PRODUCTION OF SUSTAINABLE BIOPLASTIC MATERIALS FROM WHEAT GLUTEN PROTEINS

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Summary

Petroleum-based products are creating a number of environmental problems. Petroleum and oil resources are also threatened to become depleted due to the massive utilization. Therefore, it is important to replace the petroleum-based products with products that are instead derived from renewable resources e.g. the replacement of petro-based plastics with bioplastics can be a good option. Wheat gluten proteins might be a promising solution to use for production of bioplastics. Wheat gluten is a cheap by-product from the bio-ethanol industry, thereby largely available and beside that, these proteins have interesting viscoelastic and thermoplastic properties. Gliadin and glutenin i.e. the two major types of gluten protein and their behavior when used to produce bio-based material are discussed in this paper.

Preface

The main emphasis of this introductory paper is to highlight the importance of bioplastics production as a substitute of petro-based plastics as the later type is a risk to environment, land and water ecosystems. Among renewable sources for bioplastics production the importance of cereal proteins especially wheat gluten regarding bioplastics production, acceptance, socio-economical and environmental impacts are also discussed in this paper.

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Introduction

Petrochemical products

Development and progress of the chemical industry in the mid 19th century is directly related to the discoveries of fossil reserves which provide raw material for the synthesis of all petrochemical products (Mecking, 2004). Overall, 90% of the raw material for the chemical industry is currently produced by fossil feed stocks in the form of petroleum and gas. This puts chemical industry in the third position as a user of oil and gas, while energy generation and transportation holds the first and second position, respectively (Mecking, 2004). As to chemical industry, the highest priority area for the utilization of fossil feed stocks is for polymer fabrication (Mecking, 2004). The annual plastic production will be increased up to 300 million tonnes by the year 2010 (Thompson *et al.*, 2009). This is due to the success of synthetic polymer to produce plastics for manufacturing of a range of household and industrial products over the last 50 years.

Petro-based plastics

Petro-based plastics are playing a key role in modern society due to their versatile nature (Momani, 2009). The petro-based plastics possess a range of divergent properties e.g. they can be rigid or elastic, breakable or resilient, transparent or brightly colored, and can have many added advantageous properties (e.g. cheap, recyclable, insulators) (Momani, 2009).

The first polymer, polyvinyl chloride (PVC) was synthesized accidentally in 1838 but was not fabricated into applicable plastic polymer at that time (Wade, 2006). The first successfully applicable polymer named bakelite, was synthesized in 1910 by Leo Baekeland (http://polymer.w99of.com/history-of-polymers/). Due to the possession of a wide range of properties almost every household as well as almost every construction equipment contain commodities that are fully or partly made up of petro-based plastic. The huge production and utilization of petro-based plastics has become a giant threat for the survival of the earth and its inhabitants. This is due to the fact that petro-based plastics are enormously affecting our globe due to its recalcitrance and disposal problems (Barnes *et al.*, 2009).

Impacts of petro-based plastics

Utilization of petroleum and energy resources

Plastic production is directly affecting the petroleum consumption due to the fact that tonnes of plastics are fabricated from petroleum products every year. Many types of plastics e.g. ethylene, propylene, and styrene are directly extracted from crude oil enhancing crude oil consumption (Gervet, 2007).

In year 2009, the total world's petroleum consumption was 98.3 million barrels per day. If this rate of consumption persists, the known oil reserves which are almost 1.24 trillion barrels will last for 41 years (Momani, 2009). The amount of oil used to accomplish plastics products is 4% of the absolute oil utilization (Hopewell *et al.,* 2009). Thus, by keeping in view the bulk petroleum utilization of the world, 4% results in a large amount of oil used for plastic production.

The European contribution to the World's plastic production is 25%, resulting in 60 million tones of plastics per year (www.plasticisland.org).

Significant amount of energy is required for the synthesis of petro-based plastics and plastic-based products. About 6% of all the energy used by American industries is utilized by the plastic industry (Momani, 2009). This results into 1.3% heat loss and it in turns make an addition of 0.5% global warming (Gervet, 2007). Amount of energy used to manufacture chemicals and petrochemicals from 1971-2004 raised by 206% i.e. 33.6 EJ/year globally (IEA, 2004).

Emissions of heat and green house gas

Beside the release of heat from the plastics industry, also CO_2 is released from the plastic industry further contributing to global warming. The release of CO_2 from plastic industry to the environment was increased by 160% from 1971-2004 resulting in a release of 1.0 Gt/year (Gielen *et al.,* 2008). As the manufacturing of plastics and plastic products is increasing day by day it is obvious that CO_2 production will also be increasing. Plastic production and manufacturing produces heat that contributes to the global warming (Nordell, 2003).

Use of crude oil in plastic manufacturing on average has produced approximately 0.38×10^{14} kWh heat from 1939-2000 and it has reached to 0.49×10^{14} kWh in 2004.

Health hazards

Increased use of petro-based plastics has created many health hazards. The major health risks that are associated with petro-based plastics are mainly result of monomers present in the plastics. These monomers are trapped in the polymer matrix during the process of fabrication and then, under certain conditions e.g. heating, may leak out (Momani 2009). For instance, styrene, a monomer can leak out from its polymer polystyrene when subjected to heating and it is assumed to be involved in endocrine disorders and cancer (EPA,http://www.epa.gov/endo/pubs/edspoverview /whatare.html).

Another example is the release of bisphenol-A on heating from the thermoplastic polymer, lexan. Bisphenol-A can be mixed up into food and is carcinogenic as well as can cause hormone disruption (McRandle, 2004).

Plastics disposal problems and inability of biodegradation

All over the world, considerable quantity of waste streams is produced from manufacturing and utilization of petro-based plastics. Municipal waste products in US comprise almost 10% of plastic (Barnes *et al.,* 2009).

The huge amount of plastics that is discarded every year end up in landfills and water. These dumps of plastics are contaminating almost every ecosystem including marine, fresh water, terrestrial and deserts posing numerous environmental problems (Thompson *et al.,* 2009).

Plastics are generally resistant to microbial degradation making them even more hazardous to the ecosystems (Domenek *et al.,* 2004). Further, the presence of plastics everywhere is a threat to the existence of wildlife. It is estimated that more than 260 species of insects, birds, reptiles and mammals have perceived disabilities in movement, feeding habits, sterility and even death due to ingestion of plastics or because they have been intertwined in plastic debris (Gregory, 2009).

The health and environmental hazards of petro-based plastics have resulted in prohibition of use of plastic bags in a large part of the European Union (EU) countries, Australia, China, the city of San Francisco and the ban was also tried in the California (Mooney, 2009).

Bioplastics- an alternative to petroleum-based plastics

Bioplastics (also known as biopolymers) are derivatives of renewable biomass resources e.g. plant proteins and starch. These biopolymers can be fabricated in many different organisms e.g. plants and microbes. These biopolymers do not cause risksdue to the fact these are biocompatible to the hosts (e.g. polyhydroxyalkonates (PHA) synthesized in bacterias). Biopolymers synthesized in microbes are mostly lipid in nature and accumulated in the form of mobile granules and help microbes to survive under stress conditions (Barnard and Sander, 1989; Sudesh *et al.*, 2000).

The scientific community is not only concentrating on exploration of resources that can be a substitute to petroleum derived plastics, but the focus has also been turning into consequences of the biodegradability of the plastics. Many research groups are continuing their efforts in order to investigate options of making bio-based plastics photodegradable. However, such plastics will not be suitable for landfill disposals as these are continuously exposed to sunlight (Zan *et al.,* 2006). One of the important objectives of the research related to synthesis of biodegradable plastics is the creation of biodegradation ability in composters or metropolitan landfills.

Sustainable production

Sustainable production enhances the quality and quantity of environmentally friendly goods and services for human kind. This is done through minimal utilization of natural resources, effective utilization of raw materials with decreased waste production. Sustainable production is possible only with the collaboration of government, industries and consumers (Falkman, 1995).

The requirements for petro-based products are expected to be doubled in upcoming years. So it is expected that the production of plastics will also increase and it will increase the environmental pollution as well. In order to tackle with the problem of environmental pollution, plastic production by utilization of renewable resources should be increased (Thompson *et al*., 2009). It will reduce the reliability of people on fossil fuels and other natural resources which are exhaustible (Willke and Vorlop, 2004). For the sake of conservation of energy resources and natural raw materials, together with decreasing of the global warming, it is now the time to replace non-renewable resources with renewable

resources. Some manufacturers are also showing interests in utilization of renewable resources for plastic production. One example is from one of the largest plastic manufacturers "DuPont", aimed to fabricate 25% of their materials by utilizing renewable resources by the year 2010 (Dupont, 2010). Plant-based bioplastics (e.g. wheat gluten-, ligno-cellulose-, and cellulose-derived bioplastics) are valuable alternatives to petroleum-based plastics due to their specific polymer formation and biodegradable abilities (Wretfors *et al.*, 2009).

Proteins

Proteins are one of the most vital nutrients, being of essential importance for human survival and life. Proteins are natural heteropolymers made up from 22 different amino acids arranged in different combinations giving rise to thousands of different proteins (Guilbert and Cuq, 2005)

Proteins in wheat and their content and composition i.e. monomeric and polymeric proteins, amount and size distribution of these proteins, etc. are the main detriments of bread-making quality (Johansson *et al*., 1993, 2001; Gupta *et al*., 1993).

Proteins (plants or animal derived) are the ideal raw material for bioplastics production due to the presence of many polar and non-polar amino acids providing a broad spectrum of functional and structural properties (Guilbert and Cuq, 2005). In addition to this, proteins are easily processable and can adhere to various substrates, so proteins can be utilized to produce blends or composites of desirable characters (Vaz *et al*., 2003).

Importance of proteins in bioplastics and agriculture packaging materials

The utilization of proteins for the formation of plastic materials was initially started in 1930's and remained active till 1940's. The principle raw proteins for plastic production during this era were mainly milk casein, soy and corn zein (Verbeek and Berg, 2009). However, the use of proteins for plastic production was slowed down because of the discovery of the opportunities to use petroleum for plastic production. The diverse utilization of petroleum for a number of products has caused scaricity of these resources at present. This situation has resulted again in an increased interest for development of proteinaceous bioplastic materials and agricultural packaging films (Cuq *et al*., 1998).

Proteins are a competent choice for bioplastics production as the proteins are able to offer opportunities for a wide range of chemical utilities. Protein serves as an ideal biomaterial with a possibility of formation of many different kinds of polymer networks (Guilbert and Cuq, 2005). Ability to form polymers with a wide range of functional properties and structural confirmations makes proteins suitable to form packaging films (Cuq *et al*., 1998). Proteins derived from plants are low in cost, renewable, abundant and biodegradable. Therefore, they are suitable as raw material to be used in the bioplastic industry (Irissin-Mangata *et al*., 2001). At present, a vast variety of agricultural and medicinal films are being made from a large

number of plants (gluten, zein etc.) and animal's derived (collagen, gelatin, myofibril etc.) proteins. For example films made from corn zein proteins are being utilized to preserve dry and fresh foods, vitamins and for medicinal purposes (controlled release of active compounds). One interesting example of such films based on zein protein is "Optaglaze" commercially produced by Opta Food Ingredients, Inc (Cuq *et al*., 1998). Mixtures of soy proteins and phenol formaldehyde resins are also used to make parts of automobiles (Jane *et al*., 1994)

Toxic impacts of chemical fertilizers are strongly polluting the soil and underground water. One solution for this problem is to entrap or spread the fertilizers in coatings with biodegradable materials like proteins (Devassine *et al*., 2002). There will be many additional benefits of using protein coated fertilizers; protein can swell and in turns can save a large number of water molecules. The proteins may also serve as an additional nitrogen source after degradation at a very low cost (Montain *et al*., 2004, Yuan *et al*., 2010).

Wheat

Wheat (*Triticum aestivum* L.) is one of the largest staple food source in the world and it is a universally grown crop (Carter, 2002).

A large part of the grown wheat (90%) is utilized for human consumption and only 10% is utilized as seeds for next crop and for industrial purposes e.g. for the production of starch, gluten, malt, dextrose (Hussain and Qamar, 2007). Wheat production reached 600 million tonnes in the year 2005. Wheat consumption has increased over time and it is predicted that its production will increase up to 840 million metric tonnes by the year 2020 (Rajaram, 2005). Wheat provides a number of nutritional components such as protein, carbohydrates, vitamin E and minerals etc. to the daily diet of the world's population. Besides the nutritional importance of wheat, it is increasingly used by the industries. Currently, wheat starch and proteins are both being utilized to make biofuel and bioplastics respectively (Cuq *et al*., 1998).

Wheat proteins and their types

According to solubility properties, wheat proteins are grouped into two major classes; non-gluten proteins and gluten proteins (Shewry *et al.*, 1986). Various protein types and properties are also listed in Table 1.

Non-gluten proteins are albumin and globulin comprising 15% of total wheat proteins. Albumins are soluble in water while the globulins are soluble in dilute salt solutions (Obsorne, 1924).

Wheat gluten proteins can be further subdivided into two groups based on their solubility and extractability in alcohols i.e. alcohol soluble (gliadins) and alcohol insoluble (glutenins) (Shewry *et al*., 2002; Wieser 2007). Gluten proteins show low solubility in water or dilute salt solutions due to presence of non-polar amino acids as compared to amino acids with polar ionizable side chains. Low solubility behavior of gluten proteins is also due to the presence of high amount of glutamine and proline residues as these residues contain non-polar side chains (Hernandez-Munoz *et al*., 2003; Lagrain *et al*., 2010).

Table 1. Wheat protein classification based on solubility (Osborne, 1924; Aykroyd and Doughty, 1970; Krull and Inglett, 1971; Wieser, 2007)

Groups	**Proteins**	**Function**	**Molecular weight**	**Solubility**	**Distribution**	**Average protein content**
Non-Gluten Proteins	Albumins	Metabolic and cytoplasmic proteins	20,000	Water	Embryo	15%
	Globulins	Storage and cytoplasmic proteins	20,000-200,000	Dilute salt solutions	Embryo and aleuronic layer	
Gluten Proteins	Gliadins	Storage proteins	30,000-60,000	Ethanol (70-80%)	Endosperm	45%
	Glutenins	Storage proteins	8000-several millions	Dilute acids/alkali solutions		40%

Gluten proteins and their properties

Gluten is a cohesive, visco-elastic proteinaceous material with strong thermoplastic properties which is isolated from wheat (Day *et al*., 2006). Gluten can be separated out as a byproduct in the isolation of starch from wheat flour by simply washing the dough with water. Gluten proteins were first isolated from wheat flour three centuries ago (Bailey, 1941).

Gluten proteins i.e. gliadins and glutenins were also named as prolamins i.e. due to presence of high number of proline and glutamine amino acids (Gianibelli *et al.,* 2001). Gluten is a huge and complex network of proteins which can be separated into almost 50 different types of proteins with two dimensional isoelectric focusing or sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) (Wrigley and Bietz, 1988; Shewry *et al*., 1986).

Gliadins

Gliadins are one of the two protein types that are present in wheat gluten. Gliadins are monomeric single chained proteins and their molecular weights range from 30,000-60,000 Daltons (Veraverbeke and Delcour, 2002). Gliadins are further divided into three structurally distinct groups i.e. α/β-, *γ*- and *ω*-types (Wieser, 2007).

Structure of α/β -, *γ*- and *ω*-types gliadins

The structural differences among the three groups of gliadins are small due to a difference in one amino acid residue. This amino acid residue difference has arisen by insertion, substitution and deletion of the amino acid (Wieser, 2007).

Two classes of gliadins i.e. α/β- and *γ*- gliadins have overlapping molecular weights in the range of 28000 to 35000 Daltons (Wieser, 2007). Both of these classes have distinct C- and N-terminal domains. The repetitive sequences rich in tyrosine, phenylalanine, glutamine and proline are frequently occurring in the N-terminal domain of the α/β - and *γ*-gliadins. The C-terminal domains of α/β - and *γ*- glidin are homologous, without repetitive structures and with very low amount of glutamine and proline residues (Grosch and Wieser, 1999). However the C-terminal domains of α/β- and *γ*- gliadins are rich in cystein residues i.e. α/β- with six and *γ*-gliadins with eight cystein residues (Grosch and Wieser, 1999). The structural confirmation of the N-terminal domain of the α/β- and *γ*-gliadins is similar to *ω*-gliadins, both of these groups

have β turns in their secondary structure (Tatham and Shewry, 1985). C-terminal domain of α/β- and *γ*- gliadins occurs in the form of α-helix and β-turns in their secondary structures.

ω-gliadins have higher molecular weights than α/β- and *γ*-gliadins in range of 40,000-55000 Daltons (Wieser, 2007). The *ω*-gliadins consist of the sequence units PQQPFPQQ and these units are rich in glutamine and proline residues (Grosch and Wieser, 1999).

Glutenins

Glutenins are the other protein type present in wheat gluten and consist of a mixture of polypeptides. The glutenin polymer is considered the largest protein in nature (Wrigley, 1996). Molecular weight of glutenin ranges from 8000 to several millions Daltons (Veraverbeke and Delcour, 2002).

Structurally, glutenins appears as polypeptides, thus peptide chains are interconnected via inter and sometimes intrachain disulphide linkages (Shewry and Tatham, 1997; Lagrain *et al*., 2010).

Glutenins are further classified into two subunits; low molecular weight glutenin subunits (LMW-GS) and high molecular weight glutenin subunits (HMW-GS) (Ye *et al*., 2006; Wieser, 2007).

Low molecular weight glutenins subunits

LMW-GS comprises 30% of all the wheat gluten proteins (Laszity *et al*., 2000). The LMW-GS consist of two domains, the N- terminal domain which is enriched of glutamine and proline residues and with sequence repetitive motifs i.e. QQQPPFS, and the C-terminal domain which is similar in structure and amino acid composition to α-, β- and γ- gliadins. LMW-GS has eight cysteine residues (Grosch and Wieser, 1999). Six of these are forming intrachain bonds and their positions are homologous to α-, β- and γ- gliadins. Two additional cysteine residues are unique to LMW-GS and are not involved in any type of bonding (Wieser, 2007).

High molecular weight glutenin subunits and their importance

The average amount of HMW-GS in the wheat grain is estimated to be 10-12% (Shewry *et al*., 2002; Weiser, 2007). However, the amount can vary due to genotypic variations e.g. gene silencing, polymorphism etc. (Shewry *et al*., 2002).

The viscoelastic properties of wheat gluten have to a large extent been attributed to the HMW-GS. Studies of wheat lines only differing in HMW-GS subunit composition have proven the role of HMW-GS in determining the viscoelastic properties of the wheat dough (Branlard and Dardevet 1985; Payne *et al*. 1988, Shewry *et al*., 2002). Variation in HMW-GS composition accounts for 45-70% of the variation in baking quality (Shewry *et al*., 2002).

The importance of HMW-GS to impart wheat gluten viscoelastic properties also indicate that transformation of wheat cultivars with increased number of HMW subunit genes can improve the strength and structure of wheat gluten (Shewry *et al*., 2002). Moreover, the use of genetic engineering and transformation of wheat cultivars with multiple copies of HMW-GS can be an important tool to improve the gluten utilization in the bioplastic industry.

Structure of HMW-GS

The HMW-GS have an extensive rod-like structure of 50-60 nm length (Li *et al*., 2006) with three discrete domains. The two N- and C- terminal domains have the form of an α- helix and posses interchain crosslinks of disulphide bonds due to the presence of cysteine residues. The third domain comprises a large central repetitive unit with conformation of regular β-turn repeats (Shewry *et al*., 1992).

The amino acid composition of the N-terminal domain consists of 81-89 and 104 amino acid residues for the x- and y-type subunit, respectively. This difference in number of amino acids residues results in a total of three cystein residues in the *x*-type and five in the *y*-type subunits (Tatham *et al*. 1984; Van Dijk *et al*. 1998).

The C-terminal domain of all HMW glutenin subunits has equal number of amino acid residues i.e. 42 with one cysteine (Shewry *et al*., 2002). The structure of the HMW-GS central repetitive domain has been studied extensively by using X-ray crystallography but these experiments were not able to generate clear diffraction pattern (Shewry *et al*., 2002). Hydrodynamic and detailed spectroscopic studies have depicted the formation of β-reverse turns in the central domain (Gilbert *et al*., 2000). Further, it has been hypothesized that the

central repetitive unit of HMW-GS has adopted a regular β-spiral structure (figure 1), which has also been confirmed by molecular modeling, viscometric analysis, and scanning tunneling microscopy (Shewry *et al* 2002).

Keeping in view the elasticity of gluten, the loop and train model for the HMW-GS has been presented by Belton in 1999. This model predicts that there is a struggle for making more hydrogen bonds between glutamine and water residue upon hydration. As the level of hydration increases, this competition will lead to the formation of mobile loop structures via the formation of hydrogen bonds between glutamine and water. The confirmation of mobile loops segments is in the form of β-turn structures. This result in breakage of many but not all interchain hydrogen bonds of adjacent HMW-GS, which results in the formation of β-sheets called as “trains” (figure 2).

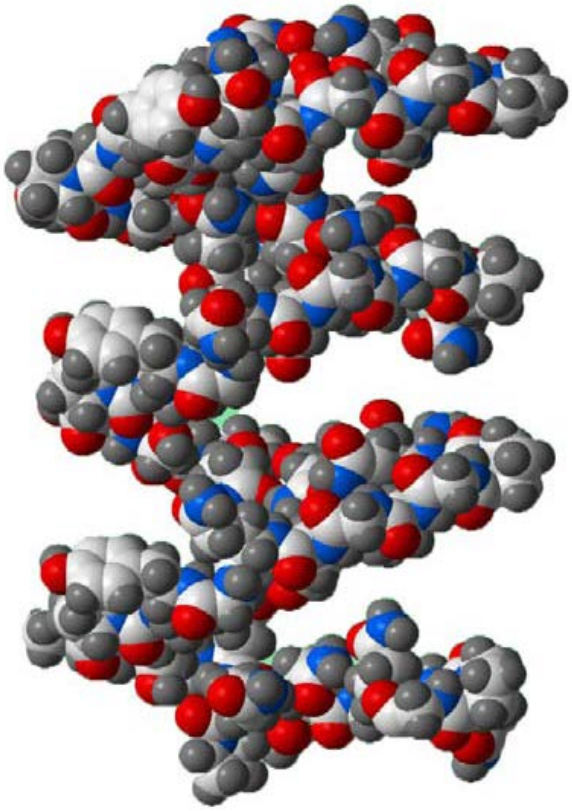

Figure 1: Molecular model developed for β- spiral structure based on the amino-acid sequence of a repetitive motifs of HMW-GS subunit *Atoms are shown in white (carbon), blue (nitrogen), red (oxygen) and grey (hydrogen) (With permission* Shewry *et al.*, 2009)

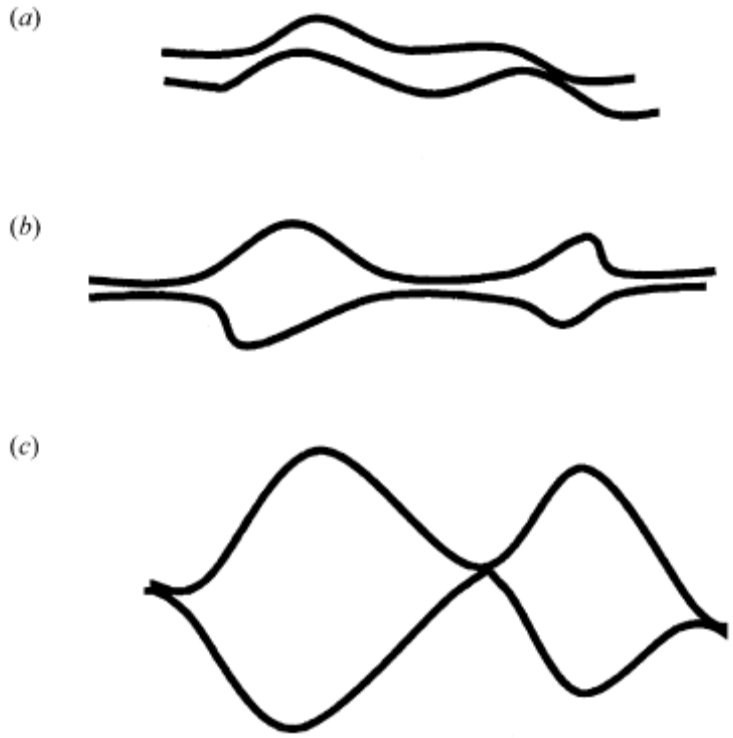

Figure 2: Model for the effect of hydration on the loop to train ratio of HMW-GS subunits. (*a*) Low hydration, disordered, close interactions; (*b*) intermediate hydration, low loop to train ratio; (*c*) high hydration, high loop to train ratio (With permission from Shewry *et al*., 2002 and Journal of *Phil. Trans R. Soc. Lond. B*).

Genes for gluten proteins

Hexaploid bread wheat species contain three set of genomes i.e. A, B and D. In wheat seed, nine loci have been found to be involved in the synthesis of gluten proteins.

Three loci *Glu-A1, Glu-B1* and *Glu-D1* on long arm of chromosomes 1A, 1B and 1D encodes the synthesis of HMW-GS (Halford *et al*. 1992; Seilmeier *et al*. 1991). Each locus with two genes thus encodes two different subunits designated *x*- and *y*-type subunits (Payne *et al*., 1987). Due to gene silencing mechanisms, bread-wheat cultivars contain 3-6 HMW-GS. (Payne *et al*., 1987; Halford *et al*. 1992; Seilmeier *et al*. 1991).

Among the two types of HMW-GS subunits the *x*-type is considered to be a more important contributor to enhance the viscoelastic and hydration properties of glutenin proteins than the *y*-type subunit (Wieser and Kieffer, 2001).

The same chromosomes i.e. 1A, 1B and 1D also contain three loci *Gli-A1, Gli-B1* and *Gli-D1* at their short arms. The three major gene families located on these loci are encoding the synthesis of ω-gliadins, *γ*-gliadins and LMW-GS subunits (Payne *et al*., 1982).
The short arms of chromosomes 6A, 6B and 6D carry three loci, *Gli-A2, Gli-B2* and *Gli-D2*. Each of these loci encodes the synthesis of α- and β-gliadins (Bietz *et al.,* 1976).

Polymerization process of wheat gluten proteins

Wheat gluten is actually a complex mixture of proteins, containing 50-100 different types of proteins. Several research groups (Sartor and Johari, 1996; Weegles *et al*., 1996; Feeney *et al*., 2003; Wellner *et al*., 2005; Li *et al*., 2006) have focused on the understanding of the detailed polymerization of gliadin and glutenin proteins. However still, it is very difficult to provide a complete and clear picture about the polymerization process of wheat gluten due to the complex network of the proteins. The improved understanding of the polymerization process is important in order to determine the best suitable combination of gluten protein for the development of bioplastics. The structural confirmation of gliadin and glutenin proteins has been studied as well. The primary, secondary and tertiary structures have been clearly depicted (Shewry *et al.,* 2002).
Polymeric proteins, i.e. LMW-GS (with more than one cystein residue) and HMW-GS are regarded as chain extender proteins and this may be a reason to their strengthening effect on wheat gluten and may contribute to stability of gluten derived products (Lee *et al.,* 1999). LMW-GS with only one cystein are also identified and these LMW-GS are known as polymeric chain terminators (Tamas *et al.,* 1998). In a recent study conducted by Hernandez-Munoz and co workers (2004), disulphide linkages were introduced in monomeric gliadins. Gliadins were polymerized with the addition of cystein and effects of this induced polymerization were evaluated by the functional properties of the derived films. These films possesed improved water vapor resistance properties due to development of intra- and interchain disulphide linkages of the gliadin proteins (Hernandez-Munoz*et al.,* 2004). Wretfors *et al.,* (2010) was able to show that addition of diamine in blends of gluten and hemp fibre increased the protein polymerization.
A complete understanding of gluten protein polymerization is required in order to develop superior quality of gluten-based plastic products.

Bioplastics

Bioplastics can be defined as derivatives of renewable biomass resources, which are largely biodegradable and may provide similar functional advantages (e.g. packaging materials) as those of traditional plastics (Song *et al.*, 2009).

At present, there is an increasing interest for the development of biodegradable plastics and agricultural packaging films derived from renewable biomass (Ye *et al*., 2006). Biopolymers (lipids, proteins, and polysaccharides) constitute the principle raw material for bioplastic production. Bioplastics derived from these biopolymers are biodegradable, renewable and environment friendly materials as compared to petro-based plastics (Murphy and Bartle, 2004).

Importance of wheat gluten proteins for bioplastic industry

Wheat gluten has a wide range of uses in food and non-food industries. Among non-food uses, gluten is used in production of cosmetics, detergents, rubber and polymer fabrication (Magnuson, 1985; Bietz and Lookhart, 1996). Gluten forms a soft and elastic solid material when it is plasticized with the addition of glycerol. Thus, the complex of wheat gluten and glycerol shows pseudo-plastic properties (Guilbert and Cuq, 2005). Wheat gluten based bioplastics are harmless and environment friendly biomaterials regardless of the technical process utilized to fabricate them (Domenek *et al*., 2004). Moreover, wheat gluten is annually renewable and a low cost material for utilization in the bioplastics industry (Lagrain *et al.*, 2010).

Wheat gluten possesses good oxygen barrier properties and thermostability when compared to other renewable materials like starch, cellulose, oils etc. (Krull and Inglett, 1971; Bietz and Lookhart, 1996; Hernandez-Munoz *et al*., 2003; Woerdeman *et al*., 2004). These properties make wheat gluten proteins a suitable raw material for the production of bioplastics. However, the water absorbing property of gluten protein material is a problem resulting in a low water vapor barrier (Cho et al., 2010). The low water vapour barrier might be overcomes by processing at high temperature which increases the high crosslinking (Pommet *et al*., 2005). Another way of reducing water absorption might be to use hydrophobic plasticizers e.g. palmitic acid chloride and succinic acid (Brauer *et al*., 2007). Lamination of poly lacticacid

(PLA) to wheat gluten has improved water vapor barrier of the plastics and can be a suitable solution for packaging of dry foods under moist or dry conditions (Cho et al., 2010).

Amino acid composition, hydration responses, various structural analysis and proposed structural/conformational models has shown considerable variations in the structure of plasticized wheat gluten. Due to amino acid compositions as well as nature and energy variations, a large number of chemical reactions are possible among gluten proteins. Thus, composition of the protein in the material enhances the possibilities of great functional variations (Pommet *et al*., 2003). These variations (e.g. elasticity, cohesiveness, biodegradability) can be utilized in the bioplastic industry. Both gliadins and glutenins impart their effects on dough made from wheat flour (Wieser, 2007). The glutenins have been found the most important determinant of gluten elastic and cohesive properties, while the gliadins are more viscous and act as a plasticizer upon hydration (Wieser, 2007).

Gluten-based bioplastics

Abundant quantity of wheat gluten is obtained as a byproduct of food processing industry at a very low cost i.e. less than $1.00/kg (Ye *et al.,* 2006). A significant quantity of wheat gluten is obtained as a byproduct when wheat is utilized for the production of bioethanol. The market for the production of biofuel from wheat gluten is expected to increase over the time and so the availability of wheat gluten will also increase (Cho *et al*., 2010). At present, wheat gluten is a readily available raw material with an annual production of almost 500,000 tonnes (Reddy and Yang, 2007).

The degradation rate of wheat gluten is among the fast degrading polymers and it has been confirmed experimentally that when gluten derived products were buried in farmland soil, these were completely degraded in 50 days (Domenek *et al.,* 2004). Due to favorable properties of wheat gluten to be fabricated into biopolymer, it is becoming a material of choice for the production of plastics and packaging films (Jerez *et al.,* 2005). Formation of complex networks with disulphide linkages upon thermosetting is the innate property of wheat gluten and this property facilitates its processing into films and plastics when it is plasticized (Cuq *et al*., 1998).

Amino acids like cysteins and dialdehydes offer crosslinking reactions between gliadins and glutenins. Water resistance and tensile strength of casting films can be improved by these

crosslinking reactions. Crosslinking is useful in order to lower the water permeability of gluten-based bioplastics (Hernandez-Munoz *et al.*, 2003, 2004).

In thermoplastic processing, bioplastic production require a complete premixing of all the components including biopolymer (e.g. protein), water and plasticizer to obtain a dough like material (Jerez *et al.*, 2005). Bioplastics have been prepared from gluten (Domenek *et al.*, 2003; Gomez-Martinez *et al.*, 2009; Song *et al.*, 2009), gliadin-rich fraction and glutenin-rich fraction (Hernandez-Munoz *et al.*, 2003; Song and Zheng, 2008; Song *et al.*, 2009) by using different plasticizers like glycerol (Domenek *et al.*, 2003; Sun *et al.*, 2007) and water (Domenek *et al.*, 2003; Gomez-Martinez *et al.*, 2009). Wheat gluten films and plastics without the addition of plasticizers are delicate and brittle. The use of plasticizer contributes to the elasticity and extensibility of gluten protein plastic materials. Plasticizers impart these effects by dropping intermolecular forces and by increasing the mobility of polymeric chains (Gontard *et al.*, 1992; Gennadios, 2002).

Hernandez-Munoz *et al.*, (2004) has developed gliadin rich films with improved water barrier properties. This has been done by cleaving the intramolecular disulphide bonds and rearrangement of structures by the formation of intermolecular disulphide bonds.

Hernandez-Munoz *et al.*, (2003), has found that biodegradable wheat films obtained from glutenin rich fractions are stronger, possess higher tensile strength and lower water vapor permeability properties as compared to those derived from gliadin rich fractions.

Gluten-based bioplastics with improved qualities

Diverse characteristics can be imparted to the bioplastics according to processing conditions and chosen formulations. Gomez-Martinez *et al.*, (2009) has shown that an increase in compression-molding temperature results in bioplastics having improved elastic properties with higher viscoelastic modulus. If the bioplastics are being designed to use in agriculture industry, addition of citric acid is advantageous to increase water absorption capacity (Gomez-Martinez *et al.*, 2009).

Preferable methods for protein processing are extrusion and compression-molding, providing fast routes for production of bioplastics (Ullsten *et al.*, 2006). However, the processing window of gluten is limited for these methods and has to be improved in order to make the technique more useful. Ullsten *et al.*, 2006 has increased the processing window of wheat

gluten by the use of salicylic acid (SA). SA possesses radical scavenging activities. It slowered the rate of crosslinking reactions and enhanced the processing window (Ullsten *et al.,* 2006). SA possesses germicidal activity in addition to radical scavenging activity and expected to enhance the shelf life of gluten-based products (Brabias and Swiatek., 1998).

Increase in molding temperature from 25 to 125 C^{o} has been shown to have promising effects on the three dimensional structural networks of gluten by increasing protein crosslinking density via disulphide linkages (Sun *et al.,* 2007). Addition of tri-thiol can also strengthen the delicate wheat gluten (Woerdeman *et al*., 2004).

The choice of plasticizer is important and some plasticizers cause aging of the protein-based films due to loss over time (Olabarrieta *et al.,* 2006). Blends of natural polymers (proteins, starch) and synthetic polymers (polycaprolactone, polylactic acid) provide an opportunity to produce biodegradable bioplastics without the addition of plasticizers (Ramkumar *et al.,* 1996). Synthetic and natural polymers, when to be blended, must be compatible in order to manufacture products with superior properties. John *et al.,* (1998) has produced biodegradable blends by mixing wheat gluten and modified polycaprolactone (PCL). PCL is a natural aliphatic polyester and it is biodegradable. PCL was modified by incorporating a reactive functional group ‘anhydride’. Anhydride increases the compatibility and reactive blending of PCL with proteins. The gluten composition in these blends was 65% and 75%, fixed by weight. The blends of gluten and modified PCL showed improved physical properties (morphology, viscosity, biodegradability) over the simple mixtures of gluten and PCL. The materials made from these blends showed stable characteristics when studied under extreme conditions of temperature in oven and freezer (John *et al.,* 1998).

Cho *et al.,* (2010) has developed compression-molded glycerol plasticized wheat gluten films. These films were laminated with polylactic acid (PLA). In addition to mechanical strengthening, PLA coating imparts two additional benefits to glycerol plasticized wheat gluten i.e. good oxygen barrier properties and prevention of loss of glycerol plasticizer over time (Cho *et al.,* 2010).

Processing of gluten

Polymer processing can be defined as the mixing and shaping of raw materials to convert them into required products with suitable properties according to end use purpose (Verbeek and Berg, 2009).

Processing routes to derive bioplastics from biomaterials (proteins, starch) with similar functional properties as petro-based plastics is important to understand. Processing, which is mostly accomplished by the application of heat and pressure is mainly dependent on the nature of the biomaterial and whether it is a thermoplastic or a thermoset material. Wheat gluten proteins possess thermoplastic properties. In a thermoplastic process, the biomaterial is first degraded or melted and thereafter shaped according to the requirement and finally cooled to set it into its new form (figure 3). Heat source for melting can be provided by radiations, conduction or mechanical work (Verbeek and Berg, 2009). Two key methods i.e. solvent-based and dry processing techniques are mainly employed for protein processing.

Commonly utilized thermoplastic techniques are extrusion, thermo-forming and injection molding. Extrusion is a dry processing method and it is the most widely used technique for the gluten-based bioplastic production. Jerez *et al.,* (2005) has compared the properties of wheat gluten films and bioplastics developed through a thermo mechanical and casting process. Bioplastics and films processed by thermo mechanical techniques possess higher thermal susceptibility as compared to casting processed bioplastics. However, bioplastics obtained from either of the process, possess a similar mechanical spectra i.e. microstructure and rheological behaviour (Jerez *et al.,* 2005).

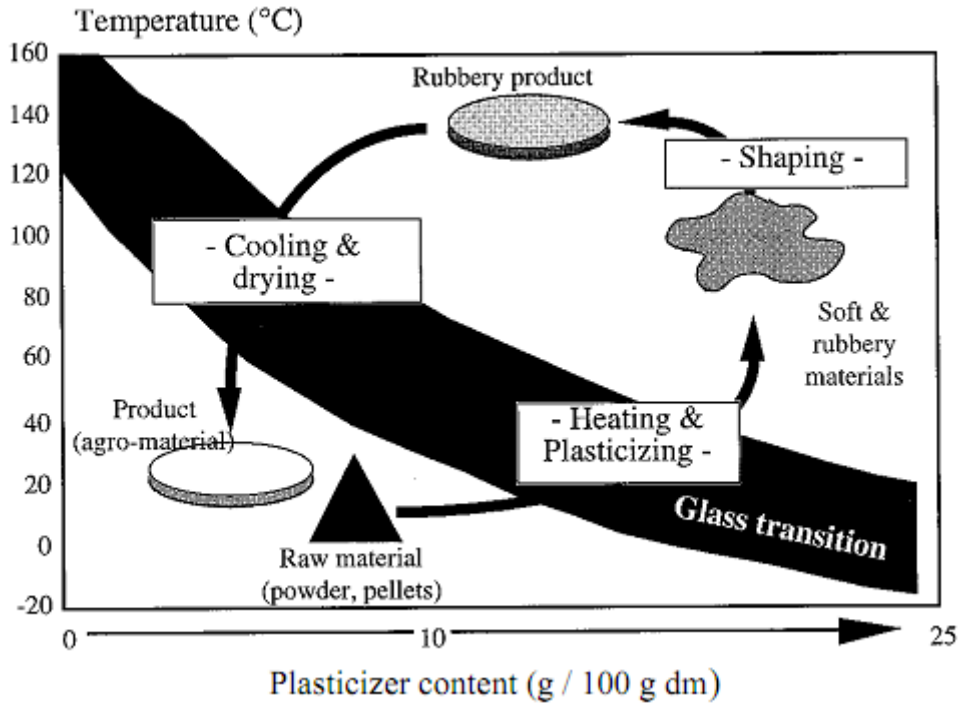

Figure 3. Thermoplastic processing of wheat gluten to form agro-packaging materials (Reproduced with permission from Cuq *et al.*, 1998 and Journal of *Cereal Chem.*)

Genetic engineering of wheat gluten

Genetic engineering techniques have been used to modify the properties of wheat gluten and its subfractions gliadins and glutenins. These techniques are used to increase or modify the expression of gluten proteins according to the need of its utilization and end products (Shewry *et al.*, 2002).

Rooke *et al.*, (1999) has developed a transgenic wheat line B73-6-1 with multiple genes of 1Dx5 encoding HMW-GS. The result is a four fold increase in expression of HMW-GS and gluten protein resulting in a dough with more strength and elasticity (Rooke *et al.*, 1999). Increased expression of 1Dx5 protein leads to formation of a highly crosslinked protein networks with increased gluten strength and elasticity (Popineau *et al.*, 2001). Mutant forms of wheat gluten have been created to study the detailed structural and functional characteristics of total gluten. At present, there are several studies conducted to evaluate the synthesis of novel peptides based on gliadins and glutenin repeat motifs by the application of a heterologous expression system. As the HMW-GS is mainly related to elastic properties of wheat gluten, it is a good target for modifications and genetic engineering of wheat proteins

(Vasil and Anderson, 1997). The clear genome, amino acid sequence and structural analysis of HMW-GS have facilitated the construction of mutants and their study in expression systems.

Use of heterologous expression systems/bacterial system for gluten protein production

Heterologous expression systems are used to study the structural and functional properties of proteins. These are the expression systems of a protein into an organism from which it is not originated (Tamas and Shewry, 2006). Traditionally, three types of expression systems as a host are being used for recombinant protein synthesis viz; *E.coli*, yeasts and cultured cells (Tamas and Shewry, 2006). The expression system must be able to grow readily in culture and should be able to reproduce proteins in bulk quantities for commercial applications.

Galili, (1989) has developed a high level expression system for the production of wheat glutenin in *E.coli* by the application of pET vectors. Maruyama *et al.,* (1998) has developed expression systems for the production of α-, γ- and *ω*-gliadins and LMW and HMW-GS subunits. This system is very useful for the expression of repetitive subunits of gluten protein. Thompson *et al.,* (1994) used baculovirus expression system for the production of gluten. However, *E.coli* based expression systems are preferable due to ease of its use, cost effectiveness and availability of large number of host strains. They give high yield and fusion technology can be applied easily. Presence of inclusion bodies in *E.coli* can also facilitate the protein purification. *E.coli* expression system is not involved in unnecessary protein modification like glycosylation and post translational modifications. Clarke *et al.,* (2003) has used *E.coli* with pET11d vector for the production of LMW-GS**.** HMW-GS expression studies have been carried out by several workers (Anderson *et al.,* 1996; Feeney *et al.,* 2001) for detailed structural studies. pET vectors have also been employed by Elmorjani *et al.,* (1997) for the expression of gliadin motif (Pro-Gln-Gln,), about 32 copies of the motif was expressed in this expression system (Elmorjani *et al.,* 1997). Detailed overview of the past studies related to expression of recombinant gluten protein subunits have been given in table 2. However, it is notable that until now, no research group all over the world has developed an expression system for the bulk production of wheat gluten.

Table 2: Overview of important studies of expression system used and further characterization of recombinant proteins (Reproduced from Tamas and Shewry, 2006).

Protein	**Expression system**	**Expression vector**	**Yield**	**Further use**	**Reference**
α-Gliadin	*Saccharomices cerevisiae*	2μ-plasmid derived pAY33	0.5-4.0mg/l	Western blotting, N-terminal sequencing	Blechl *et al.,* (1992)
γ-Gliadin	*Saccharomices cerevisiae*	pKV4	3-5 mg/l	Western blotting, N-terminal sequencing	Pratt *et al.,* (1991)
LMW-GS	*E.coli*	pET3a	40-100 mg/l	SDS-PAGE, western blotting	Patacchini *et al.,* (2003)
HMW-GS 1Dx2	*E.coli*	pET3a	7% of total protein	SDS-PAGE	Galili (1989)
HMW-GS 1Dx2 (modified in lenght)	*E.coli*	pET3a	20–40 mg/l	SDS-PAGE, RP-HPLC	D'Ovidio *et al.,* (1997)
HMW-GS 1Dx2, 1Dx5, 1Dy10, 1Dy12	*E.coli*	pET3a	10-20% total protein in fermenter	mixograph	Bekes *et al.,*(1994) and Dowd and Bekes (2002)
Subunit 1Dx5, M*r* 58,000 peptide	*E.coli*	pET17b	20mg/l	mixograph, SDS-PAGE, surface properties, MALDI-TOF MS	Buonocore *et al.,* (1998) and Gilbert *et al.,* (2000)
Gliadin-related peptides	*E.coli*	pEQ_2, $pET21d_2$, pET21d2,	15-20% total protein	SDS-PAGE, UV and FT-IR spectroscopy	Elmorjani *et al.,* (1997) and Sourice *et al.,* (2003)
HMW-GS related peptides	*E.coli*	pET32d	6-10mg/l	SDS-PAGE, western blotting, CD and FT-IR spectroscopy	Feeney *et al.,* (2001) and Wellner *et al.,* (2006)

Biocomposites

Biocomposites are blends of two biomaterials, one is a biodegradable polymer and the other is a biodegradable filler. Biocomposites are made in order to achieve improved performance, which is not possible by either of the component alone. There is considerable interest of several research groups to make biodegradable composites from biopolymers like starch (Gaspar *et al.,* 2005) and wheat gluten (Yang *et al.,* 2011; Ye *et al.,* 2006).

Ye *et al.,* (2006) has manufactured biodegradable composites of wheat gluten and basalt fibres. El-Wakil, (2009), has studied the formation of biocomposites formed by the combination of wheat gluten, alkalized lignin and sodium silicates. The resulting materials exhibits increased tensile strength, uniformity, low thermal expansion and high glass transition temperature.

Kim, (2008) has developed a new technology for the formation of biocomposites at the room temperature without the need of extrusion or processing at high temperature. It requires very minute amount of biomaterials like wheat gluten. This is possible by utilizing the strong adhesive properties of corn protein 'zein'. This technology saves time, energy and cost of production due to minimal utilization of resources (Kim, 2008). Yang *et al.,* (2011) has studied and prepared biocomposites materials by mixing wheat gluten and rice proteins. In this study, reducing and crosslinking agents were used to improve the crosslinking and tensile strength of the two blended proteins.

Sustainability of gluten-based bioplastics

As in the initial sections related to petro-based plastics, sustainability problems of petro-based plastics have been clearly depicted. Plastics, derived from fossil fuels are largely unsustainable due to their social, environmental and health damaging effects (Poole *et al.,* 2009). So, considerable efforts are made on the development of sustainable bioplastics production from biomaterials (Nordhoff *et al.,* 2007). To be regarded as a truly sustainable resource, the biomaterial must possess the following properties.

- Limited utilization of resources (energy, cost, biomaterial)
- Material should be renewable
- Should be biodegradable or compostable

- Should be able to produce locally to avoid environment and economic effects of transportation
- Must possess sustainable character throughout the lifecycle of material; growing of biomass resource, polymer production, conversion to biodegradable plastic product, end user consumption (Sustainable bioplastic guidelines, 2007).

As described in figure 4, sustainability of bioplastics must be depicted at all the three levels; Social, economic and environment. So, the gluten as a resource material and its derived biodegradable films and bioplastics could be sustainable as they confer no long term effects on environment and could be adorable at social and economic levels.

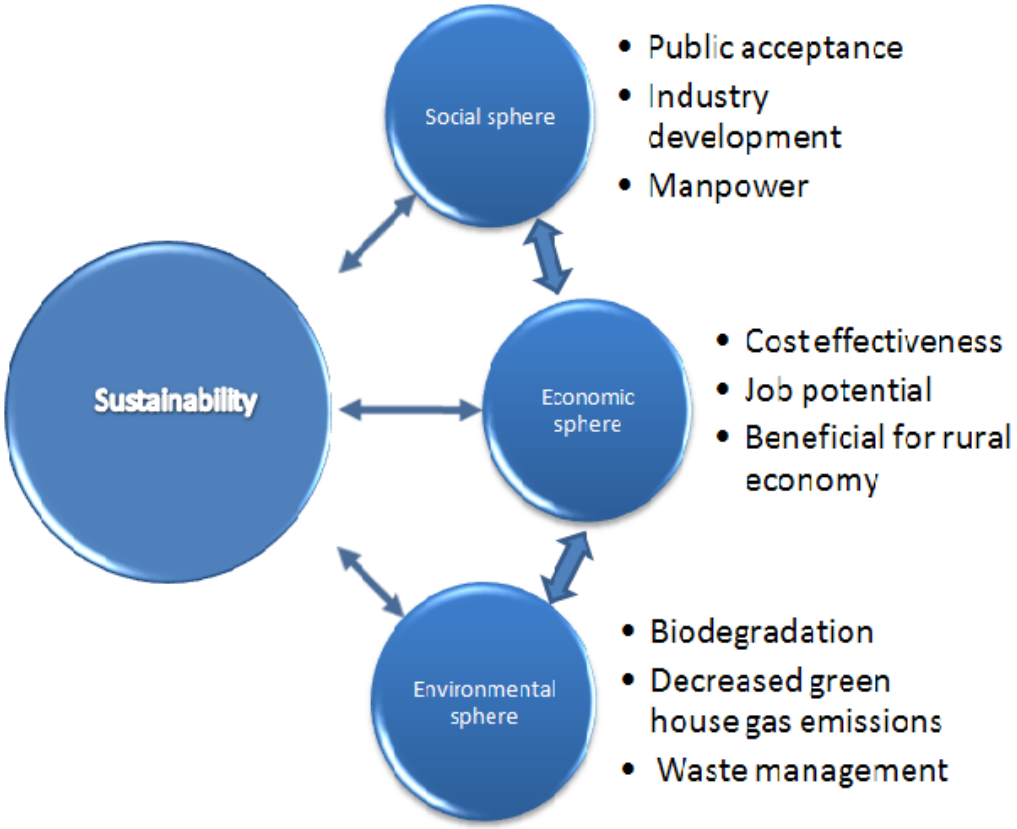

Figure 4: Concept of sustainability at economic, social and environmental level

Bioplastics vs. petroleum-based plastics

Bioplastics must confer some advantages in order to be used as commodity products over petroleum derived plastics. Table 3 represents a brief comparison of bioplastics with petroleum derived plastics as related to sustainability.

Table 3: Comparison of bioplastics with petroleum derived plastics (Industrial use for crops: Bioplastics www.hgca.com)

Properties	**Bioplastics**	**Petro-based plastics**
Renewable	Yes or partially	No
Sustainable	Yes	No
Break down in the Environment	Biodegradable and/or compostable	Usually undegradable Some degradable by polymer oxidation
Polymer range	Biopolymers (lipids, proteins, starch etc.), Bacterial polymers (Polyhydroxyalkanoates, polyhydroxybutyrate)	Extensive
Green house gas emissions	Low emission	High emission
Utilization of fossil fuels	Limited utilization	High utilization
Agriculture land utilization	Expected to increase	No utilization

Social and environmental impacts of bioplastics

Utilization of bioplastics has a number of advantages over petroleum-based plastics. Bioplastics confers important beneficial effects both at the social and environmental level. Some of their impacts and benefits to the environment and communities are listed below.

Reduced CO_2 emission

Petroleum derived synthetic polymers and plastic products are a big source of pollution, so biodegradable plastics derived from renewable biomass resources are an excellent alternative

(Sun *et al*., 2008). The amount of CO_2 released from one metric ton of bioplastic is 0.8-3.2 metric tonnes less than that released by petroleum derived plastics (Heath, 2007).

Rising fuel prices and depletion of fossil fuels

Reliance on fossil fuels for various industrial and domestic purposes is increasing. As a consequence their cost is increasing and their availability as a raw material will be decreased soon. So, development and innovations in bioplastic industry is essential in order to cope with the shortage of fossil fuels (Garrain *et al*. 2007). It is estimated that if the total amount of plastic which is utilized by the world is replaced with bioplastics, a total of 3.5 million barrels of oil reserves can be saved per day (Momani, 2009).

Economic Benefits

Bioplastics are economically beneficial because they can be fabricated by utilizing the available machinery used to synthesize the traditional plastics (Gomez-Martinez *et al*., 2009). Production of bioplastics can be cost effective if the amount of biopolymer produced in plant is high and the biomass which is left over is utilized for energy generation (Kurdikar *et al*., 2001). Gluten-based bioplastics are highly cost effective due to low price and abundant availability of the gluten and the option to utilize the existing plastic processing machinery. It is predictable that price of bioplastics and its derived products will keep on decreasing with time due to its competition with conventional plastic industries and development of new processing routes (European bioplastics, 2008). The bioplastic industry can also contribute to create the new job potentials and can boost up the rural economy due to increasing demand of agricultural crops.

Sustainable activity

Bioplastics are debated strongly to be sustainable, however there processing toward sustainability is slow. It may be due to the fact, that raw material for bioplastic production is derived from crops, so at present it is in limited supply as compared to raw material of conventional plastics. The use of genetically engineered plants and bacteria for the raw material production is arguable for public acceptance as they may create instability in ecosystems (Gaskell *et al*., 2006).

Biodegradation

Biodegradation of bioplastics means degradation of materials in nature by the action of microbes via enzymatic reactions (Mostafa *et al*., 2010). The use of biopolymer at industrial level can be approved more environment friendly when compared to synthetic petro-based polymers due to its biodegradability (Grifin, 1994). Different biopolymers undergo different changes in the biodegradation process according to their chemical structures and the type of soil in which they are buried (Mostafa *et al*., 2010).

Challenges for the bioplastic industry

Bioplastics also possess some disadvantages and at present are not providing a perfect solution to the problems created by petroleum derived products. The most challenging point for bioplastic production is not to violate the potential food sources. This obligation can be overcome by utilizing the non-food resources for the purpose. These are called as second generation bioplastics. However, these must be processable via common processing routes like extrusion, compression and injection-molding (Verbeek and Berg, 2009). Some bioplastics (e.g. derived from bacterial polymer polylactic acid) are only biodegradable in controlled conditions of temperature and humidity. This limitation must be overcome and bioplastics must be able to degrade in landfills (Matsuura *et al*., 2008). Cost of aliphatic polymers like polylactic acid must be reduced (present cost is between $2 and $5/1b) in order to compete with synthetic polymers (Yang *et al*., 1996). However, agriculture raw materials like wheat gluten, starch, corn zein and soy proteins are cheap and available in large quantity, but plastics produced from them are still brittle, highly viscous and hydrophilic. Therefore, they must be produced with a plasticizer (John *et al*., 1998). Bioplastics, when subjected to biodegradation under anaerobic conditions release methane in landfills. In order to compete with the problem and to produce valuable composts for the soil improvement, bioplastic products should be collected separately from other non-biodegradable materials and then can be composed at industrial level (Song *et al*., 2009).

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References

Anderson, O.D., Kuhl, J.C. and Kuhl, A.T. 1996. Construction and expression of a synthetic wheat storage protein gene. *Gene,* 174: 51–58.

Aykroyd, W.R. and Doughty, J. 1970. Wheat in human nutrition. FAO Nutr Stud., 23: 1-163.

Bailey, C. H. 1941. A translation of Beccari's lecture 'concerning grain' 1728. *Cereal Chem.*, 18: 555–561.

Barnard, G. N. and Sander, J. K. 1989. The poly-b-hydroxybutyrate granule in vivo. A new insight based on NMR spectroscopy of whole cells. *J Biol Chem*., 264: 3286-3291.

Barnes, D. K. A., Galgani, F., Thompson, R. C. and Barlaz, M. 2009. Accumulation and fragmentation of plastic debris in global environments. *Phil. Trans. R. Soc*., 364, 1985–1998.

Bekes, F., Anderson, O.D., Gras, P.W., Gupta, R.B., Tam, A., Wrigley, C.W. and Appels, R. 1994. The contribution to mixing properties of 1D glutenin subunits expressed in a

bacterial system. In: Henry, R., Ronalds, J. (Eds.), Improvement of Cereal Quality by Genetic engineering. Plenum Press, New York, USA, pp. 97–104.

Belton, P. S. 1999. On the elasticity of wheat gluten. *J. Cereal Sci.*, 29: 103-107.

Bietz, J. A. Huebner, F.R., Sanderson, J.E. and Wall, J.S. 1976. Wheat gliadin homology revealed through N-terminal amino acid sequence analysis. *Cereal chem.,* 54: 1070-1083.

Bietz, J. A. and Lookhart, G. L. 1996. Properties and non-food potential of gluten. *Cereal Foods World,* 41: 376-382.

Blechl, A.E., Thrasher, K.S., Vensel, W.H. and Greene, F.C., 1992. Purification and characterization of wheat alpha-gliadin synthesized in the yeast, Saccharomyces cerevisiae. *Gene,* 116: 119–127.

Brabias, B. and Swiatek, L. 1998. *Pharmaceut. Pharmacol. Lett.* 8: 81-83.

Branlard, G. and Dardevet, D. 1985. Diversity of grain protein and bread wheat quality. II. Correlation between high molecular weight subunits of glutenin and flour quality characteristics. *J. Cereal Sci*., 3: 345-354.

Brauer, S., Meister, F., Gottlober, R.-P. and Nechwatal, A. 2007. Preparation and thermoplastic processing of modified plant proteins. *Macromol. Mater. Eng., 292*: 176-183.

Buonocore, F., Bertini, L., Ronchi, C., Bekes, F., Caporale, C., Lafiandra, D., Gras, P., Tatham, A.S., Greenfield, J., Halford, N.G. and Shewry, P. R. 1998. Expression and functional analysis of Mr 58,000 peptides derived from the repetitive domain of HMW subunit 1Dx5. *J. Cereal Sci.*, 27: 209-215.

Carter, B. 2002. The Importance of Wheat Quality, Agricultural horizons, Cooperating Agencies: Washington State University, U.S. Department of Agriculture, and Eastern Washington Counties. Cooperative. 1-3.

Cho, S-W., Gallstedt, M. and Hedenqvist, M. S. 2010. Properties of Wheat Gluten/Poly(lactic acid) Laminates. *J. Agric. Food Chem*., 58: 7344–7350.

Clarke, B.C., Phongkham, T., Gianibelli, M.C., Beasley, H. and Bekes, F. 2003. The characterization and mapping of a family of LMW-gliadin genes: effects on dough properties and bread volume. *Theor.Appl. Genet*., 106: 629-635.

Cuq, B., Gontard, N. and Guilbert, S. 1998. Proteins as Agricultural Polymers for Packaging Production. *Cereal chem.,* 75: 1-9.

D'Ovidio, R., Anderson, O.D., Masci, S., Skerritt, J. and Porceddu, E. 1997. Construction of novel wheat high-Mr glutenin subunit gene variability: modification of the repetitive domain and expression in *E. coli. J. Cereal Sci.*, 25: 1–8.

Day, L., Augustinab, M. A., Bateybc, I. L. and Wrigley, C. W. 2006. Wheat-gluten uses and industry needs. *Trends in Food Sci. & Technol.*, 17: 82-90.

Devassine, M., Henry, F., Guerin, P., and Briand, X. 2002. Coating of fertilizers by degradable polymers. *Int. J. Pharmacogn.*, 242: 399-404.

Domenek, S., Feuilloley, P., Gratraud J., Morel, M. H. and Guilbert S. 2004. Biodegradability of wheat gluten based bioplastics. *Chemosphere* 54, 551-559.

Domenek, S., Morel, M. H., Redl, A. and Guilbert, S. 2003. Rheological investigation of swollen gluten polymer networks: Effects of process parameters on cross-link density. *Macromol. Symp*., 200: 137-145.

Dowd, C. and Bekes, F. 2002. Large-scale expression and purification of HMW-glutenin subunits. *Protein Expression purify.*, 25: 97-104.

Dupont. 2001. Press release: Genencor International and Dupont expand R&D collaboration to make key biobased polymer. http://www1.dupont.com/NASApp/dupontglobal/corp/index.jsp?page=/content/US/en_US/news/product/2001/pn03_12_01.html

Elmorjani, K., Thievin, M., Michon, T., Popineau, Y., Hallet, J. N. and Gueguen, J. 1997. Synthetic genes specifying periodic polymers modelled on the repetitive domain of wheat gliadins: conception and expression. *Biochem. Biophys. Res. Commun.*, 239: 240–246.

El-Wakil, N. A. 2009. Use of Lignin Strengthened with Modified Wheat Gluten in Biodegradable Composites. *J. Appl. Polym. Sci.*, 113: 793-801.

EPA, what are endocrine disruptors. http://www.epa.gov/endo/pubs/edspoverview/whatare.htm.

European Bioplastics. 2008. www.european-bioplastics.org.

Falkman, E. G. 1995. Sustainable P roduction and Consumption : A Business Perspective. Waste mangement international. 1-32.

Feeney, K. A., Tatham, A. S., Gilbert, S. M., Fido, R. J., Halford, N.G. and Shewry, P.R. 2001. Synthesis, expression and characterisation of peptides comprised of perfect repeat motifs based on a wheat seed storage protein. *Acta biochim. biophys. Sin.*, 1546: 346–353.

Feeney, K. A., Wellner, N., Gilbert, S. M., Halford, N. G., Tatham, A. S., Shewry, P. R. and Belton, P. S. 2003. Molecular structure and interactions of repetitive peptides based on

wheat glutenin subunits depend on chain length. *Biopolymers (Biospectroscopy)*, 72:123-131.

Galili, G. 1989. Heterologous expression of a wheat high molecular weight glutenin gene in Escherichia coli. *Proc. Natl. Acad. Sci*., 86: 7756-7760.

Garrain, Daniel, Vidal, R., Martinez, P., Franco, V. and David, C. T. 2007. LCA of biodegradable multilayer film from biopolymers. Paper presented at 3rd International Conference on Life Cycle Management.

Gaskell, G., Allansdottir, A., Allum, N., Corchero, C., Fischler, C., Hampel, J., Jackson, J., Kron-berger, N., Mejlgaard, N., Revuelta, G., Schreiner, C., Stares, S., Torgersen, H. and Wagner, W. 2006. Europeans and biotechnology in 2005: patterns and trends, Eurobarometer. European Commission's Directorate-General for Research, Brussels, Belgium.

Gaspar, M., Benko, Z. S., Dogossy, G., Reczey, K., and Czigany, T. 2005. Reducing water absorption in compostable starch-based plastics. *Polym. Degrad. Stabil.* 90: 563-569.

Gennadios, A. 2002. Protein-Based Films and Coatings, CRC Press, New York, 2002, pp. 69-115.

Gervet, B. 2007. The use of crude oil in plastic making contributes to global warming.

Gianibelli, M. C., Larroque, O. R., MacRitchie, F. and Wrigley, C. W. 2001. Biochemical, Genetic, and Molecular Characterization of Wheat Endosperm Proteins. American Association of Cereal Chemists, Inc. 1-20.

Gielen, D., Newman, J. and Patel, M. K. 2008. Reducing Industrial Energy Use and CO2 Emissions: The Role of Materials Science. *Harnessing Materials for Energy*, 33. 817-833.

Gilbert, S. M., Wellner, N., Belton, P. S., Greenfield, J.A., Siligardi, G., Shewry, P. R. and Tatham, A.S. 2000. Expression and characterisation of a highly repetitive peptide derived from a wheat seed storage protein. *Acta biochim. biophys. Sin.*, 1479: 135–146.

Gómez-Martínez, D., Partal, P., Martínez, I. and Gallegos, C. 2009. Rheological behaviour and physical properties of controlled-release gluten-based Bioplastics. *Bioresour. Technol.*, 100: 1828-1832.

Gontard, N., Guilbert, S. and Cuq J. L. 1992. Edible wheat gluten films: Influence of the main process variables on film properties using response surface methodology. *J. Food Sci.*, 57: 190-195.

Gregory, M. R. 2009 Environmental implications of plastic debris in marine settings-entanglement, ingestion, smothering, hangers-on, hitch-hiking and alien invasions. *Phil. Trans. R. Soc. B.* 364: 2013-2025.

Grifin, G. J. L. 1994. Editor. Chemistry and technology of biodegradable polymers. London: Blackie Academic & Professional.

Grosch, W. and Wieser, H. 1999. Redox reactions in wheat dough as affected by ascorbic acid. *J. Cereal Sci.* 29: 1-16.

Guilbert, S and Cuq, B. 2005. Material formed from proteins. In: C. Bastioli, Editor, *Handbook of Biodegradable Polymers*, Rapra Technology Limited, United Kingdom.

Gupta, R. B., Khan, K., and MacRitchie, F. 1993. Biochemical basis of flour properties in bread wheats. I. Effects of variation in the quantity and size distribution of polymeric protein. *J. Cereal Sci*., 18: 23-41.

Halford, N. G., Field, J. M., Blair, H., Urwin, P., Moore, K., Robert, L., Thompson, R., Flavell, R. B., Tatham, A. S. and Shewry, P. R. 1992. Analysis of HMW glutenin subunits encoded by chromosome 1A of bread wheat (Triticum aestivum L.) indicates quantitative effects on grain quality. *Theor. Appl. Genet*., 83: 373-378.

Heath, N. 2007. Bio-plastics: Turning Wheat And Potatoes into Plastics. *Science articles*.

Hernandez-Munoz, P., Kanavouras, A., Perry, K. W. and Gavara R. 2003. Development and Characterization of Biodegradable Films Made from Wheat Gluten Protein Fractions. *J. Agric. Food Chem.*, 51: 7647-7654.

Hernandez-Munoz, P., Lagaron, J. M., Lopez-Rubio, A. and Gavara, R. 2004. Gliadins polymerized with Cysteine: Effects on the Physical and Water Barrier Properties of Derived Films. *Biomacromolecules*, 5: 1503-1510.

Hopewell, J., Dvorak, R. and Kosior, E. 2009. Plastics recycling: challenges and opportunities. *Phil. Trans. R. Soc. B.*, 364: 2115–2126.

http://polymer.w99of.com/history-of-polymers/

http://www.bpf.co.uk/Oil_Consumption.aspx.

Hussain and Qamar. 2007. Wheat genomics, challenges and alternative strategies. *Proc. Pak. Acad. Sci.*, 44: 305-306.

Industrial use for crops: Bioplastics www.hgca.com.

International Energy Agency (IEA) (2004), *Oil* Crises and Climate Challenges: 30 Years of Energy Use in IEA Countries, IEA/OECD, Paris, France.

Irissin-Mangata, J., Bauduin, G., Boutevin, B. and Gontard, N. 2001. New plasticizers for wheat gluten films. *Eur. Polym. J.*, 37: 1533-1541.

Jane, J., Lim, S., Paetau, I., Spence, K., and Wang, S. 1994. Biodegradable plastics made from agricultural biopolymers. in: Polymers from Agricultural Coproducts. M. L. Fishman, R. B. Friedman, and S. J. Huang, eds. ACS Symposium Series 575: Chicago. pp 92-100.

Jerez, A. A., Partal, P., Martinez, A. I., Gallegos, C. and Guerrero, A. 2005. Rheology and processing of gluten based bioplastics. *Biochem. Engineering J.*, 26: 131-138.

Johansson, E., Henriksson, P., Svensson, G. and Heneen, W. K. 1993. Detection, chromosomal location and evaluation of the functional value of a novel highMr glutenin subunit found in Swedish wheats. *J. Cereal Sci*., 17: 237–245.

Johansson, E., Kuktaite, R. and Prieto-Linde, M. L. 2001. Improving wheat quality by modifying protein composition. Proceedings of the Latvian Academy of Sciences Section B, Natural Exact and Applied Sciences, 55: 185-190.

John, J., Tang, J. and Bhattacharya, M. 1998. Processing of biodegradable blends of wheat gluten and modified polycaprolactone. *Polymer*, 39: 2883-2895.

Kim, S. 2008. Processing and properties of gluten/zein composite. *Bioresource Technol.*, 99: 2032-2036.

Krull, L. H. and Inglett G.E. 1971. Industrial uses of gluten. *nm sci. today*, 16: 232.

Kurdikar, D., Fournet, L., Slater, S. C., Paster, M., Gruys, K. J., Gerngross, T. U. and Coulon, R. 2001. Greenhouse Gas Profile of a Plastic Material Derived from a Genetically Modified Plant. *J Ind. Eco*., 4:107-122.

Lagrain, B., Goderis, B., Brijs, K. and Jan, A. 2010. Delcour. Molecular Basis of Processing Wheat Gluten toward Biobased Materials. *Biomacromolecules*, 11: 533-541.

Laszity, R., Tomoskozi, S., Haraszi, R., Revay, T. and Karpati, M. 2000. Is the role of HMW-GS is decisive in determination of baking quality of wheat? *Wheat Gluten, R. soc. chem.,* 38-42.

Lee, Y. K., Beke s, F., Gras, P., Ciaffi, M., Morell, M. K. and Appels, R. 1999. The low molecular weight glutenin subunit proteins of primitive wheats. IV. Functional properties of products from individual genes. *Theor. Appl. Genet*., 98: 149-155.

Li, W., Dobraszczyk, B. J., Dias, A. and Gil, A. M. 2006. Polymer Conformation Structure of Wheat Proteins and Gluten Subfractions Revealed by ATR-FTIR. *Cereal chem.,* 83: 407-410.

Magnuson, K. M. 1985. Uses and functionality of vital wheat gluten. *Cereal Foods World*, 30: 79-181.

Maruyama, N., Ichise, K., Katsube, T., Kishimoto, T., Kawase, S., Matsumura, Y., Takeuchi, Y., Sawada, T. and Utsumi, S., 1998. Identification of major wheat allergens by means of the Escherichia coli expression system. *European J. of Biochem.,* 255: 739–745.

Matsuura, E., Ye, Y. and He, X. 2008. Sustainability Opportunities and Challenges of bioplastics. School of Engineering. Blekinge Institute of Technology. Karlskrona, Sweden.

McRandle, P. 2004. Plastic Water Bottles, *National Geographic*, 101.

Mecking, S. 2004. Nature or Petrochemistry?—Biologically Degradable Materials. *Angew. Chem. Int. Ed*., 43: 1078 –1085.

Momani, B. 2009. Assessment of the Impacts of Bioplastics: Energy Usage, Fossil Fuel Usage, Pollution, Health Effects, Effects on the Food Supply, and Economic Effects Compared to Petroleum Based Plastics. Worcester Polytechnic Institute, 1-58.

Montain, L., Dez, I. and Madec, P. 2004. Development of new composites material, carriers of active agents, from biodegradable polymers and wood. *C.R. Chim.*, 7: 635-640.

Mooney, B.P., 2009. The second green revolution? Production of plant-based biodegradable plastics. *Biochem. J*. 418: 219-232.

Mostafa, H. M., Sourell, H. and Bockisch, F. J. 2010. The Mechanical Properties of Some Bioplastics Under Different Soil Types for Use as a Biodegradable Drip Tubes. *Agricultural Engineering International: the CIGR Ejournal*., 12: 1-16.

Murphy, R and Bartle, I. 2004. Summary report, biodegradable polymers and sustainability: insight from lifecycle assessment. National non food crop centre, Uk.

Nordell, B. 2003. Thermal Pollution Causes Global Warming. *Global and Planetary Change*. 38: 305-312.

Nordhoff, S., Höcker, H. and Gebhardt, H. 2007. Renewable resources in the chemical industry– breaking away from oil? *Biotechnol. J.*, 2: 1505-1513.

Olabarrieta, I., Cho, S. W., Gallstedt, M.., Sarasua, J. R., Johansson, E. and Hedenqvist, M. S. 2006. Aging properties of films of plasticized vital wheat gluten cast from acidic and basic solutions. *Biomacromolecules*. 7: 1657–1664.

Osborne, T. B. 1924. The vegetable proteins, 2nd edn. London:Longmans, Green and Co.

Patacchini, C., Masci, S., D'Ovidio, R. and Lafiandra, D. 2003. Heterologous expression and purification of native and mutated LMW-GS from durum wheat. *Journal of Chromatograph*, 786: 215–220.

Payne, P. I., Holt, L.M., Krattiger, A. F. and Carillo, J. M. 1988. Relationship between seed quality characteristics and HMW glutenin composition determined using wheat grown in Spain. *J. Cereal Sci*., 7: 229-235.

Payne, P. I., Holt, L.M., Lawrence, G.J. and Law, C.N. 1982. The genetics of gliadin and glutenin, the major storage protein of wheat endosperm. *Qualitas Plant. Pl. Fds . Hum. Nutr*., 31: 229-241.

Payne, P. I., Nightingale, M. A., Krattiger, A. F. and Holt, L.M. 1987. The relationship between HMW glutenin sub-unit composition and the breadmaking quality of British grown wheat varieties. *J. Sci. Food Agric*., 40: 51-65.

Pommet, M., Redl, A., Guilbert, S. and Morel, M.-H. 2005. Intrinsic influence of various plasticizers on functional properties and reactivity of wheat gluten thermoplastic materials. *J. Cereal Sci*., 42: 81-91.

Pommet, M., Redl, A., Morel, M. H., Domenek, S. and Guilbert, S. 2003. Thermoplastic processing of protein-based bioplastics: Chemical engineering aspects of mixing, extrusion and hot molding. *Macromol. Symp*., 197: 207–217.

Poole, A. J., Jeffrey, S. C., and Mickey, G. H. 2009. Environmentally Sustainable Fibers from Regenerated Protein. *Biomacromolecules*, 10: 1-8.

Popineau, Y., Deshayes, G., Lefebvre, J., Fido, R., Tatham, A. S. and Shewry, P. R. 2001. Prolamin aggregation, gluten viscoelasticity, and mixing properties of transgenic wheat lines expressing 1Ax and 1Dx high molecular weight glutenin subunit transgenes. *J. Agric. Food Chem*., 49: 395-401.

Pratt, K. A., Madgwick, P. J. and Shewry, P.R., 1991. Expression of a wheat gliadin protein in yeast (Saccharomyces cerevisiae). *J. Cereal Sci.*, 14: 223–229.

Rajaram, S. 2005. Role of conventional plant breeding and biotechnology in future wheat production. *Turk J Agric*, 29: 105-111.

Ramkumar, D. H. S., Bhattacharya, M., Vaidya, U. R., Hakkainen, M., Albertsson, A. C. and Karlsson, S. 1996. Properties of injection moulded Starch/synthetic polymer blends-i. Effect of Processing parameters on physical properties. *Eur. Polym. J.* 32: 999-1010.

Reddy, N. and Yang, Y. 2007. Novel Protein Fibers from Wheat Gluten. *Biomacromolecules*, 8: 638-643.

Rooke, L., Bekes, F., Fido, R., Barro, F., Gras, P., Tatham, A. S., Barcelo, P., Lazzeri, P. and Shewry, P. R. 1999. Overexpression of a Gluten Protein in transgenic Wheat Results in Greatly Increased Dough Strength. *J. Cereal Sci.*, 30: 115-120.

Sartor, G. and Johari, G. P. 1996. Polymerization of a Vegetable Protein, Wheat Gluten, and the Glass-Softening Transition of Its Dry and Reacted State. *J. Phys. Chem.*, 100: 19692-19701.

Seilmeier, W., Belitz, H. D. and Wieser, H. 1991 Separation and quantitative determination of high-molecular-weight sub-units of glutenin from different wheat varieties and genetic variants of the variety. Sicco. Z. Lebensm. *Unters. Forsch.*, 192: 124-129.

Shewry, P. R. and Tatham, A. S. 1997. Disulphide bonds in wheat gluten proteins. *J. Cereal Sci.* 25: 207-227.

Shewry, P. R., D'Ovidio, R., Lafiandra, D., Jenkins, J. A., Mills, E. N. C. and Bekes, F. 2009. Wheat grain proteins. In *Wheat: chemistry and technology, 4th ed.* K. Khan and P. R. Shewry, eds, 223-298. AACC International, St Paul.

Shewry, P. R., Halford, N. G., and Tatham, A. S. 1992. The high molecular weight subunits of wheat glutenin. *J. Cereal Sci*., 15:105-120.

Shewry, P. R., Halford, N. G., Belton, P. S. and Tatham, A. S. 2002. The structure and properties of gluten: an elastic protein from wheat grain. *Phil. Trans. R. Soc. Lond. B*., 357: 133-142.

Shewry, P. R., Tatham, A. S., Forde, J., Kreis, M. and Miflin, B. J. 1986. The classification and nomenclature of wheat gluten proteins: A reassessment. *J. Cereal Sci*., 4: 97-106.

Song, Y. Zheng, Q. and Zhang, Q. 2009. Rheological and mechanical properties of bioplastics based on gluten- and glutenin-rich fractions. *J. Cereal Sci.*, 50: 376-380.

Song, Y. and Zheng, Q. 2008. Preparation and properties of thermo-molded bioplastics of glutenin-rich fraction. *J. Cereal Sci.*, 48: 77-82.

Sourice, S., Nisole, A., Gue ´guen, J., Popineau, Y. and Elmorjani, K. 2003. High microbial production and characterization of strictly periodic polymers modelled on the repetitive domain of wheat gliadins. *Biochem. Biophysic. Res. Commun*., 312: 989–996.

Sudesh, K., Abe, H. and Doi, Y. 2000. Synthesis, structure and properties of polyhydroxyalkanoates: biological polyesters. *Prog Polym Sci*., 25: 1503-1555.

Sun, S., Song, Y. and Zheng, Q. 2008. Morphology and mechanical properties of thermo-molded bioplastics based on glycerol-plasticized wheat gliadins. *J. Cereal Sci.*, 48: 613-618.

Sun, S., Song, Y. and Zheng, Q., 2007. Morphologies and properties of thermo-molded biodegradable plastics based on glycerol-plasticized wheat gluten. *Food Hydrocolloids*, 21: 1005-1013.

Tamas, L. and Shewry, P. R. 2006. Heterologous expression and protein engineering of wheat gluten proteins. *J. Cereal Sci*, 43: 259–274.

Tamas, L., Bekes, F., Greenfield, J., Tatham, A. S., Gras, P.W., Shewry, P. R. and Appels, R. 1998. Heterologous expression and dough mixing studies of wild-type and mutant C hordeins. *J. Cereal Sci.*, 27: 15-22.

Tatham, A.S., Shewry, P.R., 1985. The conformation of wheat gluten proteins. The secondary structures and thermal stabilities of α-, β-, *γ*- *ω*-gliadins. *J. Cereal Sci.*, 3: 104–113.

Tatham, A. S., Shewry, P. R. and Miflin, B. J. 1984. Wheat gluten elasticity: a similar molecular basis to elastin? *FEBS Lett.*, 177: 205-208.

Thompson, R. C., Moore, C. J., Saal, F. S., and Swan, H. S. 2009. Plastics, the environment and human health: current consensus and future trends. *Phil. Trans. R. Soc. B., 364*: 2153–2166.

Thompson, S., Bishop, D.H.L., Madgwick, P., Tatham, A.S. and Shewry, P.R. 1994. High-level expression of a wheat LMW glutenin subunit using a baculovirus system. *J. Agric. Food Chem,*. 42: 426–431.

Ullsten, N. H., Gallstedt, M., Graslund, A., Johansson, E. and Hedenqvist, M. S. 2006. Enlarged processing window of plasticized wheat gluten using salicylic acid. *Biomacromolecules.* 7: 771-776.

Van Dijk, A. A., van Swieten, E., Kruize, I. T. and Robillard, G. T. 1998. Physical characterisation of the N-terminal domain of high-molecular weight glutenin subunits Dx5 from wheat. *J. Cereal Sci*., 28: 115-126.

Vasil, I. K. and Anderson, O. D. 1997. Genetic engineering of wheat gluten. *Trends in plant sciences.* 2: 1360-1385.

Vaz, C. M., Fossen, M., Tuil, R. F., Graff, L. A., Reis, R. L. and Cunha, A. M. 2003. Casein and soybean protein-based thermoplastics and composites as alternative biodegradable polymers for biomedical applications. *J. Biomed. Mater. Res.*, 65: 60-70.

Veraverbeke, W. S. and Delcour, J. A. 2002. Wheat protein composition and properties of wheat glutenin in relation to breadmaking functionality. *Crit. Revl. Food Sci. Nutr*., 42: 179-208.

Verbeek, C. J. R. and Berg, L. E. V. 2009. Recent Developments in Thermo-Mechanical Processing of Proteinous Bioplastics. *Recent Patents on Materials Science*, 2: 171-189.

Wade, L.G. 2006. Organic Chemistry, 6th Ed., p. 1222, Pearson Prentice Hall. British Plastics Federation, *Oil Consumption.*

Weegels, P. L., van de Pijpekamp, A. M., Graveland, A., Hamer, R. J. and Schofield, J. D. 1996. Depolymerization and re-polymerization of wheat glutenin during dough processing relationships between glutenin macropolymer content and quality parameters. *J. Cereal Sci*., 23: 103-111.

Wellner, N., Marsh, J., Savage, A., Halford, N.G., Shewry, P.R., Mills, E.N.C. and Belton, P.S. 2006. Comparison of repetitive sequences derived from high molecular weight subunits of wheat glutenin, an elastomeric plant protein. *Biomacromolecules,* 7**:** 1096-1103.

Wellner, N., Mills, E. N. C., Brownsey, G., Wilson, R. H., Brown, N., Freeman, J., Halford, N. G., Shewry, P. R. and Belton, P. S. 2005. Changes in proteisecondarystructure during gluten deformation studied by dynamic Fourier transform infrared spectroscopy. *Biomacromolecules*, 6: 255-261.

Wieser, H. 2007. Chemistry of gluten proteins. *Food Microbiol.*, 24: 115-119.

Wieser, H. and Kieffer, R. 2001. Correlations of the amount of gluten protein types to the technological properties of wheat flours determined on a micro-scale. *J. Cereal Sci.*, 34: 19-27.

Willke, T. and Vorlop, K. D. 2004. Industrial bioconversion of renewable resources as an alternative to conventional chemistry. *Appl. Microbiol. Biotechnol*, 66: 131–142.

Woerdeman, D. L., Veraverbeke, W. S., Parnas, R. S., Johnson, D., Delcour, J. A., Verpoest, I. and Plummer, C. G. J. 2004. Designing New Materials from Wheat Protein. *Biomacromolecule*, 5: 1262-1269.

Wretfors, C., Cho, S-W., Hedenqvist, M. S., Marttila, S., Nimmermark, S. and Johansson, E. 2009. *J. Polym. Environ*., 17: 259-266.

Wretfors, C., Cho, S-W., Kuktaite, R., Hedenqvist, M. S., Marttila, S., Nimmermark, S. and Johansson, E. 2010. Effects of fiber blending and diamines on wheat gluten materials reinforced with hemp fiber. *J. Mater Sci*., 45: 4196–4205.

Wrigley, C. W. 1996. Giant proteins with flour power. *Nature*, 381: 738-739.

Wrigley, C.W. and Bietz, J. A. 1988. Proteins and amino acids. In: Pomeranz Y. (Ed.), Wheat—Chemistry and Technology, vol. 1. St. Pau American Association of Cereal Chemistry, pp. 159–275.

www.plasticisland.org

Yang, Z., Bhattacharya, M. and Vaidya, U. R.. 1996. *Polymer*, 37: 2137.

Yang, Y-Y., Zhang, K-Z., Songa, Y-H. and Zheng, Q. 2011. Preparation and properties of wheat gluten/rice protein composites plasticized with glycerol. *Chinese J. Polymer Sci.*, 29: 87-92.

Ye, P., Reitz, L., Horan, C. and Parnas, R. 2006. Manufacture and Biodegradation of Wheat gluten/basalt Composite Material. *Journal of Polymers and the Environment*, 14: 1-7.

Yuan, Q., Lu, W. and Pan, Y. 2010. Structure and properties of biodegradable wheat gluten/attapulgite nanocomposite sheets. *Polymer Degradation and Stability*, 95: 1581-1587.

Zan, L., Fa, W. and Wang, S. 2006. Novel photodegradable low-density polyethylene–TiO2 nanocomposite film. *Environ. Sci. Technol*., 40, 1681–1685.

Zubris, K. A. V. and Richards, B. K. 2005. Synthetic fibers as an indicator of land application of sludge. *Environ. Pollut.* 138, 201–211.