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Doctorial thesis

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Abstract

Due to the health effects of dietary fibre, as well as its water-holding capacity (WHC), food products are commonly fortified with fibre-rich sources. However, to be able to design food products with specific textural properties that also appeal to the consumer, it is important to understand and to characterise the physicochemical and chemical properties of the fibre. In this study the composition of dietary fibre, the solubility of pectin, the microstructure, rheological properties and water-holding capacity, as well as the sensory properties of fruit and vegetable suspensions and fibre-amended sausages, have been investigated. The plant materials used were tomato, apple, carrot and potato pulp. The variation in these properties following different kinds of processing, i.e. homogenisation and heat treatment, was also studied.

The fibre suspensions responded differently to high-pressure homogenisation. This could be due to the fundamentally different inherent microstructures of the samples, probably originating from different proportions of soluble and insoluble fibre. A high content of insoluble pectin and a high perceived crispiness/graininess was found in suspensions with a microstructure consisting of large cell clusters and aggregates (carrot and potato pulp). The microstructure of these suspensions is degraded to smaller clusters by homogenisation, but retains their original cellular shape. However, suspensions with a microstructure consisting of single cells and cell fragments (tomato and apple), were more easily degraded by homogenisation, and contained lower amounts of insoluble pectin. The latter type of microstructure exhibited a higher WHC, elastic modulus and sensory perceived melting and slipperiness.

The main effect of heat treatment was on the solubility of the dietary fibre. In carrot and apple suspensions where β-elimination was favoured by heating, an increase was seen in the amount of soluble pectin together with a decrease in the mean particle size of the insoluble fibre. With the decrease in the insoluble material, a significant decrease in the WHC was seen for both apple and carrot. Heat-treated potato pulp

suspensions were affected differently, since starch remaining in the matrix started to swell, which led to a difference in the particle size distribution and morphology. There was also a significant increase in WHC with heating, probably resulting from the swelling of the starch.

Low-fat sausages containing potato pulp exhibited greater firmness than the sausage without any fibre, according to a compression test. It is suggested that the high content of insoluble fibre in the potato pulp forms a strong fibrous network in the meat protein network of the sausages, thereby increasing the firmness.

Populärvetenskaplig sammanfattning

Livsmedelsverket rekommenderar att vi ska äta 25-35 g kostfiber per dag. De flest har dock svårt att få i sig den mängden, då det motsvarar mer än 1 kg morötter. Genom att berika olika vardagsprodukter med kostfiber kan man öka intaget. För att dessa produkter ska konsumeras måste de ha liknande, eller godare, smak och konsistens som produkten utan kostfiber. Därför bör kostfiberns sammansättning och egenskaper ändras så att bästa möjliga konsistens kan uppnås. Syftet med denna studie har därför varit att förstå hur olika processer kan påverka kostfibers egenskaper så att de på bästa sätt kan berika köttprodukter som t.ex. korv.

Kostfiber finns bland annat i frukt och grönsaker. I denna studie har några vanliga frukter och grönsaker använts för att studera processpåverkan: äpple, tomat, morot och potatispulpa. Dessa fiberkällor har gjorts till puré och sedan blandats i en vätska till en suspension för att kunna undersöka hur bra de nätverk som fibrerna bildar är. Ett bra nätverk behövs för att fibern ska kunna bidra till önskad konsistens hos det berikade livsmedlet samt ge en ökad vattenhållande förmåga.

Ett sätt att ändra konsistensen av kostfiber är att finfördela frukten och grönsakerna genom homogenisering. Då pressas fiberblandningen med högt tryck genom en smal spalt vilket resulterar i att partiklarna i blandningen blir mindre. Resultatet av denna process kan undersökas genom att studera fibrerna i mikroskop. Där avslöjades den stora skillnaden mellan de olika kostfiberkällorna. Potatispulpa, som är en biprodukt efter stärkelsetillverkning, och morot består av många, små celler som sitter ihop i stora aggregat. Tomat och äpple däremot består av större enskilda celler. Den stora skillnaden kan förstås då kostfiberinnehållet mättes. Kostfiber är ett samlingsnamn för flera olika ämnen som inte tas upp av magen och kommer huvudsakligen från cellväggar. Dessa består huvudsakligen av cellulosa, hemicellulosa och pektin. Lösligheten hos pektin kan få stora konsekvenser för strukturen. I potatispulpa och morot finns mycket olösligt pektin som fungerar som klister mellan cellerna och håller ihop dem. När fibrerna homogeniseras går tomatceller lättare sönder än potatispulpa eftersom det inte finns lika mycket olösligt pektin som håller ihop dem. Som konsekvens bildar tomat- och äppleblandningarna starkare och mindre kompakta nätverk som kan hålla vatten bättre än morot och potatispulpa.

De flesta livsmedel värmebehandlas i någon del av tillverkningen. Om man har berikat produkten med kostfiber behöver man veta hur konsistensen ändras med värmebehandling. Vid värmning av kostfiber kan nämligen lösligheten hos pektin ändras, vilket påverkar egenskaperna hos fibern. Äpple- och morotsuspensioner påverkades på liknande sätt av värmning; då de värmdes till hög temperatur under lång tid bröts de olösliga pektinkedjorna ner till lösligt pektin. Eftersom det främst är de olösliga fibrerna som bildar nätverk kunde dessa värmebehandlade fiber inte hålla lika mycket vatten som de obehandlade fibrerna. Suspensionerna med potatispulpa

påverkades annorlunda vid värmning. De innehöll ca 15 % stärkelse som började svälla under värmningen. Därmed ökade den vattenhållande förmågan vid samtliga studerade värmebehandlingar.

För att förstå hur de olika egenskaperna hos kostfiber påverkar konsistensen fick en smakbedömningspanel provsmaka de olika fiberblandningarna. Konsistensen bedömdes genom fem olika parametrar: grynig, krispig, salvig, smältbar och tjock. Desto mer salvig och smältbar en fiberblandning var, desto starkare nätverk och mer lösligt pektin hade den. Blandningar med stora partiklar uppfattades å andra sidan som krispiga och gryniga. Morot och potatispulpa upplevdes som mer krispig och grynig, medan äpple och tomat ansågs vara mest salviga och smältbara. De suspensioner som hade ett starkt nätverk av lösligt pektin i vattenfasen (tomat och potatispulpa) upplevdes som tjocka.

Slutligen testades hur potatispulpa påverkar konsistensen hos en korv med låg fetthalt. För att få en rättvis jämförelse hölls de flesta parametrar som påverkar vattenhållande förmågan konstanta, som tillsatt stärkelse och kvoten mellan vatten och protein. Korv med potatispulpa var fastare än referenskorv utan kostfiber. Detta beror troligtvis på att potatispulpans olösliga fiber kan bilda ett nätverk som resulterar i en fastare konsistens. Det var i övrigt inga skillnader mellan de olika korvarna. Så genom att tillsätta potatispulpa till korvsmet kan man få i sig mer fiber samtidigt som man äter en god korv.

List of papers

This thesis is based on the following papers, which will be referred to in the text by their Roman numerals. The papers are appended at the end of the thesis.

I. Physicochemical characterisation of the dietary fibre matrix in fruit and vegetable suspensions
Åberg H., Nyman M. and Tornberg E.
Submitted for publication January 2009

II. Physicochemical characterisation of fruit and vegetable suspensions
I: Effect of homogenisation
Åberg H. and Tornberg E.
Submitted for publication October 2009

III. Physicochemical characterisation of fruit and vegetable suspensions
II: Effect of heat treatment
Åberg H., Wikberg J. and Tornberg E.
Submitted for publication October 2009

IV. Effects of physicochemical properties on the sensory perception of the texture of homogenised fruit and vegetable suspensions
Åberg H., Hall C. and Tornberg E.
Submitted for publication October 2009

V. Heat-treated and homogenised potato pulp suspensions as additives in low-fat sausages
Åberg H., Montelius C. and Tornberg E.
Manuscript

The author's contributions to the papers

I. The author performed all the experimental work, took an active part in the evaluation of the results and wrote the major part of the paper.

II. The author designed the study together with the co-authors, performed all the experimental work, took an active part in the evaluation of the results and wrote the major part of the paper.

III. The author designed the study together with the co-authors, performed most of the experimental work, evaluated the results and wrote the major part of the paper.

IV. The author designed the sensorial study together with the co-authors, evaluated the results and wrote the major part of the paper.

V. The author designed the experiments together with the co-authors, evaluated the results and wrote the major part of the paper.

Abbreviations and symbols

Abbreviations

ANOVA	Analysis of variance
DF	Dietary fibre
DM	Degree of methylation
GC	Gas chromatography
HM pectin	High-methoxy pectin
IM	Insoluble material
LM pectin	Low-methoxy pectin
LVER	Linear viscoelastic region
PC	Principal component
PCA	Principal component analysis
PME	Pectin methyl esterase
PSD	Particle size distribution
SM	Soluble material
WHC	Water-holding capacity

Symbols

G′	Pa	Elastic modulus
G″	Pa	Viscous modulus
d_{32}	µm	Surface-area-weighted mean particle size
d_{32} (s)	µm	Surface-area-weighted mean particle size in the interval 0.1-100 µm
d_{32} (l)	µm	Surface-area-weighted mean particle size in the interval 100-900 µm
d_{43}	µm	Volume-weighted mean particle size
tan δ	-	Phase angle

Contents

1. Introduction

Dietary fibre (DF) is found in the cell wall of fruit and vegetables, and is defined as indigestible polysaccharides and lignin [1]. DF consists mainly of cellulose, hemicellulose, lignin and pectin (Fig. 1). Cellulose, a linear β(1-4)-linked polymer of glucose, forms insoluble microfibrils by hydrogen bonding between the chains [2]. Cellulose contributes to a high extent to the structure and strength of the cell wall [3]. The other components of DF are more heterogeneous in their chemical structure. Hemicellulose is a generic term for several polysaccharides; β(1-4)-linked glucose, xylose or mannose residues are found in the backbones. These polymers are often linked to cellulose by hydrogen bonds, and are thus insoluble in water, however, the solubility varies due to variations in structure and composition [4, 5]. The most abundant hemicellulose in fruit and vegetables is xyloglucan, consisting of a backbone of glucose with α(1-6)-linked xylose side-chains. Lignin is formed by oxidative cross-linking of the phenylpropane units of coniferyl, p-coumaryl and sinapyl alcohols. The rigid structure of the aromatic rings makes lignin highly water-insoluble [2].

Pectins are a complex group of cell wall polysaccharides, in which two different domains are normally seen. The smooth region consists of α(1-4)-linked galacturonic acid residues, while the so-called "hairy" region is especially rich in highly branched rhamnose [6]. The carboxyl group of the galacturonic acid residue can be esterified with a methyl group, which contributes considerably to the properties of the pectin chain. Pectin is found surrounding the hemicellulose and cellulose matrix in the cell wall (Fig. 1), as well as in the middle lamella connecting adjacent cells [4]. Depending on the binding with other cell wall components, pectin can be either soluble or insoluble in water. The contents of the different components vary in different DF sources.

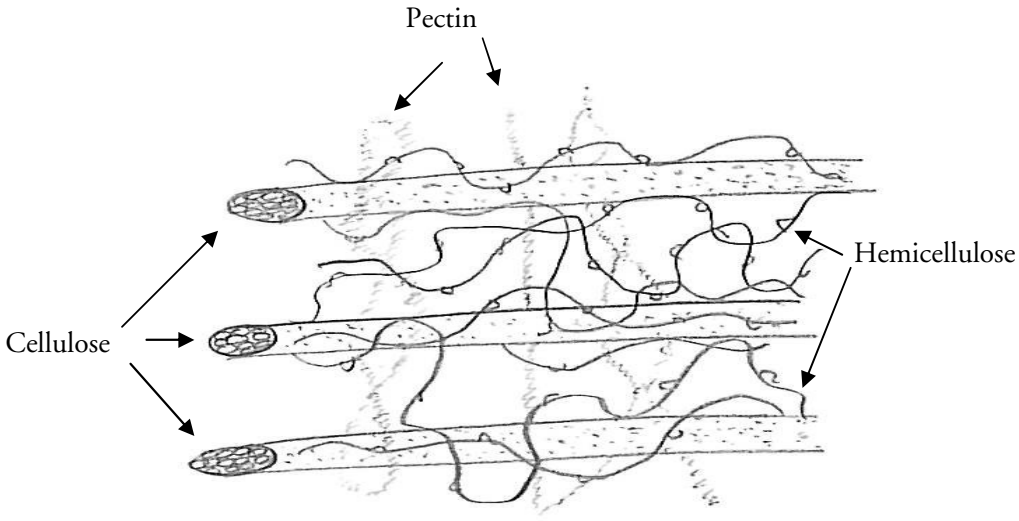

Figure 1. Schematic illustration of the cell wall (adapted from Carpita & Gibeaut [7]).

A suspension consists of dispersed solid particles in a continuous phase. When the solid particles consist of polymers, such as the polysaccharides from DF, at a sufficiently high concentration, a network can be formed. These suspensions have both solid-like and liquid-like properties, i.e. they exhibit viscoelastic behaviour [8]. The physicochemical, physiological and sensory properties of suspensions are dependent on the network formed by these materials.

The recommended daily intake of dietary fibre is 25-35 g. However, this is rarely achieved in populations of the Western world [9, 10]. An increase in the amount of DF in the diet may prevent some of today's most common diseases: high blood cholesterol, colon cancer and diabetes [11]. Different types of DF affect health in different ways: soluble DF delays or prevents the absorption of macromolecules in the stomach, and is often fermented by the colonic bacteria to produce short-chain fatty acids, while insoluble DF is more resistant to the colonic microflora, thus having a bulking effect [10, 12, 13]. However, this subdivision of soluble and insoluble DF is not always valid, for example, some insoluble DF can be degraded in the colon [10].

The physiological effects of ingested DF prevent the previously mentioned diseases in several ways [10]. Apart from the solubility of DF, other changes in the physicochemical properties by processing may also affect the physiological response [14]. The particle size of DF determines the transit time through the gastrointestinal tract. Hydration properties affect the bulking capacity, while rheological properties affect the absorption [15].

The physicochemical properties of DF are not only important in relation to the effect on human physiology, but they also govern technological properties such as the texture of a food product [16]. The addition of DF to other food products could be one way of increasing its consumption. However, the organoleptic appeal to the consumer must not be sacrificed [17]. Therefore, we must improve our knowledge of the behaviour of DF when used as an additive in food. If the chemical composition and physicochemical properties of DF could be correlated to its sensory properties, it may be possible to predict the texture of the final product when adding DF.

1.1 Objectives

The general aim of this work was to achieve a better understanding of the effects of processing of fruit and vegetable suspensions by investigating the content and composition of DF, and the physicochemical and sensory properties. The specific objectives were:

- to investigate the composition of the soluble and insoluble fractions of DF and their correlation with some physicochemical properties of four fruit and vegetable suspensions in both unprocessed (Paper I) and high-pressure homogenised suspensions at two concentrations of insoluble material in the fibre sources (Paper II)

- to study the change in pectin content and physicochemical properties when three fruit and vegetable suspensions were subjected to various kinds of heat treatment favouring pectin methyl esterase activity and/or β-elimination (Paper III)

- to investigate the effect of fibre source, concentration and degree of homogenisation on the sensory properties of the texture of four fruit and vegetable suspensions, and to investigate the correlations of these textural properties to some physicochemical variables and the composition of DF (Paper IV)

- to investigate how different processing of potato pulp suspensions affects a meat protein network when added to sausages, with regard to sensory characteristics, instrumental firmness and process loss (Paper V).

2. Dietary fibre sources

Fruit and vegetables are good sources of DF since they contain both soluble and insoluble DF [18]. The four fruits and vegetables used in the various parts of this study were carrot, potato pulp, apple and tomato. These materials were chosen since they have different plant physiological background where for example potato pulp contains starch and tomato has a high content of soluble DF. The chosen fruit and vegetables is used in industrial processing and, for parts of the study, the DF sources used were already pre-processed on an industrial scale to the extent that the enzymatic activity of the fruit and vegetables was minimised. The raw materials were suspended for the various processes and measurements, which are summarised in Fig. 2.

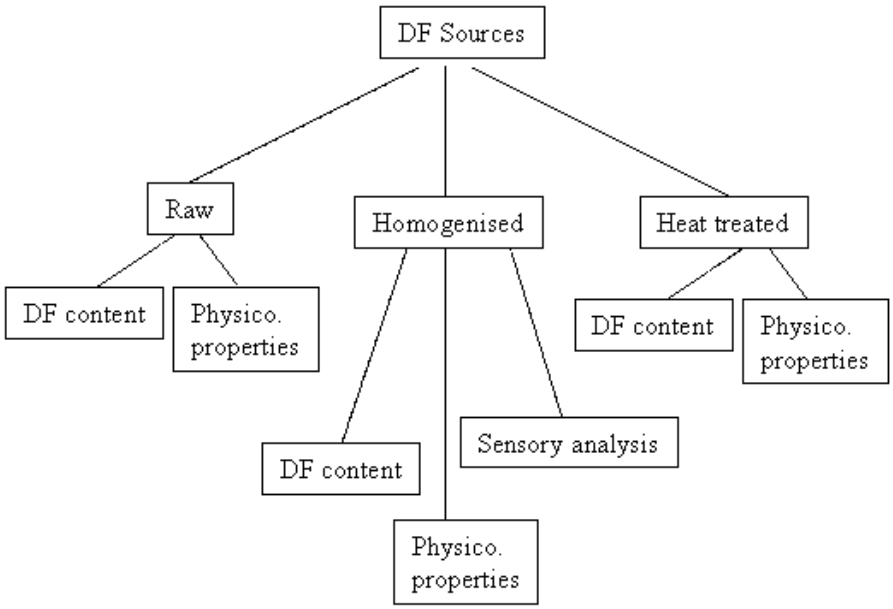

Figure 2. Overview of the processes and measurements used in the study (physico. = physicochemical)

Carrot is traditionally grown in Sweden since it is a cool weather crop. Carrots are sown during spring and harvested in the autumn [19, 20]. Carrots are cultivated on about 1800 ha in Sweden, which makes it one of the most-cultivated horticultural products in Sweden, with 90 000 tons being harvested in 2007 [21]. Carrots can be eaten raw, but are also processed and sold as juice, pickles, marmalade or frozen. The carrot used in the present study was provided in frozen cubes by Magnihill AB, Sweden, both mildly heat treated (93-96ºC, 6-6½minutes) (Papers I, II & IV) and unprocessed (Paper III). The carrot cubes were thawed and chopped in a food processor (Electrolux AB, Sweden) before mixing with the dispersion media to a suspension.

Potato has been used in Sweden to produce starch since the 1870s. The amount of potatoes used for starch production in Sweden has doubled since the 1960s, to almost 300 000 ton in 2008 [22]. All starch production nowadays takes place in the south of Sweden. In the 1980s the by-product, potato pulp, was recognised as a useful product [23], mainly because of its high content of DF. It has since been sold wet as animal feed, or dried as a food additive under the name Potex, in which about 12-15% of the starch remains in un-gelatinised form. Potato pulp, both dried (Papers I, II & IV) and wet (Papers III & V), from Lyckeby Stärkelsen, Sweden was used in this study.

The tradition of planting apple trees outside Kivik in the south of Sweden to make juice goes back to 1888. Today, 1400 ha is covered with apple trees in Sweden, and 21 000 tons of apples were harvested in 2007 [21]. Apples are eaten raw or used for juice or apple sauce production. The mashed apples used in this study were kindly provided by Kiviks Musteri AB, Sweden. The apples were blanched (85ºC, 5 min) (Papers I, II & IV) or frozen (Paper III) after the industrial grinding and mixing with bacteriostats.

Tomatoes can be grown outdoors in Sweden, but most are produced in greenhouses, and the production is rather small (17 000 tons harvested from 500 000 m^2 in 2007) [21]. Most of the tomatoes eaten in Sweden come from the Mediterranean countries, where Spain and Italy are high producers (3 700 000 and 6 000 000 tonnes, respectively in 2007) [24]. Although a small proportion of tomatoes is sold for raw consumption, a great deal are processed. Heat-treated tomatoes are sold as crushed tomatoes or as paste, from which ketchup is made. The tomato paste used in this study was provided by Procordia Foods, Sweden. The tomatoes were delivered as hot break tomato paste (28-30° Brix) (Papers I, II & IV).

3. Processing of fibre suspensions

A large proportion of the food products in supermarkets today have been processed in some way. The purpose of processing varies considerably: to ensure a safe product with low microbiological activity, to prolong shelf-life by stabilising the components, to render new physiological properties, to change the taste and texture, or a combination of these [25, 26]. A combination of several effects is often obtained by processing. In many cases, the DF is processed before being added to the product, and is then processed again when the product is processed.

DF is usually analysed after severe extraction procedures [27]. These procedures can cause changes in the plant material that can affect both the physicochemical properties and the physiological response [28, 29]. In this study, unrefined fruits and vegetables were therefore chosen as the starting material, where possible. In the experiments involving no prior heat treatment (Paper I, II & IV), the potato pulp was dried to minimise the enzymatic activity. Drying was carried out at a low temperature to prevent the starch granules from gelatinising. In the experiments involving heat treatment the same potato pulp was used, but it was not dried before use. The other fibre sources were blanched before used in the experiments without prior heat treatment, as described in Section 2.

When adding DF derived from fruit and vegetables it is important to change the size of the macromolecules and to promote or inhibit enzymatic activity [30]. Different methods can be employed to reduce the particle size of DF. If the DF is dried it can be ground to the desired particle size [31]. For non-dried sources a decrease in the particle size can be achieved by, for example, mixing, blending or homogenisation. Moderate temperatures can be used to increase the enzymatic activity, and thus improve the texture of a fibre suspension [32]. High temperatures can be used to terminate enzymatic activity and increase the rate of depolymerisation [33]. Many

products must also be heat treated in order to kill pathogenic or spoilage microorganisms. The different processes and the type of fruit and vegetables used in the present study are summarised in Table 1.

Table 1. Fruit, vegetables and processes investigated

DF source	Process
Paper I	
Carrot, apple, tomato and dried potato pulp[2]	None[1]
Paper II	
Carrot, apple, tomato and dried potato pulp[2]	Homogenisation[1]
Paper III	
Carrot, apple and potato pulp	Heat treatment
Paper IV	
Carrot, apple, tomato and dried potato pulp[2]	Homogenisation[1]
Paper V	
Potato pulp	Homogenisation and heat treatment

[1]The samples were blanched to minimise enzymatic activity
[2]Dried potato pulp (Potex)

3.1 Homogenisation

High-pressure homogenisation decreases the particle size and changes the shape of the aggregates. It is achieved by pumping a suspension through a thin slit. The pressure drop across the slit causes the flow to become turbulent, which creates eddies. These eddies are believed to break down the particles, in combination with cavitation created by the low pressure in the slit [34, 35]. Homogenisation has been applied to emulsions such as mayonnaise and milk since about 1950 [25]. Homogenisation to

change the properties of fibre suspensions has traditionally been used mainly in the processing of tomatoes [36] and in the production of fruit juices [37]. Recently, high-pressure techniques have been used to homogenise components other than the traditional food components [25]. A decrease in molecular weight has been observed in soluble fibre such as pectin [38], xanthan gum [39] and methylcellulose [40] when subjected to high-pressure homogenisation.

The effect of homogenisation is determined by the pressure applied. A pressure of 17 MPa has been seen to lead only to a small decrease in the molecular weight of pectin, compared with the effect of 124 MPa [38]. The number of passages through the homogeniser also affects the particle size of the suspension. The coarse, insoluble particles, larger than 10 μm, of a tomato paste suspension have been found to decrease from 73% of the non-homogenised suspension, to 47% after the first passage, and to 28% after two passages in a lab-scale homogeniser at 90 bar [41]. The molecular weight of soluble xanthan gum dispersions has also been found to decrease with increasing number of passages. The reduction was more pronounced after the first few passages, when a 42.4% decrease in molecular weight was seen. However, the decrease between the 16^{th} and 20^{th} passages was only 7.7% [39]. The concentration of insoluble particles is also of importance when subjecting a fibre suspension to homogenisation. When the content of water -insoluble solids in a tomato paste suspension was increased from 0.5% to 1.3%, the effect on particle size reduction was found to decrease dramatically [41]. The viscosity of the continuous phase can also affect the efficiency of the homogenisation; a Newtonian solution with a high viscosity decreases the effect [42].

Homogenisation affects foremost the particle size and shape, which can affect the physicochemical properties [30]. Homogenisation affects the rheological properties of a suspension, such as the viscosity and elastic modulus, however, the reported changes in viscosity are somewhat inconsistent. A decrease in viscosity has been reported when soluble DF, such as pectin and xanthan gum, was homogenised [38, 39], while

homogenised tomato paste suspension and strawberry sauce showed enhanced rheological properties, compared with the non-homogenised suspension [43]. It thus appears that homogenisation has detrimental effects on the rheological properties of soluble fibre, but positive effects on insoluble fibre. However, this seems not to apply to all insoluble fibre suspensions, since apple sauce has been found to have a lower apparent viscosity after homogenisation [44].

A lab-scale valve homogeniser [45] was used in this study (Papers II & IV). The homogeniser was used at a maximum of 90 bar, however, the effect on particle size distribution has been reported to be similar to the capacity of other equipment at a pressure of about 200 bar [46]. Samples were homogenised batch-wise to ensure that all the particles in the suspensions had passed through the slit the same number of times. The number of passages for each fibre suspension can be seen in Table 2. The number of passages was chosen arbitrarily, with approximately twice the number of passages between H1 and H2.

Table 2. The number of passages in the homogenising equipment corresponding to treatments H1 and H2

Fibre source	Conc. IM[1] (%)	H1 (number of passages)	H2 (number of passages)
Apple	0.8	4	10
	1.2	5	11
Tomato	0.8	9	23
	1.2	11	25
Potato pulp	0.8	4	8
	1.2	5	10
Carrot	0.8	8	18
	1.2	8	21

[1]IM: Insoluble material
From Paper II

3.2 Heat treatment

When fruits or vegetables are heat treated, several reactions, both chemical and enzymatic, occur. The component in DF that is most affected by heat treatment is pectin [47]. Two main chemical changes occur in pectin during heat treatment: de-esterification and depolymerisation.

Pectin methyl esterase (PME) is an enzyme endogenous to most fruits and vegetables. It acts by removing the methyl group on the galacturonic acid residue in the pectin backbone, thus decreasing the degree of methylation (DM), thereby increasing the reactivity with Ca^{2+} ions, cross-linking several pectin chains [6]. PME is active at room temperature; however, an increase in the demethylation rate of nearly 100 times has been seen in green beans and tomatoes when the temperature was raised from 25ºC to 65ºC [48]. At 80ºC the PME activity is significantly reduced [32, 33].

Depolymerisation can occur through two different pathways: β-elimination or acid hydrolysis. The pectin chain is cleaved through β-elimination when the hydrogen at C-5 is removed and a double bond is induced between C-4 and C-5 [49, 50]. This reaction occurs only for esterified galacturonic residues, and increases with increasing DM [51]. Pectin depolymerisation through β-elimination has been demonstrated at temperatures down to 50ºC, however, the rate increases considerably above 80ºC [49]. The rate of β-elimination is also affected by pH: the higher the pH, the higher the rate of depolymerisation [52]. At pH below 4.5, the cleavage of the pectin chain has been shown to be primarily caused by acid hydrolysis [53]. A prerequisite for acid hydrolysis is that the pectin has a low DM (<5%). For pectin with a higher DM β-elimination is the dominant reaction above pH 3.8 [54].

Often, a combination of various kinds of heat treatment is required to make a vegetable product both safe for consumption and appealing to the consumer. An increase in firmness has been seen when first activating PME, before increasing the

temperature to kill pathogens. This was stated as being due to the cross-linking with calcium, increasing the number of bonds between pectin chains in the cell wall [32, 55]. However, it could also be due to the decreased rate of β-elimination occurring with a lower DM [56]. Subjecting fruit and vegetables directly to a high temperature to sterilise or pasteurise them could have detrimental effects on the texture [57, 58].

In the present study, three different kinds of heat treatment were used to study changes compared with a reference sample (Papers III & V). The reference sample was heated to 90ºC for 5 minutes to minimise PME activity. The suspensions were heated for 2 hours at 85ºC to favour β-elimination, at 65ºC for 40 minutes to enhance PME activity, and for 5 minutes at 90ºC to minimise the enzymatic activity. A combination of the two effects was achieved in the last heat treatment, which consisted of heating for 40 minutes at 65ºC, followed by heating at 85ºC for 2 hours. The different kinds of heat treatment are summarised in Table 3.

Table 3. Heat treatment of the fibre suspensions

Heat treatment	Heating pad	Water bath	Objective
HT0	90-95°C, 5 min	-	To minimise the PME activity
HT1	Up to 85°C	85°C, 2 hours	To favour β-elimination.
HT2	-	65°C, 40 min + 90°C, 5 min	To activate PME and then stop PME activity
HT3	-	65°C, 40 min + 85°C, 2 hours	To activate PME then favour β-elimination

From Paper III

The suspensions were heated in glass bottles with the lid semi-closed. The samples heated to 65ºC were heated in a water-bath, while the samples heated to the higher temperatures (85 and 90ºC) were first heated on a heating pad to increase the temperature rapidly in order to minimise the time at lower temperatures where PME is active.

4. Statistical evaluation

Univariate statistical tests, such as analysis of variance (ANOVA), were performed using Minitab (release 14, Minitab Inc., State Collage, Pennsylvania, USA). Both one-way ANOVA and the general linear model were used. Significance between mean values was tested with Student's t-test, and was defined as $p<0.05$. Correlations between variables were tested with the Pearson correlation (r, Minitab). Multivariate statistical analysis was carried out by principal component analysis (PCA) using Unscrambler (release 9.0, Camo Software, Norway).

To fully elucidate the influence of different parameters such as processing and concentration (design variables) on the measured properties, a suitable experimental design must be employed. If any of the design variables has more than two levels a full factorial design must be used [59]. In a full factorial design, all levels of the design variables are combined, i.e. all the design variables are varied in the same experimental study, as can be seen in Fig. 3 for one source of fibre in the study on the influence of homogenisation (Paper II). The results of a full factorial design can then be analysed with a linear model, as well as the interactions between the parameters, with for example ANOVA.

Different full factorial experimental designs were employed for the various parts of the study. In the homogenisation experiments fibre from four different sources were used, at two concentrations and three degrees of homogenisation, rendering 4x2x3=24 samples (Papers II & IV). Four different kinds of heat treatment were carried out on fibre suspensions made from three fibre sources: 3x4=12 samples (Paper III). The fibre additive in the sausage study was subjected to four different kinds of heat-treatment, and the suspension was homogenised or not: 4x2=8 samples (Paper V).

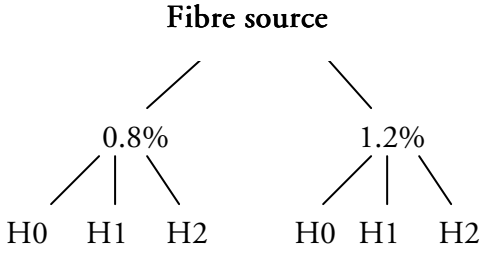

Figure 3. Full factorial design with one fibre source, two concentrations and three degrees of homogenisation

When analysing the homogenised samples with the general linear model (Paper II) the model parameters were degree of homogenisation, concentration and fibre source. Another model was constructed for each fibre source with the degree of homogenisation and concentration as parameters. Least-square means were obtained for the different parameters from the general linear model. Least-square means are within-group means appropriately adjusted for the other effects in the model.

To study the influence of fibre source and the various kinds of heat treatment on the physicochemical properties and the pectin content and solubility of the fibre suspensions, a general linear model was used (Paper III). The influence of heat treatment on each fibre source was also studied (one-way ANOVA).

ANOVA was carried out in the general linear model with degree of homogenisation, concentration, panel members and replicates as design parameters for each fibre source and sensory attribute analysed in the descriptive analysis (Paper IV). To correlate the sensory analysis with the physicochemical properties and the composition of DF, principle component analysis (PCA) was performed. The category variables were degree of homogenisation, fibre source and concentration.

When studying the influence of fibre addition on the low-fat sausage one-way ANOVA was used (Paper V). The sausages containing potato pulp were then further studied with the general linear model to check for significant effects of the processing (homogenisation and heat treatment, as well as their interaction). PCA was performed to study the correlation between the sensory properties of the sausages.

5. Characterisation of dietary fibre content and composition

The composition of DF can change during processing. For example, redistribution of insoluble to soluble fibre has been seen when blanching carrots at 98-100ºC for 1-3 minutes [26]. A change in composition can, in turn, affect the physicochemical properties [60]. Thus, it is important to measure the DF content in order to explain observed changes in fibre suspensions after processing.

There are several methods of measuring the DF content [61-65]. In the present work the Uppsala method as described by Theander et al. was chosen [63], where the different monosaccharides originating from cellulose, hemicellulose and pectin were quantitatively analysed using gas chromatography (GC). Galacturonic acid, found in the pectic backbone, was determined by colorimetry (Paper I-III). Lignin was not quantified in the present study, mainly because of its low content in fruit and vegetables [63].

The effects of processing such as homogenisation on the viscosity of a suspension will differ, depending on whether the suspension consists of mainly soluble or insoluble DF [38, 43]. Therefore, it has been suggested that the DF be divided into two fractions depending on its solubility in water, to provide a better understanding of the differences in the material [66]. A limitation of the Uppsala method is that the soluble and insoluble components are not separated. However, fractionation can be carried out before sample preparation for GC, allowing the soluble and insoluble components to be analysed separately. It is thus imperative that the analytical method does not influence the solubility of the fibre, by heating or enzymatic treatment of the suspensions, before separation into soluble and insoluble fractions [5]. In this study, a mild separation method with minimal influence on the solubility was used (Paper I). The samples were mixed with water to form a suspension containing 2% dry matter

of the fruit or vegetable, and were stirred overnight at 7ºC. The solution was centrifuged at 3000 *g* for 20 minutes. If the pellet was not totally separated from the supernatant, the solution was filtered using water suction and a 1F filter (Munktell, Sweden). The two fractions, soluble material (SM) and insoluble material (IM), were then freeze-dried until they contained less than 15% moisture, and these fractions were used for further characterisation of the insoluble and soluble DF. The analytical process is summarised in Fig. 4.

Pectin with a high DM (>50%) is considered high-methoxy (HM) pectin, whereas in low-methoxy (LM) pectin less than 50% of the carboxyl groups are esterified [67]. These two different groups of pectin, HM and LM, have essentially different gelling mechanisms (Fig. 5). Generally, HM pectins form gels by hydrophobic interactions with methyl esters in the presence of sugar at a low pH, while LM pectin chains are linked to each other by Ca^{2+} interactions at the carboxyl group [6, 67]. However, it has also been shown that HM pectin, up to a DM of 80%, can gel through electrostatic interactions with calcium ions [68].

The DM of the pectic backbone was determined with spectrophotometry after removing the methyl group by saponification [69] and then converting the methanol to formaldehyde using alcohol oxidase (Paper II). The formaldehyde was coloured through a reaction with Purpald using the methodology described by Anthon and Barrett [70] and Diaz et al. [53] with a minor modification in that citrate buffer was used (pH 6.5, 100 mM) instead of phosphate buffer. The amount of methanol in the original sample was determined by comparison with a methanol calibration curve at 550 nm. The DM was calculated as a percentage of the amount of galacturonic acid in the sample.

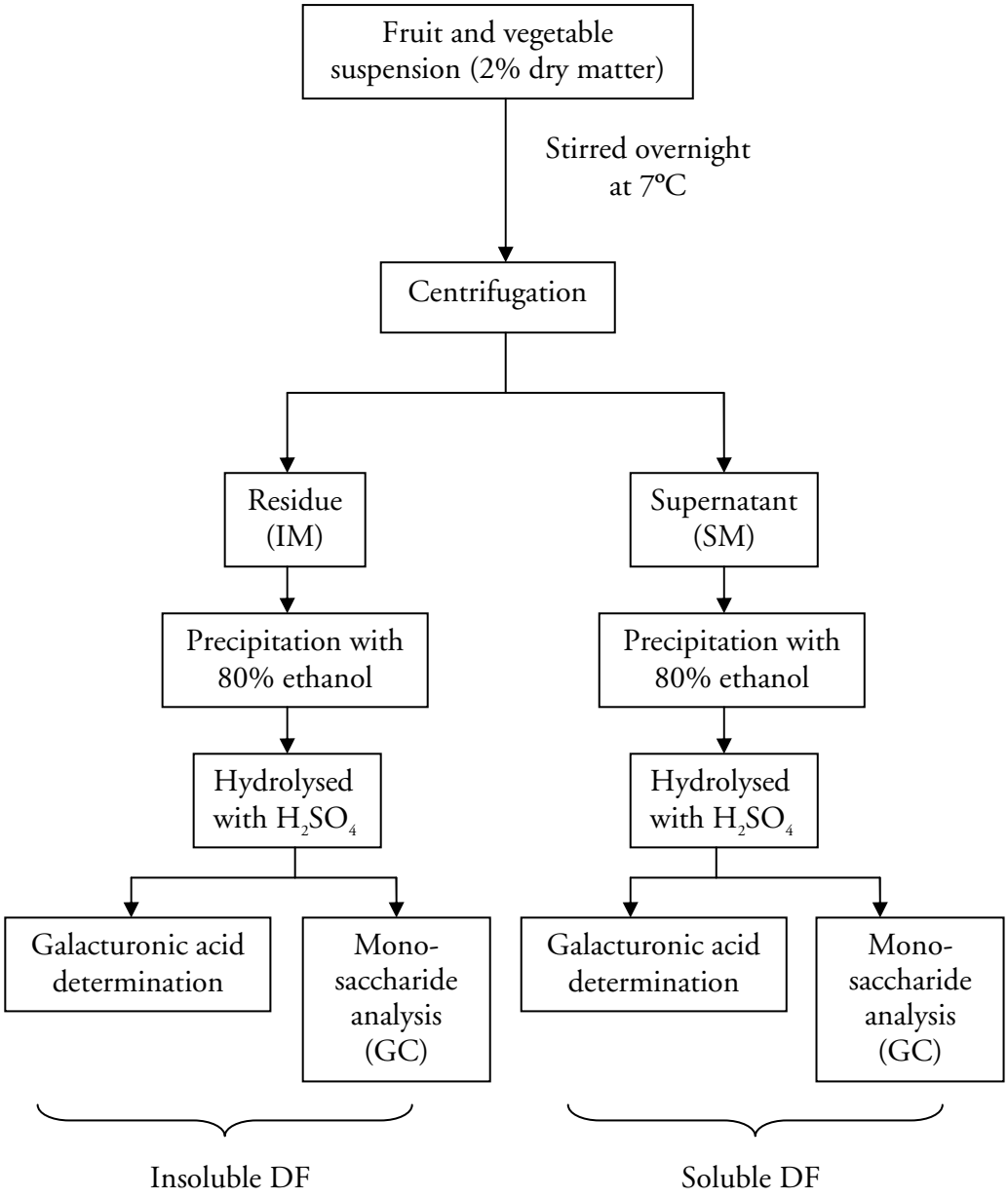

Figure 4. Illustration of the analysis of DF using the Uppsala method

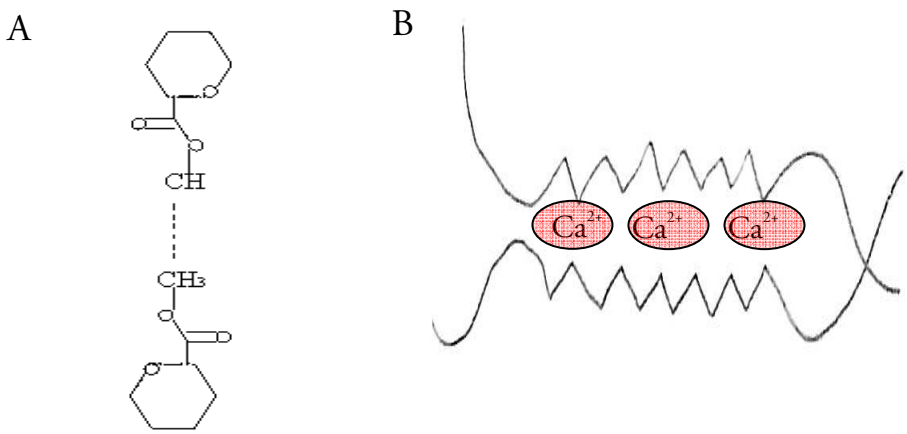

Figure 5. Gelling mechanism of pectin: A) hydrophobic interactions, B) Ca^{2+} interactions

5.1 Influence of homogenisation

The difference between the four DF sources used in the present study becomes apparent when studying Fig. 6, where the contents of soluble and insoluble DF are shown. Potato pulp stands out from the other DF sources due to its high content of insoluble DF. This is because the potato pulp is a by-product of starch manufacturing, and most of the soluble components, including the starch, have been removed by repeated washing. The total DF content in the other three sources is similar; however, apple and tomato contain considerably higher amounts of soluble DF than carrot.

The change in total DF with degree of homogenisation was not significant for any of the DF sources. There was a minor, but significant, increase in soluble DF in the two homogenised potato pulp samples compared with the non-homogenised sample. A small increase in insoluble DF was seen in the homogenised tomato samples compared with the unprocessed sample.

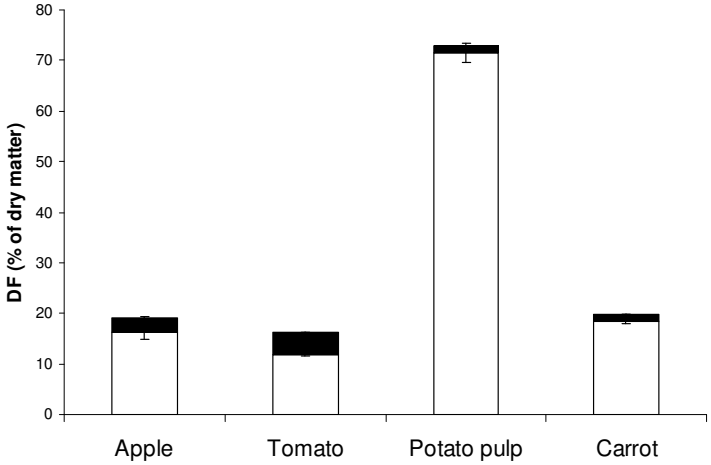

Figure 6. Soluble (shaded) and insoluble (unshaded) dietary fibre in four DF sources

The total contents of DF in the materials studied corresponded well to the amounts previously reported for apple [63], carrot [63, 71], potato pulp [63, 72, 73], and tomato [74]. However, compared with other studies the soluble DF was considerably lower in the present study [26, 72, 74, 75]. A decrease in solubility has been seen in other studies with decreasing analytical temperature. For carrot, the soluble DF has been seen to vary from 2.8% to 11.0% at temperatures ranging from 38ºC to 100ºC [26, 61, 76]. Since the temperature used in the present study during the mild separation was even lower (7ºC), the low content of soluble DF of 1.5% could be expected. The solubility of DF is also pH- and buffer-dependent [77, 78]. This may be another reason for the differences between the present results and those of other studies, in which buffers were used.

The main part of the soluble DF originates from the pectic backbone (galacturonic acid). The content of insoluble pectin is noticeably higher than the soluble equivalent in potato pulp, apple and carrot (Fig. 7). Only in tomato are there similar amounts of galacturonic acid in the two fractions. In the potato pulp and apple samples, the soluble pectin increased somewhat at the highest degree of homogenisation (from 1.7

to 1.8% for apple and from 0.3 to 0.4% for potato pulp). The change in the content of soluble and insoluble pectin was not significant in the other samples.

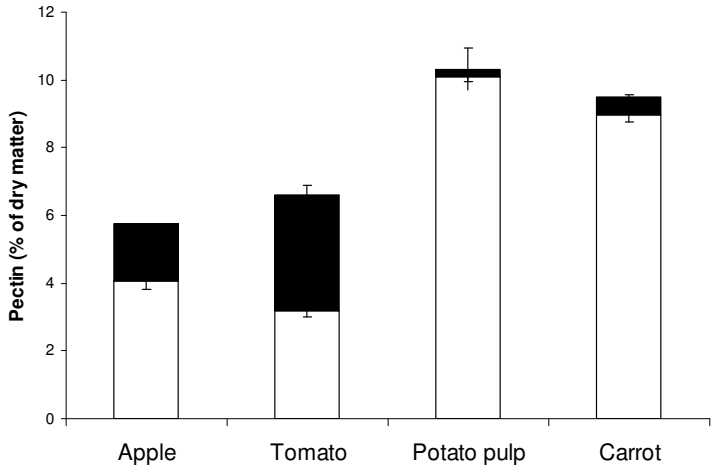

Figure 7. Soluble (shaded) and insoluble (unshaded) pectin in the four DF sources

The degree of methylation of the insoluble pectin differed considerably between the different sources. Only about 25% of the carboxyl groups in the apple fibre was methylated, which means that the pectin is a LM pectin. Carrot fibre, on the other hand, is a HM pectin, as about 90% of the carboxyl groups was esterified. In tomato and potato pulp fibre almost half of the galacturonic acid monomers were esterified (~ 40%). The variation in DM in relation to the degree of homogenisation for the different fibre sources was not significant, with the exception of potato pulp, where the non-homogenised sample had a significantly higher DM than the homogenised samples (58%, versus 43% and 41% for H0, H1 and H2, respectively). Homogenisation thus appears to have only a minimal effect on the DM. However, a significant difference in DM was seen between the fibre sources (ANOVA, $p<0.001$).

5.2 Influence of heat treatment

Since the component of DF most affected by heat treatment is pectin, it was decided to analyse only the changes in pectin content when investigating the effect of heat treatment (Paper III). Since none of the samples had a DM below 5, it was assumed that β-elimination was the prevailing mechanism for depolymerisation at the higher temperature [53, 54].

No general trend for the content of insoluble pectin was observed regarding heat treatment when all fibre sources were analysed. However, when analysing each fibre source separately heat treatment had a significant influence ($p<0.01$), but the trend was different for the different fibre sources.

Statistical analysis by one-way ANOVA showed that the content of insoluble pectin was significantly influenced by fibre source ($p<0.001$), as was seen in the homogenised fibre suspensions. The heat treatment favouring β-elimination (HT1 and HT3) caused solubilisation of the pectin in apple and carrot suspensions (Fig. 8). There was a significant difference between HT0 and HT1 in both insoluble and soluble pectin content in apple and carrot suspensions, with a drastic increase in soluble pectin. Following the heat treatment where both low and high temperatures were used (HT3), a difference was seen between apple and carrot regarding the insoluble pectin content. Since carrot pectin has a higher DM than apple pectin, it may have been more affected by the low heat treatment promoting PME activity. Thus, a significant decrease in depolymerisation and subsequent solubilisation due to β-elimination is seen in the carrot samples, while the apple samples, with a considerably lower DM, showed no change in depolymerisation between HT1 and HT3. The insoluble pectin content of potato pulp was affected differently by heating, compared with the other two fibre sources studied. Here, no difference was seen

between HT0 and HT1. However, there was an unexpected decrease in insoluble pectin after HT2 and HT3.

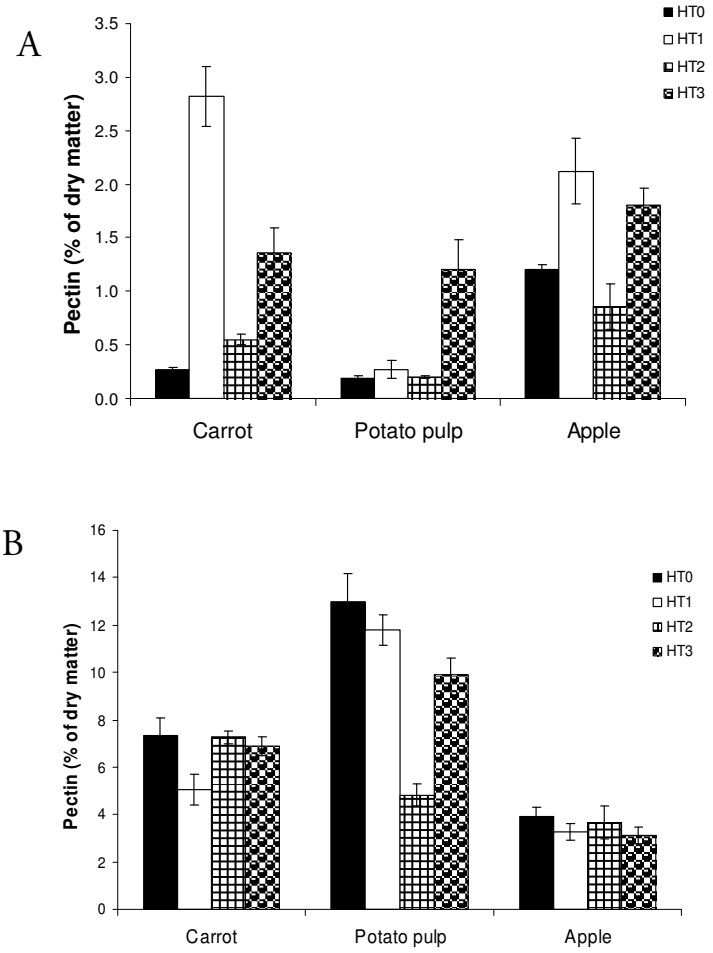

Figure 8. Pectin content in heat-treated fibre suspensions. A) Soluble and B) insoluble pectin (see Table 3 for explanations of the heat treatment).

The reference treatment, HT0, and the treatment favouring PME activity, HT2, led to a lower content of soluble pectin in carrot and apple than HT1 and HT3, where β-elimination was favoured ($p<0.001$). Soluble pectin in the potato pulp samples showed a small change; the content was only increased in the sample exposed to

HT3, while the contents of soluble pectin in the carrot and apple samples were significantly different from those in the reference samples following all other kinds of heat treatment. This shows that all kinds of heat treatment increased the soluble pectin content in carrot and apple, but it is substantially more difficult to solubilise potato pectin.

When considering the composition and content of DF before and after the two processes studied, homogenisation and heat treatment, it was found that the changes were small compared with the vast differences between the original compositions of the different DF sources.

6. Influence of processing on physicochemical properties

When fibre suspensions are processed, changes can occur in the physicochemical properties. These changes originate from modification of both the chemical composition and the physical state [79]. These changes are complex and not yet fully understood [16]. Some important physicochemical parameters of fibre suspensions are the microstructure and the rheological properties and water-holding capacity [16, 79].

To measure the physicochemical properties, the DF sources were suspended in either a pectin solution (Papers I & II) or water (Paper III). The pectin used was a LM citrus pectin (Pectin Classic CU 701, Herbstreith & Fox KG, Neuenbürg, Germany) at a concentration of 2% (dry matter) in water. DF suspensions with pectin as the continuous phase were also used for sensory evaluation (Paper IV) (Section 7.1), and mixtures containing salt, sugar and vinegar were prepared to prolong shelf-life and increase palatability (Table 4).

Table 4. The recipes for the various fibre suspensions used in sensory analysis

	Conc. IM (%)	Fruit/ vegetable purée (g)	Added Water (g)	Salt (g)	Sugar (g)	Vinegar essence (12%) (g)	Pectin (2% gel in water) (g)
Apple	0.8	120.9	-	-	-	-	345.0
	1.2	120.9	-	-	-	-	193.3
Tomato	0.8	55.9	4.1	5.0	30.0	15.0	390.0
	1.2	55.9	4.1	5.0	30.0	15.0	223.3
Potato pulp	0.8	5.1	44.9	5.0	30.0	15.0	400.0
	1.2	5.1	44.9	5.0	30.0	15.0	233.3
Carrot	0.8	103.4	6.6	2.5	20	13	354.5
	1.2	103.4	6.6	2.5	20	13	187.8

6.1 Microstructure

A change in particle size affects the physicochemical properties of a fibre suspension [30, 41, 80]. Not only the size is important, but the shape and degree of aggregation also affect the properties. Light microscopy and laser diffraction have been shown to be two useful, complementary tools when studying changes in particle microstructure [41]. These techniques were used in the present work to study the morphology and size distribution of the particles.

At a magnification of 20x large conformational features can be elucidated, such as the degree of aggregation of the fibre particles (Fig. 9A). Increasing the magnification to 50x gives a more detailed view, and each particle can be observed (Fig. 9B).

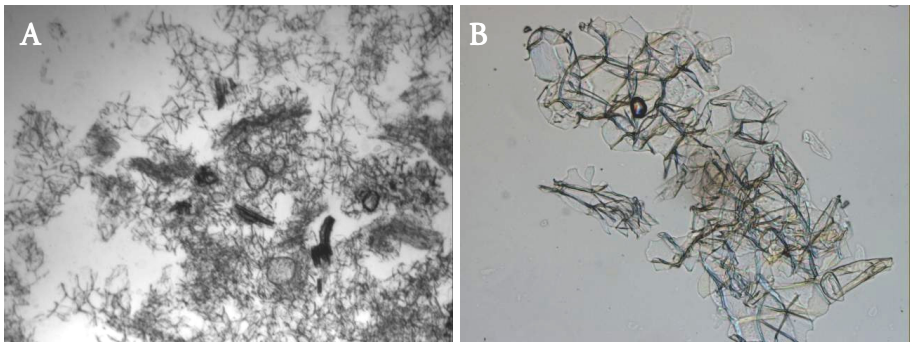

Figure 9. Microscopic images of potato pulp at A) 20x magnification and B) 50x magnification

The morphology of the fibre suspensions differed. Apple and tomato suspensions consisted mainly of whole, single cells and cell fragments, as can be seen in Fig. 10A and B, while carrot and potato pulp suspensions consisted of small cells arranged in large clusters (Fig. 10C and D) (Paper I). The potato pulp was dried prior to suspension, and it can be seen that it had a highly aggregated structure compared with the other DF sources, which were not dried.

The particle size distribution (PSD) of the insoluble part of fibre suspensions can be studied by laser light diffraction. The laser light is scattered by the particles in a dilute solution, resulting in a distinctive diffraction pattern. This pattern is then transformed into the size distribution using optical methods such as Fraunhofer diffraction [81]. Since this method is most suitable for spherical particles, the results should only be regarded as an estimate of the actual particle size, since the fibre suspensions studied here consist of cylindrical cells, fibrous particles and cell clusters. The Fraunhofer method has, however, been used previously to measure particle sizes in tomato dispersions [41, 43].

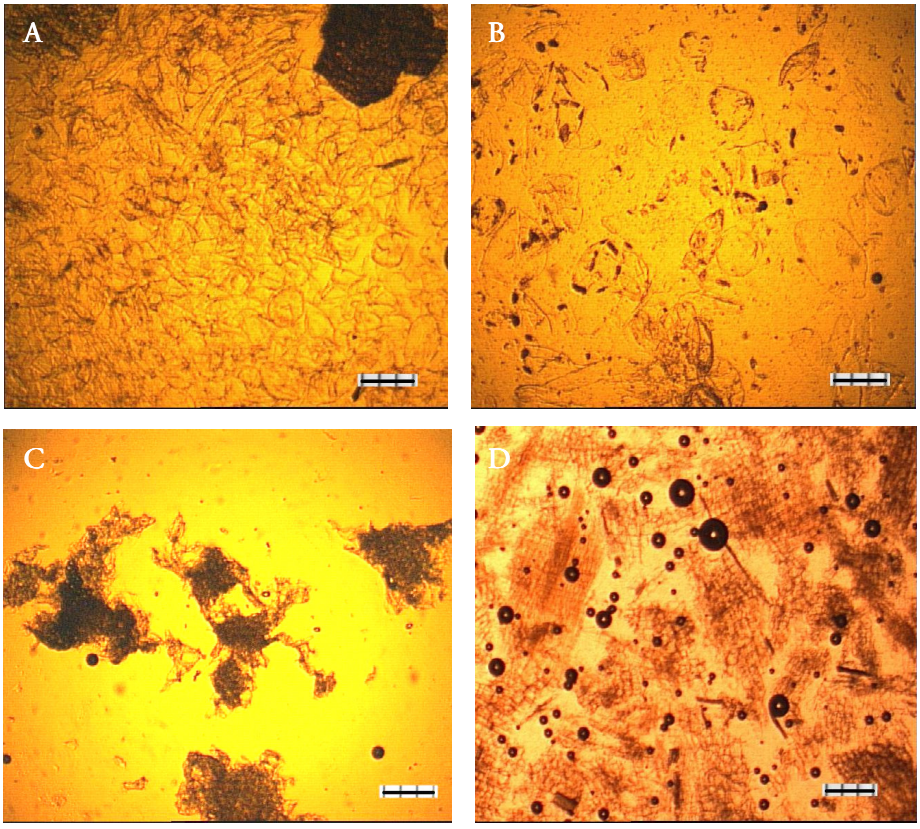

Figure 10. Micrographs of A) apple, B) tomato, C) potato pulp and D) carrot suspensions at a magnification of 20x. The scale bar is 300 µm (from Paper I).

The apparatus used in the present study, a Coulter LS130 particle analyser (Beckman Coulter, High Wycombe, UK), can measure particle sizes in the range 0.1-900 μm. Particles larger than 900 μm are therefore not included in the calculations of the mean diameter. This has a noticeable effect on the volume-weighted PSD where the distribution is truncated just after the maximum peak, whereas for the surface-area-weighted distribution a less truncated distribution was seen (compare Fig. 11A and 12A). There is also a considerable difference between the fibre sources: the potato pulp PSD curve has a cut-off at 7.6 volume % while the curve for tomato suspension is truncated at 0.5 volume % (Fig. 11A and B).

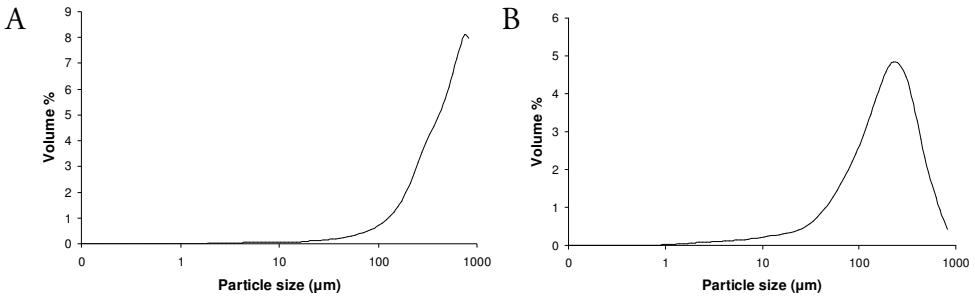

Figure 11. Volume-weighted particle size distribution of fibre suspensions of A) potato pulp and B) tomato

The PSDs of fruit and vegetable suspensions are polydispersed. A mean diameter is usually calculated, either a volume-weighted diameter (d_{43}) or a surface-area-weighted diameter (d_{32}):

$$d_{32} = \frac{\sum_i n_i d_i^3}{\sum_i n_i d_i^2} \quad [\mu m] \quad (1)$$

$$d_{43} = \frac{\sum_i n_i d_i^4}{\sum_i n_i d_i^3} \quad [\mu m] \quad (2)$$

where n_i is the percentage of particles of diameter d_i in each size class [82]. The largest particles in the suspension have the greatest influence on the volume-weighted mean diameter, while the smaller particles also play a roll in the surface-area-weighted mean diameter. For fruit and vegetable suspensions a bi- or trimodal distribution is often seen for the surface-area-weighted PSD (Fig. 12). A way to describe the PSD is to calculate the surface-area-weighted mean diameter for two fractions: the small fraction below 100 µm and the large fraction above 100 µm, by integrating the PSD curve between 0.1 and 100 µm, and 100 and 900 µm (Papers I & II). These values, d_{32} (s) and d_{32} (l), show the influence of small and large particles, respectively, to an even greater degree than d_{32} and d_{43}.

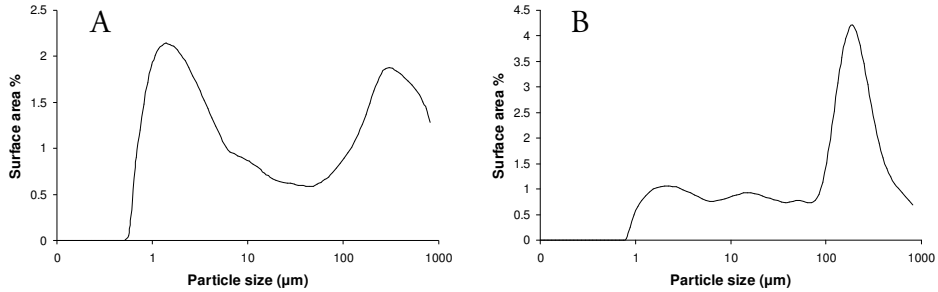

Figure 12. Surface-area-weighted particle size distributions of fibre suspensions. A) mainly bimodal distribution of potato pulp particles and B) mainly trimodal distribution of apple particles.

6.1.1 Changes due to processing

As expected, there was a significant decrease in the mean diameter following homogenisation, for both d_{43} and d_{32} (Paper II) (Table 5). After homogenisation a greater decrease was seen in d_{32} of the tomato suspensions (48%) than the potato pulp suspensions (4%). Micrographs of the fibre suspensions showed that the large aggregates in the potato pulp suspension (Fig. 10C) were broken down by homogenisation (Fig. 13) rendering rather large, non-aggregated cell clusters. In the tomato suspension, the cells were broken down into smaller cell fragments, leading to

a considerable decrease in d_{32}. Carrot and apple suspensions were also affected to different degrees by homogenisation. The cell clusters seen in the unprocessed carrot were somewhat smaller after homogenisation, although clusters remained. Although there was a decrease in the mean diameter of particles in the apple suspensions following homogenisation (Table 5), little can be deduced from the micrographs (Fig. 13).

Table 5. The least-square mean diameters of different PSD variables as a function of degree of homogenisation. Statistically significant differences are indicated by the asterisks.

	d_{32} (µm)	d_{43} (µm)	d_{32} (s) (µm)	d_{32} (l) (µm)
Apple	***	***	***	***
H0	164.5	358.6	20.6	279.1
H1	129.0	230.0	30.7	207.6
H2	114.6	196.2	33.1	190.8
Tomato	***	***	***	***
H0	158.7	356.0	22.1	300.0
H1	82.4	273.7	20.5	244.5
H2	62.9	229.8	19.8	224.1
Potato pulp	*	***	***	***
H0	162.4	473.1	16.4	383.3
H1	155.7	340.8	23.8	298.9
H2	139.6	289.4	25.5	264.3
Carrot	***	***	***	***
H0	182.9	447.1	15.2	362.3
H1	138.3	281.1	25.0	246.3
H2	122.0	226.7	29.5	213.0

* $p<0.05$, ** $p<0.01$, *** $p<0.001$
See Table 2 for explanations of H0, H1 and H2.

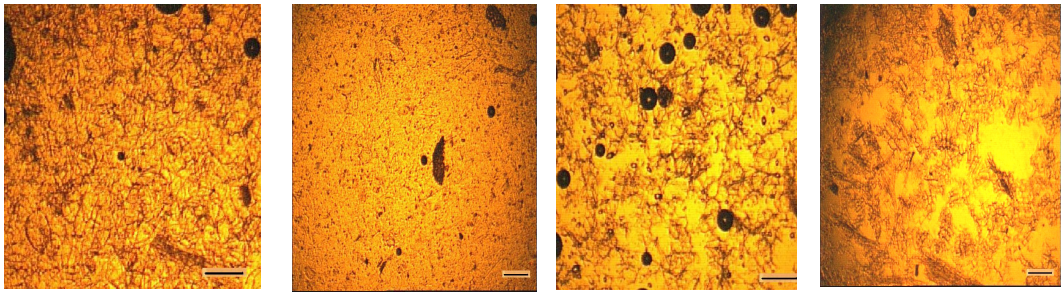

Figure 13. Micrographs (20x) of highly homogenised (H2) fibre suspensions. The scale bars are 300 µm (From Paper II)

The d_{32}(l) follows the same pattern as d_{43}, decreasing with degree of homogenisation (Table 5). This was anticipated since d_{43} mainly reflects the influence of the large particles. An increase in the mean value of small particles was observed in apple, potato pulp and carrot suspensions (Table 5). This could be due to the aggregation of the small particles in these suspensions. An increase in d_{32}(s) was, however, not seen in the tomato suspensions, indicating no aggregation of the small tomato particles.

The difference in the effects of homogenisation on the microstructure could be due to the difference in pectin content and solubility. A large proportion of the insoluble pectin is found in the middle lamella, where it acts as a glue between adjacent cells. For the fibre sources containing high amounts of insoluble pectin, such as carrot and potato pulp suspensions, the morphology consists of cell clusters, even after homogenisation, whereas tomato suspensions, containing lower amounts of insoluble pectin, are more easily degraded by homogenisation. The soluble pectin may prevent the aggregation of the small particles, since no aggregation of the small particles was seen in the tomato suspensions, which contain high amounts of soluble pectin. This was not the case in the three other fibre sources studied. Although homogenisation did not change the proportions of soluble or insoluble pectin significantly in three out of four of the fibre suspensions, the original contents could still be a major factor in

determining the extent to which different fibre sources are degraded by homogenisation.

Table 6. Surface-area-weighted mean (d_{32}) and volume-weighted mean (d_{43}) particle sizes of the differently heat-treated fibre suspensions

	d_{32}	d_{43}
Carrot		
HT0	203.8 ± 0.30[a]	500.2 ± 2.32[a]
HT1	201.2 ± 3.35[abc]	486.1 ± 1.22[b]
HT2	203.4 ± 0.34[b]	494.9 ± 1.55[c]
HT3	199.4 ± 1.01[c]	486.1 ± 2.17[c]
Apple		
HT0	198.7 ± 1.92[a]	486.9 ± 10.84[a]
HT1	185.3 ± 2.13[b]	426.2 ± 6.74[b]
HT2	199.8 ± 1.74[a]	484.2 ± 5.80[a]
HT3	188.1 ± 6.79[b]	427.6 ± 20.64[b]

See Table 3 for explanations of the heat treatment.
The letters a-c in the same column for each fibre source indicate significant differences ($p<0.05$).
From Paper III

Heat treatment caused a less pronounced effect on the PSD of carrot and apple suspensions than did homogenisation (Paper III). A decrease in the mean diameter was seen in the samples subjected to high temperatures, promoting β-elimination (Table 6). Depolymerisation of insoluble pectin, due to β-elimination, could cause the break-up of cell clusters since insoluble pectin is found in the middle lamella, connecting the cells. There were, however, no visible morphological changes in the fibre network of carrot and apple suspensions.

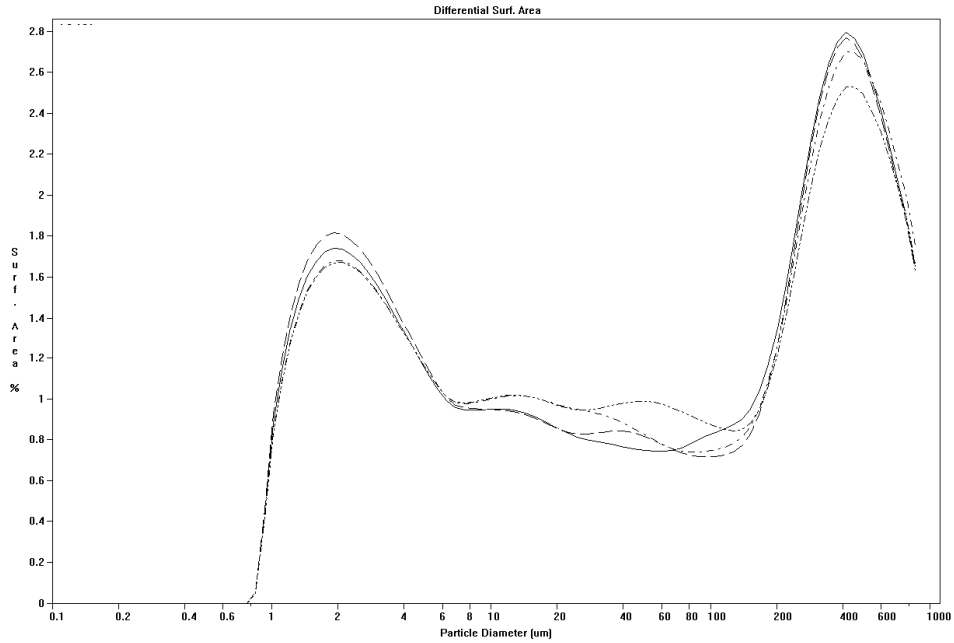

Figure 14. The surface-area-weighed particle size distributions of the differently heat-treated suspensions: HT0 (—), HT1 (– –), HT2 (– - –) and HT3 (– - - –) (see Table 3 for explanations). From Paper III.

A change in the microstructure can be observed in both the PSD curves (Fig. 14) and in the micrographs (Fig. 15) of the heat-treated potato pulp suspensions. This change originates from the starch remaining in the potato pulp (12-15% according to the manufacturer). The swelling of the starch granules with heating increases the mean diameter of the insoluble particles in the suspensions. At elevated temperatures, the starch will swell to a higher extent, but with prolonged heating amylose will start to leak out of the structure, decreasing the size of the swollen starch granules [83], thus reducing the mean diameter of the suspensions following HT1 and HT3 compared with HT2. However, all kinds of heat treatment increased the d_{43} and decreased the d_{32} of the suspensions, compared with the reference heat treatment (HT0) ($p<0.001$). The fibre network of potato pulp suspensions seems not to be changed significantly by heating, as deduced from the micrographs (Fig. 15).

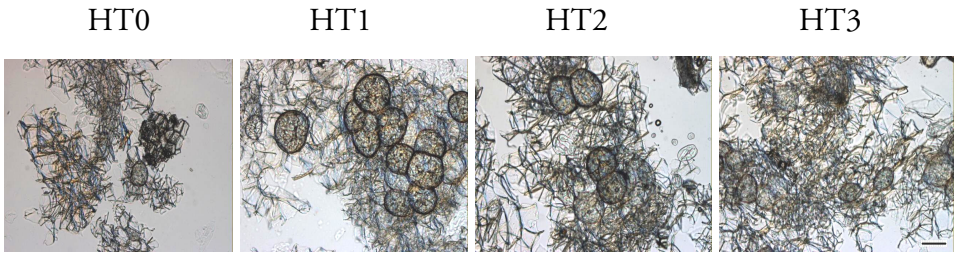

Figure 15. Micrographs of the differently heat-treated samples of potato pulp suspensions. For explanations of the heat treatment, see Table 3. The bar corresponds to 100 µm. From Paper III.

6.2 Rheological properties

When fibre suspensions are to be used as additives in food products, a strong network is often required to produce the optimal texture. The properties of the viscoelastic network in a fibre suspension can be determined by rheological measurements. Previously, the viscosity originating from steady shear measurements was the parameter measured. However, in recent publications measurements of parameters arising from dynamic oscillatory forces, such as G′ and G″ (the elastic and viscous modulus, respectively) and tan δ (the phase angle, G″/G′) are encountered more often [8, 41, 84]. These properties better describe a semi-solid food, e.g. a gel, since the viscosity of most foods shows the same trend: it decreases with increasing shear rate. Measurements using steady shear often cause breakdown of the network structure, while measurements under smaller, oscillatory forces can provide information on network properties, such as the number of junction points, the shear force required for gel breakage, and uniformity in bond strength [85, 86]. When G′ exceeds G″ (tan $\delta<1$) the viscoelastic suspension has essentially solid-like properties, i.e. the deformation when subjected to a shear force is mainly recoverable [87]. When fibre suspensions are used as a texturizer in food products, high elastic properties are usually required.

The magnitude of G′ and G″ is dependent on the frequency, temperature and the applied stress or strain [8]. It is important to minimise the structural damage to the network during measurements. Therefore, a constant frequency of 1 Hz was used to measure the rheological parameters in the present study (Papers I-III). It has been shown that the frequency dependency at low frequencies is linear for several fibre suspensions, with only a slight increase in elastic modulus [29, 30, 43, 88]. Since the purpose of this study was to compare the different fibre sources, and the effects of concentration and processing, it was decided to only carry out the less time-consuming stress sweeps. The temperature was maintained at 20ºC to minimise the effects of extrinsic factors.

A rheogram, such as that shown in Fig. 16, was obtain through dynamic oscillatory stress sweeps from 0.1 to 100 Pa, using a controlled-stress StressTech rheometer, (Reologica AB, Lund, Sweden). The vane geometry was used to prevent sedimentation and slippage problems [8]. Values of G′ and G″ in the linear viscoelastic region (LVER), i.e. where the moduli are independent of stress, were determined from the rheogram, together with the yield stress (shear stress at the end of the LVER) and the shear stress when G′ had decreased to half the magnitude measured in the LVER.

6.2.1 Elastic modulus

The elastic modulus was greater than the viscous modulus for all the fibre sources studied (Paper I-III), implying that the properties of the suspensions were more elastic than viscous, i.e. some sort of fibre network had been formed.

The change in elastic modulus resulting from homogenisation was considerable. An increase of approximately 2.5 times was seen in the tomato, potato pulp and carrot suspensions following the lower degree of homogenisation, H1. The difference

between the effects of H1 and H2 was small for these fibre suspensions. No increase in the elastic modulus was seen in the apple suspensions following H1, and the increase after H2 was not as pronounced as for the other suspensions. Heat treatment, in contrast, had only minor effects on the elastic modulus of the fibre suspensions.

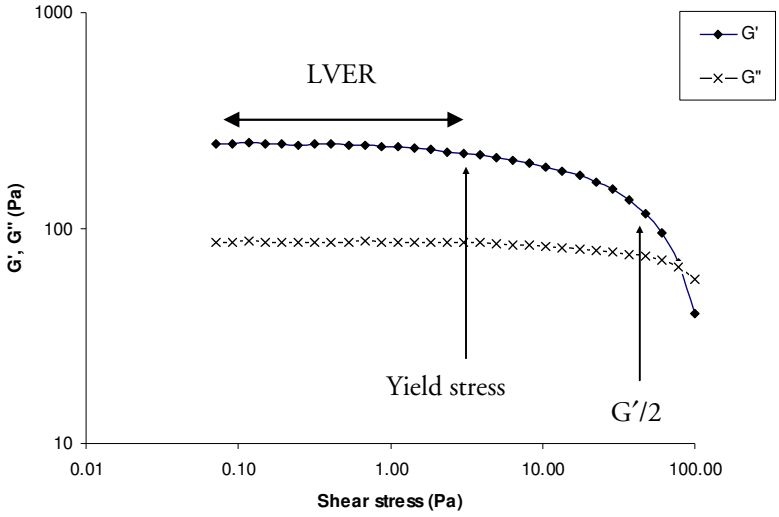

Figure 16. Rheogram of a 1.2% potato pulp suspension.

Different concentrations of the insoluble material in the fibre suspensions were investigated. At the lower concentrations of 0.8 and 1.2% IM, the continuous phase consisted of a soluble pectin dispersion (2% dry matter) (Papers I-II), while for the higher concentration used in the heat treatment study (3% IM) the fibre sources were mixed with water (Paper III). The potato pulp suspensions also differed in that at the lower concentrations the potato pulp was dried and aggregated potato pulp was used, while at 3% the pulp was not dried prior to suspension. Drying of plants has been shown to dramatically affect the rheological properties when re-dispersed [89]. There was, however, a trend towards an exponential increase in the elastic modulus with increased concentration (Fig. 17). The exponential fit was good for both apple (r^2=0.96) and carrot (r^2=0.93). The somewhat poorer fit for the potato pulp (r^2=0.90)

may be explained by the difference in material used for the various concentrations, as explained above. The linear fit for tomato is, however, poor (r^2=0.32). A possible explanation of this can be found when examining the elastic modulus of the tomato samples more closely. The elastic modulus of the tomato suspensions was only measured for the two lower concentrations in the part of the study where the effect of homogenisation was investigated. As stated above, homogenisation increases the elastic modulus considerably at both concentrations. Thus, no clear relation was found between concentration and G′ as the dominating factor is homogenisation. For the other sources of DF, however, concentration seemed to be the dominating factor since the elastic modulus was also measured for the higher concentration (3%).

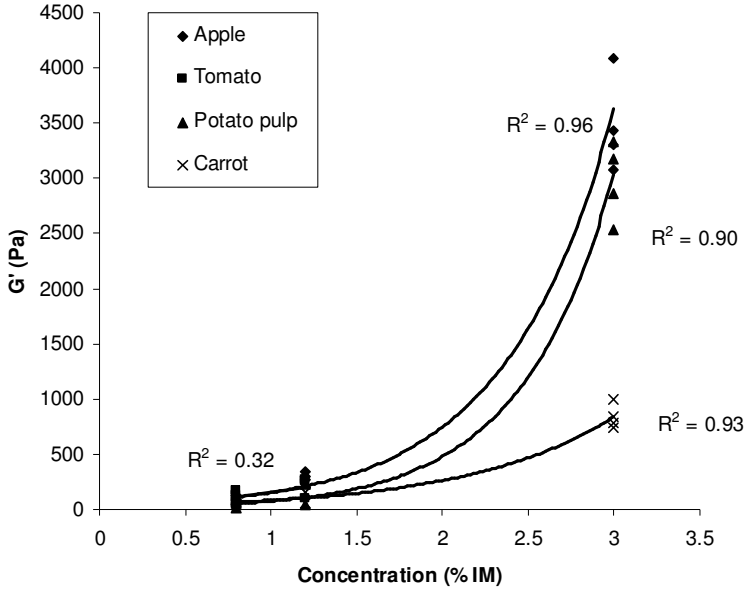

Figure 17. The effect of concentration on the elastic modulus of dietary fibre from different sources.

The elastic modulus varied with the source of the fibre and the increase with fibre concentration was also source dependent. The untreated apple and tomato fibre suspensions at a concentration of 0.8% have approximately the same elastic modulus

(Paper I). However, at the higher concentration (1.2%) the elastic modulus of the apple suspension was about three times higher than that of the tomato suspension, despite the fact that the elastic modulus of the tomato suspension was doubled at the higher concentration. Substantial differences can be seen in the concentration dependencies of the elastic modulus between apple and carrot suspensions, in that the dependence for apple is much steeper than that for carrot (Fig. 17). The increase in elastic modulus in potato pulp suspensions differs slightly from the other fibre sources studied in that at the low concentration potato pulp suspensions had a lower elastic modulus than carrot, while at the highest concentration used in this study (3%) it approached the elastic modulus of the apple suspensions. This is probably due to the fact that the potato pulp was dried before being suspended for the measurements at the lower concentrations, while this was not the case at the higher concentration. The carrot suspensions consistently exhibited the lowest elastic modulus, while the apple suspensions exhibited the highest.

6.2.2 Yield stress and shear stress at G′/2

In this work, yield stress was defined as the shear stress at the end of the LVER, i.e. at the point where the suspension breaks. If a high shear stress is required to break the suspension, the yield stress is high. The shear stress when the elastic modulus has decreased to half its initial value is a measure of how brittle or ductile the suspension is. If the decrease in G′ occurs over a broad range of shear stresses, giving a high value of shear stress at G′/2, the suspension is considered ductile. The ductility/brittleness of a suspension is thought to depend on the number of different strengths of the bonds within the network. The more uniform the strength of the bonds, the lower the shear stress required to break the gel [85].

The effect of processing on the yield stress of the apple, carrot and potato suspensions was negligible. Neither heat treatment, nor homogenisation changed the initial values

significantly. However, the yield stress of the homogenised tomato suspension increased considerably with increasing degree of homogenisation (Fig. 18). Heat treatment had no significant effect on shear stress at G′/2 (Fig. 19); however, homogenisation increased the shear stress at G′/2 for all fibre sources studied, except for carrot suspensions.

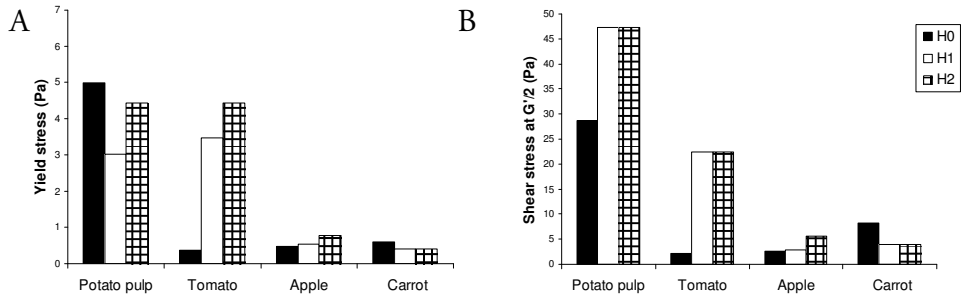

Figure 18. Yield stress (A) and shear stress at G′/2 (B) of the four fibre suspensions at an IM concentration of 1.2% following homogenisation (see Table 2 for explanations)

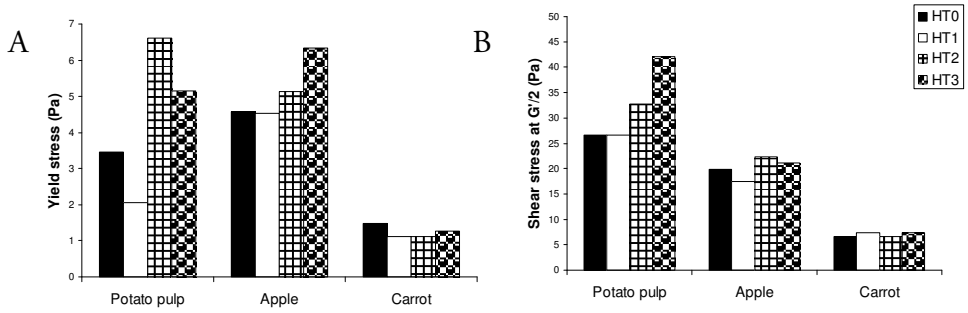

Figure 19. Yield stress (A) and shear stress at G′/2 (B) of three fibre suspensions at an IM concentration of 3% following heat treatment (see Table 3 for explanations)

Regarding the effect of concentration, only minor changes in yield stress and shear stress at G′/2 were observed with increasing concentration of the carrot, tomato and potato pulp suspensions, implying that the strength and type of bonds are similar at different concentrations and with different dispersion media (0.8 and 1.2% IM in

soluble pectin (Papers I & II), compared with 3% IM in water (Paper III)). However, a different pattern was seen for the apple suspensions, namely an increase in both yield stress and shear stress at G′/2 with concentration (Fig. 18 and Fig. 19). At the lower concentrations apple suspensions were the most brittle under shear stress. However, at 3% IM the yield stress and the shear stress at G′/2 were almost as high as in the potato pulp suspensions, the most ductile of the suspensions studied.

6.2.3 Elastic modulus of the continuous phase

In part of the study the concentration of IM was either 0.8 or 1.2%, dispersed in a 2% pectin gel. However, the composition of the continuous phase will differ since the fruits and vegetables used have varying concentrations and contents of soluble material. Therefore, the rheological properties of the continuous phase were measured (Papers I & II).

To measure the elastic modulus of the continuous phase of the fibre suspensions, the samples were centrifuged at 50 000 *g* for 20 minutes (Beckman Optima, Le-80K Ultra Centrifuge, Fullerton, California, USA) and measurements were made on the supernatant using a concentric cylinder cell (CC15). Dynamic oscillatory stress sweeps were made from 0.1 to 100 Pa with the StressTech rheometer mentioned above.

The continuous phase of the apple suspensions had the lowest elastic modulus, while that from potato pulp had the highest, perhaps due to the presence of soluble starch granules in the solid matter (Fig. 20). Concentration and homogenisation had no significant effect on the elastic modulus of the continuous phase for apple, carrot and potato pulp suspensions. The elastic modulus of the continuous phase of tomato suspensions was, however, highly affected by both concentration ($p<0.001$) and homogenisation ($p<0.01$). This may be due to the small particles present in the

continuous phase after centrifugation. At the concentration of 1.2% IM, the stronger network could perhaps trap more of these particles and thus decrease the elastic modulus of the continuous phase. Homogenisation leads to a greater proportion of small particles, which will increase the concentration of particles in the continuous phase, thus increasing the elastic modulus of the continuous phase of the tomato suspension.

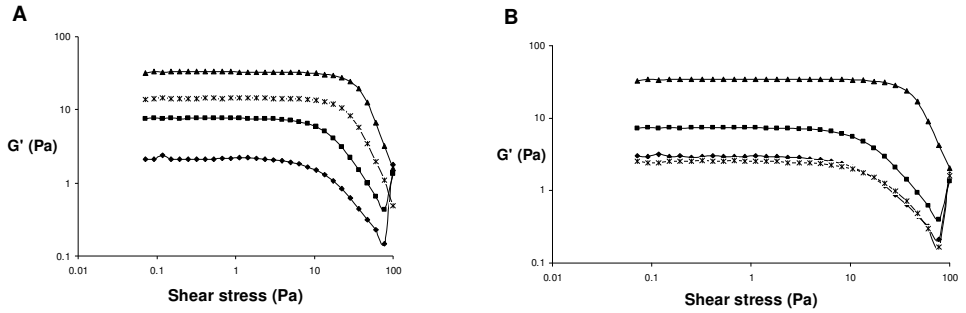

Figure 20. The elastic modulus, G′, of the continuous phase of the fibre suspensions. A) 0.8% suspensions; B) 1.2% suspensions. ♦ - apple; x - tomato; ▲- potato pulp; ■ – carrot. From Paper I.

6.3 Water-holding capacity

The hydration capacity of fibre suspensions is a measure of the water retained inside the fibre network under certain conditions [80]. The water is not usually strongly bound to the network, but merely trapped inside it. There are several ways of measuring the hydration capacity of a fibre suspension. Some of the more common methods found in the literature are briefly described below.

Swelling. The bed volume technique has been used in several studies [30, 90-92]. The sample is allowed to swell in an excess of water for several hours in a measuring cylinder. The volume of the swollen fibre suspension is then expressed as the volume per gram of dry sample.

Water-holding capacity. The generic term for the water retained by a sample after excess water has been removed is the water-holding capacity [15]. The water is either removed by gravity and filtration, or by centrifugation [30, 90]. In some studies the fibre is hydrated for a short time, ~2 hours [29, 93], while in others the fibre is kept in excess of water for a considerably longer time, up to 16 hours [90, 91]. Several names are used synonymously for this in the literature, such as water-binding capacity and water-retention capacity [15].

Water absorption. During adsorption measurements the kinetics of water up-take is studied [15]. A common method of measuring water absorption is by capillary pressure [30, 91, 93]. The sample is placed on a glass filter above a chamber containing water, and the adsorption is measure by weighing the sample before and after exposure to the water.

As the purpose of this work was to investigate various fibres in suspensions and their effect on food products, the water-holding capacity (WHC) was measured. For tomato suspensions 110 000 *g* had previously been used to separate the insoluble from the soluble material [41]. However, at pre-studies for the present work, no differentiation between the various fibre suspensions could be achieved with this high rotational speed. Therefore it was decided to measure at 50 000 *g* (20 minutes, 20ºC). The volume of the pellet was calculated according to Equation 3.

$$WHC = \frac{V_s}{V_t} \cdot 100 \qquad [\%] \qquad (3)$$

where V_s is the volume of the wet pellet and V_t is the total volume of the suspension calculated according to Bayod et al. [82].

6.3.1 Changes due to processing

Homogenisation increased the WHC of carrot and especially tomato suspensions, while no significant increase was seen in apple and potato pulp suspensions following homogenisation (Paper II). The increase in WHC of the potato pulp suspensions due to heat treatment was probably due to the swelling of the starch granules remaining in the fibre network (as can be seen in Fig. 15), which led to the retention of water in the network (Paper III). A decrease in WHC was observed in the apple and carrot suspensions subjected to heat treatment favouring β-elimination. This could be due to the decrease in insoluble pectin found in these samples, since the amount of insoluble material may influence network formation so as to decrease the WHC [41]. A significant relationship ($p<0.05$, $r = 0.70$) was found between the insoluble pectin and the WHC for the heat treated suspensions.

The effects of concentration and dispersion medium on WHC differed among the fibre sources studied. For the tomato suspensions, the WHC increased with concentration by more than 50% when using a 2% pectin solution as the dispersion medium. This strong relation between IM and WHC has been observed previously in tomato suspensions [82]. Apple suspensions were not greatly affected by concentration and/or the dispersion medium ($p>0.05$ in one-way ANOVA). For carrot and potato pulp suspensions the WHC was enhanced with increasing concentration ($p<0.01$ and $p<0.001$, respectively). At the lower concentrations, 0.8 and 1.2%, potato pulp had a lower WHC than carrot. However, at the higher concentration, and when water was used as the continuous phase, the potato pulp suspensions exhibited a higher WHC than the carrot suspensions. This may be due to the difference in degree of aggregation of the dried and non-dried potato pulp, the latter being less aggregated and used for the higher concentration of 3% IM. At the lower concentrations a dried, aggregated potato pulp was used, possibly impairing the WHC.

6.4 Correlation of physicochemical properties

The correlations between the physicochemical properties of the suspensions studied and the composition of DF for the heat-treated samples (Paper III) were mainly univariate for each fibre source, and have been discussed for each measured property above. However, the samples subjected to homogenisation (Paper II) showed more multivariate behaviour. To correlate the physicochemical properties of the suspensions and the composition of DF, PCA was carried out. The result is presented in Fig. 21.

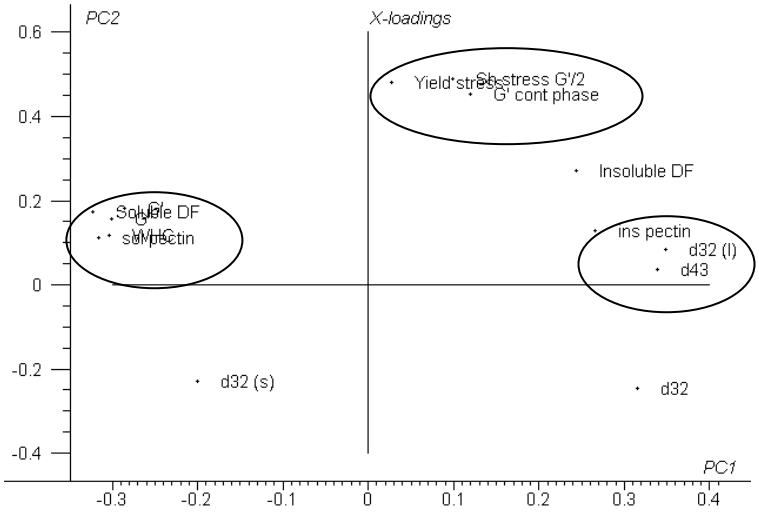

Figure 21. PCA loading plot of the physicochemical properties and the composition of DF of the homogenised fibre suspensions. From Paper II.

The first principal component (PC) was found to explain 41% of the variation. Two groups of variables can be seen along PC1. To the right in Fig. 21, the large particles (d_{32}(l) and d_{43}) and the content of insoluble pectin are correlated. On the left-hand side, it can be seen that soluble pectin, WHC and the elastic modulus are negatively correlated to large particles and insoluble pectin. The larger the particles in the suspensions, the lower the WHC ($p<0.01$, $r = -0.88$). This shows that a less

aggregated microstructure has a higher WHC and higher elastic modulus than a suspension containing large clusters.

The content of insoluble pectin did not change significantly with homogenisation for most fibre sources. Therefore, the negative correlation between insoluble pectin and WHC and G′ in the PCA plot only reflects the microstructure of the different fibre sources studied. The content of insoluble pectin is more likely to be positively correlated to these two physicochemical properties, as was observed when the content of insoluble pectin was varied in each fibre source separately, in the case of the heat-treated suspensions (Paper III). An increase in water-insoluble fibre has previously been seen to enhance the WHC and the elastic modulus of tomato suspensions [41]. The concentration of IM was also found to be positively correlated to the elastic modulus when each fibre source was investigated separately (Fig. 17).

The second PC (explaining 23% of the variation) shows a positive correlation between the elastic modulus of the continuous phase, the shear stress at G′/2, and yield stress ($r = 0.83$ and $r = 0.73$, respectively and $p<0.001$). It appears that the gel in the continuous phase has few junction points (low G′). The network of insoluble fibres is also highly influenced by the shear stress applied, leading to brittle behaviour. However, when the continuous phase has a higher elastic modulus, the LVER is longer, and the suspension is broken down over a broader range of shear stresses. This shows that the continuous phase affects the breakage properties of the suspension, but not the elasticity of the suspension at rest, since the elastic modulus of the suspension is orthogonal to the G′ of the continuous phase, implying that they are not correlated.

7. Sensory properties

Sensory analysis is the scientific field of measuring the human response to products based on the senses of sight, smell, taste, touch and hearing. Sensory analysis has been used in the food industry to develop new, tasty products since the 1940s [94]. Today, it is used in both marketing and product development. Three different types of sensory evaluation are recognised: discrimination, descriptive and hedonic analysis [95].

In discrimination testing products are compared with each other to identify differences. Common discrimination tests are the triangle test (find the sample that is different from the other two), the duo-trio test (find the sample that matches the reference) and paired comparisons (e.g. find the saltiest sample) [95].

In descriptive evaluation, a sensory panel consisting of people with excellent abilities to distinguish between the parameters being tested, is employed. For example, if the members of the panel are to evaluate the taste of a food product, their ability to differentiate between a sweet and non-sweet sample should be better than chance [94]. It is thus not the affective response of the sensory panel that is desired, but a more objective and quantitative description of the food product. Before descriptive testing is carried out, the panel undergoes product-specific training to ensure that the members understand the attributes used for evaluation.

The degree of liking of a product is tested by hedonic analysis. In contrast to descriptive evaluation, untrained consumers are asked to score a product subjectively. A common method is to use a nine-point hedonic scale, extending from dislike extremely to like extremely [95].

The texture of food products has not been studied as long as other properties, such as taste and smell. In 1963 a definition of texture was proposed by Szczesniak [96]. Popular nomenclature was correlated to textural parameters such as hardness, viscosity/elasticity, particle size and shape. These definitions are still in use in texture analysis today [97]. Texture analysis of semi-solid foods, such as fibre suspensions, has been performed previously. A beverage containing different types of β-glucan was perceived as less thick when the β-glucan had a lower molecular weight and was more processed [98]. Concentrated suspensions of soluble polysaccharides such as alginate, pectin and carrageenan have been evaluated by descriptive analysis, showing a correlation between the viscosity and perceived thickness [99]. Regarding suspensions containing insoluble DF, texture analysis has mainly been carried out in applications where DF was added to vanilla-flavoured custard [100] and béchamel sauce [101].

There is a lack of understanding regarding the relationship between the textural properties of DF and its physicochemical properties [16, 102]. DF from fruit and vegetables has been used in several applications [72, 101, 103, 104], but little information can be found in the literature regarding the sensory perception of fibre suspensions. An improved understanding of how the textural attributes of fibre suspensions are related to their physicochemical properties such as PSD, elastic modulus and WHC would help us predict the texture of foods containing DF as an additive.

7.1 Descriptive analysis of fibre suspensions

Fibre suspensions of tomato, carrot, apple and potato pulp, at concentrations of 0.8 and 1.2% IM, homogenised to different degrees (see Tables 2 and 4) were sensorily evaluated by descriptive analysis (Paper IV). Descriptive analysis has previously been used to investigate the perception of viscous drinks with fibre added [105] and was

thus considered to be a suitable means of studying the textural perception of fibre suspensions.

A professional panel of 9 members (Procordia food, Eslöv, Sweden) with considerable experience of both the scoring system and descriptive testing was used. The panel had been engaged in testing different sauces, dressings and ketchups weekly for several years, and underwent product-specific training for 12 hours prior to the test sessions. Attributes describing the texture were chosen in consultation with the head of the panel. Table 7 describes the procedure for the evaluation of each of the attributes thick, grainy, slippery, crispy and melting. The intensity scale employed for each of the descriptors was an unstructured line on a computer screen, where the left end indicated no intensity and the right end very high intensity.

Table 7. Procedure for the evaluation of the sensory attributes

Attribute	Evaluation procedure
Thick	Place the product on your tongue and press the tongue up towards your palate. Evaluate how much force is needed.
Grainy	Place the product on your tongue and press it up towards your palate. Evaluate the irregular particles when you move your tongue against your palate.
Slippery	Place the product on your tongue and press it up towards your palate. Then lower your tongue. Evaluate how much of the product remains on your palate and whether it forms a lubricating layer on the tongue and palate.
Crispy	Put a spoonful of the sample into your month and chew. Evaluate how much noise the product makes when you chew it for the first and second time with your molars.
Melting	Place the product on your tongue and press it up towards your palate. Evaluate how the product spreads.

From Paper IV

The samples were tested during four sessions. Suspensions of one fibre source were tasted at each session. Three replicates of each sample were given to the panel to test: non-homogenised suspensions and suspensions subjected to two degrees of

homogenisation, at the two concentrations specified above. The panellists were seated in individual booths and were served the samples in thirty-ml plastic cups with lids. The samples were served at room temperature in a random, balanced monadic order [106], i.e. the samples were served one by one.

The FIZZ system (Biosystemes, France) was used to collect the sensory data. The score on the unstructured line was then transformed into numbers, where no intensity (the left end) equals 0, and very high intensity (the right end) equals 10. The results of the sensory evaluation for the different descriptors are shown as mean values in Figs 22-26 .

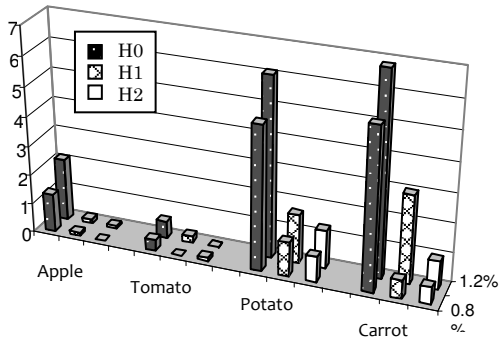

Figure 22. Mean values from the sensory analysis of the attribute **grainy**.

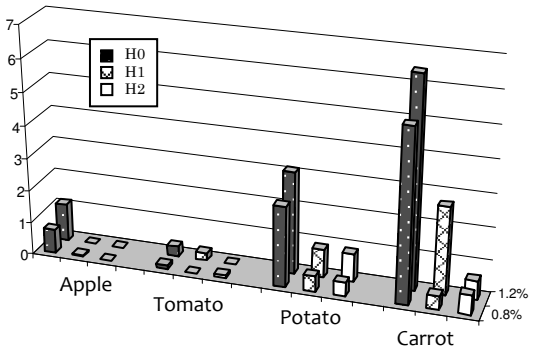

Figure 23. Mean values from the sensory analysis of the attribute **crispy**.

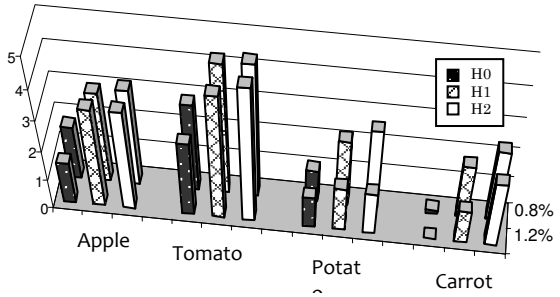

Figure 24. Mean values from the sensory analysis of the attribute **slippery** (the axis of the concentrations are reversed for the sake of clarity).

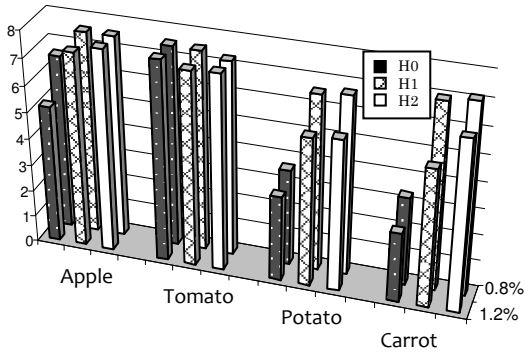

Figure 25. Mean values from the sensory analysis of the attribute **melting** (the axis of the concentrations are reversed for the sake of clarity).

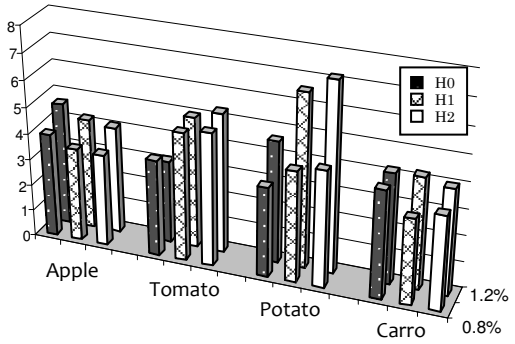

Figure 26. Mean values from the sensory analysis of the attribute **thick**.

A high score indicates a strong sensation for the attribute in question. For the descriptors grainy, crispy and thick, a trend towards stronger sensations at the higher concentration was observed ($p<0.001$). A decreasing trend was seen for slippery ($p<0.05$) and melting ($p<0.001$).

Homogenisation also had a strong influence on most of the measured descriptors. Non-homogenised samples were perceived as significantly more grainy and crispy, while homogenised samples were deemed to be more slippery and melting ($p<0.001$). This was especially true for carrot and potato pulp suspensions, which hade high initial values of the attributes grainy and crispy, and low perceived melting and slippery sensations. The effect of homogenisation on the attribute thick was also significant for most fibre sources; however, the perceived thickness of tomato and potato pulp suspensions increased with degree of homogenisation, while in the homogenised apple suspensions it decreased.

An ANOVA for each of the sensory attributes was carried out taking all the samples into account. The concentration of insoluble material had the least effect on all the sensory attributes, except for thick, where the concentration had the greatest influence. Fibre source had the greatest influence on the perception of slippery, while homogenisation had most effect on the perceptions of grainy, crispy and melting. However, the fibre source also had a considerable influence on these three attributes.

There was a significant influence of panel member on the sensory results ($p<0.001$), which has been reported previously [107]. However, there was no significant difference between the replicate samples for individual assessors, and the effects of homogenisation and concentration on most descriptors were still significant, as described above.

Principle component analysis was performed to investigate possible correlations between the sensory variables and the measured physicochemical properties (Fig. 27).

Two-thirds (66%) of the variation in the material was explained by the first two PCs. In the loading plot (Fig. 27A) slippery, melting, G′, WHC and soluble pectin, seen on the left in the plot, are correlated. A strong network with a high elastic modulus and high WHC seems to create a sensation of melting and slipperiness, perhaps by covering the mechanoreceptors in the mouth.

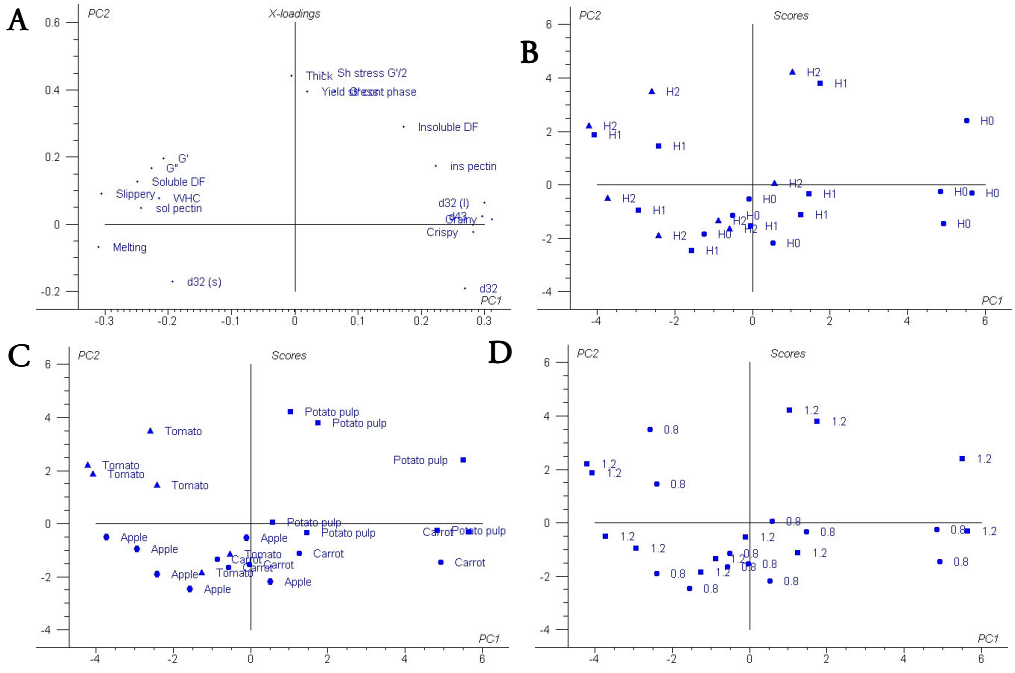

Figure 27. PCA plots of PC1 and PC2 explaining 66% of the variance. A) Loading plot for the physicochemical variables, sol = soluble, ins = insoluble. B) Score plot for homogenised samples. C) Score plot for fibre source. D) Score plot for insoluble material concentration. From Paper IV.

On the right-hand side of PC1 crispy, grainy, d_{43}, d_{32}(l) and, to some extent, insoluble pectin and d_{32}, are correlated. Larger particles/aggregates in the fibre suspensions give a crispier and grainier sensation, which has been reported previously by Engelen et al. for silica particles and polystyrene spheres in vanilla custard [102]. Engelen et al. proposed that adding particles decreased lubrication in the mouth, which increased

friction and thus the sensation of roughness. A difference in the perception of soft and hard particles was also reported. Since a soft particle will be deformed by chewing, it will not be perceived as being as rough as a hard particle [102]. This was also seen for the fibre suspensions studied in the present study, where tomato suspensions were found to be easily broken by homogenisation, implying a softer material giving more slippery and melting sensations. The opposite was observed for carrot suspensions, in which the cellular structure was maintained after homogenisation, and these were perceived as crispier and grainier.

Along the PC orthogonal to PC1, i.e. for PC2, thick is correlated to some extent to yield stress, shear stress at G′/2 and the elastic modulus of the continuous phase ($p<0.01$, $r = 0.22$, 0.70 and 0.43, respectively). The correlation between sensory attributes of different semi-solid foods similar to thick (e.g. firmness, hardness and thickness) and yield stress has been investigated by several authors [108-110], who found a clear relation between the perceived thickness and the yield stress, consistent with the findings in this study.

When the score plots (Fig. 27B, C) were studied in relation to the loading plot, a relationship was revealed between the morphology and the sensory parameters grainy/crispy and melting/slippery. Carrot and potato had a more intact cell structure both before and after processing, whereas after homogenisation apple, and especially tomato, showed a fibrous morphology with more cell fragments. The structure of the fibre suspensions changes along PC1, from cellular aggregates at the left-hand end to more single cells and fibrous microstructure at the right-hand end. The perception of a grainy, crispy texture may be the result of a microstructure of clusters of the fibres, whereas melting and slippery sensations resulted from single cells and a fibrous structure. No trends regarding the concentration could be elucidated in the first two PCs (Fig. 27D).

8. Dietary fibre as an additive in low-fat sausage

Several studies have been carried out in which different kinds of DF have been added to meat systems in order to enhance the protein network after fat removal. In most of these studies, soluble fibre such as gums [111, 112], pectin [113] or fructans [114-116] was used. Some studies have been carried out with fruit or vegetable fibre containing both soluble and insoluble DF [101, 117-119]. The effect on the texture was diverse: some products to which DF had been added showed a softer texture, while others showed increased firmness.

Table 8. Pre-processing of the potato pulp fibre suspensions

Sample	Homogenisation	Heat treatment
NH0	Non-homogenised	HT0
NH1	Non-homogenised	HT1
NH2	Non-homogenised	HT2
NH3	Non-homogenised	HT3
H0	Homogenised	HT0
H1	Homogenised	HT1
H2	Homogenised	HT2
H3	Homogenised	HT3

Homogenised: 5 passes at 90 bar in a high-pressure valve homogeniser.
Heat treatment is explained in Table 3.

The desired property of a sausage is a firm elastic network that retains a sufficient amount of water [120]. Homogenisation was shown to increase the elastic modulus of the potato pulp suspension (Section 6.2.1) and made the perceived texture smoother and less grainy (Section 7.1). There was also a trend towards increasing elastic modulus when the samples were heat-treated to activate PME. Potato pulp was

suspended in half the added water in the sausage recipe, using a household mixer (Fasett, Sweden), to obtain a fibre suspension. The suspensions were then heat-treated and homogenised according to Table 8. The suspensions were then used in the preparation of sausage batters according to the recipes in Table 9.

All the sausages made had the same basic composition: a water/protein ratio of approximately 7.5:1, a potato starch addition of 4%, a fat content of the cooked sausages of approximately 2%, and 0.6% total DF in the sausages containing potato pulp. These are the properties that affect the texture to the greatest extent in a sausage and should thus be kept constant [83, 121].

The ingredients were mixed in a food processor (Braun, Germany) for five minutes. The temperature of the meat batter was kept below 12ºC. The batter was then packed in plastic tubes with lids (Ø = 3.7 cm) and stored under refrigerated conditions. After a total resting time of 2½ hours after batter preparation the sausages were boiled in a water bath to a centre temperature of 75ºC, (approximately 45 minutes). The water bath was always at least 5ºC higher than the centre temperature of the sausages (ΔT cooking). The temperature in the sausages was measured by thin thermocouples inserted through a small hole in the plastic lid.

Table 9. Recipes for the reference sausage and the sausages to which potato pulp was added

Ingredient (g)	Reference	Potato pulp
Water/ice	46.18	36.2
Meat[1]	47.25	49.4
Spices and additives[2]	2.27	2.27
Potato starch	4.0	4.0
Vegetable additive	-	8.4
Total	100.0	100.0

[1] 60% pork (4.2% fat) and 40% beef (4.9% fat) (Ugglarps AB)

[2] Black pepper (0.1 g), nitrite salt (0.72 g), vacuum salt (1.28 g), ascorbic acid (0.02 g), polyphosphate (0.15 g)

From Paper V.

Loss due to sausage processing was calculated as the difference in weight before and after boiling of the sausages (% of initial weight). To measure the loss of water and fat during processing, meat batter and sausage (5 g) were dried at 102ºC overnight and the water loss (a) and the fat loss (b) calculated according to Equations 4 and 5, respectively.

$$a = W_i - \frac{W_f \cdot (100 - PL)}{100} \quad [\%] \quad (4)$$

$$b = PL - a \quad [\%] \quad (5)$$

where W_i the water content of the batter, W_f the water content of the cooked sausage and PL the process loss. To measure the frying loss, a 1 cm thick slice of sausage was fried in a pan at 174ºC for 2 minutes on each side (centre temperature 72-73ºC). The frying loss was expressed in terms of the weight after frying as a percentage of the

weight before frying. The texture (firmness) was measured with an Instron Universal Testing Machine. The maximum force needed to compress a 10 mm^3 cube of sausage by 30% at a speed of 1 mm/s was measured.

No significant differences were seen in any of the losses between sausages with and without potato pulp. However, the addition of potato pulp to the sausages afforded them a significantly firmer texture than the sausage without fibre ($p<0.001$). The potato pulp consists mainly of insoluble material (Fig. 6) and the estimated concentration in the batter was calculated to be 4.3%. The high concentration of IM in the potato pulp is thought to create a network enhancing the meat protein network. A stronger network structure in the sausage may lead to a greater resistance to compression, and thus greater firmness of the sausages containing potato pulp.

An interaction was found between homogenisation and heat treatment in the frying loss of the sausages containing potato pulp ($p<0.001$) (Fig. 28). In the sausages containing potato pulp suspensions minimally heat treated (HT0), and those heat-treated to enhance PME activity (HT2), a decrease in frying loss was seen with homogenisation. However, in sausages containing suspensions heat treated to favour β-elimination (HT1 and HT3) the frying loss increased with homogenisation. As discussed in Section 6.1, homogenisation causes a decrease in the size of the large potato pulp clusters and this appears to affect the efficiency of the heat treatment. The methyl groups may have been more exposed to PME after homogenisation, decreasing the DM and creating carboxyl groups. When more carboxyl groups are charged, interactions with Ca^{2+} ions can link pectin chains to form a firm network [32], thus more water is retained in the structure. Another explanation of the decrease in frying loss seen for HT2 compared with HT1 and HT3 could be due to the starch granules in the potato pulp network. This starch (around 15%) will probably be released by homogenisation. At the shorter heating times (HT0 and HT2) the starch is swollen but not disintegrated, as happens when amylose leakage occurs at higher

temperatures. The swollen starch granules retain more water, hence lowering the frying loss.

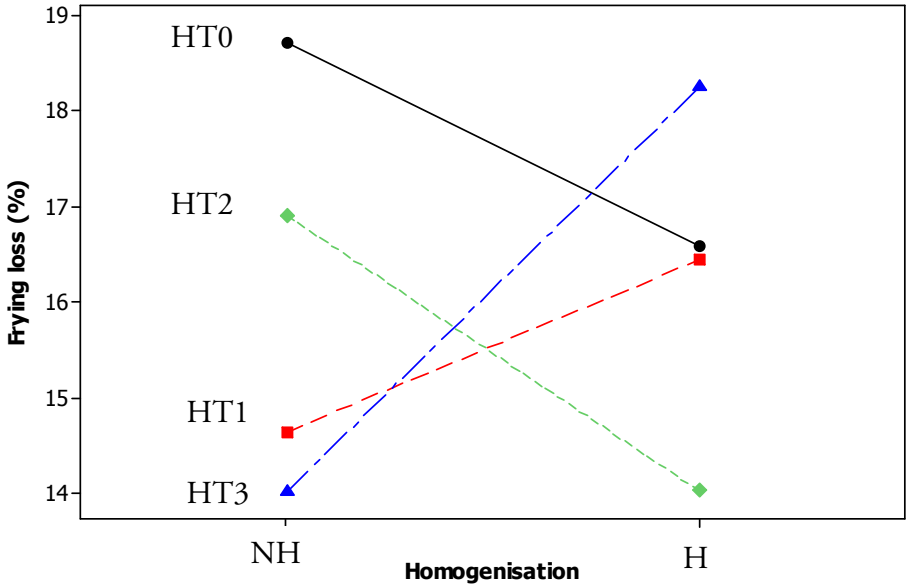

Figure 28. Interaction plots between homogenisation and heat treatment for frying loss of sausages containing potato pulp. NH = non-homogenised, and H = homogenised. Heat treatments: see Table 3. From Paper V.

The firmness of the sausages containing potato pulp was affected by homogenisation and heat treatment in a similar way to frying loss (Fig. 29). For the sausages containing potato pulp suspensions following HT0 and HT2, an increase in firmness was seen with homogenisation, probably due to the same mechanism as that proposed above for the decrease in frying loss. A stronger network may resist compression leading to greater firmness.

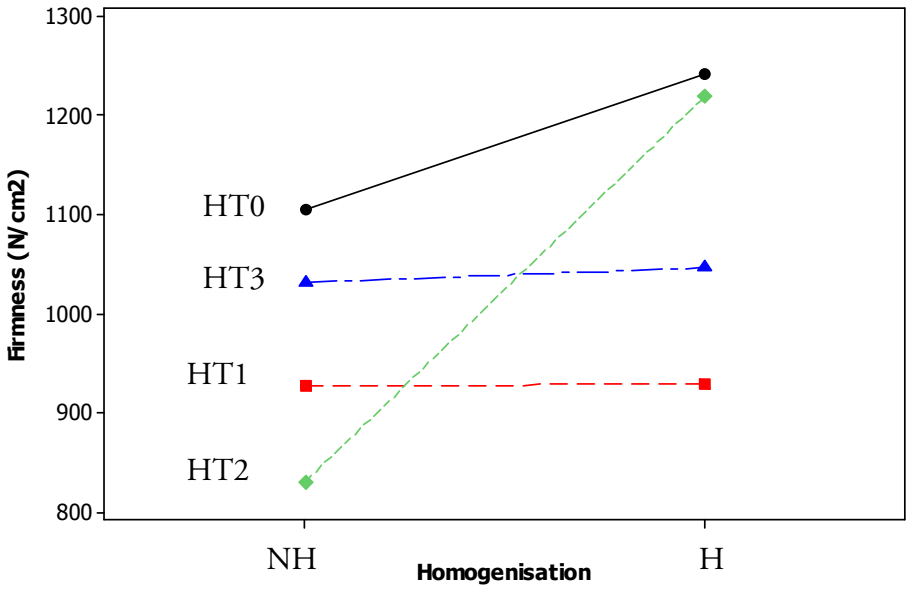

Figure 29. Interaction plots between homogenisation and heat treatment for firmness of sausages with potato pulp. NH = non-homogenised, and H = homogenised. Heat treatment: see Table 3. From Paper V.

An untrained panel evaluated pieces of fried sausage allowed to cool to room temperature, hedonically. Sixteen participants from a pool of 34 attended each of the six sessions. The panel evaluated crumbliness, compactness, juiciness, meat-taste intensity, off-flavour and their total impression of the sausages on a scale from 1 to 9, according to the explanation of the attributes given in Table 10. On each occasion, 6 or 7 pieces of sausage were sampled, labelled with a three-digit number. The sausages were served on a white paper plate, in a random order; each assessor tasting the sausages in a different order to prevent position error [94]. The tests were carried out in individual booths with normal lighting, and the assessors were given a glass of water to rinse their mouths between samples.

Table 10. Attributes and ratings used in the hedonic sensory analysis of sausages, with and without fibre additives

Attribute	Rating	Scale
Crumbliness	Is the sausage smoother, or more particulate than what you consider a normal sausage should be?	1 – Smooth 9 – Grainy
Compactness	Does the sausage fall apart, or is it more strongly held together than what you consider a normal sausage should be?	1 – Not compact 9 – Very compact
Juiciness	Does the sausage release more liquid when chewed, or is it drier than you consider a normal sausage should be?	1 – Dry 9 – Very juicy
Meat-taste intensity	Does the sausage have more or less meat taste than what you consider a normal sausage should have?	1 – None 9 – Very high
Off-flavour	Does the sausage have more or less off-taste than what you consider a normal sausage should have?	1 – None 9 – Very high
Total impression	What is you total impression of the sausage, do you like/ dislike the sausage?	1 – Very bad 9 – Very good

From Paper V

Two sensory dimensions were seen for the sausages when subjected to PCA (Fig. 30). High juiciness was correlated to a low perceived compactness of the sausage ($p<0.001$, $r = -0.28$). The reference sausage was found to be less compact and more juicy than the sausages containing potato pulp. As discussed above in relation to the firmness measurements, a high content of IM appears to create a strong network, leading to a sausage with a firmer texture.

In the other sensory dimension, off-flavour was negatively correlated to meat-taste intensity and total impression ($p<0.001$, $r = -0.37$ and -0.43). The meat-taste intensity was also associated with total impression ($p<0.001$, $r = 0.53$), which has also been found in a previous study of low-fat sausages [122]. In this sensory dimension, no significant difference was seen between the reference sausage and those containing potato pulp, i.e. the taste of the sausages was considered equally good. The mean

value of the total impression of all the sausages was above 5, which indicates that all the sausages were more liked than disliked.

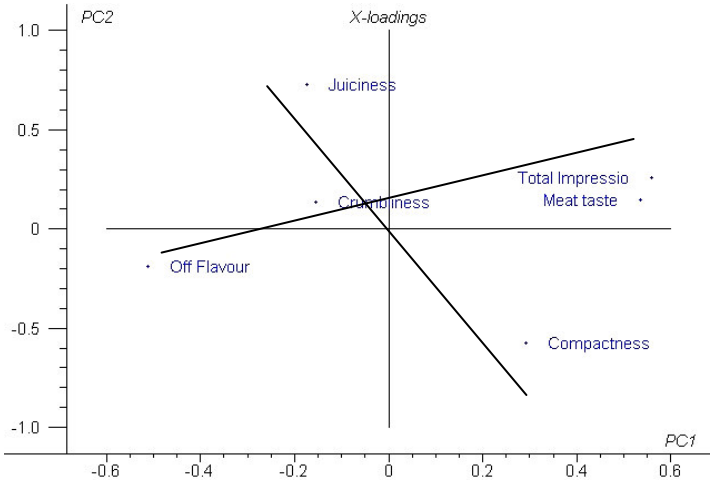

Figure 30. PCA of the sensory attributes explaining 53% of the variation in the first two PCs (From Paper V)

9. Conclusions

The processing of fruit and vegetable fibre suspensions affects their physicochemical properties, the composition of the DF and the sensory characteristics. A strong relation was found between the pectin solubility and the way in which the fibre suspensions are affected by processing. The concentration of insoluble material in the fibre suspensions was also found to be of great importance for physicochemical properties such as the rheological parameters and the water-holding capacity.

The effect of homogenisation on fibre suspensions depended on the microstructure of the fibre source, which in turn was governed by the composition of the soluble and insoluble fibre. Fruit and vegetable suspensions showed closely associated large cell clusters, probably due to the high content of insoluble pectin in the cells. When subjected to homogenisation, these cell clusters were more difficult to break than the single cells and cell fragments in DF suspensions with a lower content of insoluble pectin. The difference in microstructure can also explain the difference in elastic modulus and WHC, as a less aggregated structure led to a higher elastic modulus and WHC.

Heat treatment of apple and carrot suspensions affected the pectin composition, which in turn gave rise to variations in the particle size distribution and WHC. In samples subjected to heating to favour β-elimination a decrease in insoluble pectin was seen. The mean particle size decreased, as did the WHC, probably due to the depolymerisation of the insoluble pectin connecting the cells. In the samples where PME was activated, an increase was seen in the elastic modulus of the apple suspension, which could be the result of strengthening of the network due to Ca^{2+} interactions between the LM pectin chains. Regarding the potato pulp suspensions, heat treatment mainly affected the remaining starch, which started to swell, thus increasing the WHC.

Sensory perception of the texture of homogenised fibre suspensions was correlated to the morphology of the fibre sources. The larger clusters or aggregates, the greater the perception of crispiness and graininess. As the microstructure changed to smaller cell clusters and single cells, and finally to cell fragments, the texture changed and became more slippery and melting. The descriptor thick was correlated to the tendency of networks in the fibre suspensions to break up and the elastic modulus of the continuous phase.

A high content of insoluble fibre in potato pulp added to make a low-fat sausage increased the firmness of the sausage. The potato pulp created a strong network, enhancing the meat protein network, thus increasing the resistance to compression. Homogenising the fibre suspensions prior to addition to the sausages enhanced the firmness even more, especially in combination with heat treatment promoting PME activity.

10. Future outlook

In this study it has been shown that different fruit and vegetable fibre sources at various concentrations are affected differently by processes such as homogenisation and heating, regarding both the composition of the DF and its physicochemical properties. This, in turn, affects the sensory perception of both the fibre suspension per se, and the texture of sausages. This work has provided further insight in this area, but more research is needed to achieve the optimal texture of protein-rich foods to which fibre suspensions have been added. Some areas that would be interesting for further study are listed below.

There are several potential applications within the field of protein-rich food enriched with DF. Hamburgers, for example, have a completely different texture and microstructure from the sausages containing DF studied in this work. It would be interesting to investigate whether the trends reported in Paper V apply to all types of protein-rich foods. The effect of different fibre sources would also be an area of interest for further research.

Relationships were found between the microstructure, the composition of the DF and the susceptibility to homogenisation for carrot, apple, potato pulp and tomato. Other fruit and vegetable fibre sources should be studied to verify this relationship.

An interesting, less studied, area is the dependence of some of the properties of the fibre suspensions, such as texture and water-holding capacity, on the concentration of the insoluble material. These properties seem to depend on both the fibre source and the dispersion medium.

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