Molecular mobility in crispy bread crust

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Abstract

Crispy foods are strongly appreciated by consumers. For products to be judged as crispy they should break in a brittle way and emit sound while being eaten. These characteristics of a crispy food product are generally lost upon plasticizer uptake (mainly water) from the environment or from a more wet part of the product. The overall objective of this thesis was to better understand the role of water in the mechanism of loss of crispness in crispy bread crusts. The model systems used in this thesis were a bread model system and a bread crust model system.

The mechanism of water transport into the bread crust sample was studied by evaluating the kinetics of water uptake by bread crust particles for two different sorption experiments, an oscillatory sorption test and a more classical sorption test in which the air relative humidity (RH) was increased stepwise. In this way the effect of experimental sorption time on the resulting rate parameters could be studied because in the stepwise sorption test each step took a factor 25 longer than in the oscillatory sorption test. The water uptake kinetics showed exponential behaviour which is often ascribed to Case II relaxation controlled diffusion. The rate parameters of the experiment where RH was stepwise increased were around a factor 10 lower than those derived from the oscillatory sorption experiments. This indicates that the experimental time used affects the outcome of the experiment.

Besides the kinetics of water uptake also the effect of water on the glass transition of model bread crusts was studied. The results were compared with sensory data. Sensory crispness scores decreased at a water activity (a_w) of 0.55 and decreased further with increasing $a_{w.}$ At an a_w higher than 0.70 sensory crispness was completely lost. The glass transition at around 20°C (room temperature) was found for an a_w of 0.70-0.75. Nuclear Magnetic Resonance (NMR) gave a transition point in the mobility of the protons of water at a_w 0.58. This supports the hypothesis that loss of crispness starts as a result of processes at a molecular level, before the macroscopic glass transition. Probably the non-bound water (water that is not directly attached to the solid matrix) is causing the loss of crispness at low a_w. At higher a_w increased mobility of initially segments and side chains of the macromolecules will start to play a role.

The separate effects of a_w and water concentration were studied by making use of the hysteresis effect in water sorption. The water content (instead of a_w) was found to be decisive for the glass transition and the transition in mobility of water as measured by NMR. However, a comparison with sensory data and data from a puncture test showed that both water content and water activity had an effect on perceived crispness and number of force and sound peaks. Possibly an inhomogeneous distribution of water and consequently crispy and less-crispy regions exist in the samples with a history of high a_w thus making them overall more crispy than samples with the same water content but higher a_w.

Bread crusts of different formulations were tested regarding their water uptake kinetics. Treatment of a bread crust with protease (sprayed on top of the bread dough) resulted in slower water uptake kinetics for bread crust particles larger than 0.25 mm. For smaller particles there was no significant difference in water uptake rates. This suggests a morphology effect.

Table of contents

Chapter 1 General introduction

1.1 Introduction

Many different types of crispy foods can be found in the supermarket. Products that are crispy range from potato crisps to bread to fish fingers to ice cream cones and vegetables. Crispy foods are strongly appreciated by consumers as was for example shown by Katz and Labuza (*1*) for saltines and popcorn. They found that consumers rate these products the same for the attribute "crispness" and the attribute "like" or "dislike". For products to be judged as crispy they should break in a brittle way and emit sound while being eaten. These characteristics of a crispy food product are generally lost upon plasticizer uptake (mainly water) from the environment or from a more wet part of the product.

The research described in this thesis was part of a larger project called "Crispy crunchy behaviour of cellular solid foods". The aim of this project was to get a better understanding of all factors involved in crispy behaviour of composite foods both at molecular, mesoscopic and macroscopic scale. Also objective methods to determine crispy/crunchy behaviour in time were developed. This should finally result in an improved shelf life of crispy composite foods. The composite foods on which this project was focused are foods with a moist interior and a dry and crispy outside. For these products the water that plasticizes the crispy parts during storage will come primarily from the moist part of the product. Two model systems were used: a deep fried snack model and a bread model system. Using these products different aspects were evaluated. The main ones were fracture behaviour, sound emission during fracture, effect of morphology on crispness, sensory evaluation of the products, effect of ingredient variation, effect of increase in water content on sensorial and instrumental measured crispness in relation to product structure and relation between crispness and mobility of water. The overall objective of this thesis was to better understand the role of water in the mechanism of loss of crispness in crispy bread crusts, with special attention to the role of the most important constituents starch and gluten. A method was developed to measure water uptake kinetics in crispy bread crust. With this method the effect of ingredient variation and morphology on water uptake rates was studied. Besides the water uptake kinetics also glass transitions were measured as a function of water content or ingredient variation and the role of the separate components gluten and starch was discussed. Results obtained were used to construct a working model explaining the role of the main factors affecting the creation and retention of crispness. This will be used to establish guidelines for the industry to improve crispness creation and retention of the crust of baked bread type and deep fried snack type products. This introduction will give an overview of the characteristics of bread crust and the different tracks of research on crispness in food products.

1.2 Understanding crispness

1.2.1 Sensory aspects

Crispy food products made from wheat flour that have been evaluated sensorially are e.g. partly dried white and extruded breads (*2, 3*) and extruded starch products (*4*). In general above a water content of 9% loss of crispness is observed at room temperature for these products.

Most products follow a sigmoidal curve for loss of crispness as a function of water activity or water content (*3, 5*). This is schematically depicted in figure 2. Katz and Labuza (*1*) evaluated potato chips, popcorn, puffed corn curls, and saltines at different water activities with respect to crispness and textural hedonic quality with a sensory panel. Critical water activities, where the products became organoleptically unacceptable, generally fell in the 0.35–0.50 a_w range. Sauvageot and Blond (*6*) found the same for breakfast cereals. Sensorial evaluation of bread crust was performed by Primo-Martín et al (*7*) who found that a bread crust lost its crispness within 2 hours when stored at 80% RH, but maintained its crispness up to 5 hours when stored at 40% RH. Modification of the bread crust with a protease had a positive effect on crispness retention.

Exact consensus on the definition of sensory characteristics like crispy and crunchy does not yet exist (*8, 9*), but it is generally accepted that they are all related to the fracture behaviour of the material (*10*). Crispy foods are firm and brittle. An abrupt failure of the product is required for a crispy sensation. Not only the mechanical properties are important but also the sound that is produced during biting (*9*).

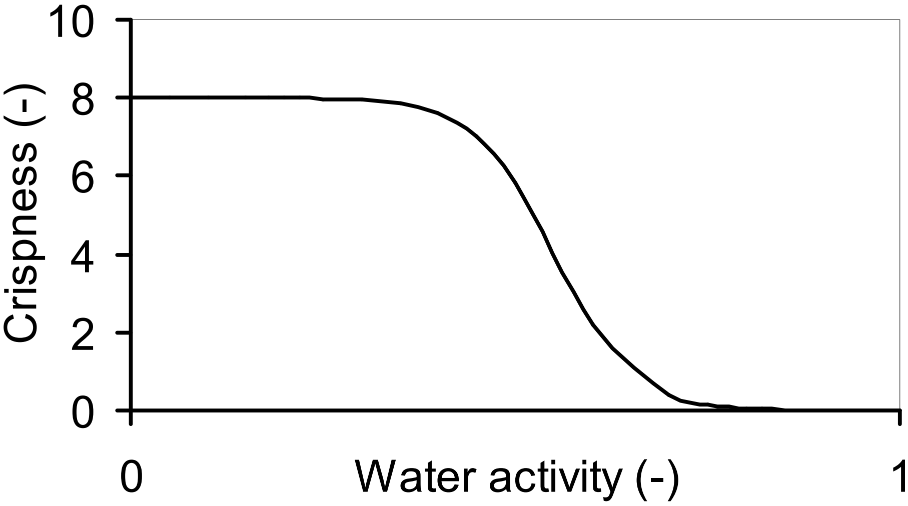

Figure 1 Schematic curve describing loss of crispness as a function of water activity.

1.2.2 Fracture behaviour

Fracture behaviour of crispy food products can be understood by realizing that all these materials contain defects or weak spots. These irregularities lead to local concentration of the stress during deformation and act as starting points for fracture (*11*). When a defect (crack) grows the stress in the material around the new formed crack tip relaxes and the stored elastic energy in it becomes available. This energy may be used partly to create new surfaces in the fracturing material, partly for reshaping of the deformed material, and partly for other processes like permanent deformation, acoustic emission both in- and outside the audible region, heat, etc (*8*). These processes can be summarized in an energy balance:

$$W = W' + W'' + W_f + W_0'' \quad (1)$$

where: W is the amount of energy per unit volume supplied to the material during deformation:

$$W = \int_{\text{path of deformation}} \sigma(\varepsilon)\, d\varepsilon \quad (2)$$

σ is the stress, ε the strain involved, W' is the part of the deformation energy that is elastically stored, W'' is the part dissipated because of flow or permanent deformation, W_f is the fracture energy and equals the specific fracture energy per area R_s ($J \cdot m^{-2}$) multiplied by the fractured area, W_0'' is the part of energy dissipated because of other processes (*8*).

During deformation of a (crispy) product energy is supplied to this material. For an ideal elastic material this energy is completely stored in the product as elastic energy (W'). If a crack starts to grow, the energy stored directly around this crack becomes available due to relaxation of the stress in the material. If the amount of energy that becomes available exceeds the energy required for the fracturing process, the crack will propagate spontaneously at high speed (*12, 13*). The minimal required crack speed to get acoustic emission is likely around 300-500 m/s (*14*). Energy dissipation processes other than due to W' directly effect the fracture process because these processes will lower the amount of energy available for the fracture process. This would result in a lower crack speed and with that less or no acoustic emission and a less brittle fracture. Sensorially, the product will be perceived as less crispy. Increased mobility of (parts of) the macromolecules present in for example a crispy bread crust might lead to extra energy dissipation and thus to decreased sensorial crispness. This increased mobility could be due to an increased plasticizer level (eg water).

1.2.3 Sound

The sound emission during fracture of a crisp food is of importance for the crispy sensation. Several authors have measured the sound emission during fracturing of e.g. toasted bread (*3*), gluten and starch extrudates (*15*) and potato chips (*16*). Roudaut et al (*3*) concluded that only the sound intensity is of interest. They also found a similar critical water content when evaluating the loss of the acoustic emission and of the sensorial crispness as a function of water content, but the sensorial loss of crispness occurred over a broader transition range than the loss of acoustic emission. Although good correlations between acoustic emisson and sensory results were found, Edmister and Vickers (*17*) also showed that consumers graded crispness the same with and without hearing the sound of breaking via the ears. This suggests that also part of the sound of the fracturing food product is bone-conducted (*18*).

Luyten and van Vliet calculated a minimum beam thickness for sound to be produced while fracturing crispy foods. They found that beams thinner than about 50-100 µm would not produce audible sound at fracture and thus will not or much less contribute to the perception of crispness. For crispness, there was also an upper limit predicted for the beam thickness. Above a thickness of a few hundred micrometers they will be perceived as hard (*14*).

1.2.4 Mobility aspects

Water migration causes hydration of initially dry components of crispy products. This results in an increase of the mobility of the macromolecules, causing a glass to rubbery transition of the amorphous regions of the macromolecules present (mainly proteins and carbohydrates). Roughly at the same water content a loss of the crispy behaviour occurs (*3*), (*19*). This situation is depicted in figure 2. At low plasticizer content and at low temperature crispy food products in general will be in the glassy state and at high moisture and /or high temperature the sample will change to a ductile state. Not only water but also small sugars, lipids and other low molecular weight molecules can act as a plasticizer. However, most authors observed that the glass transition temperature was much higher than the sensorial transition temperature for crispness (*2, 3, 20*). Le Meste et al (*21*) related the sensorial transition temperature for crispness to sub-Tg relaxations such as observed in biopolymers and low molecular weight sugars (β and γ relaxations). Their origin is still being discussed. As observed in polysaccharides they could correspond to rotation of lateral groups (γ relaxation at low temperature) or to local conformation changes of the main chain (β relaxation) (*22*).

With respect to storage stability, it is important to know not only the total uptake of water at a certain environmental relative humidity (RH) (as determined by thermodynamics) but also to know the kinetics of water uptake. This is because the faster the water uptake, the faster the loss of crispiness. Water uptake rates can be influenced by structure like for example porosity, beam thickness and beam structure of the crust as well as by its composition (e.g. interaction of water with the food constituents). The effect of addition of ingredients was for example tested by adding fat to sponge cake. This fat decreased the water migration kinetics, likely due to hindering of the migration of water into the solid matrix (*23*). The effect of porosity on moisture transfer kinetics have been studied for dough (*24*), bread crumb (*25*) and for sponge cake (*23, 26, 27*). In general researchers found a higher effective diffusion coefficient with increasing porosity. This was related to a change in the water transport mechanism (*23*). In low porosity foods, diffusion of the liquid water is considered to be the main process of water transport whereas in high porosity foods water vapor transport through the open pores will also determine the water uptake kinetics (*28*).

Fick's law is often used to describe water uptake rates for food systems.(*29, 30*). Fick's first law describes water diffusion related to Brownian motions. The diffusion coefficient depends on local water concentration (*31*). For example Roman-

Gutierrez et al. (*30, 32*) studied the rate of water uptake of a hard and a soft wheat flour and of different flour components. Diffusion coefficients of water in starch or wheat flour systems were determined using Fick's law (water uptake vs time). They found significantly higher apparent diffusion coefficients for the water-insoluble pentosans (1.5 x 10^{-13} m^2/s) at 25°C and 60% RH than for all other components (starch, gluten, water-soluble pentosans and wheat flour). For these components the apparent diffusion coefficients were between 0.5 x 10^{-15} and 5 x 10^{-15} m^2/s under the same conditions.

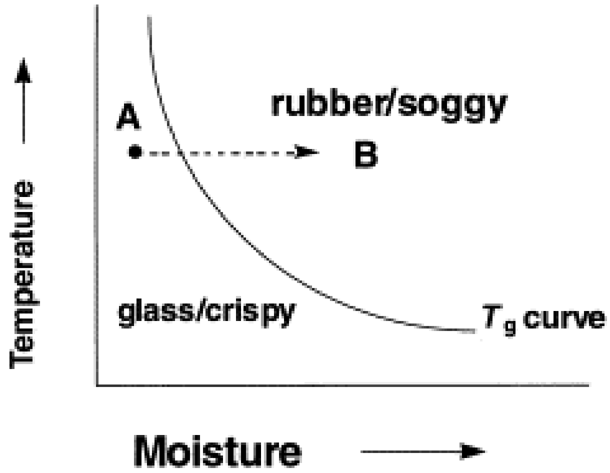

Figure 2. Schematic path in the state diagram for a crispy cracker absorbing water from the atmosphere at constant temperature, showing the crisp (A) to rubbery (B) phase transition (*28*).

1.3 Bread and bread components

Bread is a staple food in many countries in the world. Some breads in western Europe like the conventional Dutch loaf are not especially known for the crispness of their crusts because these breads are stored in plastic bags where they loose crispness very quickly. However other types of bread like a French baguette or a German "Kaiser brödchen" are known for their crispness. The quality of the crusts of these breads mainly deteriorate because of the water uptake from the crumb below and as a result toughening of the crust. For these type of products a longer shelf life in terms of crispness can be of importance (for instance this could mean baking bread 2 times a day instead of 3 times a day, or less waste of bread due to loss of quality). Figure 3 shows a schematic picture of a slice of bread with directions of water transport. A crust of a fresh bread contains around 12% water and the crumb of a fresh bread around 46% water (*7*).

The main ingredient of bread is wheat flour. Wheat flour mainly consists of the starchy endosperm of the wheat kernel. The main components of the flour are carbohydrates, proteins, lipids and non-starch polysaccharides. They will be briefly discussed below.

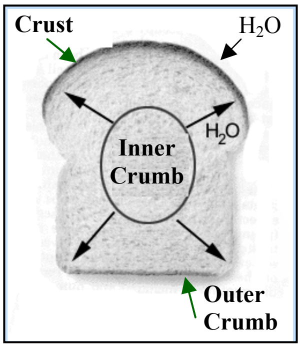

Figure 3. Schematic picture of a slice of bread. The directions of the transport of the water after baking are indicated with arrows.

1.3.1 Wheat flour components

1.3.1.1 Starch

A wheat flour contains around 80% starch (based on dry matter). The native starch granules are between around 5 and 55 µm in size and consist of two different types of polymers: amylose (around 30%) and amylopectin (around 70%). Amylose is a linear molecule of α-1,4 linked D-glucopyranosyl units and amylopectin is a branched molecule of α-1,4 linked D-glucopyranosyl units with about one α-1,6 link each 20 monomer units (*33*).

During milling some starch granules can be mechanically damaged. Aerated breads such as baguettes and pan breads are very sensitive to starch damage. The optimal amount of damaged starch is between 5 and 8%. This will give the highest bread volume. Damaged starch improves the hydration potential of the flour and thus leads to more water in the crumb after baking. However, to much water can lead to sticky dough that can't be handled or processed. Damaged starch granules are also more easily accessible to enzymes like amylases. The enzymes will increase the amount of maltose and dextrins in the dough. This will lead to more CO_2 and alcohol production by the yeast and a more intense crust coloration due to caramelization and Maillard's reaction.(*34*).

In excess of water, gelatinization of the starch granules can take place upon heating. If not enough water is available gelatinization will not or only partly take place. Gelatinization means that first the linear amylose molecules will leak out of the granule and in the presence of enough water and heat (52-85°C) the starch granule will fall apart (*33*). This process occurs in bread crumb during baking because there is enough water and the temperature is high enough.

1.3.1.2 proteins

The second largest component present in wheat flour is protein (content 10-15%). The largest part of these proteins have the unique properties that enable the production of a viscoelastic dough with good gas holding abilities and the preparation of a proper bread due to the fact that they are mainly water insoluble. Osborne (*35*) classified the wheat flour proteins into four groups based on their differences in solubility: albumins that are soluble in water, globulins soluble in salt solutions, gliadins soluble in 70% ethanol and glutenins partly soluble in dilute acid or alkali. The major part of wheat proteins (80%) consists of gliadins and glutenins, with the glutenins being the proteins that primarily provide the structure to the dough (*36*). The gluten proteins have heat-setting properties and undergo crosslinking reactions in the temperature range 70-160°C which results in the formation of crumb and crust (*37*).

1.3.1.3 Other components

Lipids and non-starch polysaccharides (NSP) are two minor components in wheat flour. The lipid content varies between 1.5 and 2.5%. Pentosans (mainly arabinoxylans (AX)) are the most important NSPs and they can influence the water absorption of the flour. Pentosans are usually divided into water extractable pentosans (WEP, mainly water extractable AX) and water unextractable solids (WUS, mainly water unextractable AX). Wheat flour contains about 2-3% pentosans of which 20-25% are water soluble (*36*).

1.3.2 The bread baking process

The basic ingredients of a bread are wheat flour, water, yeast and salt. These components are mixed together and kneaded into a dough. Mixing has in this context three functions: (1) mixing of the ingredients, (2) developing the gluten in the dough so far that it results in a continuous gluten network with optimal gas retaining properties and (3) including air into the dough which will form the nuclei of the gas cells that grow out during the proofing and the baking stage. After mixing, the dough is left to rest for a certain time at a slightly elevated temperature (30°C) in order for the yeast to produce CO_2. This CO_2 enhances the growth of the gas cells during the proofing and the first part of the baking stage. During the later part of the baking gas cell growth is mainly brought about by gas expansion and the formation of water vapour. During baking of bread the dough surface is exposed to high temperatures (> 200 °C) and quick dehydration of the dough surface will take place. The temperature in the interior of the dough will rise much slower (up to 90-95°C at the end of the baking) and not so much dehydration will take place. This means that the crust will be set much earlier than the crumb. Whitworth and Alava (*38*) have followed the bread baking process by x-ray computed tomography (CT) during baking. They showed that when the crust of the bread became firm enough to resist the expansion of the dough, the dough was compressed against the crust, resulting in a relatively dense crust structure compared to the structure of the crumb. Zanoni et al (*39*) have followed the temperature profile of a bread crust during baking. They showed that the temperature of the bread crust rises up to around 150°C at the end of the baking process (after 30 minutes). Similar results were found for the crust temperature of rolls at the end of the baking process (*40*). The temperature of the centre of the bread reached around 95°C at the end of the baking process. The latter temperature rise is sufficient to gelatinize the starch in the dough and set the crumb structure. Under the high temperature conditions of the crust, starch and proteins are involved in different kinds of reactions (dextrinization, caramelization, non-enzymatic browning reactions, thermal degradation, etc.) that lead to irreversible changes. These reactions result in the formation of a brown colour on the crust and aromatic substances that give the typical smell of a fresh baked bread (*41*). Due to the quick loss of water from the crust the starch in the crust is not completely gelatinized (*40, 42*). This lead the latter authors to the assumption that a bread crust is a network of gluten protein with starch granules embedded. This was supported by CSLM observations as shown in figure 4 (*7, 43*). The left side of the picture shows the crust, where the intact starch granules (green) can be seen, embedded in the gluten network (red). The structure is relatively dense on the crust side of the picture. Moving to the right in the picture, the air bubbles become less compressed and the structure less dense. The intact starch granules disappear and a transition to a mixed network of starch and protein can be seen when moving to the crumb side of the picture.

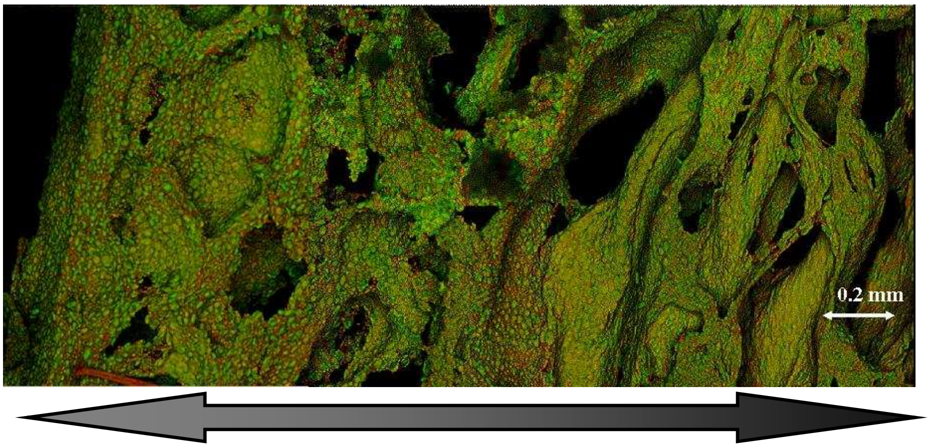

Figure 4. 3D CSLM image of a transverse section of bread crust. The left side of the image corresponds to the crust (light grey) and the right side (black) to the crumb. Starch is stained (green) using FITC and proteins (red) using Rhodamine B.

1.4 Scope

A better understanding of the process behind loss of crispness is necessary to improve the time that a composite food with a crispy part maintains this crispy part. With respect to storage stability, it is important not only to know the total uptake of water at a certain environmental relative humidity (RH) (as determined by thermodynamics) but also to know the kinetics of water uptake. This is because the faster the water uptake, the faster the loss of crispiness. Therefore in this thesis the water uptake kinetics are determined to evaluate the speed at which the loss of crispness takes place. In addition properties like water proton mobility, glass transition, sensorial properties and water content-water activity relations are evaluated at steady state conditions. This is to obtain better understanding of how water uptake affects crispness. Ingredient variations are applied to evaluate the effect of composition and structure on above mentioned properties.

1.5 Outline of this thesis

In Chapter 2 and 3 a methods are described allowing the analyses of water sorption uptake rates of bread crusts. The way water is transported into the bread crust crumbles is discussed. Chapter 2 differentiates between the water transport to the crust and the water transport into the solid matrix. Chapter 3 describes the water transport mechanism into the crust. Chapter 4 describes the relation between the sensorial loss of crispness of a model bread crust and the glass transition temperatures as determined by differential scanning calorimetry and phase transition analyses. A comparison is made between a whole crust and the separate main components gluten and starch. In Chapter 5 the relation between water content and water activity is discussed with respect to sensorial loss of crispness. At equal water activity a food material like bread crust can contain different amounts of water depending on the history of the sample. This effect is called hysteresis. This is the basis for this chapter in which crispness at different water activity-water content combinations is evaluated. Chapter 6 describes the effect of ingredient variation and morphology on the water uptake kinetics. Here solutions for slowing down the water kinetics are suggested. Finally in Chapter 7 a general discussion is given.

References

1. Katz, E. E.; Labuza, T. P., Effect of water activity on the sensory crispness and mechanical deformation of snack food products. *Journal of Food Science* **1981,** 46, 403-409.
2. Fontanet, I.; Davidou, S.; Dacremont, C.; Meste, M. l., Effect of water on the mechanical behaviour of extruded flat bread. *Journal of Cereal Science. 1997;* **1997,** 25, (3), 303-311.
3. Roudaut, G.; Dacremont, C.; le Meste, M., Influence of water on the crispness of cereal-based foods: acoustic, mechanical, and sensory studies. *Journal of Texture Studies* **1998,** 29, (2), 199-213.
4. Valles Pamies, B.; Roudaut, G.; Dacremont, C.; le Meste, M.; Mitchell, J. R., Understanding the texture of low moisture cereal products: mechanical and sensory measurements of crispness. *Journal of the Science of Food and Agriculture* **2000,** 80, (11), 1679-1685.
5. Peleg, M., A mathematical model of crunchiness/crispness loss in breakfast cereals. *Journal of Texture Studies* **1994,** 25, (4), 403-410.
6. Sauvageot, F.; Blond, G., Effect of water activity on crispness of breakfast cereals. *Journal of Texture Studies* **1991,** 22, 423-442.

7. Primo-Martín, C.; van de Pijpekamp, A.; van Vliet, T.; de Jongh, H. H. J.; Plijter, J. J.; Hamer, R. J., The role of the gluten network in the crispness of bread crust. *Journal of Cereal Science* **2006,** 43, (3), 342-352.

8. Luyten, H.; Plijter, J. J.; van Vliet, T., Crispy/crunchy crusts of cellular solid foods: a literature review with discussion. *Journal of Texture Studies* **2004,** 35, (5), 445-492.

9. Roudaut, G.; Dacremont, C.; Pamies, B. V.; Colas, B.; le Meste, M., Crispness: a critical review on sensory and material science approaches. *Trends in Food Science and Technology* **2002,** 13, (6-7), 217-227.

10. Vincent, J., The quantification of crispness. *Journal of the Science of Food and Agriculture* **1998,** 78, (2), 162 - 168.

11. van Vliet, T.; Walstra, P., Large deformation and fracture behaviour of gels. *Faraday Discussions* **1995,** 101, 359 - 370.

12. Atkins, A. G.; Mai, Y. M., Elastic and plastic fracture. *Ellis Horwood, Chichester* **1984**.

13. van Vliet, T.; Luyten, H., Fracture mechanics of solid foods. *New physico-chemical techniques for the characterization of complex food systems, edited by E. Dickinson, Chapman & Hall, Cambridge, UK* **1995**, p157-176.

14. Luyten, H.; Van Vliet, T., Acoustic emission, fracture behavior and morphology of dry crispy foods: A discussion article. *Journal of Texture Studies* **2006,** 37, 221-240.

15. Attenburrow, G. E.; Davies, A. P.; Goodband, R. M.; Ingman, S. J., The fracture behaviour of starch and gluten in the glassy state. *Journal of Cereal Science* **1992,** 16, (1), 1-12.

16. Vickers, Z. M., Sensory, Acoustical and Force-Deformation Measurements of Potato Chip Crispness. *Journal of Food Science* **1987,** 52, (1), 138-140.

17. Edmister, J. A.; Vickers, Z. M., Instrumental acoustic measures of crispness in foods. *Journal of Texture Studies* **1985,** 16, (153-167).

18. Dacremont, C.; Colas, B.; Sauvageot, F., Contribution of air- and bone conduction to the creation of sounds perceived during sensory evaluation of foods. *Journal of Texture Studies* **1991,** 22, 443-456.

19. Slade, L.; Levine, H., A food polymer science approach to structure-property relationships in aqueous food systems: non-equilibrium behavior of carbohydrate-water systems. *In Water Relations in Foods (L. Slade and H. Levine, eds.) pp. 29–101, Plenum press, New York.* **1991**.

20. Meste, M. l.; Roudaut, G.; Davidou, S., Thermomechanical properties of glassy cereal foods. *Journal of Thermal Analysis* **1996,** 47, 1361-1375.

21. Meste, M. l.; Champion, D.; Roudaut, G.; Blond, G.; Simatos, D., Glass transition and food technology: a critical appraisal. *Journal of Food Science;* **2002,** 67, (7), 2444-2458.

22. Montes, H.; Mazeau, K.; Cavaille, J. Y., The Mechanical beta relaxation in amorphous cellulose. *Journal of Non-Crystalline Solids* **1998,** 235-237, 461-421.

23. Roca, E.; Guillard, V.; Guilbert, S.; Gontard, N., Moisture migration in a cereal composite food at high water activity: Effects of initial porosity and fat content. *Journal of Cereal Science* **2006,** 43, 144-151.

24. Fu, Y.-C.; Tong, C. H.; Lund, D. B., Moisture migration in solid food matrices. *Journal of Food Science* **2003,** 68, (8), 2497-2503.

25. Roberts, J. S.; Tong, C. H.; Lund, D. B., Drying kinetics and time-temperature distribution of pregelatinized bread. *Journal of Food Science* **2002,** 67, (3), 1080-1087.

26. Guillard, V.; Broyart, B.; Bonazzi, C.; Guilbert, S.; Gontard, N., Moisture diffusivity in sponge cake as related to porous structure evaluation and moiture content. *Journal of Food Science* **2003,** 68, (2), 555-562.

27. Roca, E.; Broyart, B.; Guillard, V.; Guilbert, S.; Gontard, N., Controlling moisture transport in a cereal poreus product by modification of structural or formulation parameters. *Food Research International* **2007,** 40, 461-469.

28. Labuza, T. P.; Hyman, C. R., Moisture migration and control in multi-domain foods. *Trends in Food Science and Technology* **1998,** 9, 47-55.

29. Becker, H. A.; Sallans, H. R., A study of internal moisture movement in the drying of the wheat kernel. *Cereal Chemistry* **1955,** 32, (3), 212-226.

30. Roman-Gutierrez, A. D.; Mabille, F.; Guilbert, S.; Cuq, B., Contribution of specific flour components to water vapor adsorption properties of wheat flours. *Cereal Chemistry* **2003,** 80, (5), 558-563.

31. Crank, J., The mathematics of diffusion, 2nd Ed. Oxford University Press: Great Britain. **1975**.

32. Roman-Gutierrez, A. D.; Guilbert, S.; Cuq, B., Distribution of water between wheat flour components: a dynamic water vapour adsorption study. *Journal of Cereal Science* **2002,** 36, (3), 347-355.

33. Damodaran, S.; Parkin, K. L.; Fennema, O. R., Fennema's food chemistry. *CRC Press, Taylor and Francis Group, Boca Raton, USA* **1996**.

34. Belderok, B., Effect of starch damage in wheat flour on dough and bread properties. *Getreide, -Mehl-und-Brot* **1982,** 36, (7), 179-183.

35. Osborne, B. G.; Voorhees, C.G., *American Chemical Journal* **1893,** 15, 392.

36. Pyler, E. J., Baking Science and Technology, Volume 1. *Sosland Publishing Company, Kansas City, USA* **1988**.

37. Kokini, J. L.; Cocero, A. M.; Madeka, H.; de Graaf, E., The development of state diagrams for cereal proteins. *Trends in Food Science & Technology* **1994,** 5, 281-288.

38. Whitworth, M. B.; Alava, J. M., Non-destructive imaging of bread and cake structure during baking. *Proceedings of the 12th International ICC Cereal and Bread Congress, Harrogate, UK, 23-26th May 2004* **2005**.

39. Zanoni, B.; Peri, C.; Pierucci, S., A Study of the Bread-Baking Process. I: A Phenomenological Model. *Journal of Food Engineering* **1993,** 19, 389-398.

40. Primo-Martín, C.; van Nieuwenhuijzen, N. H.; Hamer, R. J.; van Vliet, T., Crystalline changes in wheat starch during the bread-making process: starch crystallinity in the bread crust. *Journal of Cereal Science* **2007,** 45, 219-226.

41. Cuq, B.; Abecassis, J.; Guilbert, S., State diagrams to help describe wheat bread processing. *International Journal of Food Science and Technology* **2003,** 38, (7), 759-766.

42. Pomeranz, Y.; Meyer, D.; Seibel, W., Wheat, wheat-rye and rye dough and bread studied by scanning electron microscopy. *Cereal Chemistry* **1984,** 61, 53-59.

43. Primo-Martín, C.; van Nieuwenhuijzen, N. H.; Hamer, R. J.; van Vliet, T., Crystalline changes in wheat starch during the bread-making process: starch crystallinity in the bread crust. *Journal of Cereal Science* **2007,** 45, 219-226.

44. Hopkinson, I.; Jones, R. A. L.; Black, S.; Lane, D. M.; McDonald, P. J., Fickian and Case II diffusion of water into amylose: a stray field NMR study. *Carbohydrate polymers* **1997,** 34, 39-47.

Chapter 2 Oscillatory water sorption test for determining water uptake behaviour in bread crust

Abstract
In this work, water sorption kinetics of bread crust are described using an oscillatory sorption test in combination with a Langmuir type equation. Both kinetic and thermodynamic information could be obtained at the same time. An advantage of applying a Langmuir type equation for a quantitative description of the water uptake kinetics is that no prior knowledge is necessary with respect to shape and surface area of the sample. It was shown that adsorption and desorption of water to the bread crust particle surface is much faster than the experimental time used (15 minutes at minimum). From this we may conclude that diffusion of water into the solid matrix is the rate limiting step in the water sorption process. The method also allows to calculate a Gibbs free energy. The method is suitable for use up to RH 60%.

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2.1 Introduction

The crispy / crunchy characteristics of crusts are closely related to the fracture behaviour of the solid matrix and the morphology of the crust. The solid matrix should break in a brittle way while sound is emitted. This character is lost at a higher water activity (*1*), (*2*). For example, for bread crust this is at a water activity above approximately 0.6 (*3*). It is especially difficult to keep a food crispy when the product has a moist interior. Water then migrates from the interior to the crust causing hydration of the crust components. This results in an increase of the mobility of the macromolecules, which causes a brittle to ductile transition of the amorphous regions of the crust (mainly proteins and carbohydrates) that were initially in the brittle state (*4*). Roughly at the same time a loss of the crispy behaviour occurs (*5*), (*6*). This makes the product less desirable for the consumer because of the loss of perceived freshness. To improve crispy behaviour, more information is required about the mechanisms underlying the deterioration of crispiness. Since it is clear that water causes the loss of crispiness, it is of interest to investigate the state of water in a food product, the kinetics of water uptake and the mechanism of water binding in a food product.

General water uptake (equilibrium conditions) is usually described by using a sorption isotherm. Several models are fitted to these curves to describe water uptake (*7*). However, with respect to storage stability it is not only important to know the total uptake of water at a certain environmental relative humidity (RH) (as determined by thermodynamics) but also to know the kinetics of water uptake. This is because: the faster the water uptake, the faster the loss of crispiness. Often researchers use Fick's law to describe water sorption kinetics (*8, 9*). Fick's first law describes water diffusion related to Brownian motions. The diffusion coefficient depends on local water concentration (*10*). Others used empirical models like the Weibull model or a mathematical model that accounts both for polymer relaxation and stochastic diffusion to describe water uptake kinetics of dry foods (*11*), (*12*), (*13*), (*14*). The Weibull model can be used to determine the process mechanism of water uptake. Cunha *et al* (*13*) did this to describe water uptake processes controlled by internal diffusion, external resistance to mass transfer and relaxation phenomena. A disadvantage of this model is the lack of a theoretical basis (*11*).

Roman-Gutierrez *et al* (*9, 15*) studied the rate of water uptake of a hard and a soft wheat flour and of different flour components. Diffusion coefficients of water in starch or wheat flour systems were determined. Roman-Gutierrez *et al* (*9*) used Fick's law to interpret sorption rates of the flour components (water uptake versus time). They did not observe a difference in initial sorption rates for the selected wheat components, except for the water-insoluble pentosans. These adsorbed water faster than all other components.

Del Nobile *et al.* (*14*) used an oscillatory sorption test for determining the water transport properties of chitosan based films. In this experiment they used cycles of oscillating water pressures. The water pressure levels around which the water pressure oscillated was for every cycle set at a higher level as well. They proposed an anomalous diffusion model and a non-ideal Fickian model to fit the experimental data, in the latter they did not take relaxation phenomena into account. The anomalous diffusion model accounts for both stochastic diffusion and polymer matrix relaxation, whereas the non-ideal Fickian model neglects the polymer matrix relaxation phenomenon (*16*). Both proposed models fitted well to stepwise water sorption data, but only the anomalous diffusion model fitted well to the oscillatory sorption data. These results suggest that when the water sorption kinetics are controlled by polymer relaxation, an oscillatory sorption test should be used instead of stepwise sorption tests. The use of an oscillatory sorption test will allow a better determination of the relationship between the water diffusion coefficient and the local water concentration (*14*).

A disadvantage of using Fick's law for describing the diffusion of water into a bread crust particle is the requirement to make an assumption regarding the particle surface to volume ratio. This is mostly done by assuming a spherical shape of the particle, which is not the case for bread crust particles. Besides that, Fick's law results in a diffusion constant of water which is an average of the contributions from the adsorption on the surface of the particles and the diffusion into the particle and assuming no matrix relaxation. When results obtained by applying Fick's law are interpreted as diffusion inside a bread particle, instant adsorption of the water at the surface of the material is assumed. Currently it is unclear whether this assumption is correct.

In this article a combination of an oscillatory sorption test and the use of a Langmuir type equation (*17*) is proposed to allow a quantitative analysis of water uptake behaviour without prior knowledge of shape and surface area of the sample. By varying the oscillation time it should in principle be possible to differentiate between water adsorption/desorption from the surface and diffusion of water into the particle.

In the oscillatory sorption experiment described in this paper the air RH oscillates between two different values. Water uptake and release during the time span of the oscillation period involved are measured with a microbalance. In contrast to the method applied by del Nobile *et al* (*14*) oscillations in RH are applied until a steady state has been reached. Water sorption kinetics can be investigated as a function of particle size, oscillation time, the A_w range studied and product properties. How the weight changes in reaction to a change in air relative humidity depends on the rate of transport to the surface, on the rate of adsorption and desorption of water at the surface of the sample and on the rate of diffusion of the water into the sample. The water sorption kinetics are described using a very simple model based on an equation equal to the equation derived by Langmuir (*17*). Besides that in this article the use of our model for the calculation of an imaginary surface area of the sample will be discussed.

2.2 Materials and methods

2.2.1 Materials

Bread crusts baked of a soft wheat flour (Soissons) were used. The flour was purchased from Meneba (Meneba Meel BV, Rotterdam, the Netherlands). The composition of the flour is shown in table 1.

2.2.2 Methods

2.2.2.1 Part-baked breads

Part-baked rusk rolls (8 cm diameter, 4 cm height) were prepared at the TNO baking laboratory (TNO Quality of life, Zeist, the Netherlands). Wheat flour (3000 g), water (1695 mL), based on farinograph water absorption (*18*), yeast (50 g), salt (70 g) and ascorbic acid (20 ppm) were mixed in a high speed mixer (Kemper SP 15, Kemper, the Netherlands) as explained below. First, all ingredients (temperature 5°C) except water were dry mixed for one minute at low speed. Next water (10°C) was added, and everything was mixed at low speed for two minutes. Finally, the dough was kneaded at high speed until a dough temperature of 26°C was reached. After mixing, the dough was allowed to rest for 15 min. Next the dough was divided and rounded. Proofing was performed at 30°C and 80% RH until a fixed volume of the dough sample (500 mL SJA) was reached. The breads were part-baked at 215 °C during 12 min in a Rototherm RE oven (Haton, the Netherlands). The part-baked breads were allowed to cool down for 30 min and frozen at -30°C and stored at –18 °C until use.

Breads were baked off in a Bakermat Mastermind oven (Leventi, Gilze, the Netherlands). The bake off conditions were as follows: pre-heating of the oven at 250°C followed by 5 s steam injection and convection heating during 5 min at 235°C.

After bake off, the breads were allowed to cool down during 0.5 h at ambient temperature (fresh bread). Next the crust (2 mm thickness) was separated from the crumb with a slicing machine (250 TC, Manconi&C, Sumirago-Varese, Italy). The crust samples were milled in a coffee grinder for 10 seconds at a time with a maximum of 3 times (to prevent heating of the sample) until a fine powder was obtained. The powder was sieved with different sieves (63µm, 0.25 mm and 0.5 mm) and the different sieve fractions (<63µm, 63µm-0.25mm and 0.25 mm-0.5 mm, called S, M and L, respectively) were freeze dried. The largest sieve fraction (>0.5 mm) was discarded. Samples were kept at room temperature in closed tubes until use.

2.2.2.2 Oscillatory sorption experiments

The oscillatory sorption experiments were performed using a VTI-SGA 100 symmetric vapor sorption analyzer (VTI Corporation, Hialeah, Florida USA). This is a continuous vapour flow sorption instrument for obtaining water and organic vapour isotherms. The instrument is equipped with a dew point analyser, and a Cahn microbalance. All tests on crust samples (5-7 mg with an approximate thickness of the powder bed of 0.5 mm) were performed at 25°C. Every experiment was started with a drying step at 50°C during 120 min (this resulted in a water content at the start of each experiment of approximately 2.5% for all samples). The oscillation experiment was performed by applying block wise oscillations of the environmental relative humidity (RH) between 60 and 80% RH, 50 and 70% RH, 40 and 60% RH and between 20 and 40% RH, respectively. The rising and falling times of each block were 4.5 minutes (figure 1). The isotherm was determined by changing the RH every 570 minutes or when the isotherm was stable (less than 0.0001% weight change in 10 minutes). The weight change of the sample (caused by the change in environmental RH) was measured in time. The measurements were stopped when the oscillation of the sample weight became steady. Two different oscillation times were used (28 and 48 minutes).

2.2.2.3 Differential Scanning Calorimetry (DSC)

Crust and crumb samples (around 15 mg) were weighed and distilled water was added to a 3:1 ratio of water to the sample in stainless steel pans (TA Instruments Inc., USA). DSC measurements were performed with a Perkin Elmer DSC 7 calorimeter (Perkin Elmer Corp., USA). Indium was used for calibration of the system. The samples were heated from 10 to 130°C at 10°C/ min. An empty stainless steel pan was used as a reference during the DSC measurement. The enthalpy was expressed in J/g of sample (*19*). Relative crystallinity (RC) was calculated as:

$$RC = \frac{\Delta H_s}{\Delta H_f} * 100 \tag{2.1}$$

where ΔH_s represents the melting enthalpy (J/g) of starch crystallites in the sample and ΔH_f represents the enthalpy (J/g) of starch gelatinization in the flour.

2.2.2.4 Protein analysis

Protein content was determined according to the DUMAS method using a NA2100 Nitrogen and Protein Analyser (CE Instruments). Methionine was used as a standard (*20*).

2.2.2.5 Total starch

The total starch content was measured using a total starch assay kit from Megazyme (α-amylase/amyloglucosidase method) (Megazyme International Ireland, Wicklow, Ireland).

Table 1. Characteristics of Soissons wheat flour (db), results from Primo-Martin *et al* (*3*).

Parameter	Value	Parameter	Value
Water absorption (%)[a]	56.5	Starch (%)	77
Moisture (%)	14.1	Damaged starch (%)	6.0
Ash (%)	0.58	Energy input (J)[b]	$290 \cdot 10^{-4}$
Protein (%)	10.9	Falling number	333

a: farinograph (Brabender), b: alveograph (Chopin).

Table 2 DSC results and protein content for different sized fractions of Soissons bread crust particles

Size	Relative cristallinity (%)	Stdev	Protein (% dm)	Stdev	Total starch (% dm)	Stdev
Large	23	3.1	10.9	-	74	1
Medium	21	1.0	10.9	0.2	73	1
Small	18	2.5	11.0	0.1	n.d.	-

2.3 Theory

Analysis by using a simple one exponential equation

Weight change ($\Delta m(t)$) was defined as follows:

$$\Delta m(t) = \frac{M(t) - M(0)}{M(0)} \quad \text{(g water/g dry weight)} \tag{2.2}$$

where M(t) and M(0) are the weight at time t and the initial weight after the drying step of the sample, respectively.
For the analysis of the uptake of water by bread crust, this process is assumed to be expressed by:

$$H_2O(gas) \overset{k_{ads}}{\underset{k_{des}}{\rightleftarrows}} H_2O(adsorbed) \tag{2.3}$$

Then, the oscillation experiment can be described by an equation equal to the Langmuir equation (*21*).
When using this equation we assume that all sorption sites are equal and that the adsorption energy is independent of the degree of coverage of the surface.
With these assumptions we can express the change in weight in time as (*21*):

$$\frac{\mathrm{d}\Delta m(t)}{\mathrm{d}t} = -k_{des}\Delta m(t) + k_{ads}P_i(\Delta m_{\max} - \Delta m(t)) \tag{2.4}$$

where k_{ads} is the apparent rate constant for adsorption (s^{-1} Pa^{-1}), k_{des} the apparent rate constant for desorption (s^{-1}), P_i is the water vapour pressure (Pa), *t* is time and Δm_{max} is the maximum weight increase. The latter parameter has only a physical meaning when the properties of the adsorbent do not change as a function of the amount of water adsorbed. In many practical cases, above a certain degree of water uptake the material will begin to swell, introducing a time dependence in Δm_{max} which will in theory eventually lead to an infinite value for Δm_{max}, because the system has become a solution. Moreover, a change in adsorbate properties due to swelling may affect the adsorption energy and with that k_{des} and k_{ads}.
The rate of the change of surface coverage due to adsorption is proportional to *P* and the amount of available surface area (i.e. number of sorption sites) (Δm_{max} - $\Delta m(t)$). Eqn. 2.4 can be solved as follows:

$$\mathrm{d}t = \frac{\mathrm{d}\Delta m(t)}{-(k_{des} + k_{ads}P_i)\Delta m(t) + \Delta m_{\max} k_{ads} P_i} \tag{2.5}$$

Integration of equation 2.5 gives:

$$t = \frac{-1}{(k_{des} + k_{ads}P_i)} \ln \frac{-(k_{des} + k_{ads}P_i)\Delta m(t) + \Delta m_{\max} k_{ads} P_i}{c} \tag{2.6}$$

$$e^{-(k_{des}+k_{ads}P)t} = \frac{-(k_{des} + k_{ads}P_i)\Delta m(t) + \Delta m_{\max} k_{ads} P_i}{c} \tag{2.7}$$

Where c is an integration constant. This gives the next expression for Δm(t):

$$\Delta m(t) = \frac{\Delta m_{\max} k_{ads} P_i}{k_{des} + k_{ads}P_i} - \frac{c}{k_{des} + k_{ads}P_i} e^{-(k_{des}+k_{ads}P_i)t} \tag{2.8}$$

When *t* is 0 then:

$$\Delta m(0)' = \frac{\Delta m_{\max} k_{ads} P_i}{k_{des} + k_{ads}P_i} - \frac{c}{k_{des} + k_{ads}P_i} \tag{2.9}$$

$$c = -\Delta m(0)' * (k_{des} + k_{ads}P_i) + \Delta m_{\max} k_{ads} P_i \tag{2.10}$$

this leads to:

$$\Delta m(t) = \left[\Delta m(0)' - \frac{\Delta m_{\max} k_{ads} P_i}{k_{des} + k_{ads}P_i} \right] e^{-(k_{des}+k_{ads}P_i)t} + \frac{\Delta m_{\max} k_{ads} P_i}{k_{des} + k_{ads}P_i} \tag{2.11}$$

where $\Delta m\,(0)'$ is the weight change at the start of the part of experiment considered. If the experiment is started from the dry state $\Delta m\,(0)'$ is 0 and when considering the water uptake kinetics for one oscillation step $\Delta m\,(0)'$ is equal to $\Delta m(t)$ at the start of the oscillation step. The fit results of the last 5 oscillation steps (when the oscillations were visually steady) were averaged to obtain a value for the adsorption/desorption of water in a single oscillation step. To avoid the need for a third adjustable parameter we assume that the thermodynamics of the upward and downward water sorption process are equal. Then we assume that the equilibrium constant

$$K = \frac{k_{ads}}{k_{des}} \tag{2.12}$$

and *Δm_{max}* are identical for both upward and downward branches of the oscillating data curves of *Δm(t)*. Note that K has the units Pa^{-1}. Consequently, fits of

$$\Delta m(t) = (\Delta m(0)' - a_0)e^{-t*a_1} + a_0 \tag{2.13}$$

to upward and down branches were carried out with a_0 and a_1 as adjustable parameters. From

$$\frac{a_{0h}}{a_{0l}} = \frac{P_h}{P_l}\frac{1+P_lK}{1+P_hK} \tag{2.14}$$

(P_h and P_l are the water vapour pressure values, and a_{0h} and a_{0l} are the fitted equilibrium weight change levels for the upward and downward oscillation branches, respectively) and

$$a_{0,i} = \frac{\Delta m_{\max} KP_i}{1+KP_i} \tag{2.15}$$

and

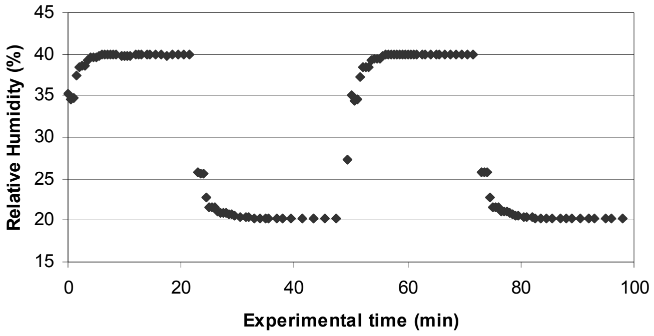
$$a_{1,i} = k_{des.i} + P_i k_{ads,i} \tag{2.16}$$

K, Δm_{max}, $k_{des,i}$ and $k_{ads,i}$ can be calculated. The subscript *i* equals '*h*' or '*l*' and refers to the upward and downward branches of the oscillating curve.

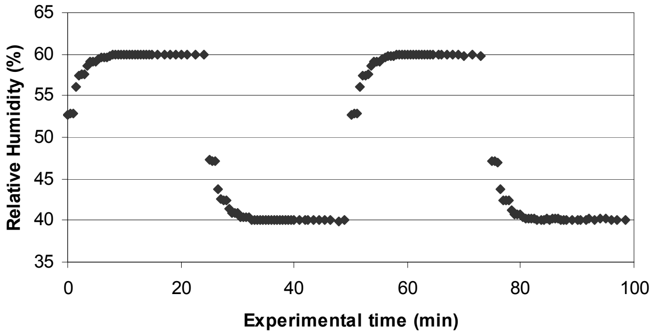
From the obtained equilibrium constant *K* and the water vapour pressure P_i the Gibbs free energy (ΔG) of the adsorption of one mole of water can be calculated as follows (*21*):

$$\Delta G = -RT \ln KP_i \tag{2.17}$$

where R is the gas constant (8.314 J/mol*K) and *T* is the temperature (298 K).



(a)



(b)

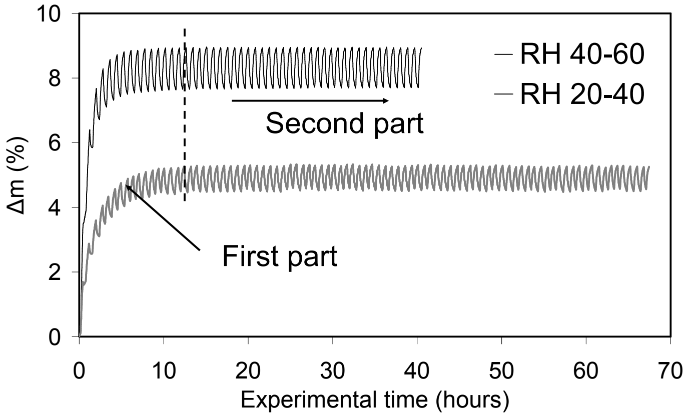
Figure 1 Change in applied relative humidity (RH) as a function of time for two relative humidity combinations in the VTI-SGA-100 analyser (a) 20 and 40% RH and (b) 40 and 60% RH respectively.

2.4 Results

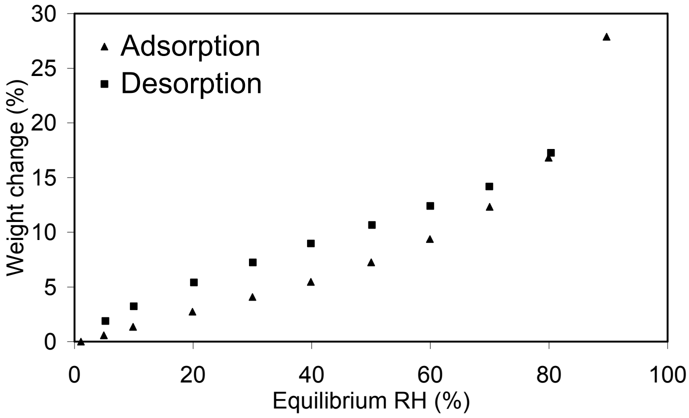
2.4.1 Reaction constants

Figure 2a shows an example of an oscillatory sorption experiment for two different relative humidities. The experiment can be divided into two parts; the first part in which a steady state is not yet reached and in which a large amount of water is taken up and a second part in which a steady oscillating state (five repetitive oscillation steps are the same within one experiment) is reached. In this study we will focus on the second part, the steady oscillating state situation. Figure 2a shows that the overall water uptake is less and proceeds at a lower rate at a lower air relative humidity as expected. The model was fitted to the separate oscillation steps in the second part of the experiment. Figure 3 shows the single exponential fit (equation 2.13) of the results obtained for Soissons bread crust with an oscillation time of 48 minutes for an upward oscillation step. Fits of the downward oscillations were similar (data not shown). From this graph can be concluded that a single exponential model fits the experimental data with an accuracy of $R^2 \cong 0.99$. Fitting of the single exponential model resulted in two kinetic parameters k_{ads}, and

k_{des}. The values for these two parameters are shown in figure 4a and 4b respectively for both the upward and downward oscillation branches. k_{ads} should not change with water content when the affinity for water does not change with water content. However, as can be seen in figure 4a, k_{ads} is lower for a higher air relative humidity range for both the upward and downward RH process. k_{des} is higher at a higher air relative humidity oscillation range (figure 4b). Both k_{ads} and k_{des} for the upward RH process are lower than for the downward RH process. At lower air RH (20-40%) this effect is clearer than at the higher air RH's (figure 5 shows the ratio for $k_{des,\ upward}/k_{des,\ downward}$, data for k_{ads} not shown). This could indicate that the material changes relatively stronger over the considered lower RH range. This complies with the fact that at lower RH the isotherm also shows the largest hysteresis effect (figure 2b). Apparently already hysteresis occurs at small changes in water content. Therefore the assumption that there are no differences between the parameters of the upward and downward oscillation branch (K and Δm_{max} are equal for upward and downward branch) cannot be maintained.



(a)



(b)

Figure 2 Oscillatory sorption experiments for bread crust particles (size large) of Soissons flour at 2 different relative humidities (40-60% and 50-70%) (a) and water sorption isotherm of Soissons bread crust large sized particles (b).

2.4.2 Gibbs free energy

The Gibbs free energy change due to water adsorption (dG) can give information on the direction of the reaction of water uptake or loss. The Gibbs free energy of the adsorption/desorption process was calculated from the ratio of $k_{ads}*P/k_{des}$. Figure 6 shows the results for Soissons bread crust with an oscillation time of 48 minutes. The Gibbs free energy goes to zero and reaches a positive value with increasing water content. An increasing Gibbs free energy with increasing RH indicates a weaker drive for water to adsorb onto crust with a higher water activity. A Gibbs free energy of zero suggests the absence of a preference for water molecules to be either in the vapour state or in the adsorbed state.

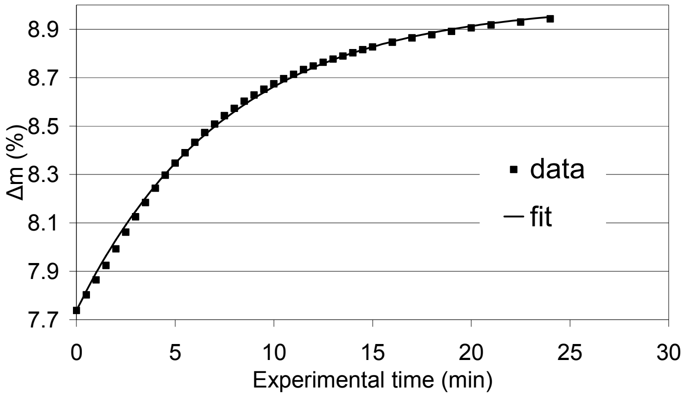
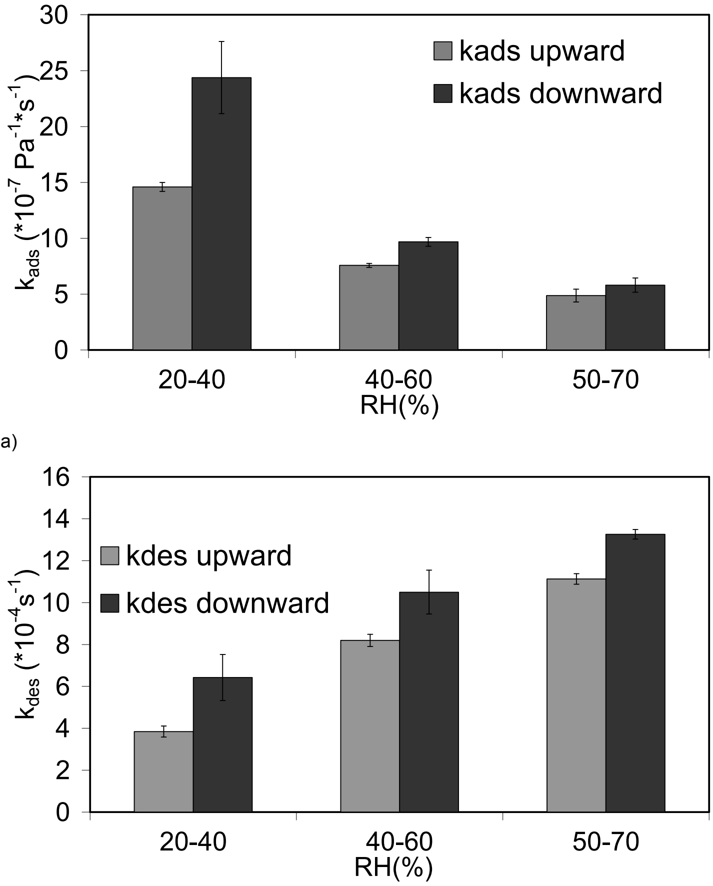


Figure 3 Fit of the single exponential type equation (eq 2.13) for large Soissons bread crust particles with a RH oscillating between 40 and 60%.

2.4.3 Effect of size

Figure 5 also shows that the difference between the upward and downward process is not influenced by the size of the particle. In other words, the activation energies for adsorption and desorption do not depend on the particle size. The other determined parameters are influenced by size. Figure 6 shows the effect of particle size on dG. dG was found to depend on the size of the particles at lower RH. In a lower air RH range dG becomes more negative with increasing sample size. Figure 7 shows that Δm_{max} increases with decreasing sample size. This indicates an increase in active “surface area” per unit mass for smaller particles. With an increasing air RH Δm_{max} also increases. This also indicates an increase in the number of sorption sites. Figure 8a and 8b show $k_{ads}*P$ and k_{des} for Soissons bread crust of different size classes of crust particles for the upward branch. $k_{ads}*P$ and k_{des} both decrease with increasing particle size. The diffusion distance of water into the particles will be longer for larger sized particles (average diameter of 0.19 mm as measured with a stereomicroscope) than for smaller sized particles (approximate average diameter of 0.01 mm) and therefore the water uptake process will go slower for larger particles.



(a)

(b)

Figure 4 k_{ads} (a) and k_{des} (b) as a function of air relative humidity (RH) for the absorption and desorption process for a large sized particle.

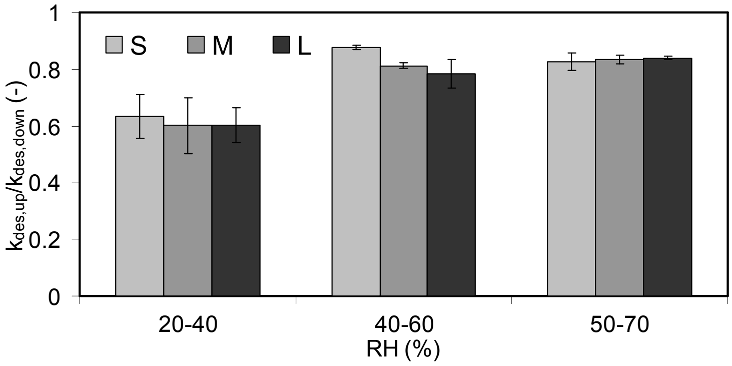


Figure 5 Hysteresis in k_{des} as a function of size (indicated) and air RH between the upward and downward branch of the RH oscillation.

2.4.4 *Effect of oscillation time*

No significant difference was found between oscillation times of 28 minutes and 48 minutes for any of the parameters (data not shown). That means that all processes that determine the kinetics and average level of water uptake have a characteristic time that is either well below 28 minutes or is much longer than 48 minutes. This conclusion is supported by the small size of the hysteresis found when comparing weight change curves at increasing and decreasing RH (i.e. the upward and downward branches of the oscillations).

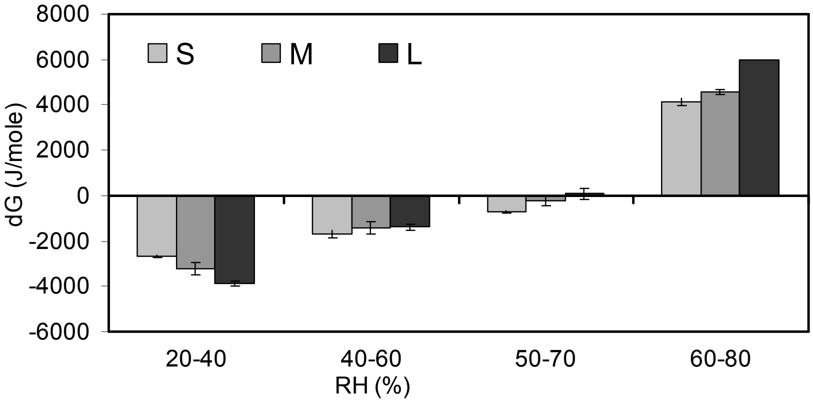


Figure 6 dG for Soissons bread crust with different particle sizes and for 3 different RH oscillation ranges.

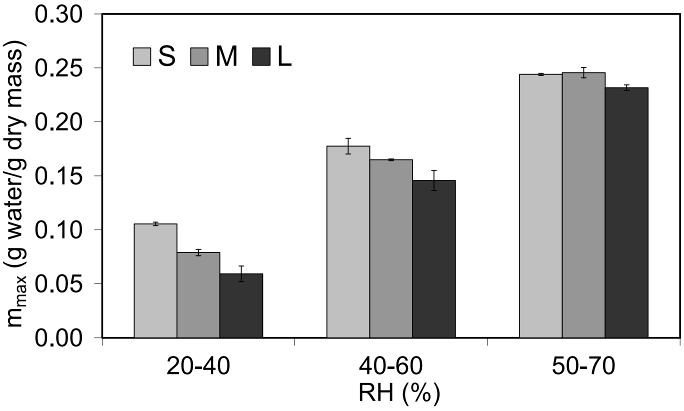
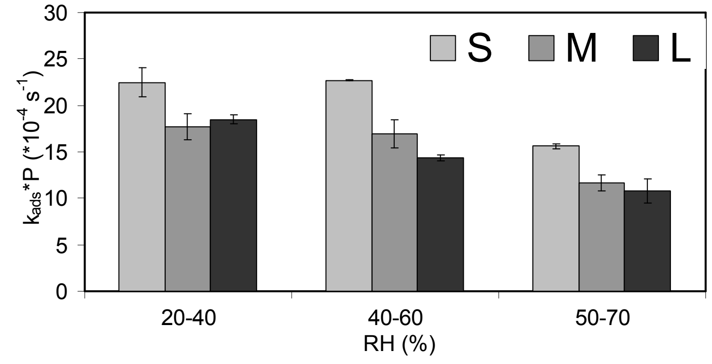
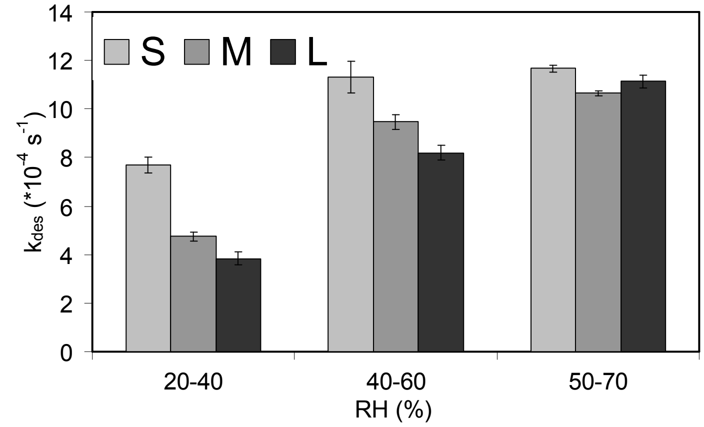


Figure 7 Imaginary maximum water uptake for Soissons bread crust with different particle sizes and for 3 different RH oscillation ranges.



(a)



(b)
Figure 8 k_{ads}*P for Soissons bread crust of different size and for different RH ranges (a), k_{des} for Soissons bread crust of different size and for different RH ranges (b).

2.5 Discussion

Our results show that it is possible to obtain new information (Δm_{max}, k_{ads}, k_{des}) from the combination of an oscillatory sorption test under steady state conditions and an analysis based on a single exponential model. The advantage of this method compared to the generally used Fick's law is that no information is required about geometry and size of the used sample because this information is included in one of the fit parameters of the model. This method allows in principle differentiation between diffusion of water into the matrix and adsorption and transport of water to the surface although we did not see the latter in our experiments.

No influence was found of the oscillation time between 28 and 48 minutes on parameters considered. Since the product also shows hysteresis, most likely we may conclude that adsorption and desorption processes at the surface and transport of water to the surface is much faster than the oscillation rates used. This means that diffusion of water into the matrix is the rate limiting step in the water uptake process. This also confirms one of the assumptions that is made when using Fick's law to describe the diffusion process; instant adsorption of the water to the surface of the product. The use of shorter oscillation times could be of interest, but with our equipment unfortunately this is not possible. The Langmuir parameters and Fickian diffusion can be linked together through k_{des}. Desorption is always activated because the particles have to be lifted from the bottom of a potential well (*21*). A higher k_{des} therefore indicates a smaller activation energy for water to jump from one place to another. Diffusion can be thought of as water molecules jumping from one place to another. The jumping distance is determined by the size of the voids/free volume in the system. For example a glassy system has more voids than a crystal, so faster diffusion (*22*). k_{des} increases with increasing RH in our bread crust particles, thus suggesting also faster diffusion of the water.

Both the adsorption and desorption rate tend to decrease with increasing particle size. Besides the difference in surface to volume ratio the smaller particles have a higher water content in the steady state (results not shown), likely due to a higher percentage of the material structurally affected by the penetration of water. Material into which water has penetrated has a higher ability to retain water and a higher water diffusivity. The latter could result in a higher k_{des} and k_{ads} for smaller particles. Δm_{max} of a smaller sized sample is larger; the surface area to volume ratio of a smaller sample is also larger and as above described this could increase the amount of water penetrating the sample and thus increasing Δm_{max}. Through the parameter Δm_{max} it is therefore possible to get an indication of the size of the active surface area per unit mass of the sample. The size effect on Δm_{max} is only a factor 2 whereas the average diameter differs by a much larger factor (approximately 10). An earlier study by image analysis (R.H. Tromp, unpublished data) showed that the 2-dimensional projections of larger sized toasted rusk roll crumb particles have a larger perimeter per surface area compared to smaller particles. In other words, the surface of larger particles is rougher than the surface of smaller particles. This would imply a relatively larger surface area for the larger particles than expected based on their diameter and the assumption of a smooth surface. Such an effect can explain the smaller difference between the Δm_{max} for the different sizes. The dependence of Δm_{max} on RH and particle size indicates that this parameter may be used as a tool to characterize the active sorption sites of particles from different bread sources. It is probably a relevant parameter to describe the difference in the rate of aging due to a change in humidity of bread crust with different ingredients. At higher water contents the differences between the Δm_{max} for the differently sized particles become smaller. This is probably caused by the fact that at a RH above 60% the assumption that all sorption sites are equal does not hold anymore.

The fact that dG is not the same for all sizes indicates that a change in the chemical composition of the sample may have occurred during sieving, although no difference in protein content, starch content or amount of gelatinized starch (100-RC) was found (table 2). The DSC thermograms of the bread crust particles showed a peak at around 70°C for all sizes which is similar to results found before (*23*). There could be a difference in non starch polysaccharide content (likely a lower content of non-starch polysaccharide in the small sized sample) which could cause a difference in dG. According to Roman-Gutierrez *et al.* (*24*) pentosans have a relatively high equilibrium water content compared to other wheat components. To clarify this point further research needs to be done.

k_{ads} and k_{des} of Soissons bread crust differ for different air relative humidities (both for the upward and downward branch) even within one oscillation experiment (figure 4a and b). This suggests that the material changes when the water content of the material changes. Due to the water uptake the material probably swells. This is also indicated by the increase in Δm_{max} with the applied RH range, which indicates an increased number of sorption sites. The variation of $\mathrm{k_{ads}}$ and $\mathrm{k_{des}}$ with RH could mean that the type or number of sorption sites changes (for example from highly reactive polar sites on the macromolecule at low relative humidity to binding sites with higher activation energies at higher relative humidities). The change in Gibbs free energy also confirms that a change in the material with a change in RH has occurred.

The hysteresis effect as shown between the upward and downward branch is not compatible with the absence of an influence of oscillation time on the considered parameters. The latter indicates that at all times the system is in equilibrium, whereas a hysteresis indicates a non-equilibrium. Further research is necessary to clarify this point. Taking smaller ranges of air RH could probably improve the experiment. These findings also indicate that the diffusion is likely not purely Fickian. More likely the diffusion mechanism lies somewhere between Fickian diffusion (case 1 diffusion) and case 2 diffusion, which also involves matrix relaxation. Case 2 diffusion occurs when the characteristic relaxation processes of the polymer are slower than that of the diffusion process and the diffusion is completely controlled by the polymer relaxation rate. This relaxation of the polymer causes a change in the effective local diffusion coefficient of the diffusing molecules. This behaviour is characterized by the presence of a sharp penetrant concentration profile that propagates into the polymer with a constant velocity (*25*). The hysteresis effect also indicates a change in the material. Then according to Del Nobile *et al.* (*14*) indeed an oscillatory sorption test should be used.

It should be noted that the ratio between the $k_{des\ upward}$ and $k_{des\ downward}$ is equal for all sizes, but changes with RH. This indicates that this parameter may be used as a material property. This ratio is lower at lower water content (RH 20-40) suggesting a difference in activation energy for the binding of water between the upward and downward branch. This indicates a more ordered state of the sample compared to the samples at higher RHs. With a larger part of the material in an ordered state this hysteresis effect will be larger and the ratio between $k_{des\ upward}$ and $k_{des\ downward}$ will be further away from 1. This ratio being 1 would mean that the material is behaving as a dilute solution. dG was calculated for the bread crust samples at an oscillation time of 48 minutes (figure 6). This table shows that the Gibbs free energy increases with increasing water content. The Gibbs free energy becomes zero somewhere in the water activity range of 0.5-0.7. This indicates a (generally broad) transition from a more adsorption dominated process to a more dilution dominated process. This water activity range is also the range at which the crispiness of the bread crust is lost (*3*). A positive value, as found at the highest RH, would mean a ’negative’ adsorption of water or an increase of the pressure of water vapour when it is exposed to a crust of a water activity of around 0.7. As this is unrealistic, it should be concluded that the single exponential-adsorption approach fails in the high RH regime (RH>60%), or that there is a change in the process that provides the energy for water adsorption. Most likely the ordered state of the bread crust at lower relative humidities finally dissolves. The point at which dG equals zero in this respect can be used as a rough indication to determine at which relative humidity the ordered state has disappeared.

As became clear from the results and as can be expected, the conditions which are assumed when using the one exponential approach were not or only approximately met during the process of water uptake by bread crust. However our approach does provide a framework within which this complicated process can be discussed.

The combination of the single exponential model approach and an oscillatory sorption tests allowed us to calculate kinetic and thermodynamic parameters at the same time under steady state or quasi equilibrium conditions.

The test shows that adsorption and desorption of water to the bread crust particle surface takes most likely place on a timescale of the order of minutes. From this we may conclude that diffusion of water into the solid matrix is the rate limiting step in the water sorption process. Besides that a method is developed that enables us to describe water sorption kinetics irrespective of the particle geometry, but with the possibility to obtain new information (k_{abs}, k_{des}) about the water sorption process. These rate parameters change with changing size of the particles and with changing water content of the sample and thus may help to understand the water sorption process and can give information on water uptake kinetics and on activation energies, necessary for the water to adsorb to or desorb from the particle. The parameter $k_{des\ upward}/k_{des\ downward}$ can be used to describe material properties since it was shown to be independent of particle size. The use of a simple one exponential model could be especially convenient for porous products, because it is difficult to determine the surface area of these samples. In this way it might be possible to compare different ingredient formulations with each other without knowing the exact particle sizes or porosities. The often used Fick's law to describe the water sorption process for example does require this information on surface area. It also appears possible to determine changes in the active sorption area (Δm_{max}) with changing particle size and water content. In order to validate the method better it will be necessary to evaluate more different materials. The results for the Gibbs free energy show that our approach cannot be used at a RH above 60% for bread crust particles.

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References

1. Meste, M. le; Champion, D.; Roudaut, G.; Blond, G.; Simatos, D., Glass transition and food technology: a critical appraisal. *Journal of Food Science* **2002,** 67, (7), 2444-2458.

2. Luyten, H.; Plijter, J. J.; van Vliet, T., Crispy/crunchy crusts of cellular solid foods: a literature review with discussion. *Journal of Texture Studies* **2004,** 35, (5), 445-492.

3. Primo-Martín, C.; van de Pijpekamp, A.; van Vliet, T.; de Jongh, H. H. J.; Plijter, J. J.; Hamer, R. J., The role of the gluten network in the crispness of bread crust. *Journal Of Cereal Science* **2006,** 43, (3), 342-352.

4. Nicholls, R. J.; Appelqvist, I. A. M.; Davies, A. P.; Ingman, S. J.; Lillford, P. J., Glass transitions and the fracture behaviour of gluten and starches within the glassy state. *Journal of Cereal Science* **1995,** 21, (1), 25-36.

5. Roudaut, G.; Dacremont, C.; le Meste, M., Influence of water on the crispness of cereal-based foods: acoustic, mechanical, and sensory studies. *Journal of Texture Studies* **1998,** 29, (2), 199-213.

6. Slade, L.; Levine, H., A food polymer science approach to structure-property relationships in aqueous food systems: non-equilibrium behavior of carbohydrate-water systems. *In Water Relations in Foods (L. Slade and H. Levine, eds.) pp. 29–101, Plenum press, New York.* **1991**.

7. Al Muhtaseb, A. H.; McMinn, W. A. M.; Magee, T. R. A., Moisture sorption isotherm characteristics of food products: a review. *Food and Bioproducts Processing* **2002,** 80, 118-128.

8. Becker, H. A.; Sallans, H. R., A study of internal moisture movement in the drying of the wheat kernel. *Cereal Chemistry* **1955,** 32, (3), 212-226.

9. Roman-Gutierrez, A. D.; Mabille, F.; Guilbert, S.; Cuq, B., Contribution of specific flour components to water vapor adsorption properties of wheat flours. *Cereal Chemistry* **2003,** 80, (5), 558-563.

10. Crank, J., The mathematics of diffusion, 2nd Ed. Oxford University Press: Great Britain. **1975**.

11. Fatima-Machado, M. de; Oliveira, F. A. R.; Gekas, V.; Singh, R. P., Kinetics of moisture uptake and soluble-solids loss by puffed breakfast cereals immersed in water. *International Journal of Food Science & Technology* **1998,** 33, (3), 225-237.

12. Marabi, A.; Livings, S.; Jacobson, M.; Saguy, I. S., Normalized Weibull distribution for modeling rehydration of food particulates. *European Food Research and Technology* **2003,** 217, (4), 311-318.

13. Cunha, L.M.; Oliveira, F.A.R.; Oliveira, J.C., Optimal experimental design for estimating the kinetic parameters of processes described by the Weibull probability distribution function. *Journal of Food Engineering* **1998,** 37, (2), 175-191.

14. Nobile, M.A. de; Buonocore, G.G.; Conte, A., Oscillatory sorption tests for determining the water-transport properties of chitosan-based edible films. *Journal of Food Science* **2004,** 69, (1), 44-49.

15. Roman-Gutierrez, A.; Sabathier, J.; Guilbert, S.; Galet, L.; Cuq, B., Characterization of the surface hydration properties of wheat flours and flour components by the measurement of contact angle. *Powder Technology* **2003,** 129, (1-3), 37-45.

16. Nobile, M. A. d.; Buonocore, G. G.; Altieri, C.; Battaglia, G.; Nicolais, L., Modeling the water barrier properties of nylon film intended for food packaging applications. *Journal of Food Science* **2003,** 68, (4), 1334-1340.

17. Langmuir, I., The adsorption of gases on plane surfaces of glass, mica and platinum. *Journal of the American Chemical Society* **1918,** 40, 1361-1403.

18. Methods, I. S., *International association for cereal chemistry. Method for using the Brabender Farinograph; No 115/1* **1991**.

19. Attenburrow, G. E.; Davies, A. P.; Goodband, R. M.; Ingman, S. J., The fracture behaviour of starch and gluten in the glassy state. *Journal of Cereal Science* **1992,** 16, (1), 1-12.

20. Adler-Nissen, J., Enzymatic hydrolysis of food proteins. *London Elsevier. Application Manual NA 200 Nitrogen and Protein Analyzer, CE Instruments* **1986**.

21. Atkins, P.; Paula, J., Atkins' Physical Chemistry, Oxford University Press, Oxford. **2002**.

22. Tromp, R. H.; Parker, R.; Ring, S. G., Water diffusion in glasses of carbohydrates. *Carbohydrate research* **1997,** 303, (2), 199-205.

23. Primo-Martín, C.; van Nieuwenhuijzen, N. H.; Hamer, R. J.; van Vliet, T., Crystalline changes in wheat starch during the bread-making process: starch crystallinity in the bread crust. *Journal of Cereal Science.* **2007***, 45(2), 219-226*.

24. Roman-Gutierrez, A. D.; Guilbert, S.; Cuq, B., Distribution of water between wheat flour components: a dynamic water vapour adsorption study. *Journal of Cereal Science* **2002,** 36, (3), 347-355.

25. Snively, C. M.; Koenig, J. L., Studying anomalous diffusion in a liquid crystal/polymer system using fast FTIR imaging. *Journal of Polymer Science: Part B: Polymer Physics* **1999,** 37, 2261-2268.

Chapter 3 The water uptake mechanism in crispy bread crust

Abstract
Crispness is an important quality characteristic of many types of food products like for example crispy rolls. Its retention is directly related to the kinetics of water uptake by the crust. In this study a method for the evaluation of the water sorption kinetics in bread crust is proposed. Two different sorption experiments were used; an oscillatory sorption test and a sorption test in which the air relative humidity (RH) was increased stepwise. These two experiments had different time scales, which made it possible to get a better understanding of the mechanisms involved.
Results show that the adsorption and desorption dynamics of the oscillatory sorption test could be described best by a single exponential in time and not by a Fickian type square root time dependence. Such an exponential behavior is often ascribed to Case II relaxation controlled diffusion. A maximum in the water uptake rate was found for a RH value between 50 and 70%. The rate parameters of the experiment where RH was stepwise increased were around a factor 10 lower than those derived from oscillatory sorption experiments. This indicates that the experimental time used effects the outcome of the experiment. This is an important factor when designing experiments for the determination of water uptake rates. In addition, also a parameter describing the time dependence of the rate parameters of the oscillatory sorption experiment was calculated (C). C was in the same range as the rate parameter of the isotherm experiment. This indicates that different (relaxation) processes are acting at the same time in the bread crust during water uptake.

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3.1 Introduction

Maintaining the sensory characteristics of crispy and crunchy foods during their shelf life can be very difficult if not impossible when the product is not in an equilibrium situation. This is the case when a crispy product is stored in a humid environment or when the crispy product is a composite food that also has a moist part. For example like a bread with a dry and crispy crust and a moist and soft crumb. Then moisture migration will take place from the environment or moist core to the dry and crispy parts. This results in a loss of sensorial crispness of the product (*1*-*3*). Since the amount of water present in the crust is an important factor in determining its crispness, the rate of loss of crispness will be related to the kinetics of water uptake by the crispy parts of the product.

When a dry solid polymer material like protein or starch is brought into contact with water, the water diffuses into the polymer and, in the case of an energetically favourable interaction between the water and the polymer, the polymer matrix swells. Diffusion involves migration of the water molecules into pre-existing or dynamically formed spaces between polymer chains. This first stage follows Fickian diffusion (*4*) for example in the case of organic vapour sorption (vinyl chloride, acetone and methanol) into glassy polymer powders (poly(vinylchloride)). Fickian diffusion in polymers is an ideal case of penetrant transport, corresponding to free diffusion of penetrant without interference of polymer chain rearrangement, ie structural relaxation (*5*). The second stage in penetrant sorption is the swelling of the matrix due to polymeric relaxations. These provide a redistribution of the free-volume elements which ultimately results in a time-dependent increase of free-volume to accommodate additional penetrant (anomalous penetrant transport). It is believed that for an initially glassy polymer matrix system both concentration gradient controlled migration (stochastic or Fickian diffusion) and relaxation or swelling controlled migration (Case II diffusion) contribute to the rate and extent of penetrant sorption (*4, 6, 7*). However, many variations exist (*5*).

Water uptake rates for food systems are often calculated with the use of Fick's law (*8, 9*) Despond et al (*7*) found that water transport in chitosan films is governed by a Fickian process for partial pressures lower than 0.4, and that in that range of partial pressures, the diffusion coefficient is concentration–dependent. At a higher water activity the rate of diffusion becomes quite similar to the rate of relaxation, leading to anomalous diffusion (which means somewhere in between Fickian and Case II diffusion). Del Nobile *et al.* (*10*) concluded for chitosan films that in the range of a_w 0.2-0.6 the characteristic time of stochastic diffusion was smaller than the characteristic time associated with polymer matrix relaxation. Therefore, polymer matrix relaxation was assumed to be the limiting step controlling water-sorption kinetics in chitosan films. Fickian diffusion was found by Hopkinson et al (*11*) for water vapour sorption into amylose pellets. For food materials no pure Case II behaviour (diffusion is very fast compared to the rate of relaxation of the solid material) was found in literature when considering vapour sorption and for bread crust no water uptake mechanism has been suggested at all.

Besides Fickian diffusion and relaxation of the polymers, other processes may play a role in the sorption process. Valiullin et al (*12*) studied the sorption hysteresis in

Vycor porous glass, which is a glass of SiO_2 prepared according to Hood and Nordberg (Hood, U.S. Patent No. 2,106,744 and Patent No. 2,286,275 #243). The observed hysteresis suggests that for a certain range of vapour pressures, the amount of a molecular species adsorbed by the porous host is higher during desorption than during adsorption, indicating a failure of the system to equilibrate over the time span studied. This phenomenon is often found in food systems as well (*13*). Valiullin et al (*12*) found for Vycor silica glass at low water vapour pressure (outside the hysteresis region) that the gas diffuses into the pore structure, forming adsorbed layers on the pore walls. The relaxation process is here essentially diffusive in character. As the water pressure increases (into the hysteresis region), the adsorbed layers finally grow together and a droplet of water may be formed in the pores. This process is called capillary condensation. This will reduce the surface area exposed to the water vapour/air mixture. The dynamics slow down dramatically and, at long times, are dominated by activated rearrangement of the water within the host material.

Wolf et al (*14*) classified the variations in hysteresis loops in foods into three general groups, namely high-sugar/high-pectin (e.g. apple), high-protein (e.g. pork) and high-starch systems (e.g. rice). Starch gives the largest hysteresis loop of the three groups**.** Several theories have been suggested to explain hysteresis in porous solid foods. Most were based on the above mentioned capillary condensation phenomena (*15*). For starch type products also activation of (additional) sorption sites due to the history of high water content was suggested as a possible explanation (*14, 16*).

In chapter 2 we used the combination of an oscillatory sorption test and a Langmuir type approach to describe water uptake. By varying the oscillation times we found that the characteristic rate of water adsorption on the surface of the bread crust is faster than approximately 15 minutes. From this it was concluded that the transport of water into the material is the rate limiting step in the water uptake process. The objective of the current paper is to obtain a more profound understanding of the mechanism of water uptake in bread crust at different water contents. An understanding of the mechanism will make it possible to develop crispy products that maintain their crispness for a longer time. Besides that it is necessary to have a good model that is representative for water uptake in bread crust in order to be able to compare the water uptake kinetics of different variations of crispy products (e.g. ingredient variation). Two different methods to investigate water uptake kinetics were compared; an oscillatory sorption test and a sorption test in which the air RH was increased stepwise. In this way the effect of experimental time could be studied. In the oscillatory sorption test water uptake and loss in model bread crusts are followed while the relative humidity (RH) of the surrounding air oscillates step-wise between two set values. First we determined the water uptake mechanism for bread crust (Fickian, anomalous or Case II). Next, the water uptake process was further studied by fitting the changes in weight with a single exponential. In addition, the effect of different RH oscillation ranges was investigated in order to better understand the relation with the sensorial loss of crispness. Oscillatory sorption data were compared with data from non-oscillatory, long term water uptake sorption isotherms to evaluate the effect of the experimental set up. Model bread crusts prepared from two fractions of an air-classified flour, a protein rich and a starch rich fraction and a bread flour were studied to check the use of the developed methods for investigating the effect of ingredient composition on water uptake kinetics.

Tabel 1 Composition of the flours that were used to make the bread crusts

Component	Starch rich flour	Protein rich flour	Soissons
Starch (% dm)	82	69	79
Protein (% dm)	9.5	17.3	12.9
Damaged starch (% dm)	6.2	10.3	4.9
Fat (% dm)	0.6	1.8	1.2
Moisture (%)	9.6	9.2	14.8
Insoluble non-starch polysaccharides (% dm)	0.1	0.03	1.5
Soluble non-starch polysaccharides (% dm)	0.47	1.01	0.42

3.2 Materials and methods

3.2.1 Materials

Model bread crusts were prepared from two air-classified fractions of a wheat flour (a protein rich and a starch rich fraction). Rusk rolls were prepared with Soissons wheat flour. The flours were purchased from Meneba (Meneba Meel BV, Rotterdam, the Netherlands). The composition of the flours is shown in table 1. Native wheat starch (Excelsior) was purchased from Avebe (Veendam, the Netherlands).

3.2.2 Methods

3.2.2.1 Preparation of model bread crust

Dough was prepared by mixing in a farinograph mixer (Brabender, Duisburg, Germany) using a 50 g mixing bowl. The formulation included flour, NaCl (2 g/100g flour) and dry yeast (1.7 g/100 g flour). For the protein rich dough and the starch rich dough, respectively, 80% and 63% water (based on Farinograph water absorption, ICC 115/1, 1991) (*18*) on flour basis was used to prepare the dough. The temperature of the added water was 22°C and the starting temperature of the mixing bowl and flour was 22.5°C and 23°C, respectively. The dough was mixed at a speed of 100 rpm until the dough temperature reached 26°C, which corresponded to the maximum in the torque-time curve. After mixing, the dough was separated into three pieces and proofed at 30°C and 90% RH for 15 minutes in a climate chamber (Wekk 0028, Weiss Enet, Tiel, The Netherlands). Next, the dough pieces were flattened by hand, rounded and again proofed at 30°C for 15 minutes. The dough was then sheeted between Teflon paper using a pasta machine (Titania, Imperia, Sant'Ambrogio di Torino, Italy) in order to get a flat piece of dough. Sheeting was repeated until a sample thickness of 2 mm was obtained (from position 1 to 4 of the pasta machine). Test pieces were cut from the dough

sheet with the help of a stainless steel 6 cm diameter ring. Next, the dough was proofed for 70 min at 30 °C and 90% RH. Finally the dough sheet was placed on a punctured stainless steel plate. This supports the dough during baking and allows moisture loss from the bottom of the sample.
The dough samples were baked in a HR83 halogen heater (Mettler Toledo, Tiel, The Netherlands). The temperature program was selected to simulate baking conditions. First, the dough was baked at 130 °C for 4 min (including 1 min to reach 130°C). Next, the temperature was increased to 160°C for 4 min (including 40 s to increase the temperature to 160°C) and finally to 185°C for 50 seconds (including 20 s to increase the temperature).

3.2.2.2 Preparation of rusk roll crust

Soissons bread crusts from a rusk roll were prepared from Soissons flour as mentioned in chapter 2, section 2.2.2.1.

3.2.2.3 Sample preparation

Crust samples were milled in an analytical grinder (type A10, IKA Labortechnik, Staufen, Germany) for 10 seconds at a time with a maximum of 3 times (to prevent heating of the sample) until a fine powder was obtained. The powder was sieved using different sieves (63µm, 0.25 mm and 0.5 mm). The different sieving fractions (<63µm, 63µm-0.25mm and 0.25 mm-0.5 mm, called S, M and L, respectively) were freeze dried. The largest sieve fraction (>0.5 mm) was discarded. From the model crust samples only the large particle size samples were used. Samples were kept at room temperature in closed tubes until use.

3.2.2.4 Oscillatory sorption experiments

Oscillatory sorption experiments were performed using a VTI-SGA 100 symmetric vapor sorption analyzer (VTI Corporation, Hialeah, Florida USA). This is a continuous vapour flow sorption instrument for obtaining water and organic vapour sorption isotherms. The instrument is equipped with a dew point analyser, and a Cahn microbalance. All tests on crust samples (5-7 mg with an approximate thickness of the powder bed of 1 mm) were performed at 25°C. Samples were stored over P_2O_5 for at least 3 days before starting the experiment resulting in an initial water content of approximately 1.5%. Every experiment was started with a drying step in the VTI at 50°C during 120 min. Oscillatory sorption experiments were performed by applying block wise oscillations of the environmental RH between 40 and 50% RH, 50 and 60% RH, 60 and 70% RH and between 70 and 80% RH, respectively, for the model crust samples. For the Soissons bread crust steps of 20% RH were taken (20-40, 40-60, 50-70 and 60-80%). The rising and falling times of each block were 4.5 minutes. The measurements were stopped when the oscillations of the sample weight became steady. Oscillation times of 23 minutes were used.

3.2.2.5 Stepwise sorption isotherm experiments

The isotherm was determined following the same procedure as described above but the RH was increased with 10% every 570 minutes or when the isotherm was stable (less than 0.0001% weight change in 10 minutes). The weight change of the sample (caused by the change in environmental RH) was measured in time.

3.2.2.6 Starch content

Total Starch. The total starch content was measured using a total starch assay kit from Megazyme (R-amylase/amyloglucosidase method) (Megazyme International Ireland, Wicklow, Ireland).

Damaged Starch The damaged starch content was measured using a damaged starch assay kit from Megazyme (α-amylase method) Megazyme International Ireland, Wicklow, Ireland).

3.2.2.7 Protein content

Protein content was determined according to the DUMAS method (*19*) using an NA2100 Nitrogen and Protein Analyser (CE Instruments). Methionine was used as a standard.

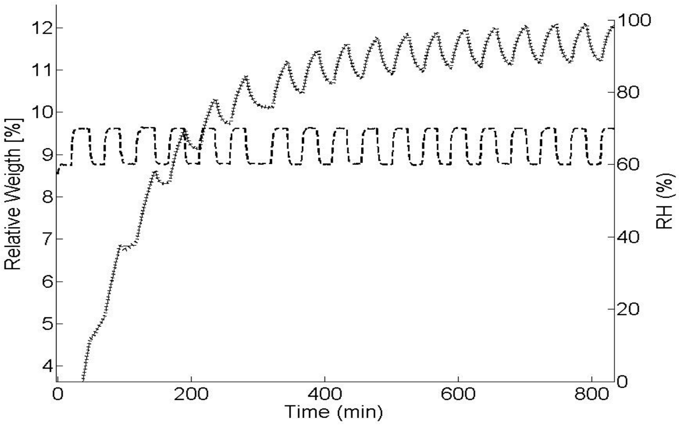
3.2.2.8 Pentosan content

Insoluble pentosans. The flour (50 g) is dispersed in water (450 ml). 50 ml of 12% SDS was added and 0.5 g dithiotreitol (DTT). The solution was stirred for 3 hours. After stirring, the solution is poured over a sieve (45 μm) and the residue is thoroughly washed with water. The contents of the sieve were collected and freeze dried and weighed. The protein content was determined with the Dumas method.

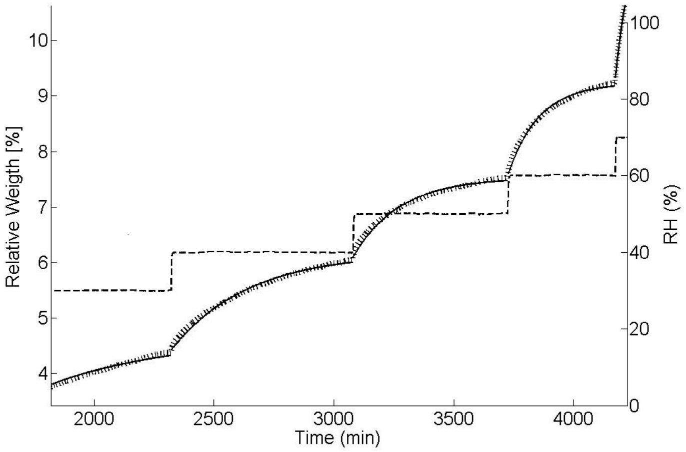
Soluble pentosans. 40 g of flour was added to a solution of 36 grams Tri-chloro-acetic acid (TCA) in 164 grams of water (18% ($^{w}/_{w}$)). The solution was stirred overnight at 4°C and next centrifuged at 10,000 rpm in a Sorvall centrifuge for 30 minutes. Cold (4°C) ethanol was added to the supernatant until a concentration of 70% (v/v). The solution was left overnight to sediment the insoluble parts and the supernatant was centrifuged again at 2,000 rpm. The residue was washed 2 times with cold 70% (v/v) ethanol to remove the TCA. Then the residue was dissolved in 40 ml 100mM citric acid buffer pH 4.6. 14 units amyloglucosidase was added and the solution was incubated at 40°C overnight to digest the remaining starch. The solution was cooked for 5 minutes, cooled down and centrifuged until a clear solution was obtained. The solution was dialyzed against distilled water (3 x 5 litre). The solution was freeze dried and the weight was determined.

3.2.2.9 Fat content

The fat content was measured with a Soxtec (Avanti 2050). Hexane was used as extraction fluid. The samples (around 10 g) were immersed in the fluid for 30 minutes and rinsed with hexane for 2 hours.



(a)



(b)

Figure 1 Relative change in weight as a function of time for a protein rich model crust during an oscillatory sorption experiment when oscillating the RH between 50 and 60% (a) and the same for part of an isotherm experiment when increasing the RH in steps of 10% (b) --- indicates the RH, ... experimental data, and full lines the fitted curve

3.3 Results and Discussion

Figure 1a shows an example of an oscillatory sorption experiment with the RH step wise oscillating between 50 and 60%. The experiment can be divided into two parts; the first part in which a steady state in the oscillation of the sample weight is not yet reached and in which a large amount of water is taken up and a second part in which a steadily oscillating state is reached. The percentage weight change ($\Delta m(t)$) was defined as follows:

$$\Delta m(t) = \frac{100 * [M(t) - M(0)]}{M(0)} \quad (\%) \tag{1.1}$$

where M(t) and M(0) are the weight at time t and the initial weight after the drying step of the sample, respectively.

Figure 1b shows part (between RH 30 and 60%) of the result of an alternative, non-oscillatory way for measuring water sorption kinetics in which the RH was increased with steps of 10% RH until 90% RH was reached. The experimental time per step (3.5-10 hours depending on how fast an apparent steady state was reached) was longer than in the oscillatory sorption experiment (around 23 minutes per step).

3.3.1 Type of sorption kinetics

The kinetics of water sorption were studied at 25°C at different relative humidities (RH). A single exponential function:

$$M(t) = [M(0) - M(\infty)]e^{-kt} + M(\infty) \tag{1.2}$$

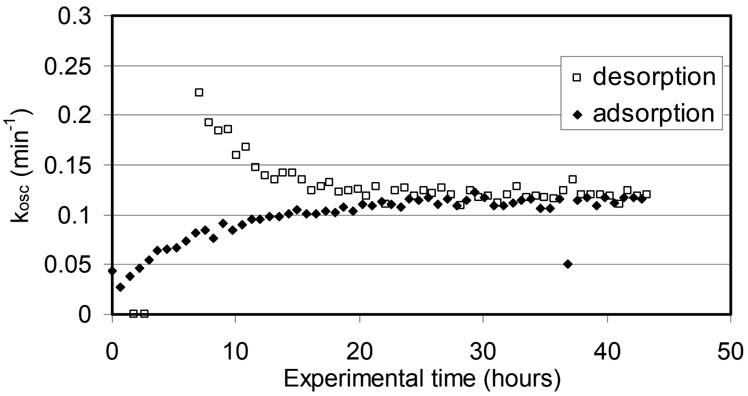
was found to be the best simple fit for the water uptake data. M(t) is the mass of the sample at time t, M(0) is the initial sample weight and M(∞) is the sample weight at apparent equilibrium. Both M(∞) and k are adjustable parameters. A double exponential improved the fit, but the two resulting k-values only differed a factor two from each other. To prevent over-interpretation of the data a single exponential fit was chosen.

Another way of analysing the weight increase in time is a power law approach. This was done by fitting the initial sorption results for a duration of 4 minutes starting 1 minute after the step wise change of the RH to:

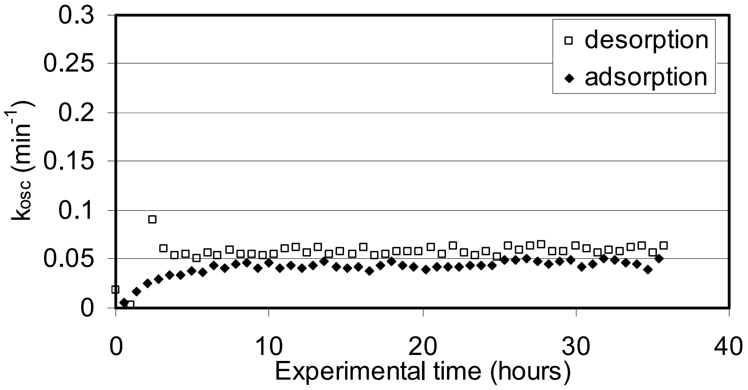
$$(M(t) - M(0)) / M(0) = r * t^n \tag{1.3}$$

r is a rate parameter and t is time. Since it took around 1 minute to obtain a stable RH we excluded the first minute from the fit. The curves were linear during these 4 minutes (around 7 data points) on a log-log scale. The fitted n-value was close to 1 for all adsorption and desorption curves indicating exponential kinetics. This suggests case II water uptake behaviour, which means that diffusion is very fast compared to the rate of relaxation of the solid material. Thus relaxation of the matrix may be the limiting step in the water uptake process. A value of 1 rules out Fickian diffusion, for which n=0.5 would be found.

A rate parameter *k* was calculated by fitting Eqn. 1.2 to the experimental data. In figure 1a and 1b the fit results are shown. The quality of fit, expressed by R^2 is better for the oscillatory sorption experiments (R^2~0.99) than for the sorption experiments with stepwise increasing RH (R^2~0.98).



(a)



(b)

Figure 2 Rate parameter k_{osc} for the separate oscillations steps of 23 minutes each as a function of the total experimental time for an oscillatory sorption experiment. Results for the adsorption and desorption steps for a protein rich model crust with RH oscillating between 40 and 50% (a) and RH oscillating between 70 and 80% (b)

3.3.2 Analysis of sorption rates

Figure 2 shows the fit results of rate parameter k for the upward and downward branch of an oscillatory sorption experiment with the RH oscillating between 40 and 50% (a) and between 70 and 80% (b). The fit results of M(∞) values for the adsorption branch were close to the measured isotherm values. The M(∞) values for the desorption branch were lower than those of the isotherm values that were obtained for water activities smaller than 0.6. This is likely due to the fact that the maximum water activity that these samples reached was lower than 0.9 (results not shown). Therefore the maximum effect of hydration did not occur. The k-values for the adsorption steps at the start of the oscillatory sorption experiment (figure 2a and b) are very low. This is most likely due to the dryness of the material which therefore has a very small free volume for water available. As a consequence, the rate of transport of water through the material will also be slow. With increasing water content the water will hydrate the polymers and the material will swell, thus creating more space for the water to move through. This will result in an increase of the k_{osc}-value until a steady state in the material is reached and the average water content is constant. The average steady state k_{osc}-value also increases with increasing water content or RH for the lower RH combinations (up to 60% RH) probably as a result of the process described above .

For the RH oscillations between 40 and 50% (figure 2a), the k values for the upward and downward branch become similar when the system reaches a steady state situation. At higher RH (figure 2b) the k-values from the upward branch have a little lower steady state value than the k-values from the downward branch of the oscillatory sorption experiment. This difference in upward and downward k values must correspond to a difference in the shapes of the upward en downward branches. Such a difference in shape is not unexpected considering the fact that in a stepwise sorption experiment with this material hysteresis is observed. The material may have changed during water uptake and will therefore behave different upon moisture loss. It could also be that the difference in upward and downward k_{osc} values is due to the difference in initial water content. This difference is much larger at higher RH. According to Bruin and Luyben (*20*) the diffusion coefficient of water increases with increasing water content from 10^{-15} m^2/s at low water content (<5% water) to 10^{-10} m^2/s at 50% water content. This means that the transport of the water out of the sample at the start of a downward branch will be faster than the transport of water into the sample at the start of an upward branch. This is because the initial water content is higher for the desorption part than for the adsorption part.

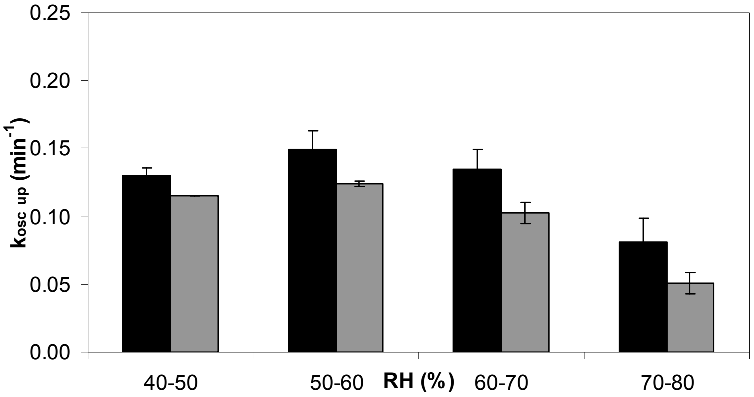
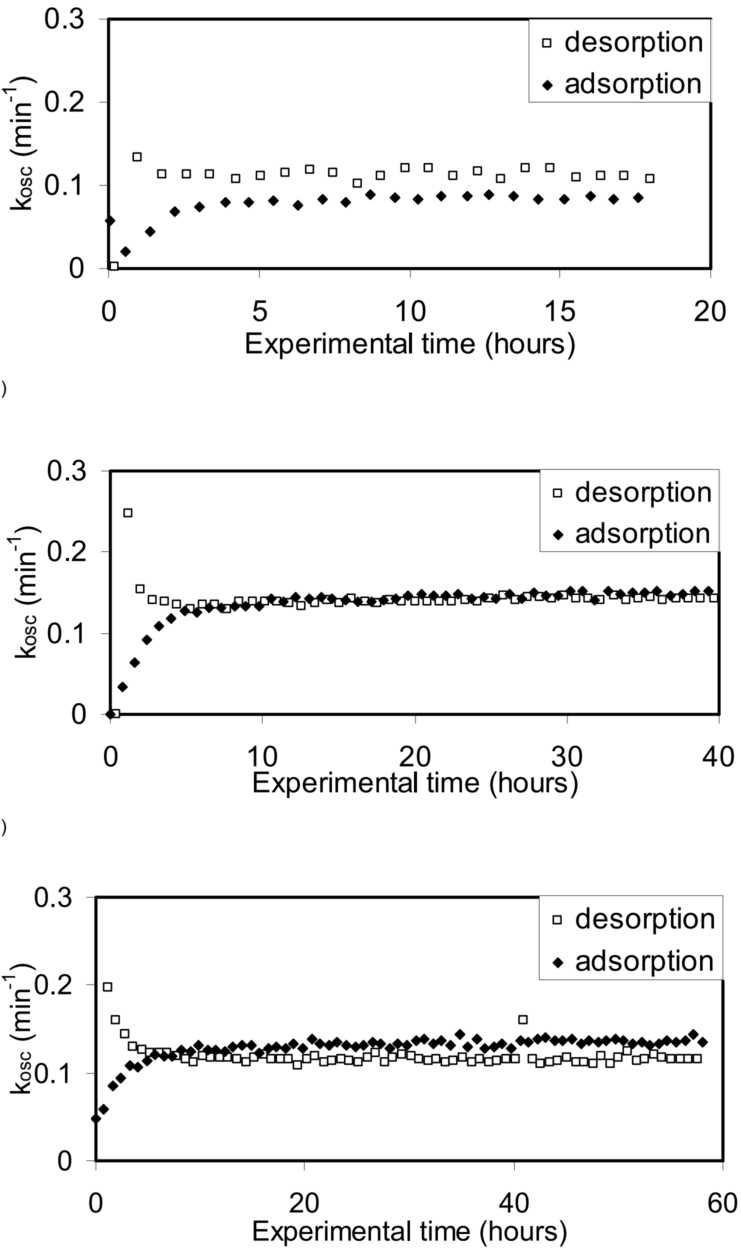


Figure 3 Average rate parameter k_{osc} for starch rich (black columns) and protein rich (grey columns) model crusts oscillating between different RHs for the adsorption branch. Average of k_{osc} for the last five adsorption steps in the steady state situation.

Figure 3 shows the k_{osc} -values for adsorption (average of the last 5 oscillation steps in the steady state situation) for particles of a protein rich and starch rich model crust. A maximum in the rate parameter k_{osc} was found for a RH value between 50 and 70%. At even higher water activities a smaller rate parameter k_{osc} was found. This is probably due to the swelling of the material with increased water content which results in a reduced effective surface area and possibly even caking of the particles. Figure 3 also shows that the protein rich model crust has smaller k values than the starch rich model crust. The reason for this is not yet fully understood. In chapter 6 the effect of ingredient variation is further studied.

For Soissons rusk roll crusts experiments were performed in the same way as for the protein rich and starch rich model crusts. However for the Soissons rusk roll crust the RH was oscillated over ranges of 20% RH difference (20-40; 40-60; 50-70 and 60-80). Also for the Soissons rusk roll crusts a single exponential could be fitted to the oscillatory sorption experiments. Figure 4a, b and c show the development of the k_{osc}-values from the start of the experiment until a steady state was reached. At high RH the rate parameter k_{osc} of the adsorption branch is lower than the k_{osc} of the desorption branch even when an apparent steady state is reached. At very low RH (20-40%) the trend is the opposite: the k_{osc}-value of the adsorption branch is higher than the k-value of the desorption branch. As mentioned before, a difference between adsorption and desorption rates at an apparent dynamic equilibrium might be the result of an interplay between hysteresis (changes in the sample during each oscillation step) and non-exponential behaviour.

The effect of particle size on the k_{osc}-values of Soissons bread crust at steady state is shown in figure 5a. A smaller particle size gives a higher k-value. This is expected since the smaller particles have a relatively larger surface area and consequently the sample will reach a steady state in less time. The k-values for the larger particle size of the Soissons bread crust are similar to the values for the model crusts.



(a)

(b)

(c)

Figure 4 Evaluation of the rate parameter k_{osc} for adsorption and desorption steps of 23 minutes in an oscillatory sorption experiment as a function of the total experimental time. Results for a Soissons rusk roll crust with RH oscillating between 60 and 80% (a); 40 and 60% (b) and 20 and 40% (c).

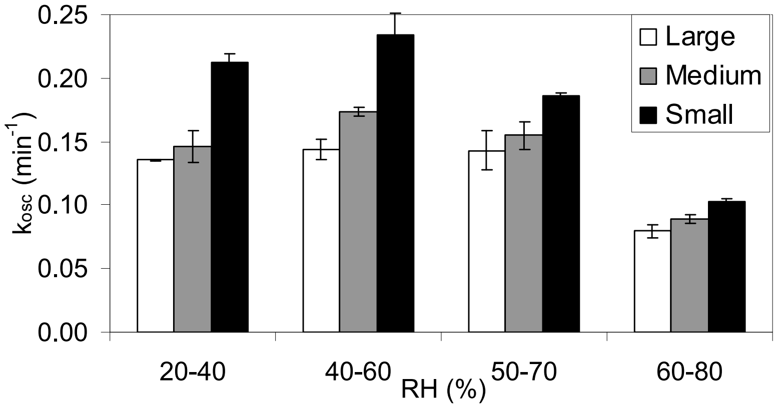


Figure 5 Effect of particle size on the average k_{osc} of the adsorption step in the steady state situation (average of the last 5 oscillation steps). Results for a Soissons rusk roll crust in an oscillatory sorption experiment. The RH was oscillated between different values. Large=0.25-0.5 mm, Medium=0.063-0.25 mm, Small=<63 µm.

3.3.3 Comparison of sorption rates measured by oscillatory sorption and standard step sorption experiments

The water uptake curves obtained during the determination of a sorption isotherm by stepwise increasing RH (figure 1b) were analyzed in the same way as the oscillatory sorption curves (by fitting an exponential function). In the sorption experiments with a stepwise increase of RH, water uptake was followed for 600 minutes for each step, whereas one step in an oscillatory sorption experiment took only 23 minutes. However also in an oscillatory sorption experiment it could take 600 minutes or more before a steady oscillating state was reached. The isotherm experiments gave rate values (k_{step}) that are around a factor 10 lower than those obtained with the oscillatory sorption experiments (k_{osc}) (figure 6). The maximum in the rate parameter calculated from an isotherm experiment is found at higher RH than for an oscillatory sorption experiment. Most likely a different relaxation time is manifested here due to the different experimental time. In the oscillatory sorption experiment the duration of an oscillation was 23 minutes. Therefore, the first 23 minutes of each step in the sorption experiment with a stepwise increasing RH was considered separately. From this early part of the sorption curve smaller k-values than those of the oscillatory sorption experiments are obtained (figure 6) although the experimental time considered was the same. This suggests that oscillating the water content in the sample changes the response of the sample to a changing RH.

Different results with changing experimental times were also found in a separate experiment in which the effect of the time per step in an experiment with a stepwise increase in RH was considered for native wheat starch. The resulting rate parameters k_{step} for adsorption are shown in figure 7. The experimental times were 60 and 300 minutes per step. From the experiment with 300 minute steps also a k_{step} was calculated from the weight change during the first 60 minutes of each step. The results show that the adsorption rate during the first 60 minutes of a 300 minute step is not the same as the sorption rate during a 60 minute step nor the same as those of a fit of the total 300 minutes. This suggests that at longer experimental times more relaxation processes take place. However fitting with two exponentials resulted in two k-values that were in the same order of magnitude as mentioned before. The equilibrium values were the same for both experiments (results not shown). This means that when designing experiments to study kinetics of water uptake the equilibration time should always be chosen carefully in accordance with the research question, taking the relevant time scales into account.

From the higher rate parameter found for the oscillatory sorption experiment, we conclude that the water that is exchanging in this experiment is more loosely bound than the water that is adsorbed and allowed to stay and redistribute during several hours in a sorption experiment where RH increases stepwise. Probably the water that determines k_{osc} involves especially water that exchanges relatively easily e.g. water that adsorbs at sites that are easily accessible, relatively close to the outside of the material and to the pores. In the isotherm experiment the sample starts for each step from a situation more close to thermodynamic equilibrium whereas in the oscillatory sorption experiment the sample is never in equilibrium. In the latter case the water will certainly not be homogeneously distributed. Additionally, probably the material changes at a very slow rate because the water uptake kinetics change with changing experimental times. This means that the material can be different depending on the history of the sample.

The maximum in the values of k_{step} and k_{osc} occurs at different values of RH, The oscillatory sorption experiment displays a maximum at a lower RH than the step experiment. The maximum in the k_{step} obtained from the first 23 minutes of the isotherm experiment is different again and shifts to a lower RH value (approximately 40%) compared to the k_{step} of the full experiment. This suggests that the maximum in the k-value depends on both the time scale of the experiment and the state of the sample. Different k-values and different positions of the maxima are expected because likely different processes will take place; water adsorbs to the surface of the particles and then migrates into the particle. Water that migrates into the dry matrix will probably do that at a different rate as compared to water migrating into the already hydrated matrix. Therefore different experimental set-ups having different experimental times lead to the description of these different processes. Besides that a variation in the relaxation times between the different molecules of the solid matrix may also result in different k-values. This is the subject of a follow-up paper, describing a multilayer water uptake model in which the different water uptake processes are taken into account (*21*).

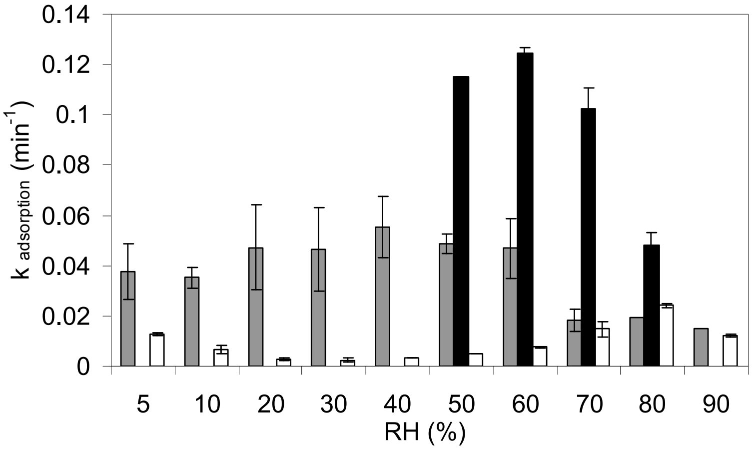


Figure 6 Rate parameter of both isotherm data (k_{step}) and oscillatory sorption data (k_{osc}) calculated from the adsorption branch for the protein rich model crust, in white $k_{step,}$ in black k_{osc}, in grey k_{step} where only the first 23 minutes of the data per step were used for the fit.

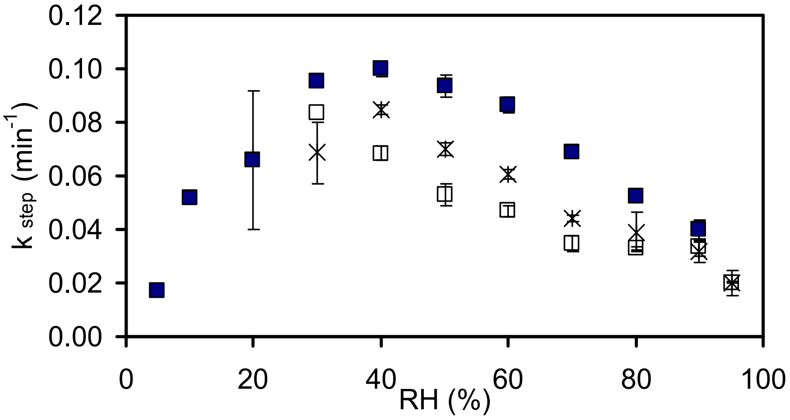


Figure 7 Effect of the experimental time per step on the k_{step} rate parameter for a sorption experiment with stepwise increase of RH for native wheat starch. Results for experimental times per step of 300 minutes (□), 60 minutes (■) and 300 minutes where only the first 60 minutes were used for the fit (ж).

3.3.4 Dependence of rate parameter k on experimental time

From figure 2a and b we calculated the time dependence of the rate parameter k_{osc} by fitting a single exponential function to these results:

$$k_{osc}(t) = k_{osc}(\infty) - [k_{osc}(\infty) - k_{osc}(0)]e^{-Ct} \tag{1.4}$$

with $k_{osc}(0)$, $k_{osc}(t)$ and $k_{osc}(\infty)$ are the value of k_{osc} at times 0, t, and at steady state, respectively, and C an overall rate parameter, which describes how fast the sorption rate changes with time. The results for C are shown in figure 8 for model crust of protein rich and starch rich flour. C increases with increasing water content which indicates that at higher water contents equilibrium is reached sooner. The characteristic time (1/C) of the experiment changes from 400 minutes at low RH (40-50% RH) to 150 minutes at higher RH (70-80% RH). Compared to k_{step}, also included in figure 8, C is smaller than k_{step}, but both rate parameters are of the same order of magnitude. It should be noted that no difference in C or in k_{step} was observed between the protein rich and starch rich model crust. Probably the difference in water uptake rates only manifests itself at specific time scales or the relatively long experimental times used to calculate C and k_{step} make the method less precise.

For Soissons rusk roll crust the results for C and k_{step} are shown in figure 9a and b. The fit (R^2~0.95) was not as good as for the starch rich and protein rich model crusts (R^2~0.98). This is not well understood yet. A minimum in C is found around 60% RH for small sized particles of Soissons rusk roll crust (figure 9a). This is not expected but might have to do with the fit which was not as good as the fit for the model crusts. For medium and large sized particles C was more or less constant for all RHs. As for the model crusts, also for the Soissons rusk roll crust the C value is about equal to k_{step} at lower RH, while at higher RH k_{step} is higher than C (figure 9b). This could be due to caking or reduction of the surface area during the oscillatory sorption experiment. The sample is constantly at a high RH during the oscillatory sorption experiment while for the isotherm experiments the high RHs involve only part of the total time. Additionally the time considered to calculate C varies between 20 and 60 hours which is longer than the experimental time per step in a stepwise sorption experiment.

This slower process with a characteristic time of 150-400 minutes is found in both the oscillatory sorption experiment and the isotherm experiment. This suggests that k_{step} that was found in the isotherm experiment and C that was found for the oscillatory sorption experiment stem from a mechanism that manifests itself in both experiments. The sample in an oscillatory sorption experiment "senses " an average water content. This will be the water from which a part will penetrate into the sample further with the slow C-value that is similar to k_{step}. No exact agreement may be expected since C is calculated for a process starting from a dry state and k_{step} for an increase of RH by 10%. Our working hypothesis is that this could be a relaxation process which is also responsible for the loss of crispness, since the time scales on which crispness is lost are in the same range (*22*).

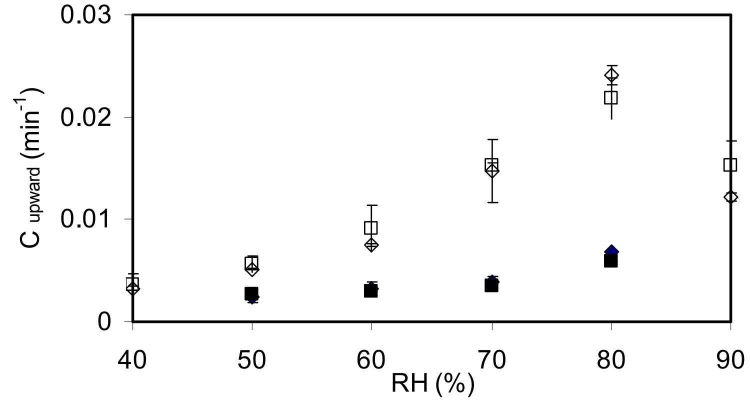


Figure 8 Rate parameter C of the oscillatory sorption experiment for PR (♦) and SR (■) model crusts compared with the rate parameters k_{step} of the isotherm experiment for PR (◊) and SR (□) model crusts (for further explanation see the text).

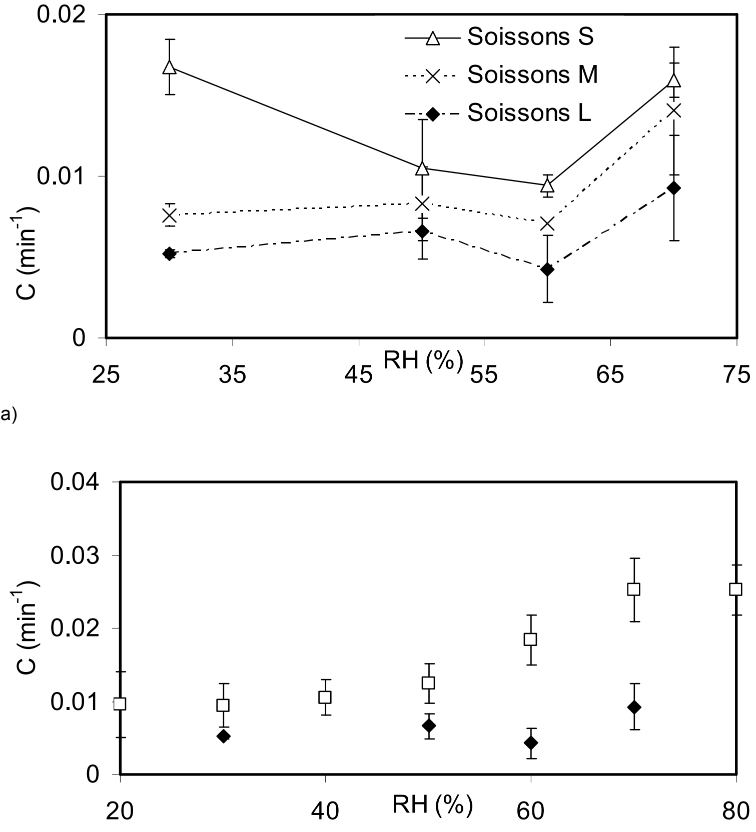


Figure 9 Effect of particle size on the rate parameter (C) for Soissons rusk roll crust at different RH combinations, S=<63 μm, M=0.063-0.25 mm, L=0.25-0.5 mm (a) and comparison of the rate parameter C for Soissons rusk roll crust (♦) with the rate parameter k_{step} of the isotherm experiment (□) for a large particle size (0.25-0.5 mm) (b).

3.3.5 Maximum of k-values and relation with glass transition

A transition point is found in sensorial crispness as well as in water mobility (NMR T_2), at a water activity of around 0.58 (chapter 4 of this thesis). However, the glass transition of bread crust at room temperature as measured with DSC and PTA is around water activities of 0.85 and 0.80, respectively (chapter 4). Aguerre and Suarez (*23*) found a maximum in the diffusion coefficient for sorghum and corn at a water content of around 0.10 kg water per kg dry solid at 40°C but they did not give an explanation for this observation. That maximum shifted to lower water contents with increasing temperatures which could point at a relation with the glass transition. The maximum in the k-values we found is at about a water activity of 0.6 (~0.11 kg water/kg dry matter) for the oscillatory sorption experiment and at 0.8 (~0.16 kg water/kg dry matter) for the sorption experiment with stepwise increasing RH. It appears, therefore, that transition point in mobility of water with increasing water content as measured with NMR in chapter 4 is found at roughly the same a_w as the maximum in the rate parameter k_{osc} as measured with an oscillatory sorption experiment and the transition point as measured with the PTA and DSC is found at roughly the same aw as the maximum in the rate parameter k_{step}.

3.4 Conclusions

A single exponential function was found to fit the water sorption data best. The rate parameters of water adsorption derived from the standard isotherm experiment (k_{step}) were around a factor 10 lower than those derived from oscillatory sorption experiments (k_{osc}). The corresponding characteristic times (1/k) were found to be around 10 and 200 minutes for the oscillatory sorption and isotherm experiment, respectively. An experiment on native wheat starch with different experimental times per step in a sorption experiment with stepwise increasing RH showed that an increase in experimental time per step decreased the value of the rate parameter. So different experimental times gave different results. This implies that different experimental set-ups can result in the description of different processes like adsorption of water to the surface of the sample or diffusion into the matrix. Furthermore also a rate parameter (C) describing the time dependence of the rate parameters of the oscillatory sorption experiment (k_{osc}) was calculated and found to be in the same range as the rate parameter of the isotherm sorption experiment (k_{step}). This indicates that at least 2 processes (with a factor 10 difference in rate) act at the same time in the bread crust during water uptake. Probably the slower water uptake process with a characteristic time of around 200 minutes is representative for the kinetics of loss of crispness. The water sorption experiments on (model) bread crust show that the transport mechanism of water into the crust particles is definitely not Fickian, but more Case II like. The oscillation experiment was sensitive enough to show a relatively small difference in rate parameters between two model crust of two different formulations.

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References

1. Luyten, H.; Plijter, J. J.; van Vliet, T., Crispy/crunchy crusts of cellular solid foods: a literature review with discussion. *Journal of Texture Studies* **2004,** 35, (5), 445-492.
2. Roudaut, G.; Dacremont, C.; le Meste, M., Influence of water on the crispness of cereal-based foods: acoustic, mechanical, and sensory studies. *Journal of Texture Studies* **1998,** 29, (2), 199-213.
3. Labuza, T. P.; Hyman, C. R., Moisture migration and control in multi-domain foods. *Trends in Food Science and Technology* **1998,** 9, 47-55.
4. Berens, A. R.; Hopfenberg, H. B., Diffusion and relaxation in glassy polymer powders: 2. Separation of diffusion and relaxation parameters. *Polymer* **1978,** 19, 489-496.
5. Wel, G. K. v.d.; Adan, O. C. G., Moisture in organic coatings-a review. *Progress in Organic Coatings* **1999,** 37, 1-14.
6. Vrentas, J. S.; Jarzebski, C. M.; Duda, J. L., A deborah number for diffusion in polymer-solvent systems. *AIChE Journal* **1975,** 21, (5), 894-901.
7. Despond, S.; Espuche, E.; Domard, A., Water sorption and permeation in chitosan films: relation between gas permeability and relative humidity. *Journal of Polymer Science: Part B: Polymer Physics* **2001,** 39, 3114-3127.
8. Roman-Gutierrez, A. D.; Mabille, F.; Guilbert, S.; Cuq, B., Contribution of specific flour components to water vapor adsorption properties of wheat flours. *Cereal Chemistry* **2003,** 80, (5), 558-563.
9. Becker, H. A.; Sallans, H. R., A study of internal moisture movement in the drying of the wheat kernel. *Cereal Chemistry* **1955,** 32, (3), 212-226.
10. Nobile, M. A. d.; Buonocore, G. G.; Conte, A., Oscillatory sorption tests for determining the water-transport properties of chitosan-based edible films. *Journal of Food Science* **2004,** 69, (1), 44-49.
11. Hopkinson, I.; Jones, R. A. L.; Black, S.; Lane, D. M.; McDonald, P. J., Fickian and Case II diffusion of water into amylose: a stray field NMR study. *Carbohydrate Polymers* **1997,** 34, 39-47.
12. Valiullin, R.; Naumov, S.; Galvosas, P.; Kärger, J.; Woo, H.; Porcheron, F.; Monson, P. A., Exploration of molecular dynamics during transient sorption of fluids in mesoporous materials. *Nature* **2006,** 443, (26), 965-968.
13. Al Muhtaseb, A. H.; McMinn, W. A. M.; Magee, T. R. A., Moisture sorption isotherm characteristics of food products: a review. *Food and Bioproducts Processing* **2002,** 80, 118-128.
14. Wolf, M.; Walker, J. E.; Kapsalis, J. G., Water vapor sorption hysteresis in dehydrated foods. *Journal of Agricultural and Food Chemistry* **1972,** 20, (5), 1073-1077.
15. McMinn, W. A. M.; Magee, T. R. A., Studies on the effect of temperature on the moisture sorption characteristics of potatoes. *Journal of Food Engineering* **1999,** 22, 113-128.
16. Berg, C. van der; Kaper, F. S.; Weldring, J. A. G.; Wolters, I., Water binding by potato starch. *Journal of Food Technology* **1975,** 10, 589-602.

17. Edmister, J. A.; Vickers, Z. M., Instrumental acoustic measures of crispness in foods. *Journal of Texture Studies* **1985,** 16, (153-167).

18. ICC-Standard methods, **1991**. International Association for Cereal Chemistry.
Method for using the Brabender Farinograph; No 115/1.

19. Sweeney, R. A.; Rexroad, P. R., Comparison of LECO FP-228 "nitrogen determinator" with AOAC copper catalyst Kjeldahl method for crude protein. *Journal-Association of Official Analytical Chemists* **1987,** 70, (6), 1028-1030.

20. Bruin, S.; Luyben, K., Drying of food materials: a review of recent developments. *In:A.S. Mujumbar, ed. Advances in Drying, Vol.1. Hemisphere, Washington* **1980**, 155.

21. Meinders, M. B. J.; Nieuwenhuijzen, N. H. van; Vliet, T. van, Modeling water sorption dynamics of cellular solid food systems. **To be submitted**.

22. Primo Martín, C.; van de Pijpekamp, A.; van Vliet, T.; de Jongh, H. H. J.; Plijter, J. J.; Hamer, R. J., The role of the gluten network in the crispness of bread crust. *Journal Of Cereal Science* **2006,** 43, (3), 342-352.

23. Aguerre, R. J.; Suarez, C., Diffusion of bound water in starchy materials: application to drying. *Journal of Food Engineering* **2004,** 64, 389-395.

Chapter 4 Relations between sensorial crispness and molecular mobility of model bread crust and its main components as measured by PTA, DSC and NMR.

Abstract

Consumer appreciation of brittle cellular foods, like bread crusts, depends on textural properties such as crispness. This crispy character is lost above a certain water activity. It is not known what exactly is happening in these crusts when water enters. For example it is unclear whether it is the changes in the starch or the gluten that initiate the loss of crispness with ageing time. In this paper the effect of water on the glass transition of model bread crusts was studied using two complementary techniques: phase transition analysis (PTA) and temperature modulated differential scanning calorimetry (TMDSC). The mobility of water was studied with Nuclear magnetic resonance (NMR). The results were compared with sensory data. Bread crusts prepared with different types of flour were tested to evaluate the effect of flour composition on crispness of model crusts equilibrated at different relative humidities. The results were compared with the results for the single flour components starch and gluten. Sensory crispness scores decreased with increasing a_w from 0.55 upwards. At a_w 0.70 sensory crispness was completely lost. Both DSC and PTA showed a transition point at an a_w of 0.70-0.75. NMR gave a transition point in the mobility of the protons of water at a_w 0.58. This supports the hypothesis that loss of crispness starts as a result of processes at a molecular level, before the macroscopic glass transition. This also suggests that non-bound water (water that is not directly attached to the solid matrix) causes the loss of crispness at low a_w. At higher a_w increased mobility of the macromolecules will start to play a role. NMR experiments with the separate flour components indicate that the T_2 transition point in starch samples occurs at a lower RH than for gluten. This could imply that starch loses crispness at lower a_w than gluten. Increased mobility of small components and side chains might induce increased energy dissipation upon deformation of the material resulting in less available energy for fracture and with that in a less crispy product.

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4.1 Introduction

Consumer appreciation of brittle cellular foods, such as bread crusts, depends on textural properties such as crispness. This crispy character is lost above a certain water activity (*1, 2*). For example, for bread crust this is at a water activity above approximately 0.6 (*3*). It is especially difficult to keep a food crispy when the product has a moist interior. Water then migrates from the interior to the crust causing hydration of the crust components. This results in an increased mobility of the macromolecules, leading to a glass to rubber transition of the main macromolecules present (mainly proteins and carbohydrates). At about the same water activity a loss of sensory crispness occurs (*4*), (*5*). Much research has been performed on glass transitions of bread-like products (toasted bread and crackers) at low moisture contents (*6*), (*7*), (*8*). They observed however that loss of crispness already occurred while the crispy material was still in the glassy state. Le Meste et al (*2*) related this to sub-Tg relaxations such as observed in biopolymers and low molecular weight sugars (β and γ relaxations), although their origin is still being discussed. Sub-Tg relaxations could correspond to rotation of lateral groups (γ relaxation at low temperature) or to local conformation changes of the main chain (β relaxation) (*9*).

Retrogradation of starch in bread is considered one of the important causes of loss of freshness of the crumb during storage (*10*),(*11*). Recently, starch retrogradation has also been investigated for bread crust (*12*). It was concluded that during baking ca. 40% of the starch in the crust does not gelatinize due to a lack of water. The fraction that does lose its crystallinity regains it by retrogradation, but only after 20 days at high water activity. Low enthalpy amylopectin retrogradation was sometimes found in bread crust after two days of storage in a plastic bag (*12*). This means that amylopectin retrogradation - the main process responsible for the staling of bread crumb- on its own cannot be responsible for crispness deterioration of the crust. A crust of bread has had a different history in terms of water content and temperature than for example extruded bread or bread toast investigated before (*6, 13*). Also, unlike bread crumb or extruded bread products, bread crust is a network of gluten protein with starch granules embedded. This is supported by CSLM observations (*3, 12*). However, it is not known what exactly is happening in these crusts when water enters. For example it is unclear whether it is the changes in the starch or in the gluten that initiate the loss of crispness with ageing time. Clearly, more information is required about the mechanisms underlying its deterioration. The aim of this study was to investigate the relation between loss of sensorial crispness in a bread crust model and the glass transitions as measured by well established techniques (DSC, NMR) and a relatively new technique (PTA). The same techniques were used to evaluate the role of gluten and starch in the process of the loss of crispness.

4.2 Materials and methods.

4.2.1 Materials

Model bread crusts were prepared using a soft wheat flour (Soissons) and two air-classified fractions of a wheat flour (a protein rich and a starch rich fraction). The flour samples were purchased from Meneba (Meneba Meel BV, Rotterdam, the Netherlands). The composition of the flour samples is shown in table 1. Dry yeast (Fermipan) was kindly supplied by DSM bakery ingredients (Delft, the Netherlands). Native wheat starch (Excelsior) was purchased from Avebe (Veendam, the Netherlands) and gluten (Protimax 137) was purchased from Avebe Latenstein (Nijmegen, the Netherlands).

4.2.2 Methods

4.2.2.1 Preparation of the model crust

Dough was mixed in a farinograph mixer (Brabender, Duisburg, Germany) using a 50 g mixing bowl. The formulation included flour, NaCl (2 g/100g flour), dry yeast (1.7 g/100 g flour) and ascorbic acid (20 ppm) for the model crust of Soissons flour. For the Soissons flour, the protein rich fraction and the starch rich fraction 55.5%, 80% and 63% water, respectively was used to prepare the dough. The temperature of the added water was 22°C and the starting temperature of the mixing bowl and flour was 23°C, 22.5°C and 23°C, respectively. The dough was mixed at a speed of 100 rpm until the dough temperature reached 26°C, which corresponded with the maximum in the torque-time curve. After mixing, the dough was separated in 3 pieces and made into model crust following the procedure as explained in chapter 3. After baking, the model crusts were left to cool to room temperature and were frozen and stored in closed plastic bags at -20°C until use.

Tabel 1. Composition of the flours, WIAX is water insoluble arabinoxylans, WEAX is water extractable arabinoxylans.

	Moisture (%)	Protein (%)	Starch (%)	WIAX (%)	WEAX (%)	Damaged starch (%)
Starch rich	12.3	9.5	82	0.18	0.47	6.2
Protein rich	10.4	17.3	69	0.05	1.01	10.3
Soissons	14.8	12.9	79	1.63	0.42	4.9

4.2.2.2 Preparation of gelatinized starch

Native wheat starch was dispersed in excess water and boiled at 90°C for 10 minutes. Next the starch was freeze dried and milled into powder with an analytical grinder (type A10, IKA labortechnik, Staufen, Germany).

4.2.2.3 Preparation of heated gluten

35 g of gluten (Protimax 137) was dissolved in 350 ml demi-water with 3.5 g NaCl added. The solution was boiled for 10 minutes at 100°C, cooled down in water and centrifuged for 6 minutes at 7500 rpm (Beckman CS-15R, Beckman Instruments

Inc., Palo Alto, CA, USA). The residue was frozen at -80°C and freeze dried before use.

4.2.2.4 RH equilibration of model crust

The crust samples for DSC and PTA measurements were milled in an analytical grinder for periods of 10 seconds with adequate pauses to prevent heating of the sample. The resulting powder was sieved using a 0.5 mm sieve and the pieces larger than 0.5 mm were discarded. Starch and gluten were also used as powders. The samples were stored in climate chambers at 22°C and equilibrated at a certain RH (40, 50, 55, 60, 65, 70, 75 and 80 %). Samples for NMR experiments (crust, gluten and starch) were also equilibrated at lower RH (0, 20 and 30% RH). All samples were equilibrated for 5 days, which was enough to obtain a steady weight.

4.2.2.5 Water activity

The water activity of the crust samples was measured using an Aqua Lab Series 3 (Decagon devices, Pullman, USA) at 22°C.

4.2.2.6 Water content

Water contents were obtained gravimetrically by drying at 105°C for 15 hours.

4.2.2.7 Low field NMR experiments

Proton relaxation measurements were made using a Minispec MQ20 20 MHz NMR analyzer (Bruker, Germany) operating at a resonance frequency of 19.95 MHz. Equilibrated samples (approximately 0.5 g each) were placed into glass vials (18 cm height and 9 mm diameter). The vials with samples were closed with a cap to limit moisture loss and measured at 25°C. The transverse relaxation (T_2) time in the ms time domain was measured using the Carr–Purcel–Meiboom–Gill (CPMG) pulse sequence, which consists of a 90° pulse followed by a train of 180° pulses to refocus the NMR signal. The 90–180 pulse spacing was set to 0.05 ms. Relaxation curves obtained from the CPMG sequence were analyzed with a Contin routine as described by Provencher (*14*) using Matlab software (Matwork version 7.1). Additionally 2 exponentials were fitted to the data.

4.2.2.8 Phase transition analyzer (PTA)

The PTA uses a combination of time, temperature, pressure, and moisture to measure the

T_g and T_m of a biopolymer sample (*15*). It consists of two sealed chambers, top and bottom, separated by an interchangeable capillary dye. For the experiments presented in this paper only the closed dye was used (the open dye is only used when evaluating flow behaviour). The two chambers house electric heaters and contain a hollow cavity that allows a cooling fluid to be used. The pistons, mounted together through sidebars, are held in a fixed position during testing. Air cylinders, mounted to the bottom of the PTA, maintain constant pressure on the sample. The sample was filled in the upper chamber, which was closed by the piston afterwards. A linear-displacement transducer measures the sample’s deformation, compaction, and flow relative to initial sample height (*16*). In this study the measurement of the T_g was performed at 100 bar and with a heating rate of 5°C per minute. The temperature at the start of the measurement was around 20°C. 1 or 1.8 g of sample was used for each measurement and the measurements were performed in duplo.

A first derivative was calculated from the displacement versus temperature data. For the transition temperatures of starch (both gelatinized and native) all peaks were considered. Only one peak was changing with water content or water activity (generally the second peak) . The maximum of this peak was then taken as the transition temperature. For the evaluation of the transition temperature of the model crusts all peaks were considered as well. Since the first peak may be due to starting up of the movement of the plunger the second peak was taken as the transition temperature. At high a_w (> 0.70) only one peak was observed

4.2.2.9 Differential Scanning Calorimetry (DSC)

Glass transition

DSC experiments were performed using a modulated DSC with nitrogen cooling (MDSC2920, TA Instruments). Aluminium sample cups of 40 µl were used with around 20 mg of grinded bread crust sample (<0.5 mm). The samples were first heated from 20 to 135°C at a speed of 10°C per minute and cooled down to -20°C in ~ 5 min in order to remove the retrogradated starch signal. Next, the analysis was performed from -20 to 120°C at a rate of 1°C per minute. The period and the amplitude of modulation were 40 s and 0.5°C. The glass transition temperature was taken from the second scan of each sample by measuring the midpoint of the shift in specific heat (dCp).

Amount of gelatinized starch

Freeze-dried crust and crumb samples were weighed and distilled water added at a 3:1 (v/w) water to sample ratio in stainless steel pans. DSC measurements were performed with a Perkin Elmer Diamond DSC calorimeter (Perkin Elmer Corp., USA). Indium was used to calibrate the system. The samples were heated from 10 to 130°C at 10°C/ min. An empty stainless steel pan was used as a reference during the DSC measurement. The enthalpy was expressed in J/g of sample (db). Relative crystallinity (D_{RC}) was calculated as:

$$D_{rc} = \frac{\Delta H_s}{\Delta H_f} * 100 \qquad (1.1)$$

where ΔH_s represents the melting enthalpy (J/g) of starch crystallites in the sample (peak 70°C) that did not gelatinize during baking and ΔH_f represents the enthalpy (J/g) of starch gelatinization in the flour.

4.2.2.10 Sensorial testing

Model crusts equilibrated at relative humidities from 40% to 80% were presented to a trained panel of 8 persons (3 males) in randomized order. The panellists were between 20 and 62 years old. The test was repeated on a second day with freshly prepared samples. Panellists were asked to score the degree of crispness of the model bread crusts during chewing. This was one of the attributes as assessed earlier (*17*). They had to mark the degree of crispness on a scale of 1 to 10 along a 15 cm line with word anchors on both ends. Pairwise comparison between means was tested using a two-sided t-test (p= 0.05) using GenStat software v 8 (VSN international Ltd., Herts, UK).

The sigmoid relationship between crispness, averaged over panelists and replicates, and the water activity was described using a Fermi-fit:

$$S = S_{\min} + \frac{C}{1 + e^{-B*(X - X_c)}} \qquad (1.2)$$

where S is the expected value of the crispy score averaged over panelists, $S_{\min}$=lower asymptote, C=attribute range (S_{max} – S_{min}), X=water activity (*18*), B =rate parameter at the transition region or steepness, and X_c= water activity water content at the point of inflection (a_{wc}). Non linear regression analysis using Matlab (Matwork, version 7.1) was used to fit the curves.

4.3 Results

4.3.1 Sensorial results

Figure 1a, b and c show the results of the sensorial panel for model bread crusts stored at different RHs. As can be seen the loss of crispness of these model bread crusts starts at a water activity of around 0.55. No difference could be detected between the different formulations for the point where the model crust starts to lose crispness. This probably means that the amount of protein present does not influence the point where crispness starts to decrease. The starting point for loss of crispness is a little lower than the a_w reported earlier for a fresh rusk roll crust (a_w 0.64-0.70) (*3*). The point at which half of the crispness is lost is around a_w 0.6 +/- 0.01 for all 3 formulations. The end of the transition range for model bread crust is at a water activity of around 0.7.

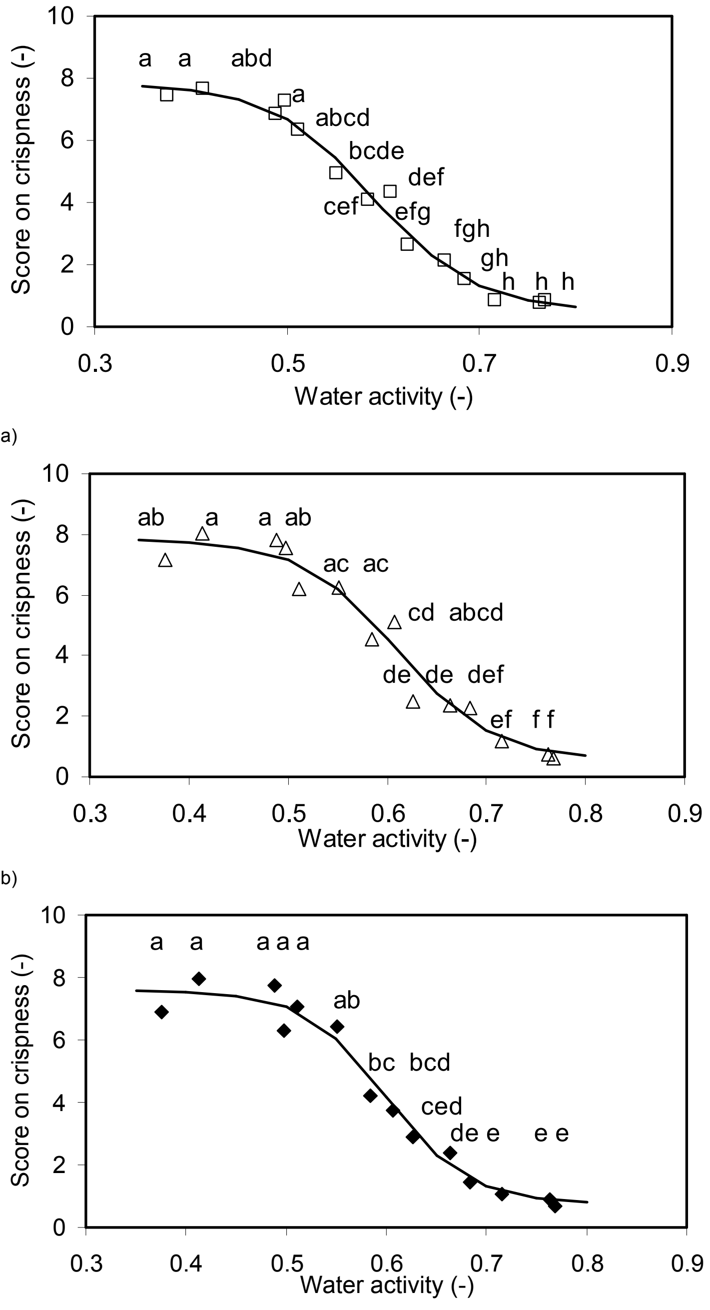
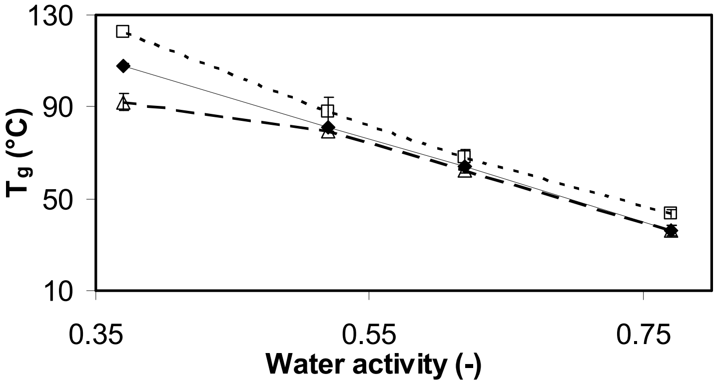
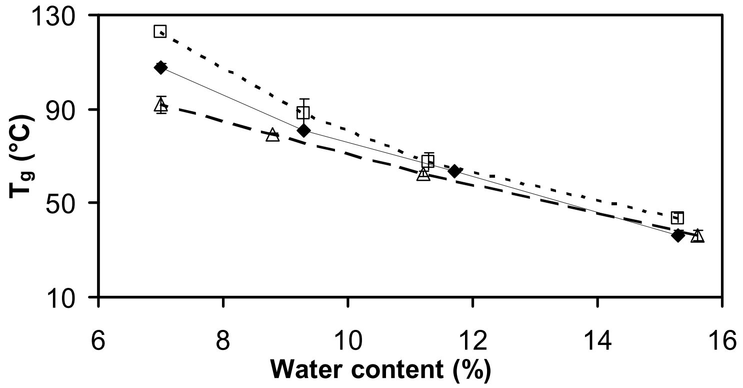


Figure 1. Effect of a_w on crispness scores of model crust of different formulations. Sensory results for model crusts of Soissons flour (a), a protein rich flour (b) and a starch rich flour (c) the solid lines indicate the Fermi-fit. Different letters indicate a significant difference ($p < 0.05$).



(a)



(b)

Figure 2. T_g of model crust as a function of a_w and water content as measured with modulated DSC. Model crust samples of Soissons (□), Starch rich (♦) and Protein rich (Δ) flour plotted against water activity (3a) and water content (3b)

4.3.2 Glass transition measurements with DSC and PTA

4.3.2.1 DSC

Figure 2 shows the results of the DSC experiment for 3 different formulations for different water contents / water activities. It shows that the glass transition temperature as measured with DSC decreases with increasing water content and a_w, reaching room temperature (22°C) at a water content of 15%. No significant difference in the T_g between the starch rich and protein rich flour model crusts could be detected at the higher water activities (a_w 0.62 and 0.77), but at the lower two water activities (a_w 0.37 and 0.52) the model crust of starch rich flour gave a significantly higher transition temperature. The transition temperature as measured with DSC for the model crust of Soissons flour was in all cases significantly higher than the transition temperature of the crust with the other two formulations.

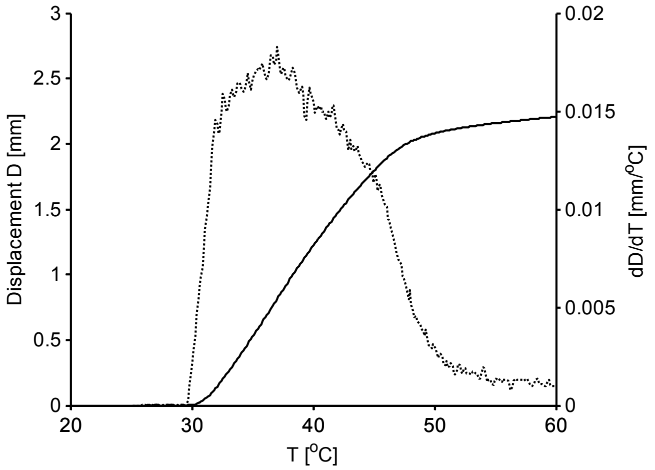


Figure 3 PTA displacement curve and derivative of the displacement curve for Soissons bread crust stored at a RH of 75%. Solid lines indicate the displacement curve with the displacement on the left y-axis, broken lines indicate the derivative curve with the derivative on the right y-axis.

4.3.2.2 PTA

In the PTA the displacement of the piston was measured as a function of temperature. With increasing temperature the displacement increases at constant pressure (figure 3). Figure 3 also shows the result in the form of a first derivative plot of the PTA data for a Soissons model crust that was equilibrated at 75% RH. The maximum in the derivative curve was taken as the transition temperature.

Figures 4a and b show the transition temperatures as measured with PTA for model crusts of the three formulations as a function of water activity and water content. In general, the transition temperature decreases linearly with increasing water content or water activity. When comparing formulations it should be noted that the starch-rich and protein-rich model crusts have similar transition points as a function of water activity, whereas the crust from Soissons flour has a transition temperature of around 10°C higher between a_w 0.65 and 0.75. Model crusts of Soissons flour contain a little higher water content (figure 5) and therefore when plotted against water content the transition temperatures of the Soissons model crust are around 10°C higher over a larger range (between 8 and 13% water content). A similar trend was found in the DSC results. Figure 4c also shows the transition temperatures of the separate components starch and gluten. The transition temperatures of both gelatinized and native starch are around 20°C higher than those of the model crusts, when plotted against water content. This is because especially the water content of the starch is very different from that of the model crusts at equal water activity (figure 5). The transition temperatures of the gluten are much closer to the transition temperatures of the model crust. When plotted against a_w no differences were observed between the transition points of the starch, gluten and model crust as measured with PTA (results not shown). A difference between the transition temperatures of native and gelatinized starch could not be detected with the PTA.

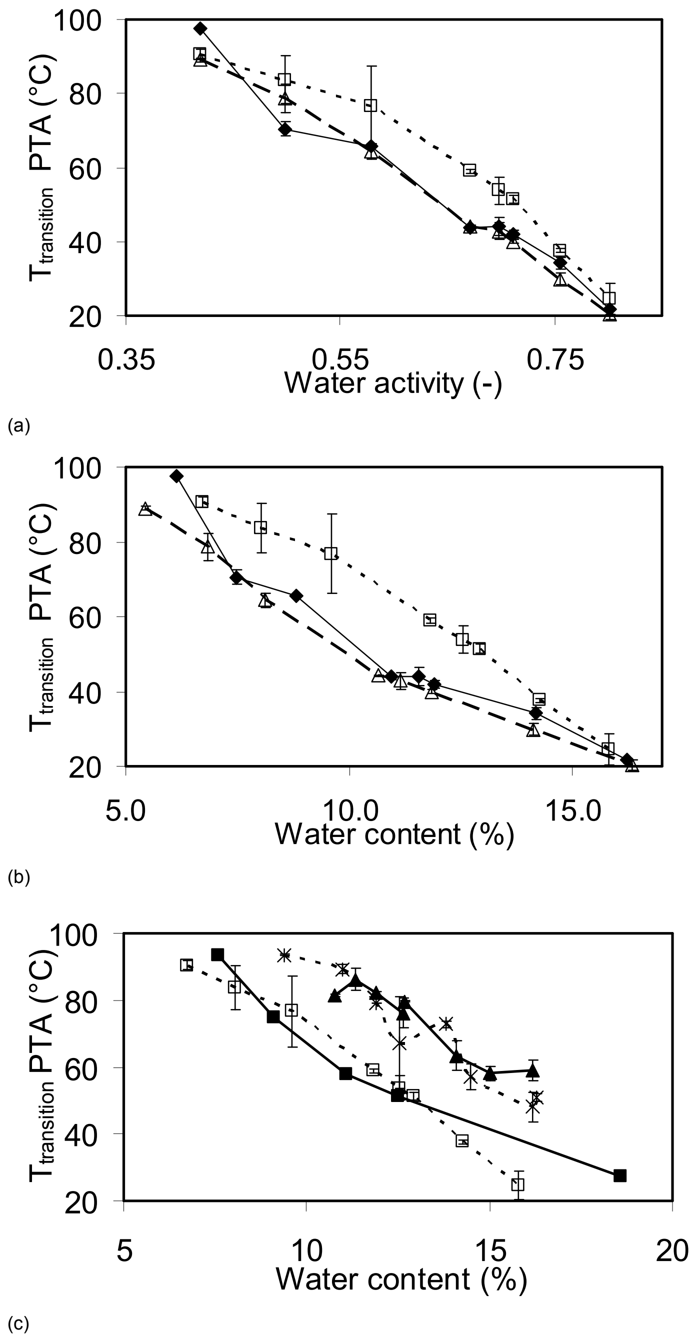


Figure 4a PTA transition temperatures for model crusts samples as a function of a_w and water content. Model crusts of Soissons (□), Starch rich (♦) and Protein rich (Δ) flour as a function of water activity (a) and water content (b) and PTA transition temperatures compared for native (▲) and gelatinized starch (x), model bread crust (Soissons) (□)and native gluten (■) as a function of water content (c).

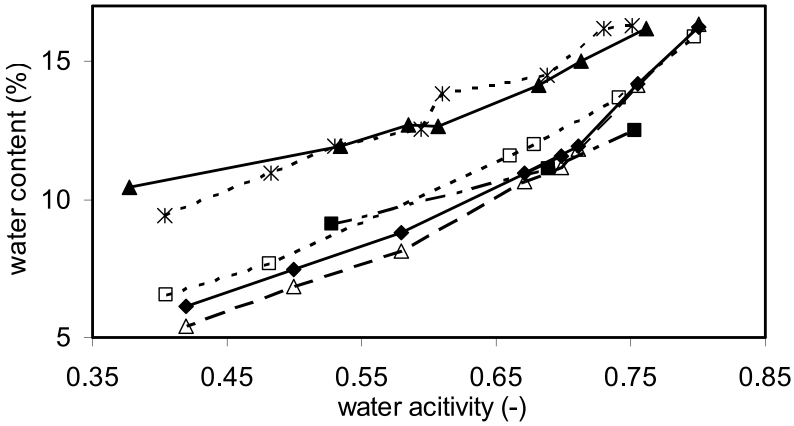


Figure 5 Relation between water content and a_w for model crusts and its main components. Water content of native (▲) and gelatinized starch (x) and model bread crust of Soissons (□), Starch rich (♦), Protein rich (Δ) flour and native gluten (■) as a function of a_w.

4.3.3 NMR

In order to evaluate the state of the water in model bread crusts and their separate components within the same water activity range, we measured the mobility of the protons in the samples with NMR. The T_2 value obtained with a CPMG pulse sequence is a good representative of proton mobility (*19*). A higher T_2 points at a higher mobility of the protons of water. The results for T_2 relaxation time of the protons of water at different water activities for Soissons model bread crusts are shown in figure 6. The T_2 value is a result of a double exponential fit. One of the exponential contributions accounted for 90% of the intensity. The T_2 belonging to this contribution is discussed here. The results of the fits to bi-exponential functions corresponded with the positions of peaks obtained from a Contin fit (i.e. an inverse La Place transform of the time domain NMR signal resulting in distributions of relaxation rates) for all samples except for the heated gluten. The transition points were at approximately the same value of a_w for the Contin fit and the two exponential fit. T_2, which reflects the mobility of protons and therefore the mobility of water, increased with increasing water activity. At lower water activity (up to a water activity of around 0.58) the T_2 value for this fraction of water protons increased very slowly with increasing water activity. This probably means that not all water-binding sites have been hydrated at a water activity below ~ 0.58 and the free sites are progressively occupied with increasing water activity. At higher water activities the T_2 value started to increase more steeply. Probably at higher a_w, only part of the water molecules present was directly bound to the polymer matrix and the other part was more affiliated with other water molecules, and therefore the mobility was higher on average. The transition point, defined as the intersection of two tangents, respectively, to the low and high a_w part of the a_w dependence in the case of T_2 coincides with the starting point of sensory loss.

Low field NMR experiments were also performed on the separate components starch, gelatinized starch and gluten (Figure 6). The transition point for starch (both native and gelatinized) is at a lower RH than the transition point of the bread crust (around a_w 0.48 and 0.52 for gelatinized and native wheat starch respectively). For gluten the transition occurs at an a_w of 0.65 (results not shown).

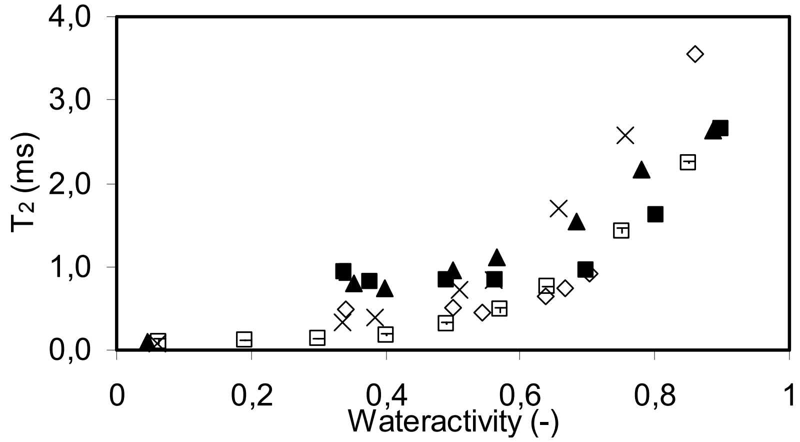


Figure 6. NMR T_2 relaxation times as a function of a_w for model crust and its main components. Results for native (▲) and gelatinized starch (x), model bread crust (Soissons) (□), native gluten (■) and heated gluten (◊).

4.4 Discussion

More information is required about the mechanisms underlying the water induced loss of crispness to improve its retention. The aim of this study was to investigate the relation between loss of sensorial crispness in a bread crust model and the glass transitions temperature. Besides that gluten and starch were investigated separately and compared with the results for bread crust in order to find out whether it is the starch or the gluten that gives the crispness to a bread crust. Model bread crusts were used because these can be reproducibly made in sufficient quantities. The alternative: isolating the crust from bread is very laborious and less reproducible, since the separation of crust and crumb is cumbersome and difficult. Typically, a bread crust is characterized by incomplete starch gelatinization, a continuous protein matrix and a low water content (3, 12). This is also a feature of the model crust although there is a difference in degree of relative cristallinity between a model crust and a real bread crust (from a crispy roll as discussed in chapter 6) as determined by DSC (14 % +/-3 for the model crust and 28% +/-9 for the real crust). This difference is expected to be too small to influence the results. We therefore regard the model bread crust as representative for a real bread crust. It should be noted that the samples were conditioned at different RH's until a steady state was reached. The kinetics of loss of crispness were not studied here, but in chapter 3 the water uptake kinetics were found to be significantly slower for the protein rich flour model crusts as measured by an oscillatory sorption experiment.

Tabel 2. Degree of gelatinization of the model crusts as measured by DSC

Model crust	Degree of gelatinization (%)
Starch rich model crust	85
Protein rich model crust	91
Soisssons model crust	86

4.4.1 DSC

Le Meste et al (*6*) were not able to detect a thermal event identifiable with a glass transition for bread and extruded bread in the water content range of 4-18%. They suggested this may be due to the heterogeneity inherent to this type of food product. With thermomodulated DSC results could be obtained for a commercial bread toast (*13*). Lower T_g values were found (about 40°C lower when plotted against water content) in the present study. This may be due to a higher content of sugars in bread toast, since sugar is often added to this type of products. Sugar may cause a lowering of the T_g (*20*). Also the starch will be completely gelatinized in toasted bread crumb and native wheat starch has a higher glass transition temperature compared to gelatinized wheat starch (*21*). From the comparison with literature data (*22-24*) we conclude that the TMDSC shows a transition of the (gelatinized) starch in our bread crust. The transition temperatures found for gluten in literature were around 25°C lower than for starch when plotted against water activity. This was expected since starch is the main component in the bread crust. A glass transition of the gluten was not observed in the bread crust samples.

4.4.2 PTA

The transition temperatures in the PTA experiment were calculated by taking the temperature at the maximum of the first derivative curve. For water activities below 0.70 this derivative curve showed more than one peak. A range of transitions may occur during the heating of the sample due to the inhomogeneity of the sample. The transition may start with the particles sliding past each other followed by a glass transition (or a range of glass transitions). It is also possible that in the beginning some artefacts occur due to the starting up of the movement of the plunger or some packing/reorganisation of the material particles. The technique turned out to be useful for the determination of a glass transition in inhomogeneous powdered products like bread crust. We always found a transition temperature with the PTA, which is for example with a DSC not always possible, as above mentioned for bread toast as measured by le Meste et al (*6*).

Figure 7 shows a comparison between TMDSC results and PTA results for one formulation (protein rich flour model crust) . It turns out that TMDSC gives a transition temperature around 10°C higher than the PTA. This is in disagreement with the findings of Bengoechea et al (*22*) who found that DSC gave a lower transition temperature than the PTA However, these results were observed for gluten whereas here a different product was compared. It must therefore be concluded, that the transition points obtained by PTA and DSC are only qualitatively comparable, but the trend as a function of water content is the same.

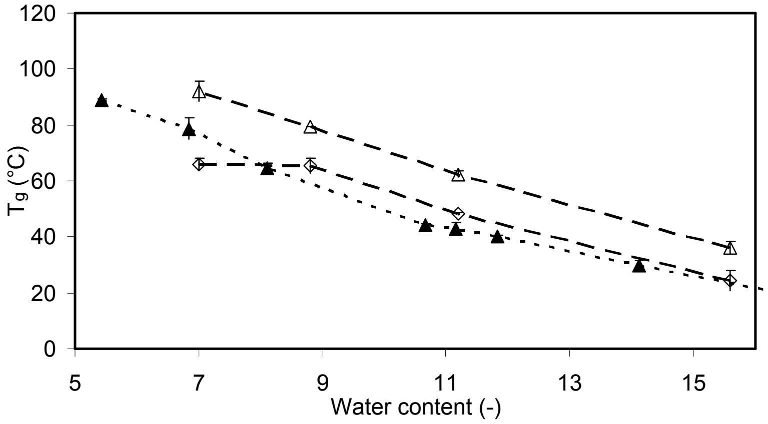


Figure 7 Transition temperatures of a protein rich model crust as a function of water content. Results for both T_g (Δ) and T_{onset} (◊) as measured with TMDSC and $T_{softening}$ (▲) as measured with a PTA.

4.4.3 NMR

The transition point between crispy and non-crispy that was obtained from the sensory panel at a water activity of around 0.55 is equal to the transition point that was found by the CPMG NMR measurements (at a water activity of 0.58). These results indicate that the free water available in crispy products causes an increase in the mobility of the macromolecules and that it is this water that is responsible for the loss of the crispness of these bread crusts. Ruan and Chen (*25*) state that the inflexion point in the NMR curve suggests a transition from the immobile state to the mobile state. They also found that the degree of firming of high-protein bars correlated with the mobility of protons associated with these proteins (*26*). This agrees with our findings.

The transition points in the NMR T_2 curve for starch and gluten are at a somewhat lower and higher RH than the transition point for model crust, respectively. The finding that the T_2 transition point of the gluten is at higher a_w than the transition point of starch is in contradiction with the reported glass transition temperatures (T_g) for starch and gluten in literature as measured with DSC or TMA (*8, 23*) or as measured with PTA in our study. The T_g for starch was generally found to be 20-30°C higher than the T_g of gluten. Similar results were found by Chinachoti (*27*) for starch and gluten and by Choi and Kerr for native wheat starch (*28*) with NMR as well. Chinachoti (*27*) did not find mobile water protons at 2% water in starch and at 7.5% water in gluten with solid state ^{1}H NMR. At 10% moisture the water in the starch sample was rather mobile whereas the water signal in gluten samples at 7.5% moisture was incorporated as part of the broad solid signal. This suggested that the mobility of water hydrating gluten is lower than the mobility of water hydrating starch.

Attenburrow et al (*29*) found the same order in transition points for gluten and starch as reported here for the T_2 transition point. They used a texture analyser to analyse the fracture behaviour of extruded starch and gluten granules. They recorded the acoustic emission when breaking the starch and gluten granules at different water contents. Gluten still emitted sound when breaking up to a higher water content than starch did. The transition points were at around 12 and 10% water for gluten and starch respectively at room temperature (corresponding to an a_w of 0.72 and 0.35 respectively). In a second study they performed a sensory test on the starch and gluten extrudates that showed that the starch extrudates lost their sensorial crispness between a water activity of around 0 and 0.23 corresponding to a water content between 1.4 and 8.1%. The gluten extrudates lost their crispness between a water activity of 0.35 and 0.57 corresponding to a water content between 7.3 and 9.3%. The sensorial loss curve was much steeper for starch than for gluten (*30*). The critical water activity for loss of crispness of gluten is much closer to the critical water activity for loss of crispness of a model crust than that of starch. This suggests that it is indeed the gluten network in the crust that gives a bread crust its crispness.

The order in transition points resulting from NMR and sensorial experiments is different from the order in transition points as found with DSC and PTA for starch and gluten (*23*). This may be due to the fact that DSC and PTA give a more macroscopic transition point, where the complete macromolecule starts to move and NMR shows increased mobility of small parts like water and probably small side chains. Our results would suggest that indeed the movement of these small parts or components cause loss of crispness. This was also suggested by Roudaut et al (*31*) in an earlier study on dry bread. Farhat et al (*32*) found indications for increased mobility of sugar in sugar-starch mixtures at water contents above 10-15%. Probably small components in the vicinity of gluten or side chains of gluten start to gain mobility at a higher a_w than small components in the vicinity of starch or side chains of starch. This increased mobility of small components and side chains might induce increased energy dissipation upon deformation of the material resulting in less available energy for fracture and with that in a less crispy product (*1*).

4.5 Conclusions

Sensorial data with respect to crispness and results from DSC and PTA were compared. There appears to be no full agreement between these methods for assessing the transition from a glassy to a more ductile state in model bread crust. On the other hand, NMR gives a transition point that is equal to the point where the model crusts start to lose sensory crispness. On this basis, it is suggested that it is the increase in mobility of water which is indicative for the loss of crispness. Also other small molecules and side chains of the larger macromolecules may become more mobile at this point. Probably these movements play a crucial role in the fracture behaviour of the crispy network in bread crust, because this increased mobility could lead to energy dissipation upon deformation of the crispy material. Only at much higher water content or water activity the mobility of the macromolecules starts to increase, resulting in a glass transition as measured with

DSC and PTA. Agreement between sensory data presented here and that of Nicholls et al (*30*) supports the conclusion that loss of sensory crispness is related to changes in the (mobility of the) gluten network in a bread crust.

Acknowledgements

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References

1. Luyten, H.; Plijter, J. J.; van Vliet, T., Crispy/crunchy crusts of cellular solid foods: a literature review with discussion. *Journal of Texture Studies* **2004,** 35, (5), 445-492.
2. Meste, M. le; Champion, D.; Roudaut, G.; Blond, G.; Simatos, D., Glass transition and food technology: a critical appraisal. *Journal of Food Science;* **2002,** 67, (7), 2444-2458.
3. Primo-Martín, C.; van de Pijpekamp, A.; van Vliet, T.; de Jongh, H. H. J.; Plijter, J. J.; Hamer, R. J., The role of the gluten network in the crispness of bread crust. *Journal of Cereal Science* **2006,** 43, (3), 342-352.
4. Roudaut, G.; Dacremont, C.; le Meste, M., Influence of water on the crispness of cereal-based foods: acoustic, mechanical, and sensory studies. *Journal of Texture Studies* **1998,** 29, (2), 199-213.
5. Slade, L.; Levine, H., A food polymer science approach to structure-property relationships in aqueous food systems: non-equilibrium behavior of carbohydrate-water systems. *In Water Relations in Foods (L. Slade and H. Levine, eds.) pp. 29–101, Plenum press, New York.* **1991**.
6. Meste, M. le; Roudaut, G.; Davidou, S., Thermomechanical properties of glassy cereal foods. *Journal of Thermal Analysis* **1996,** 47, 1361-1375.
7. Nikolaidis, A.; Labuza, T. P., Use of dynamic mechanical thermal analysis (DMTA), Glass transitions of a cracker and its dough. *Journal of Thermal Analysis* **1996,** 47, 1315-1328.
8. Huang, V. T.; Haynes, L.; Levine, H.; Slade, L., Glass transitions in starch, gluten and bread as measured by dielectric spectroscopy and TMA methods. *Journal of Thermal Analysis* **1996,** 47, 1289-1298.
9. Montes, H.; Mazeau, K.; Cavaille, J. Y., The Mechanical beta relaxation in amorphous cellulose. *Journal of Non-Crystalline Solids* **1998,** 235-237, 461-421.
10. Hugh-Iten, S.; Handschin, S.; Conde-Petit, B.; Escher, F., Changes in Starch Microstructure on Baking and Staling of Wheat Bread *Lebensmittel Wissenschaft und Technologie* **1999,** 32, (5), 255-260.
11. Morgan, K. R.; Furneaux, R. H.; Stanley, R. A., Observation by solid-state 3C CP MAS NMR spectroscopy of the transformations of wheat starch associated with the making and staling of bread. *Carbohydrate Research* **1992,** 235, 15-22.
12. Primo-Martín, C.; van Nieuwenhuijzen, N. H.; Hamer, R. J.; van Vliet, T., Crystalline changes in wheat starch during the bread-making process: starch crystallinity in the bread crust. *Journal of Cereal Science* **2007,** 45, 219-226.

13. Braga, A. L. M.; Cunha, R. L., Plasticization and antiplasticization by small molecules in brittle cellular food: TMDSC and Mechanical Properties. *International Journal of Food Properties* **2004,** 7, (1), 105-120.

14. Provencher, S. W., Contin: A general purpose constrained regularization program for inverting noisy linear algebraic and integral equations *Computer Physics Communications* **1982,** 27, (3), 229-242.

15. Strahm, B.; Plattner, B.; Huber, G.; Rokey, G., Application of food polymer sience and capillary rheometry in evaluating complex extruded products. *Cereal Foods World* **2000,** 45, (7), 300-302.

16. Plattner, B.; Strahm, B.; Rausch, K., The phase transition analyzer and its impact on extrusion processing of foodstuffs. *ASAE Annual International Meeting, Sacramento, California, USA: ASAE, St. Joseph, MI;* **2001,** ASAE Meeting Paper Number 01-6067.

17. Dijksterhuis, G.; Luyten, H.; de Wijk, R.; Mojet, J., A new sensory vocabulary for crisp and crunchy dry model foods applied. *Food Quality and Preference* **2007,** 18, (1), 37-50.

18. Matsunaga, K.; Kawasaki, S.; Takeda, Y., Influence of physicochemical properties of starch on crispness of tempura fried batter. *Cereal Chemistry* **2003,** 80, (3), 339-345.

19. Choi, S. G.; Kerr, W. L., Effects of chemical modification of wheat starch on molecular mobility as studied by pulsed 1H NMR. *Lebensmittel Wissenschaft und Technologie* **2003,** 36, (1), 105-112.

20. Kalichevsky, M. T.; Blanshard, J. M. V., The effect of fructose and water on the glass transition of amylopectin. *Carbohydrate Polymers* **1993,** 20, 107-113.

21. Zeleznak, K. J.; Hoseney, R. C., The glass transition in starch. *Cereal Chemistry* **1987,** 64, (2), 121-124.

22. Bengoechea, C., Arrachid, A., Guerrero, A., Hill, S.E. and Mitchell, J.R., Relationship between the glass transition temperature and the melt flow behavior for gluten, casein and soya. *Journal of Cereal Science* **2007,** 45, (3), 275-284.

23. Laaksonen, T. J.; Roos, Y. H.; Labuza, T. P., Comparison of the use of desiccators with or without vacuum for water sorption and glass transition studies. *International Journal of Food Properties* **2001,** 4, 545-563.

24. Micard, V.; Morel, M. H.; Bonicel, J.; Guilbert, S., Thermal properties of raw and processed wheat gluten in relation with protein aggregation. *Polymer* **2001,** 42, 477-485.

25. Ruan, R.; Chen, P., Nuclear Magnetic Resonance Techniques. *IN: Bread Staling, Ed. Chinachoti, P., Vodovotz, Y.* **2001**.

26. Lin, X.; Ruan, R.; Chen, P.; Chung, M.; Ye, X.; Yang, T.; Doona, C.; Wagner, T., NMR State Diagram Concept. *Journal of Food Science* **2006,** 71, (9), 136-145.

27. Chinachoti, P., NMR dynamics properties of water in relation to thermal characteristics in bread. *In: D.S. Reid, The properties of water in foods, ISOPOW 6. Blackie Academic & Professional, London* **1998**, 139-158.

28. Choi, S. G.; Kerr, W. L., 1H NMR studies of molecular mobility in wheat starch. *Food Research International* **2003,** 36, (4), 341-348.

29. Attenburrow, G. E.; Davies, A. P.; Goodband, R. M.; Ingman, S. J., The fracture behaviour of starch and gluten in the glassy state. *Journal of Cereal Science* **1992,** 16, (1), 1-12.

30. Nicholls, R. J.; Appelqvist, I. A. M.; Davies, A. P.; Ingman, S. J.; Lillford, P. J., Glass transitions and the fracture behaviour of gluten and starches within the glassy state. *Journal of Cereal Science.* **1995,** 21, (1), 25-36.

31. Roudaut, G.; Maglione, M.; Meste, M. le, Relaxations below glass transition temperature in bread and its components. *Cereal Chemistry* **1999,** 76, (1), 78-81.

32. Farhat, I. A.; Mitchell, J. R.; Blanshard, J. M. V.; Derbyshire, W., A pulsed 1H NMR study of the hydration properties of extruded maize-sucrose mixtures. *Carbohydrate Polymers* **1996,** 30, 219-227.

Chapter 5 Water content or water activity, what rules crispy behaviour?

Abstract

A dry crust loses its crispness when water migrates into the crust. It is not clear if it is the amount of water absorbed or the water activity (a_w) that leads to a loss of crispness. The hysteresis effect observed when recoding a water sorption isotherm allowed us to study effects of a_w and moisture content separately. All experiments were carried out on model bread crusts made from Soissons bread flour. The effect of water content and water activity on the glass transition of model bread crusts was studied in detail using two complimentary techniques: phase transition analysis (PTA) and Nuclear magnetic resonance (NMR). The results were compared with sensory data and results from a puncture test which provided data on acoustic emission and fracture mechanics during breaking of the crusts. The water content of the crust was found to be decisive for the transition point as measured by PTA and NMR. However both water content and water activity had an effect on perceived crispness and number of force and sound peaks. From this may be concluded that the distribution of the water in the samples with a history of high water content is more inhomogeneous which results in crispy and less-crispy regions thus making them overall more crispy than samples with the same water content but higher $a_{w.}$

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5.1 Introduction

It is well known that crispness of dry cellular foods is lost when the water activity or water content of the products rises above a critical value. Sensorial evaluations of crispy food products have been performed on e.g. crispy breads (*1*), model bread crusts (chapter 4 of this thesis), toasted rusk roll (*2*) and extruded starch products (*3*). In general, above a water content of 9% loss of crispness is observed in these products. Katz and Labuza (*4*) evaluated potato chips, popcorn, puffed corn curls, and saltines at different water activities with respect to crispness and textural hedonic quality with a sensory panel. Critical water activities (a_c), where the products became organoleptically unacceptable, generally fell in the 0.35–0.50 a_w range. This corresponded to critical water contents of 4.2-7.0% db. Sauvageot and Blond (*5*) found the same for breakfast cereals. Water causes hydration of the components of the crispy products. This results in an increase of the mobility of the macromolecules, which causes a glass to rubber transition of the amorphous regions of the macromolecules present (mainly proteins and carbohydrates) that were initially in the glassy state. Roughly at the same time a loss of the crispy behavior occurs (*1, 6*). Several authors studied the relation between the glass transition of bread-like products at low moisture contents and the sensorial loss of crispness in more detail (*7-9*). They found that loss of crispness already occurred while the crispy material was still in the glassy state, which is remarkable because it suggests that increased mobility of the macromolecules is not causing the loss of crispness.

The aim of this study was to investigate the relation between loss of sensorial crispness in a bread crust model with a specific emphasis on the role of water content and water activity. It is important to know whether the water content or the water activity should be used to explain loss of crispness. Water activity will determine the direction of water migration, but water content may finally determine the level of crispness. Samples of bread crust were brought at different water contents and equal water activities by making use of the fact that different water contents can be achieved by varying the RH trajectory (hysteresis effect). Here, different water activities at the same water content or *vice versa* were obtained by either exposing a dry sample to the desired water vapour pressure, or by exposing it to 90% air humidity first, and than dry back under the same conditions as the first sample. For example the crust of a protected atmosphere packaged part-baked bread will follow the trajectory of high RH and then drying back when baked-off, whereas a bread that is baked completely in one step will not show this hysteresis effect.

The hysteresis effect implies that products with a similar water activity can have a different water content and *visa versa* depending on their history. Wolf et al (*10*) classified the variations in hysteresis loops in foods into three general groups, namely high-sugar/high-pectin (eg apple), high-protein (eg pork) or high-starch systems (eg rice). Starch gives the largest hysteresis loop of the three groups. Several theories have been suggested to explain hysteresis for porous solid foods. Most were established on the basis of capillary condensation phenomena (*11, 12*), but for starch type products also activation of sorption sites due to the history of high water content was suggested as a possible explanation (*10, 12, 13*).

The glass transition temperature of the samples studied were measured by a Phase Transition Analyser (PTA) (*14*). By this technique a transition temperature in the mechanical behaviour of the material is measured. This technique gives more constant results than for example differential scanning calorimetry, which not always shows a T_g in dry inhomogeneous products (*8*). The PTA is especially suitable for powders. The state of the water in the samples was determined by low field NMR spectroscopy. This is a spectroscopic technique based on the magnetic properties of atomic nuclei and is often used to monitor motional properties of molecules by analysing the relaxation characteristics of the NMR active nuclei, such as ^1H. With respect to protons from water, a difference can be made between bound (short T_2) and free (long T_2) water.

The sensorial difference between the different samples was evaluated by a sensory panel. A texture analyzer was used to evaluate the fracture mechanics and acoustic emission of the different samples.

5.2 Materials and methods.

5.2.1 Materials

Bread crusts were prepared using a wheat flour from cultivar Soissons. The flour was purchased from Meneba (Meneba Meel BV, Rotterdam, the Netherlands). The composition of the flour is shown in table 1.

Table 1. Composition of the Soissons flour (analysed according the methods mentioned in chapter 3)

Moisture (%)	Protein (% db)	Starch (% db)	Lipids (% db)	Damaged starch (% db)
14.8	12.9	79.3	1.0	4.9

5.2.2 Methods

5.2.2.1 Preparation of the model crust

Dough was mixed in a farinograph mixer (Brabender, Duisburg, Germany) using a 50 g mixing bowl. The formulation included flour, NaCl (2 g/100g flour), dry yeast (1.7 g/100 g flour) and ascorbic acid (20 ppm). 55.5% water on flour basis was used to prepare the dough. The temperature of the added water was 22°C and the starting temperature of the mixing bowl and flour was 23°C. The dough was mixed at a speed of 100 rpm until the dough temperature reached 26°C, which corresponded with the maximum in the torque-time curve. After mixing, the dough was separated into three pieces and made into model crust following the procedure explained in chapter 3. After baking, the model crusts were left to cool to room temperature, wrapped in plastic bags and frozen and stored at -20°C until use.

5.2.2.2 RH equilibration of model crust

The crust samples for NMR and PTA measurements were milled in an analytical grinder (type A10, IKA Labortechnik, Staufen, Germany) for 2-3 times 10 seconds (to prevent heating of the sample) until a fine powder was obtained. The powder was sieved using a 0.5 mm sieve. Pieces larger than 0.5 mm were discarded. For the sensory test the model crust samples were presented as such to the sensory panel. The samples were stored in climate chambers at 22°C and 90% RH for 1 night and then put at 40 and 50% RH and 22 °C for 5 days. The reference samples were stored immediately at 40, 50, 57 and 63% RH for 5 days. For NMR and PTA experiments additional samples were stored immediately at 6, 19, 30, 78 and 85% RH.

5.2.2.3 Amount of retrograded starch as determined by DSC

Around 8 mg of powdered and equilibrated model crust samples (<0.5 mm) were weighed into stainless steel pans. DSC measurements were performed with a Perkin Elmer Diamond DSC calorimeter (Perkin Elmer Corp., USA). Indium was used to calibrate the system. The samples were heated from 20 to 100°C at 5°C/ min. An empty stainless steel pan was used as a reference during the DSC measurement. The enthalpy (*15*) of the amylopectin retrogradation was determined (peak at 55°C) and expressed in J/g of sample (db).

The results are shown in table 2. Storage for 5 days at a higher relative humidity (57 and 63%) had more effect on the extent of the retrogradation of starch than storage for one day at 90% RH even though the samples had an equal water content after 5 days. There was no difference between the amount of retrograded starch of the samples stored at 40 and 50% RH with a history of one day at 90% RH compared to the samples that were stored at 40 and 50% RH from a dry state.

Table 2. Enthalpy of the retrogradation peak at 55°C as measured by DSC at a speed of 5°C per minute.

RH sample	dH at 55°C	RH sample	dH at 55°C
40% (a_w 0.4)	0.9	50% (a_w 0.5)	0.8
90%>40% (a_w 0.4)	0.8 +/- 0.2	90%>50% (a_w 0.5)	0.8 +/- 0.0
57% (a_w 0.57)	2.0	63% (a_w 0.63)	1.9 +/- 0.2

5.2.2.4 Water activity

The water activity (*16*) of the crust samples was measured using an Aqua Lab Series 3 (Decagon devices, Pullman, USA) at 22 °C.

5.2.2.5 Water content

Water contents were obtained gravimetrically by drying at 105°C for 15 hours.

5.2.2.6 Low field NMR experiments

Proton relaxation measurements were made using a Minispec MQ20 20 MHz NMR analyzer (Bruker, Germany) operating at a resonance frequency of 19.95 MHz. After 4 days equilibration samples (approximately 0.5 g each) were placed into glass vials (18 cm height and 9 mm diameter) and left in the climate chambers for another day. The vials were then sealed with a cap to prevent moisture loss and NMR measurements were performed at 25°C. The transverse relaxation (T_2) time in the ms time domain was measured using the Carr–Purcel–Meiboom–Gill (CPMG) pulse sequence, which consists of a 90° pulse followed by a train of 180° pulses to refocus the NMR signal. The 90–180 pulse spacing was set to 0.05 ms. Relaxation curves obtained from the CPMG sequence were analyzed by fitting a 2 exponential to the relaxation curve.

The Free Induction Decay (FID) experiment was performed to obtain information about the ratio between solid and mobile protons. The FID was recorded with a recycle delay of 1.5 s. An average of 64 scans was taken. The following expression was fitted to the FIDs:

$$F(t) = Ae^{\frac{-a^2t^2}{2}}\frac{\sin(bt)}{bt} + Be^{\frac{-t}{T_{2m}}} \qquad (1.1)$$

In this equation the parameters A and B represent the contributions of the immobile and mobile protons in the sample. Parameter T_{2m} is the spin-spin relaxation time of the mobile proton fraction. The NMR spectrum of the immobile proton fraction is assumed to be a rectangular line shape with a total width *2b*, convoluted with a Gaussian line shape with a standard deviation given by parameter *a* (*17, 18*).

5.2.2.7 Phase transition analyzer (PTA)

The PTA uses a combination of time, temperature, pressure, and moisture to measure the

T_g and T_m of a biopolymer sample (*19*). It consists of two sealed chambers, top and bottom, separated by an interchangeable capillary dye. For the experiments presented in this paper only the closed dye was used (the open dye is only used when evaluating flow behaviour). The two chambers house electric heaters and contain a hollow cavity that allows a cooling fluid to be used. The pistons, mounted together through sidebars, are held in a fixed position during testing. Air cylinders, mounted to the bottom of the PTA, maintain constant pressure on the sample. The sample was brought in the upper chamber, which was closed by the piston afterwards. A linear-displacement transducer measures the sample's deformation, compaction, and flow relative to initial sample height (*20*).. In this study the T_g was measured at 100 bar and a heating rate of 5°C per minute. The temperature at the start of the measurement was around 20°C. 1 g of sample was used for each measurement. The glass transition temperature was taken as the temperature where the derivative of the displacement temperature plot was a maximum.

5.2.2.8 Sensorial testing

A two alternatives forced choice (2AFC) discrimination test was performed with 17 panellists (3 males) in duplo on 2 different days. The panellists were between 34 and 64 years old. First they were trained by asking them to order 3 model crusts with a water activity of 0.4; 0.64 and 0.85 on crispness. During the test they were asked to point out the most crispy sample of two. They were offered Soissons model crusts of equal a_w, but of different water contents and model crusts of equal water content but of different a_w. The water contents and water activities are shown in table 3. The number of selected samples in the 2AFC test was tested for significance by using a normal approximation of the binomial distribution (the results of the 2 days were taken together resulting in a number of panellists (n) of 34)

5.2.2.9 Mechanical and acoustic measurements

Mechanical testing was performed using a texture analyzer (TA-XT Plus, Stable Micro Systems Ltd., Surrey, UK).-Pieces (different sizes) of model crust were taken for the measurement. The pieces were placed on a solid metal plate and punctured with a cylinder of 2 mm diameter at a speed of 0.1 mm/s. This speed was chosen to discriminate as many fracture events as possible. 10 samples were analyzed for each condition.

The sound emitted during fracturing was recorded simultaneously. For this purpose, an acoustic sensor (Bruel &Kjær 4189 prepolarized free-field ½" microphone plus a 2671 Deltatron preamplifier microphone, Nærum, Denmark) with a frequency band of 6.3 Hz up to 20 KHz and a sensitivity of 50 mV/Pa was used. A fixed distance of 4.5 cm between the model crust and the microphone was used for sound recording. The analogue sound signal and the data of the texture analyzer were digitized using a Brüel & Kjaer Front-end A/D converter system (type number 2827) at a sampling rate of 65 kHz. All fracture and sound tests were performed inside an anechoic acoustic isolated chamber to avoid interference with an external source of sound. Recording and initial signal analyses were performed using Brüel & Kjær Pulse Labshop Software (version 7.0). Both the number of sound pulses and force peaks were counted and a linear distance was calculated for the force-time curve according to Meinders et al (*21*) using Matlab software (Matwork version 7.0.4).

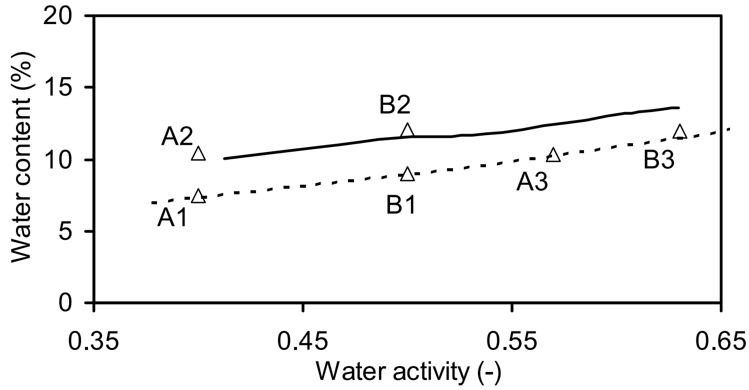


Figure 1. Sorption isotherm of bread crust indicating the samples used in this study. Part of the sorption (broken line) and desorption (solid line) isotherms are shown. The sorption isotherm was started at 40% RH. The desorption curve was started at 90% RH. A1-A3 and B1-B3 indicate the samples as mentioned in the text.

5.3 Results

Samples were evaluated with different techniques to check the effect of water content and water activity on the glass transition temperature as measured with the PTA, water mobility and sensorial, mechanical and acoustic properties of crispy model bread crusts. Figure 1 shows the water activity and water contents of the two series (A and B) that were used for the tests. The reference samples are on the adsorption part of the isotherm and the high water content samples on the desorption part of the isotherm.

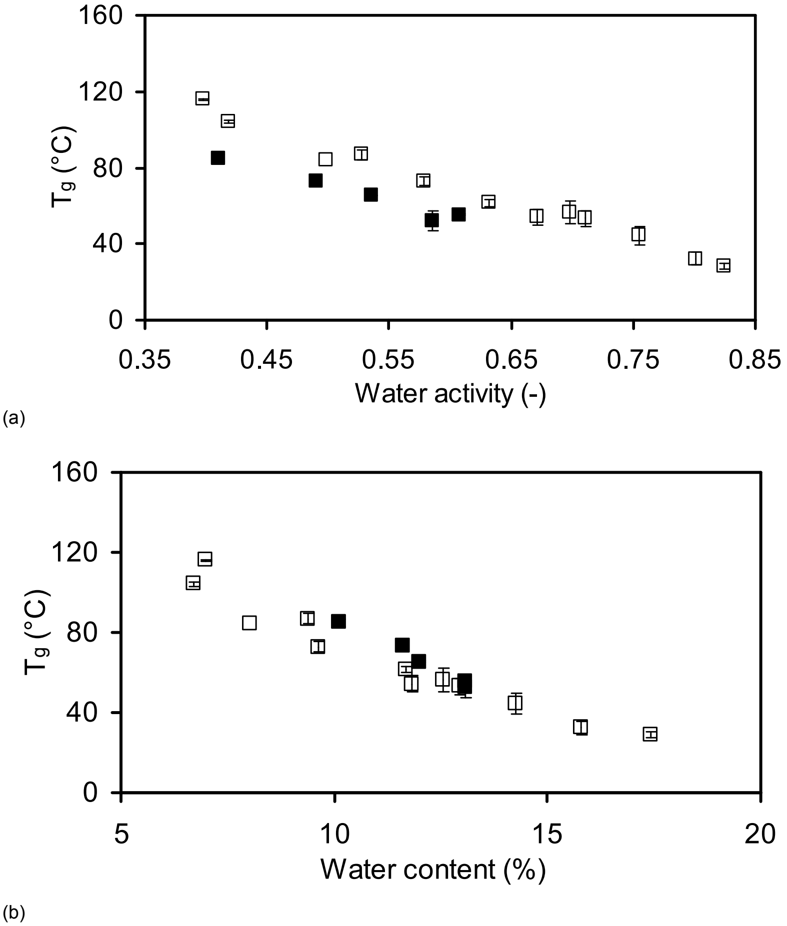


Figure 2a and b. Glass transition temperatures as a function of a_w and water content as measured by the PTA. Results for the reference model crust (□) and model crust with a history of high a_w (■)as a function of a_w (a) and water content (b).

5.3.1 PTA

With the PTA the glass transition temperature of the bread crust particles was evaluated. Samples of different water contents (between 7 and 18%) and a_w (between 0.4 and 0.82) were tested. The water content difference between two samples at the same water activity varied from 3.4% (at the lower RH) to 2% at 65%RH (figure 1). In figure 2 the resulting transition temperatures of the different samples are shown. Figure 2a shows the PTA transition temperatures against water activity and in figure 2b the same transitions temperatures are plotted against water content. The glass transition temperature as measured with PTA decreased with increasing water content and water activity. The PTA results were comparable with the DSC results as obtained in chapter 4 for model crusts of Soissons flour. Figure 2a shows that the samples with a higher water content had a significantly lower glass transition temperature at equal a_w. Figure 2b shows that the difference in water activity of the sample had no influence when comparing the glass transition temperatures of samples with equal water content but different a_w.

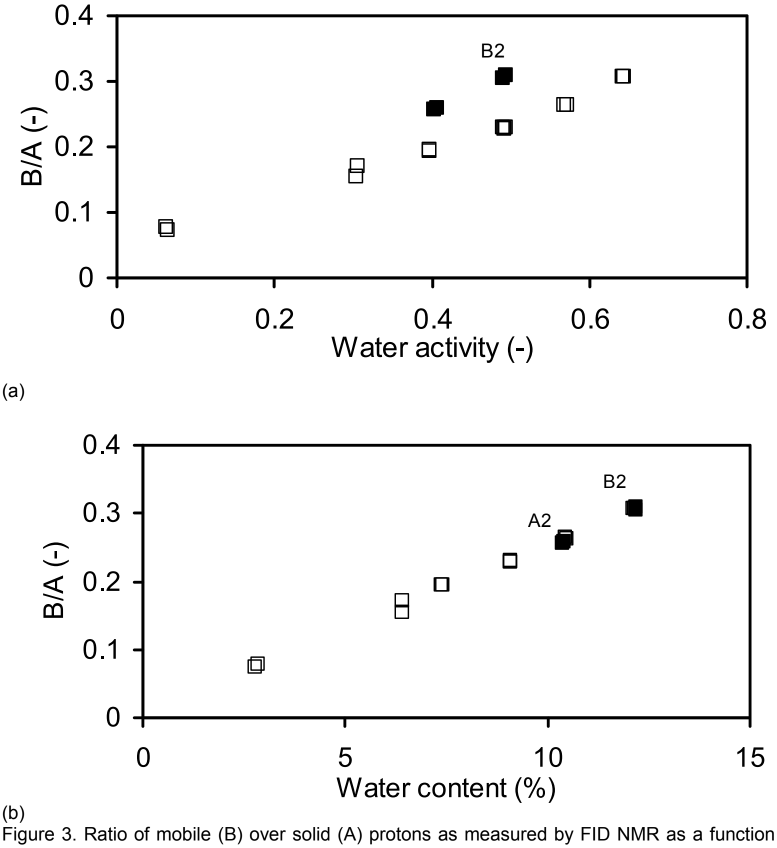


Figure 3. Ratio of mobile (B) over solid (A) protons as measured by FID NMR as a function of a_w and water content. Results for the reference model crust (□) and model crust with history of high a_w (samples A2 and B2) (■) as a function of a_w (a) and water content (b).

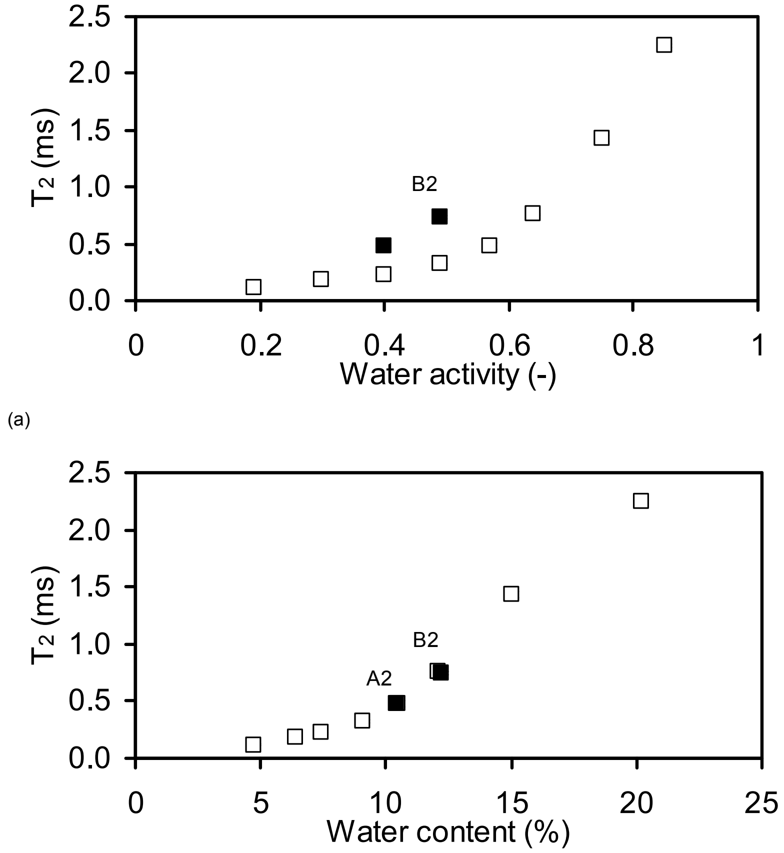
Table 3. Water content of Soissons model crust samples

RH **Sample**	Water content	RH **Sample**	Water content
40% (a_w 0.4) **A1**	7.5 +/- 0.0	50% (a_w 0.5) **B1**	9.0 +/- 0.2
90%>40%(a_w0.4) **A2**	10.4 +/- 0.1	90%>50% (a_w 0.5) **B2**	12.1 +/- 0.0
57% (a_w0.57) **A3**	10.3 +/- 0.2	63% (a_w 0.63) **B3**	12.0 +/- 0.1

5.3.2 NMR

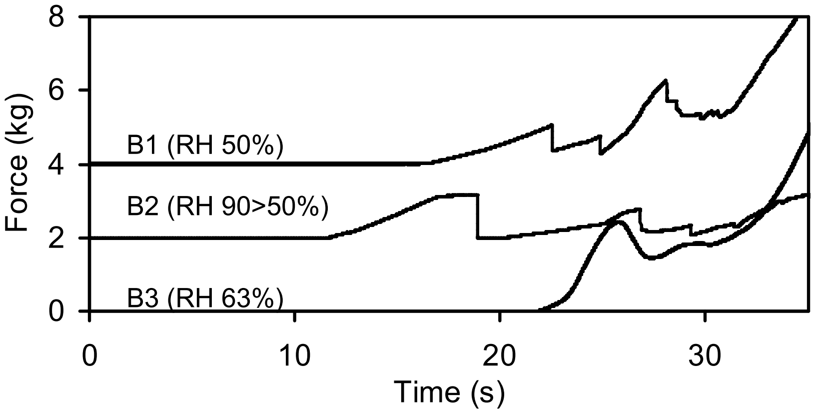
NMR was used to obtain information about the mobility of the (water) protons in the model crust in relation to water content and water activity. A single-pulse free-induction-decay (FID) experiment was used to study the ratio of mobile protons (B) over immobile protons (A). The mobile protons are associated with water and the immobile protons with the solids (e.g. starch) (*22*). The results are shown in figure 3. This ratio increases with increasing water content and a_w. At a fixed water content, this ratio turned out to be unaffected by a variation in water activity.

For the single-pulse experiment a T_{2m} relaxation time associated with the mobility of water was calculated as well. However, as the decay in this region is more affected by field inhomogeneity, a CPMG pulse sequence was subsequently used to study this component (*22*). Figure 4 shows the T_2 relaxation time from a CPMG experiment for the different samples. The protons of the water molecules with a higher mobility will have longer magnetic relaxation times. The T_2 value is a result of a double exponential fit. The shown value for T_2 corresponds to around 90% of the total amplitude. T_2 (mobility of protons and therefore mobility of water) increases with increasing water activity. At lower water activity (up to a water activity of around 0.58) the T_2 value for this fraction of water protons is increasing very slowly with increasing water activity. This probably means that at a molecular level not all water-binding sites have been hydrated at a water activity below ~ 0.58. At higher water activities the T_2 value starts to increase more rapidly. Likely, only part of the water molecules present will be directly bound to the polymer matrix, and therefore the mobility will be higher on average. Figure 4a and b show that at a fixed water content, this relaxation time is unaffected by a variation in water activity.

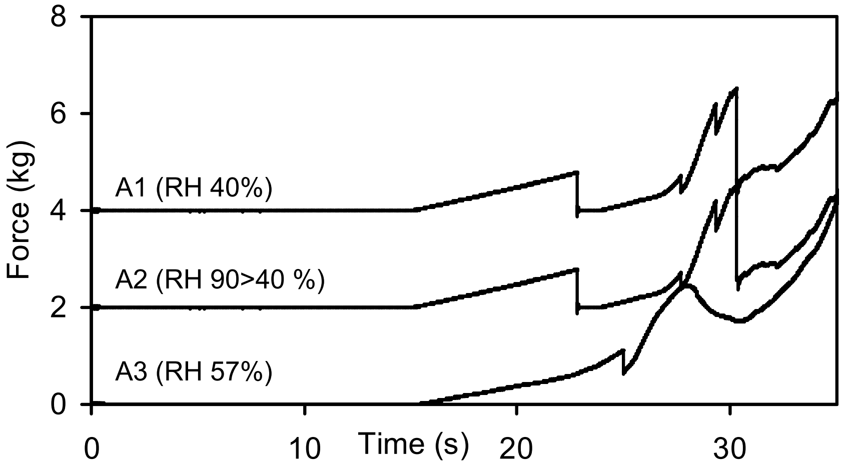


(b)

Figure 4 CPMG NMR T_2 relaxation time as a function of a_w and water content. Results for the reference model crust (□) and model crust with history of high a_w (samples A2, B2) (■). A larger T_2 indicates a higher mobility of the water protons. Samples as a function of water activity (a) or water content (b).



(a)



(b)

Figure 5 Force-time curves as measured with a puncture test with a needle by a texture analyser. Results for samples A1, A2 and A3 (a) and B1, B2 and B3 (b). For clarity reasons the upper lines are shifted along the y-axis.

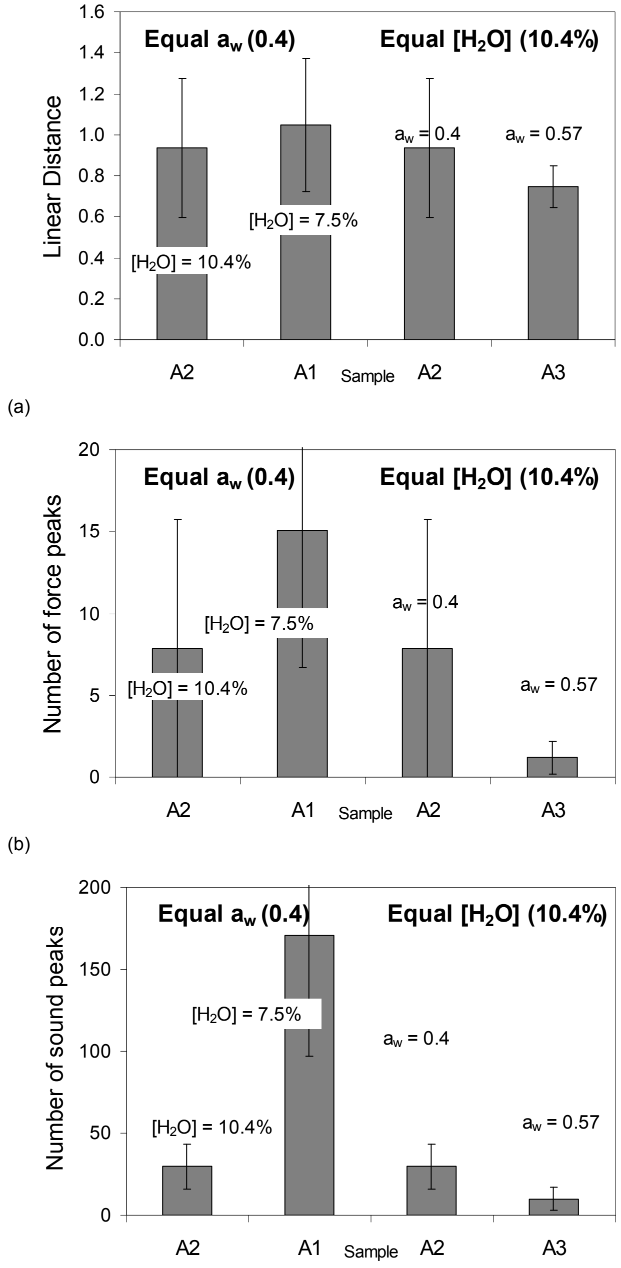
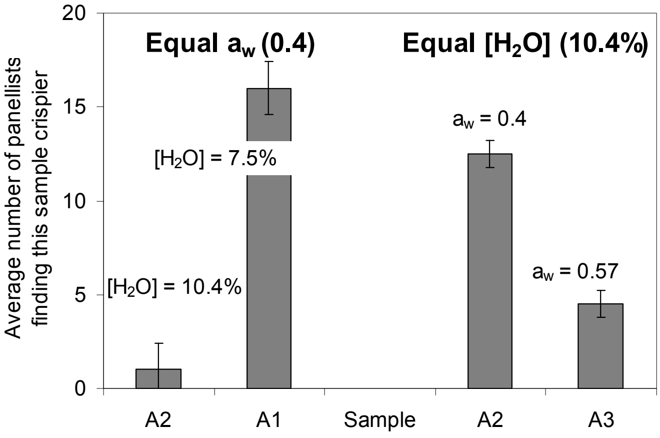


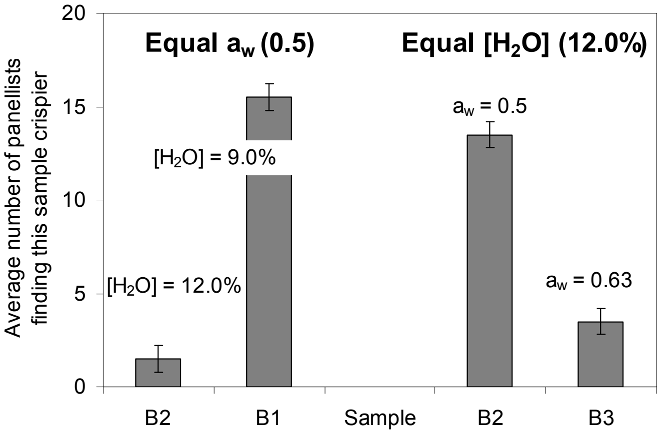
Figure 6 Effect of water content and a_w on the linear distance (LD) and number of force and sound peaks in a puncture test. Results for LD (a), number of force peaks (b) and number of sound peaks (c) for reference model crusts (A1 and A3) and model crusts with a history of high a_w (A2) as measured with a needle by a texture analyzer. On the left a comparison at equal a_w on the right a comparison at equal water content.

5.3.3 Texture Analyser

In order to evaluate the effect of water content and water activity on the mechanical properties and acoustic emission during breaking of the model crusts, a puncture test was performed. Several pieces of model bread crust were punctured and the sound and force signal was recorded. The results of the force-time signal during puncturing of a model crust with a needle are shown in figure 5a for the set of samples stored at 40% and 57% RH and in figure 5b for the set of samples stored at 50% and 63% RH. As can be seen, the three different samples all show different behaviour, especially the samples stored at 57 and 63% RH show more bending then breaking behaviour. To evaluate the results in more detail the linear distance and the number of force and sound peaks were counted for all samples and the results are displayed for the series with samples A in figure 6a, b and c, respectively. For the series with the samples B the trend was the same (results not shown). The linear distance is a measure to evaluate the jaggedness of the curve. It is defined as the length of an imaginary line joining all points on the graph between in this case the point at which the force curve starts to increase (when the needle hits the sample, generally after around 15 seconds) until 38 seconds which is about the end of the experiment. A highly jagged line will have a longer linear distance and is expected to correspond to a more crispy product than a smooth line (*23*). Figure 6a showed no significant difference between the linear distances of the samples, but the average linear distance was shorter for the samples A3 and B3 which had a high water content and a high a_w. However, the results for the number of sound and force peaks did show significant differences. The samples stored at 40 and 50% RH (A1 and B1) showed the most force and sound events whereas the sample stored at 57 and 63% RH (A3 and B3) showed the lowest number of force and sound events. For the latter samples the fracture was actually more bending rather than breaking as can be seen by the rounding (lack of sharp peaks) in the force versus time curve for these samples (figure 5). The behaviour of samples stored at 90% RH and later dried back to 40 or 50% RH (A2 and B2) was intermediate.



(a)



(b)

Figure 7. Contribution of water content and a_w to crispness perception as measured in a "two alternatives first choice test". Average number of panellist finding the specified Soissons model crust sample the crispier one of the two for series A (a) and B (b). Comparison for equal a_w on the left and for equal water content on the right.

5.3.4 Sensorial test

The two sets of samples were tested by a panel with respect to their difference in sensorial crispness. During each test two samples of Soissons model crust were compared. In the first set we compared samples with equal a_w but different water contents. For this purpose one sample was stored at 90% RH and dried back to a certain a_w (A2 or B2) and the other sample was brought immediately at this set a_w from a dry state (a_w ~0.2) (A1 or B1). In a second test we compared samples with equal water content but different a_w. For this purpose the samples stored at 90% RH and dried back to 40 or 50% RH (A2 or B2) were compared with samples that were brought at a similar water content but higher a_w from a dry state (A3 or B3).

The results of the sensory test are presented in figure 7. Subjects selected samples with low water content as more crispy for samples with equal a_w (n=34, x (number of subjects choosing higher water content as less crispy) = 32 and 31, $p \leq 0.00000033$, for A1 and B1 compared to A2 and B2, respectively). Subjects selected samples with low a_w as more crispy for samples with equal water content (n=34, x=25 and 27 $p \leq 0.0018$, for the samples A2 and B2 compared to A3 and B3, respectively). The first results indicate that the water content is determining crispness. On the other hand the second result suggests that crispness depends on the water activity. These results followed the same trend as the results from the puncture test in the sense that both water content and water activity seems to play a role in determining crispness.

5.4 Discussion

A dry crust looses its crispness when water migrates into the crust. It is not clear if it is the amount of water absorbed or the water activity that leads to loss of crispness. The hysteresis effect observed when recording a water sorption isotherm allowed us to study effects of a_w and moisture content separately. Information on the mobility of water and the macromolecules was obtained, as well as information on the sensorial and textural properties of the model crusts. The results show that the sensorial crispness and instrumentally determined fracture behaviour may depend on both the water content and the water activity, or, in other words on the history of the sample. According to the NMR results the water in the samples with a history of high a_w is still in a 'more free' state than the sample with the same a_w, but a lower water content. The water in the samples with a history of high a_w is probably more inhomogeneously distributed than in the samples with equal water content but higher a_w. This also means that the regions where cracks can be stopped are more inhomogeneously distributed. As a consequence regions with different crispness might be present in the sample resulting in an apparently overall more crispy product.

For starch type products the hysteresis effect is often explained as the creation of more water binding places due to the history of the sample that has been at RH 90% and dried back. This would explain the lower measured water activity. If there are more binding places for water the water will on average be more tightly bound resulting in a lower water activity. The fact that a history of high a_w has no effect on the mobility of the water protons at equal water content suggest that hysteresis in bread crust may not be caused by an increased number of binding places for water in the sample. Possibly the a_w is, as well as the water, not homogeneously distributed in the sample with a history of high a_w. More research has to be done before a more definitive conclusion can be drawn.

Labat (*24*) investigated the effect of starch crystallinity on the number of force peaks as measured with a puncture test. She concluded that the retrograded samples were characterised by a higher number of peaks than the reference samples. The former are therefore likely to be more crispy. In bread crust only part of the starch is gelatinized and therefore only part of the starch can retrogradate (*25*). DSC results (table 3) show that only the samples that were stored at higher RH (57 and 63%) showed a significantly higher amount of retrograded starch (expressed as ΔH_r) than the other samples. However these samples were rated as the least crispy. Therefore we do not expect that retrogradation of the starch has had a clear effect on the outcome of our experiments. Why there is no difference in retrogradation between the samples with a history of one day storage at 90% and the samples stored at 40 and 50% RH from a dry state is not clear.

The different water activities of samples with equal water content had no influence on the glass transition temperature as measured with the PTA. This may be expected because the transition temperatures are quite elevated and some transfer of the water may already have taken place due to the high pressure put on the sample and the increase in temperature.

5.5 Conclusions

Results from sensory and texture analyses in which the effect of water content and water activity on the crispness of model crusts is evaluated show that the history of the sample plays a role in sensory perception of crispness. The glass/rubbery transition temperatures as measured by PTA and the mobility of the protons as measured by NMR are directly related to the water content. Samples with a different a_w but equal water content give equal results. Possibly an inhomogeneous distribution of water and consequently crispy and less-crispy regions exist in the samples with a history of high a_w thus making them overall more crispy than samples with the same water content but higher a_w.

Acknowledgement

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References

1. Roudaut, G.; Dacremont, C.; le Meste, M., Influence of water on the crispness of cereal-based foods: acoustic, mechanical, and sensory studies. *Journal of Texture Studies* **1998,** 29, (2), 199-213.
2. Primo-Martín, C.; Castro-Prada, E. M.; de Wijk, R.; Vereijken, P. F. G.; Meinders, M. B. J.; van Vliet, T., Effect of structure on the sensory characterization of the crispness of toasted rusk roll, **submitted**.
3. Valles Pamies, B.; Roudaut, G.; Dacremont, C.; le Meste, M.; Mitchell, J. R., Understanding the texture of low moisture cereal products: mechanical and sensory measurements of crispness. *Journal of the Science of Food and Agriculture* **2000,** 80, (11), 1679-1685.
4. Katz, E. E.; Labuza, T. P., Effect of water activity on the sensory crispness and mechanical deformation of snack food products. *Journal of Food Science* **1981,** 46, 403-409.
5. Sauvageot, F.; Blond, G., Effect of water activity on crispness of breakfast cereals. *Journal of Texture Studies* **1991,** 22, 423-442.

6. Slade, L.; Levine, H., A food polymer science approach to structure-property relationships in aqueous food systems: non-equilibrium behavior of carbohydrate-water systems. *In Water Relations in Foods (L. Slade and H. Levine, eds.) pp. 29–101, Plenum press, New York.* **1991**.

7. Huang, V. T.; Haynes, L.; Levine, H.; Slade, L., Glass transitions in starch, gluten and bread as measured by dielectric spectroscopy and TMA methods. *Journal of Thermal Analysis* **1996,** 47, 1289-1298.

8. Meste, M. le; Roudaut, G.; Davidou, S., Thermomechanical properties of glassy cereal foods. *Journal of Thermal Analysis* **1996,** 47, 1361-1375.

9. Nikolaidis, A.; Labuza, T. P., Glass transition state diagram of a baked cracker and its relationship to gluten. *Journal of Food Science* **1996,** 61, (4), 803-806.

10. Wolf, M.; Walker, J. E.; Kapsalis, J. G., Water vapor sorption hysteresis in dehydrated foods. *Journal of Agricultural and Food Chemistry* **1972,** 20, (5), 1073-1077.

11. McMinn, W. A. M.; Magee, T. R. A., Studies on the effect of temperature on the moisture sorption characteristics of potatoes. *Journal of Food Engineering* **1999,** 22, 113-128.

12. Al Muhtaseb, A. H.; McMinn, W. A. M.; Magee, T. R. A., Moisture sorption isotherm characteristics of food products: a review. *Food and Bioproducts Processing* **2002,** 80, 118-128.

13. van den Berg, C.; Kaper, F. S.; Weldring, J. A. G.; Wolters, I., Water binding by potato starch. *Journal of Food Technology* **1975,** 10, 589-602.

14. Bengoechea, C., Arrachid, A., Guerrero, A., Hill, S.E. and Mitchell, J.R., Relationship between the glass transition temperature and the melt flow behavior for gluten, casein and soya. *Journal of Cereal Science* **2007,** 45, (3), 275-284.

15. Sidhu, J. S.; Caceres, P. G.; Behbehani, M., Measurement of starch properties during staling of Arabic bread. *Starch starke* **1997,** 49, (5), 180-186.

16. Matsunaga, K.; Kawasaki, S.; Takeda, Y., Influence of physicochemical properties of starch on crispness of tempura fried batter. *Cereal Chemistry* **2003,** 80, (3), 339-345.

17. McBrierty, V. J.; Packer, K. J., *In: Nuclear magnetic resonance in solid polymers,*
Cambridge University Press, Cambridge, UK **1993**.

18. van den Dries, I. J.; van Dusschoten, D.; Hemminga, M. A., Mobility in maltose-water glasses studied with H-1 NMR. *Journal of Physical Chemistry B* **1998,** 102, (51), 10483-10489.

19. Strahm, B.; Plattner, B.; Huber, G.; Rokey, G., Application of food polymer sience and capillary rheometry in evaluating complex extruded products. *Cereal Foods World* **2000,** 45, (7), 300-302.

20. Plattner, B.; Strahm, B.; Rausch, K., The phase transition analyzer and its impact on extrusion processing of foodstuffs. *ASAE Annual International Meeting, Sacramento, California, USA: ASAE, St. Joseph, MI;* **2001,** ASAE Meeting Paper Number 01-6067.

21. Meinders, M. B. J.; Castro Prada, E. M.; Vliet, T. van, Quantification of single events during fracture of cellular solid foods. **To be submitted**.

22. Choi, S. G.; Kerr, W. L., Effects of chemical modification of wheat starch on molecular mobility as studied by pulsed 1H NMR. *Lebensmittel Wissenschaft und Technologie* **2003,** 36, (1), 105-112.

23. Nalesnik, C. A.; Onwulata, C. I.; Tunick, M. H.; Phillips, J. G.; Tomasula, P. M., The effects of drying on the properties of extruded whey protein concentrates and isolates. *Journal of Food Engineering* **2007,** 80, 688-694.

24. Labat, E., Effect of starch crystallinity on the mechanical properties of dry and hydrated baked cereal foams. *Poster 359 at the AACC Annual Meeting, September 17-20 2006, San Francisco, USA.* **2006**.

25. Primo Martín, C.; van Nieuwenhuijzen, N. H.; Hamer, R. J.; van Vliet, T., Crystalline changes in wheat starch during the bread-making process: starch crystallinity in the bread crust. *Journal of cereal science* **2007,** 45, 219-226.

Chapter 6 Effect of ingredient variation and modification on water uptake kinetics in crispy bread crusts

Abstract

The aim of this study was to evaluate the effect of ingredients, crust modifications by enzymes and porosity on water uptake and water uptake kinetics in crispy bread crust. Bread crusts of different formulations were tested regarding their water uptake kinetics. The addition of xylanase and amylase to the dough formulation did not show an effect on water uptake kinetics in the bread crust. Neither did the addition of arabinoxylans to the model crust. The addition of lecithin or a lipase to the dough or spraying of a protease on the dough surface on the other hand did influence the water uptake kinetics. These modifications all act on the morphology of the bread crust and therefore it was concluded that morphology is an important factor in water uptake kinetics of bread crusts. The water uptake rates of a model crust (with a more dense structure) were also compared with those of a crispy roll crust. The water uptake rates of the model crust were found to be slower which points at a morphology effect as well.

N.H. van Nieuwenhuijzen, C. Primo-Martín, T. van Vliet, R.J. Hamer, to be published.

6.1 Introduction

Maintaining the sensory characteristics of crispy and crunchy foods during shelf life can be difficult. When a crispy product is stored in a moist environment or when the crispy product is part of a composite food that includes a moist part, moisture migration will take place from the environment or the moist part of the product to the dry and crispy part. This usually results in loss of sensorial crispness of the product (*1*-*3*). Since the amount of water present in a crispy product is an important factor for crispness, the rate of loss of crispness will be related to the kinetics of water uptake in the crispy parts of the product. Water uptake rates can be influenced by structure like for example porosity, beam thickness and beam structure of the crust as well as by its composition (e.g. interaction of water with the food constituents). The effect of addition of ingredients was for example tested by adding fat to sponge cake. This fat decreased the water migration kinetics, likely due to hindering of the migration of water into the solid matrix (*4*). The effect of porosity on moisture transfer kinetics have been studied for dough (*5*), bread crumb (*6*) and for sponge cake (*4, 7, 8*). In general researchers found a higher effective diffusion coefficient with increasing porosity. This was related to a change in the water transport mechanism (*4*). In low porosity foods, diffusion of the liquid water is considered to be the main process of water transport whereas in high porosity foods water vapor transport through the open pores will also determine the water uptake kinetics (*3*). Primo-Martín et al (*9*) showed that the coarseness of a food structure (here a toasted rusk roll) can have an effect on the crispy sensation in a steady state with respect to water content and water activity. A more coarse structure gave a more crispy sensation. However for the rate of loss of sensorial crispness in a composite food the water uptake kinetics are highly important. No studies were found on the effect of porosity on the water uptake kinetics of dry crispy food products like bread crust.

The hydration of wheat flour and its isolated components have been studied extensively (*10*). Roman-Gutierrez et al (*10*) measured the diffusion coefficient of a soft and a hard wheat flour and its separate components starch, gluten and pentosans by analysing water uptake data at different RH's and measuring porosity and particle size of all samples with a mercury porosimeter and laser granulometry, respectively. They found significantly higher apparent diffusion coefficients for the water-insoluble pentosans (1.5×10^{-13} m^2/s) at 25°C and 60% RH, than all other components (starch, gluten, water-soluble pentosans and wheat flour). For these components the apparent diffusion coefficients were between 0.5×10^{-15} and 5×10^{-15} m^2/s under the same conditions. Following the same line of reasoning the kinetics of water uptake by a crispy bread crust will also be related to the composition of the bread crust. The aim of this study is to evaluate the effect of ingredient addition, modification of flour components by enzymes and porosity on water uptake and water uptake kinetics in crispy bread crust.

6.2 Materials and Methods

6.2.1 Material

All breads were prepared with Soissons wheat flour. Flour was purchased from Meneba (Meneba Meel BV, Rotterdam, the Netherlands). The composition of the flours is shown in table 1, chapter 3 of this thesis. Native wheat starch (Excelsior) was purchased from Avebe (Veendam, the Netherlands).

6.2.2 Methods

6.2.2.1 Preparation of gelatinized starch

Native wheat starch was dispersed in excess water and boiled at 90°C for 10 minutes. Next the starch was freeze dried and milled into powder with an analytical grinder (type A10, IKA labortechnik, Staufen, Germany).

6.2.2.2 Preparation of gluten as used for the isotherm measurements

Dough was prepared by mixing in a farinograph mixer (Brabender, Duisburg, Germany) using a 50 g mixing bowl. The formulation included flour, NaCl (2 g/100g flour) and ascorbic acid (20 ppm). 55.5% water on flour basis was used to prepare the dough. The temperature of the added water was 22°C and the starting temperature of the mixing bowl and flour was 22.5°C. The dough was mixed at a speed of 100 rpm until the dough temperature reached 26°C, which corresponded to the maximum in the torque-time curve. The dough was left to rest for 1 hour at 30°C in a closed box. Next the dough was washed with tap water and later with 2% NaCl solution until the washing water was clear and all the starch was removed. The gluten were frozen at -80°C and freeze dried. The samples were milled and sieved with a 63μm sieve and the fraction <63μm was used for the measurement. The sample was stored over P_2O_5 at 22°C until use for minimally 3 days.

6.2.2.3 Model crust preparation

Model crust which is a baked thin layer of dough was prepared as explained in chapter 3 with one exception that instead of 20 ppm ascorbic acid, accidentally 200 ppm ascorbic acid was used. Model breads (crispy rolls), which are small white breads, were prepared according to Primo-Martín et al (*11*). Three enzymes were used according to Primo-Martín et al (*11*): Lipopan F BG, a lipase which acts on polar lipids (phospholipids and galactosyl glycerides) and on triglycerides, provided by Novozymes (Denmark), Bakezyme P 500 BG, a fungal *alpha*-amylase from *Aspergillus oryzae* and Bakezyme BXP 5001 a bacterial xylanase provided by DSM Bakery Ingredients (Heerlen, The Netherlands). These enzymes were mixed into the dough. A protease (Profix 200L, an endo-protease, Papain, EC 3.4.22.2 provided by Quest Int, Ireland) was used as well, but this enzyme was sprayed on top of the bread crust (1g/30 ml) according the method of Primo-Martín et al. (*12*). For the breads with lecithin a concentration of 1% on flour basis was used. Model bread crusts were also prepared with additional water extractable (medium viscosity WEAX) and water insoluble arabinoxylans (WIAX. Both arabinoxylans were from wheat flour (Megazyme International Ireland Ltd., Bray, Ireland) and added in a concentration of 3% by substitution of flour and keeping the water content constant.

The crust from a crispy roll was isolated by scraping off around 1 mm of the crust with a sharp knife if necessary the white crumb was scraped off. This first 1 mm was the most dense and dry part of the crust. Model crusts were used completely. Crust samples were milled in an analytical grinder (type A10, IKA Labortechnik, Staufen, Germany) for 10 seconds at a time with a maximum of 3 times (to prevent heating of the sample) until a fine powder was obtained. The powder was sieved with different sieves (63µm, 0.25 mm, 0.5 mm and 0.71 mm). The different sieving fractions (<63µm, 63µm-0.25mm, 0.25 mm-0.5 mm and 0.5-0.71 mm, called S, M, L and XL, respectively) were freeze dried. The largest sieve fraction (>0.71 mm) was discarded because these sizes were undefined. In an earlier study (*13*) no significant differences in starch and gluten content were detected between the sieve fractions S, M and L therefore we do not expect differences here either. Samples were stored at room temperature over P_2O_5 until use (with a minimum of 3 days).

6.2.2.4 Confocal scanning laser microscopy

Two fluorescent probes (fluorescein 5-isothiocyanate (FITC, 0.85% in water) and Rhodamine B (0.15% in water) were used to stain the bread samples (crumb and crust). Fluorescent probes were added as aqueous solutions (1.67 ml of each solution per 10 g of flour) to the mixing water during bread making. At the concentrations used, FITC stains starch (green), while Rhodamine B reacts more specifically with protein (red). The samples were studied using a Leica TCS SP (Leica Microsystems, Heidelberg, Germany) CSLM with an Ar/Kr laser. The excitation wavelengths used were 488 and 568nm for FITC and Rhodamine B, respectively, and the emission maxima were at 518 and 625 nm. Images from the separated channels were overlain to allow a simultaneous imaging of starch and protein.

6.2.2.5 Sorption experiments

Sorption experiments were performed using a VTI-SGA 100 symmetric vapour sorption analyzer (VTI Corporation, Hialeah, Florida USA). This is a continuous vapour flow sorption instrument for obtaining water and organic vapour adsorption isotherms. The instrument is equipped with a dew point analyser, and a Cahn microbalance. 5-7 mg was weighed in the measuring cup which gave an approximate thickness of the powder bed in the sample cup of 1 mm. Also a SPS11-10µ (Project Messtechnik, Ulm, Germany) moisture sorption analyser was used in order to speed up the amount of experiments that could be done. For this machine approximately 200 mg was weighed into the weighing cups and 11 samples could be measured at the same time. The sample was spread out on the bottom of the cup forming a single layer. All tests on crust samples were performed at 25°C. Every experiment was started with a drying step at 50°C during 120 min. The isotherm was determined by changing the RH every 800 minutes or when the isotherm was stable (less than 0.0001% weight change in 10 minutes). The weight change of the sample (caused by the change in environmental RH) was measured in time.

6.2.2.6 *Gelatinized starch content as measured by DSC*

Around 8 mg of crust sample or flour were weighed in stainless steel pans. Water was added at a ratio of 1:4. DSC measurements were performed with a Perkin Elmer Diamond DSC calorimeter (Perkin Elmer Corp., USA). Indium and gallium were used to calibrate the system. The samples were heated from 20 to 100°C at 10°C/ min. An empty stainless steel pan was used as a reference during the DSC measurement. The enthalpy was expressed in J/g of sample (db). Relative crystallinity (D_{RC}) was calculated as:

$$D_{RC} = \frac{\Delta H_s}{\Delta H_f} \cdot 100 \tag{1.1}$$

where ΔH_s represents the melting enthalpy (J/g) of starch crystallites in the sample (peak 70°C) that did not gelatinize during baking and ΔH_f represents the enthalpy (J/g) of starch gelatinization in the flour.

6.2.2.7 *Porosity measurements*

Porosity, ε (%), is a measure of the void spaces in a material. It is the portion of the non-solid volume of the total volume of a material, and is defined by the formula:

$$\varepsilon = \frac{V_t - V_s}{V_t} * 100(\%) \tag{1.2}$$

Where, V_t = total volume (cm^3) and V_s = volume of the solid (cm^3). The volume of the solid material (V_s) was determined by using an Ultrapycnometer 1000 (Quantachrome cooperation, Ankersmid, Oosterhout, The Netherlands), by measuring the difference in pressure between the measuring cell (containing the sample) and the reference cell when blowing nitrogen gas through.

The total volume of the sample (solid material plus air inside the sample) was determined by under oil weighing. For this purpose, a balance was used (±0.01 g), capable of weighing by a hook underneath. For that, a wire mesh basket with lid (dimensions 3x7x7 cm, 8 mm openings between wires) which included the sample was connected to the hook. A pan containing arachis oil (room temperature) was placed underneath the balance. The density of the oil (ρ_{oil} (g/cm^3)) was determined using an aerometer. The density of the sample is calculated using the following formula:

$$\rho_{sample} = \frac{m_{air}}{m_{air} - m_{oil}} x \rho_{oil} \tag{1.3}$$

In which, m_{air} is the weight of the sample in the air and m_{oil} is the weight of the sample measured in the oil. V_t is calculated by using:

$$V_t = \frac{m_{air}}{\rho_{sample}} \tag{1.4}$$

6.2.2.8 Calculations

The kinetics of water sorption were studied at 25°C at different relative humidities (RH). A single exponential function

$$M(t) = [M(0) - M(\infty)]e^{-kt} + M(\infty) \tag{1.5}$$

was found to fit to the water uptake data best (chapter 3). M(t) is the mass of the sample at time t, M(0) is the initial sample weight and M(∞) is the fitted sample weight at equilibrium. k is the water uptake rate parameter (min^{-1}). Both M(∞) and k are adjustable parameters.

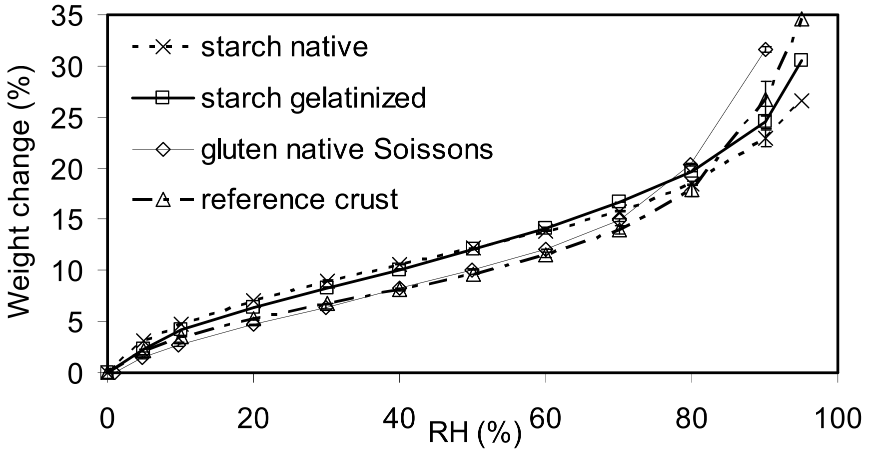


Figure 1 Weight change as a function of storage RH of the separate components of wheat flour; starch (gelatinized and ungelatinized) and native gluten and of the baked crust. All particles are smaller than 63 µm.

6.3 Results and discussion

6.3.1 VTI vs SPS11

In order to speed up the experiments 2 different water sorption apparatus were used; VTI-SGA 100 symmetric vapour sorption analyzer (VTI Corporation, Hialeah, Florida USA) and a SPS11-10µ (Project Messtechnik, Ulm, Germany) moisture sorption analyser. The latter allows the measurement of 11 samples at the same time whereas the first one only can measure one sample at a time. The VTI machine may be more sensitive than the SPS11 due to a more sensitive balance (the VTI measures 4 decimals in mg versus 2 decimals in mg for the SPS11; a factor 100 difference). On the other hand only very small amounts of sample were used (6-7 mg) whereas with the SPS11 samples of around 200 mg were used (a factor 30 more). This makes measurements with the latter instrument less sensitive for inhomogeneities in the sample. Standard deviations for the difference between duplo measurements on the water uptake kinetics of reference bread crusts vary from 3 to 20% of the k value for the SPS11 and 3 to 40% for the VTI for the larger particle sizes. With the SPS11 the sample was spread on the bottom of a round cup (approximately 5.5 cm in diameter) with a bed thickness of a single layer of particles whereas in the VTI (sample cup with a maximum diameter of 1 cm) the sample forms a bed of particles (with a thickness of around 1 mm). The time that the air around the sample needs to equilibrate was 4.5 and 12 minutes for the VTI and SPS11, respectively.

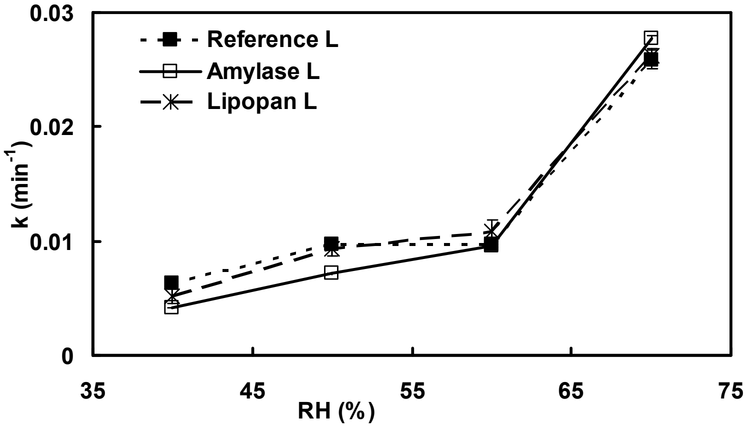


Figure 2a. Effect of amylase and lipase treatment of the crispy roll dough on the water uptake kinetics of the baked crispy roll crust as measured with the SPS11. Results for particle size L (0.25-0.5 mm) as a function of RH.

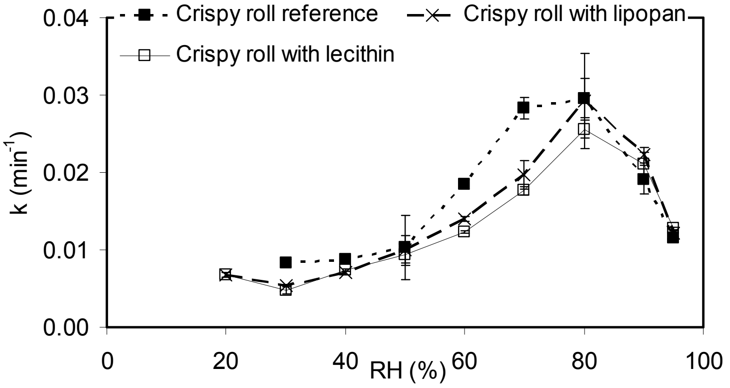


Figure 2b. Effect of lipase treatment and the addition of lecithin of a crispy roll dough on the water uptake kinetics of a baked crispy roll crust as measured with the VTI. Results for particle size L, 0.25-0.5 mm as a function of RH.

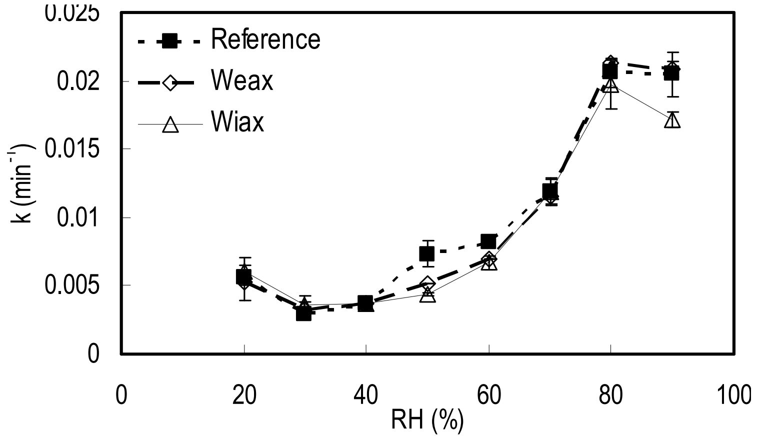


Figure 2c Effect of the addition of water extractable arabinoxylans (Weax) and water insoluble arabinoxylans (Wiax) on the water uptake kinetics of model crust particles (size 0.5-0.71 mm) as a function of RH and as measured with the VTI. Weax and Wiax indicate the substitution of flour with 3% Weax or Wiax, respectively, the water content of the dough was kept the same.

6.3.2 Equilibrium isotherms of the separate crust components

Isotherms of the separate components of the flour were measured and the results are shown in figure 1. Both native and gelatinized starch pick up more water than gluten and bread crust in an isotherm experiment up to 70% RH. Although bread crust consists of 70% starch, the sorption isotherm of the bread crusts is apparently influenced by the isotherm of gluten. Also an interaction between the crust components may affect sorption behavior of the crust.

6.3.3 Effect of ingredients on water uptake kinetics

The kinetics of water uptake of the separate components will be very important for the rate at which crispness of bread crust is lost. However, we will refrain from analyzing the water uptake kinetics from the separate components here because the difference in diffusion distance (size) per sample makes the comparison of the data very difficult. Therefore we concentrated on the modification of whole crust and measured the water uptake kinetics of particles of these crusts. In order to study the effect of variations in the properties of ingredients on the kinetics of water uptake, bread dough or bread crust was modified by adding enzymes. A rate parameter k was calculated by fitting Eqn. 1.3 to the experimental data. The quality of fit, expressed by R^2 was minimally 0.97. Figure 2a shows the effect of a lipase treatment and amylase treatment on the water uptake kinetics (k-value) of the bread crusts as measured with the SPS11. A higher k-value means faster water uptake. Amylase breaks down the damaged starch in the dough into smaller sugar molecules during fermentation and the gelatinized starch during baking. This results in more food for the yeast (higher dough volume), more substrate for the Maillard reaction and a lower water binding capacity of the smaller molecules compared to the original starch. The bread crust was indeed more dark compared to the reference crust after baking. The lipase used in this study is specific for polar lipids. Lipopan F BG hydrolyzes the endogenous wheat polar lipids (phospholipids and galactosyl glycerides) into the corresponding more polar monoacyl lipids. These polar lipids are surface active and will have a positive effect on the stability of the gas cells in the dough and with that on the porosity of the crust (*14*).

Figure 2a does not show an influence of the added enzymes on the water uptake kinetics of the bread crusts. However, when measuring the same samples with the VTI, a significantly lower k-value was observed at 60 and 70% RH for the lipase treatment compared to the reference bread (figure 2b). A bread crust treated with amylase was not tested in the VTI. The addition of lecithin was also tested in an experiment with the VTI. Also lecithin gave slower water uptake kinetics at 60 and 70% RH (figure 2b). This is not unexpected since the reaction products of the lipase treatment are expected to have a similar emulsifying effect as the phospholipids from lecithin. Primo-Martin et al (*11*) measured the porosity of the bread crusts with added lipase. For lipase the porosity was 57% (+/- 2%), for lecithin the porosity was also measured and found to be 54% (+/- 1) versus 49% (+/- 3%) for the control. Both treatments thus increased the porosity of the crust. Image analyses of the outer 2.5 mm of the lipase treated crust, performed according to the method as described by Tromp et al (*15*), also showed that the lipase crust was more coarse and had a more irregular structure compared to the reference bread crust (results not shown). However, an increase in porosity as measured by Roca et al (*4*) was expected to give an increase in speed of water uptake. It could be that more fats moved to the surface as a result of the lipase treatment than in an untreated bread and that these fats possibly deteriorated the “wetting” properties of the surface. The k-values differ a factor 1.3 at 60% RH which could improve the time that the crispness remains with a factor 1.3 as well, this could be an extra 35 minutes crispness based on an average shelf life of a crust of 2 hours (*12*).

At a RH above 80% (around 14% water) likely some swelling and caking of the particles is taking place. This will result in a decrease in surface area of the particle bed and with that in slower water uptake kinetics. A similar trend with a peak in water uptake rates between 9 and 13% water content was found by Roca et al (*8*) for sponge cake.

Also the effect of a xylanase, and the effect of the addition of arabinoxylans (both soluble and insoluble) was tested on model bread crusts. Xylanase hydrolyses water insoluble arabinoxylans. This will decrease the molecular weight of the arabinoxylans and increase their solubility. No effect on water uptake kinetics was found for a xylanase treated model crust as measured by the SPS11 (results not shown). Xylanase treatment of a bread crust did not have an effect on crispness nor on the porosity of the crust as measured by under oil weighing (*11*). Beforehand the breakdown of arabinoxylans was expected to have an effect on the water uptake kinetics due to the decreased ability of the arabinoxylans to absorb water. The addition of water soluble arabinoxylans and water insoluble arabinoxylans to model crusts also did not show an effect on water uptake kinetics as measured by the VTI (figure 2c). Neither was there an effect on the apparent equilibrium values of the isotherm experiment. The fact that there is no effect of xylanase or of the addition of arabinoxylans on the rate of water uptake may suggest that only fatty ingredients can influence the water uptake kinetics.

6.3.4 Effect of porosity on water uptake kinetics

Figure 3a shows the difference between the water uptake kinetics of a model and a crispy roll crust of Soissons flour. At a RH above 50% the kinetics of water uptake from the model crust were slower than the kinetics of water uptake of the crispy bread crust. This was measured with the SPS11. With the VTI the same materials were measured but the particle size was smaller (0.25-0.5 mm instead of 0.5-0.71 mm). However, the differences in the rate parameter k were more clear as can be seen in figure 3b. To explain these data the porosity of the materials was determined. A difference in porosity between model crusts and rusk roll crusts was found, the porosity of a Soissons model crust was around 28 +/- 5%% and the porosity of a crispy roll crust was 49 +/-3% (*11*).This implies that the model crust is much more dense than the real bread crust. Figure 3c shows a picture of a model crust and a bread crust. The air bubbles in a model bread crust seem much bigger and possibly the lamella are thicker, but this is very difficult to judge by the eye. So far no objective method for measuring lamella thickness was established, but X-ray tomography may be a solution for this problem (*16, 17*).

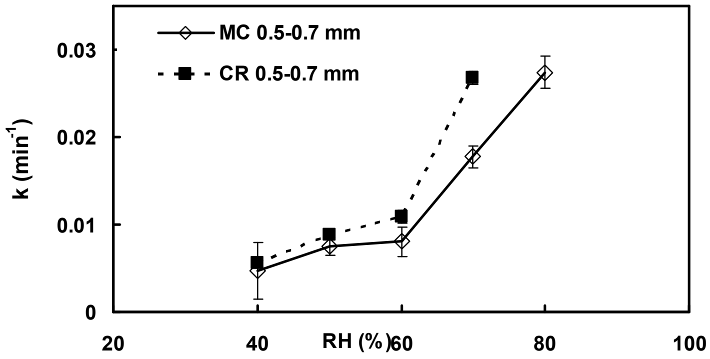


Figure 3a. Comparison of the water uptake kinetics of model crust (MC) with crispy roll crust (CR) particles as a function of RH for a crust particle size of 0.5-0.7 mm as measured with the SPS11.

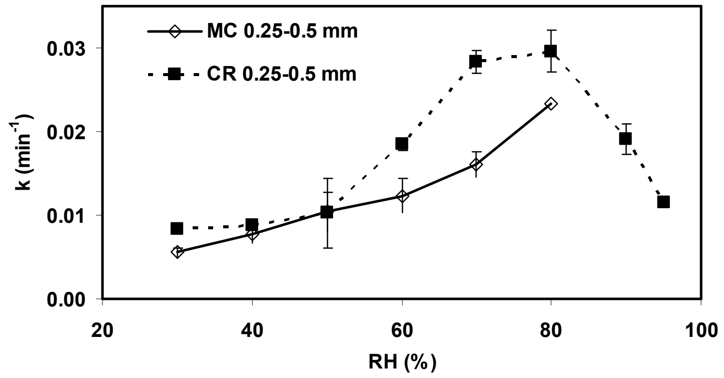


Figure 3b Comparison of the water uptake kinetics as a function of RH of model crust (MC) with crispy roll crust (CR) for a crust particle size of 0.25-0.5 mm as measured with the VTI.

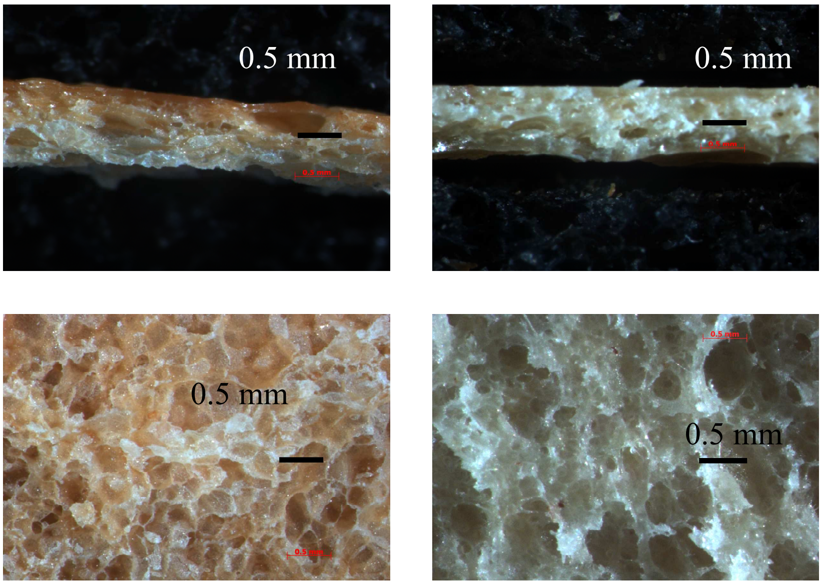


Figure 3c. Light microscopy pictures of a crispy roll bread crust (left) and a model bread crust (right) from Primo-Martín et al. (*18*). On top a cross section, on the bottom the underside of the crust.

There is a small difference in degree of relative cristallinity as determined by DSC (14 % +/-3 for the model crust and 28% +/-9 for the crispy roll crust for the Soissons flour , but this is likely not enough to be the reason for the difference in water uptake kinetics. Cryomilled samples, in which porosity effects are removed, did not show a difference between water uptake kinetics of model and crispy roll bread crust (results not shown). However, it should be noted that for these samples the fits with equation 1.3 were not very good (R^2 of 0.94-0.96).
The water uptake rates are 0.008 or 0.011 min^{-1} for the step from 50 to 60% RH for the model crust and the crispy roll crust, respectively. This corresponds to a decrease in the water uptake rate parameter by a factor 1.3 which means that it will take 1.3 times as long for the sample to reach the critical water content. Following from this instead of 90 minutes (1/k) 125 minutes are needed before half of the water that can go in will go in. With the same information one could argue that probably the beams in a model crust are 1.2 times as thick as in a crispy roll crust (following roughly from Fick's law). A structure with thicker beams may result in a slower water uptake rate and could therefore be a tool for enlarging the time that a crispy crust remains crispy.

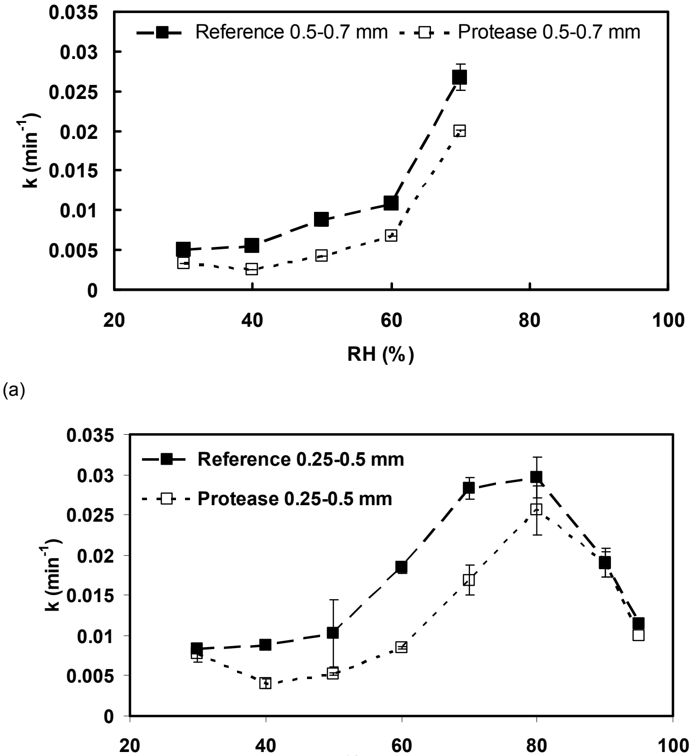


Figure 4 Effect of protease spraying on the bread dough surface before baking on the water uptake kinetics of a Soissons crispy roll crust. Results for a particle size of 0.5-0.71 mm as measured with the SPS 11 (a) and for a particle size of 0.25-0.5 mm as measured with the VTI (b).

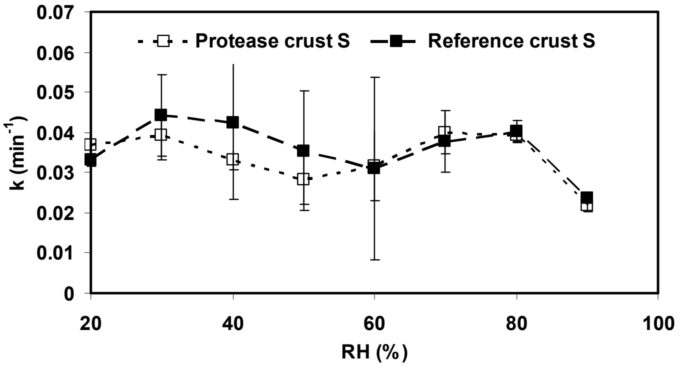
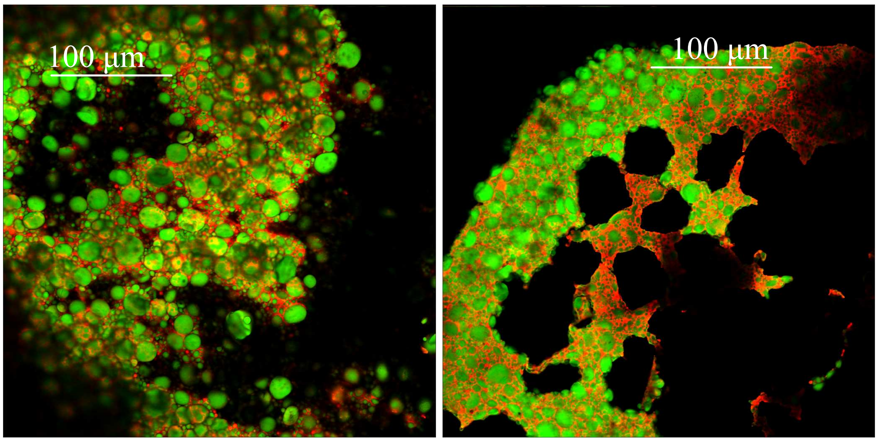


Figure 5. Effect of cryomilling (ruling out porosity effects) on the water uptake kinetics of a protease treated bread crust compared with a reference bread crust. Results as a function of RH as measured with the VTI.

6.3.5 Effect of protease on the water uptake kinetics in a Soissons crispy roll crust

Figure 4 and 5 show the effect of protease spraying on the bread dough surface on the water uptake kinetics of the bread crusts. The samples with a larger sample size (0.5-0.7 mm) studied by the SPS11 showed an obvious difference in water uptake kinetics (figure 4a). The protease treated crust showed significantly slower water uptake kinetics than the reference crust. This may be due to a difference in porosity, beam thickness or a difference in the solid material properties between the protease treated crust and the reference crust (e.g. in the degree of air inclusion in the solid beams). Also for the large particle size (0.25-0.5 mm), studied by the VTI the protease crust is slower regarding water uptake (figure 4b) than the reference crust. In order to differentiate between the effect of morphology and the effect of chemical composition on the water uptake kinetics also bread crusts particles with a smaller particle size (<63 um) were measured to rule out morphological effects. When using this small size we expect that the kinetics are only influenced by the chemical composition of the particles. The results are shown in figure 5. No significant difference was found between the protease treated crust and the reference crust. However, it should be noted that with a smaller particle size the differences in water uptake kinetics between the duplicate measurements were much larger resulting in bigger standard deviations. This might be due to the fact that the packing of the particles in the measuring cup starts to become more important resulting in more deviations in the measurements. This makes it difficult to conclude whether or not an effect of the chemical composition is involved in the water uptake kinetics. No difference was found in the steady state values between the two sizes (results not shown).

Figure 6a and b show a CSLM picture of a rusk roll bread crust and a rusk roll bread crust which has been treated with protease. The pictures show the crust surface from above They show that the protease treatment resulted in holes in the crust of 50-100 µm with very sharp edges. Likely the protease has locally broken down the gluten network and with that the protease has changed the morphology of the crust. However, porosity measurements did not show a significant difference in porosity between protease treated bread crusts and a reference bread crust (porosity of 46 +/- 2% and 47 +/- 2%, respectively). Primo-Martín et al found a positive effect of protease spraying on the initial crispness of bread crust and also a positive effect of protease spraying on the retention of crust crispness. The better crispness was found to be directly related to a lower water content and water activity directly after baking (*12*). Two possible explanations can be given for the mechanism that lowers the initial water content of the crust after baking and maintains the lower water content for a longer time in these protease treated crusts. First, the holes in the crust could make it more easy for the water to evaporate from the crust during baking, resulting in a lower initial water content in the crust and the outer crumb when the breads leave the oven. A lower water content in the crust of a protease treated bread was indeed found (*12*). Secondly the density of the solid matrix could have increased because the breakdown of the gluten matrix by the protease will reduce the gas holding capacity of the dough before and during baking, which could have resulted in less gas inclusion in the breams of the protease treated crust. This would make the beams more dense and likely slow down water uptake rates in the crust while storing the bread. In chapter 3 we concluded that the speed of water uptake was determined by the relaxation of the matrix. It can also be that due to a difference in the chemical composition these relaxation rates changed. In chapter 4 was concluded that it is the gluten matrix that likely gives the crispness to the product, therefore slower water uptake rates in the gluten matrix could help improving the time a bread crust is crispy.



(a) (b)

Figure 6 CSLM images from the bread crust surface stained for starch (green) and protein (red) for a reference bread crust (a) and a bread crust sprayed with protease (b)

6.4 Conclusions

No effect of xylanase and amylase treatment on water uptake kinetics was found. Neither for the addition of arabinoxylans to the bread crust. On the other hand a difference in water uptake kinetics was found between model crust and a rusk roll crust and also between a protease treated crust and a reference crust. Also a lipase treatment and lecithin addition to the dough slowed down the water uptake kinetics. Both lipase and the addition of lecithin will have an effect on the stability of the gas cells in the dough, which could therefore point at a morphology effect. Especially the protease treatment of the bread crust worked well for maintaining crispness in the crust. This can be explained in several ways. First the initial water content of the treated crust is lowered which will also slow down further water uptake kinetics. Secondly the break down of the gluten may have resulted in a more dense matrix due to the decreased gas holding capacity of the matrix. Probably also the beams are thicker resulting in a larger diffusion distance and slower overall water uptake rates. Above conclusion suggest that a more detailed investigation of the effect of morphology on crispness (retention) would be a good subject for a further study. Especially a method to analyze beam thickness and gas cell size distribution is needed.

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References

1. Luyten, H.; Plijter, J. J.; van Vliet, T., Crispy/crunchy crusts of cellular solid foods: a literature review with discussion. *Journal of Texture Studies* **2004,** 35, (5), 445-492.
2. Roudaut, G.; Dacremont, C.; le Meste, M., Influence of water on the crispness of cereal-based foods: acoustic, mechanical, and sensory studies. *Journal of Texture Studies* **1998,** 29, (2), 199-213.
3. Labuza, T. P.; Hyman, C. R., Moisture migration and control in multi-domain foods. *Trends in Food Science and Technology* **1998,** 9, 47-55.
4. Roca, E.; Guillard, V.; Guilbert, S.; Gontard, N., Moisture migration in a cereal composite food at high water activity: Effects of initial porosity and fat content. *Journal of Cereal Science* **2006,** 43, 144-151.
5. Fu, Y.-C.; Tong, C. H.; Lund, D. B., Moisture migration in solid food matrices. *Journal of Food Science* **2003,** 68, (8), 2497-2503.
6. Roberts, J. S.; Tong, C. H.; Lund, D. B., Drying kinetics and time-temperature distribution of pregelatinized bread. *Journal of Food Science* **2002,** 67, (3), 1080-1087.
7. Guillard, V.; Broyart, B.; Bonazzi, C.; Guilbert, S.; Gontard, N., Moisture diffusivity in sponge cake as related to porous structure evaluation and moiture content. *Journal of Food Science* **2003,** 68, (2), 555-562.
8. Roca, E.; Broyart, B.; Guillard, V.; Guilbert, S.; Gontard, N., Controlling moisture transport in a cereal poreus product by modification of structural or formulation parameters. *Food Research International* **2007,** 40, 461-469.
9. Primo-Martín, C.; Castro-Prada, E. M.; de Wijk, R.; Vereijken, P. F. G.; Meinders, M. B. J.; van Vliet, T., Effect of structure in the sensory characterization of the crispness of toasted rusk roll. **Submitted**.
10. Roman-Gutierrez, A. D.; Mabille, F.; Guilbert, S.; Cuq, B., Contribution of specific flour components to water vapor adsorption properties of wheat flours. *Cereal Chemistry* **2003,** 80, (5), 558-563.
11. Primo-Martín, C.; de Beukelaer, H.; Hamer, R. J.; van Vliet, T., Fracture behaviour of bread crust: Effect of ingredient modification. **Submitted**.
12. Primo-Martín, C.; van de Pijpekamp, A.; van Vliet, T.; de Jongh, H. H. J.; Plijter, J. J.; Hamer, R. J., The role of the gluten network in the crispness of bread crust. *Journal of Cereal Science* **2006,** 43, (3), 342-352.
13. van Nieuwenhuijzen, N. H.; Tromp, R. H.; Hamer, R. J.; van Vliet, T., Oscillatory water sorption test for determining water uptake behavior in bread crust. *Journal of Agricultural and Food Chemistry* **2007,** 55, 2611-2618.
14. Primo-Martín, C.; Hamer, R. J.; de Jongh, H. H. J., Surface Layer Properties of Dough Liquor Components: Are They Key Parameters in Gas Retention in Bread Dough? *Food Biophysics* **2006,** 1, (2), 83-93.

15. Tromp, R. H.; Primo-Martin, C.; Zedde, R.; Koenderink, N., Quantifying the morphology of bread. *Proceedings of Bubbles in Food 2: Novelty, Health and Luxury International Conference* **2007**.
16. Babina, P.; Della Valle, G.; Chiron, H.; Cloetens, P.; Hoszowska, J.; Pernot, P.; Réguerre, A. L.; Salvo, L.; Dendievel, R., Fast X-ray tomography analysis of bubble growth and foam setting during breadmaking *Journal of Cereal Science* **2006,** 43, (3), 393-397.
17. Lim, K. S.; Barigou, M., X-ray micro-computed tomography of cellular food products. *Food Research International* **2004,** 37, (10), 1001-1012.
18. Primo-Martín, C.; Sozer, N.; Vliet, T. van, Effect of water activity on crispness of a model (bread) crust.

Chapter 7 General discussion

7.1 Introduction

Consumer appreciation of brittle cellular foods, such as bread crusts, depends on textural properties such as crispness. This crispy character is lost above a certain water activity (*1, 2*). For example, for bread crust this is at a water activity above approximately 0.6 (*3*). It is especially difficult to keep a food crispy when the product has a moist interior. Water then migrates from the interior to the crust causing hydration of the crust components. This results in an increase of the mobility of initially segments of the macromolecules and finally the whole molecule, which causes a glass to rubber transition of the main macromolecules present (mainly proteins and carbohydrates) that were initially in the glassy state. Roughly at the same time a loss of the crispy behaviour occurs (*4*), (*5*). To improve crispy behaviour of bread crusts (e.g. prolong the time a crust stays crispy), more information is required about the mechanisms underlying its deterioration. The overall objective of this thesis was to better understand the role of water in the mechanism of loss of crispness in crispy bread crusts. The kinetics of water uptake were studied because these mainly determine the time a crispy product stays crispy. Besides that the different aspects of mobility of water and the relations between water and other ingredients present in bread crusts were evaluated.

7.2 Methods

Bread crust is an inhomogeneous product, both regarding composition and dimensions. This makes it difficult to relate water uptake kinetics to product properties. For example Roman-Gutierrez et al have measured porosity and particle size of starch, gluten and non-starch polysaccharides (*6*) and used this information to determine a diffusion coefficient by Fick's law. However, porosity and particle size do not give enough information to define the diffusion distance. Water uptake kinetics will also depend on beam thickness and interconnections between gas cells. Besides that, more information is required about the water uptake process and the parameters that are important for the kinetics. Since the samples are inhomogeneous different relaxation processes are expected and with that probably also different water uptake rates. The combination of experiments with an oscillating RH and a stepwise increasing RH made it possible to understand the relevant processes (e.g. is the water transport to the crust particles or the water transport into the matrix time limiting?). In addition this combination of experiments allows a more thorough understanding of the methods used, the relevant time scales of the water uptake process and with that of the relevant time scales needed for our experiments. Next to the kinetic information also knowledge is necessary about the relation between crispness and the glass transition. This relation has been studied for bread crumb and toasted breads (*7, 8*), but not for bread crusts. In this chapter our results will be placed in a somewhat broader perspective. First our model system is discussed. Next, based on the results presented in this thesis and literature some suggestions are made on how to improve the time a crispy bread crust remains crispy. Finally some calculations will be presented on the morphology of the bread crust, based on the effect of particle size on water uptake kinetics.

7.3 Materials

Part of the work in this thesis has been done on model bread crusts instead of real bread crusts because it was more easy to prepare a model crust than to isolate the crust from breads. This was necessary for measurements that need to be performed under steady state conditions like the determination of the glass transition temperature and the sensory evaluation as a function of water activity or water content. Besides that the model crusts allowed quicker testing of the effect of variations in ingredient formulation on for example glass transition and water uptake kinetics. A model bread crust is a thin layer of dough that has been baked in a Halogen oven. The result is a 1.5 mm thick model crust with a slight brown colour which is characteristic for a bread crust. There is a small difference in degree of relative cristallinity between a model crust and a real bread crust from Soissons flour as determined by DSC (14 % +/-3 for the model crust and 28% +/-9 for the real crust), but this difference is expected to be too small to influence the results. Also a difference in porosity between model crusts and rusk roll crusts was found, the porosity of a Soissons model crust was around 28 +/- 5% and the porosity of a crispy roll crust was 49 +/-3% (*9*). Primo-Martín et al found that the coarseness of rusk rolls was influencing crispness perception. However, the difference in coarseness did not significantly change the critical water activity at which 10% or 50% of the initial crispness was lost (*10*). Therefore it is expected that the sensorial transition point as found for the model crusts as a function of water activity and water content should be comparable with that of a real crust.

7.4 Main findings

In this thesis both the mobility of water and the mobility of the macromolecules in bread crust is discussed. The mobility of water is discussed in chapter 2,3 and 6 and the mobility of the macromolecules in chapter 4 and 5. In Chapter 2 and 3 the mechanism by which the water moves through the crust was evaluated and methods were developed for the analysis of the speed of water uptake. The water uptake curves could be described by a single exponential. It was concluded that the transport of water into the matrix was the rate limiting step and that the transport mechanism by which the water migrates through the solid matrix is most likely a Case II type mechanism of water migration. Chapter 4 and 5 focused on the mobility of the macromolecules by measuring glass transition temperatures and relating this mobility to sensory data. Here it was concluded that the loss of sensory crispness was more related to an increase in the mobility of the water and possibly other small solutes and polymer side chains than to an increase in the mobility of the entire macromolecules. The measured glass transition temperatures were much higher than room temperature in the range of water contents where the main loss in crispness was observed. The water activity at which the crispness of model crust was lost was in the same range as the water activity at which gluten extrudates lost their crispness (*11*). Therefore it could be concluded that the gluten matrix likely is responsible for crispness and loss of crispness in bread crust. This fits in with the conclusion of Primo-Martin et al who found that gluten forms a continuous phase in bread crust (*3*). Chapter 5 also shows that the history of the sample in terms of water content and a_w is important for the crispness behaviour. It is possible to have different water contents in the system at one a_w (for example due to a hysteresis effect; a history of high water content) and still maintain a certain degree of crispness. This is likely due to a different distribution of the water through the crust depending on the history of the sample. In chapter 6 the effect of ingredient modification or the addition of certain ingredients is evaluated. It was found that the treatment of crispy roll crust with a protease resulted in slower water uptake rates in bread crust particles larger than 0.25 mm. Also when comparing model crust particles with crispy roll crust particles (particles larger than 0.25 mm) a difference was found. The model crust particles took up water at a slower rate. As model crust and crispy roll crust differ in morphology, these results suggest that morphology (beam thickness, air bubble size distribution) could be important for the rate of water uptake.

7.5 Crispy bread crust design

7.5.1 Composition, ingredients and crust modifications

As far as the ingredients and modifications are concerned some differences in water uptake rates have been found, but only for components or modifications that influence the gas cell distribution in the bread dough. These are components and modifications that are used as bread structure improver. For example lecithin slows down water uptake rates by a factor 1.5 at a RH between 60 and 70%. Lecithin is a component that stabilizes the air bubbles in the dough and likely also in the crust. Xylanase or the addition of extra arabinoxylans (water extractable or water soluble) did not influence the water uptake rates, but these components also did not have an influence on crust porosity. A comparison between the water uptake kinetics of a model crust and a real bread crust shows that a model crust has a factor 1.5-2 slower water uptake rates at 60 and 70% RH. This is likely due to a larger beam thickness of the model crust, pointing at the importance of morphology for crispness retention. A difference of a factor 1.2-1.6 in water uptake rates could be detected between model crusts of a protein rich and a starch rich model crust in an oscillatory sorption experiment. The protein rich model crust had a slower water uptake rate. This could be due to a more dense structure of the protein rich model crust which is again a morphology effect. From this we may conclude that if there is an effect of ingredients on water uptake rates the reason for it may be a change in morphology.

For bread crust we may conclude that a water activity below 0.55 is required to rate it as crispy. In table 1 crispy/non-crispy transition water contents and water activities at room temperature (22°C) of bread crust and the separate components starch and gluten are tabulated. These data, which are partly from this thesis and partly from literature, are based on results obtained by using different analytical techniques. Loss of sensorial crispness occurs before the actual glass transition, but at more or less the same water contents as the transition point in NMR T_2 curves. This indicates that the increased mobility of the water and probably also of other small molecules, short chain segments or polymer side chains are a signature of the loss of crispness. The mobility of the water associated with gluten increases at a higher water activity than the mobility of the water associated with starch. This corresponds with data from Nicholls et al (*11*) for the sensorial crispness retention in gluten and starch extrudates. This would indicate that a gluten network is necessary to retain a crispy crust structure because this gluten network can keep its crispness up to a higher water activity than when only starch would be used. When comparing gluten and starch, the glass transition (*12*) differs in a way opposite to that of the T_2 transition. This confirms that the glass transition has little to do with the start of sensorial loss of crispness.

On the basis of the NMR results in this thesis, the NMR method may be suggested as a good tool to analyse different components like other plant-derived proteins (zein, soy protein, lupine protein, gliadin) with respect to the relation between water activity and proton mobility. In this way ingredients could be scanned for their potential to form a crispy network and maintain that up to a higher water activity.Roudaut et al (*4*) investigated the sensory loss for extruded flat bread and found a critical water content (water content where 50% of the crispness is lost) of 10.5% water. Primo-Martín et al (*10*) found a critical water activity of 0.57-0.59 for toasted rusk roll, corresponding to 9.1-9.7 % water. In chapter 4 we found a critical water activity of 0.6, which corresponds to 11.5% water for a bread crust. Thus also in a system with 100% gelatinized starch like a toasted rusk roll or toasted bread, crispness can be maintained above the critical water content of 5% or water activity of 0.1-0.3 of the starch extrudates as measured by Nicholls et al. and Valles Pamies et al. (*11, 15*). Probably the amount of gelatinized starch in this type of products is not so important for the critical water content or a_w as long as an intact gluten network is present. However, more gelatinised starch in the bread crust could result in an initially higher water content in bread crust after baking, which would not be beneficial for crispness retention. The slightly lower transition water content found by Roudaut et al for bread toast and Primo-Martín et al. for bread crust (*4, 10*) compared to model crust could be due to extra ingredients that were added to the bread and rusk roll formulations like fat or sugar. In both studies commercial products were used.

7.5.2 Water uptake

The transport of water into the matrix was the rate limiting step in the water uptake process (chapter 2). In chapter 3 we found that in bread crust particles probably case II water transport is taking place and not Fickian diffusion. This means that the relaxation time of the matrix determines the time it takes for water to move into the sample. Slowing down the water uptake process would be possible by using ingredients that make it difficult for water to move through, but so far we have no examples of biopolymer ingredients that have such an effect. Figure 1 depicts schematically the effect of both starting conditions (in terms of water content) and kinetics of water uptake on the time in which the critical water content is reached.

Both lowering water uptake kinetics and starting with a lower water content will increase the time during which a product is crispy. With a lower initial water content it will take more time before the critical water content is reached because more water has to move in. From figure 2 can be concluded that in the region of water activities of a fresh bread crust and a stale bread crust (between an a_w of 0.4-0.6) the water uptake kinetics increase with increasing water content. Between a_w 0.4 and 0.6 the rate parameter increases from 0.0055 to 0.011 min^{-1}, which is a factor 2 increase in water uptake rate. This would correspond to a decrease from 180 to 90 minutes of the time that half of the water that can go in will go in. Starting with a low water content is therefore in two ways favourable for increasing the time a crust is crispy:

- More water has to move in before the critical water content is reached
- The kinetics of water uptake will at least initially be slower.

Protease spraying on the bread dough surface has been proved to help lowering the initial water content of a bread crust after baking (*3*) and as a consequence retention of crispness during a longer time.

In addition to the amount of water also the way the water is distributed can have an effect on crispness as was shown in chapter 5. A history of high water content will make a bread crust less crispy when dried back to a certain water activity. These results show that when designing crispy products it is important to consider both water content and water activity. During the production process a product prepared from a dry state will give a more crispy result at a certain water activity than a product which had a history of a high water content and was dried back. Therefore an extra drying step after bread baking would probably work less good for maintaining crispness in the crust than making sure that enough water is lost during the baking process in the oven. Wetting a product and drying back to get it crispy should be avoided. This provides an extra complication for improving the quality of part baked breads that are stored in a freezer or packaged under protected atmosphere and baked off later. For the breads stored in the freezer this could be avoided by cooling and freezing the breads fast while the crust is still dry. The cooling down could for example be done by vacuum cooling (*16*) because this resulted in a lower water content of the crust after cooling .

Tabel 1 Critical water content and water activity at 22°C, e.g. water content corresponding to a T_g at 22°C for gelatinized starch is 22% an the corresponding a_w is 0.88 as measured with DSC. Both data from this thesis and literature. Tg is the glass transition temperature, DSC is differential scanning calorimetry, PTA is phase transition analyser, NMR is nuclear magnetic resonance, T_2 is a proton relaxation time, associated with the mobility of water (for further explanation see chapter 4), TA is texture analyser.

Product	T_g DSC [H_2O] a_w	T_g PTA	Sensorial transition	Transition content NMR T_2 (ms)	Transition water content TA (acoustic emission, i.e. sound during fracture)
Model crust	>15.3 >0.77	15.8 0.80	10.0 0.55	10 0.58	≤7.5 (0.1 mm/s) ≤0.40
Gluten (heated)	16.4 0.85 (*12*)	18.6 (vital) 0.95 (*13*)	7.3-9.3 0.35-0.57 (*11*)	11 0.70	11.8 (0.8 mm/s) 0.72 (*14*)
Starch (gelatinized)	22 ~0.88 (*12*)	>16 >0.75	~5 0.1 (*11*) 0.31 (*15*)	11.1 0.48	8.1 (0.8 mm/s) 0.23 (*14*)

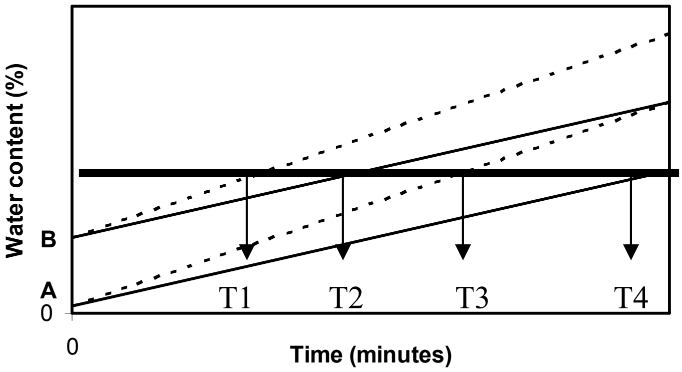

Figure 1 Schematic picture of the effect of initial water content and water uptake rates on the time it takes to reach the critical water content (thick line). Slow water uptake rates are indicated by the solid lines, fast water uptake rates by the dashed line. Examples for starting at a low initial water content (A) and a higher initial water content (B) are shown. T1-T4 indicate the time until the critical water content is reached.

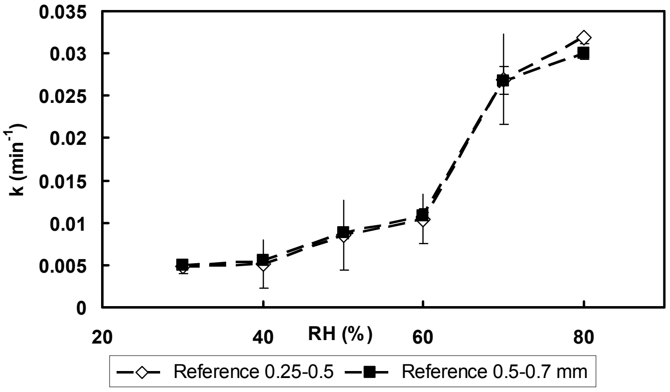

Figure 2. Rate parameter k as a result of a single exponential fit for water uptake of a Soissons crispy roll reference crust for 2 different crust particle sizes (size 0.5-0.7 mm and 0.25-0.5 mm) as measured with the SPS 11 (see chapter 6).

7.5.3 Effect of morphology

Indirect estimation of beam thickness

Figure 2 shows the effect of particle size on the rate parameter of the water uptake process by grinded crusts for larger particle sizes of a Soissons crispy roll crust; 0.25-0.5 mm and 0.5-0.7 mm. There is no difference between the rate parameters of the two samples. When looking microscopically to these particles this could be expected since the particles have the shape of thin sheets, with a thickness that likely corresponds to a beam/film thickness or the relevant length scale of the crust. The fraction 0.5-0.7 are just bigger sheets but with probably the same the thickness as the 0.25-0.5 mm particles. This is depicted in figure 3c. One piece of the structure can contain a beam with a thickness of for example 250 µm. With respect to the diffusion time it will not matter if it is a small piece like in figure 3c or part of the hole structure like in the open bubble structure of figure 3b. However if this beam is fractured again the diffusion distance can become smaller. We found an effect of the size on the water uptake kinetics (see chapter 2 and 3) between grinded crust particles of 0.25-0.5 mm and the fraction of particles with a smaller size (63 µm to 0.25 mm and smaller than 63 µm). From this a length scale relevant for the time dependence of the water uptake could be estimated of 250 to 500 µm. Between this fraction and a fraction with particles larger than 500 µm the kinetics did not change. Luyten and van Vliet estimated that the beams, that define one of the probably several length scales determining the optimum morphology for crispness, should be between 100 and 350 µm based on information derived from fracture mechanics, acoustic emission and physiological aspects of humans (*17*). Beam size and the length scale relevant for the water uptake kinetics appear to be in the same range. Similar information was obtained for model crust particles. Here particles with a size between 0.5 and 0.7 do have slower water uptake kinetics than particles with a size between 0.25 and 0.5 mm (results not shown). When the relevant length scale governing the water uptake process can indeed be identified with the size of the particles, it could be suggested that beams are thicker in the model crust. This could be expected since the model crust appears to be more dense and sensorially harder than a bread crust. These type of measurements could be used as an indirect method for the estimation of the beam size of bread crust particles. The two hours that it takes for water in a freshly baked loaf of bread to move from the bread crumb into the crust suggests that the bread crust structure must be very open. The distance, the water has to travel in figure 3a to reach the upper beam, if the water is coming from the bottom is twice as long as in figure 3b. In 2 hours the water likely only passes one beam.

The diffusion coefficient of water in a bread crust (10-15% water) was estimated to be 10^{-12}-10^{-13} m^2/s (*18*). The average distance x travelled by a diffusing water molecule can be estimated by:

$$x = \sqrt{6Dt}$$

D is the diffusion coefficient and t is time. With a beam thickness of 190 µm and, D estimated at $5*10^{-13}$ m^2/s, the time needed for the water to reach the middle of the beam is 120 minutes. The time it actually takes to loose crispness in a bread crust is around 120 minutes (*3*). Assuming a beam thickness of 190 µm is therefore consistent with experimental observations. So thicker beams would help slowing down water uptake.

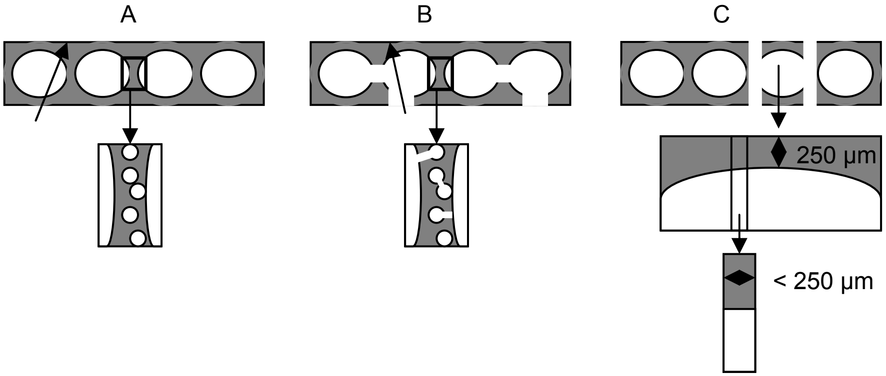

Figure 3 Schematic picture of the shortest travelling path for a water molecule to reach the upper layer. In grey solid matrix, in white open spaces. 3a indicates a foam and the zoom indicates the air bubble distribution in the beams, 3b indicates an -towards the bottom side-open structure, figure 3c indicates a fractured structure. The upward pointing arrow indicates the shortest water diffusion path.

7.6 Recommendations

Morphology was found to be important for the rate of water uptake and also for the perception of crispness. Therefore it would be of interest to further characterise the bread crusts with respect to beam thickness, porosity, air bubble structure and the extent to which different gas cells are in contact with each other. Different techniques that could be used are CSLM, X-ray tomography and mercury porosimetry. Closed structures where pores are not connected to each other are favourable, so it would be of interest to investigate how this type of structure could be obtained. Further research on the analysis of the water uptake curves, the effect of experimental times and the different processes going on in a bread crust during water uptake could be useful to better understand the water uptake mechanism. Another subject of study could be the diffusion of water in the crumb. The speed with which the water moves through the crumb to the crust will also influence the time a crust stays crispy. It would also be of interest to look more into detail to the effect of water distribution on crispness, e.g. with MRI, and how this knowledge could be applied to design crispy crust.

References

1. Luyten, H.; Plijter, J. J.; van Vliet, T., Crispy/crunchy crusts of cellular solid foods: a literature review with discussion. *Journal of Texture Studies* **2004,** 35, (5), 445-492.
2. Meste, M. le; Champion, D.; Roudaut, G.; Blond, G.; Simatos, D., Glass transition and food technology: a critical appraisal. *Journal of Food Science;* **2002,** 67, (7), 2444-2458.

3. Primo-Martín, C.; van de Pijpekamp, A.; van Vliet, T.; de Jongh, H. H. J.; Plijter, J. J.; Hamer, R. J., The role of the gluten network in the crispness of bread crust. *Journal of Cereal Science* **2006,** 43, (3), 342-352.

4. Roudaut, G.; Dacremont, C.; le Meste, M., Influence of water on the crispness of cereal-based foods: acoustic, mechanical, and sensory studies. *Journal of Texture Studies* **1998,** 29, (2), 199-213.

5. Slade, L.; Levine, H., A food polymer science approach to structure-property relationships in aqueous food systems: non-equilibrium behavior of carbohydrate-water systems. *In Water Relations in Foods (L. Slade and H. Levine, eds.) pp. 29–101, Plenum press, New York.* **1991**.

6. Roman-Gutierrez, A. D.; Mabille, F.; Guilbert, S.; Cuq, B., Contribution of specific flour components to water vapor adsorption properties of wheat flours. *Cereal Chemistry* **2003,** 80, (5), 558-563.

7. Fontanet, I.; Davidou, S.; Dacremont, C.; Meste, M. le, Effect of water on the mechanical behaviour of extruded flat bread. *Journal of Cereal Science. 1997;* **1997,** 25, (3), 303-311.

8. Roudaut, G.; Maglione, M.; Meste, M. le, Relaxations below glass transition temperature in bread and its components. *Cereal Chemistry* **1999,** 76, (1), 78-81.

9. Primo-Martín, C.; Beukelaer, H. d.; Hamer, R. J.; van Vliet, T., Fracture behaviour of bread crust: Effect of ingredient modification. **Submitted**.

10. Primo-Martín, C.; Castro-Prada, E. M.; de Wijk, R.; Vereijken, P. F. G.; Meinders, M. B. J.; van Vliet, T., Effect of structure on the sensory characterization of the crispness of toasted rusk roll **Submitted**.

11. Nicholls, R. J.; Appelqvist, I. A. M.; Davies, A. P.; Ingman, S. J.; Lillford, P. J., Glass transitions and the fracture behaviour of gluten and starches within the glassy state. *Journal of Cereal Science. 1995;* **1995,** 21, (1), 25-36.

12. Laaksonen, T. J.; Roos, Y. H.; Labuza, T. P., Comparison of the use of desiccators with or without vacuum for water sorption and glass transition studies. *International Journal of Food Properties* **2001,** 4, 545-563.

13. Bengoechea, C., Arrachid, A., Guerrero, A., Hill, S.E. and Mitchell, J.R., Relationship between the glass transition temperature and the melt flow behavior for gluten, casein and soya. *Journal of Cereal Science* **2007,** 45, (3), 275-284.

14. Attenburrow, G. E.; Davies, A. P.; Goodband, R. M.; Ingman, S. J., The fracture behaviour of starch and gluten in the glassy state. *Journal of Cereal Science* **1992,** 16, (1), 1-12.

15. Valles Pamies, B.; Roudaut, G.; Dacremont, C.; le Meste, M.; Mitchell, J. R., Understanding the texture of low moisture cereal products: mechanical and sensory measurements of crispness. *Journal of the Science of Food and Agriculture* **2000,** 80, (11), 1679-1685.

16. Primo-Martín, C.; Beukelaer, H. de; Hamer, R. J.; van Vliet, T., Fracture behaviour of bread crust: Effect of bread cooling conditions. **Submitted**.

17. Luyten, H.; van Vliet, T., Acoustic emission, fracture behavior and morphology of dry crispy foods: A discussion article. *Journal of Texture Studies* **2006,** 37, 221-240.

18. Bruin, S.; Luyben, K., Drying of food materials: a review of recent developments. *In:A.S. mujumbar, ed. Advances in Drying, Vol.1. Hemisphere, Washington* **1980**, 155.

Summary

Many different types of crispy foods can be found in the supermarket. Products that are crispy range from potato crisps to bread to fish fingers to ice cream cones and vegetables. Crispy foods are strongly appreciated by consumers. For products to be judged as crispy they should break in a brittle way and emit sound while being eaten. These characteristics of a crispy food product are generally lost upon plasticizer uptake (mainly water) from the environment or from a more wet part of the product. The work described in this thesis was part of a big project aimed at obtaining a better understanding of all factors involved in crispy behaviour of composite foods both at molecular, mesoscopic and macroscopic scale. The overall objective of this thesis was to better understand the role of water in the mechanism of loss of crispness in crispy bread crusts. Especially the different aspects of mobility of water and the relations between water and other ingredients present in bread crusts were evaluated. Besides that the kinetics of water uptake were studied because these mainly determine the time a crispy product stays crispy. This should finally result in an improved shelf life of crispy composite foods.

The focus of this thesis was on composite foods with a moist interior and a dry and crispy outside. For these products the water that plasticizes the crispy parts during storage will come primarily from the moist part of the product. The model systems used in this thesis were a bread model system and a bread crust model system. As part of this thesis work methods were developed to measure water uptake kinetics in crispy bread crust. With these methods the effect of ingredient variation and morphology on water uptake rates were studied. Besides the water uptake kinetics also glass transition temperatures were measured as a function of water content or ingredient variation and the role of the main components of wheat flour, gluten and starch was discussed. Results obtained were used to construct a working model explaining the role of the main factors affecting crispness creation and retention. This will be used by other sub/projects of the main project to establish guidelines for the industry to improve crispness creation and retention of the crust of baked bread type products.

In chapter 2 and 3 the mechanism of water uptake in bread crust is discussed. In chapter 2 the water sorption kinetics of bread crust are described using an oscillatory sorption test in combination with a Langmuir type equation. Both kinetic and thermodynamic information could be obtained at the same time. An advantage of applying a Langmuir type equation for a quantitative description of the water uptake kinetics is that no prior knowledge is necessary with respect to shape and surface area of the sample. It was shown that adsorption and desorption of water to the bread crust particle surface is much faster than the experimental time used (15 minutes at minimum). From this we may conclude that diffusion of water into the solid matrix is the rate limiting step in the water sorption process. The method also allows to calculate a Gibbs free energy. The method is suitable for use up to RH 60%. In chapter 3 the mechanism that controls the water transport into the solid matrix is studied further by evaluating the kinetics of water uptake by bread crust particles for two different sorption experiments, the oscillatory sorption test

described in chapter 3 and a more classical sorption test in which the air RH was increased stepwise. In this way the effect of experimental sorption time on the resulting rate parameters could be studied because in the stepwise sorption test each step took a factor 25 longer than in the oscillatory sorption test. Results show that the adsorption and desorption dynamics of water into the solid matrix of the oscillatory sorption test could be described best by a single exponential in time and not by a Fickian type square root time dependence. Such an exponential behaviour is often ascribed to Case II relaxation controlled diffusion. A maximum in the water uptake rate was found for a RH value between 50 and 70%. The rate parameters of the experiment where RH was stepwise increased were around a factor 10 lower than those derived from the oscillatory sorption experiments. This indicates that the experimental time used affects the outcome of the experiment. This was confirmed by a test where the RH was increased stepwise for native wheat starch. The equilibration time per step was 300 or 60 minutes. The water uptake at equilibrium was equal for both experiments, but the kinetics were faster for the experiment with steps of 60 minutes. This is an important factor when designing experiments for the determination of water uptake rates. In addition, also a parameter describing the time dependence of the rate parameters of the oscillatory sorption experiment was calculated (C). C was in the same range as the rate parameter of the isotherm experiment. This indicates that different (relaxation) processes are acting at the same time in the bread crust during water uptake. A significant difference in the rate parameter value of the single oscillation steps was found between model crusts made of two different fractions of an air-classified flour, one enriched in protein, the other enriched in starch. The model crust of the starch rich fraction gave the higher value.

In chapter 4 the effect of water on the glass transition of model bread crusts was studied using three complementary techniques: phase transition analysis (PTA), temperature modulated differential scanning calorimetry (TMDSC) and nuclear magnetic resonance (NMR). The results were compared with sensory data. Bread crusts prepared with different types of flour were tested to evaluate the effect of ingredients on crispness of model crusts equilibrated at different relative humidities. The results were compared with the results for the single flour components starch and gluten. Sensory crispness scores decreased at an a_w of 0.55 and decreased further with increasing $a_{w.}$ At an a_w higher than 0.70 sensory crispness was completely lost. Both DSC and PTA showed a glass transition temperature at around 20°C (room temperature) for an a_w of 0.70-0.75. This supports the hypothesis that loss of crispness starts as a result of processes at a molecular level, before the macroscopic glass transition. NMR gave a transition point in the mobility of the protons of water at a_w 0.58. This points in the direction of non-bound water (water that is not directly attached to the solid matrix) causing the loss of crispness at low a_w. At higher a_w increased mobility of initially segments and side chains of the macromolecules will start to play a role. NMR experiments on the single components indicate that the transition point for mobility of water in starch samples is at a lower a_w than for gluten. This would imply that crispness of starch is lost at lower a_w than crispness of gluten. Comparison with literature suggested that the model bread crusts maintain their crispness up to the same point as gluten extrudates. This would mean that the crispness of bread crust is mainly related to the gluten network.

Chapter 5 discusses the relation between loss of sensorial crispness in a bread crust model on the one hand and the water content and a_w on the other hand. All experiments were carried out on model bread crusts of a Soissons bread flour. The model crust samples were brought at the same a_w, but different water contents by making use of the hysteresis effect in water sorption. This was done to study the separate effects of a_w and water content. The effect of water content and water activity on the glass transition of model bread crusts was studied in detail using two complimentary techniques: PTA and NMR. The results were compared with sensory data and results from a puncture test which provided data on acoustic emission and fracture mechanics during breaking of the crusts. The water content (instead of a_w) was found to be decisive for the transition point as measured by PTA and NMR. However, a comparison with sensory data and data from a puncture test showed that both water content and water activity had an effect on perceived crispness and number of force and sound peaks. Possibly an inhomogeneous distribution of water and consequently crispy and less-crispy regions exist in the samples with a history of high a_w thus making them overall more crispy than samples with the same water content but higher a_w.

In chapter 6 bread crusts of different formulations were tested regarding their water uptake kinetics. Amylase treatment and xylanase treatment did not induce a conclusive change in the water uptake kinetics of crust particles of a crispy roll. Also the addition of arabinoxylans (both soluble and unsoluble) to model bread crust did not change the water uptake kinetics. However, treatment of a bread crust with protease (sprayed on top of the bread dough) resulted in slower water uptake kinetics for bread crust particles larger than 0.25 mm. For smaller particles there was no significant difference in water uptake rates. However, the standard deviations were much larger than for the larger particles. This was likely due to a larger effect of the packing of the particles in the measuring cup, for the smaller particles. The water uptake rates of a model crust (with a more dense structure) were compared with those of a crispy roll crust. The water uptake rates of the model crust were found to be slower which points at a morphology effect.

In chapter 7 some tools are discussed for designing crispy bread crusts. Lowering the initial water content in a bread crust is an important tool. Lowering the initial water content (for example by protease spraying) works in two ways. First more water can move into the crust before the critical water content is reached. Secondly the kinetics of water uptake will be slower at a lower water content. Based on the fact that a bread crusts looses its crispness in around 120 minutes, a diffusion coefficient from literature and the rate parameters for water uptake found in this thesis, the thickness of a beam in a bread crust was estimated to be around 200 μm. No effect of the amount of gelatinized starch was found on the critical water activity above which the crispness is lost.

Samenvatting

In de supermarkt worden diverse krokante producten verkocht. Deze producten variëren van aardappel chips tot vissticks tot ijshorentjes en knapperige groenten. Krokante producten worden zeer gewaardeerd door de consument. Om een product krokant te kunnen noemen moet deze bros breken en moet er geluid vrijkomen tijdens het eten. Deze eigenschappen van krokante producten gaan verloren als het product vocht opneemt uit de lucht of van een vochtig onderdeel van het product. Het werk dat in dit proefschrift beschreven wordt was onderdeel van een groter project (het “Crispy / Crunchy project”). Het doel van dit grotere project was om de factoren die van invloed zijn op krokant gedrag van levensmiddelen beter te begrijpen. Dit zowel op microscopisch, mesoscopisch en macroscopisch niveau. Het doel van dit proefschrift was de rol van water in het verlies van krokantheid beter te begrijpen. Met name de beweeglijkheid van water en de relatie tussen water en de verschillende andere componenten in de broodkorst waren het onderwerp van deze studie. Ook de kinetiek van water opname werd onderzocht omdat dit mede bepaald hoe lang een krokant product krokant blijft. Uiteindelijk moet dit resulteren in een langere houdbaarheid van krokante producten.

Het model product voor dit proefschrift is een samengesteld product met een droog en krokant deel en een zacht en vochtig deel. Het water dat ervoor zorgt dat de korst nat wordt komt in dat geval uit het product zelf. In dit proefschrift zijn twee model systemen gebruikt, een model korst en de korst van een model brood. Voorts werden methoden ontwikkeld om de water opname kinetiek te kunnen meten in krokante broodkorsten. Met deze methoden werd ook het effect van ingrediënt variatie en morfologie op de kinetiek van water opname onderzocht. Naast de kinetiek van water opname werden ook glas overgangen bepaald als functie van water activiteit en vochtgehalte. De rol van de verschillende componenten van bloem (zetmeel en gluten) aanwezig in brood korst werd bediscussieerd. De resultaten werden gebruikt om een model te ontwikkelen dat de rol van verschillende factoren in krokantheid en het verlies van krokantheid van broodkorsten kan verklaren.

In hoofdstuk 2 en 3 wordt het mechanisme van water opname beschreven. In hoofdstuk 2 wordt de wateropname kinetiek in broodkorst beschreven door het gebruik van een oscillatorische sorptie test in combinatie met een Langmuir-achtige vergelijking. Dit leverde tegelijkertijd zowel kinetische als thermodynamische informatie op. Een voordeel van het gebruik van een Langmuir-achtige vergelijking voor de kwantitatieve beschrijving van de kinetiek van water opname is dat het niet noodzakelijk is de vorm en het oppervlak van de deeltjes te weten. Uit de oscillatorische sorptie experimenten kan geconcludeerd worden dat het transport van water naar het oppervlak toe vele malen sneller is dan de duur van het experiment (15 minuten per stap). Hieruit kunnen we concluderen dat de diffusie van water in de vaste stof matrix de limiterende stap is in het water opname proces. Met deze methode kan ook een Gibbs vrije energie berekend worden. De methode werkt tot een luchtvochtigheid van 60%. In hoofdstuk 3 wordt het mechanisme beschreven waardoor het water de vaste stof indringt. Hiervoor zijn twee verschillende sorptie experimenten gebruikt: een waarbij de RH steeds geoscilleerd werd en een waarbij de RH in stappen steeds verhoogd werd. Op deze manier kon het effect van de duur van het experiment ook geëvalueerd worden. In het oscillatorische sorptie experiment duurde elke stap 25 minuten, in het experiment met stapsgewijze verhoging van de luchtvochtigheid duurde elke stap ongeveer 600 minuten. De resultaten laten zien dat de adsorptie en desorptie van het water in de vaste stof het best beschreven kan worden met een enkele exponentieel in de tijd en niet met een Fick achtige wortel-tijds afhankelijkheid. Dit gedrag wordt vaak toe geschreven aan Case II relaxatie gestuurde diffusie. Een maximum in de water opname kinetiek (snelheids parameter k) werd gevonden voor een luchtvochtigheid tussen de 50 en 70% voor het oscillatorische water opname experiment en voor een luchtvochtigheid van 80% voor het stapsgewijze water opname experiment. De k was een factor 10 lager voor het experiment met een stapsgewijze toename in RH dan voor het experiment met de oscillerende RH. Dit geeft aan dat de tijd die gebruikt is per RH stap de uitkomst van het experiment beïnvloedt. In een experiment met natief zetmeel (stapsgewijze verhoging van de RH) werd dit bevestigd. Elke stap duurde 60 of 300 minuten. Het water gehalte in stabiele toestand was voor beide experimenten gelijk, maar de water opname ging sneller voor het experiment met stappen van 60 minuten. Dat betekent dat de experimentele tijd belangrijk is voor het ontwerpen van experimenten voor het meten van vochtopname snelheden. Uit de k-waarden als functie van de tijd voor een oscillatorisch sorptie experiment is een tweede snelheids parameter (C) bepaald. Deze was van dezelfde ordegrootte als de k-waarde van het experiment met stapsgewijze verhoging van de RH. Dit duidt erop dat meerdere relaxatieprocessen tegelijkertijd plaats hebben. Tussen modelkorstjes van twee fracties van een gewindzifte bloem (een eiwitrijke en zetmeelrijke fractie) werd een significant verschil in de k-waarden gevonden voor het oscillatorische sorptie experiment. De model korsten van de zetmeel rijke fractie van de bloem hadden een hogere k-waarde.

In hoofdstuk 4 werd het effect van water op de glasovergang van model brood korsten bestudeerd met behulp van drie complementaire technieken: Phase Transition Analysis (PTA), Temperature Modulated Differential Scanning Calorimetry (TMDSC) en Nuclear Magnetic Resonance (NMR). De resultaten werden vergeleken met sensorische data. Brood korsten van fracties van bovengenoemde gewindzifte bloem en van een broodbloem (Soissons) werden getest om het effect van ingrediënten op krokantheid van model korsten bij verschillende luchtvochtigheid te kunnen bestuderen. De resultaten werden vergeleken met die van de aparte bloem componenten zetmeel en gluten. Sensorische krokantheid ging verloren boven een water activiteit van 0.55 en daalde verder met toenemende $a_{w.}$ Bij een a_w hoger dan 0.7 was de krokantheid volledig verdwenen. Zowel DSC als PTA lieten een glas overgang zien bij kamer temperatuur (20°C) voor een a_w van 0.7-0.75. NMR geeft een overgang in de mobiliteit van protonen van water bij een a_w van 0.58. Dit wijst in de richting van niet-gebonden water (water dat niet direct aan de matrix vast zit) dat het verlies van krokantheid veroorzaakt. Dit ondersteunt de hypothese dat verlies van krokantheid al begint voor de macroscopische glasovergang, dus al op moleculair niveau (bijvoorbeeld met mobiliteit van water en kleine zijgroepen). NMR experimenten op gluten en zetmeel apart laten zien dat de a_w op het omslag punt veel hoger is voor gluten dan voor zetmeel. Dit suggereert dat de krokantheid van zetmeel producten al bij veel lagere a_w achteruit gaat dan voor producten van gluten. Vergelijking met literatuur laat zien dat dit ook inderdaad het geval is. Dit betekent dat het behoud van krokantheid vooral afhangt van het gluten netwerk.

Hoofdstuk 5 gaat over de relatie tussen verlies van sensorische krokantheid van een model korst aan de ene kant en het water gehalte en de a_w aan de andere kant. Alle experimenten werden uitgevoerd met een model korst systeem van Soissons brood bloem. De model korst monsters werden op gelijke a_w gebracht, maar met behulp van het hysterese effect werden ze op verschillend vochtgehalte gebracht. Het doel was om het effect van a_w en water gehalte apart van elkaar te bestuderen. Het effect van water gehalte en a_w op de glas overgang van de model korsten is bestudeerd met de PTA, het effect op de mobiliteit van water met NMR en het effect op de mechanische en akoestische eigenschappen met een Texture Analyser (TA). Ook werd een sensorische test uitgevoerd. Het water gehalte (in plaats van a_w) bepaalde de resultaten van de PTA en NMR. Aan de andere kant werden de resultaten van de sensorische test en de TA (puncture test) bepaald door zowel water gehalte als $a_{w.}$ Mogelijk ontstaat er in de hysterese monsters een inhomogene verdeling van het water waardoor er krokante en minder krokante omgevingen ontstaan met een overall krokanter resultaat tot gevolg.

In hoofdstuk 6 werden verschillende variaties in ingrediënten in broodkorst getest op snelheid van water opname. Amylase en xylanase behandelingen leken geen effect te hebben op de snelheid van wateropname van korsten van harde witte bolletjes. Ook het toevoegen van arabinoxylanen (zowel oplosbare als onoplosbare) aan model korst had geen effect op de snelheid van water opname. Behandeling van broodkorsten met protease (gesproeid op het deeg) resulteerde in een lagere water opname kinetiek voor broodkorst deeltjes groter dan 0.25 mm. Voor kleinere deeltjes kon geen effect van de protease behandeling op de snelheid van vochtopname gevonden worden. Hier moet wel bij opgemerkt worden dat de standaard deviaties veel groter waren voor de kleine deeltjes dan voor de grote deeltjes. Dit komt waarschijnlijk doordat de kleine deeltjes veel dichter op elkaar gepakt in het meet cupje zitten dan de grote deeltjes en dat hierdoor de diffusieafstand gedeeltelijk bepaald kan zijn door het vullen van het cupje. Ook werden de water opname snelheden van model korst vergeleken met die van een echte korst. De opname van water ging langzamer voor een model korst dan voor een echte korst, waarschijnlijk doordat de modelkorst dikkere lamellen had en een wat dichtere structuur.

In hoofdstuk 7 worden een aantal handvaten gegeven voor het ontwerpen en behouden van krokante structuren. Allereerst is het van belang het vochtgehalte in de korst vanaf het begin zo laag mogelijk te houden. Het besproeien van het deeg met een protease oplossing kan hierbij helpen. Een laag start vochtgehalte zorgt ervoor dat er meer water in de korst kan worden opgenomen voor de kritische grens bereikt is. Bovendien zorgt een lager vochtgehalte ook voor een lagere water opname snelheid. De dikte van een lamel in een broodkorst kan berekend worden met behulp van diffusie coëfficiënten uit de literatuur en het feit dat een broodkorst in ongeveer 120 minuten zijn krokantheid verliest. Hieruit volgt een lamel dikte in broodkorst van ongeveer 200 μm.

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Neleke

Curriculum Vitae

Nelly Hermina (Neleke) van Nieuwenhuijzen werd op 20 januari 1978 geboren te Melissant. In 1996 behaalde zij haar VWO diploma aan de Regionale Scholen Gemeenschap Middelharnis. Aansluitend studeerde zij levensmiddelentechnologie aan de Wageningen Universiteit. Afstudeervakken heeft zij uitgevoerd bij de vakgroep proceskunde en bij Nizo food research. Stages werden gelopen bij de McGill University in St-Anne-de-Bellevue, Quebec, Canada en bij Delizza B.V. te Kruiningen. Dit resulteerde eind 2001 in een diploma levensmiddelentechnologie. Neleke vervolgde haar carrière als product- en procesontwikkelaar bij Delizza te Kruiningen en later als product technoloog bij Nutricia te Cuijk. Per 1 november 2003 werd zij aangenomen als promovenda bij het toenmalige Wageningen Center for Food Sciences (tegenwoordig TI Food and Nutrition) in het “Crispy Crunchy” project. Het werk uitgevoerd tussen november 2003 en november 2007 staat beschreven in dit proefschrift.

List of Publications

N.H. van Nieuwenhuijzen, M.B.J. Meinders, R.H. Tromp, R.J. Hamer and T. van Vliet, The water uptake mechanism in crispy bread crust, to be submitted.

N.H. van Nieuwenhuijzen, R.H. Tromp, J.R. Mitchell, C. Primo-Martín, R.J. Hamer and T. van Vliet, Relations between sensorial crispness and molecular mobility of model bread crust and its main components as measured by PTA, DSC and NMR, to be submitted.

N.H. van Nieuwenhuijzen, C. Primo-Martín, M.B.J. Meinders, R.H. Tromp, R.J. Hamer and T. van Vliet, Water content or water activity, what rules crispy behaviour?, to be submitted.

N.H. van Nieuwenhuijzen, C. Primo-Martín, T. van Vliet and R.J. Hamer, Effect of ingredient variation and modification on water uptake kinetics in crispy bread crusts, to be submitted.

M.B.J. Meinders, N.H. van Nieuwenhuijzen, R.H. Tromp, R.J. Hamer and T. van Vliet, Water sorption and transport in dry crispy bread crust, submitted.

N.H. van Nieuwenhuijzen, R.H. Tromp, R.J. Hamer and T. van Vliet, Oscillatory water sorption test for determining water uptake behaviour in bread crust, Journal of Agricultural and Food Chemistry, V55, p2611-2618, 2007.

C. Primo-Martín, N.H. van Nieuwenhuijzen, R.J. Hamer, T. van Vliet, Crystalline changes in wheat starch during the bread-making process: starch crystallinity in the bread crust, Journal of cereal science, V45(2), p219-226, 2007.

H.S. Ramaswamy, N.H. van Nieuwenhuijzen, Evaluation and modelling of two-stage osmo-convective drying of apple slices, Drying Technology, V20(3), p651-667, 2002.

N.H. van Nieuwenhuijzen, M.R. Zareifard and H.S. Ramaswamy, Osmotic drying kinetics of cylindrical apple slices of different sizes, Drying Technology, V19(3&4), p525-545, 2001.

Overview of completed training activities

Discipline specific activities

Vlag PhD week, VLAG, 2004

Miniworkshop crispy / crunchy behaviour of food products, TIFN (former WCFS), 2004

Workshop on water in foods, Nestle, Lausanne, 2004

Young Cereal Scientists workshop, Cereals and Europe, Dublin, 2004

PhD trip to Japan, Food Chemistry, 2004

Science of Baking, University of Reading, 2005

Young Cereal Scientists Workshop, Cereals and Europe, Vienna, 2005

Physical Modelling, WIMEK, 2006

Workshop on water in foods, Nestle, Brussels, 2006

Practical period at Nottingham Univeristy (Food Structuring Group), Loughbourrough, 2006

PhD trip to Belgium, France and United Kingdom, Food Chemistry, 2006

ISOPOW 2007 meeting, Bangkok, 2007

General courses

Afstudeervak organiseren en begeleiden, OWU, 2004

Scientific writing, CENTA, 2005

Presentation Skills, TIFN /Bob de Groof, 2005

Debating course, TIFN/Debat en dialoog, 2006

Career assessment, Meijaard en Meijaard, 2006

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