Pasta as an example for structure and dynamics of carbohydrate rich food materials

A literature review

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Contents

1. Introduction .................................................. 7

I. PASTA: FROM RAW MATERIAL TO PRODUCT ............... 9

2. Material ....................................................... 10
   2.1. Durum wheat components .................................. 10
       2.1.1. Starch .............................................. 12
       2.1.2. Protein ........................................... 15
       2.1.3. Interaction of starch and gluten .............. 17
       2.1.4. Non-starch polysaccharides/ Dietary fibres ... 19
       2.1.5. Minor components: lipids, colour pigments, ash .... 21
   2.2. Alternatives to durum wheat and non-traditional ingredients .... 22

3. Processing ...................................................... 26
   3.1. Durum wheat processing .................................. 28
   3.2. Pasta production ......................................... 28
       3.2.1. Influence of processing on selected parameters .... 32
   3.3. Pasta cooking and post-cooking .......................... 32
       3.3.1. Influence of cooking conditions ................... 33

II. METHODS ....................................................... 35

4. Introduction Part II Methods .................................. 36

5. Analysing the microstructure ................................ 37
   5.1. Microscopy ............................................... 37
       5.1.1. Light microscopy (LM) ............................. 37
       5.1.2. Confocal laser scanning microscopy (CLSM) ... 40
       5.1.3. X-ray computed tomography (XRT) ............. 42
       5.1.4. Scanning electron microscopy (SEM) ........... 42
       5.1.5. Transmission electron microscopy (TEM) ....... 44
       5.1.6. Atomic force microscopy (AFM) .................. 44
       5.1.7. Image analysis ................................... 45
   5.2. Other methods ............................................ 45

6. Analysing the macrostructure ................................ 47
   6.1. Rheological methods ..................................... 47
       6.1.1. Rheological properties of flours and doughs .... 47
       Rapid viscoanalyser (RVA) ............................ 48
       Falling number ......................................... 48
       Farinograph and amylograph .......................... 48
       Alveograph ............................................. 48


| 6.1.2. Rheological properties of gluten | 48 |
| 6.1.3. Texture analysis | 49 |
| 6.2. Thermal analysis | 49 |
| 6.2.1. Differential scanning calorimetry (DSC) | 49 |
| 6.2.2. Dynamic mechanical analysis (DMA) | 50 |
| 6.3. Other methods | 50 |

| 7. Analysing water transport and mobility | 51 |
| 7.1. Water transport and its modelling | 51 |
| 7.1.1. Water transport mechanisms | 51 |
| 7.1.2. Theoretical kinetic models of water transport | 52 |
| 7.1.3. Kinetic models applied to pasta | 53 |
| Mixing and drying | 53 |
| Rehydration process | 53 |
| 7.2. Nuclear magnetic resonance and magnetic resonance imaging | 56 |
| 7.3. Near-infrared reflectance spectroscopy (NIR) | 59 |
| 7.4. Hyperspectral imaging | 59 |
| 7.5. Gravimetrical analysis | 60 |
| 7.6. Other methods | 60 |

| 8. Analysing quality parameters | 61 |
| 8.1. Colour | 62 |
| 8.2. Texture | 62 |
| 8.3. Composition analysis | 65 |

| 9. Concluding remarks | 66 |

Bibliography 67
### Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>AACC</td>
<td>American Association of Cereal Chemists</td>
</tr>
<tr>
<td>AFM</td>
<td>Atomic force microscopy</td>
</tr>
<tr>
<td>AX</td>
<td>Arabinoxylans</td>
</tr>
<tr>
<td>BF</td>
<td>Bright field</td>
</tr>
<tr>
<td>CLSM</td>
<td>Confocal laser scanning microscopy (also known as CSLM)</td>
</tr>
<tr>
<td>DMA</td>
<td>Dynamic mechanical analysis</td>
</tr>
<tr>
<td>DIC</td>
<td>Differential interference contrast microscopy</td>
</tr>
<tr>
<td>DSC</td>
<td>Differential Scanning Calorimetry</td>
</tr>
<tr>
<td>HMW-GS</td>
<td>High molecular weight glutenin subunits</td>
</tr>
<tr>
<td>HT</td>
<td>High temperature</td>
</tr>
<tr>
<td>LMW-GS</td>
<td>Low molecular weight glutenin subunits</td>
</tr>
<tr>
<td>LM</td>
<td>Light microscopy</td>
</tr>
<tr>
<td>LOX</td>
<td>Lipoxygenase</td>
</tr>
<tr>
<td>LT</td>
<td>Low temperature</td>
</tr>
<tr>
<td>MRI</td>
<td>Magnetic resonance imaging</td>
</tr>
<tr>
<td>NIR</td>
<td>Near-infrared reflectance</td>
</tr>
<tr>
<td>NMR</td>
<td>Nuclear magnetic resonance</td>
</tr>
<tr>
<td>OCT</td>
<td>Optimal cooking time</td>
</tr>
<tr>
<td>QD</td>
<td>Quantum dots</td>
</tr>
<tr>
<td>RS</td>
<td>Resistant starch</td>
</tr>
<tr>
<td>SEM</td>
<td>Scanning electron microscopy</td>
</tr>
<tr>
<td>TEM</td>
<td>Transmission electron microscopy</td>
</tr>
<tr>
<td>TPA</td>
<td>Texture profile analysis</td>
</tr>
<tr>
<td>VHT</td>
<td>Very high temperature</td>
</tr>
<tr>
<td>WE-AX</td>
<td>Water extractable arabinoxylans</td>
</tr>
<tr>
<td>WU-AX</td>
<td>Water unextractable arabinoxylans</td>
</tr>
<tr>
<td>XRT</td>
<td>X-ray tomography (also known as microtomography, μ-CT)</td>
</tr>
</tbody>
</table>
1. Introduction

Pasta

Nowadays, pasta is an universal food mainly made from wheat, but also from rice and other cereals. Pasta has a century-old history with roots in China and Italy, however, the industrial revolution did not start before the 1950s (De Vita, 2009). Even in Italy the consumption of pasta rose first after this time, from being a food for feast days to now everyday use. The consumption rose to top today about 25 kg per capita and year. For comparison: In Sweden 9 kg per capita and year are consumed. Pasta is offered in manifold ways - as fresh pasta, instant pasta, dried pasta or as noodles.

However, it is still not completely understood how the cooking performance and textural properties of pasta are formed and influenced by every step of the production chain - starting from the choice of the raw material over the production itself to end with how to keep the product warm after cooking.

Objective

The main aim of this PhD project is to study the interdependence of raw materials, process conditions, structure and final quality properties of carbohydrate-rich food systems with the emphasis on pasta products.

The vision of the project is in the long run to study which microstructure properties are

- Important for properties in pasta in ready-to-eat meals
- Needed for a pasta which can be prepared in the microwave without prior cooking
- Needed for a pasta with a high level of dietary fibres and wholegrain

The main goal is to control the pasta quality by choice of raw material and components, how the process can be adjusted depending on quality demands, as well as to create structures starting from certain raw material conditions.

- To design microstructures in order to create desired texture properties
- To control mass transport at different heating profiles
- To design composite material with defined quality properties

This literature review shall sum up the current knowledge on (durum) wheat and dietary fibre as a raw material, the manufacturing process as well as on modifications of the raw material or process to improve the quality of pasta products. Furthermore, methods to analyse certain parameters such as microstructure and water migration shall be described as well.

\[^1\text{http://www.internationalpasta.org/index.php?cat=22&item=7&lang=2, accessed 02/2012.}^1\]
Part I.

PASTA: FROM RAW MATERIAL TO PRODUCT
2. Material

Generally, dried pasta is produced by mixing milled durum wheat, water and optional ingredients such as eggs, tomato, spinach or other functional ingredients (Cubadda, 1993). However, in several countries some restrictions apply. For instance, it is only allowed to use durum wheat for dried pasta sold in Italy, France or Greece; to use common wheats is considered to be an adulteration (for fresh pasta, however, it is allowed; Peressini et al., 2000; Sissons, 2008).

Kratzer (2007) defines pasta processing from a material science point of view as a transformation of a "cell structured natural composite of the major components starch and protein and minor constituents [...] into a biopolymer composite material consisting of a continuous protein phase with starch granules as a filler." The major components shall be described in this chapter. The composition of starch and protein are described in individual subsections. As they are often analysed at the same time, their influence on pasta quality is described in a subsequent subsection.

2.1. Durum wheat components

On a global scale, bread or common wheats (Triticum aestivum) are the most common cultivated wheat crops with a share of about 95% (Fuad and Prabhasankar, 2010). Pasta can be made of common wheat, however the preferred crop are durum wheats (Triticum durum) (Cubadda, 1993). From the wheat crop, only the seeds - the kernels or grain - are used for food production. The main parts of a grain are the bran, the starchy endosperm and the germ (Figure 2.1A shows a schematic illustration of a common wheat grain; deviant to the illustration durum wheat has no hairs of brush).

The bran consists of several protective cell layers from the pericarp (epidermis, hypodermis, endocarp), testa, nucellar and aleurone layers (Kill and Turnbull, 2001; Figure 2.1 B+C). The endosperm consists mainly of starch granules and protein bodies, which are grouped in a cellular structure surrounded by thin cell walls. It is the energy storage of the grain. The share of starch increases within the endosperm from the outer regions of the subaleurone layer to the central region. The germ is the embryo of the grain and thus is rich in vitamins, minerals, antioxidants and dietary fibre (Manthey and Schorno, 2002).

To achieve homogeneous flours only the endosperm is used and thus bran and germ are often removed during milling. However, they are used in speciality products such as whole-grain food products (Manthey and Schorno, 2002). The remaining endosperm is milled to flour and for common and durum wheat these flours are called farina and semolina, respectively. There compositions are given in Table 2.1. The numbers shown represent only an average, as quality characteristics vary within common and durum wheat types, especially concerning protein content. In general, only varieties of common wheat with a rather high protein content can be used for pasta making (Cubadda, 1993). Alternatives to durum wheats are mentioned in Chapter 2.2. Several methods have been developed to assess the pasta making quality of grains which include visual appearance, defects or the weight of 1000 kernels and after milling parameters such as ash content, speck count, colour or particle size distribution besides the analysis of the quantity of the major components (Sissons, 2008).
Figure 2.1.: Structure elements of a common wheat grain. (A) illustration of a grain, (B) light micrograph of an embedded section (length of scale bar is unknown), (C) fluorescence micrograph from bran layers. Figures reproduced from Slavin et al. (2000), Autio and Salmenkalliomarttila (2001) and Poutanen (2012).
Table 2.1.: Selected representative properties of durum wheat (semolina) and common wheat (farina) flours

<table>
<thead>
<tr>
<th>Unit</th>
<th>Durum wheat</th>
<th>Common wheat</th>
<th>Sources</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carbohydrates [%]</td>
<td>72.8</td>
<td>78.0</td>
<td>Anonymous (2010)</td>
</tr>
<tr>
<td>Damaged starch [%]</td>
<td>11-12</td>
<td>7-10</td>
<td>Eliasson and Larsson (1999)</td>
</tr>
<tr>
<td>Amylose [%]</td>
<td>26-28</td>
<td>23</td>
<td>Vansteelandt and Delcour (1999); Hesk and Lee (2003)</td>
</tr>
<tr>
<td>[%]</td>
<td>0-40</td>
<td></td>
<td>Sissons (2008)</td>
</tr>
<tr>
<td>Protein [%]</td>
<td>12.7</td>
<td>10.6</td>
<td>Anonymous (2010)</td>
</tr>
<tr>
<td>[%]</td>
<td>11-16</td>
<td></td>
<td>Kill and Turnbull (2001)</td>
</tr>
<tr>
<td>Gluten proteins [%]</td>
<td>0.6-0.86</td>
<td>(60)-80</td>
<td>Gil-Humanes et al. (2011)</td>
</tr>
<tr>
<td>Gliadin:Glutenin</td>
<td></td>
<td>0.72</td>
<td>Rao et al. (2001); Aravind et al. (2011); Létang et al. (1999)</td>
</tr>
<tr>
<td>Water [%]</td>
<td>12.7</td>
<td>10.5</td>
<td>Anonymous (2010)</td>
</tr>
<tr>
<td>Dietary fibre [%]</td>
<td>3.9</td>
<td>1.9</td>
<td>Anonymous (2010)</td>
</tr>
<tr>
<td>Lipids [%]</td>
<td>1.0</td>
<td>0.5</td>
<td>Anonymous (2010)</td>
</tr>
<tr>
<td>Ash [%]</td>
<td>0.8</td>
<td>0.4</td>
<td>Anonymous (2010); Eliasson and Larsson (1999)</td>
</tr>
<tr>
<td>Kernel size length [mm]</td>
<td>7</td>
<td></td>
<td>Troccoli et al. (2000)</td>
</tr>
<tr>
<td>Kernel size perimeter [mm]</td>
<td>15</td>
<td></td>
<td>Troccoli et al. (2000)</td>
</tr>
<tr>
<td>Granule B-type [µm]</td>
<td>3-5</td>
<td>2-3</td>
<td>Soh et al. (2006), Pérez and Bertoft (2010)</td>
</tr>
<tr>
<td>Granule C-type [µm]</td>
<td>&lt; 1</td>
<td></td>
<td>Wilson et al. (2006)</td>
</tr>
</tbody>
</table>

1 Percentage of specific major component

2.1.1. Starch

Granular organisation

On the nanoscale, starch is composed of the two polysaccharides amyllose and amyllopectin and are both based on the monomer (1->4) α-D-glucans. These two polysaccharides are synthesized in granular form, leading to an ultrastructure of starch existing over several length scales (from nm to up to 40 µm) as it is illustrated in Figure 2.2 (Pérez and Bertoft, 2010). They are formed during biological synthesis together with non-starch molecules such as phosphates and lipids (Conde-Petit, 2003; Tang et al., 2006).

Amylose is a long-chain molecule with a molecular weight of 10^5 to 10^6, which is not or only slightly branched and has partly a helical and double-helical structure (Delcour et al., 2010). The highly branched amyllopectin (1->6 linkages) with a high molecular weight of 10^7 to 10^8, in contrast, is constituted in major parts of crystalline and amorphous lamellae (length of about 10 nm, Figure 2.2B, Tang et al., 2006). The crystalline lamellae consists of the double-helical side chains of the amyllopectin, whereas the amorphous lamellae are the branching zones of amyllopectin. The extent of crystallinity is dependent on the amount of amyllose and is ranging between 15 % for high-amyllose starches and 40-50 % for waxy, i.e. non-amyllose starches (Copeland et al., 2009). It is also dependent on moisture content, with an observed maximum of crystallinity at 27 % (Blazek, 2008). The crystallinity is based upon the close arrangement of six or seven double-helices. Two types of crystallinity structure patterns can be distinguished in wheat starch, as it can be analysed by X-ray diffraction (Figure 2.2A and B, Blazek, 2008; Tang et al., 2006). Type A has more structured double-helices than type B and differs also in the chain length of amyllopectin.
and in the amount of structural water within the crystal. Wheat as other cereals tend to have a crystallinity pattern of type A (Copeland et al., 2009). The crystallinity of native starch granules can be shown with polarized light, as the crystallinity leads to birefringence, and that can be seen as a so called Maltese cross (Pérez and Bertoft, 2010).

The next higher level of structure are blocklets, which are formed of several lamellae amyllopectin molecules (length up to 120 nm) (Gallant et al., 1997). These ordered structures are interrupted both by amylose, which is mostly surrounding the blocklets and by slightly branched amylose-like (LC) amyllopectin. The LC amyllopectin molecules (up to 13 % of all amyllopectin) can have a length of several blocklets (Tang et al., 2006). According to the same authors, these semi-crystalline blocklets are the basic unit in starch granule formation and exist in two forms, 'normal' and 'defective' blocklets. They can be correlated to the highest level of starch organisation within the granule, the observed growth rings. These growth rings are now seen as being a none-continuous structure with alternating hard and soft shells, consisting of crystalline and semi-crystalline regions of a size of 120-400 nm (Blazek, 2008; Pérez and Bertoft, 2010). The hard shells are formed of normal blocklets (common wheat: 100 nm wide) and the soft shells of the defective blocklets (25 nm wide) (Pérez and Bertoft, 2010). The defective organization comes from non-branching amyllopectin molecules (Tang et al., 2006). These zones of defective blocklets could be seen as a correspondent to the amorphous channel concept of Gallant et al. (1997). For common wheat, the existence of channels has been demonstrated using CLSM (Kim and Huber, 2008). Channels were present in type-A- and type-B-granules (definition: see next subsection) and in waxy and normal common wheat starches. Hydration ceased the visualisation of the channels. For durum wheat starch, channels were yet not presented. Their existence can be assumed, however, as during starch heating amylose is leaking within the granule both to the surface and the inner groove region (shown in pasta by Heneen and Brismar, 2003).

Blocklets have yet not been extracted and characterised, but there is evidence for a level of structure between the growth rings and amyllopectin lamellae and blocklets are offering a valuable model for the position of amylose in the granules (Pérez and Bertoft, 2010). Within the granule, amylose is randomly distributed and radial orientated. Its content is richer at the periphery of the granules. A smaller part of amylose may be involved in lipid complexes and there exists larger amylose molecules as well, which are non-leachable and which are thought to interact with the double-helical zones of amyllopectin (Pérez and Bertoft, 2010). The aforementioned starch-lipid complexes consists of helical amylose including fatty acids, monoglycerides or linear alcohols, while amyllopectin do probably form these complexes only weakly or not at all (Blazek, 2008).

A further structural element is currently disputed: On the surface of the granules, pores might be visible and be connected to the amorphous channels. Tang et al. (2006) call the pores artefacts, while Pérez and Bertoft (2010) claim their existence. Furthermore, there might be a correlation between the number of pores and the digestibility of the starch granule (Pérez and Bertoft, 2010). Undisputed are the components on the starch granule surface. Surface lipids (free lipids and starch-amylose complexes) and proteins can moderate the starch functionality (Blazek, 2008). Delcour et al. (2000a) demonstrated, that the removal of surface proteins led to accelerated hydration of starch granules.

Kernel organisation

Starch granules are organised within the native kernel in a cellular structure. Wheat starch is commonly said to have a bimodal distribution with larger oval type-A-granules and smaller round type-B-granules (Soh et al., 2006). Some researchers claim that there exists a third category of type-C-granules, which are even smaller than type B (Wilson
Figure 2.2: (A) Starch organisation over several length scales – from granular to molecular level (reproduced from Gallant et al., 1997).

(B) Detailed scheme of the organisation of amyllopectin, amylose, lipids and minor components in blocklets (reproduced from Tang et al., 2006).
C-granules make up to 80% of all granules when counted in numbers, but are negligible when counted as volume percentage. The volume share of A- and B-granules in durum wheat is according to the measurements done by Wilson et al. (2006) between 80-90% A-granules and 10-20% B-granules, respectively. While A-granules start to grow soon after the anthesis, B- and C-granules start to grow later. The average chain length of amylopectin molecules is shorter in B-granules than in A-granules (Copeland et al., 2009). Changing the ratio of A:B granules towards more B-granules results in higher and faster water absorption, because B-granules have a higher surface to volume ratio (Soh et al., 2006). According to a summary of Soh et al. (2006), it is not fully clear whether a change in the content of A- and B-granules leads to a change in the content of amylose in total. Some studies stated that A-granules compared to B-granules contained 4-10% more amylose, while other studies could not detect significant differences. Probably caused by a higher lipid content in B-granules, the gelation temperature is higher in B- than in A-granules (Delcour et al., 2010).

Comparison of durum wheat starch and common wheat starch

Zweifel (2001) summarised the outcome of several publications, which studied the differences between starches from durum wheat and common wheat (see also Table 2.1). In durum wheat starch the amylose content is slightly higher than in common wheat starch, and shows a lower gelatinisation temperature (Delcour et al., 2010; Vansteelandt and Delcour, 1999). Furthermore, durum wheat starch has a higher water binding capacity. Zweifel (2001) concluded that these data suggests a less compact starch granule structure in durum wheat. However, durum wheat lacks a certain protein (puroindoline), which induces a stronger interaction of proteins with the starch granule surface and is making the kernels very hard (Delcour et al., 2010). This may be the reason, that starch damage during milling is higher in durum wheat than in common wheat. Compared to common wheat, especially the type-A-granules of durum wheat are smaller and the volume share of them are higher (Wilson et al., 2006). Finally, according to Vansteelandt and Delcour (1999), durum wheat starch and common wheat starch show about the same protein content, but differ in lipid content.

2.1.2. Protein

The heterogeneous wheat protein is classified according to extractability into four fractions: water soluble albumins, globulin (soluble in salt solution), gliadins (soluble in ethanol) and glutenins (soluble in dilute acids, Troccoli et al., 2000). Albumins and globulins represent the minor part (15-20%) and have emulsifying properties. The major part of wheat proteins consists of gliadin and glutenin Delcour et al. (2012). The latter two proteins form the composite gluten, which is seen as probably being the most important factor in determining pasta quality as they can form a viscoelastic network.

Glutenin is further separated into high and low molecular weight subunits (HMW-GS and LMW-GS, respectively). LMW-GS has a ratio of 60-80% of all glutenins and its molecular mass is in the magnitude of 3-5*10^5 (Sissons, 2008). The equivalent value for HMW-GS is 8-12*10^5, while the molecular mass of glutenin can exceed 10^6. This indicates that glutenin is a huge polymer, bonding together the subunits by intermolecular disulphide bonds and it is formed in a helical structure (Sissons, 2008; Zweifel, 2001).

Gliadins (MW 2.5-7.5*10^5) are classified based on their electrophoretic mobility into α-, β-, γ- and δ-gliadins (Kontogiorgos, 2011; Sissons, 2008). The monomeric Gliadins contain only intra disulphide bonds and the interaction with the gluten polymer is happening via non-covalent forces (Sissons, 2008).
In the dry grain the gluten is accumulated in protein bodies (Figure 2.3A) which are quantitatively and qualitatively unevenly distributed in the kernel (Tosi et al., 2011; Gil-Humanes et al., 2011). Protein concentration is higher in the sub-aleurone cells and lower in the central starchy endosperm cells. Furthermore, two types of protein bodies (small and large) are formed in the starchy endosperm. Small bodies are rich in gliadins and LMW-GS whereas large protein bodies are rich in HMW-GS (Tosi et al., 2011). Some protein bodies can even form a continuous matrix enclosing starch granules.

Hydrated gluten forms a continuous network. Kontogiorgos (2011) suggest a new hierarchical model for the structure of the hydrated gluten (Figure 2.3 B). At molecular level individual glutenins and gliadins are interacting via various physical and covalent forces. Depending on the conditions of e.g. ratio of protein fractions and hydration status, there happens a transition from fibrils to a continuous phase where gluten polymers form sheets. These resulting sheets can be seen as the building block of the gluten network and it aggregates may be embedded. The side-by-side arrangement of the sheets gives a nanoporous structure with “confined” water entrapped into the sheets and interacting strongly with the gluten matrix and “bulk” water surrounding it. Between the sheets nanocapillaries are formed. Depending on the packing of the sheets, a three-dimensional network is formed. Finally, the gluten network on the macroscale can show various morphologies.

Speaking of rheological properties, glutenin is responsible for elastic properties in a dough, while gliadin acts as a plasticizer being responsible for viscous properties, such as dough extensibility (Sissons et al., 2007). The term gluten strength is a measure for the balance between the two gluten subfractions, i.e. between viscosity and elasticity (Sissons, 2008). Gluten strength can be measured with the gluten index method and a higher gluten index indicates a stronger gluten (see also Chapter 6.1.2). There seems to be a tendency for a higher share of glutenin in stronger gluten varieties (Rao et al., 2001).
2.1.3. Interaction of starch and gluten

Hydrothermal induced changes: Glass transition and gelatinisation/denaturation

In its native state and at room temperature, both starch and gluten are in a glassy state and are organised in crystalline structure. They are very limited in their water uptake with a decreasing solubility for the starch components from amylopectin, to amylose and amylose-lipid complexes (Conde-Petit, 2003). Depending on temperature and water content starch and gluten can change from a glassy state to a rubbery state reversibly (Figure 2.4) via a so called glass transition. A second, irreversible transformation can occur at higher temperatures: Starch gelatinisation and protein denaturation.

Starch When sufficient moisture is available, native starch gelatinise during heating above the gelatinisation temperature, losing its crystallinity and structural organization (Copeland et al., 2009). At the same time there is a significant increase in molecular water mobility (Cuq et al., 2003). Water is absorbed first to the amorphous zones and if there are channels, water gets first to the inside and diffuses out from the inside (Copeland et al., 2009; Fannon et al., 2004). This hydration process can be quite fast, with a half time of 7s (Lemke et al., 2004). The loosely bound amylose molecules start to leach out of the granule. Starch molecules can swell to a certain amount, before they are disrupted, caused by the induced stress. Both glass transition and gelatinisation lead to a considerable increase in viscosity. Observing the melting process of starch using DSC measurements, an additional peak appears at higher temperatures than the peak corresponding to starch gelatinisation. This is attributed to the melting of amylose-lipid complexes (Conde-Petit, 2003, not shown in Figure 2.4). The shown temperature curves may in reality not be as exact as they are presented. Because of the heterogeneity of starch and gluten, the transitions are occurring in a certain temperature range (Cuq et al., 2003).

Gluten Through the addition of water, gluten transforms from the glassy state to become rubbery, elastic and is forming a network through inter-molecular bonds (Kill and Turnbull, 2001). Already at a water content of 15 %, the glass transition occurs below room temperature and hence gluten is in the rubber state at the conditions used for pasta dough preparation. At temperatures above 60 °C, again as a function of moisture, hydrated gluten is forming three-dimensional aggregates through the establishment of covalent bonds (protein-protein cross links; Sissons, 2008). This thermosetting is irreversible (Cuq et al., 2003). Gluten bond formation both during processing and cooking is described in more detail elsewhere (Wagner et al., 2011; Bock and Seetharaman, 2012).

In starch and gluten, the structural transformations occur at similar moisture and temperature conditions. Hence, the transformations are often competitive in regard to water. This can lead to an uneven water distribution among the components in pasta eg. during cooking (Petitot et al., 2009a). It is interesting to note, that while starch becomes soluble during gelation, gluten becomes during the network formation insoluble (Pagani et al., 2007). Therefore, the kinetics of these processes determine the extent of starch swelling.

Technological studies describing the effects of starch and gluten in pasta processing

To understand the influence of the wheat components on pasta quality, several authors carried out reconstitution studies. That means, they fractioned the semolina into gluten (or even several gluten fractions) and starch (variation in starch granule distribution) and combined the fractions in varying compositions together (Delcour et al., 2000a,b; Sissons et al., 2002). Sissons et al. (2007) altered the gluten composition and showed that glutenin
increased and gliadin decreased dough strength in a dough made of both reconstituted flour and semolina base. HMW-GS increased the dough strength of the base, while LMW-GS decreased it. The dough strength changes did not alter spaghetti texture, however.

An increased share of 32-44 % B-granules resulted in an improved pasta quality, with an increased firmness, reduced stickiness and reduced cooking loss (Soh et al., 2006). The dough strength decreased above a share of 32 % B-granules. The authors speculated, that too many B-granules would need too much water creating an imbalance in the dough in water distribution. Another study concentrated on the effect of starch digestion. For a share of 40 % B-granules, they found slightly lower starch digestion compared to the control (Ara vind et al., 2011).

To increase the amylose content, Soh et al. (2006) used high-amylose maize starch. Therefore, the results could be different, when using amylose of durum wheat instead. However, they showed that increased concentrations of amylose led to a more extensible dough, lower water uptake of the pasta and increased firmness. According to the authors, higher amounts of amylose lead to more tightly packed starch granules, which could be under swelling more resistant to deformation. They concluded that the decreased water uptake might change the sensory perception negatively. The observations on varying amylose content is in accordance with studies from Gianibelli et al. (2005) and Vignaux et al. (2005), which found inferior cooking properties for reduced amylose content. An explanation could be that starch granules of low amylose content deteriorate physically more easily and during cooling they form aggregates, but no network. This would result in a soft structure (Tan et al., 2006).

The reconstitution method in itself has some limitations, as through fractioning the material properties are changed. Sissons et al. (2002) showed, that the dough strength increased in a reconstituted pasta sample compared with an non-reconstituted sample.

Another approach was used by Wood (2009). Chickpea-fortified spaghetti was produced and compared with standard durum wheat spaghetti to study the mechanisms for pasta quality. Her findings were, that pasta firmness is influenced more by the composition and content of gluten than of protein content in total. Additionally, the protein-polysaccaride matrix rather than the starch composition is important for cooking loss, while cooking loss and stickiness do not necessarily correlate very strongly. Finally, increased protein and amylose contents decreased pasta stickiness.
### 2.1.4. Non-starch polysaccharides/ Dietary fibres

#### Classification of dietary fibres

Dietary fibre is a not well defined term according to Lunn and Buttriss (2007). It is a collective term for a mixture of substances differing in chemical and physical composition which all exert some kind of physiological effect. This physiological effect is mostly some sort of reduced digestibility by the human digestion. Dietary fibre includes soluble and insoluble fibres, but also resistant starch and additives such as guar gum (BeMiller, 2010). Dietary fibre are differentiated according to their solubility in a buffer at a defined pH or according to their fermentability in an in-vitro system representing human alimentary enzymes (Dhingra et al., 2011, see also Figure 2.5, Lunn and Buttriss, 2007). Insoluble fibres are cellulose, hemicellulose and the non-carbohydrate cell component lignin. Common soluble fibres are pectin, gums, mucilages, but also inulin and beta-glucan (Dhingra et al., 2011). Resistant starch (RS) is a category for undigested starches and starch-degraded products. There are four sources for RS: Physically inaccessible starch for amylase as it is entrapped in a food matrix (RS1), Starch granules resisting hydrolysis because of its nature (e.g. uncooked starch; RS2), retrograded starch (RS3) and modified starch inaccessible to enzymes (RS4) (BeMiller, 2010). For RS, an extended review on the effect of processing on the content of RS in cereals was written by Alsaar (2011).

Important physico-chemical properties of dietary fibres include particle size, surface area characteristics, porosity and hydration properties such as swelling and water retention capacity (Meuser, 2008). Also the structure of a dietary fibre will determine its properties. E.g. cellulose can bind rather low amounts of water (due to its fibril structure) whereas arabinoxylans have a high capacity to bind water. These physico-chemical properties are also dependent on environmental conditions (temperature, pH, ionic strength) and they are modified during processing (grinding, drying, heating, extrusion; Dhingra et al., 2011).

#### Dietary fibres in the wheat grain

Non-starch polysaccharides can be found in the wheat grain in the germ, the bran and in the cell walls of the endosperm (Sissons, 2008). The cell walls consist of cellulose, hemicellulose, lignin, and beta-glucan. A major component within hemicellulose are arabinoxylans (AX) which are divided into water extractable (WE-AX) and water unextractable (WU-AX) (BeMiller, 2010; Sissons, 2008). Wheat bran is an important example for the category of dietary fibre. It is often used as an ingredient in nutritional-value-added products as it is composed of about 48 % dietary fibre, 16 % starch, 18 % proteins, 5 % fat, 5 % sugars and 6 % ash (Meuser, 2008). Bran consists of several tissues with varying properties and the structure has been shown in more detail by Surget and Barron (2005).

![Non-digestible carbohydrate](image)

Figure 2.5.: Suggested overview to classify dietary fibre by Lunn and Buttriss (2007)
Technological studies describing the effects of various fibres in pasta processing

Due to its nutritional profile, there is a great interest to learn more about how wheat bran affects products. Only recently, several studies have examined the effects of bran and bran fractions being incorporated into pasta/noodles (Ara vind et al., 2012a; Chen et al., 2011; Chillo et al., 2008; Kaur et al., 2012; Shiau et al., 2012; Sudha et al., 2012; West et al., 2013a) or bread (doughs) (e.g. Almeida et al., 2013; Curti et al., 2013; Majzoobi et al., 2013). However, bran often induces a strong aroma (due to phenolic acids) and a changed texture with increased cooking loss and decreased firmness (West, 2012). What induces the texture change is not fully understood. Tudorica et al. (2002) argue for fresh pasta that soluble fibres are included into the protein network of the pasta, while insoluble fibres such as bran are disrupting it. In the case of dried pasta, SEM images showed that the bran particles where not in contact with the protein matrix and thus disrupted the network (Bustos et al., 2011a; Manthey and Schorno, 2002). Others argue that it might depend on the amount and type of fiber (germ particles destroyed the protein network to a larger extent than bran particles; Aravind et al., 2012a) and on the process conditions (Villeneuve and Gelinas, 2007). One recent study rejects the theory of gluten network destruction for the case of bread (Noort et al., 2010). Instead, the authors argue that the fibres interact physically or chemically with the gluten and thus hinder gluten aggregation.

Several approaches have been reported to counteract the deteriorating effect of bran inclusion. Heat treated bran (Sudha et al., 2011), wholegrain flour milled to rather large particle sizes (Gauthier et al., 2006) and pasta dried at high temperatures (West et al., 2013b) helped to maintain the desired texture. A review discusses in a broader context various techniques to increase the bioactive potential of wheat bran (e.g. through milling, fermentation or enzymes; Mateo Anson et al., 2012).

According to de Noni and Pagani (2010) it has yet not be shown, that individual enzymes influence the quality of pasta. However, added endoxylanases (EC 3.2.1.8) hydrolysed the xylan backbone in arabinoxylans and could thus convert some of the water unextractable AX to water extractable AX forms. High doses of the enzyme led to less leaching of soluble AX during pasta cooking in water and retaining thus a higher amount of soluble dietary fiber in the product (Ingelbrecht et al., 2001; Brijs et al., 2004). Increasing the amount of WE-AX, increased the water absorption, but the pasta texture was not influenced (Turner et al., 2008). During cooking of pasta, only minor amounts of AX were released into the cooking water, but this amount can increase during overcooking (Sissons, 2008).

Besides the addition of further ingredients to the dough, also technological variations can influence the amount and effect of dietary fibres. Coarsely and finely grounded dietary fibre-rich flours will show a different behaviour during rehydration (Gauthier et al., 2006; Meuser, 2008; Shiau et al., 2012). During thermal treatments such as cooking, fibre-protein complexes are formed in wheat bran increasing the amount of dietary fibre, too (Dhingra et al., 2011). Meuser (2008) reminds that only highly purified dietary fibres will have a light colour and will be free from an unpleasant taste. Unpurified fibres, instead, are often of yellow to brownish colour and have a bitter to characteristic flavour.

Extrusion can increase the water solubility of dietary fibre. Whether this has a significant effect on wheat bran fibre seems to depend on process conditions and material properties such as particle size (Robin et al., 2012).

Resistant starch is heat sensitive and its content might increase slightly during extrusion, however decreases significant during cooking of pasta (Gelencsér et al., 2010; Alsaafar, 2011). During storage, the RS content of food products can be modified by applying temperature cycles (Alsaafar, 2011). For a gelatinised starch, the temperature cycle between 4°C and 30°C led to the formation of amylopectin crystals resulting in a reduced digestibility.
Finally, also non-dietary fibre components can effect the digestibility: Amylose-lipid-complexes have been shown to have a lower digestibility. They occur not only naturally, but can also be formed during gelatinisation of starch when lipids are present (Alsaaffar, 2011).

2.1.5. Minor components: lipids, colour pigments, ash

Lipids exists in the wheat grain in two forms: Starch bound lipids, forming amylose-inclusion complexes and non-starch lipids, which can be divided into free and bound lipids (Sissons, 2008). 64% of all lipids in semolina are free lipids, which appear to be evenly distributed in the protein network. These free lipids in the endosperm are mainly triglycerides and other nonpolar lipids such as hydrocarbons, mono- and diglycerides as well as free fatty acids (Pomeranz, 1988). An extensive list of the individual types of wheat lipids can be found in a review on lipids in bread making (Pareyt et al., 2011).

Amylose within the amylose-lipid-complexes is prevented from leaching during gelatinisation and thus may reduce stickiness in starch (Blazek, 2008). According to Sissons (2008), not much is known about composition changes of lipids during pasta processing. However, it is known, that lipids interact during dough mixing especially with the gluten network and that a removal of all lipids led to a pasta with increased stickiness and cooking loss (Sissons, 2008).

The Colour pigments, giving pasta its yellow colour, are named carotenoids and are grouped into carotenes, unsaturated hydrocarbons and xanthophylls (major component is lutein; Sissons, 2008; Troccoli et al., 2000). Carotenoids are located in the outer layer of the kernel and to lesser extent in the endosperm. The insensitivity of the carotenoids in the pasta products is influenced by several factors such as storage time and process conditions, but the most important factor is the activity of the oxidative degradation by lipoxygenase (LOX) (Troccoli et al., 2000). LOX oxidises free fatty acids such as carotenes. Also the activity of peroxidase in wheat has been reported, leading to a brown colour (Troccoli et al., 2000) and peroxidase may also affect rheological properties (Pomeranz, 1988). According to the same authors, innumerab e other enzymes can be found in wheat, but which probably have no great impact in cereal processing. For instance, in pasta production, α-amylase is of minor importance (Sissons, 2008). Amylase is existing mainly in preharvest sprouted grain and even if this grain is used, the enzyme activity is further reduced during drying and cooking.

Another category of components is ash, the residual after combusting the wheat. Ash consists mainly of minerals and the ash content varies within the kernel, with significant higher amounts in the bran as in the endosperm (Troccoli et al., 2000). The ash content is most important in milling, as the content is correlated to semolina yield. Furthermore, ash content influences the colour of the semolina, with more ash giving a browner colour (Troccoli et al., 2000). The ash content is in general higher in durum wheat than in common wheat (Pagani et al., 2007).

They are no components in itself, but mycotoxins are toxic metabolites from fungi present in durum wheat grains, too. The content depends on the growing conditions and mycotoxins can mainly be found in the bran layer (Brera et al., 2013). Adequate cleaning, debranning and milling processes can reduce the mycotoxin content (Cheli et al., 2013). Drying and finally cooking of pasta further reduces the mycotoxin content (Brera et al., 2013).
2.2. Alternatives to durum wheat and non-traditional ingredients

The oldest enrichments were probably colouring plant material from tomato, spinach or carrots (Cubadda, 1993). Also the usage of egg has a long history. Nowadays, there are more widespread applications available. A list of tested supplemental flours is given in Table 2.2. This table also includes a short-list of available dietary fibres, as well as some recently tested enrichments. In the comprehensive review on non-traditional ingredients written by Fuad and Prabhasankar (2010), the authors included a chapter on additives such as emulsifiers, organic acids or alginate. As using additives in dried pasta is prohibited in Europe by law, they will not be covered in detail.

Three main drive forces could be identified within the research for supplements or replacements to durum wheat pasta:

- Substitute durum wheat with local available grains
- Increase nutritional value
- Decrease allergenicity, in particular replace gluten

In markets with a developed pasta production, such as the United States or the European Union, mainly durum wheat is used for pasta production. However, especially researchers in non-traditional pasta consuming countries such as Iran or India are working on finding alternatives to durum wheat to be able to use the local grown species, in most cases varieties of common wheat (e.g. Aalami and Leelavathi, 2008; Fuad and Prabhasankar, 2012; Heneen and Brismar, 2003; Jyotsna et al., 2004). In a recent study, common wheat and emmer were analysed for its feasibility for the production of pasta (Fuad and Prabhasankar, 2012). The usage of common wheat led to an inferior pasta quality, as it has been reported before (Heneen and Brismar, 2003). The cooking quality and sensory perception of emmer based pasta was comparable to durum pasta. Spelt wheat (*Triticum aestivum ssp. spelta*) is especially used in the ecological food market and is similar in its properties to durum wheat (Fuad and Prabhasankar, 2010). It is notable, that even for other grains like rice, experiments are carried out to test additives such as gluten or gum arabic to achieve texture properties comparable to pasta (Raina et al., 2005).

Another major goal is to increase the nutritional value of the pasta while maintaining good cooking properties. One possibility is to use flours with an optimized nutritional profile, which is often derived from other crops. An alternative is to add purified components such as fibres. In fact, most of the listed examples in Table 2.2 seek to improve the nutritional value by increasing protein content, decreasing the glycaemic response or even increasing the content of omega-3-fatty acids (Iafelice et al., 2008). Other approaches, not listed in the Table, include enrichments using kamut, sprout finger millet, amaranth seed or even shrimp meat. It seems, that almost every ingredient could be suitable. An extensive list of dietary fibre containing ingredients suitable for extruded cereal products including their respective fibre concentration can be found in the review of Robin et al. (2012).

Maybe of higher importance in history, egg pasta amounts nowadays for only a small part of the market, especially for dried pasta. Therefore, studies on the influence of egg on the quality of dried egg pasta are rare (Materazzi et al., 2008; Schreurs et al., 1986). Research on fresh egg pasta is more frequent (e.g. Akillioglu and Yalcin (2010); Alamprese et al. (2005, 2009); Fratianne et al. (2012); Nouviance et al. (2008); Zardetto and Dalla Rosa (2009); Hager et al. (2012a)). Egg has also been used as a supplemental for non-gluten pasta based on buckwheat starch (Alamprese et al., 2007) and on oat and teff flour (Hager et al., 2013).
Currently, there is not only a striving for pasta with higher nutritional value, but also allergens shall be decreased (Krishnan and Prabhasankar, 2012). One approach is to use lactobacilli to ferment pasta dough so that the non-tolerated gliadin fractions got hydrolysed (Di Cagno et al. 2005). Other studies evaluate the properties of amaranth, buckwheat, soy, maize and quinoa for the production of gluten-free pasta (Mastromatteo et al., 2011; Schoenlechner et al., 2010). In a recent and comprehensive review for gluten replacements in bread-making, further ingredients and technological adjustments such as high hydrostatic pressure are discussed (Zannini et al., 2012). For the case of pasta, Marti and Pagani (2013) reviewed different technologies to pre-treat flours. Petitet et al. (2009a) discuss the implications of modern process modifications on starch digestibility and allergenicity. As an example, very high temperature drying induces a higher protein aggregation, which may encapsulate starch granules stronger. This would give a lower glucose response during digestion.

Hager et al. (2012b) screened 33 commercial gluten-free pasta brands from several European countries. They recommend further product and process improvements, as the tested gluten-free pasta brands showed different colours and higher cooking loss as well as higher stickiness than durum wheat pasta.

Several of the before mentioned ingredients have made its way to the market, at least in the United States. Pszczola (2010) has written a market review for what he calls "better-for-you" pastas.
Table 2.2: List of selected flours and other non-traditional ingredients tested for pasta production

<table>
<thead>
<tr>
<th>Category</th>
<th>Component</th>
<th>Examples for (functional) properties</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheats</td>
<td>Whole meal of durum wheat</td>
<td>Lower firmness and higher cooking loss, however effect not so pronounced when pasta was dried at high temperature</td>
<td>Biarno et al. (2008); Mantleby and Schorno (2002)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Common wheat</td>
<td>Similar properties to durum could be achieved through additives</td>
<td>Fauel and Prabhurame (2012); Jaywena et al. (2004)</td>
</tr>
<tr>
<td></td>
<td>Spelt</td>
<td>Similar properties to durum</td>
<td>Mazzarelli et al. (2002)</td>
</tr>
<tr>
<td></td>
<td>Emmer</td>
<td>Similar properties to durum</td>
<td>Fauel and Prabhurame (2012)</td>
</tr>
<tr>
<td></td>
<td>Barley</td>
<td>Darker colour, higher fibre content, higher cooking loss</td>
<td>El-Fahmy et al. (2010)</td>
</tr>
<tr>
<td></td>
<td>Buckwheat</td>
<td>Gluten-free</td>
<td>Mantleby and Hall (2007); Versarco et al. (2011)</td>
</tr>
<tr>
<td>Other</td>
<td>Oat</td>
<td>Increase nutritional value</td>
<td>Chillo et al. (2009); De Pilli et al. (2013)</td>
</tr>
<tr>
<td>flours</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Lupin</td>
<td>Higher protein + fibre content, slight changes in cooking properties, stickier at high lupin amounts</td>
<td>Jaywena and Nasser-Abbas (2012)</td>
</tr>
<tr>
<td></td>
<td>Corn</td>
<td>Similar properties (if pregelatinised)</td>
<td>Pagani et al. (2007)</td>
</tr>
<tr>
<td></td>
<td>Flaxseed</td>
<td>Increase alpha-linolenic acid</td>
<td>Lee et al. (2003)</td>
</tr>
<tr>
<td></td>
<td>Amaranth</td>
<td>Increase nutritional value</td>
<td>Fiorica et al. (2013)</td>
</tr>
<tr>
<td></td>
<td>Split pea, faba bean</td>
<td>Minor changes in microstructure, improved nutritional value</td>
<td>Pedret et al. (2010)</td>
</tr>
<tr>
<td></td>
<td>Pea protein</td>
<td>Higher protein content</td>
<td>Mercier et al. (2011); Zhao et al. (2006)</td>
</tr>
<tr>
<td></td>
<td>Soy</td>
<td>Lower cooking loss, similar sensory characteristics</td>
<td>Biarno et al. (2011)</td>
</tr>
<tr>
<td>Insoluble</td>
<td>Wheat bran</td>
<td>&lt;15% similar sensory characteristics</td>
<td>Aravind et al. (2012a); Kaur et al. (2012); Shaivu et al. (2012)</td>
</tr>
<tr>
<td>fibres</td>
<td>Germ</td>
<td>Higher cooking loss, darker colour, similar overall acceptability</td>
<td>Tarzi et al. (2012)</td>
</tr>
<tr>
<td></td>
<td>Oat bran</td>
<td>Lower GI, negative cooking properties, reduced firmness at &gt;5%, &lt;5% similar sensory characteristics</td>
<td>Bustos et al. (2011b,a)</td>
</tr>
<tr>
<td></td>
<td>Hemi-/Cellulose</td>
<td>From bamboo fibre; decreased firmness</td>
<td>Beeman and Tuckeries (2007)</td>
</tr>
<tr>
<td></td>
<td>Pea fibre</td>
<td>Slightly decreased firmness, unchanged stickiness</td>
<td>Beeman and Tuckeries (2007); Tuckeries et al. (2002)</td>
</tr>
<tr>
<td>Soluble</td>
<td>Guar gum</td>
<td>Lower GI, high water absorption + cooking loss, softer + stickier, less yellow</td>
<td>Aravind et al. (2012c); Tuckeries et al. (2002)</td>
</tr>
<tr>
<td>fibres</td>
<td>Inulin</td>
<td>Lower GI, higher cooking loss, inulin was lost to cooking water</td>
<td>Aravind et al. (2012d); Beeman et al. (2004); Manno et al. (2009)</td>
</tr>
<tr>
<td></td>
<td>β-glucan</td>
<td>Depending on type of β-glucan rice in cooking loss and stickiness, softer pasta</td>
<td>Aravind et al. (2012b); Cleary and Beeman (2011); Chenery and Beeman (2000)</td>
</tr>
<tr>
<td></td>
<td>Carboxymethylcellulose (CMC)</td>
<td>Lower GI, comparable cooking properties</td>
<td>Aravind et al. (2012c); Chillo et al. (2000)</td>
</tr>
<tr>
<td></td>
<td>Hydroxypropyl cellulose (HPC, E463)</td>
<td>Improved taste, less stickiness, longer OCT</td>
<td>Majmoudi et al. (2011b)</td>
</tr>
<tr>
<td>Category</td>
<td>Component</td>
<td>Examples for (functional) properties</td>
<td>References</td>
</tr>
<tr>
<td>---------------</td>
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<td>-----------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Starches</td>
<td>Resistant starch, type II and IV</td>
<td>Lower GI, partly improved textural properties after cooking</td>
<td>Alsaar (2011); Bustos et al. (2011a); Sozer et al. (2008); Vernaza et al. (2012)</td>
</tr>
<tr>
<td></td>
<td>Pregelatinised starch</td>
<td>Acted as an structuring agent for non-durum wheat flours</td>
<td>Chiklo et al. (2009)</td>
</tr>
<tr>
<td>Other</td>
<td>Egg</td>
<td>Improved colour and taste</td>
<td>e.g. Fratini et al. (2012)</td>
</tr>
<tr>
<td>ingredients</td>
<td>Beef heart</td>
<td>Improved nutritional value, higher firmness</td>
<td>Dharmadhikari et al. (2009)</td>
</tr>
<tr>
<td></td>
<td>Blue-green algae</td>
<td>Green colour, lower cooking loss, higher firmness and swelling index, improved sensory characteristics</td>
<td>Zouari et al. (2011)</td>
</tr>
<tr>
<td></td>
<td>Broccoli powder</td>
<td>Significant swelling of broccoli particles, could be controlled with hydrocolloids</td>
<td>Silva et al. (2013)</td>
</tr>
<tr>
<td></td>
<td>Cassava</td>
<td>Increase nutritional value in gluten-free pasta</td>
<td>Piozzi et al. (2013)</td>
</tr>
<tr>
<td></td>
<td>Fatty acids</td>
<td>Similar properties at low concentrations, at higher concentrations affected sensory characteristics</td>
<td>Iafeles et al. (2008)</td>
</tr>
<tr>
<td></td>
<td>Fennugreek seed powder</td>
<td>Decreased cooking loss and stickiness, at high concentrations sensory characteristics affected</td>
<td>Jyothi et al. (2011)</td>
</tr>
<tr>
<td></td>
<td>Fermented pasta dough</td>
<td>Increase in vitamin B2</td>
<td>Capozzi et al. (2011)</td>
</tr>
<tr>
<td></td>
<td>Mustard protein isolate</td>
<td>Higher protein content, improved firmness, decreased cooking loss and stickiness</td>
<td>Aliacem Sedeghi and Bhagya (2008)</td>
</tr>
<tr>
<td></td>
<td>Peanut</td>
<td>Darker colour and higher cooking loss; effect reduced when hydrocolloids were used or when dried at high temperature</td>
<td>Howard et al. (2011)</td>
</tr>
<tr>
<td></td>
<td>Seaweed</td>
<td>Increase in fucoxanthin</td>
<td>Prabhauadar et al. (2010)</td>
</tr>
<tr>
<td></td>
<td>Transglutaminase</td>
<td>Reduced polymer solubility, increased cooking quality of poor durum wheat varieties</td>
<td>Tades et al. (2007); Aalam et al. (2008); Sioones et al. (2010)</td>
</tr>
<tr>
<td></td>
<td>Unripe banana flour</td>
<td>Higher water absorption and increased chewiness, darker colour</td>
<td>Agrawal et al. (2009); Krishnan and Prabhauadar (2010)</td>
</tr>
<tr>
<td></td>
<td>Vegetable puree</td>
<td>Decreased cooking loss; sensory characteristics negatively affected</td>
<td>Recha et al. (2013)</td>
</tr>
<tr>
<td></td>
<td>Whey protein concentrate</td>
<td>White colour, mashy strand quality, highly increased stickiness, sensory characteristics could be improved by using hydrocolloids</td>
<td>Prabhauadar et al. (2007)</td>
</tr>
</tbody>
</table>

1 In comparison to durum wheat pasta
3. Processing

Only a few process steps are necessary to produce pasta. However, it is crucial to execute them in a proper way to achieve a sufficient result. A flow chart illustrates the process including SEM images for some intermediate products (Figure 3.1). Additionally, reported tested variations for each unit operation are listed in Table 3.1.

Table 3.1.: Unit operations, important parameters and possible technique variations in pasta processing.

<table>
<thead>
<tr>
<th>Unit operation</th>
<th>Equipment/Parameter</th>
<th>Reported technique variations¹</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grain preparation</td>
<td>Cleaning</td>
<td></td>
<td>Kill and Turnbull (2001)</td>
</tr>
<tr>
<td>Milling</td>
<td>Tempering</td>
<td></td>
<td>Delcourt et al. (2010)</td>
</tr>
<tr>
<td></td>
<td>Particle size</td>
<td>&lt; 350 μm</td>
<td>Rubin (2007)</td>
</tr>
<tr>
<td>Mixing</td>
<td>Pre-mixing</td>
<td></td>
<td>Pagani et al. (2007)</td>
</tr>
<tr>
<td></td>
<td>Kneading</td>
<td>Single-screw, co-rotating twin-screw, conveyor belt</td>
<td>Pagani et al. (2007)</td>
</tr>
<tr>
<td></td>
<td>Vacuum quality</td>
<td></td>
<td>Carini et al. (2010); Leardi-Vernière and Feillet (1999)</td>
</tr>
<tr>
<td></td>
<td>Water temperature</td>
<td></td>
<td>Kill and Turnbull (2001)</td>
</tr>
<tr>
<td></td>
<td>Time</td>
<td>20 s -15 min</td>
<td>Ait Kaddour and Cuq (2011); Rubin (2007)</td>
</tr>
<tr>
<td>Extrusion</td>
<td>Dough movement</td>
<td>Cylinder-plunger, Single-screw, <strong>Double-screw</strong></td>
<td>Kratzer (2007)</td>
</tr>
<tr>
<td></td>
<td>Extruder speed</td>
<td></td>
<td>Wojtowicz and Mosiecki (2011)</td>
</tr>
<tr>
<td></td>
<td>Die material</td>
<td>Bronze, <strong>teflon</strong></td>
<td>Lucchini et al. (2008)</td>
</tr>
<tr>
<td></td>
<td>Shape size/form</td>
<td></td>
<td>Carini et al. (2009)</td>
</tr>
<tr>
<td></td>
<td>Re-extrusion</td>
<td></td>
<td>Chillo et al. (2010)</td>
</tr>
<tr>
<td></td>
<td>Temperature</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sheeting</td>
<td>Hot air</td>
<td><strong>Convection-heat forced</strong>, Vacuum drying, microwave</td>
<td>Beradi and Koda (2010); Altaf and Maslam (2005); De Fili et al. (2009)</td>
</tr>
<tr>
<td></td>
<td>Pre-treatment</td>
<td>Controlled pressure drop</td>
<td>Manche-Roumeq and Allaf (2005)</td>
</tr>
<tr>
<td></td>
<td>Temperature/Time</td>
<td>50-100°C</td>
<td>Cukic et al. (2007); Zweickel et al. (2003)</td>
</tr>
</tbody>
</table>

¹Bold text shows the current technological standard

Opposed to pasta, (asian) noodles are often based on common wheat flour instead of durum wheat. Certain types of noodles are made of mung bean starch only, combined with additives such as salts, fat or gums (Oh et al., 1983). In addition, the processing of noodles is more varied. After mixing, the noodles are usually sheeted and cut followed by to be either dried, fried, steamed or directly boiled directly. In the case of steaming, the intermediate product can again be either dried, frozen or fried. The steaming process is especially used to produce instant noodles (Oh et al., 1983).
Figure 3.1.: Flow chart of the processing of pasta. SEM micrographs show (I) semolina, (II) extruded pasta, (III) the surface and (IV) cross section of dried spaghetti at high temperature. Images were assembled by Petitot et al. (2009a).
Still, in the next section the description is concentrated on the processes on for dry pasta production as dry pasta is by far the most sold type of pasta product in Europe (Serventi and Sabban, 2002). The production process of durum wheat based pasta is reviewed in great detail by Kill and Turnbull (2001) and Rubin (2007), but shall be briefly described in the next sections.

3.1. Durum wheat processing

The aim of the milling process is to separate the endosperm from the germ and the bran of the wheat kernel to get a maximum of semolina at the best quality (Kill and Turnbull, 2001). Milling comprises therefore the following steps: Cleaning of the wheat, tempering (or debranning) to loosen the bran layers as well as milling and purifying to achieve semolina of the right particle size.

The cleaning process is - besides checking the grain at the delivery - a measure to ensure product integrity and is used to remove impurities such as sticks, stones, metals or straw as well as seeds, other grains and broken or infected kernels. Between a first and a second cleaning, water is added to adjust the kernel to the right hardness, moisture content and to loosen the bran layers before milling (Kill and Turnbull, 2001).

The milling process is a combination of grinding, sifting and blending to reduce milling passages. To grind preprocessed kernels, they are led over several rolls which are categorised according to their roughness into breakers, reduction rolls, steel screens and purifiers (Cubadda et al., 2009). The breakers open the kernels and shall separate the carbohydrate-rich endosperm from bran and germ. The reduction rolls are mainly used to adjust the particle size whereas a larger number of purifiers is used to remove remaining bran particles (Delcour et al., 2010).

Traditionally, coarse semolina was used for pasta production with a particle size from 200 µm to 630 µm (Kill and Turnbull, 2001). A trend can be seen towards smaller particle size to reduce hydration time as smaller particles absorb water faster than bigger particles. Due to the hardness of the durum wheat kernels the amount of starch damage is higher compared to other wheats and is increasing with smaller particle size fractions and the ash content is increasing as well (Delcour et al., 2010; Feillet et al., 2000). Whether this has negative effects on pasta quality, was disputed in recent years (Sissons, 2008). Older studies showed, however, that the amount of starch damage correlated to the amount of cooking loss and gave poor surface characteristics (referred to by Dexter and Marchylo, 2000).

3.2. Pasta production

Pasta is produced by three main steps: Semolina is polymerised during mixing, the material is compacted and formed by means of sheeting or extruding and this structure is stabilised by drying (Kratzer, 2007). The impact of extrusion and drying on the intermediate products are visualised by SEM images (Figure 3.1; Petitot et al., 2009a). Semolina shows a compact structure with distinct starch granules and few of them are entrapped into a protein matrix. After extrusion, starch granules are embedded into a protein matrix and are aligned along the flow direction. Some granules might be slightly swollen. After drying, starch granules are deeply embedded into the protein matrix in the pasta strand while they are associated with the protein film at the surface. Some cracks and small holes are visible in the protein film at the surface, too, probably due to shrinking and tensions during drying.
Figure 3.2: Microstructure of durum pasta dough before and after various types of extrusion. (A) unprocessed endosperm, (B) cylinder-plunger system (C) single-screw extruder (D) double-screw extruder. Reproduced from Kratzer (2007)

Light microscopy was also used to show the change in microstructure during extrusion (Kratzer, 2007; Zweifel et al., 2003). The organisation of starch granules into cellular structures and the cell walls themselves are clearly noticeable in semolina (Figure 3.2A), while the cellular structure is lost after extrusion (Figure 3.2B-D). Depending on the type of extruder, starch granules are more or less compactly organised and cell walls cannot be seen any more.

Mixing Commonly, semolina particles and water are pre-mixed at high-speed to ensure a homogeneous particle wetting (Kill and Turnbull, 2001). Afterwards the mixture is kneaded for some minutes at lower speed to form a dough and especially to form the gluten network. To achieve a homogeneous dough a narrow particle size distribution is necessary to reduce the risk of the formation of non-wetted particles (white spots) (Kill and Turnbull, 2001). A smaller particle size is preferred to reduce the mixing time. Semolina with a particle size below 250 µm can be mixed in 5 min compared to the coarse semolina which needs 15 min to mix (Rubin, 2007). An alternative, integrated mixing system was introduced some years ago which uses co-rotating twin-screws, leading to intensified mixing and kneading. The mixing time could be reduced to 20 sec (Kill and Turnbull, 2001).

Vacuum in the mixing zone prevents oxidation of semolina natural pigments and the intrusion of air bubbles, which gives a better shine of the pasta product (Pagani et al., 2007). Additionally, temperature should be controlled during dough preparation with an optimum temperature between 35 and 40°C (Kill and Turnbull, 2001).

Extrusion The formed dough is moved under kneading towards the extrusion zone. The dough is compacted and pressure is built up. The pressure depends on dough moisture and temperature as well as on die resistance which in turn depends on the extrusion area and speed (Kill and Turnbull, 2001).

Extrusion can increase the water solubility of dietary fibre and solubility increases with higher specific mechanical energies (Robin et al., 2012). However, stickiness increases with higher energy input during extrusion, too (Kratzer, 2007). Thus, the energy input should be just high enough to form an homogeneous pasta dough (Kratzer, 2007).
Temperature should be kept below 50°C to prevent gluten aggregation already in the extruder as this network would be destroyed by the high shear forces at the die. In general, shear forces (e.g. from worn dies) should be kept at a minimum, as they increase damage of starch granules (Petitot et al., 2009a).

Commonly two types of dies are available: Bronze and teflon. While teflon dies produce a smooth texture and even surface, the surface of pasta extruded through bronze dies is rough. The effect was quantified by Lucisano et al. (2008). The bronze die extruded spaghetti had a higher porosity and a reduced breaking strength of the dried spaghetti of 20-30%. The type of drying cycle and semolina particle size distribution had an influence as well, but to a minor extent.

The type of shaping influences the cooking properties (shown for fresh pasta). Laminated products were more yellow and had a lower cooking loss than extruded products Carini et al. (2009). The authors assumed that the milder conditions during lamination led to a softer, less extensible material which retained solids in a better way.

Sissons (2008) discusses the influence of the type of gluten on the extrusion. Strong gluten wheats lead to less sticky doughs facilitating the extrusion. This is specially important for thin-walled instant pasta. Fresh pasta, however, needs a more extensible dough and thus a weaker gluten quality.

The quality of non-durum based pasta can often be increased by using pre-gelatinised starch. Multiple re-extrusion of the dough can be one way to increase the degree of dough gelatinisation. This was tested for amaranth, quinoa and oat flours (Chillo et al., 2010). The degree of dough gelatinisation did in fact increase for oat and quinoa dough, but not for amaranth. With increasing re-extrusion number, the breaking strength of the dry dough samples was increased. The colour of the samples were improved, but overall sensorial characteristics of the cooked spaghetti samples remained the same.

**Drying** Drying shall reduce the moisture content from roughly 30% to below 12.5% to make pasta a stable product (Pagani et al., 2007). The local water concentration in the dough differs during drying, which can induce internal stress, potentially leading to fractures and cracks (Migliori et al., 2005). Drying is therefore seen as the critical step during processing. In a very recent study, Zhang et al. (2013) showed that spaghetti shrank faster in radial than in axial direction during drying in an experimental drying chamber. Additionally, the spaghetti shrank torsional, non-central directed in the beginning and linear, central directed later (Figure 3.3). These findings may explain the arising of internal stress and cracks.
Drying profiles including phases of high temperatures shall lead to higher product quality and can generally compensate to some extent raw materials of inferior quality (Zweifel et al., 2003; Petitot et al., 2009a). In addition, high temperature drying can reduce the drying time (Petitot et al., 2009a). Zweifel et al. (2003) analysed various drying profiles (Figure 3.4) and concluded that drying at high temperature at a late stage gave the best product quality (compared to low-temperature- and early high-temperature drying). The protein network was preserved through promoted protein denaturation to a dense and continuous protein network encapsulating starch granules and thus swelling of starch granules was reduced. Compared to early high-temperature drying, late high-temperature drying stabilised the protein network to such an extent that the network was still visible even in the external zone of overcooked pasta.

High temperature drying can make up for inferior protein composition (Del Nobile et al., 2003b). However, if the temperature is too high and the polymerisation reaches a point, where the proteins get too rigid to expand, it will result in an inferior pasta, too (Decour et al., 2010). High temperature drying also increases the risk for heat damages of the dried pasta - mainly off-colours and off-flavours and reduced nutritional value of the proteins with the breakdown of lysine due to the formation of Maillard reaction products (de Noni and Pagani, 2010; Peressini, 2011). It may also affect the allergenicity of the proteins (Petitot et al., 2009a).
3.2.1. Influence of processing on selected parameters

Grant et al. (1993) studied the influence of several processing parameters on pasta cooking quality. Sprouting decreased the firmness of cooked spaghetti, however, this effect could be prevented to some extent with high-temperature drying. Regrinding of the semolina increased the amount of damaged starch (Grant et al., 1993).

The amount of damaged starch is of importance for the cooking properties as it increases water absorption and is more susceptible to enzyme attack (Sissons, 2008). Damaged starch is in general formed due to mechanical forces during kneading and extruding (Delcour et al., 2010). However, also the semolina can influence the amount of damaged starch with a smaller granulation size tending to lead to increased levels (de Noni and Pagani, 2010).

Active enzymes can form reducing sugars and a higher amount of damaged starch lead to the formation of more reducing sugars (de Noni and Pagani, 2010). During high-temperature drying these reducing sugars in turn can be converted together with free amino groups into products of the Maillard reaction (e.g. furosine), giving an unfavourable brown product colour (Sissons, 2008). However, the Maillard reaction can be controlled by reducing the content of reducing sugars during processing. This can be achieved by using semolina with low alpha-amylase activity, by milling to larger granulation or by controlling the moisture content during drying (Peressini, 2011; de Noni and Pagani, 2010).

The dough should not be overdeveloped during mixing and extrusion. The optimal time for this is generally found empirically. However, Ait Kaddour and Cuq (2011) studied the opportunity of using NIR as an automated tool to determine the optimal end point of mixing semolina with water. They showed that raw NIR spectra could be correlated to the size of the agglomerated particles. Second NIR spectra could be correlated to chemical changes during the wetting phase. Finally, through mathematical modelling, a NIR time could be determined which corresponded to the time defined as being necessary to achieve a constant particle distribution in the mixer.

Bran inclusion modifies the drying kinetics and the equilibrium moisture is different in bran-rich than in bran-free semolina during drying (Villeneuve and Gélinas, 2007).

Factors important for ready-to-eat pasta meals (such as capacity to hold sauce, wholeness and weight constancy) are discussed by Kindt et al. (2008).

3.3. Pasta cooking and post-cooking

During pasta cooking, starch granules absorb water and swell. This increases the volume and pressure on the protein network (Delcour et al., 2010). With increasing temperature, two endothermic transitions take place. First starch gelatinises and at a higher temperature amylase-lipid complexes dissociate (Petitot et al., 2010). Starch gelatinisation and gluten polymerisation occur at the same time and are competitive in regard to the absorbed water as well as they are controlled by the water penetration inside the pasta (Petitot et al., 2009a). Depending on the amount of starch swelling, amylase can leach out of the granules and the starch granules disintegrate. This can induce excessive cooking loss and increased stickiness (Delcour et al., 2010). The later depends on the protein network, with a strong network preventing the leakage and dissolving of starch granules (Zweifel et al., 2003; Bruneel et al., 2010). Honeycomb-like structure at the surface of partly and fully cooked spaghetti could be detected (Heneen and Brismar, 2003; Sung and Stone, 2005). In any case, the cooking generates a concentrically change from the centre to the surface in the microstructure (Figure 5.2). This gradient in the change of microstructure and moisture content with a firm core is often referred to as 'al dente'. Delcour et al. (2010) summarised the cooking process by arguing that the transformation of starch is a hydration-driven gelatinisation process in the outer layer while it is a heat-induced crystallite melting in the
centre of the pasta. This process is characterised by two diffusion coefficients for the two different transformations (Cunningham et al., 2007).

Heneen and Brismar (2003) studied the microstructure of cooked pasta made from durum or common wheat. They argue that during cooking of pasta, large voids appear around the swelling starch granules and granules are flattened in the intermediate region. Later in the process starch granules are fusing. The authors suggest that due to the larger size of common wheat granules, more granules will fuse. This results in an insufficient, discontinued protein network.

Storing cooked and pre-cooked pasta deteriorates their sensorial properties as the uneven water distribution events out and thus reduces the firmness (McCarthy et al., 2002; Wood, 2009; Olivera and Salvadori, 2012). The deterioration effect can be controlled by the storage process. Irie et al. (2004) treated cooked spaghetti samples differently after production: samples were dried, frozen, or stored at moderate or chilled conditions. Dried and frozen spaghetti showed a clear moisture gradient from surface to core with a low core moisture content of below 15%. Fresh and chilled spaghetti showed a gentle moisture gradient whereas one week stored spaghetti did barely show any gradient. Mechanical properties followed the tendencies in the moisture gradient, with higher forces needed to break dried and frozen spaghetti and lower forces for the chilled spaghetti. Faster product freezing can remain better initial quality during frozen storage (Olivera and Salvadori, 2011).

Also the choice of raw materials can influence the properties after storage. Chickpea-fortified spaghetti remained firmer than the control (Wood, 2009). Furthermore, spaghetti maintained firmer when seasoned after cooking with sodium chloride or monosodium glutamate as those salts temporarily absorbed excess water at the surface of the spaghetti (Horigane et al., 2009).

3.3.1. Influence of cooking conditions

The cooking water composition affects texture properties of pasta. Both increased pH and water hardness result in higher stickiness values (Malcolmson and Matsuo, 1993). Weak acidic pH seems to be optimal. Distilled water has a pH of 6 (as it binds carbon dioxide) and is thus not recommended to compare different pasta samples. It might minimize differences which are more apparent when cooking pasta in tap water (Malcolmson and Matsuo, 1993; Cole, 1991). Instead, artificial water with a standardized water composition has been recommended, but not often used (Matsuo et al., 1992). Another standard (AFNOR Standard NF-V 03-714) recommends mineral water mixed with 0.7 % sodium chloride (referred by Delcour et al., 2000a).

Salted (sodium chloride) cooking water influences pasta cooking properties, too. Higher salt contents decrease the water absorption of pasta samples, resulting in longer cooking times (Majzoobi et al., 2011a; Sozer and Kaya, 2003, 2008; Ogawa and Adachi, 2013). Ogawa and Adachi (2013) assume that the sodium ion positively hydrates and thus binds some water molecules and due to its larger ion diameter hinders water diffusion. Salt stabilizes also the protein structure by increasing the hydrophobic interactions between the gluten units (Peressini et al., 2000; Sozer and Kaya, 2008). Salt improves the sensorial perception and an increases in salt concentration increase the hardness and adhesiveness of cooked spaghetti, too (Sozer and Kaya, 2008).

The amount of absorbed water depends on the water temperature, too (Del Nobile and Massera, 2002). Below 40°C pasta strands increased in weight to 160% of its dry weight. For temperatures above 40°C (here: 60, 80, 100°C) the weight increased in the stationary phase to about 300, 400 and above 500%, respectively.

Finally, microwave cooking can reduce cooking loss and better retain the yellow colour than cooking in boiling water while firmness decreases(Cocci et al., 2008).
Part II.

METHODS
4. Introduction Part II Methods

Food systems are organised at several length scales, which can be analysed and visualized by a variety of methods. For the example of starch as food component, Conde-Petit (2003) structured several methods into nano-, micro and macroscale. Other used varying terms and introduced an additional mesoscale (Trystram, 2011; van der Sman and van der Goot, 2009). Starch granules and gluten are cited as examples for mesoscale structures by van der Sman and van der Goot (2009).

In the following chapters, several methods are briefly described, which were or could be used to analyse the structural elements of pasta and its raw materials over several length scales. Additionally, suitable methods to observe water and moisture migration are mentioned. Some of the methods can be used in several ways, for example SEM and TEM can either be used as an imaging or as a spectroscopic tool. Therefore, the following classification may not be completely stringent, but lists methods rather in the context where they are most often (potentially) used in carbohydrate research.
5. Analysing the microstructure

5.1. Microscopy

Microscopy techniques use microscopes to enlarge the image of objects, which could otherwise not been seen in the same detail with the eye. Thus it is possible to gain knowledge about structural properties. In optical and electron microscopy, electromagnetic radiation (light) or an electron beam is interacting with the specimen, whereas in scanning probe microscopy the interaction is based on atomic forces of the specimen.

In general, microscopical methods can also be distinguished whether they are applied destructively or non-destructively and whether the surface of the specimen is analysed or a inner layer of it.

5.1.1. Light microscopy (LM)

Light microscopy is one of the oldest analytical method as its working principle is rather simple. A light beam is sent through a set of lenses and the sample to receive an enlarged image of the structure of the sample on a detector. The image is formed by several sources of contrast such as absorption, scattering and reflection of the illumination. Diffraction limits the image resolution theoretically at about the half of the length of the incident light beam - using visible light this gives a limit of about 200 nm. This limit is in practice in the magnitude of μm due to sample preparation and properties. Resolution limits are lower when using electron microscopy, but light microscopy offers an useful range for analysing food products (Autio and Salmenkalliomarttila, 2001).

Based on different contrast mechanisms, several optical microscopy set ups have been developed and frequently used ones are shown in Figure 5.1 (Cisek et al., 2009).

- **Bright field (BF) light microscopy** Bright light coming from the light source is focused through the condenser lens on the specimen, from where the light is collected through the microscope objective, which also creates a magnified intermediate image. Through the ocular the image is magnified further and the light is focused again on a detector.

- **Dark field light microscopy** excludes the unscattered light ray through an specifically designed annular aperture. It enhances contrast in unstained specimens and can display elements of low light intensity, but which scatter and diffract the light.

- **Phase contrast** can evolve when light rays are travelling through materials of varying refractive index or thickness. This results in different light path lengths and the interference of the different phases of these light paths can form the contrast. To enhance the occurring phase contrast, an angular aperture is used before the condenser and a special phase plate behind the objective,

- **Polarised light microscopy** A polarising filter (polariser) beneath the condenser aligns the light rays into one single plane. A second filter (analyser) above the objective is positioned in right angle to the polariser. Thus, only polarised light will
Table 5.1.: Commonly used staining agents for carbohydrates in (A) bright-field light microscopy and (B) fluorescence microscopy. Adapted from Autio and Laurikainen (1997), Autio and Salmenkalliomarttila (2001) and Aguilera and Stanley (1999), except for the references mentioned in the foot notes.

(A) BRIGHT-FIELD LIGHT MICROSCOPY

<table>
<thead>
<tr>
<th>Component</th>
<th>Stain</th>
<th>Colour</th>
</tr>
</thead>
<tbody>
<tr>
<td>Starch</td>
<td>Iodine solution</td>
<td>Black, violet</td>
</tr>
<tr>
<td>Amylose</td>
<td>Iodine solution</td>
<td>Blue</td>
</tr>
<tr>
<td>Amylopectin</td>
<td>Iodine solution</td>
<td>Beige, brown</td>
</tr>
<tr>
<td>Protein</td>
<td>Light green</td>
<td>Green</td>
</tr>
<tr>
<td>Lipid</td>
<td>Sudan III / IV</td>
<td>Red</td>
</tr>
<tr>
<td></td>
<td>Oil Red O</td>
<td>Red</td>
</tr>
<tr>
<td></td>
<td>Sudan Black B</td>
<td>Blue-black</td>
</tr>
<tr>
<td>Cellulose</td>
<td>Thionin</td>
<td>Violet</td>
</tr>
<tr>
<td>Lignin</td>
<td>Phloroglucin</td>
<td>Red</td>
</tr>
<tr>
<td>Pectin</td>
<td>Ruthenium red</td>
<td>Rose</td>
</tr>
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</table>

(B) FLUORESCENCE MICROSCOPY

<table>
<thead>
<tr>
<th>Component</th>
<th>Stain</th>
</tr>
</thead>
<tbody>
<tr>
<td>Starch</td>
<td>Periodic acid Schiff’s (PAS)</td>
</tr>
<tr>
<td>Protein</td>
<td>ANS, Fuchsin acid, Texas red, FITC, Thiazine red</td>
</tr>
<tr>
<td></td>
<td>Rhodamine red(^a)</td>
</tr>
<tr>
<td>Glutenin</td>
<td>Tetraethyl-rhodamine B(^b)</td>
</tr>
<tr>
<td>Gliadin / Gluten subunits</td>
<td>Immunostaining(^c)</td>
</tr>
<tr>
<td>Lipid</td>
<td>Nile blue, Nile red</td>
</tr>
<tr>
<td>β-glucan</td>
<td>Calcofluor, Congo red</td>
</tr>
<tr>
<td>Cell wall comp.</td>
<td>Congo red(^d)</td>
</tr>
<tr>
<td>Lignin</td>
<td>Crystal violet/erythrosin B</td>
</tr>
<tr>
<td>Arabinoxylans</td>
<td>Immunostaining(^a)</td>
</tr>
</tbody>
</table>

\(^a\)Kratzer (2007), \(^b\)Li et al. (2004), \(^c\)Tosi et al. (2011), \(^d\)Alminger et al. (2012)

Figure 5.1.: Comparison of several methods in optical microscopy. O, ocular; CO, microscope objective; S, sample; CL, condenser lens; LS, light source; AA, annular aperture; PP, phase plate; A, analyser; P, polariser; N, Normaski prism; F1/2, Transmitting filter. Reproduced from Cisek et al. (2009)
Figure 5.2: Light micrographs of cooked spaghetti and stained with Fast Green and iodine. Starch is coloured violet/brown, while gluten is coloured green. \textit{Left image} specimen thickness 2\,µm. \textit{Right image} specimen thickness 9\,µm, Scale bar 25\,µm. Reproduced from Heneen and Brismar (2003) and Zweifel et al. (2003)

be detected which has been rotated when passing through the sample. The rotation is induced by anisotropic structures showing birefringence. This is the case for e.g. ungelatinised starch granules (their appearance under polarised light is called Maltese cross) or for fibrils and cell walls of plants.

\textbf{Differential interference contrast microscopy (DIC)} The polarised light ray is split into two parallel rays (caused by a prism) and travels through the specimen in such a way, that the split light ray paths are phase shifted. With a second prism the rays are combined again, giving the image a three-dimensional appearance (Cisek et al., 2009). This method is suitable to visualise differences in thickness or orientation in the specimen.

\textbf{Fluorescence microscopy} uses the effect that certain components will - after being illuminated with light of a certain wavelength - emit light of a longer wavelength. The technique is explained in more detail in the next section (see Section 5.1.2).

\textbf{Sample preparation}

In thin biological samples absorption and scattering have generally low effects. Therefore, to enhance contrast in optical microscopy, staining agents are often used. An overview of commonly used agents in staining components in carbohydrate-rich systems is given in Table 5.1. However, some types of microscopy, such as polarised light microscopy, do not need staining at all.

Further sample preparation is dependent on the resolution required. A rule of thumb is that the thinner the specimen, the better the resolution will be as scattering and absorption of light is reduced. To achieve a specimen thickness of about 1\,µm, the structure of the sample has to be fixed. Glutaraldehyde and Osmiumtetraoxid are common fixation agents. After the specimen has been embedded in a resin solution and completely polymerised, it can be sliced using a microtome. As an example, Heneen and Brismar (2003) prepared specimens with a thickness of 2\,µm. An alternative to embedding is cryosectioning, that is to freeze the specimen and then slice it. Sample thickness of 10\,µm can be obtained (Hug-Iten et al., 1999; Zweifel et al., 2003). A faster option resulting in a larger thickness is to smear the liquid or solid sample onto an object glass (Langton and Hermansson, 1989). Examples for the effect of specimen thickness on scattering are shown in Figure 5.2)

An exception to the rule of thumb is polarised light microscopy, where thicker samples are useful (at least when analysing pasta). In polarised light microscopy, the intensity of the transmitted light is increasing proportional with the sample thickness. Del Nobile et al.
(2003b) used samples with a thickness of about 1 mm (and such samples can be quickly prepared by cutting uncooked and cooked spaghetti strands with a razor blade).

**Application in research**

Cisek et al. (2009) list several examples for the use of phase contrast, dark field microscopy and DIC in analysing cells. For pasta, however, no examples in the literature could be found. Autio and Salmenkalliomarttila (2001) described bright field light microscopy as an useful tool for grain research. It offers the opportunity of targeted staining of grain components and can visualise structural changes of the material. Within the broader topic of pasta research, light microscopy has been used to compare pasta made of different wheat types or other flour types (Heneen and Brismar, 2003; Petitot et al., 2010), to study the influence of drying on pasta texture (Zweifel, 2001; Zweifel et al., 2003) and to visualise the change of starch structure during cooking of spaghetti (Cunin et al., 1995). Because of the easiness to discriminate amylopectin and amylose when staining with iodine, the method is also often used to analyse starch phenomena only (Kratzer, 2007; Langton and Hermansson, 1989; Sriakao et al., 2006; Wellner et al., 2011). A common dimension for bright field light microscopy images showing pasta structures is at about 20 to 200 μm. It is possible to display the complete cross section of e.g. a spaghetti, at least when using low magnifications (Del Nobile et al., 2003b). This makes this method suitable to give an overview over a pasta sample. There are also hot stage microscopes available which have a heatable sampler holder and thus allowing it to follow changes in the microstructure of the product at elaborated temperatures (Kratzer, 2007).

**5.1.2. Confocal laser scanning microscopy (CLSM)**

CLSM combines in the often used epi-fluorescence mode two concepts - fluorescence and eliminating out-of-focus light rays. Light of a specific wavelength is sent to the specimen where the fluorophores of the specimen will emit light of a longer wavelength (fluorescence). The emitted light is separated from the excitation beam by a dichroic mirror, which is a wavelength filter and is collected on scanning mirrors. Close to the light source and the detector pinholes are inserted in conjugated planes to the sample to eliminate the light rays which are not in focus in the focal plane (Figure 5.3).

![Figure 5.3.: Schematic working principle of a CLSM](source: Danh at the English language Wikipedia under CC-BY-SA 3.0, accessed 06/12)
Figure 5.4: Upper Row CLSM of cooked pasta made from (a) durum wheat and (b) split pea flours. Fibre fragments in (d) split pea pasta and (e) durum wheat pasta. Al, aleurone, Per pericarp, cell, cellular structure. All samples were stained with calcifluor.

Lower Row CLSM of cooked pasta in (a) central, (b) intermediate and (c) external region of a spaghetti strand. Proteins were stained with fuchsin acid and appear white. Dark areas can be connected to starch granules. Extracted from Petitot et al. (2010) and Petitot et al. (2009a).

The epi-reflection mode of the CLSM allows to analyse the surface and topography of samples, as the reflected laser light is collected as the signal. To increase contrast the sample can be sputtered with a thin metal film (as in SEM; Dürrenberger et al., 2001).

The resolution limit is slightly better than light microscopy. Preparation effort is depending on the sample and often minimal (Aguilera and Stanley, 1999). For instance, samples showing autofluorescence, such as parts of the cell wall as aleurone and pericarp structures and in general fibres, do not need further preparation. An example is shown in Figure 5.4 (Petitot et al., 2010). The main source for autofluorescence are polyphenolic compounds like lignin and ferulic acid (Autio and Salmenkalliomarttila, 2001). Using fluorescents (Table 5.1) or labelled antibodies can increase the spectra of analysable components, which are otherwise not showing autofluorescence. It is also possible to bleach samples to remove autofluorescence (Zweifel et al., 2003). To analyse cell wall structures, Dornez et al. (2011) tested several immunolabeling techniques. Immunolabelling offered the possibility to stain very specific certain cell wall material. However, it is a complex technique and time-consuming. Dornez et al. (2011) recommend to consider what the purpose of the labelling shall be and whether immunolabelling or a common fluorescent should be preferred.

The change in the protein structure during processing has been analysed by several authors (Dürrenberger et al., 2001; Fardet et al., 1998; Zweifel et al., 2003; Petitot et al., 2009b). The protein distribution has been shown in both wheat grain and cooked pasta (Figure 5.4; Petitot et al., 2009a; Tosi et al., 2011). CLSM was also used to show the influence of dietary fibres on pasta quality (Petitot et al., 2010; Aravind et al., 2012a,d).
Quantum dots (QDs) are a recent development to be used as substitute markers for proteins. QDs are semiconductive nanoparticles, which are fluorescent and their colour are depending on the particle size. Thus, the colour can easily be controlled by varying the size. Furthermore, QDs are resistant to photobleaching. At a recent conference, Sozer and Kokini (2011) presented their first results on using QD to mark proteins in bread.

Fluorescence recovery after photobleaching (FRAP)

A focused laser beam is concentrated on a spot of a specimen for a short time to bleach the fluorescently labelled molecules in this area. This will decrease the fluorescent signal significantly. After stopping the laser beam, fluorescent-active molecules will diffuse in this area, which can be measured by an increased signal. By analysing the drop and the time of the recovery of the signal, the diffusion coefficient can be calculated. The working range of FRAP is given to be between 0.1 and 100 \( \mu \text{m}^2 \text{s}^{-1} \) for the diffusion coefficient (Descout et al., 2010).

Photo activation

Photo activation is the vice-versa of FRAP. Instead of bleaching, fluorescent particles are activated by a concentrated laser beam. Again, the movement of the particles can be tracked by e.g. CLSM. This method is especially suitable in aqueous systems.

5.1.3. X-ray computed tomography (XRT)

XRT is another non-invasive technique to create cross-section images of an object, using X-rays. The cross-sections can later be recalculated to achieve a 3D-model of the object. Besides of XRT, the term \( \mu \)-CT is frankly used, with micro referring to the pixel size and CT standing for computational tomography. The image contrast is based on absorption differences of the X-rays of the elements of the object (e.g. whether they are solid, liquid or gaseous) and thus, density variations can be determined. Observations are possible under environmental conditions without prior sample preparation. The axial and lateral resolution is in size range of some \( \mu \text{m} \) (van Dalen et al., 2003). Mohoric et al. (2009) used the technique to visualise the porous structure of rice kernels after cooking. The authors claim that the technique is especially useful for analysing in 3D porous microstructures under low moisture conditions. Another study combined XRT with magnetic resonance imaging (MRI) to identify structural elements in a food system which cause fast hydration (Weglarz et al., 2008). They baked a probably starch-based food system with crunchy inclusions and determined its structure by XRT in the dry state. Then they analysed the hydration in real-time using a fast spin-echo imaging method and were able to to correlate moisture content deviations to the type and form of the inclusion.

A recent study used XRT to visualise the internal shrinkage in spaghetti during drying (Zhang et al., 2013). The authors describe the technical prerequisites and calculations necessary to visualise the effect of drying; among others, they used added aluminium particles to increase the contrast in the dense pasta dough.

5.1.4. Scanning electron microscopy (SEM)

The working principle of SEM: A focused, incident beam of electrons hits the sample and by interacting with the atoms of sample, knocks out electrons from it. These knocked out electrons will have a certain energy depending on the atomic shell they were released from and can be detected using several methods. Electrons can be emitted as X-ray, as secondary electrons, they can be backscattered or they can be transmitted. Each type of
electron interaction can be registered with specific detectors and the detection of secondary electrons (describing reflected electrons) is most common. The image of the sample is created by the collected electrons at the detector and is formed through a raster scan of the focused beam over the surface of the sample.

The image in a SEM is formed based on differences in contrast. Contrast mechanisms are based on the surface topography, magnetism, atomic number, conductivity and crystallographic orientation. SEM in general can be used to analyse surfaces of a specimen. Depending on the detector, the SEM has a high depth of focus, showing the topography of the specimen in semi 3D.

Several factors influence the spatial resolution and this includes beam spreading effects, the incident electron beam diameter, signal to noise ratio and instabilities. A higher voltage in the microscope leads to better contrast and higher resolution and thicker samples can be used. However, higher voltage can also lead to sample destruction. Finally, the resolution depends on the quality of the sample preparation, too.

In general, the specimen has to be conductive or needs to get a conductive layer. This is often done by sputtering a gold layer onto the specimen which also increases the contrast. Alternatively, low voltage SEM can be used to avoid charging of the material (Heneen and Brismar, 2003; Langton and Hermansson, 1989). Furthermore, water has to be removed before SEM analysis. The sample can be dehydrated after a fixation step or solidified by freezing. Another limitation is the need of a certain quality of a vacuum to avoid interactions of the electron beam with molecules in the gas phase. If this is not possible, environmental SEM (ESEM) has been developed. This method gives a lower resolution,
but can be used without special treatment in e.g. biological samples. Petitot et al. (2010) used ESEM to visualise the starch granule distribution from various flours used for pasta production (Figure 5.5).

The resolution limit of SEM is between 5 and 10 nm and the scanned area is often between 50 and several 100 µm in each dimension.

Scanning electron microscopy is widely used to understand phenomena in pasta. The first papers using SEM to analyse the structure of pasta have been published in the 70s and 80s (Dexter et al., 1978; Matsuo et al., 1978; Pagani et al., 1986; Resmini and Pagani, 1983). It is often used when the influence of new ingredients on the structure shall be compared, especially on the protein network (Aalami and Leela vathithi, 2008; Aravind et al., 2012a,d; Fuad and Prabhasankar, 2012; Majzoobi et al., 2011b; Manno et al., 2009; Prabhasankar et al., 2010; Sung and Stone, 2005). The influence of processing on the pasta product was shown as well using SEM (Cocci et al., 2008; Fardet et al., 1998; Lucisano et al., 2008; Manthey and Schorno, 2002; Zweifel, 2001). Cunningham et al. (2007) used SEM to demonstrate the effects of starch swelling on the pasta surface during pasta cooking. An example for the change from dried to cooked pasta is given in Figure 5.5 (Purnima et al., 2012) and for the intermediate products during processing in Figure 3.1 (Petitot et al., 2009a).

5.1.5. Transmission electron microscopy (TEM)

The working principle is similar to SEM, however in TEM, the electron beam is send through an ultrathin sample as the resolution is inversely proportional to the sample thickness. Instead of topography, the inner structure of the sample is visualised as the image is formed from the interactions of the electrons with the specimen as the beam passes through. Besides the thickness of the specimen, a higher voltage source results in shorter wavelengths which in turn increases resolution. Especially for biological samples, however, it has to be taken into account, that a too high voltage can destroy the sample.

TEM offers the best resolution compared to LM and SEM, with a limit of down to some nm, but requires extensive sample preparation. Specimens have to be very thin to avoid multiple scattering and are often below 100 nm in thickness (e.g. Pagani et al., 1986). Resmini and Pagani (1983) presented freeze-fracturing as a preparation technique for analysing dry and cooked pasta. TEM was used to show to subunits in gelatinised starch granules and lipid inclusions in the protein matrix (Pagani et al., 1986). Besides the before-mentioned studies, TEM has not been widely used to analyse the microstructure of pasta.

TEM is mentioned to study the growth rings of hydrolysed starch granules in the review of Pérez and Bertoft (2010). To name another field, TEM offers opportunities to show isolated wheat components at great detail. For an isolated crude extract of high molecular weight (HMW) glutenins, a novel nanostructure could be shown (Mackintosh et al., 2009).

5.1.6. Atomic force microscopy (AFM)

AFM uses a tip to scan a surface at the atomistic level. While the tip is moved mechanically over the specimen, the tip will interact with the specimen. These movement of tip can be measured with a laser beam. The deviation of the beam is registered with a detector and this is used to form an image of the topography of the specimen (Figure 5.6). The resolution limit is below nm and it can be applied both under environmental conditions and especially in liquids, too. A drawback of the technique is that there are lots of possible artefacts and that AFM can be used only for rather smooth surfaces.
Albeit being a rather new technique and only being available for a little more than twenty years, a vast number of studies has been published on analysing biological samples and there, among others, the starch structure (Paananen, 2007). A recent study analysed the surface structure of buckwheat starch (Neethirajan et al., 2012). The surface of pasta dried at different temperatures was analysed by AFM as well (Zweifel, 2001). Pores were visible at the surface in the study of Ogawa et al. (2011) (Figure 5.6).

5.1.7. Image analysis

Image analysis summarises techniques to extract meaningful information from and to quantitatively analyse more data in images. The analysis is often based on pattern recognition, geometry or signal processing. A good introduction into the topic is offered by Russ (2006).

To name some examples from recent studies: Using grey level granulometry, the protein network distribution was studied in dough (Jekle and Becker, 2011) and in pasta (Petitot et al., 2010). Others determined the granule size distribution of wheat starch granules (Wilson et al., 2006) and the pore size in pasta dough (Thorvaldsson et al., 1999). A recent study proposed three different techniques to distinguish the surface texture of cooked pasta samples (Fongaro and Kvaal, 2013).

5.2. Other methods

Spectroscopic methods have in common to study the interaction between matter of the sample and radiated energy. The energy source can vary and can be among other light, electrons or X-ray. It is often used for chemical component analysis of a material. Typical methods include Light scattering, Near-infrared spectroscopy (NIR or FT-IR), nuclear-magnetic resonance (NMR, Mariette (2009)) and X-ray microanalysis using SEM or TEM.

Crystallinity level of starch can be determined with a X-ray diffractometer (Baiano et al., 2006). In general, Lopez-Rubio and Gilbert (2009) recommend the increased usage of neutron scattering for determination of structural elements at the nm scale, e.g. for starch structures.

Finally, some chemical methods shall be briefly listed: Protein distribution and determination of gluten fractions have been done by size-exclusion high-performance liquid chromatography (SE-HPLC, Icard-Vernière and Feillet, 1999; Rao et al., 2001). The extent of protein polymerisation was determined by diluting the sample in sodium dodecyl sulphate (SDS) solution, i.e. SDSEP (Lagrain et al., 2008). The amount which is unextractable
in the solution corresponds to the degree of protein polymerisation and it has been used to analyse protein modifications during drying and cooking (Bruneel et al., 2010).

Petitot et al. (2010) list in their study the current methods to determine the contents of carbohydrates, protein, lipids, ash and dietary fibre. For the specific purpose of analysing wheat grain and flours, a publication of the Wheat Marketing Center of Kansas State University offers a valuable source (Anonymous, 2008).
6. Analysing the macrostructure

6.1. Rheological methods

Rheological measurements are frequently used to describe viscoelastic materials. The main principle is that a force is applied to the material and the response of the material is measured. The force can be applied constant (creep), as a sinusoidal force (oscillation) or as a rotational force and it can be applied non-destructive (small amplitude measurement) or destructive (large amplitude measurement). Various material properties can be deduced from the different types of measurement. The properties are further depending on the time, temperature and the frequency of the force applied.

Commonly three parameters are determined: the storage modulus describing elastic properties of a material \(G'\), the loss modulus describing viscous properties \(G''\) and the relation of \(G'\) to \(G''\) giving an indication of the viscoelastic behaviour of the material (\(\tan \delta\)). A thorough introduction into rheology can be found elsewhere (Schramm, 1995).

Within the area of pasta research, viscoelastic or mechanical properties can be determined of components such as gluten or starch, of doughs and of the final pasta product and can be correlated to pasta cooking quality. For example, Cafieri et al. (2010) determined and modelled the elastic modulus and tensile strength during cooking of spaghetti strands, whereas Sozer and Daligc (2007) used relaxation and creep data to model rheological characteristics for varying spaghetti types. Relaxation and dynamic oscillatory tests were carried out to evaluate changes during storage of frozen lasagne (Olivera and Salvadori, 2011). Other used an Instron Universal testing machine to determine the elastic modulus of cooked pasta strands during extension (Baiano et al., 2006).

6.1.1. Rheological properties of flours and doughs

Rheometers were developed to measure rheological properties with great accuracy and offering a wide range of available techniques. For example, a dough for fresh pasta was placed in a parallel plate geometry and a time sweep as well as a frequency sweep were applied using a defined oscillating shear force (Peressini et al., 2000). Zhu et al. (2011) summarised the outcome of other studies which used creep data to determine rheological properties of pasta products.

Rheometers offer a great accuracy, but they demand a careful sample preparation and measurement procedure. Therefore, several devices have been developed to quickly and easily analyse some empirical rheological properties of flours and doughs. The American Association of Cereal Chemists developed standard methods\(^1\) for several of the below-mentioned devices. Some further empirical tools for analysis of all from flours to pasta are collected in a so called wheat flour book, issued by the North American Export Grain Association\(^2\).

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\(^1\)http://www.aaccnet.org/approvedmethods/toc.aspx, accessed 03/2012.

Rapid viscoanalyser (RVA)

RVA analyses flour starch properties. A starch sample is mixed with water and the slurry is heated under stirring to 95°C. The resistant to shear is recorded over time (Copeland et al., 2009). From the recorded curve peak viscosity can be determined. Vansteelandt and Delcour (1999) extended the measurement by applying an isothermal step at 95°C and then cooling the slurry to 50°C. In this way they could also determine the retrogradation behaviour of the analysed starch.

Falling number

Sprout damages and enzyme activity in flours can be revealed using the concept of the falling number. Flour and water are mixed to form a slurry and are placed in a tube with a falling stirrer. The slurry is stirred with a falling stirrer and is heated at 100°C in a water bath. The time it takes for the stirrer to reach the bottom is recorded as the falling number in seconds. Low falling numbers indicate high enzyme activity or sprout damages.3

Farinograph and amylograph

The farinograph and the amylograph are devices developed by the company Brabender to determine flour and resulting dough properties. Flour and water are mixed and the shear resistance over time during kneading at a constant temperature is recorded. Generally, a hockey-stick shaped form of the curve can be seen and can be related to the gluten strength of the flour. Some empirical parameters can be measured such as developing time (time to reach maximum resistance), stability (time duration of constant resistance), mixing tolerance index (MTI, describing the difference in resistance at the peak value and 5 min later) and the level of water absorption (Gazza et al., 2011).

Alveograph

Doughs can be analysed with the alveograph. A dough sample is deformed under pressure to an increasing thin bubble until it breaks. Through recorded alveograms tenacity and elasticity of the dough and the relation of these two parameters can be determined (Sissons, 2008).

6.1.2. Rheological properties of gluten

An arbitrary, but often used method to determine the gluten strength is the gluten index, which categorises the gluten into qualities from inadequate to excellent (Sissons, 2008). According to a standardised method4, gluten is isolated by washing it from the semolina using an automatic washing apparatus. Then the wet gluten is placed on a special sieve in a centrifuge. After centrifugation, gluten passed through and gluten remaining on the sieve is weighed and set into relation. The more gluten is remaining on the sieve, the stronger the gluten is regarded (Rao et al., 2001).

Through drying the wet gluten and weighing the remaining, the dry gluten content can be determined as a percentage of the wet gluten content. The difference between the weights of dry and wet gluten is referred to as water-binding capacity.

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4 http://www.icc.or.at/standard_methods/158, accessed 03/2012.
Figure 6.1: DSC of milled, non- and partly cooked spaghetti for varying times (cooking time in min) showing apparent heat capacity by unit mass of dry matter. Extracted from Riva et al. (2000)

6.1.3. Texture analysis

Texture analysis can be used to analyse mechanical properties of pasta products. For example, Gonzalez et al. (2000) used a texture analyser to determine the initial slope of the resistance force and peak force during cutting. Both values were negatively correlated with cooking time. However, they also decreased during holding of the cooked pasta sample. Both observations can be correlated to a more uniform moisture distribution.

In general, texture analysis is used to determine texture parameters such as firmness, elasticity or stickiness (Sissons, 2008; Cubadda et al., 2007). How this is done is described in more detail in Section 8.2.

6.2. Thermal analysis

6.2.1. Differential scanning calorimetry (DSC)

DSC is a thermoanalytical method used to determine physical transformations, especially phase transitions. The working principle: A small amount of the milled sample is placed in excess water in a small pan and is sealed. A second pan is filled with water only. Both lids are then heated at linear increasing temperature (constant heat rate) and the heat transfer of both lids is recorded. During phase transitions a difference will occur and this difference is referenced to the temperature (see the peak in Figure 6.1). A limitation of the method is that the results are dependent on the heating rate.

Within pasta analysis it is mostly used to determine the level of starch gelatinisation. Gelatinisation is an endothermic reaction resulting in a peak in the DSC diagram as seen in Figure 6.1. The longer pasta is cooked, the more starch is already gelatinised and thus smaller gelatinisation peaks will be recorded in the DSC analysis (Figure 6.1; Riva et al., 2000). Amylose, amylopectin and starch have different gelatinisation temperatures and therefore DSC can be used to determine the concentration of these components (Vansteelandt and Delcour, 1999).
6.2.2. Dynamic mechanical analysis (DMA)

DMA is a technique to determine the viscoelastic properties of a solid material. The sample is placed between two grips and then an oscillating torsional or axial force is applied. Most often, temperature or frequency sweeps can be carried out.

The temperature sweep was used to determine the glass transition temperature in pasta in dependency of the water content (Takhar et al., 2006). The glass transition is determined as the temperature region where the storage modulus is sharply decreasing. The same authors predict that DMA can help to predict the effect of drying and storage temperatures. In another study, DMA was used to analyse the influence of the water content on the structural changes in pasta dough during heating (Thorvaldsson et al., 1999).

6.3. Other methods

Pore size distribution has been measured by mercury intrusion porosimetry (e.g., Ogawa et al., 2011; Petitot et al., 2010). By applying a pressure to a pasta sample, mercury is pressed into it. Based on the weight increase, the volume taken in by mercury can be calculated.
7. Analysing water transport and mobility

7.1. Water transport and its modelling

Water is a major part of all foods and its distribution steers food properties. In the context of pasta products, especially the water absorption and desorption processes are of extra interest. During the life-time of a pasta product water migration occur during mixing of the dough, drying and cooking of the pasta and finally during storage of the cooked pasta. A lot of work has been carried out to model these sorption processes. The modelling is complicated by the fact that the sorption is happening at the same time as swelling/shrinking and physical modifications of the structural elements, i.e. gelatinisation of starch and denaturation of gluten (Del Nobile et al., 2003a). To ease the mathematical description of the phenomena, water migration is most often described in one dimension only. That is also why spaghetti and lasagne plates are used so abundant, as the radial profile of the spaghetti and the large surface compared to the thickness of lasagne, respectively, leads to a water migration in predominantly just one dimension.

7.1.1. Water transport mechanisms

![Figure 7.1: Schematic illustration of three approaches to describe water motions. Reproduced from Schmidt (2007)](image)

The qualitative movement of water and its local distribution and availability can be analysed at various length scales and with various conceptual approaches. Schmidt (2007) classified three types of water movements (Figure 7.1). At the macromolecular level, the concepts of water activity and sorption isotherms have been developed and are used widely especially as both parameters are useful to describe spoilage or deterioration of foodstuff...
of medium and high moisture content during storage (Labuza and Atunakar, 2007). However, for dried pasta these parameters are of minor importance as dry pasta has a rather low water activity and is a microbiological stable food. Still, a study was published on determining the sorption isotherms for macaroni at varying humidity levels at ambient temperatures (Arslan and Togrul, 2005).

For pasta, water transfer mechanisms at the microstructure level are more interesting to study. Some general concepts for transport mechanisms in porous media are discussed by de Oliveira Romera et al. (2012) (Figure 7.2). Pasta is seen as a porous medium where water transport is led by liquid diffusion during drying (Cubadda, 1993). However, it is a medium of low porosity where the large majority of pores is in the size of 2-50 nm and only a minor fraction in the size of 0.05-2 μm (Petitot et al., 2010).

Possible transport mechanisms during hydration for foodstuff in porous media can comprise in general diffusion and capillary flow (Saguy et al., 2011). Others developed a model based on the assumption that the hydration is happening as a fluid transport in a porous medium that can swell (Zhu et al., 2011). Ogawa et al. (2011) combined sorption kinetics experiments with pore analysis and concluded that the rate of hydration was governed by the diffusion through the pores of the spaghetti as the hydration rate was assumed to be higher than the diffusion rate of water.

Studies which relate water transport to structural elements (such as starch, gluten, air or bran) could not be found.

The other two approaches shown in Figure 7.1 - molecular water mobility and polymer science approach - are discussed in Section 7.2 and 2.1.3, respectively.

![Figure 7.2: Moisture transfer mechanisms in porous media. Reproduced from de Oliveira Romera et al. (2012)](image)

### 7.1.2. Theoretical kinetic models of water transport

The driving force of a stationary, continuous diffusion is the chemical potential or vapour pressure difference between two regions and is described by Fick’s first law (Case I Fickian diffusion, Labuza and Atunakar, 2007; Horigane et al., 2006):
\[ J = -D \frac{\partial c}{\partial x} \]

**J** Flux \([\text{mol s}^{-1} \text{m}^2]\); or for water can be written as \([g\text{H}_2\text{O} \text{ s}^{-1} \text{m}^2]\)

**D** Diffusion coefficient \([\text{m}^2 \text{s}^{-1}]\)

**\(\frac{\partial c}{\partial x}\)** Concentration gradient \([\text{mol m}^{-4}]\)

**c** Concentration; or moisture content \([\text{mol m}^{-3}]\)

**x** Distance of diffusion \([\text{m}]\)

The diffusion coefficient is material dependent. If the concentrations are varying over time, Fick's second law (case II Fickian diffusion) can be applied, which is valid for in-stationary diffusion:

\[ \frac{\partial c}{\partial t} = D \frac{\partial^2 c}{\partial x^2} \]

Another concept introduced the term water demand (WD), which is defined as the difference between the existing and the maximum moisture content (Fukunoka et al., 2000).

### 7.1.3. Kinetic models applied to pasta

In general, most models applied to pasta are based on experimental analysis. This is most often done by either direct methods such as gravimetric measurements (Cafieri et al., 2008, 2010; Cunningham et al., 2007; Del Nobile et al., 2003a) or indirect methods such as studying the moisture profile by NMR/MRI (Carini et al., 2012; Gonzalez et al., 2000; Hills et al., 1996, 1997; Horigane et al., 2006; Abd Karim et al., 2000; McCarthy et al., 2002; Xing et al., 2007) or NIR (De Temmerman et al., 2007; Ait Kaddour and Cuq, 2011; Zardetto and Dalla Rosa, 2006). Most models have been developed for the process of drying (De Temmerman et al., 2008; Hills et al., 1997; Migliori et al., 2005; Ponsart et al., 2003) and cooking (among others Cafieri et al., 2008; Del Nobile et al., 2003a; Fasano et al., 2011; Ogawa et al., 2011; Zhu et al., 2011), respectively.

**Mixing and drying**

The reactivities during mixing were described by Oulahna et al. (2011). They described mixing as a two step process: The first step is the spreading-wetting of the solid surface and the second the liquid diffusion within the grain. The authors modelled the reactivities using sorption isotherms (in dependence of relative humidity).

Drying is a simultaneous process of mass and heat transfer (Migliori et al., 2005). According to Hills et al. (1997), the drying process is a non-fickian process, as there is a sharp moisture gradient and the outer layer acts as a barrier to moisture loss. Others, however, could use Fickian diffusion to predict average moisture contents during drying and they could fit the model to experimental data (De Temmerman et al., 2008). Thus the mass transfer could be expressed by a diffusion coefficient and the heat transfer by desorption isotherms.

**Rehydration process**

The rehydration process occurring during cooking and overcooking depends on four main phenomena according to Del Nobile et al. (2003b): 1. melting kinetics of crystalline starch domain, 2. water diffusion, 3. relaxation of the macromolecular matrix (swelling) and 4. residual deformation release.
The phenomena are described and explained in the following two paragraph according to the model of the same reference (Del Nobile et al., 2003b). Water diffusion is controlling water uptake at the beginning of the rehydration process. The diffusion occurs through two phenomena at the same time: A. Molecular diffusion (related to Brownian movement) is driven by the concentration gradient of water (low in the core of the pasta and high at the surface). B. Macromolecular matrix relaxation driven by the disequilibrium of the local system. The second effect means that when water hits the macromolecular matrix (that is starch and protein), the matrix does not take up all the water needed to reach its equilibrium immediately. The matrix swells over time and the kinetics of the swelling is dependent on the level of disequilibrium.

Melting of starch crystals requires a minimum temperature, but is then a fast process compared to the processes mentioned before. The fourth phenomena (release of residual deformation) takes into account that the continuous protein phase is during drying frozen into a state which is not in equilibrium. During hydration and melting of the starch crystals the reformation can occur which reduces the macromolecular matrix. However the fourth phenomena is outweighed by the amount of water taken up, which leads to an increase in spaghetti size during the rehydration process.

Several models predicted a moisture profile in e.g. spaghetti as shown in Figure 7.3. The rapid increase at low moisture contents indicate that first during starch melting the matrix takes up significant amount of water (Del Nobile et al., 2003a). Furthermore, it is of importance for the models that, as temperature uniformity is reached within seconds, the temperature within a solid pasta product can be considered to be uniform and to be equal to the surrounding boiling temperature (Fasano et al., 2011).

Fickian diffusion is only valid, when no other effects occur at the same time (e.g. it might not be valid near the glass transition; Villeneuve and Gelinas, 2007). When water migration in pasta shall be described, the other before mentioned effects have either to be ignored or to be taken into account to some extent. Several authors based there modelling on Fick’s second law, but adapted it to various degrees. McCarthy et al. (2002) and Horigane et al. (2006) assumed for the whole cooking process a constant diffusion coefficient whereas Cafieri et al. (2008) separated it into two coefficient constants; one for the undercooked region and one for the fully gelatinised region.

In a recent paper, Zhu et al. (2011) stated that in earlier studies it was often tried to model water absorption in swelling foodstuffs by adding empirical factors to the classical models. The authors used the empirical data of Cafieri et al. (2008) and developed a finite
Table 7.1.: Reported diffusion coefficients for pasta products

<table>
<thead>
<tr>
<th>Product</th>
<th>Parameter</th>
<th>$D [\mu m^2 s^{-1}]$</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spaghetti, gelatinised parts</td>
<td>Boiled 10 min</td>
<td>550</td>
<td>Horigane et al. (2006)</td>
</tr>
<tr>
<td></td>
<td>Boiled 12 min</td>
<td>490</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Boiled 10 min, hold 60 min</td>
<td>24</td>
<td></td>
</tr>
<tr>
<td>Wheat noodles</td>
<td>Boiled 6 min</td>
<td>654</td>
<td>Maeda et al. (2009)</td>
</tr>
<tr>
<td></td>
<td>Boiled 10 min</td>
<td>569</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Boiled 15 min, hold 30-120 min</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td>Lasagne</td>
<td>Boiled 11 min</td>
<td>290</td>
<td>McCarthy et al. (2002)</td>
</tr>
<tr>
<td></td>
<td>Boiled 15 min</td>
<td>310</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Boiled 11 min, during holding</td>
<td>16</td>
<td></td>
</tr>
<tr>
<td>Spaghetti, gelatinised</td>
<td>Boiled 11 min</td>
<td>670</td>
<td>Zweifel (2001)</td>
</tr>
<tr>
<td>Spaghetti, fully gelatinised</td>
<td>Boiled 20 min</td>
<td>960</td>
<td></td>
</tr>
<tr>
<td>Spaghetti, ungelatinised</td>
<td>Model parameter</td>
<td>- 600</td>
<td>Del Nobile et al. (2003b)</td>
</tr>
<tr>
<td>Spaghetti, fully gelatinised</td>
<td>Model parameter</td>
<td>- 3000</td>
<td></td>
</tr>
<tr>
<td>Spaghetti, fully gelatinised</td>
<td>Model parameter</td>
<td>630</td>
<td>Caferi et al. (2008)</td>
</tr>
<tr>
<td>parts</td>
<td>Model parameter</td>
<td>2000</td>
<td>Fukuoka et al. (2000)</td>
</tr>
</tbody>
</table>

Element analysis taking into account the simultaneous effects of water absorption and viscoelastic deformation. The porosity was also considered, as according to the authors, dried pasta is a dehydrated system and pores can have been created during dehydration. Thus, the food system is unsaturated from the beginning. The moisture transport during the hydration process is the sum of diffusion and viscoelastic effects and thus, the water transport is dominated by non-Fickian diffusion (Zhu et al., 2011).

The model of Caferi et al. (2008) is a simplified version of Del Nobile et al. (2003a) who tried to describe all the occurring processes separately. Recently, a rather complex model with moving boundaries was presented (Fasano et al., 2011). They defined three boundaries: The outer surface, the gelatinisation and the water front and modelled parameters for every zone.

It was shown in another study, that the diffusion coefficient is also dependent on the temperature and the dependency can be adequately described with the Arrhenius equation (Cunningham et al., 2007; Ogawa et al., 2011). A selected list of reported diffusion coefficients for some pasta products during cooking as well as during warm holding is given in Table 7.1.

All the models so far concentrated mostly on the geometrical aspect of the hydration process. The direct influence of certain raw materials on the pasta hydration process, however, has not been modelled yet. Del Nobile et al. (2003b) used lab-manufactured spaghetti using different wheat types, but could not relate any difference to the properties of the raw materials used.

It seems, that for so far the influence of raw materials on the pasta hydration process is described only qualitatively and based solely on empirical research.

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7.2. Nuclear magnetic resonance and magnetic resonance imaging

Nuclear magnetic resonance spectroscopy (NMR) allows to determine physical and chemical properties of molecules and related properties such as mass transport or compound concentrations in a sample (e.g. moisture content). Magnetic resonance imaging (MRI) is an imaging application of NMR; it adds a spatial information to NMR signals, thus images can be established (Falcone et al., 2006).

Both techniques are non-invasive and need often only little sample preparation. The techniques use magnetic nuclei (such as charged protons) which absorb and re-emit electromagnetic radiation in a magnetic field. The hydrogen proton ($^1$H) is frequently used (hydrogen is abundant in organic compounds and in water), but charged protons of various elements can be analysed. The protons are aligned to the same spin orientation in a strong magnetic field (Falcone et al., 2006). They are excited and their orientation is shifted in steps of 90° by applying for a short time a pulse frequency (consisting of a superimposed magnetic field gradient). When the pulse frequency stops, the protons will relax (turn back to original orientation) and during this relaxation emit a response signal, which depends on their physicochemical environment (Cabrer et al., 2005). By varying the strength of the superimposed magnetic field, spatial localisation of the recorded signal is possible. Signal intensity depends on proton density as well as relaxation times (T1, T2) and the intensity can be modulated by the experimental parameters repetition time (TR), angle of the excitation pulse and echo time (TE; Cabrer et al., 2005). A detailed description of the technique can be found elsewhere (e.g. Schmidt, 2007).

The relaxation time can be defined as the specific time for the protons to rotate back to the equilibrium. Two types of proton relaxation time, longitudinal (T1) and transverse (T2), can be measured and can be related to the molecular water mobility. Varying relaxation time constants can be correlated to more mobile and more bound water fractions, with longer relaxation times being associated with more mobile water or higher moisture content, respectively (Figure 7.4; Wang et al., 2004; Hills et al., 1996). T1 and T2 are furthermore influenced by the temperature (Mohoric et al., 2004). Through variation of the experimental parameters of repetition time and echo time, the peak intensity can be modulated. Longer echo times reduce generally signal amplitude (Ruan and Chen, 1998).

MRI is the preferred method to study water migration. Signal intensity is directly correlated to proton density which in turn gives the correlation to water content. Ruan and Chen (1998) made the analogy to conventional microscopy, that while in microscopy staining agents are giving the contrast, in MRI protons determine the contrast.

Figure 7.4.: The example illustrates association of relaxation times and moisture content. Numbers indicate moisture content in a soft wheat pasta sample. Reproduced from Hills et al. (1996)
Three steps are necessary to develop moisture content maps from NMR measurements. At first, a scan of the sample is carried out (with a resolution of a certain number of voxels [a 3D-pixel]) and the signal intensity for each voxel at various echo times is recorded. Via Fourier transformation of the intensity values of the sequential images, T2 maps can be calculated. The relation between moisture content and T2 values is then established by a calibration curve. The calibration is carried out by determining T2 values for samples with known moisture content, using the same NMR equipment as for the latter experiments. In the case of pasta, the resulting calibration curves showed both linear and non-linear relationships, depending on the moisture content (see Figure 7.5, Horigane et al. (2006); Kratzer (2007)). An intermediate T2 map and a resulting moisture profile is shown in Figure 7.6 and for comparison a moisture content map in Figure 7.7 (Irie et al., 2004; Horigane et al., 2006).

The voxel size determines the image resolution. In many applications, the spatial resolution is much higher than the longitudinal resolution making it difficult to correlate MRI-results to specific effects of the microstructure of a sample. In the example of Horigane et al. (2006), spatial resolution was $78 \times 78 \, \mu m^2$, but the longitudinal resolution
Figure 7.7.: Moisture content maps of spaghetti (A) cooked for 2-14 min and (B) cooked for 12 min and hold for 0.5-15 hours. Reproduced from Horigane et al. (2006)

1,000 μm. The resolution of an image can be improved by increasing the number of scans. However, the limiting factor is the time to record the scans (in the above-mentioned example scan time was at around 2 min). The use of MRI is further limited at low moisture contents, as the signal to noise ratio becomes problematic (Ruan and Chen, 1998).

MRI has been used to study changes in moisture profiles in spaghetti during drying (Hills et al., 1997; Xing et al., 2007) and during cooking and post-cooking (Hills et al., 1996; Horigane et al., 2006, 2009; Irie et al., 2004; McCarthy et al., 2002; Bonomi et al., 2012; Sekiyama et al., 2012). Kratzer (2007) and Kratzer et al. (2008) used prepared wheat endosperms of a well-defined cylinder shape to determine the moisture profiles in excess of water and during limited availability of water at various temperatures.

The MRI method has been expanded to observe water migration in real time using the example of rice kernels (Figure 7.8; Mohoric et al., 2004). Convection in the water would influence the NMR measurement. Therefore, they built a set-up where they could raise the water temperature to 90ºC through a heated air flow. After reaching the temperature, the heater was turned off to minimise convection. The images series showed that neither the microstructure nor the water diffusion were uniform. Finally, the authors developed based on the data a 3D model of water diffusion in rice.

Within pasta analysis, MRI measurements seem to dominate over NMR measurements. Nevertheless, a recent study analysed non-conventional pasta sample ingredients using NMR (Carini et al., 2012). They added soy and carrot ingredients to a standard fresh pasta dough formulation and measured how the T1 and T2 relaxation times were changed. The reference pasta showed two peaks which were attributed to protons of the gluten (0.1-1 ms) and starch domain (2-80 ms), respectively. The gluten peak disappeared for carrot and soy flour formulations and the authors discussed the possibility that this could be correlated to an improper formation of the gluten network.
Figure 7.8: (A) Set up to measure water migration in real time. The rice kernel is embedded in glass wool to fix its position. The tube is filled with excess water. (B) Time series of the central slice of the rice kernel during cooking. Numbers indicate time in minutes since the start of the cooking. Reproduced from Mohoric et al. (2004)

7.3. Near-infrared reflectance spectroscopy (NIR)

NIR spectroscopy uses the near-infrared region of the electromagnetic spectrum. Through the radiation, covalent molecules are excited and an absorption spectrum can be measured. Water is strongly absorbing in the NIR spectrum and therefore, the method can be used to determine moisture contents, but can also describe physical and chemical changes during wheat product processing (Ait Kaddour and Cuq, 2011). Thorvaldsson et al. (1999) measured the local moisture content in a pasta dough during heating. Other developed an in-line NIR technique to determine the average moisture content of freshly extruded pasta and suggested to use the technique as a process control. Advantages of NIR are that it is a non-invasive and fast technique (De Temmerman et al., 2007). Also others want to implement NIR for process control and could distinguish differences in the dough structure (Zardetto and Dalla Rosa, 2006). Ait Kaddour and Cuq (2011) reviewed the usage of NIR in a broader context of wheat product processing.

7.4. Hyperspectral imaging

Hyperspectral imaging has similarities with NIR, but uses a broader range of the electromagnetic spectrum. Several sensors detect the information from different parts of this range in various images and the images are then combined. A proof of concept has been shown to determine contact-less the cooking front in potatoes during cooking (Figure 7.9, Nguyen Do Trong et al. (2011)) and could potentially be used as a rapid monitoring tool.

Figure 7.9: Progress of water migration into potato during cooking analysed with hyperspectral imaging. Extracted from Nguyen Do Trong et al. (2011)
7.5. Gravimetical analysis

By comparing the weight of dry samples with the weight of cooked samples for varying times, hydration kinetics can be established. A similar approach can be used to describe drying kinetics by recording the weight of samples dried for varying times.

Several groups analysed the water uptake of pasta samples during cooking (Cafieri et al., 2010; Petitot and Micard, 2010; Del Nobile et al., 2003b) and Figure 7.10 is showing a typical curve. Measurable parameters besides the weight increase include the determination of the increase in length and in radial size.

7.6. Other methods

The starch in a pasta gelatinises during cooking and loses its crystallinity. This can be visualised using cross-polarised light and hence the position of the water front during cooking can be shown (Del Nobile et al., 2003b).

Gelatinised starch has different texture properties compared to native starch. Again, this can be used to determine the position of the water front or the area gelatinised starch during cooking of pasta. Puncture tests could theoretically be used to determine within a sample the point of the fastest change in measured resistance force which would mark the position of the water front.
8. Analysing quality parameters

The term quality is not a well-defined term and thus it can describe several concepts. In an attempt to define pasta quality, Sisson (2008) collected several factors grouped into three categories of raw material, recipes and processing. For the case of durum wheat, Troccoli et al. (2000) showed that quality parameters can vary dramatically between interested groups along the value chain (Figure 8.1). This chapter concentrates on quality parameters which are important for consumers as well as how these parameters can be determined.

In general, the major quality parameters for the perception of a food are appearance, texture and flavour. For pasta they are appearance and textural properties (Cole, 1991).

Pagani et al. (2007) list requirements from Italian consumers especially concerning the appearance and the cooking quality of pasta: Pasta shall have a typical yellow colour and have an absence of black specks, white spots and fractures or fissures. Concerning the cooking quality, the pasta shall have an optimum consistence as well as being non-sticky and non-bulky. A firm and elastic structure of the cooked pasta is often referred to as “al dente” quality, however this attribute is not an universal preference. The optimal cooking time preferred by consumers can vary dramatically between countries (Kill and Turnbull, 2001) and in some countries such as Brazil in general softer textures are preferred as there pasta is often produced using farina (Marchylo et al., 2004). In such countries, 'al dente' would be experienced as an undercooked state.

In a broader context also the taste and the tolerance to overcooking are included as quality parameters important for consumers (Martinez et al., 2007).

There is no explicit definition for the cooking performance of pasta and hence, in general only a random selection of the below-mentioned parameters is used to describe cooking performance in the various studies. Bruneel et al. (2010) stated in a summary that cooked pasta of good quality shows high levels of water absorption, low cooking loss and a good texture comprising high firmness and low stickiness.

The properties of the cooked pasta depend strongly on the cooking time (and the time until the assessment). A reference for the cooking time was established by using the optimal

<table>
<thead>
<tr>
<th>Durum wheat quality for:</th>
<th>Seed company</th>
<th>Grain dealer</th>
<th>Farmer</th>
<th>Milling industry</th>
<th>Pasta industry</th>
<th>Consumer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Varietal purity</td>
<td>Yield</td>
<td>Semolina yield</td>
<td>Protein content</td>
<td>Cooking quality</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Germination</td>
<td>Grain yield/ quality and stability</td>
<td>Ash content</td>
<td>Gluten quality</td>
<td>Visual appeal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Protein content</td>
<td>Grain uniformity</td>
<td>Granulation and particle size of semolina</td>
<td>Qualitative standard of pasta product</td>
<td>Good Price/ Quality ratio</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Test weight</td>
<td>Test weight</td>
<td>Yellow index</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grain moisture</td>
<td>Grain moisture</td>
<td>Impurity</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 8.1.: Principal aspects of quality for interested groups working with durum. Reproduced from Troccoli et al. (2000)
cooking time (OCT). The American Association of Cereal Chemists defined OCT as the time for a pasta strand to lose its white uncooked core (AACC, 2000). This is determined by pressing a small strand between two glass plates and record the time when the white spot in the core just disappears.

8.1. Colour

Colour attributes are commonly measured with the help of a colorimeter and defined in the L,a,b colour space (also CIELAB). A sample is enlightened with a defined illuminant and the same instrument records light absorbance intensities in comparison to a white spot. Brightness, yellowness and redness are represented by the values L, a and b, respectively, and the values are ranging from 0 to 100 (L) and -1 to +1 (a and b; Tang et al., 1999).

Colour measurements are especially used to instrumentally determine the yellowness of raw and cooked pasta samples (Zweifel, 2001; Majzoobi et al., 2011a; Debbouz and Doetkott, 1996). The yellowness depends on the natural carotenoid content of the semolina, the degradation of the carotenoids by LOX and processing conditions such as storage time of the grain, type of extrusion and drying temperatures (Fu et al., 2011). Fu et al. (2011) showed in their study that pasta yellowness can easily be predicted by analysing the yellowness of the dough.

The undesirable brown component in pasta can be measured as the extent of brownness (Feillet et al., 2000). Brownness is seen to be the opposite to brightness (100-L) and is due to inherent brownness of the semolina as well as the result of Maillard reactions which occur at higher rates during drying at high temperatures (Feillet et al., 2000).

8.2. Texture

The below-mentioned parameters can be determined with a sensory panel or by instrumental techniques and both require standardised methods (Marchylo et al., 2004). E.g. one often standardised parameter is to cook pasta in boiling water in a ratio of 1:10 (Marchylo et al., 2004).

Three of the most often used texture parameters for cooked pasta are described by Kill and Turnbull (2001) using a sensorial definition:

- **Firmness** is the initial resistance to penetration offered by the pasta as it is bitten.
- **Elasticity** describes how pasta breaks down in the mouth on further chewing.
- **Stickiness** is the overall mouth feel of the pasta together with any residual starch left in the mouth after swallowing.

Further parameters can be used for sensory analysis Sozer and Kaya (2003):

- **Adhesiveness** can be evaluated by pressing spaghetti to the palate and determine the force needed to remove it with the tongue.
- **Bulkiness** is the extent to which strands of pasta stick together.
- **Chewiness** is the energy required to chew to the state ready for swallowing
- **Cohesiveness** is the degree to which the sample holds together after chewing
- **Hardness** is the resistance of cooked pasta to the compression by teeth
- **Resilience** is the rate of recovery after deformation.
Sensory analysis is carried out as a panel test and the panel often uses a scale of 1 to 9 with higher numbers indicating a higher quality for respective parameter (Baiano et al., 2006; Sozer and Kaya, 2003). In one study a total score of the sum of several parameters (in this case: stickiness, bulkiness and firmness) was created to describe a total score of cooking quality (Marconi et al., 2000). Besides the mentioned texture parameters other parameters such as aroma are rarer measured. In fact, aroma seems to be of importance only when wholegrain flour or other cereals are incorporated into the pasta dough (Baiano et al., 2008; Feillet et al., 2000; Aravind et al., 2012a).

Manthey and Dick (2012) concluded that sensory tests are useful for internal quality control and to monitor quality changes with time. However, the results may vary between individual testers. The wish to develop more objective tests was therefore one main driving force to develop instrument tests, according to Manthey and Dick (2012). In several studies methods were developed to measure individual quality parameters such as stickiness or firmness (e.g. Dexter et al., 1983; Oh et al., 1983; Sissons et al., 2008b) and a standard method exists to determine firmness (AACC, 2000).

One important example for an instrumental test is the texture analyser, which is an instrument that moves forms of well defined geometries (such as shear blades, plates, balls or needles) at constant speed into and out of a sample and measures continuously the appearing resistance force to record the force over distance. In a texture profile analysis (TPA) several texture parameters can be calculated from this measurement Sozer and Kaya (2003). Five spaghetti strands are aligned side by side and the geometry used is often a shear blade or a plastic tooth (Anonymous, 2008; Martinez et al., 2007; Epstein et al., 2002). Martinez et al. (2007) used this setup to determine the parameters firmness, stickiness and with a slightly changed setup hardness, adhesiveness, springiness, cohesiveness, resilience as well chewiness as as follows:

![Texture Profile Analysis](https://example.com/texture_profile_analysis.png)

Figure 8.2.: Illustration of a texture profile analysis. Reproduced from Epstein et al. (2002)
- **Firmness** was determined as the maximum peak force during the single compression of the spaghetti at low speed until 70% strain (the higher the value, the firmer the spaghetti).

- **Stickiness** was determined by compressing the samples in a similar way until a certain force was reached, hold for 3s and retracted. Stickiness were then defined as the maximum peak force to separate the probe from the samples (higher values indicated stickier samples).

- To determine the following parameters, the samples were compressed twice at higher speed with a time delay of 2 seconds. The first compression was carried out until maximum force was reached and the second compression to 70% of the distance of the first compression (see for comparison Figure 8.2; the figure illustrates a similar texture profile analysis, which differs in how far the compressions shall work (Epstein et al., 2002)).

- **Hardness** was defined as the peak force during the first compression and

- **Adhesiveness** as the negative area during the first probe retraction.

- **Springiness** was defined as to which extent the spaghetti strand moved back to its undeformed state when the force was removed. It was measured as the ratio between the distance of the first peak and the second peak.

- **Cohesiveness** was calculated as the ratio of the areas under the first and second peak, respectively.

- **Resilience** was the ratio of the area under the second half of the first peak to the first half of the same peak.

- **Chewiness** was the product of hardness, cohesiveness and springiness (Martinez et al., 2007; Tang et al., 1999).

In contrast to the before mentioned parameters, elasticity can be measured using a tension test (Tudorica et al., 2002).

It should be noted that Sissons et al. (2008a) question the repeatability of the aforementioned AACC method to measure firmness. In the study of Sissons et al. (2008a) firmness was measured with several instruments in different laboratories and results showed that there was in general a good agreement between laboratories as well as instruments, but in detail some rank variations for various measured samples occurred. In another study the authors analysed various test parameters (such as cooking time, water temperature, cooling time, rest time, number and position of strands, crosshead speed) and suggested a new standardised method (Sissons et al., 2008b). The improvement can be seen in test parameter optimisation and especially in the fact that spaghetti strands were not aligned in contact, but were spaced 1 cm apart. In another work, several probe types were tested to make the AACC method also suitable for short pasta goods such as macaroni (Manthey and Dick, 2012).

The AACC method to determine firmness seems not to be widespread. It rather seems, that researcher use their own methods adapted to the equipment available for them and this is especially true for other parameter such as stickiness or hardness (e.g. Lee et al., 2002; Cubadda et al., 2007; Dexter et al., 1983). However, this makes the comparison of the results more difficult.

Very recently, Fongaro and Kvaal (2013) presented a different approach to evaluate the surface texture of pasta. They used a standard document scanner to acquire images from
cooked pasta samples. They extracted then some image parameters (such as grey level) and could use multivariate analysis to correlate some of the parameters with texture properties. The authors conclude that this set up might be used as an instrument for pasta quality assessment.

8.3. Composition analysis

In the AACC method 66-50 there are some methods described to analyse quantitatively changes in the composition of pasta (AACC, 2000). Very common is to determine the amount of residue in the cooking water, the so called cooking loss (Sissens, 2008).

\[
\text{Cooking loss} \text{ [%]} = \frac{\text{Weight of drained residue in cooking water}}{\text{Weight of dried spaghetti}} \times 100
\]

A higher cooking loss is generally considered to correlate with a lower pasta cooking quality (Tarzi et al., 2012). Cooking loss is influenced by the water composition used (Malcolmson and Matsuo, 1993), does not correlate necessarily with stickiness (Tudorica et al., 2002; Malcolmson and Matsuo, 1993) and maybe misleading when comparing bran-rich and bran-free pasta samples as soluble components can originate from the bran (Edwards et al., 1995).

An alternative to the cooking loss is to determine total organic material (TOM), which is defined as the material released from a pasta surface during exhaustive rinsing (D’Egidio et al., 1982; Cubadda et al., 2007). The higher the amount of TOM, the lower is the cooking quality. TOM describes especially the cooking quality of pasta dried at low temperatures, as cooking quality is dominated by surface characteristics (D’Egidio et al., 1993).

Determining the amount of water absorption or in general the cooked weight (as described in Section 7.5) is another quality parameter as well as the degree of changes in size during cooking (Baiano et al., 2006).
9. Concluding remarks

Pasta quality is influenced by numerous interdependent factors and their implications on pasta production are far from being fully understood. Pasta is a product which has taken a steady development. For a long time, most of the development happened in Italy as pasta played a special role in Italian diet even centuries ago. One indicator for this was the formation of guilds of pasta makers in the sixteenth century in some Italian cities. This led in turn to a professionalised production. Around this time, for example, the wine press known since ancient times was adapted to be able to extrude pasta dough (Serventi and Sabban, 2002). From this starting point more and more equipment was developed and nowadays the once artisanal pasta production is dominated by fully automated industrial production processes (Serventi and Sabban, 2002; Pagani et al., 2007). The development is still ongoing, as it for example has been shown recently by Bühler who presented a new drying line. In this line starch and gluten shall be kept in a rubbery state through most of the drying to reduce the induced stress\(^1\).

These developments and new demands led to new research questions. How can new ingredients and alternatives to durum wheat be incorporated into a pasta product while keeping a high quality? How can the process be adapted to achieve products which keep a high quality even if they are not consumed directly after cooking? Which internal structure is necessary and how is the microstructure influenced by the various process steps?

Several analytical tools are available to study the microstructure of pasta and there is an ongoing adaption of the techniques to achieve better analysis capabilities. As an example, MRI is a very suitable non-destructive method to study the water distribution and movement in pasta, but until now the resolution is not at a level to correlate the local water distribution to components such as single starch granules.

The same is true for modelling. There is a wish to model the processes during manufacturing and cooking to both better understand the underlying mechanism as well as to be able to reduce experiments. However, for so long most models are based on empirical research and the transition to physically-based models (allowing for more transferable know-how) is only happening slowly (Saguy et al., 2011).

Even in testing the finished products, there is a drive to improve the instrumental techniques in order to have an automated alternative to sensory panels.

In conclusion, even after centuries of empirical improvements and research, the field of carbohydrate-rich foods such as pasta still offers plenty of questions yet to be solved.


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Traditional Italian Products from Wheat and Other


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