1. Introduction

Vital gluten can be characterized as a complex mixture consisting of the two main fractions named Gliadine and Glutenine. These fractions determine the physical and chemical properties and they are responsible for the solubility behavior of gluten. The lack of solubility in aqueous solutions is one of the major limitations of its more enlarged use in the food industry. The solution properties are caused by the acid-base properties of the amino acids present, their number, their sequences as well as by the three-dimensional nature of the polypeptide chains.

The high amount of non polar amino acids like leucine, proline and glutamine and the lack of many ionizable side chains such as lysine, arginine, glutamic and aspartic acid together with the hydrogen bond interactions between glutamine and asparagine side chains are the main factors which are responsible for the typical properties of gluten [1-7].

Using gluten in technical fields of application like a cobinder in paper coating colors, like gluten-based hydrolysates and detergents it is necessary to modify this protein for satisfying the technical requirements. Modification of gluten and determination of important technical properties in the different fields of technical applications mentioned above are the content of this paper.

2. Gluten-based cobinders in paper coating colors

2.1 Technical requirements for cobinders in coating colors

Raw paper is a monolayered, flat material consisting of fibres and can be described by a roughened surface characteristic. Important characteristics of high quality paper are excellent appearance and good printability. This can be obtained by coating raw paper with a slurry consisting of the three following basic elements: a mineral coating color pigment, a binder system (binder and cobinder) and other additives which are necessary to prevent troubles in production [8].

Good mechanical stability, good coating and rheological properties, favorable water retention characteristics are important for the suitability of a coating color. These properties are particularly affected by the binding system.

The most important technical requirements of an industrial cobinder are [10,11]

- high pigment binding strength
- control of water retention
- control of viscosity
- control of immobilization of the coating color
- good compatibility with other components of the coating color
- good solubility in water
- low foaming tendency
- good printability of the coated paper
- favourable optical and mechanical properties.

To fulfill these special technical conditions vital gluten must be modified. One possible modification is the type of chemical acylation.
2.2 Chemical modification of gluten
2.2.1 Acylation

Gluten can be easily acylated under mild alkaline conditions with the anhydrides of succinic-, maleic- and acetic acid [12,13].

These reactions are acylations and share the common feature of a nucleophilic attack at an sp²-hybridized or trigonal C-atom. The acylation occurs especially at the free amino groups of the gluten molecule but also the SH, the phenolic OH group, and the imidazole residues of histidine are reactive partners.

Acylation of histidine or cystine, for example, is only observed very rarely because the reaction products tend to hydrolyse very quickly. Serine and threonine do not react in aqueous solutions because their nucleophilic properties are low. The reaction products of these functional groups are unstable and easily became cleared under the conditions chosen. Therefore, it is only the modification of the amino group that builds up stable derivatives.

The most important difference in the performed acylation reactions are the changes in the net charge on the protein. Whilst the reaction with acetic anhydride only converts the positive ammonium ion to non ionizable, neutral amide group, reaction with cyclic carboxylic acid anhydrides results in an additional negative charge via the carboxyl group.

A change in the forces of attraction and repulsion within the polypeptide chain can be expected as a result of different modifications [14,16]. This leads to a change in physical and chemical properties demonstrated by the nitrogen solubility index (NSI), gel permeation chromatography and viscosity measurements [17,18].

The adaptation of gluten for this technical requirement was done by optimizing temperature, reaction time and reagent concentra-

Table 1: Results of testing gluten based cobinders under industrial conditions

<table>
<thead>
<tr>
<th>Mixture</th>
<th>eta mPa's</th>
<th>WRB sec</th>
<th>Glance</th>
<th>IGT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Industrial system</td>
<td>110</td>
<td>21</td>
<td>42</td>
<td>3</td>
</tr>
<tr>
<td>Reference system I</td>
<td>536</td>
<td>87</td>
<td>38</td>
<td>3-4</td>
</tr>
<tr>
<td>Reference system II</td>
<td>1230</td>
<td>76</td>
<td>37</td>
<td>3</td>
</tr>
<tr>
<td>Gluten Suc.</td>
<td>620</td>
<td>49</td>
<td>40</td>
<td>3</td>
</tr>
<tr>
<td>Gluten Ac.</td>
<td>1040</td>
<td>34</td>
<td>35</td>
<td>3-4</td>
</tr>
<tr>
<td>Gluten Ma.</td>
<td>1350</td>
<td>28</td>
<td>42</td>
<td>5</td>
</tr>
</tbody>
</table>

Gluten Suc.: succinylated gluten
Gluten Ac.: acetylated gluten
Gluten Ma.: maleylated gluten

The first column shows the different systems under investigation. The systems were characterized by the properties of viscosity, water retention behavior, glance measured by specular 45° reflectance and the picking up resistance.

Regarding the viscosity of the products ase- on the viscosity is very high but it is still within the range of application of the commercial products. The range of values for the WRB of the industrial and the reference systems I and II are within the range of 21 to 87 sec. Comparable values are measured for the gluten derivatives.

Regarding the values of glance and picking up resistance it can be seen that only the picking up resistance of maleic modified gluten differs clearly from the other systems under investigation.
Comparing the results of technical standard cobinders with our gluten derivatives it can be seen that the main technical requirements are fulfilled by modified gluten especially by gluten derivatives modified with succinic anhydride.

3. Gluten-based hydrolysates

It is well known that products with good surface-active properties (emulsifiers and detergents) can be prepared by condensation of fatty acids, fatty acid chlorides or fatty acid methylesters with polypeptides derived from proteins (especially collagen) by hydrolysis [22-25]. Therefore, we looked at hydrolysate of gluten in this field of application.

3.1 Preparation of gluten by hydrolysates

The conditions and the methods of preparing gluten hydrolysates are shown in Fig. 1.

![Figure 1: Methods and conditions of preparing gluten hydrolysate](image)

Vital gluten was stirred in an alkaline medium under controlled conditions at different pH, temperature and reaction time. The treatment leads to a different degree of desamination. The reaction mixture was separated by centrifugation and the gluten hydrolysates were received by spray drying the supernatant.

The efficiency of this hydrolysation technique can be estimated by testing the functional properties such as:

- nitrogen solubility index (NSI)
- emulsifying activity (EA)
- emulsifying stability (ES)
- rheological properties
- foaming properties

3.2 Characterization of gluten hydrolysates

3.2.1 Nitrogen solubility index (NSI)

The solubility behavior of gluten hydrolysates were characterized by determining the nitrogen solubility index [26,27], which is expressed as:

\[
\text{nitrogen content in the supernatant at different pH-values} \bigg/ \text{total nitrogen content at different pH-values}
\]

The results are shown in Fig. 2.
3.2.2 Emulsifying properties

The determination of emulsifying properties \([28,29]\) can also be used as an indicator for the degree of hydrolysis.

3.2.2.1 Emulsifying activity (EA)

This property was determined with a slight modification according to the method developed by YASUMATSU \([28]\).

7 g protein were added to 100 ml of water and stirred at low speed till having achieved a homogenous distribution of protein in water. 100 ml of oil was added and the mixture was blended by an ultraturrax at high speed for one minute. Afterwards the resulting emulsion was centrifuged at 7,000 rpm for 5 minutes.

The emulsifying capacity was expressed as

\[
EA(\%) = \left(\frac{\text{height of emulsified layer}}{\text{height of total layer in the centrifugal tube}}\right) \times 100
\]

3.2.2.2 Emulsifying stability (ES)

The emulsifying stability was determined like the emulsifying activity except that the emulsion was heated to 80 °C for 30 min subsequently cooled before centrifugation at 7000 rpm for 5 min.

The stability was measured as

\[
ES(\%) = \left(\frac{\text{height of the layer after heating}}{\text{height of the total layer in the centrifuge tube}}\right) \times 100
\]

Table 2 summarizes the results of the emulsifying properties.

---

Figure 2: Nitrogen solubility index (NSI) as a function of different preparation conditions of gluten hydrolysates

Hydrolysate prepared by treating vital gluten at pH 8 shows a good solubility over the whole range between pH 1 und pH 12. A quite different behavior of solubility was received by preparing gluten hydrolysates at pH 10 and pH 12. Gluten hydrolysates prepared at pH 10 possess a small solubility within the range of pH 4.5-8. Also decreasing solubility of the sample treated at pH 12 can be detected within the range of pH 4.5-6. At pH values higher than 8 these two samples show a behavior of solubility which is comparable to those of the hydrolysates treated at pH 8.

These results illustrate that the solubility behavior is directly connected with the methods of preparation.
Table 2: Emulsifying properties as a function of different conditions of preparation

<table>
<thead>
<tr>
<th>Time of treatment min</th>
<th>Emulsifying activity %</th>
<th>Emulsifying stability %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 8</td>
<td>pH 10</td>
</tr>
<tr>
<td>30</td>
<td>40.0</td>
<td>79.5</td>
</tr>
<tr>
<td>60</td>
<td>59.5</td>
<td>81.5</td>
</tr>
<tr>
<td>120</td>
<td>54.2</td>
<td>83.0</td>
</tr>
</tbody>
</table>

Regarding the pH dependence at 30 min, it can be seen that increasing pH leads to an increase of emulsifying activity. This trend is also valid for 60 and 120 min. of alkaline treatment. For each pH, no correlation exists between the emulsifying activity and the duration of treatment.

The same behavior is reflected by the values determined for the emulsifying stability. There is a trend of enhanced emulsifying stability with increasing pH, whereas no trend can be measured regarding the time dependence of pH.

3.2.3 Foaming properties

In addition to emulsifying properties, foam expansion and foam stability are of technical importance [30]. These parameters were measured by the stirring method as follows.

An aqueous solution of 1% hydrolysate was stirred at 2000 rpm for 2 minutes. The suspension was filled into a 100 ml cylinder and the foam volume was defined as "foam expansion." The residual foam volume after 30 minutes was determined and the result was designated as "foam stability." The results of our tests are shown in Table 3.

The foam expansion behaves like the emulsifying properties, i.e., for each reaction time an increase of the values with increasing pH can be observed.

A quite different behavior was found for the foam stability. The time of treatment at different pH-values doesn't have any significant influence on foam stability but the change of pH is very important for foam stability. The foam stabilities of systems prepared of gluten hydrolysates at pH 10 are very poor.

4. Gluten-based detergents

As mentioned before [3.1], it is well known from the literature that acylated protein hydrolysates, especially collagen derivatives, are used as detergents. In this respect, we tried to substitute hydrolysates of collagen by hydrolysates of gluten in the acylation reaction.

The acylation of gluten hydrolysates was done with dodecenylsuccinic anhydrid described in Fig. 3.

Figure 3: Reaction pathway of acylating gluten hydrolysates
The reaction leads to mono- and diacylated products, which can be characterized by an additional carboxylic group according to the number of reactive centers [31].

The following reaction parameters were varied:

- gluten hydrolysates
- temperature
- concentration
- pH-value.

The efficiency of the reaction was tested by comparing the foaming volume according to DIN 53902. This method was used for screening and optimizing our different experiments.

Comparing the acylated gluten hydrolysates with technical relevant systems, especially those based on collagen, it can be seen that the use of modified gluten hydrolysates produces a foaming volume with satisfactory quality characteristics (Fig. 4).

Systems showing technical relevant foaming volumes were furthermore characterized by the determination of the critical micelle concentration (CMC) which is the main criterium for detergents [32]. The CMC was measured by the method of interfacial tension [33-36].

Reaching the CMC, substances with surface active properties form micelles. From this point the interfacial tension stays constant.

Such a point can be measured by our acylated gluten derivatives. Therefore these systems can be regarded as detergents. More intensive characterizations of these systems are the subject of consecutive experimental work.

5. Conclusion

Examples are given demonstrate the possibility to use gluten respectively modified gluten in non food industry. The results are received only on a laboratory scale. The application of these gluten-based products on industrial scale will be a question of economy.

These products have to compete with a lot of synthetic products and they will only be used in industry if the technical properties of these products are much better than those of synthetic ones and, or if from these products profit may be expected.
With respect to the enormous expansion of the production of wheat starch in future connected with an increasing amount of the by-product gluten it is expected that the price of vital gluten will fall and, therefore, the application of natural products like gluten in non food industry will get more importance.

6. Literature Cited


NON-TRADITIONAL APPLICATIONS OF WHEAT GLUTEN

J.M. VEREIJKEN, H.J. KUIPER and J.C. KOK
Agrotechnological Research Institute (ATO-DLO),
PO box 17, 6700 AA Wageningen, the Netherlands

ABSTRACT

In our research aimed at the development of new applications of wheat gluten two approaches are being followed: a) the special functional properties of gluten are exploited and b) new functional properties are conferred to gluten. The latter is achieved by modification of the proteins and is the subject of this paper. Results are reported obtained by three types of modification, i.e. succinylation, deamidation and proteolytic hydrolysis. By these modifications the solubility of gluten at neutral pH can be increased from 5% to 70% (by succinylation) or to 90% (by deamidation and proteolytic hydrolysis). Foaming properties are also improved by these modifications. Some of the modified glutens have foaming properties superior or equal to those of egg white.

INTRODUCTION

The production of wheat gluten has increased considerably in the last decade. In the European Community (EC) its production has doubled; in the Netherlands the production has even risen by a factor of five and amounts about 40,000 tons per year. The total world production capacity in 1988 was estimated to be about 330,000 metric tons (1). The main outlet for gluten is in the bread baking industry, in which gluten is used as an additive to improve the breadmaking quality of wheat flour.

In view of the increased production of gluten and to decrease the dependency of the wheat starch industry on the use of gluten as a breadimprover, development of new applications of gluten is required. Industrial uses of proteins are dependent on their set of functional properties, i.e. their properties relevant for a given application. To develop new applications, two approaches can be followed:

a. explication of the special functional properties of wheat gluten,

b. conferring new functional properties to wheat gluten.

Amongst industrial proteins, wheat gluten is unique because it possesses viscoelastic properties. The traditional application, as a breadimprover, is based on this functional property. However, other applications based on this property can be foreseen. An example is the development of environmentally safe coatings and films based on wheat gluten.