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Celiac Disease: Advances in Treatment via Gluten Modification

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Celiac disease (CD) is an autoimmune enteropathy that occurs in genetically susceptible individuals carrying the prerequisite genetic markers HLA DQ2 or DQ8. These genetic markers are present in approximately 30% of the population, and the worldwide prevalence of CD is estimated to be approximately 1%–2%. Currently a gluten-free diet is the only treatment for CD, but novel therapies aimed at gluten modification are underway. This review will discuss gluten-based therapies including wheat alternatives and wheat selection, enzymatic alteration of wheat, oral enzyme supplements, and polymeric binders as exciting new therapies for treatment of CD.

Keywords: Gluten; Treatment; Celiac; Autoimmune; Therapies.

When gluten is ingested, gastric and pancreatic proteases cleave gluten into peptide fragments, some of which are immunogenic in celiac disease (CD). Gluten is a group of wheat proteins composed of gliadins, high-molecular-weight glutenin subunits (HMW-GS), and low-molecular-weight glutenin subunits (LMW-GS).[1] The gliadins tend to be more immunogenic in CD and are further subdivided into α, β, γ, and ω fractions. Within these subfractions, the α-gliadin has been shown to be the most toxic, with β, γ, and ω having decreasing toxicity.[2–4]

Glutamine residues in α-gliadin undergo deamidation to glutamic acid via tissue transglutaminase (tTG), thereby increasing the immunogenicity of some gluten-derived peptides. Presentation of the immunogenic deamidated gliadin peptides by HLA DQ2 or HLA DQ8 molecules on antigen-presenting cells (APCs) to T cells results in the activation of T cells and the production of inflammatory cytokines (Figure 1). The subsequent inflammation ultimately causes small intestinal injury that is characterized by villous atrophy, crypt hyperplasia, and increased intraepithelial leukocytosis.[4] The severity of disease is highly variable and ranges from asymptomatic to severe malnutrition. Complications of continued gluten exposure include increased risk of lymphoma, osteoporosis,[5] anemia, infertility,[6,7] neurologic problems, and cancer.[7]

The current mainstay of treatment for CD is a gluten-free diet (GFD); however, as many as 50% of patients with CD do not achieve histologic remission with a GFD.[5] Daily consumption of as little as 50 mg of gluten, the equivalence of 1/100th of a slice of bread, may contribute to the persistence of mucosal damage.[5,8] It is estimated that patients who are adherent to a GFD will consume on average 5–50 mg of gluten each day as a result of gluten contamination.[8] These concerns and observations have given way to increased interest in therapeutics that may either replace or act in conjunction with a GFD to help diminish the accidental gluten exposures that may account for continued disease activity.

What Are the Findings?

Wheat Alternatives and Generating Celiac-Safe Wheat

Gluten replacement. One gluten-based therapeutic approach is to use grains that do not have the immunogenic proteins found in wheat-derived gluten (Figure 1). One such grain that has garnered some interest as a wheat substitute is sorghum, which is a cereal grain related to maize. It has been consumed for thousands of years in Africa and Asia. More recently, sorghum has been used to make multiple wheat-free products, including breads, tortillas, cookies, and flatbreads that do not have discolorations or odd tastes. In vitro organ cultures do not show an increased production of inflammatory markers indicative of the activation of the innate or adaptive immune systems. In vivo challenge for 5 days in 2 patients with CD did not show onset of gastrointestinal symptoms or changes in serum anti-tTG levels.[9] Additional studies assessing morphologic changes and larger populations will be needed before further recommendations can be made on the safety of sorghum for patients with CD. Gluten contamination of sorghum used commercially has been an issue.

Gluten removal. Another approach is to use breeding and hybridization of different wheat species to remove the immunogenic gluten-derived proteins. There are significant differences in the levels of T-cell stimulatory epitopes in the gluten of the different wheat species and cultivars,[4] and T-cell assays have suggested that there may be a way to alter or breed new varieties of wheat that contain fewer toxic peptides. However, one of the obstacles to rendering it gluten-free is the role that gluten plays in the viscoelasticity and polymerization of the bread.[1]

Genes encoding the various components of gluten are predominantly contained on chromosomes 1 and 6 of the 3 homologous genomes of hexaploid bread wheat (*Triticum aestivum*) (AABBDD). Genes located within the Gli-2 locus of the short arm of chromosome 6D encode most of the α-gliadins.[1,10] Removal of this locus

Abbreviations used in this paper: APCs, antigen-presenting cells; ASP, aspergillopepsin; CD, celiac disease; GFD, gluten-free diet; HMW-GS, high-molecular-weight glutenin subunits; IFN, interferon; LMW-GS, low-molecular-weight glutenin subunits; P(HEMA-*co*-SS), poly (hydroxyethyl methacrylate-co-styrene sodium sulfonate); tTG, tissue transglutaminase.

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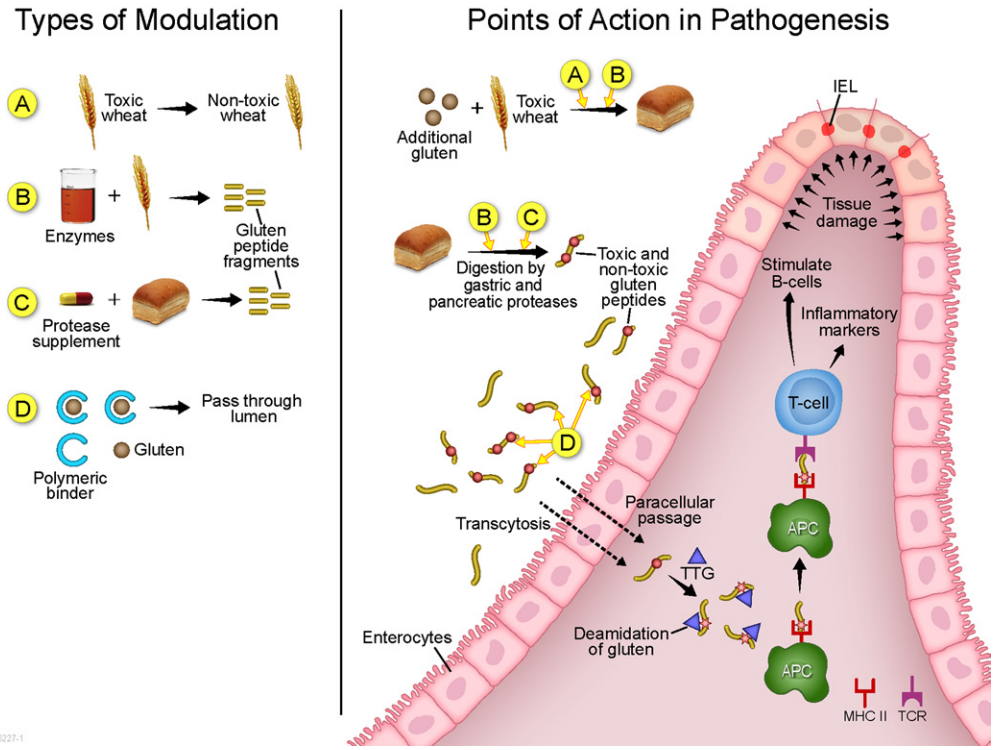

Figure 1. On the *left side* are the 4 categories in which gluten can be modified: (*A*) gluten removal via wheat mutagenesis, (*B*) enzymatic pretreatment of wheat flour, (*C*) oral enzyme supplements, and (*D*) polymeric binders. On the *right side* are the points in which the pathogenesis of CD that these 4 categories would act. IEL, intraepithelial lymphocyte.

in the Chinese Spring cultivar of *Triticum aestivum* results in the removal of T-cell stimulatory epitopes but also changes the mechanical properties of wheat. Removal of the ω-gliadin, γ-gliadin, and LMW-GS loci from chromosome 1 of the D-genome removed T-cell stimulatory epitopes but does not appear to negatively affect mechanical properties of wheat.[1]

C173 is an experimental wheat line derived from breeding 2 mutant plants with spontaneous deletions of several gliadins and glutenins, specifically Gli-A2, Gli-D1, and Gli-D3. In vitro studies with duodenal mucosal biopsies from treated and healed CD patients showed that C173 does not decrease the villous-to-crypt ratio but does increase the release of the inflammatory cytokines interferon (IFN)-γ and interleukin-2 as well as the production of interleukin-10 and anti-tTG antibodies in the collected supernatant.[3] The lack of morphologic changes would suggest then that deleting several gliadins and glutenins reduces the toxicity of this wheat; however, the continued production of inflammatory cytokines is of great concern. Therefore, C173 may not be appropriate for CD patients.

Enzymatic Pretreatment of Wheat Flour

Fermentation of wheat with sourdough lactobacilli and fungal proteases is another method of rendering wheat less toxic (Figure 1). Specific combinations of lactobacilli and fungal proteases result in complete hydrolyzation of gluten in wheat flour.[11] When treated with this hydrolyzed wheat flour, duodenal mucosa from CD patients do not produce IFN-γ mRNA greater than the level produced from duodenal mucosa from healthy controls.[12] The lactobacillus-treated wheat sourdough can be mixed with nontoxic flours (ie, oat, millet, and buckwheat flours) to produce a bread that has texture similar to wheat sourdough breads. This fermented bread does not appear to increase intestinal permeability when consumed by patients with CD.[11] When mixed with buckwheat to form pasta, this fermented wheat had lower stickiness and firmness in comparison with standard durum semolina wheat; however, these differences did not prohibit its manufacture and did not affect odor or taste.[13] In a randomized trial of 6 patients, those who ingested 200 g per day of fully hydrolyzed baked goods (containing 8 parts per million residual gluten) did not experience symptoms or morphologic or serologic changes after 60 days.[14] Hydrolyzed wheat flour still maintains its mechanical properties so that it can be used to make bread, pasta, and sweets.[13,14] A similar study involving ingestion of 200 g per day of fully hydrolyzed sweet baked goods did not show a change in clinical complaints, serology, or intestinal permeability.[12]

Another enzymatic pretreatment method uses transamidation to render the α-gliadin derived peptides nonimmunogenic. This enzymatic reaction can be achieved via incubation of commercial wheat flour with microbial transglutaminase and lysine methyl ester. When this altered flour is incubated for 48 hours with T-cell lines derived from duodenal biopsies from 12 adults with CD, IFN-γ expression is less than nonaltered flour, and binding of immunogenic peptides to DQ2 is decreased.[15]

Ultimately, it may not be feasible to alter or select a variety of wheat completely devoid of gluten without compromising its mechanical properties; however, current research suggests that less toxic wheat could be designed and may be tolerated better in patients with CD. Conceivably, this less toxic wheat could be used in conjunction with the oral enzyme supplements and polymeric binders. Also unlikely is that these special varieties of wheat could be a financially sound alternative to industrial wheat.

Oral Enzyme Supplements

Gluten is rich in glutamine and proline residues, the latter of which plays a crucial role in directing tTG-mediated deamidation of the glutamine residues and thereby increasing their affinity to bind to HLA DQ2 and DQ8. These residues are