

Research Insight

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Innovative Approaches in Wheat Starch and Gluten Separation: Techniques, Functional Modifications, and Emerging Applications

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Abstract This study analyzes the separation technologies of wheat starch and gluten, including wet separation, dry separation, and emerging technologies such as ultrasound-assisted separation, enzymatic separation, microfluidic separation, and the synergistic separation of electric and magnetic fields. The advantages and limitations of different methods are discussed in detail. Additionally, the impact of separation processes on the functional properties of starch and gluten is explored. The study finds that combining ultrasound and enzymatic methods can effectively improve separation efficiency while reducing damage to the protein and starch structures. Furthermore, the study summarizes functional modification technologies, such as physical (microwave, γ -rays), chemical (esterification, oxidation), and biological (enzymatic hydrolysis) modifications, and their role in optimizing the functionality of wheat starch and gluten. The emerging applications of these modifications in fields such as food, environmental materials, and biomedicine are also analyzed, including low-GI foods, biodegradable packaging, and biomaterial scaffolds. This study provides scientific evidence and necessary references for the separation and high-value utilization of wheat starch and gluten. **Keywords** Wheat starch; Gluten; Separation technologies; Functional modification; Green processing

1 Introduction

Wheat is one of the most widely planted food crops in the world, not only a staple food for people, but also an important industrial raw material. In the process of wheat processing, the two key components separated - wheat starch and gluten (gluten protein) - are widely used in various fields such as food, medicine, and even construction. High purity wheat starch, with thickening, gel and stabilization functions, is an important source of food thickeners, as well as an important raw material for bioplastics and pharmaceutical excipients. The unique viscoelasticity of gluten allows it to play a critical role in baking, plant-based foods, and functional protein products (Van Der Borght et al., 2005; Day et al., 2006).

At present, with the increasing application of wheat starch in food thickeners, bioplastics, adhesives, and pharmaceutical excipients, the demand for high-purity starch in the market is also continuing to grow. At the same time, improving separation efficiency can not only obtain purer starch, but also minimize gluten contamination. The efficient separation and purification of starch and gluten has become an important step in enhancing the processing value of wheat and achieving product customization (Peigabardost et al., 2008). However, traditional separation techniques such as wet grinding are not efficient and the separation effect is not ideal. Therefore, researchers are gradually turning their attention to more advanced and intelligent new separation technologies.

In recent years, some innovative separation technologies have gradually entered people's vision, such as the "shear flow migration technology", which can greatly improve separation efficiency and product purity (Peigabardost et al., 2008). In addition, new treatment methods such as "water phase ozone modification" have also shown the potential to change the structure and function of gluten, and can be customized to meet the needs of different industries (Fan et al., 2024). Not limited to wheat, Al-Hakkak and Al-Hakkak (2007) also proposed a method for extracting starch and gluten from non wheat plants, which provides new ideas for the cross species application of separation technology and the comprehensive utilization of plant resources.



With the continuous advancement of technology, separation efficiency and product functionality are gradually improving, and the economic value of starch and gluten is also increasing. This study will systematically review the development history and latest achievements of these new technologies, with a focus on innovation in separation processes, new ideas for functional modification, and emerging applications in the food, pharmaceutical, and new materials industries. I hope that through these analyses, we can provide reference for the industry on the current challenges, technological breakthroughs, and future trends related to wheat separation, and help promote the sustainable and high value-added development of wheat processing.

2 Composition and Structural Characteristics of Wheat Starch and Gluten

2.1 Structural characteristics of wheat starch

Wheat starch is the primary carbohydrate component in wheat flour, and its granules are mainly composed of a mixture of two polysaccharides: amylose and amylopectin. The ratio of these two polysaccharides affects the functional properties of starch, such as water absorption, swelling power, and gelatinization. Li et al. (2020a) found that high amylose wheat starch (HAWS) has a higher content of amylose and a lower degree of branched starch chain branching. It has a more linear molecular structure compared to ordinary wheat starch, manifested by stronger HAWS viscosity and stability. Due to the looser arrangement of polymers within the granules, HAWS also shows stronger water absorption capacity.

The crystalline structure of wheat starch is generally classified as A-type, which is characteristic of amylose. This structure is marked by relatively weak crystallinity and poor water solubility; however, it remains stable even under treatments such as ultrasonication (Karwasra et al., 2020). Karwasra et al. (2020) suggested that the A-type crystalline structure of wheat starch contributes to its relatively high gelatinization temperature and distinct pasting properties, which ultimately affect the quality of wheat-based products. During processing, the surface properties of wheat starch granules - including surface lipids and proteins - influence the ease of starch separation. Furthermore, after processing, the relative crystallinity of starch may change, which further affects its functional properties such as swelling power and oil absorption capacity (Wang et al., 2021).

2.2 Composition and functional properties of wheat gluten

Wheat gluten is a protein complex primarily composed of two types of proteins: glutenin and gliadin. It also forms the viscoelastic network essential for the unique dough properties of wheat flour. Gliadin and glutenin provide complementary functional properties, jointly determining the extensibility and elasticity of the dough. Meanwhile, their ratio significantly influences the pasting properties, thermal stability, and structural characteristics of gluten-starch mixtures. Furthermore, changes in the glutenin-to-gliadin ratio also alter the viscosity and thermal stability of the mixture, ultimately affecting the quality of wheat-based products (Li et al., 2020b).

The functional properties of gluten, including extensibility and elasticity, largely depend on its protein structure. For example, the disulfide bonds within gluten and the secondary structures of its proteins, such as α -helices and β -sheets, together determine its ability to form a network. This protein structure can be modified through mechanical, chemical, or enzymatic treatments. Liu et al. (2021) found that physical modification methods, such as planetary ball milling, can disrupt the natural structure of gluten, reducing its particle size and decreasing its crystallinity. These structural changes further alter the surface hydrophobicity and foaming capacity of gluten, expanding its potential applications in food products.

2.3 Interfacial binding mechanism between starch and gluten

Starch granules are embedded within the gluten network, forming a complex structural matrix that ultimately affects the physical properties of the dough. Therefore, the interaction between starch granules and gluten proteins is one of the key factors determining dough properties. The particle size distribution of starch granules, particularly the ratio of A-type to B-type granules, significantly influences this interaction. Studies have shown that the higher the B/A/porosity ratio (i.e., the greater the proportion of B-type granules), the more stable the dough tends to be, which is attributed to the closer binding between starch granules and the gluten network (Gao et al., 2020; Yu et al., 2021). Figure 1 compares the starch granule morphology and gluten network formation characteristics of three wheat varieties: Xinong 979, Zhengmai 7698, and Xinong 836. When the starch granules



are evenly distributed and have smooth surfaces, gluten chains can more easily wrap around the granules, forming a continuous network (Li et al., 2023). In addition, Li et al. (2023) also found that the presence of smaller starch granules can enhance the binding strength between gluten and starch, leading to a more stable dough structure.



Figure 1 Starch granules and the protein network structure in the three wheat varieties (Adopted from Li et al., 2023) Image caption: Images of isolated starch granules and the protein network structure in dough samples produced from Xinong 979 (A,D,G), Zhengmai 7698 (B,E,H), and Xinong 836 (C,F,I). (A-C) Morphology of starch granules observed by scanning electron microscope at a magnification of 1 000×. (D-F) Original stained images of dough obtained by laser scanning microscopy, where the scale bar represents 50 μ m. (G-I) Gluten network structure processed with AngioTool, where white represents the junctions, blue represents the protein skeleton, yellow represents the protein outline/area, and the black area is filled with starch granules (Adopted from Li et al., 2023)

During the dough formation process, the interaction between starch and gluten is mainly regulated by their physicochemical properties. The gluten network forms a continuous three-dimensional structure, while starch granules act as fillers distributed within it. The stability of this network depends on the combined actions of disulfide bonds and hydrogen bonds, which determine the dough's viscoelasticity (Zhang et al., 2018; Peng et al., 2022). Moreover, strong intermolecular hydrogen bonds can form between high-molecular-weight gluten subunits, further enhancing the dough's elasticity and increasing its strength, leading to stability (Shewry et al., 2002).

The thermal properties and gelatinization characteristics of starch also affect the interaction between starch and gluten. A stronger protein-starch interaction can inhibit starch gelatinization, thereby improving the dough's



stability and resistance to enzymatic degradation (Peng et al., 2022). In addition, the physicochemical properties of starch, such as amylose content and granule morphology, also play an indispensable role in influencing the rheological properties of dough. These factors determine the processing performance of wheat products (Cao et al., 2019).

3 Traditional Separation Techniques for Wheat Starch and Gluten

3.1 Wet separation

The dough washing method (dough washing method, DWM) is one of the traditional techniques for separating wheat starch and gluten. The specific process involves mixing wheat flour with water to form a dough, which is then allowed to rest. After sufficient fermentation, the dough is washed with water. Since gluten is insoluble in water, during the washing process, starch particles are carried away by the water, thus achieving the separation of starch and gluten. Additionally, factors such as water temperature and the flour-to-water ratio affect the effectiveness of the washing method. They directly relate to the yield and purity of both starch and gluten during the separation process (Yöndem-Makascioğlu et al., 2002).

Process A and process B are two common procedures in wet separation. Process A typically uses a hydrocyclone separation system. The specific process is as follows: wheat flour is mixed with water, and a hydrocyclone is used to separate the slurry. At this point, starch is washed out with clean water, while gluten, being insoluble in water, spontaneously aggregates into blocks, allowing it to be separated by screening. This process can save water and is more efficient for processing large amounts of flour. However, it requires more complex equipment and precise control of process parameters. Process B, represented by the traditional Martin method, is more manual and relies mainly on hand washing or simple mechanical assistance to separate the dough. Process B requires less investment in equipment, making it more suitable for small-scale production. However, it involves higher labor intensity, and the separation yield and purity are not as high as those of processes using modern equipment for wet separation (Witt and Goldau, 2000).

In wet separation, more advanced technologies include liquid flow classification and centrifugal separation. These methods rely on differences in the density and particle size of gluten and starch. Liquid flow classification uses the impact and dispersion of water flow to separate starch and gluten based on their settling velocities. Centrifugal separation uses centrifugal force to effectively separate gluten, which has a lower density and larger particles, from starch, which has a higher density and smaller particles (Sayaslan, 2004). These technologies are favored in industrial production for their high efficiency and ability to produce high-purity starch and gluten. However, they require high investment in equipment and expertise in operation (Sayaslan, 2004; Van Der Borght et al., 2005).

3.2 Dry separation

Traditional dry separation techniques for wheat starch and gluten include dry milling and sieving. The principle is to grind wheat into powder and use sieving technology to separate based on particle size. However, Remadnia et al. (2014) argue that due to the limitations in effectively separating fine particles, this method is not very efficient in separating high-nutrient components.

Air classification is also a common dry separation technology, which uses differences in particle size and density between starch and gluten to separate them by high-speed airflow. Although air classification can effectively increase the protein content, it cannot achieve high-purity separation due to issues such as powder contamination, equipment fouling, and low overall separation efficiency (Assatory et al., 2019; Silventoinen et al., 2020).

Electrostatic separation utilizes the charge difference between particles and is a more innovative dry separation technique. This method generates different charges on the particles through friction, and then separates them in an electric field based on the charge differences. However, the separation efficiency of this method is affected by particle aggregation effects, which can be minimized by optimizing airflow and field strength (Remadnia et al., 2014; Wang et al., 2015). Although electrostatic separation has potential, the presence of a protein layer on the surface of starch particles often interferes with the separation. Therefore, directly separating starch and gluten from the original flour remains a challenge.



3.3 Limitations of traditional technologies

Traditional separation technologies, especially wet separation, face issues such as high energy consumption and the need for large amounts of water. Additionally, research by Assatory et al. (2019) indicates that such processes typically require long drying times and the use of organic solvents, which may damage the natural structure and functionality of the separated starch and gluten. In contrast, dry techniques such as flow classification and electrostatic separation reduce energy and water consumption, but still have shortcomings in achieving high-purity separation (Assatory et al., 2019; Silventoinen et al., 2020).

A major limitation of traditional techniques is the trade-off between achieving high purity in the separation and maintaining the functional properties of the components. Although dry methods can preserve the natural structure and functionality of the separated components, they often struggle to produce high-purity products (greater than 90%) due to issues like powder agglomeration and low yield. Additionally, mechanical forces involved in processes like milling may alter the structural properties of gluten, affecting its functional quality (Liu et al., 2021).

4 Innovative Separation Technology for Wheat Starch and Gluten Powder

4.1 Ultrasound assisted separation

Ultrasonic assisted separation is an innovative technology that utilizes high-frequency sound waves to disrupt the composite interface between starch and gluten powder. Its principle is to generate cavitation bubbles in the medium by applying ultrasonic wave. When the foam collapses violently, micro jet flow and shock wave will be generated, which can physically destroy the matrix of starch and gluten, thus promoting the separation of starch and gluten. The separation efficiency of breaking the bond between starch and gluten powder using the mechanical action of ultrasound is higher than traditional methods (Van Der Borght et al., 2005; Peigabardoust et al., 2008).

The efficiency of ultrasound assisted separation is related to the intensity of ultrasound. Higher ultrasound intensity increases the energy input of the system, enhances cavitation, and thus improves separation efficiency. However, when the ultrasound intensity is within the optimal range, excessively high intensity may lead to degradation of gluten powder quality or excessive fragmentation of starch particles, thereby negatively affecting the purity and yield of the isolate (Zalm et al., 2009; Liu et al., 2021).

4.2 Enzymatic separation

Enzymatic separation utilizes selective enzymatic hydrolysis to disrupt the protein starch network. Specific enzymes, such as proteases, target peptide bonds in gluten proteins, weaken the gluten network, and promote the release of starch granules. The advantage of this method is that it can finely regulate specific components of the gluten network without affecting the starch, thereby maintaining the integrity and functionality of the separated starch (Van Der Borght et al., 2005).

Optimizing the combination of enzymes can improve separation efficiency by selecting appropriate types and concentrations of enzymes, customizing the enzymatic hydrolysis process, and achieving maximum separation efficiency in the shortest possible time. At the same time, factors such as enzyme activity, temperature, pH value, and reaction time must be taken into account to ensure that the enzyme can effectively decompose the gluten network while maintaining starch quality (Van Der Borght et al., 2005; Al-Hakkak and Al-Hakkak, 2007).

4.3 Microfluidic separation technology

Microfluidic separation technology is an efficient and promising method for separating starch and gluten particles. It utilizes microfluidic channels to achieve nanoscale separation, which has advantages in maintaining the functional characteristics of separation components. This technology is based on passive sorting by particle size, which helps to avoid the common clogging problem in traditional separation methods. By changing the boundary interfaces in microfluidic arrays, uniform flow patterns can be achieved, thereby improving the separation efficiency of the entire array (Inglis, 2009). This precise control at the nanoscale ensures that the functional properties of starch and gluten powder are preserved, making it an efficient technology that requires high-purity and high-quality separation components for application.



4.4 Joint separation of electric and magnetic fields

The combined use of electric and magnetic fields provides an innovative method for separating wheat starch and gluten powder. Electric field separation utilizes the difference in surface charge between starch and gluten particles. This method can selectively separate them based on their charge characteristics and can be finely adjusted to optimize separation efficiency. In addition, magnetic field separation involves the use of adaptive magnetic separation agents that can selectively bind to a certain component, thereby promoting its removal from the mixture. This dual method not only improves the accuracy of the separation process, but also reduces the need for mechanical processing, which may lower the quality of gluten powder (Peigabardoust et al., 2008; Zalm et al., 2009). By combining these two fields, the separation process becomes more efficient and causes less damage to the functional properties of gluten powder, making it suitable for the production of high-quality gluten powder.

5 Effects of Separation Processes on Functional Properties of Starch and Gluten 5.1 Changes in starch functional properties

The integrity and crystallinity of starch granules are significantly influenced by various separation processes. For instance, defatting treatments on wheat starch result in more pores on the granule surface, particularly in A-type starch, while B-type granules remain largely unchanged. This treatment does not alter the X-ray diffraction pattern but decreases the crystallinity degree, affecting the starch's structural properties (Li et al., 2016). Similarly, the presence of damaged starch (DS) from mechanical milling impacts the granular integrity, reducing gelatinization enthalpy and altering the viscosity profile, which is solvent-dependent (Teobaldi et al., 2022). Furthermore, the interaction between A-type and B-type starch granules with gluten during dough mixing shows that B starch granules enhance gluten network formation, while A starch granules tend to separate, affecting the overall dough structure (Li et al., 2021).

The gelation properties and pasting behavior of starch are also affected by separation processes. The addition of protein-glutaminase-treated glutenin and gliadin to starch decreases viscosity parameters and alters the gelation properties, resulting in a more compact starch granule morphology (Chen et al., 2018). The removal of surface lipids from starch granules through defatting changes the peak, trough, final, and setback viscosities, as well as gelatinization transition temperatures and enthalpy, indicating significant alterations in pasting behavior (Li et al., 2016). Additionally, the presence of oligosaccharides like fructooligosaccharides (FOS) and xylooligosaccharides (XOS) can inhibit starch retrogradation and modify pasting properties by reducing crystallinity and increasing pasting temperature (Su et al., 2020b). The interaction of starch with gluten and sucrose also modifies pasting profiles, with sucrose increasing the onset temperature of gelatinization and altering the viscoelastic properties of starch gels (Teobaldi et al., 2021).

5.2 Changes in gluten functional properties

The separation processes significantly impact the reformation of disulfide bonds and the reconstruction of protein networks in gluten. During the separation, disulfide-sulfhydryl exchange reactions are promoted, which are crucial for the reformation of gluten's protein network. However, the establishment of new bonds can be restricted due to prior cross-links in the material, particularly in harshly separated gluten (Ceresino et al., 2019). The presence of pentosans and enzymes like xylanase can further influence the re-agglomeration of gluten proteins, affecting the size and aggregation of glutenin macropolymer particles, which are essential for the protein network's integrity (Wang et al., 2004b).

The solubility and water-holding capacity of gluten are affected by the separation process. For instance, the use of water unextractable solids (WUS) can negatively impact gluten yield and its rheological properties, although these effects can be mitigated by increasing mixing time and water content (Wang et al., 2003). Additionally, the emulsifying properties of gluten are influenced by the separation process, as the interaction between gluten and other components like pentosans can alter its functional characteristics (Wang et al., 2004a).

5.3 Functional advantages of environmentally friendly separation processes

Environmentally friendly separation processes, such as those involving mild conditions or the use of enzymes, have shown positive effects on the retention of gluten's functional properties. For example, mildly separated



gluten has been found to improve dough development parameters compared to harshly separated gluten, indicating better retention of functional properties (Ceresino et al., 2019). Moreover, the use of green modification methods, such as UV irradiation and thermal treatment, has been shown to effectively modify starch properties while maintaining functional integrity, suggesting similar potential benefits for gluten (Kurdziel et al., 2019). These processes help preserve the structural and functional properties of gluten, making them advantageous for sustainable food production.

6 Functional Modification of Wheat Starch and Gluten

6.1 Physical and chemical modification of starch

Physical modification of wheat starch includes techniques such as microwave, ultrasound, and γ -ray treatments. These methods alter the structure and functional properties of starch without the use of chemicals. For example, heat treatment, as a form of physical modification, is used to change the starch properties in wheat flour, enhancing its application in the food industry by improving texture and mouthfeel. Van Rooyen et al. (2022) found that forced convection continuous stirring baking (FCCT) is an efficient heating method that uses convective heating to ensure uniform heat distribution in the grains. The formation of a temperature gradient affects the degree of starch gelatinization, protein denaturation, and enzyme inactivation efficiency. At the same time, moisture diffuses from the interior to the surface of the grains and eventually evaporates, reducing the moisture content and improving storage stability (Figure 2). Repeated and continuous heat-moisture treatments (RHMT and CHMT) are also used to modify the molecular, morphological, and physicochemical properties of wheat starch, resulting in changes such as increased solubility and altered gelatinization temperatures (Su et al., 2020a). These physical modifications can improve the functional properties of starch, such as enhanced resistance to digestion and increased adsorption capacity (Xie et al., 2019).



Figure 2 Schematic of convection dry heat treatment of wheat kernels during force convection continuous tumble (FCCT) roasting by means of heat and mass (moisture diffusion) transfer (Adopted from Van Rooyen et al., 2022)

Chemical modification of wheat starch includes methods like acid hydrolysis, oxidation, and esterification. These techniques are used to introduce new functional groups into the starch molecules, thereby altering their physicochemical properties. For example, oxidation and cross-linking with agents like hydrogen peroxide and sodium phytate can significantly modify the structural and functional characteristics of wheat starch, enhancing properties such as solubility, swelling power, and freeze-thaw stability (Sun et al., 2017). Chemical modifications like oxidation and esterification are particularly effective in improving the physicochemical properties of starch, making it suitable for various industrial applications, including drug delivery systems (Masina et al., 2017). Additionally, enzymatic modifications, such as those involving the GtfB enzyme, can reduce retrogradation rates and modify the molecular weight and amylose content of wheat starch (Li et al., 2018).

6.2 Functional modification of gluten

Crosslinking is a pivotal modification technique used to enhance the thermal stability of gluten. The use of microbial transglutaminase (MTGase) following partial hydrolysis with Alcalase has been shown to significantly increase the storage modulus and thermal denaturation temperature of wheat gluten. This process unfolds the compact gluten structure, increasing β -sheet content and surface hydrophobicity, which in turn improves molecular flexibility and exposes additional glutamine sites for crosslinking (Wang et al., 2016). Additionally,



aqueous ozone (AO) treatment has been found to increase cross-linking in glutenin macropolymer (GMP), stabilizing the secondary and tertiary structures of gluten, which enhances its viscoelastic properties (Fan et al., 2024).

Adjusting the hydrophilic and hydrophobic balance of gluten can significantly improve its emulsification properties. For instance, the use of corn syrup in hydrothermal treatment has been shown to affect the emulsifying properties of gluten. Although higher temperatures can adversely affect emulsification, the addition of corn syrup helps restore emulsion stability, particularly at lower pH levels (Singh et al., 2006). This adjustment is crucial for enhancing the functional properties of gluten in various food applications.

Enzymatic hydrolysis is a key method for developing bioactive peptides from gluten. Continuous hydrolysis in an enzymatic membrane reactor (EMR) allows for the efficient production of low-molecular-weight peptides with antioxidant activities. This method overcomes the limitations of batch reactions, such as inconsistent product quality and low productivity, by providing a stable and homogeneous permeate fraction (Cui et al., 2011). The peptides produced through this process have potential applications in health-promoting food products.

6.3 In-situ functional modification during separation

Innovative approaches that integrate separation and modification processes can enhance the functional properties of gluten. For example, the use of aqueous ozone not only improves the yield and purity of separated gluten but also induces specific molecular modifications that enhance its functional properties (Fan et al., 2024). Such integrated strategies are crucial for developing more efficient and sustainable wheat processing technologies. Additionally, the combination of heat-moisture treatment and enzymatic hydrolysis has been shown to improve the structural and physicochemical properties of wheat starch, suggesting potential applications for similar strategies in gluten modification (Xie et al., 2019).

7 Diversified Applications of Wheat Starch and Gluten

7.1 Applications in the food industry

Wheat starch is widely utilized in the food industry due to its functional properties such as thickening, gelling, and anti-aging. These properties are enhanced through various modification techniques, including physical and non-thermal methods, which improve the thermal stability and reduce retrogradation of starch, making it more suitable for diverse food applications (Han et al., 2020). The use of modified starches is crucial in developing food products with desired textures and shelf-life stability (Raghunathan et al., 2020).

Wheat gluten is a key ingredient in noodle products, plant-based meats, and baked goods due to its viscoelastic properties, which are essential for dough formation and texture. The cohesiveness and elasticity provided by gluten are primarily due to the presence of gliadin and glutenin, which form a strong network through covalent and non-covalent bonds (Zhang et al., 2022). Innovative processing techniques, such as the use of aqueous ozone, have been shown to enhance the yield and quality of gluten, further expanding its application potential in the food industry (Fan et al., 2024).

7.2 Functional foods and health products

The development of low-glycemic index (GI) foods is a growing area of interest, with modified starches playing a significant role. Techniques such as enzymatic and physical modifications are employed to alter the digestibility of starch, thereby reducing its GI and making it suitable for functional foods aimed at health-conscious consumers (Han et al., 2020; Raghunathan et al., 2020). These modifications help in creating food products that can aid in better blood sugar management.

Wheat gluten can be enzymatically hydrolyzed to produce bioactive peptides with antioxidant and antihypertensive properties. These peptides are increasingly being incorporated into functional foods to enhance their health benefits. The use of continuous enzymatic membrane reactors has been shown to efficiently produce these peptides, which exhibit strong antioxidant activities, making them valuable for health-oriented food products (Cui et al., 2011).



7.3 Biomaterials and eco-friendly products

The development of biodegradable films and packaging materials from wheat starch and gluten is gaining significant attention due to environmental concerns associated with synthetic plastics. Wheat starch, being abundant and cost-effective, is an excellent candidate for creating eco-friendly packaging solutions. It forms thermoplastic materials and films that can be enhanced with natural plasticizers, making it suitable for sustainable food packaging applications (Lauer and Smith, 2020; Liu et al., 2022; Onyeaka et al., 2022). The incorporation of gluten into these materials further enhances their mechanical properties, allowing for the production of bioplastics that can be reinforced with natural fibers to create biocomposites (Lagrain et al., 2010; Zhang et al., 2022). These starch-gluten composites offer a promising alternative to conventional petrochemical-based polymers, providing a renewable and biodegradable option for food packaging (Cheng et al., 2021; Alibekov et al., 2024).

Starch-gluten composites are increasingly being utilized in food packaging due to their ability to form strong, flexible films that can protect food products while being environmentally friendly. The combination of starch and gluten results in materials with improved mechanical strength and barrier properties, essential for effective food packaging (Abdillah and Charles, 2021; Su et al., 2022). These composites can be tailored to include functional additives, such as antioxidants and antimicrobials, to further enhance their protective capabilities and extend the shelf life of packaged foods (Bangar et al., 2021; Liu et al., 2022). The ongoing research and development in this area aim to overcome current limitations and expand the applicability of starch-gluten composites in the packaging industry (Cheng et al., 2021; Alibekov et al., 2024).

7.4 Biomedicine and tissue engineering

In the field of biomedicine, starch-gluten-based materials are being explored for their potential in tissue engineering applications. The biocompatibility and biodegradability of these materials make them suitable candidates for developing biological scaffolds that can support cell growth and tissue regeneration. The structural properties of gluten, particularly its viscoelasticity and ability to form stable networks, are advantageous in creating scaffolds that mimic the extracellular matrix, providing a conducive environment for cell proliferation and differentiation (Lagrain et al., 2010; Zhang et al., 2022).

Functional gluten peptides are being investigated for their potential use as drug carriers in biomedicine. These peptides can be engineered to enhance drug delivery by improving the solubility and stability of therapeutic agents. The unique properties of gluten, such as its ability to form covalent and non-covalent bonds, allow for the development of peptide-based carriers that can effectively encapsulate and release drugs at targeted sites within the body. This innovative application of gluten peptides holds promise for improving the efficacy and safety of drug delivery systems in medical treatments (Lagrain et al., 2010; Zhang et al., 2022).

8 Future Trends and Research Directions

8.1 Development of smart separation technologies

The integration of sensor-based monitoring and real-time process control in wheat starch and gluten separation processes is a promising area for future research. This approach can enhance the precision and efficiency of separation techniques by providing continuous feedback and adjustments during processing. For instance, the use of non-conventional approaches such as portable NIR devices has been explored for monitoring wheat sprouting, which could be adapted for real-time monitoring of separation processes (Grassi et al., 2018). This technology allows for the assessment of chemical composition and technological changes, which could be crucial for optimizing separation parameters.

Artificial intelligence (AI) can play a significant role in optimizing separation parameters by analyzing large datasets to identify patterns and predict outcomes. AI can assist in fine-tuning the conditions under which separation processes occur, potentially leading to improved yields and quality of separated components. The use of AI in conjunction with sensor data could lead to more adaptive and efficient separation processes, reducing waste and improving product quality.



8.2 Function-oriented separation concepts

Future research should focus on developing separation technologies that preserve the functional properties of gluten and starch based on their intended applications. For example, the use of aqueous ozone has been shown to enhance the yield and purity of gluten while maintaining its viscoelastic properties, suggesting that targeted modifications can improve both the quality and functionality of separated components (Fan et al., 2024). This approach ensures that the separated products meet specific functional requirements for various industrial applications.

Reverse engineering of separation processes, where the desired functional properties of the end product dictate the separation method, is an emerging trend. This concept involves designing separation processes that are tailored to produce components with specific functional attributes, such as enhanced viscoelasticity or improved foaming capacity. For instance, the modification of gluten using planetary ball milling has been shown to improve its functional properties, indicating the potential for reverse design in separation processes. By focusing on the end-use requirements, researchers can develop more efficient and application-specific separation technologies.

8.3 Integration of green separation and carbon reduction technologies

The development of low-energy, low-pollution, and water-saving separation processes is a critical area of focus in the separation of wheat starch and gluten. Innovative methods such as shear flow separation have shown promise in reducing water usage while maintaining gluten quality, offering a more sustainable alternative to traditional methods (Peighambardoust et al., 2008). Additionally, the use of aqueous ozone (AO) in wheat flour processing has been highlighted for its potential to enhance product quality while being eco-friendly, suggesting a pathway towards more sustainable processing techniques. These advancements align with the broader goal of establishing sustainable separation technology standards that minimize the carbon footprint of industrial processes.

8.4 Functional complementarity and synergistic utilization of starch and gluten

Research into the synergistic enhancement mechanisms in composite materials and functional foods is gaining traction. The modification of wheat gluten using techniques such as planetary ball milling has been shown to improve its functional properties, expanding its application potential in various food products. Furthermore, the continuous enzymatic membrane reactor (EMR) has been effective in producing bioactive peptides from modified wheat gluten, demonstrating the potential for synergistic utilization of gluten in functional foods. These studies underscore the importance of understanding and leveraging the functional complementarity of starch and gluten to develop innovative food products with enhanced nutritional and functional benefits.

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Conflict of Interest Disclosure

The authors affirm that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

References

- Abdillah A., and Charles A., 2021, Characterization of a natural biodegradable edible film obtained from arrowroot starch and iota-carrageenan and application in food packaging, International Journal of Biological Macromolecules, 191: 618-626. https://doi.org/10.1016/j.ijbiomac.2021.09.141
- Al-Hakkak J., and Al-Hakkak F., 2007, New non-destructive method using gluten to isolate starch from plant materials other than wheat, Starch Stärke, 59(3-4): 117-124.

https://doi.org/10.1002/STAR.200600564

- Alibekov R., Urazbayeva K., Azimov A., Rozman A., Hashim N., and Maringgal B., 2024, Advances in biodegradable food packaging using wheat-based materials: fabrications and innovations, applications, potentials, and challenges, Foods, 13(18): 2964. <u>https://doi.org/10.3390/foods13182964</u>
- Assatory A., Vitelli M., Rajabzadeh A., and Legge R., 2019, Dry fractionation methods for plant protein, starch and fiber enrichment: a review, Trends in Food Science & Technology, 86: 340-351. https://doi.org/10.1016/J.TIFS.2019.02.006



Bangar S., Purewal S., Trif M., Maqsood S., Kumar M., Manjunatha V., and Rusu A., 2021, Functionality and applicability of starch-based films: an eco-friendly approach, Foods, 10(9): 2181. https://doi.org/10.3390/foods10092181

Cao X., Tong J., Ding M., Wang K., Wang L., Cheng D., Li H., Liu A., Liu J., Zhao Z., Wang Z., and Gao X., 2019, Physicochemical properties of starch in relation to rheological properties of wheat dough (Triticum aestivum L.), Food Chemistry, 297: 125000. https://doi.org/10.1016/j.foodchem.2019.125000

Ceresino E., Kuktaite R., Sato H., Hedenqvist M., and Johansson E., 2019, Impact of gluten separation process and transglutaminase source on gluten based dough properties, Food Hydrocolloids, 87: 661-669.

https://doi.org/10.1016/J.FOODHYD.2018.08.035

Chen B., Zhang B., Li M., Xie Y., and Chen H., 2018, Effects of glutenin and gliadin modified by protein-glutaminase on pasting, rheological properties and microstructure of potato starch, Food Chemistry, 253: 148-155. https://doi.org/10.1016/j.foodchem.2018.01.155

Cheng H., Chen L., Mcclements D., Yang T., Zhang Z., Ren F., Miao M., Tian Y., and Jin Z., 2021, Starch-based biodegradable packaging materials: A review of their preparation, characterization and diverse applications in the food industry, Trends in Food Science and Technology, 114: 70-82. https://doi.org/10.1016/J.TIFS.2021.05.017

- Cui J., Kong X., Hua Y., Zhou H., and Liu Q., 2011, Continuous hydrolysis of modified wheat gluten in an enzymatic membrane reactor, Journal of the Science of Food and Agriculture, 91(15): 2799-2805. https://doi.org/10.1002/jsfa.4524
- Day L., Augustin M., Batey I., and Wrigley C., 2006, Wheat-gluten uses and industry needs, Trends in Food Science & Technology, 17(2): 82-90. https://doi.org/10.1016/J.TIFS.2005.10.003
- Fan X., Jiang J., Wang J., Liu C., Shang J., and Zheng X., 2024, Aqueous ozone effects on wheat gluten: yield, structure, and rheology, Journal of Food Science, 89(10): 6283-6295.

https://doi.org/10.1111/1750-3841.17324

- Gao X., Tong J., Guo L., Yu L., Li S., Yang B., Wang L., Liu Y., Li F., Guo J., Zhai S., Liu C., Rehman A., Farahnaky A., Wang P., Wang Z., and Cao X., 2020, Influence of gluten and starch granules interactions on dough mixing properties in wheat (Triticum aestivum L.), Food Hydrocolloids, 106: 105885. https://doi.org/10.1016/j.foodhyd.2020.105885
- Grassi S., Cardone G., Bigagnoli D., and Marti A., 2018, Monitoring the sprouting process of wheat by non-conventional approaches, Journal of Cereal Science, 83: 180-187.

https://doi.org/10.1016/J.JCS.2018.08.007

Han Z., Shi R., and Sun D., 2020, Effects of novel physical processing techniques on the multi-structures of starch, Trends in Food Science & Technology, 97: 126-135.

https://doi.org/10.1016/j.tifs.2020.01.006

- Inglis D., 2009, Efficient microfluidic particle separation arrays, Applied Physics Letters, 94(1): 013510. https://doi.org/10.1063/1.3068750
- Karwasra B., Kaur M., and Gill B., 2020, Impact of ultrasonication on functional and structural properties of Indian wheat (Triticum aestivum L.) cultivar starches, International Journal of Biological Macromolecules, 164: 1858-1866.

https://doi.org/10.1016/j.ijbiomac.2020.08.013

- Kurdziel M., Łabanowska M., Pietrzyk S., Sobolewska-Zielińska J., and Michalec M., 2019, Changes in the physicochemical properties of barley and oat starches upon the use of environmentally friendly oxidation methods, Carbohydrate Polymers, 210: 339-349. https://doi.org/10.1016/j.carbpol.2019.01.088
- Lagrain B., Goderis B., Brijs K., and Delcour J., 2010, Molecular basis of processing wheat gluten toward biobased materials, Biomacromolecules, 11(3): 533-541

https://doi.org/10.1021/bm100008p

Lauer M., and Smith R., 2020, Recent advances in starch-based films toward food packaging applications: Physicochemical, mechanical, and functional properties, Comprehensive Reviews in Food Science and Food Safety, 19(6): 3031-3083. https://doi.org/10.1111/1541-4337.12627

Li C., Dhital S., Gilbert R., and Gidley M., 2020a, High-amylose wheat starch: Structural basis for water absorption and pasting properties, Carbohydrate Polymers, 245: 116557.

https://doi.org/10.1016/j.carbpol.2020.116557

- Li L., Liu Z., Li X., Chu X., Yang W., Wang B., Xie Y., and Li X., 2023, Superior gluten structure and more small starch granules synergistically confer dough quality for high amylose wheat varieties, Frontiers in Nutrition, 10: 1195505. https://doi.org/10.3389/fnut.2023.1195505
- Li M., Liu C., Zheng X., Hong J., Bian K., and Li L., 2021, Interaction between A-type/B-type starch granules and gluten in dough during mixing, Food Chemistry, 358: 129870.

https://doi.org/10.1016/j.foodchem.2021.129870

Li M., Yue Q., Liu C., Zheng X., Hong J., Li L., and Bian K., 2020b, Effect of gliadin/glutenin ratio on pasting, thermal, and structural properties of wheat starch, Journal of Cereal Science, 93: 102973. https://doi.org/10.1016/j.jcs.2020.102973



- Li W., Gao J., Wu G., Zheng J., Ouyang S., Luo Q., and Zhang G., 2016, Physicochemical and structural properties of A-and B-starch isolated from normal and waxy wheat: effects of lipids removal, Food Hydrocolloids, 60: 364-373. https://doi.org/10.1016/J.FOODHYD.2016.04.011
- Li X., Fei T., Wang Y., Zhao Y., Pan Y., and Li D., 2018, Wheat starch with low retrogradation properties produced by modification of the GtfB enzyme 4,6-α-glucanotransferase from *Streptococcus thermophilus*, Journal of Agricultural and Food Chemistry, 66(15): 3891-3898. https://doi.org/10.1021/acs.jafc.8b00550
- Liu D., Zhao P., Chen J., Yan Y., and Wu Z., 2022, Recent advances and applications in starch for intelligent active food packaging: a review, Foods, 11(18): 2879.

https://doi.org/10.3390/foods11182879

- Liu Z., Zheng Z., Zhu G., Luo S., Zhang D., Liu F., and Shen Y., 2021, Modification of the structural and functional properties of wheat gluten protein using a planetary ball mill, Food Chemistry, 363: 130251. <u>https://doi.org/10.1016/j.foodchem.2021.130251</u>
- Masina N., Choonara Y., Kumar P., Du Toit L., Govender M., Indermun S., and Pillay V., 2017, A review of the chemical modification techniques of starch, Carbohydrate Polymers, 157: 1226-1236.

https://doi.org/10.1016/j.carbpol.2016.09.094

Onyeaka H., Obileke K., Makaka G., and Nwokolo N., 2022, Current research and applications of starch-based biodegradable films for food packaging, Polymers, 14(6): 1126.

https://doi.org/10.3390/polym14061126

- Peighambardoust S., Hamer R., Boom R., and Goot A., 2008, Migration of gluten under shear flow as a novel mechanism for separating wheat flour into gluten and starch, Journal of Cereal Science, 48(2): 327-338. https://doi.org/10.1016/J.JCS.2007.10.005
- Peng P., Wang X., Liao M., Zou X., Ma Q., Zhang X., and Hu X., 2022, Effects of HMW-GSs at *Glu-B1* locus on starch-protein interaction and starch digestibility during thermomechanical processing of wheat dough, Journal of the Science of Food and Agriculture, 103(4): 2134-2145. https://doi.org/10.1002/jsfa.12340
- Raghunathan R., Pandiselvam R., Kothakota A., and Khaneghah A., 2020, The application of emerging non-thermal technologies for the modification of cereal starches, Lwt, 138: 110795.

https://doi.org/10.1016/j.lwt.2020.110795

- Remadnia M., Kachi M., Messal S., Oprean A., Rouau X., and Dascalescu L., 2014, Electrostatic separation of peeling and gluten from finely ground wheat grains, Particulate Science and Technology, 32(6): 608-615. <u>https://doi.org/10.1080/02726351.2014.943379</u>
- Sayaslan A., 2004, Wet-milling of wheat flour: industrial processes and small-scale test methods, LWT Food Science and Technology, 37(5): 499-515. https://doi.org/10.1016/J.LWT.2004.01.009
- Shewry P., Halford N., Belton P., and Tatham A., 2002, The structure and properties of gluten: an elastic protein from wheat grain, Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences, 357(1418): 133-142. https://doi.org/10.1098/RSTB.2001.1024
- Silventoinen P., Kortekangas A., Ercili-Cura D., and Nordlund E., 2020, Impact of ultra-fine milling and air classification on biochemical and techno-functional characteristics of wheat and rye bran, Food Research International, 139: 109971. https://doi.org/10.1016/j.foodres.2020.109971
- Singh H., Macritchie F., Kim Y., Madl R., and Herald T., 2006, Functional modification of vital wheat gluten with corn syrup using hydrothermal treatment, Journal of the Science of Food and Agriculture, 86(2): 251-257. <u>https://doi.org/10.1002/JSFA.2341</u>
- Su C., Li D., Wang L., and Wang Y., 2022, Biodegradation behavior and digestive properties of starch-based film for food packaging-a review, Critical Reviews in Food Science and Nutrition, 63(24): 6923-6945. https://doi.org/10.1080/10408398.2022.2036097
- Su C., Zhao K., Zhang B., Liu Y., Jing L., Wu H., Gou M., Jiang H., Zhang G., and Li W., 2020a, The molecular mechanism for morphological, crystal, physicochemical and digestible property modification of wheat starch after repeated versus continuous heat-moisture treatment, Lwt, 129: 109399. https://doi.org/10.1016/j.lwt.2020.109399
- Su H., Tu J., Zheng M., Deng K., Miao S., Zeng S., Zheng B., and Lu X., 2020b, Effects of oligosaccharides on particle structure, pasting and thermal properties of wheat starch granules under different freezing temperatures, Food Chemistry, 315: 126209. <u>https://doi.org/10.1016/j.foodchem.2020.126209</u>
- Sun F., Liu J., Liu X., Wang Y., Li K., Chang J., Yang G., and He G., 2017, Effect of the phytate and hydrogen peroxide chemical modifications on the physicochemical and functional properties of wheat starch, Food Research International, 100(Pt 1): 180-192. https://doi.org/10.1016/j.foodres.2017.07.001
- Teobaldi A., Barrera G., Sciarini L., and Ribotta P., 2021, Pasting, gelatinization, and rheological properties of wheat starch in the presence of sucrose and gluten, European Food Research and Technology, 247: 1083-1093. https://doi.org/10.1007/s00217-021-03689-y
- Teobaldi A., Barrera G., Severini H., and Ribotta P., 2022, Influence of damaged starch on thermal and rheological properties of wheat starch and wheat starch-gluten systems in water and sucrose, Journal of the Science of Food and Agriculture, 103(3): 1377-1384. https://doi.org/10.1002/jsfa.12231



- Van Der Borght A., Goesaert H., Veraverbeke W., and Delcour J., 2005, Fractionation of wheat and wheat flour into starch and gluten: overview of the main processes and the factors involved, Journal of Cereal Science, 41(3): 221-237. <u>https://doi.org/10.1016/J.JCS.2004.09.008</u>
- Van Rooyen J., Simsek S., Oyeyinka S., and Manley M., 2022, Holistic view of starch chemistry, structure and functionality in dry heat-treated whole wheat kernels and flour, Foods, 11(2): 207.

https://doi.org/10.3390/foods11020207

- Wang J., Wit M., Boom R., and Schutyser M., 2015, Charging and separation behavior of gluten-starch mixtures assessed with a custom-built electrostatic separator, Separation and Purification Technology, 152: 164-171. <u>https://doi.org/10.1016/J.SEPPUR.2015.08.025</u>
- Wang K., Luo S., Cai J., Sun Q., Zhao Y., Zhong X., Jiang S., and Zheng Z., 2016, Effects of partial hydrolysis and subsequent cross-linking on wheat gluten physicochemical properties and structure, Food Chemistry, 197(Pt A): 168-174. <u>https://doi.org/10.1016/j.foodchem.2015.10.123</u>
- Wang M., Hamer R., Vliet T., Gruppen H., Marseille H., and Weegels P., 2003, Effect of water unextractable solids on gluten formation and properties: Mechanistic considerations, Journal of Cereal Science, 37(1): 55-64. https://doi.org/10.1006/JCRS.2002.0478
- Wang M., Van Vliet T., and Hamer R., 2004a, Evidence that pentosans and xylanase affect the re-agglomeration of the gluten network, Journal of Cereal Science, 39(3): 341-349.

https://doi.org/10.1016/J.JCS.2003.12.003

- Wang M., Van Vliet T., and Hamer R., 2004b, How gluten properties are affected by pentosans, Journal of Cereal Science, 39(3): 395-402. https://doi.org/10.1016/J.JCS.2004.02.002
- Wang Z., Ma S., Sun B., Wang F., Huang J., Wang X., and Bao Q., 2021, Effects of thermal properties and behavior of wheat starch and gluten on their interaction: a review, International Journal of Biological Macromolecules, 177: 474-484. <u>https://doi.org/10.1016/j.ijbiomac.2021.02.175</u>
- Witt W., and Goldau H., 2000, Modern methods of separation the components of wheat, Żywność Nauka Technologia Jakość. Suplement, 2(23): 244-265.
- Xie Y., Li M., Chen H., and Zhang B., 2019, Effects of the combination of repeated heat-moisture treatment and compound enzymes hydrolysis on the structural and physicochemical properties of porous wheat starch, Food Chemistry, 274: 351-359.

https://doi.org/10.1016/j.foodchem.2018.09.034

- Yöndem-Makascioğlu F., Dik T., and Kincal N., 2002, Separation of bread wheat flours into starch and gluten fractions: effect of water temperature alone or in combination with water to flour ratio, Journal of the Science of Food and Agriculture, 82(4): 414-420. <u>https://doi.org/10.1002/JSFA.1062</u>
- Yu L., Guo L., Liu Y., Ma Y., Zhu J., Yang Y., Min D., Xie Y., Chen M., Tong J., Rehman A., Wang Z., Cao X., and Gao X., 2021, Novel parameters characterizing size distribution of A and B starch granules in the gluten network: Effects on dough stability in bread wheat, Carbohydrate Polymers, 257: 117623.

https://doi.org/10.1016/j.carbpol.2021.117623

Zalm E., Goot A., and Boom R., 2009, Influence of process conditions on the separation behaviour of starch-gluten systems, Journal of Food Engineering, 95(4): 572-578.

https://doi.org/10.1016/J.JFOODENG.2009.06.038

Zhang D., Mu T., and Sun H., 2018, Effects of starch from five different botanical sources on the rheological and structural properties of starch-gluten model doughs, Food Research International, 103: 156-162.

https://doi.org/10.1016/j.foodres.2017.10.023

Zhang M., Jia R., Ma M., Yang T., Sun Q., and Li M., 2022, Versatile wheat gluten: functional properties and application in the food-related industry, Critical Reviews in Food Science and Nutrition, 63(30): 10444-10460.

https://doi.org/10.1080/10408398.2022.2078785



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