

Genetically Engineered Organisms

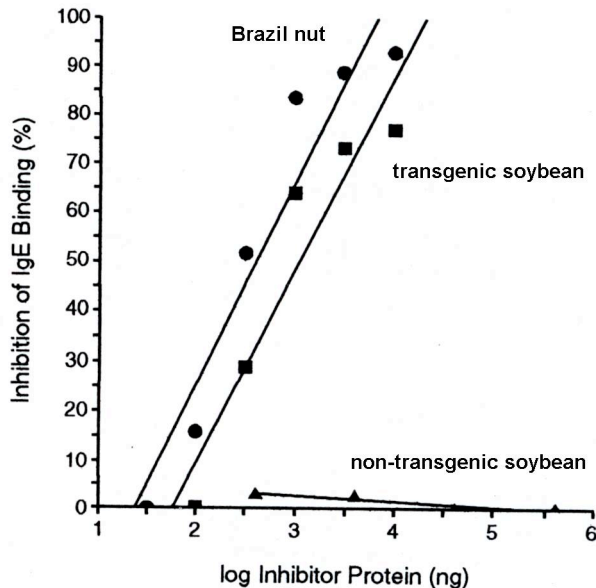


Figure 1. Results of Radioallergosorbent Assays with Extracts of Non-transgenic Soybean (triangle), Transgenic Soybean (square), and Brazil Nut (circle). Concentration of 2S albumin (in nanograms, log scale) in each dilution of extract in relation to percent inhibition of IgE binding. Read the x-axis as "Concentration of substance" and read the y-axis as "Potential allergic response via RAST test." (From Nordlee et al. 1996. Identification of a Brazil-nut allergen in transgenic soybeans. *The New England Journal of Medicine* **334**: 688-692. Copyright © 1996, Massachusetts Medical Society. All rights reserved.)

Soybeans are nutritional superstars by providing essential dietary amino and fatty acids. In addition, consumption of soy products have been shown to reduce cholesterol levels, and reduce risks of kidney and heart disease, osteoporosis, and possibly some cancers. However, soybeans are not a "complete" protein source for people and animals since soybeans lack the essential amino acid methionine ("essential" here means that animals cannot create methionine themselves). To rectify this deficiency, plant biotechnologists used methods of recombinant-DNA technology to insert a gene into a strain of soybeans to enable them to synthesize this missing amino acid.

The problem with this approach was that the original source of the gene for methionine, "2S albumin," came from Brazil nuts (*Bertholletia excelsa*). Many people are allergic to Brazil nuts and their reaction can range from severe rashes to anaphylactic shock. The study we focus on here by Nordlee et al. (1966) addresses this issue.

The purpose of the study by Nordlee et al. (1996) was to determine the extent to which the transgenic soybeans, containing the Brazil nut 2S albumin gene, caused allergic reactions similar to Brazil nuts.

Figure 1 is based on data from a radioallergosorbent test (RAST) in which a sample of blood from a potentially allergic person is checked for allergic sensitivity to specific substances. The approach used by the RAST is to determine whether specific IgE antibodies in serum drawn from sensitized subjects are able to recognize the protein of interest, in this case the 2S albumin from Brazil nuts.

Discuss the following:

Start by taking 5 minutes to interpret the Figure. Run through your ten steps for yourself; you may wish to provide a short list of answers to those questions to help you answer the questions below.

Discuss answers to the following:

1. What were the main research questions the scientists were asking?
2. What was the benefit of conducting this test to determine the allergenicity of transgenic soybeans?
3. Contrast the slopes of the 3 treatments; what conclusions can you draw about the relative allergenicity of the transgenic soybean?
4. Did Pioneer Hi-Bred International (Dupont) make the right decision to abandon this research program? What were some alternatives?
5. The ethical question of whether transgenic foods should be labeled is hotly debated. Can you assess the value or importance of this experiment within the context of bioethics? Should governments require labeling of genetically engineered foods?
6. According to statements made by FDA Commissioner Dr. J. D. Henney (Thompson 2000, see [Background to Figure Set 1](#)) "we have no scientific evidence to indicate that any of the new proteins introduced into food by biotechnology will cause allergies." Based on your understanding of Nordlee et al. (1996), is Dr. Henney correct? Explain your response.

Glossary of terms

Allergy- overreaction of the immune system to specific substances called allergens (such as pollen or bee stings) that in most people result no symptoms. Allergies often involve IgE (one of the 5 types of immunoglobulins produced by humans) antibodies.

Allergen- an antigen that produces allergic reactions by inducing formation of IgE.

Antibody- proteins, produced by the immune system, that recognize a foreign substance and starts a process of removal of the foreign material from the body.

Antigen- a substance that stimulates the production of an antibody (see Allergen)

Immune system- a system in mammals that recognizes and then eliminates or neutralizes foreign substances.

Immunoglobulin- a group of proteins active in the immune system that serve as antibodies. They work by binding to foreign antigens.

BIO 103 Transgenics activity.

Work together in groups of four. Pair with one other person in the group. In each pair, one person should work on the problem while the other coaches. Choose who will have each role for the first two figures then switch roles for the two figures

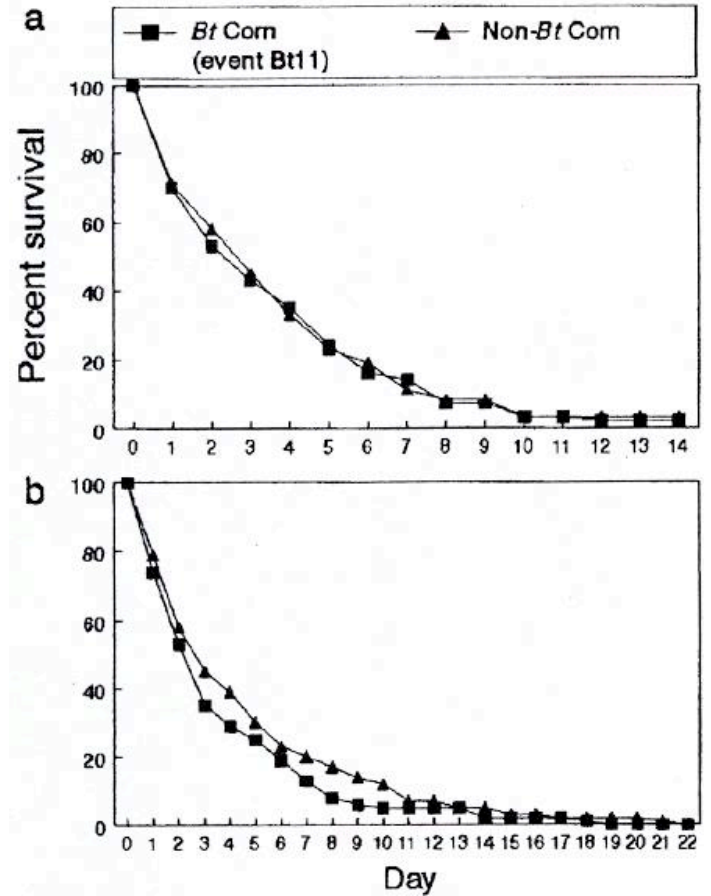
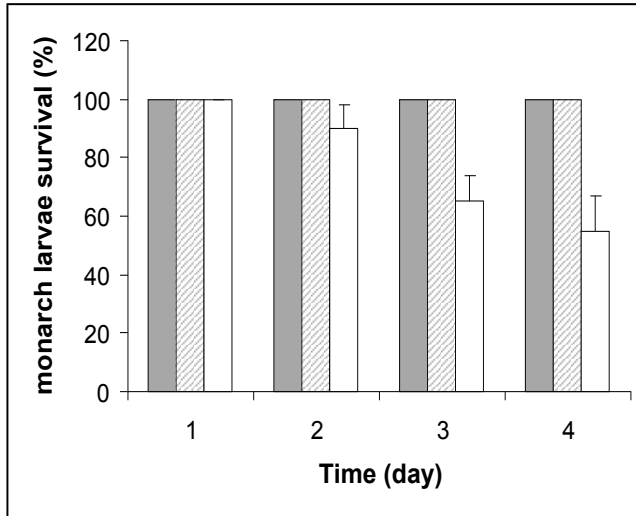


Figure 2a. The survival of second to third-instar monarch larvae was tested. Three milkweed leaf treatments were conducted: leaves with no pollen (grey), leaves treated with untransformed corn pollen (hatched), and leaves dusted with pollen from Bt corn (white). The mean survival rate is based on the proportion of larvae surviving in five replicates of each treatment (from Losey, H. E., L. S. Rayor, and M. E. Carter. 1999. Transgenic pollen harms monarch larvae. *Nature* 399: 214. © 1999 Nature Publishing Group.)

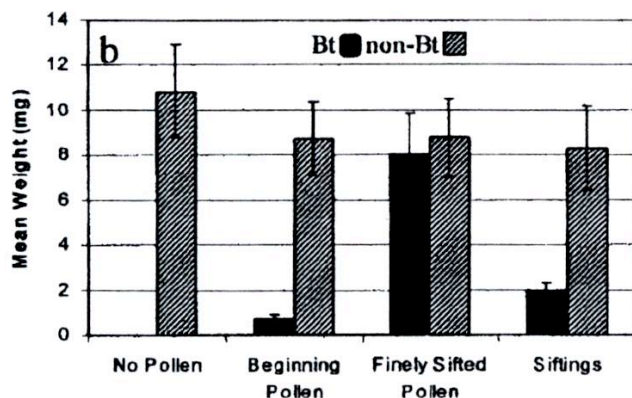
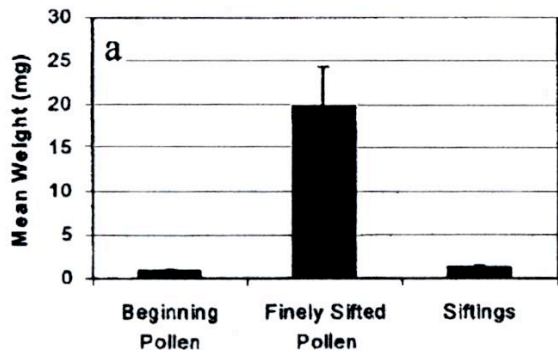


Figure 2b. Survival curves for monarch larvae placed in and near Bt and non-Bt corn fields. Survival curve (a) is based on data from Iowa and survival curve (b) is based on data from New York (from Stanley-Horn, D. E. et al. 2001. Assessing the impact of Cry1Ab-expressing corn pollen on monarch butterfly larvae in field studies. *Proceedings of the National Academy of Sciences* 98: 11931-11936, © 2001 National Academy of Sciences, U.S.A.)

Figure 2c. These graphs depict the mean weight of monarch larvae after feeding on treated milkweed for 96 hours. Treatments included: (a) pollen from hybrid G8539Bt that was processed with 250- μ m sieve (beginning), pollen from same hybrid processed with 90- μ m sieve (finely sifted), and siftings remaining after the initial sample was finely sifted; and (b) no pollen, pollen from hybrid N7070 with Bt inserted, and pollen from hybrid N7070 (non-Bt that were processed in the same manner as pollen in a) Hellmich, R. L. 2001. Monarch larvae sensitivity to *Bacillus thuringiensis* - purified proteins and pollen. *Proceedings of the National Academy of Sciences* early edition: 1-6, © 2001 National Academy of Sciences, U.S.A.)

First, examine Figure 2a. In this experiment, stems of milkweed leaves were put into tubes containing water. Five three-day-old monarch larvae (instar is a stage of development) were placed on each leaf.

In Figure 2a, compare larval survival after feeding on leaves with Bt pollen with those leaves dusted with untransformed pollen or the control leaves with no pollen.

Do you think the evidence indicates that differences in monarch larvae survival on leaves dusted with Bt pollen is due to the effects of Bt pollen?

Describe and interpret Figure 2b. Here the researchers sought to determine if there were significant differences in the numbers of larvae surviving in Bt corn fields and non-Bt corn fields in two different states, in order to analyze the toxicity of transgenic pollen on monarch larvae.

Does this new study discredit the original study? Explain why or why not.

Figure 2c is from an experiment conducted a couple years after the first study. It shows results of a contamination study. In this case researchers studied whether monarch larvae were affected by contaminants in the samples by comparing the varying levels of sifting of the pollen. Examine both figures.

How do larvae whose diet contained beginning pollen or siftings of the contaminated pollen compare with larvae that consumed finely sifted pollen (most contaminants were removed)?

What conclusion about the cause of the monarch larvae's reactions to the samples can you draw from this figure?

Does this new study discredit the original study? Explain why.

Is the overall implication of this study similar to that of Figure **2b**'s study?

What investigation(s) would you propose next?

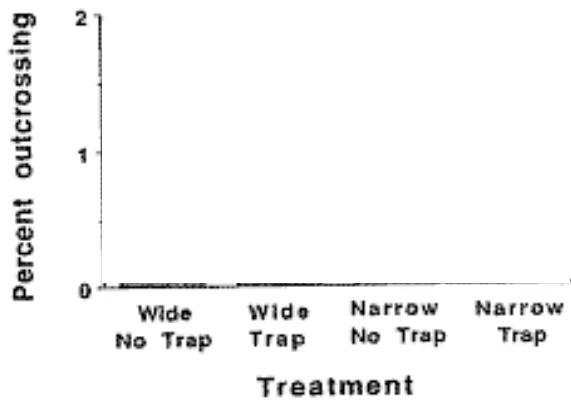
Check your answers with the other pair in your group.

We have discussed the allergenicity of genes engineered into crops and whether crops engineered to resist pests might also impact non-target insects. Ecologists are also concerned about cross-pollination of transgenic plants with closely related weeds; plants are well known to hybridize across species and often produce fertile offspring. If weeds can mate with genetically engineered crop plants, their offspring may inadvertently express traits carefully engineered into those crops and become widespread pest- or herbicide- resistant weeds. Agricultural scientists are hard at work trying to discover ways that this cross-pollination can be prevented.

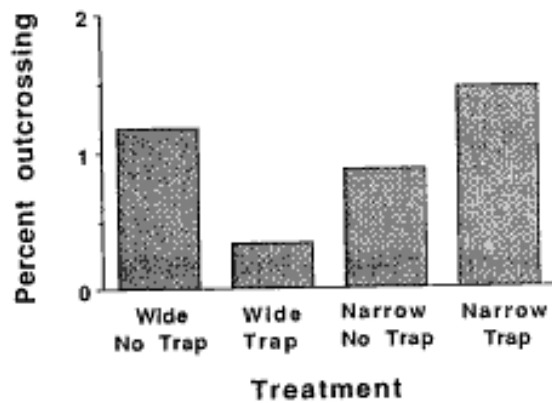
Morris and colleagues conducted an experiment to test the effectiveness of wide and narrow zones surrounding crop plants that are either free of plants (no trap) or planted with non-transgenic border plants (trap). They planted transgenic canola plants as the crop, and examined the frequency with which cross-pollinations occurred with closely related wild radish plants, strategically placed in a ring around the transgenic crop. Pollinators (bees, especially) were allowed to roam freely.

What is the dependent variable?
 What is the independent variable?

Draw what the graph would look like if barren zones were most effective in preventing the escape of transgenic pollen and increasing zone width improves effectiveness of pollen transfer prevention.



The effect of isolation zone width (narrow= 4m, wide = 8m) and of trap beds on the overall rate of gene escape from the transgenic canola plot. From Morris et. al.1994. Do barren zones and pollen traps reduce gene escape from transgenic crops? Ecological Applications 4: 157-165.



Which seems to be the most effective strategy for reducing the escape of transgenic pollen?

If there were statistically significant differences among our treatments, what could you say about the value of *P* in a statistical test comparing percent outcrossing among the treatments?

Extra credit: How might these results differ if we were using wind-pollinated corn, instead of insect-pollinated canola?