

Cooking with gluten: an exploration into the immune system and Celiac disease

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Purpose: Today we will learn about the immune response to food and how this results from eating gluten. As a hands-on project we will make both gluten-free and gluten-containing brownies. This will also allow us to talk about experimental design and the scientific method. After baking we will use an immuno assay kit to test for the presence of gluten in both brownie types.

Experimental group: Gluten-Free Brownie Recipe

- 1 1/2 cups sugar; [Baker's Special Sugar](#) or superfine sugar, if you have it
 - 1/2 cup butter
 - 1/2 teaspoon salt
 - 1 teaspoon gluten-free vanilla extract
 - 3/4 cup (2 1/2 ounces) Dutch-process cocoa or baking cocoa; we prefer the flavor of Dutch-process ("European-style")
 - 3 large eggs
 - 3/4 cup King Arthur [Gluten-Free Multi-Purpose Flour](#) or brown rice flour blend*
 - 1 teaspoon baking powder
 - 1 cup chocolate chips, optional
 - 1 cup chopped nuts, optional
 - *See recipe for this blend below.
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Instructions

1. Preheat the oven to 350°F. Grease an 8" square pan or 9" round pan; either should be at least 2" deep.
2. Place the sugar, butter, and salt in a microwave-safe bowl or saucepan. Heat over medium heat, stirring with a heatproof spatula until the butter melts and the mixture lightens in color. This step helps melt the sugar, which will give the brownies a shiny crust.
3. If you've heated the sugar and butter in a saucepan, transfer the mixture to a bowl; otherwise, just leave the hot ingredients right in their microwave-safe bowl. Blend in the vanilla and cocoa, then add the eggs and mix until shiny.

4. Blend in the flour or flour blend and the baking powder. Stir in the chips and/or nuts, if you're using them.
5. Pour the batter into the prepared pan, spreading it to the edges.
6. Bake the brownies for 33 to 38 minutes, until the top is set; and a cake tester or toothpick inserted in the center comes out clean or nearly so, with perhaps a few wet crumbs, or a tiny touch of chocolate at the tip of the tester.
7. Remove from the oven and cool for about 15 minutes before cutting. Once the brownies are cool, cover tightly with plastic.
8. Yield: 16 brownies.

Control Group: Gluten-Containing Brownie Recipe

- 1/2 cup butter
- 1 cup white sugar
- 2 eggs
- 1 teaspoon vanilla extract
- 1/3 cup unsweetened cocoa powder
- 1/2 cup all-purpose flour
- 1/4 teaspoon salt
- 1/4 teaspoon baking powder

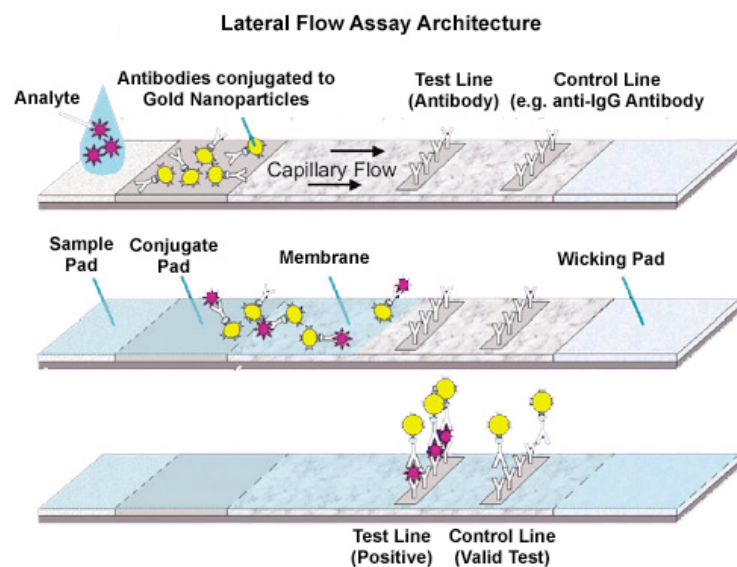
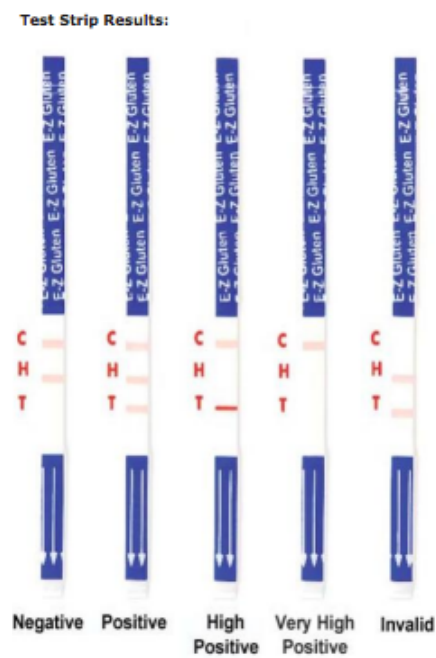
1. Preheat oven to 350 degrees F (175 degrees C). Grease and flour an 8-inch square pan.
2. In a large saucepan, melt 1/2 cup butter. Remove from heat, and stir in sugar, eggs, and 1 teaspoon vanilla. Beat in 1/3 cup cocoa, 1/2 cup flour, salt, and baking powder. Spread batter into prepared pan.
3. Bake in preheated oven for 25 to 30 minutes. Do not overcook.
4. To Make Frosting: Combine 3 tablespoons softened butter, 3 tablespoons cocoa, honey, 1 teaspoon vanilla extract, and 1 cup confectioners' sugar. Stir until smooth. Frost brownies while they are still warm.

Time for some Science: Testing for Gluten

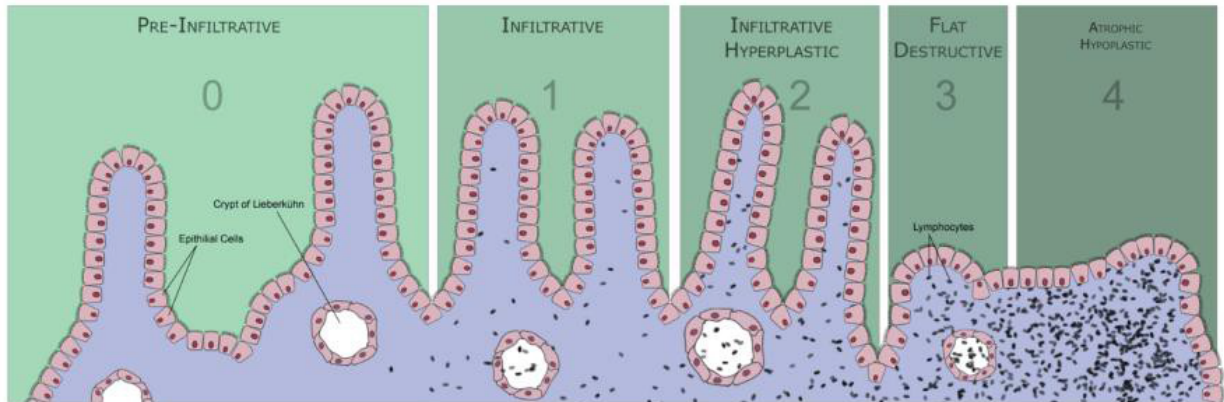
This test is called an immuno assay. It used antibodies bound to a pad to 'catch' passing gluten as it is pulled up the strip due to capillary action. Then the bound gluten is detected and a line appears on the pad accordingly.

EZ Gluten Protocol:

1. Measure out 1 level spoonful of your sample using the measuring spoon provided.
2. Add the sample to the vial containing extraction solution.
3. Cap the vial of extraction solution and mix by inversion for 2 minutes.
4. Set the vial vertically to let the contents settle for 5 minutes.
5. Transfer 10 drops of supernatants into the test tube.
*Use the cap to hold the test tube
6. Using the test strip from the silver foil pouch, insert it into the test tube with arrows pointing down.
7. Leave the test strip in the test tube for at least **10 minutes**.
8. Remove the strip and hold it alongside the picture to read the result.
*For best resolution, lay and dry the strip for 5 minutes before reading results.



UPPER JEJUNAL MUCOSAL IMMUNOPATHOLOGY



Shag carpet vs. linoleum tile

Scientific Method

Hypothesis: _____

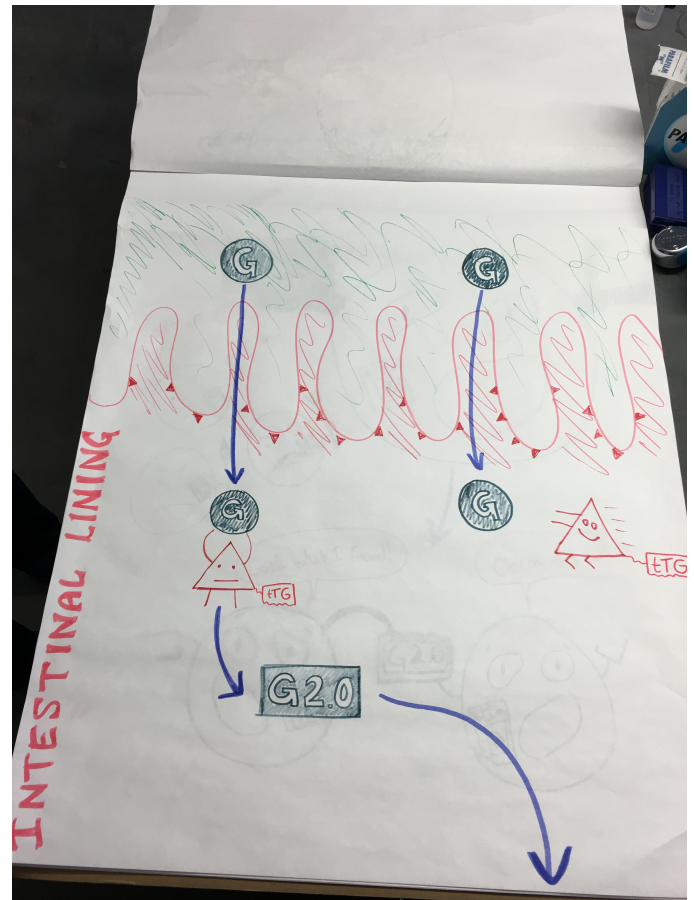
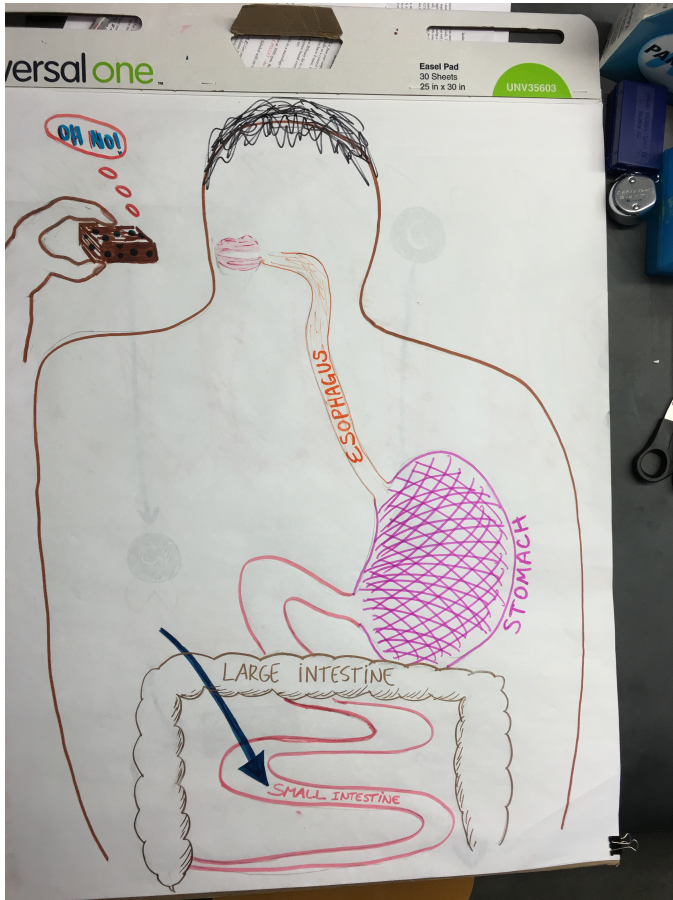
Experimental group: _____

Control group: _____

Results (observations): _____

Conclusions (thoughts): _____

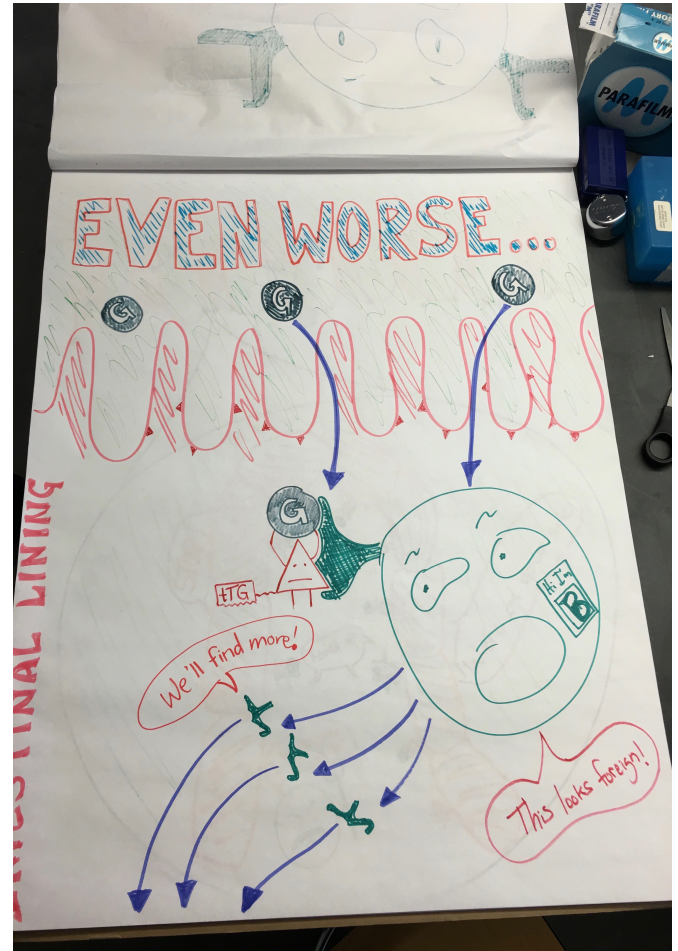
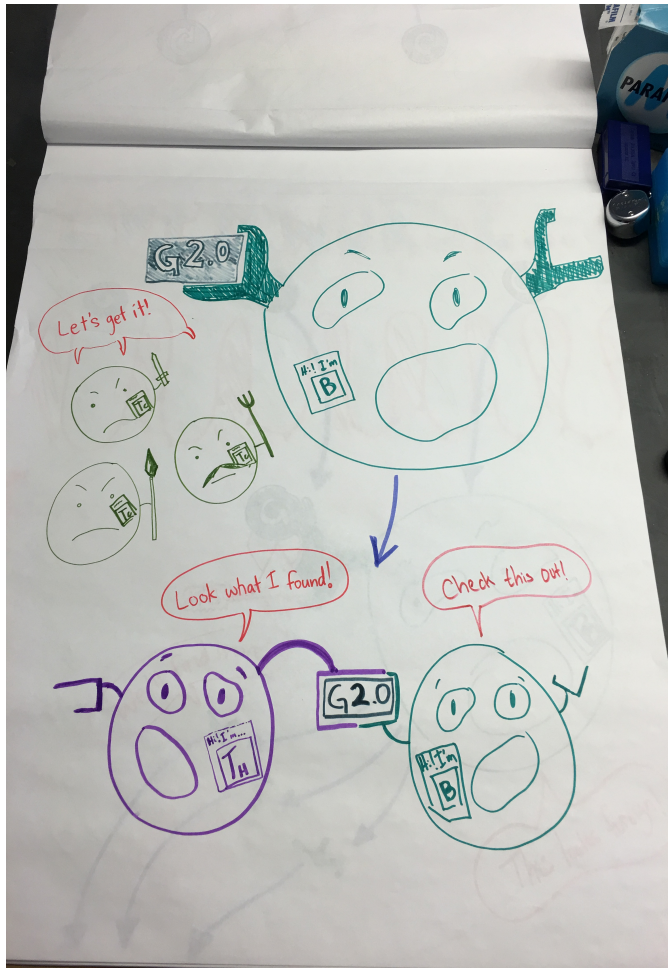
A Brief Explanation of the Immune Response to Gluten



When we eat brownies, they travel down our esophagus, into our stomach and, later, into our small intestine. Do you know anyone who can't eat gluten?

Enzymes in our gut cells (the wavy shag carpet cells), convert a special amino acid (building block) of gluten into a different form (let's call it gluten 2.0). Sometimes, this enzyme, tTG, (Mr. Triangle here) gets stuck on the gluten 2.0 after it converts it.

In some cases there exists a specific "flavor" of arm (MHC) on the body's cells that detects this Gluten 2.0, and calls the body's police department (the T cells).

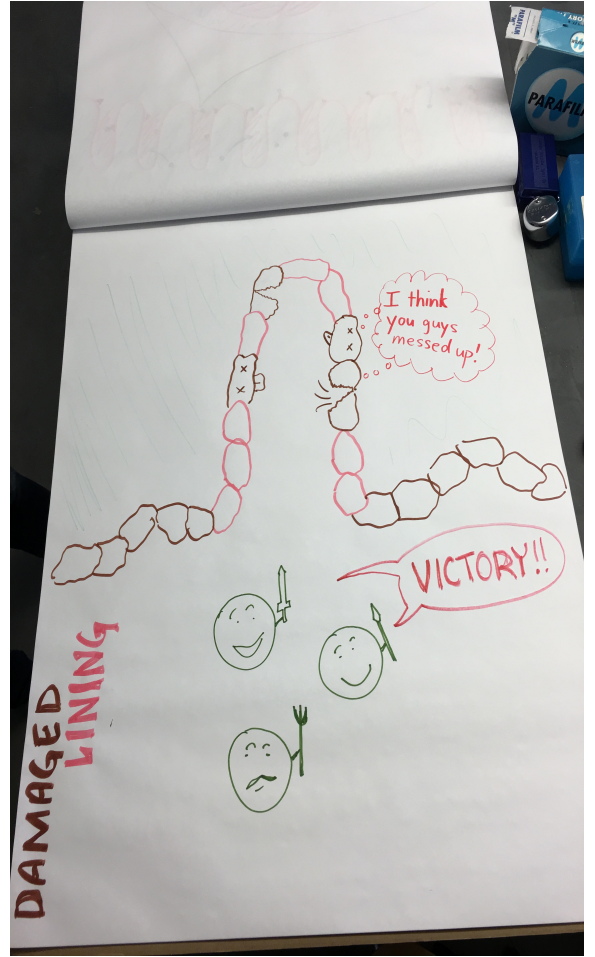


In an even worse scenario, the gluten bound to the tTg looks foreign to the B cell and it produces little 'magnets' to stick to the foreign looking complex. These 'magnets' are called antibodies and they go looking for more gluten-tTG.

If they can find any of the gluten-tTG complex, they bind and signal the soldiers of the immune system (Tc cells) to come kill any cell with tTG.

This is very bad for the body because *our intestinal cells* have tTG, and get killed. This ruins the shag carpet of our guts. This is detrimental to our health because it leads to poor absorption of nutrients (look at panel number 3 and 4 in the upper jejunal mucosal immunopathology figure).

Because the immune system targets its own cells, it is called an autoimmune disorder. Can you think of any other autoimmune disorders? _____



What are two different cell types we learned about today?

What do the two cell types do?

What can we do to make sure our friends with celiac disease don't eat gluten?