± 2.70^{BC}	4.53 ± 1.41^{CD}	11.52 ± 6.96^{AB}	0.992
0.5	Untreated	37.23 ± 2.22^{D}	16.05 ± 3.05^{ABC}	10.53 ± 1.33^{A}	0.950
	PEF	77.97 ± 2.99^{B}	5.31 ± 1.56^{CD}	20.93 ± 5.90^{A}	0.992
	T60	60.56 ± 1.77^{C}	6.93 ± 0.79^{C}	10.24 ± 1.88^{A}	0.985
	PEF-T60	91.66 ± 2.56^{A}	1.28 ± 0.35^{D}	13.22 ± 8.41^{AB}	0.996

The initial relative hardness represents the relative hardness after 5 min come up time, during which the texture can already soften. In case the turgor pressure isn't completely lost by a pretreatment, the turgor pressure can become completely lost due to thermal destabilisation of the cell membrane during the come up time. Temperatures above 50 °C also facilitate the gelatinisation of starch (Laborde & Padella-Zakour, 2003). Additionally, chemical depolymerisation of pectin through beta-elimination can already occur from temperatures of 80 °C and higher, but this contribution will be negligible during the come up time. Potato tissue treated in 0% Ca^{2+} had a higher initial relative hardness if pretreated with PEF or the combination pretreatment (PEF-T60), with the hardest texture being observed after the combination pretreatment (**Table 1**). In the case of 0.05% Ca^{2+}, the initial relative hardness of tissue submitted to a PEF, a mild thermal, and a combination pretreatment was significantly harder than the initial relative hardness of untreated potato tissue. Again, the hardest texture was observed after the combination pretreatment. The initial relative hardness of the tissues treated in 0.5% Ca^{2+} increased in the order

Untreated < T60 < PEF < PEF-T60. It can be observed that the initial relative hardness is higher if the treatments were performed in Ca^{2+} rich media.

After fitting the initial relative hardness estimates to a linear model (not shown), it becomes clear that both the pretreatments and the calcium concentration have a significant effect (α=0.05) on the initial relative hardness. The initial relative hardness increases in the order Untreated^{C} < T60^{B} = PEF^{B} < PEF-T60^{A}, with different letters in superscript referring to significant different effects on the initial relative hardness. Furthermore, an increasing calcium concentration results in a harder texture.

During the thermal treatment at 95 °C hardness will decrease by pectin depolymerisation through beta-elimination and by pectin solubilisation. However, the pectin demethylesterification and ionic crosslinking, that may result from the different pretreatments, can slow down the depolymerisation reaction, and hence retard the tissue softening. The rate constant of each curve was estimated (**Table 1**) and can be used to calculate the rate of texture degradation at a certain holding time. No significant differences between rate constants could be observed after cooking in 0% Ca^{2+}, although the untreated sample shows the highest texture degradation rate constant. Also after the treatments in 0.05% Ca^{2+} no significant difference between rate constants could be found, although the combination pretreatment seemed to result in the smallest texture degradation rate constant. The texture degradation of tissue treated in 0.5% Ca^{2+} was significantly slower after the combination pretreatment. However, in general, it seems that the texture during cooking remains harder in the order Untreated < T60 < PEF < PEF-T60 (except for 0.05% Ca^{2+}) (**Figure 3**). Moreover, the rate constants were smaller if a medium with a higher Ca^{2+} concentration was used, in particular after the mild thermal pretreatment and the combination pretreatment.

The rate constants were fitted to a linear model (not shown). As was already observed in **Table 1**, the rate constant decreases significantly with increasing calcium concentration. The effect of the variable 'pretreatment' on the rate constant, on the other hand, was not significant according to the F-test (Pr>F = 0.0883). However, according to the t-test, the different pretreatments decrease the rate constant in the order Untreated^{A} ≥ PEF^{AB} ≥ T60^{BC} = PEF-T60^{BC}. Therefore, both the mild thermal pretreatment and the combination pretreatment result in a significantly smaller rate constant, in comparison with the untreated tissue.

On the other hand, the effect of the pretreatments and the Ca^{2+} concentration on the residual relative hardness was negligible (**Table 1**). The residual relative hardness after the PEF and the combination pretreatment had a relatively high standard error for all Ca^{2+} concentrations tested. This may be due to the fact that only a few datapoints could be used to estimate this parameter (**Figure 3**). These pretreatments might result in a harder texture after prolonged heating, but these observations could not be confirmed by comparison of the 95% confidence intervals.

According to the linear model fitting the residual relative hardness values for different pretreatments and Ca^{2+} concentrations (not shown), the residual relative hardness significantly increases with increasing Ca^{2+} concentration. No significant effect of the variable 'pretreatment' could be found using the F-test (Pr>F = 0.1085), but the t-test showed an increasing residual relative hardness in the order Untreated^{C} ≤ T60^{BC} = PEF-T60^{BC} ≤ PEF^{AB}. Therefore, only a significant effect of the PEF pretreatment on the residual relative hardness, in comparison with the untreated tissue, could be found. The insignificant effect of the combination pretreatment on the residual relative hardness is probably due to its relatively high standard error.

It needs to be stressed that the evolution of the cooking curve depends on all three parameters (initial relative hardness, rate constant and residual relative hardness). Whereas the effect of the different pretreatments on the initial relative hardness was less clear in carrot tissues (Moens et al., 2020), the pretreatments had a significant effect on the initial relative hardness of potato tissue. This can be explained by the presence of (partially) gelatinised starch in potato cells after the mild thermal pretreatment, firming the texture. However, more significant differences between rate constants could be found for carrot tissue, even when cooked in demineralised water (Moens et al., 2020). These differences between vegetable tissues can, at least partially, be attributed to the lower ratio of HG to RG-I in potato pectin (Ralet et al., 2016). If the pretreatments would affect HG structure, it may be that these structural changes have a lower impact on the cooking behaviour of potato tissue. Moreover, the gelatinised starch in potato cells promotes cell separation and counteracts the adhesive function of pectin, which may be altered due to the different pretreatments.

However, with pectin changes being the rate determining process during the cooking process, it is interesting to investigate whether the pretreatments have an effect on the DM and ionic crosslinking of pectin in potatoes. The different pretreatments may facilitate endogenous PME activity, decreasing the pectin DM. Demethylesterified GalA residues may bind Ca^{2+} ions, especially when Ca^{2+} is available in excess, and form Ca^{2+} crosslinks that affect the solubilisation and depolymerisation of pectin during cooking. Therefore, the effect of the pretreatments and the Ca^{2+} concentration on the pectin DM, and the effect of the pretreatments on ionic crosslinking was investigated in Section 3.3 and Section 3.4, respectively.

3.3 Pectin demethylesterification

The DM of pectin from untreated and PEF and/or mild thermally pretreated tissue in media with 0% and 0.5% Ca^{2+} was determined (**Figure 4**). The PEF pretreatment reduced the DM significantly in case of both Ca^{2+} concentrations. Although the exact mechanism of the promotion of PME activity after such pretreatment needs further investigation, it is hypothesised that the electroporation of the cell membrane, and possibly of other membranes surrounding cell organelles such as the tonoplast, enhances enzyme-substrate contacts. An enhanced transport of intracellular (monovalent) cations to the cell wall may help deliberate PME from pectin chains and, hence, promote PME activity, or a higher availability of PME at the cell wall due to migration of the enzyme from the cytoplasm and its organelles to the cell wall may explain the reduced DM. The elevated concentration of Ca^{2+} in the treatment medium, on the other hand, had no significant effect on the DM. Too high concentrations of divalent cations may even inhibit PME activity as pectin becomes less accessible if ionically crosslinked (Christiaens et al., 2016). A reduction in pectin DM after a PEF pretreatment was also observed in carrot vascular tissue in the studies of Moens et al. (2020) and Moens, Huang, et al. (2021). The effect of the PEF pretreatment on the DM of pectin from carrot cortex was less pronounced and may be attributed to the lower conductivity (related to ionic content).

The mild thermal pretreatment had no significant effect on the DM (**Figure 4**). Possibly, the 20 min incubation at 60 °C was not long enough to significantly reduce the DM, or the elevated temperature might (partially) inactivate PME. Previous work showed that at this temperature the PME labile fraction and (to a smaller extent) the stable fraction may be inactivated (Moens, De Laet, et al., 2021). This makes it difficult to use a thermal pretreatment that at the same time affects the cell membrane integrity (allowing Ca^{2+} migration) and avoids PME inactivation. In carrots, on the other hand, a significant reduction of the DM was observed after a mild thermal pretreatment for pectin in both cortex and vascular tissue (Moens et al., 2020). A different thermal stability of PME from these vegetables may explain these different observations (Anthon & Barrett, 2002).

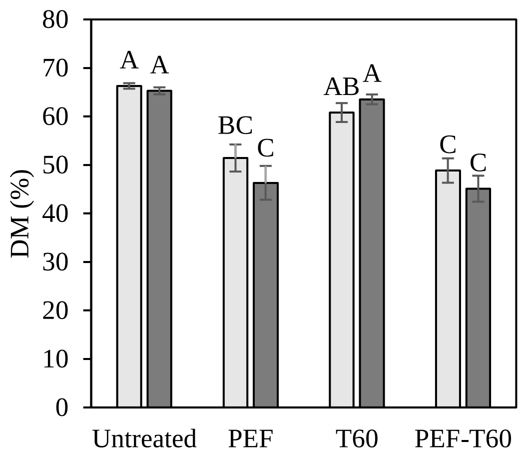

Figure 4 Degree of methylesterification (DM) of pectin from potato tissue that is subjected to no pretreatment (Untreated), a pulsed electric field pretreatment (PEF), a mild thermal pretreatment at 60 °C (T60), and a combination pretreatment (PEF-T60), and soaked in 0% Ca^{2+} (■) or 0.5% Ca^{2+} (■) medium. Error bars represent standard errors on the DM measurements. Significant differences in DM values between pretreatments and calcium concentrations are indicated with different letters, based on the results of the Tukey HSD test (α=0.05).

The combination pretreatment resulted in a pectin DM similar to the DM observed after the PEF pretreatment (**Figure 4**). It is clear that the mild thermal pretreatment has no additional effect on the pectin DM. A reduced DM after a combination pretreatment was also observed for pectin in carrot cortex (Moens et al., 2020) and especially in vascular tissue (Moens et al., 2020; Moens, Huang, et al., 2021).

Although the pectin DM seemed slightly lower after no pretreatment, the PEF and the combination pretreatment in case of the highest Ca^{2+} concentration, and the pectin DM seemed slightly higher after the mild thermal pretreatment in case of the highest Ca^{2+} concentration (**Figure 4**), no significant effect of the Ca^{2+} concentration on the PME activity could be demonstrated.

The reduction of the pectin DM after the PEF pretreatment is in correspondence with the harder texture during cooking after the PEF pretreatment (**Figure 3**). However, the rate constant of PEF treated tissue was not significantly lower than the rate constant of untreated tissue, for all Ca^{2+} concentrations tested (**Table 1**). The formation of ionic crosslinks, especially in the case of high Ca^{2+} concentrations, and the high initial relative hardness might explain the firmer texture during cooking.

The insignificant effect of the mild thermal pretreatment on the pectin DM was in line with the observed rate constant after such pretreatment, which was not significantly different from the rate constant of untreated tissue for all Ca^{2+} concentrations tested (**Table 1**). According to the linear model in Section 3.2, on the other hand, the rate constant was significantly affected by the mild thermal pretreatment. The presence of ionic and/or covalent crosslinks may (partially) explain the lower rate constant and the harder texture during cooking after a mild thermal pretreatment, compared to the texture during cooking of untreated tissue (**Figure 3**). Moreover, the position of the cooking curves at higher relative hardness values may be explained by the higher initial relative hardness, especially in the case of high Ca^{2+} concentrations.

Although the combination pretreatment resulted in a significant reduction of DM, the rate constant of PEF and mild thermally pretreated tissue was only significantly lower than the rate constant of untreated tissue if the treatments were performed in 0.5% Ca^{2+} (**Table 1**). The linear model (Section 3.2), on the other hand, suggested a significant effect of the combination pretreatment on the rate constant. However, the firmer texture during cooking after the combination pretreatment can’t be completely explained by a reduction in pectin DM, as the combination pretreatment resulted in a DM value similar to the DM after the PEF pretreatment, and the PEF pretreatment resulted in a less firm texture during cooking (**Figure 3**). Therefore, the presence of ionic pectin crosslinks is discussed in Section 3.4.

An exponential correlation between the pectin DM and the rate constant k ($k = k_{DM0}*exp(a_{DM*}DM)$) was found for 0% and 0.5% Ca^{2+} (**Figure 5** and **Table 2**). Again, it seems that the rate constant is lower in the case of 0.5% Ca^{2+} medium. However, k_{DM0} is not significantly different between Ca^{2+} concentrations. The large error on this estimate may be explained by the lack of datapoints at very low

DM values. Also a large error on the estimates of a_{DM} can be observed, and no significant difference in sensitivity of the rate constant of texture degradation at 95 °C to pectin DM (a_{DM}) between calcium concentrations can be demonstrated.

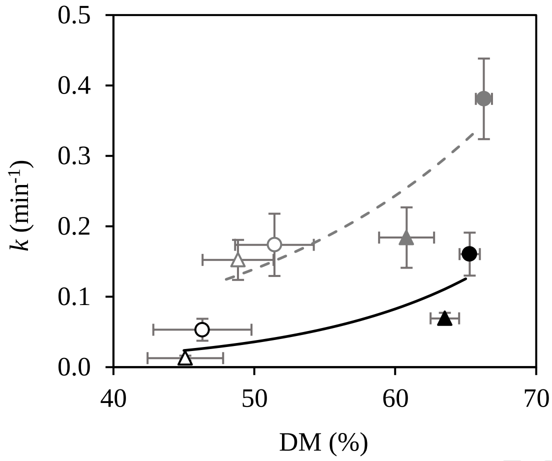

Figure 5 Exponential relationship between the degree of methylesterification (DM) of pectin and the rate constant k of texture degradation at 95 °C of tissue that was submitted to no pretreatment (● Untreated), a pulsed electric field pretreatment (○ PEF), a mild thermal pretreatment at 60 °C (▲ T60), a combination pretreatment (△ PEF-T60), and soaked in media with 0% Ca^{2+} (dashed line, grey symbols) or 0.5% Ca^{2+} (full line, black symbols). Vertical error bars represent the standard error on the estimates of the rate constant k and horizontal error bars represent the standard error on the DM measurements.

Table 2 Parameter estimates and their standard error of the exponential model, describing the relationship between the pectin degree of methylesterification (DM) and the rate constant of texture degradation at 95 °C (k) in media with 0% Ca^{2+} and 0.5% Ca^{2+}. k_{DM0} = the rate constant of texture degradation at 95 °C if DM = 0%, a_{DM} = exponential factor expressing the sensitivity of k for the DM, R^2_{adj} = adjusted coefficient of determination. Significant differences in parameter estimates between calcium concentrations are indicated with different letters in superscript, based on the overlap of their 95% confidence intervals.

Calcium concentration (%)	$\boldsymbol{k_{DM0}}$ (10^{-2} min^{-1})	$\boldsymbol{a_{DM}}$ (10^{-2} $\%^{-1}$)	$\boldsymbol{R^2_{adj}}$
0	0.86 ± 1.14^{A}	5.58 ± 2.15^{A}	0.954
0.5	0.06 ± 0.22^{A}	8.34 ± 6.33^{A}	0.843

To investigate the effect of the different pretreatments on ionic crosslinking of pectin, the bound calcium content and R value of potato cell wall material from untreated and pretreated potato tissue was compared for the highest Ca^{2+} concentration (Section 3.4).

3.4 Pectin-calcium crosslinking

The cell wall bound calcium content was determined for cell wall material from untreated and pretreated potato tissue, soaked or pretreated in 0.5% Ca^{2+} (**Figure 6** (a)). Both the PEF and the combination pretreatment significantly increased the proportion of GalA units that is bound to calcium. This observation can be explained by the reduced pectin DM in **Figure 4** after the same types of pretreatment: more demethylesterified GalA units are available to bind Ca^{2+} ions. The bound calcium content of cell wall material from mild thermally pretreated tissue seems higher than the bound calcium content of cell wall material from untreated tissue, but this difference is insignificant.

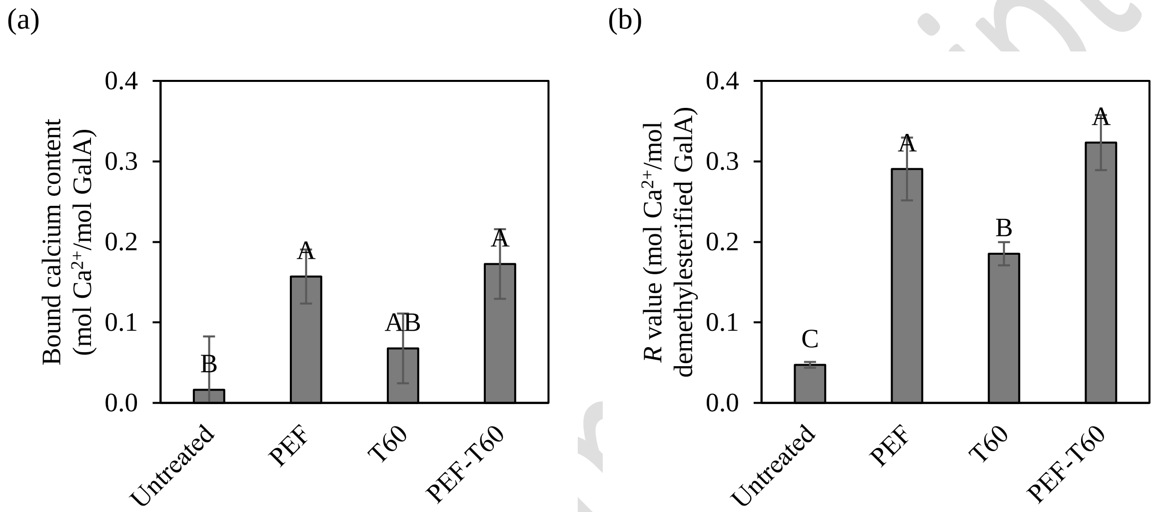

Figure 6 Bound calcium content (a) and *R* value (b) of potato cell wall material obtained after no pretreatment (Untreated), a pulsed electric field pretreatment (PEF), a mild thermal pretreatment at 60 °C (T60), a combination pretreatment (PEF-T60), and soaking in 0.5% Ca^{2+}. The error bars represent the standard deviations on the parameters. Significant differences in bound calcium content or *R* value between pretreatments are indicated with different letters, based on the overlap of their 95% confidence intervals.

On the other hand, the *R* value significantly increased after the mild thermal pretreatment (**Figure 6** (b)), meaning a larger proportion of demethylesterified GalA residues is bound to calcium. As the DM of pectin from mild thermally pretreated tissue was not significantly lower than the DM of pectin from untreated tissue (**Figure 4**), this difference in *R* value may be explained by an enhanced Ca^{2+} migration to the cell wall as a result of the thermal destabilisation of the cell membrane at 60 °C, whereby the Ca^{2+} ions bind existing demethylesterified GalA residues. The *R* value is highest after the PEF and the combination pretreatment. This suggests that these pretreatments not only result in the lowest pectin DM, but also in enhanced migration of Ca^{2+} ions towards the cell wall, binding the existing and newly formed demethylesterified GalA residues. As was already suggested in Section 3.1, the PEF pretreatment may have a larger effect on the membrane integrity than the mild thermal pretreatment. Moreover, the *R* values higher than 0.25 indicate that pectin chains are dimerised following an ordered egg-box model, resulting in a stronger gel and firmer texture (Celus et al., 2018; Fraeye et al., 2010). The DM (**Figure 4**) and *R* values (**Figure 6** (b)) were similar after the PEF and the combination pretreatment. Therefore, they might not be sufficient to completely explain the observed differences in texture during cooking (**Figure 3**). The formation of covalent crosslinks during the different pretreatments can't be excluded.

The observations in **Figure 6** are in line with the results obtained by Moens, Huang, et al. (2021) for carrot vascular alcohol insoluble residue (AIR), whereby the bound calcium content increased after the mild thermal pretreatment and, especially, after the PEF and the combination pretreatment. The *R* value of carrot vascular AIR, on the other hand, increased to the same extent after the three types of pretreatment. Probably, the intrinsic Ca^{2+} content of the carrot tissue was not high enough to bind the newly formed demethylesterified GalA residues, whereas in this experiment an excess of Ca^{2+} became available at the cell wall. However, as the carrot experiment was performed without calcium soaking, the bound calcium content and *R* value of carrot cell wall material might increase in the presence of additional Ca^{2+} ions.

As the exponential correlation of the rate constant of texture degradation at 95 °C and the pectin DM was not fully adequate for 0.5% Ca^{2+} (**Table 2**), the bound calcium content and the *R* value were correlated to the rate constant ($k = k_{i0}*exp(a_i*i)$, with i = bound calcium content (BCC) or *R* value). The rate constant decreases exponentially with increasing bound calcium content or increasing *R* value (**Figure 7** and **Table 3**). From the adjusted coefficient of determination it is clear that these parameters have a higher explanatory value for the cooking behaviour of potato tissue in 0.5% Ca^{2+} than the pectin DM (**Table 2**).

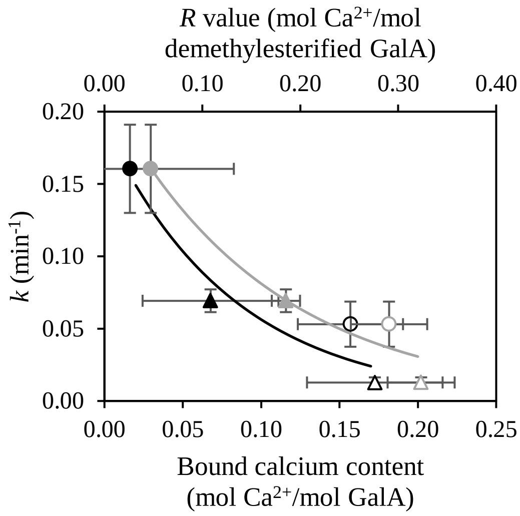

Figure 7 Exponential relationship between the bound calcium content (black) and the *R* value (grey) of potato cell wall material and the rate constant *k* of texture degradation at 95 °C of potato tissue that was submitted to no pretreatment (● Untreated), a pulsed electric field pretreatment (○ PEF), a mild thermal pretreatment at 60 °C (▲ T60), a combination pretreatment (△ PEF-T60), and soaked in 0.5% Ca^{2+} medium. Vertical error bars represent the standard error on the estimates of the rate constant *k* and horizontal error bars represent the standard deviation of the bound calcium content and *R* value.

Table 3 Parameters estimates and their standard error of the exponential model, describing the relationship between the bound calcium content (BCC) and *R* value of potato cell wall material and the rate constant of texture degradation at 95 °C in medium with 0.5% Ca^{2+}. k_{BCC0} = the rate constant of texture degradation at 95 °C if BCC = 0 mol Ca^{2+}/mol GalA, a_{BCC} = exponential factor expressing the sensitivity of *k* for the BCC, k_{R0} = the rate constant of texture degradation at 95 °C if *R* value = 0 mol Ca^{2+}/mol demethylesterified GalA, a_R = exponential factor expressing the sensitivity of *k* for the *R* value, R^2_{adj} = adjusted coefficient of determination.

Bound calcium content (*BCC*)	
$\boldsymbol{k_{BCC0}}$ (10^{-2} min^{-1})	19.01 ± 3.21
$\boldsymbol{a_{BCC}}$ (mol GalA/mol Ca^{2+})	-12.16 ± 3.55
$\boldsymbol{R^2_{adj}}$	0.960
***R* value**	
$\boldsymbol{k_{R0}}$ (10^{-2} min^{-1})	21.43 ± 2.97
$\boldsymbol{a_R}$ (mol demethylesterified GalA/mol Ca^{2+})	-6.07 ± 1.21
$\boldsymbol{R^2_{adj}}$	0.978

4. Conclusion

This paper investigated the effect of PEF technology on the texture evolution of potato tissue during subsequent cooking. More exactly, the cooking behaviour of untreated and PEF and/or mild thermally pretreated potato tissue was compared in media with different Ca^{2+} concentrations. In general, the initial relative hardness increased in the order Untreated < T60 = PEF < PEF-T60. Although the texture during cooking tended to be harder in the order Untreated < T60 < PEF < PEF-T60, almost no significant differences between rate constants, corresponding to the different pretreatments, could be found. Only a significantly slower cooking behaviour could be observed after the combination pretreatment in 0.5% Ca^{2+} medium. Linear modelling of the rate constants suggested an effect of the mild thermal pretreatment and the combination pretreatment. The effect of the different pretreatments on the residual relative hardness was negligible. It is clear that the actual evolution of texture during cooking is determined by the interplay of the relative hardness obtained after the pretreatment, the rate constant, and the residual relative hardness after prolonged cooking. Additionally, the presence of Ca^{2+} tended to increase the relative hardness and to reduce the rate constant.

Only the PEF and combination pretreatment reduced the pectin DM significantly, and independently of the Ca^{2+} concentration used. Accordingly, the bound calcium content and *R* value of potato cell wall material increased after these types of pretreatment (0.5% Ca^{2+}). The mild thermal pretreatment had no significant effect on the pectin DM, but seemed to facilitate the binding of Ca^{2+} ions (0.5% Ca^{2+}) to existing demethylesterified GalA residues. The rate constant of texture degradation was exponentially correlated with the pectin DM (0-0.5% Ca^{2+}), and the bound calcium content and *R* value (0.5% Ca^{2+}).

In the case potatoes are thermally processed to inactivate endogenous enzymes and micro-organisms and a firm 'fresh-like' texture is desired after the thermal process, the introduction of a PEF pretreatment, as such or combined with a mild thermal pretreatment, may be considered. However, the texture preserving effect of the pretreatment is less pronounced than in carrot vascular tissue (Moens et al., 2020). The use of treatment media with high calcium concentrations might help to better preserve texture of PEF pretreated potato tissue during thermal processing. Moreover, the PEF pretreatment may also reduce the energy requirements to cut the fresh tissue. In the case potatoes are thermally processed to soften the tissue for reasons of palatability, special attention has to be paid to the texture of the final product if a PEF pretreatment is introduced to reduce the cutting force of the fresh tissue. Upon thermal processing, the PEF pretreated tissue may remain harder than untreated tissue. The complete inactivation of PME and/or the use of treatment media with low Ca^{2+} concentrations might be helpful to limit the texture preserving effect of the PEF pretreatment during thermal processing.

Declaration of competing interest

The authors declare no conflict of interest.

Acknowledgement

This research was partly funded by VLAIO (Flanders Innovation and Entrepreneurship, Belgium) in the context of a Baekeland mandate (HBC.2016.0591).

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