CASE TEACHING NOTES
for
"The Case of the 'Tainted' Taco Shells"

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INTRODUCTION / BACKGROUND

This case study discusses some of the issues associated with the use of genetically modified plants, including ecological risks, resistance, and allergenicity. This case was originally developed for use with a biochemistry course taught in the Chemistry Department at Wabash College. It would also be appropriate for use in a non-majors general, organic, or biochemistry course as well as a general biology course.

The case is presented in two editions. The General Edition is appropriate for non-majors general, organic, and biochemistry (GOB), general biology, or cellular biology courses, and emphasizes the basic biochemistry and scientific ethics issues associated with genetically modified foods. It could also be used at the beginning of a biochemistry course. The Advanced Edition probes the same issues discussed in the General Edition in further detail, and is appropriate for an upper level biochemistry, cell biology, or genetics class.

Both the General and Advanced Editions use the case study as a launching point to read and discuss primary and secondary literature articles. The General Edition set of papers and questions is appropriate for use with non-science majors. The articles require little, if any, prior experience with scientific literature and use very basic methods. All of the articles are available on the Internet (see references for links). The General Edition of the case is appropriate for liberal arts schools, as it touches on many of the ethical issues associated with science, including making decisions in the absence of complete information, balancing the needs of the few versus the many, and economics versus health issues. The Advanced Edition includes more advanced questions regarding the biochemistry of how genetically modified plants "work."

Prior Student Knowledge

Edition	*Pre- or co-requisite concepts*
General	• Central dogma of molecular biology • Gel electrophoresis • Enzymes
Advanced	• Transcription and translation • Restriction enzymes • PCR • Protein structure analysis tools* • Enzyme kinetics and inhibition*

*The depth of this prerequisite knowledge can be altered by changing the questions and/or selected articles.

Objectives

After completing this case study, all students should:

- Understand how a transgenic plant is made.
- Feel comfortable reading a primary literature article (at an appropriate level for the class).
- Understand the issues surrounding the use of genetically modified foods, including environmental effects and concerns about resistance.
- See how scientists have to deal with uncertainty and risk-assessment.
- Understand why Roundup is toxic to plants but not to animals.
- Understand the methods used to predict allergenicity.

Additionally, advanced students should:

- Understand the biochemical mechanism of *Bt* toxicity towards insects.
- Classify Roundup's inhibitor type based upon structural and kinetic data.
- Identify the mutations that could lead to development of Roundup resistant weeds.
- Understand and use the methods to predict allergenicity.

CLASSROOM MANAGEMENT

I use this case study during the DNA unit at the end of the semester in conjunction with a laboratory experiment where we test corn meal samples for the presence of genetic modifications (submitted to *Journal of Chemical Education*, July 21, 2003). The pairing of the case study with a laboratory activity has worked very well, as it provides a context for the laboratory activity and allows students to productively use the "waiting" times in the laboratory. This approach is also an easy way to add a case to a course without having to take time out of the traditional lecture period.

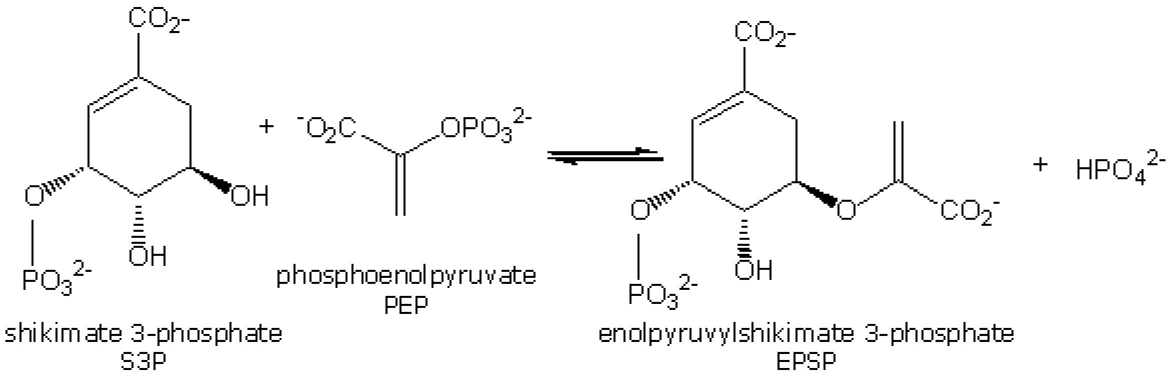
I have also used the case at the beginning of a biochemistry course by using the general set of papers, then discussing the advanced papers as the various topics they deal with were covered in class (e.g., the glyphosate inhibition of EPSPS during enzyme kinetics—see sample small group exercise at the end of this section). Alternatively, one could return to the case several times throughout the semester, either using the full paper assignments or in-class exercises based on the papers.

When the case is initially handed out, the situation is role-played by several students who take the parts of the various characters in the case scenario, and then the questions at the end of the story (except for the flippant one) are discussed. Students can review what they already know about genetic modifications in answering the first question. The safety question can be followed up with questions such as, "What do we mean by safe? Safe for whom?" This will lead to the environmental issues discussed in the papers. The third question ties in the economic issues discussed in the USDA bulletin, as well as the long-term possibilities of resistance. The groups then work on understanding their assigned primary literature papers. During the discussion period, each group presents their results and then the entire group returns to the initial questions, as well as extensions such as:

- Why do different scientists obtain different answers to questions such as "are genetically modified foods safe?"
- What is an acceptable level of risk? Is it okay to not know the long term outcome?
- People sometimes complain that scientists tell them one thing this week, and the opposite the next week. Why does this happen? How should consumers deal with this ambiguity?

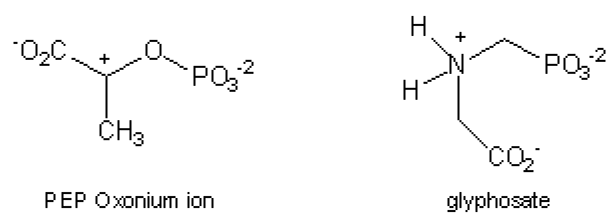
Sample Small Group Exercise Used When Integrating Biochemistry Level Papers in Class

The herbicide Round-Up, also known as glyphosate, works by inhibiting 5-enolpyruvylshikimate 3-phosphate synthase, or EPSPS for short. EPSPS catalyzes the following reaction:



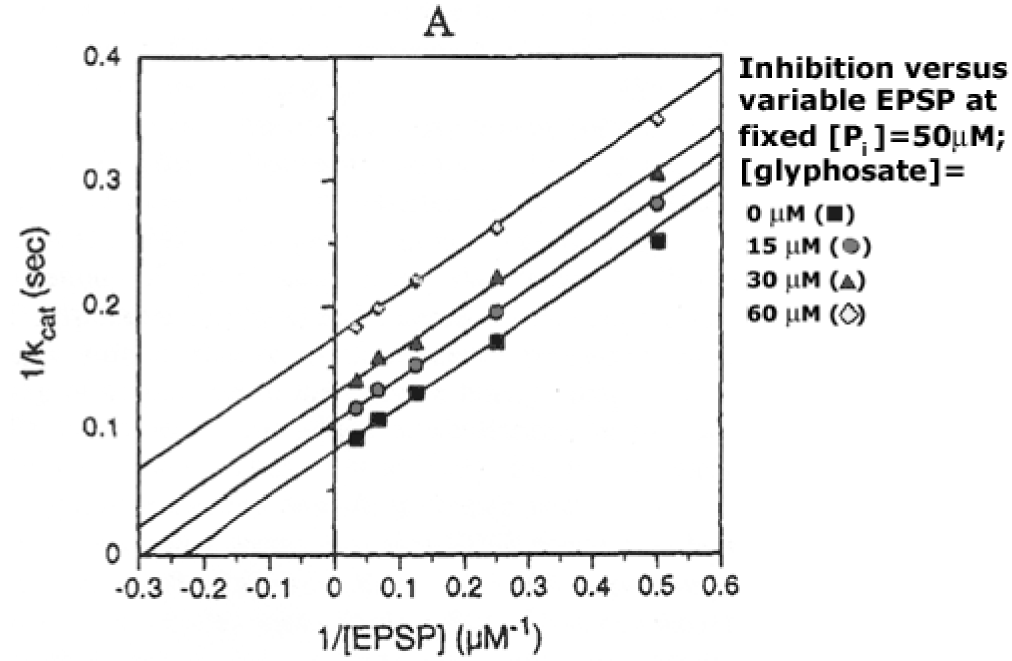
1. The crystal structures of EPSPS have been determined with and without S3P, and have PDB codes of 1G6T and 1EPS, respectively. Use Protein Explorer to look at these structures (http://www.proteinexplorer.org). Does EPSPS follow the "lock and key" or "induced fit" model of enzyme-substrate binding? Explain.

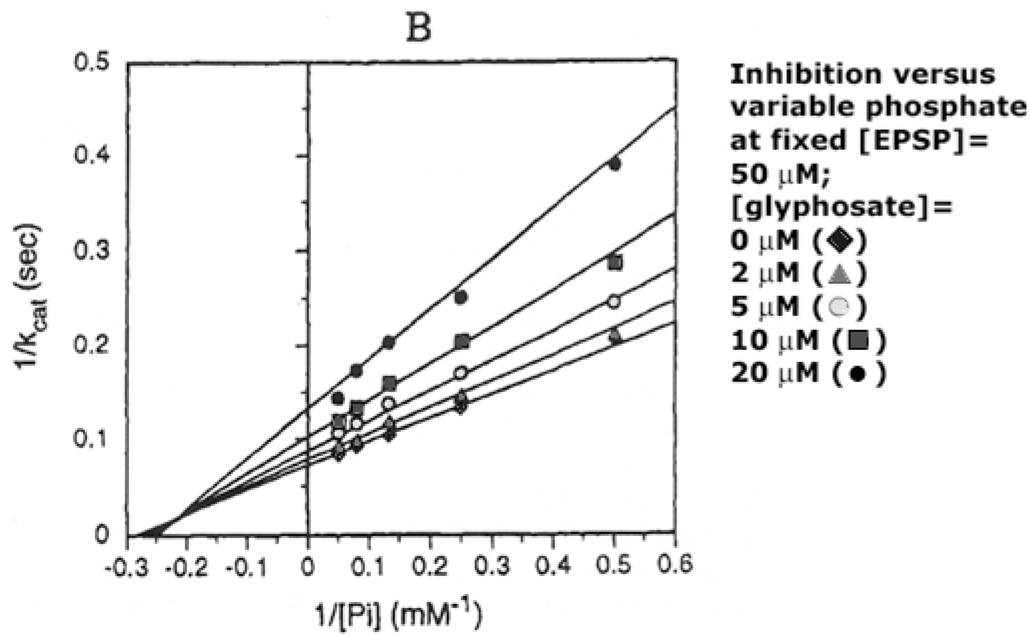
2. It is believed that the mechanism includes the formation of a PEP oxonium ion, shown below. Glyphosate is believed to be a good inhibitor because it resembles the transition state of the reaction. Use structural arguments to support this statement, and predict what kind of inhibitor you expect glyphosate to be (competitive or uncompetitive).



3. Sammons *et al.* (*Biochemistry*, vol. 34, pp. 6433–40, 1995) investigated the ability of glyphosphate to catalyze the reverse reaction. In one set of conditions (Figure A below), EPSP was varied while phosphate was constant; in Figure B, phosphate was varied while EPSP was constant. What kind of inhibitor is glyphosate in relation to EPSP? In relation to phosphate? Explain.

Figures A and B display Lineweaver-Burke plots for inhibition of *E. coli* EPSP synthase by glyphosate. Experiments were performed in a 1-cm path length rectangular cell for variable phosphate (total volume = 1.0 mL) and a 10-cm path length cylindrical cell for variable EPSP (total volume = 4 mL). (Figures adapted from Sammons *et al.*, 1995, copyright © 1995 American Chemical Society. Used with permission.)





BLOCKS OF ANALYSIS

The biggest challenge facing farmers is controlling weeds and diseases while maintaining crop yield and quality. It is important that these objectives are met cheaply, as the average acre yields 128 bushels of corn and the average price of corn has been $2.30/bushel over the last 10 years (USDA 2003). That $295/acre needs to pay for the cost of seed, equipment (a new combine costs $100,000), fertilizer, weed control, and the labor of the farmers (salary for the year!). Considering an acre is 4,840 square yards, the most practical methods for achieving good weed and disease control, crop yield, and quality are mass-methods. Historically, this has meant the widespread use of pesticides and herbicides to control unwanted insects and weeds. A new approach to these issues is through the production of genetically modified plants. In fact, 75% of soybeans and 34% of corn planted in 2002 were genetically modified (USDA 2002).

There are two common types of genetically modified crops: insect resistant (*Bt*) and herbicide resistant (Round-Up Ready). "*Bt*" is short for *Bacillus thuringiensis*, a soil bacterium whose spores contain a protein (Cry), which is broken down in an insect's gut to release a toxin, known as a delta-endotoxin. This toxin binds to and creates pores in the intestinal lining, resulting in an ion imbalance, paralysis and after a few days, death of the insect (Colordao State University n.d.). Several different Cry genes, also known as "*Bt* genes," have been identified. They are effective against different orders of insects and affect the insect gut in slightly different ways. The most common herbicide resistance products are the "Round-Up Ready" (RR) plants produced by Monsanto. Roundup® is the commercial name for glyphosate, an inhibitor of an enzyme required for amino acid synthesis in plants. RR plants contain the bacterial version of this enzyme, which is not affected by glyphosate.

To make a genetically modified plant, several components are needed in addition to the gene for the protein of interest. First, a method for getting the gene into the plant is needed. Most commonly a plasmid from *Agrobacterium tumefaciens* called Ti is used. Alternatively, a "gene gun" can be used. Once the gene is in the plant, it must be correctly transcribed and translated. This requires a promoter sequence and species-specific codon usage. Usually a powerful promoter from Cauliflower Mosaic Virus (CaMV 35 S) is used.

There are several environmental questions about the use of transgenic foods. In the case of *Bt* plants, there is concern about the effect of the toxin on non-target organisms such as butterflies via pollen. There is also the possibility that insects will become resistant to the *Bt* toxins. Resistance is also an issue with the Round-Up Ready plants; as glyphosate is repeatedly sprayed on fields, weeds may evolve to not be sensitive to the herbicide. This is already the case with horseweed in the United States and goosegrass in Malaysia.

There are also concerns in regard to human health. Although the human digestive system does not contain the receptors for the *Bt* toxins nor the conditions for proteolysis into the active toxin form, these proteins are not normally found in the human diet. Consequently, there is concern that *Bt* toxins might be allergens. The genes which have been approved for use in human food products denature under high heat and are easily digested by gastric enzymes, so there is relative confidence that they will not cause food allergies. Recently, Taco Bell brand taco shells were removed from the market for containing an unapproved Cry protein, Cry9C. This protein is heat stable and resistant to degradation by trypsin, two characteristics of potential food allergens (Hileman 2003). However, these characteristics do not guarantee allergenicity, so sequence-based methods are also being developed.

ANSWER KEY

Answers to the questions posed in the case study are provided in a separate answer key to the case. Those answers are password-protected. To access the answers for this case, go to the key. You will be prompted for a username and password. If you have not yet registered with us, you can see whether you are eligible for an account by reviewing our password policy and then apply online or write to answerkey@sciencecases.org.

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