

# Modified mushrooms may yield human drugs

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Mushrooms might serve as biofactories for the production of various beneficial human drugs, according to plant pathologists who have inserted new genes into mushrooms.

"There has always been a recognized potential of the mushroom as being a choice platform for the mass production of commercially valuable proteins," said Charles Peter Romaine, who holds the John B. Swayne Chair in spawn science and professor of plant pathology at Penn State. "Mushrooms could make the ideal vehicle for the manufacture of biopharmaceuticals to treat a broad array of human illnesses. But nobody has been able to come up with a feasible way of doing that."

Dr. Romaine and his colleague, Xi Chen, then a post-doctoral scholar at Penn State and now a Syngenta Biotechnology Inc. research scientist, have developed a technique to genetically modify *Agaricus bisporus* -- the button variety of mushroom, which is the predominant edible species worldwide. One application of their technology is the use of transgenic mushrooms as factories for producing therapeutic proteins, such as vaccines, monoclonal antibodies, and hormones like insulin, or commercial enzymes, such as cellulase for biofuels.

"Right now medical treatment exists for about 500 diseases and genetic disorders, but thanks to the human genome project, before long, new drugs will be available for thousands of other diseases," Dr. Romaine said. "We need a new way of mass-producing protein-based drugs, which is economical, safe, and fast. We believe mushrooms are going to be the platform of the future."

To create transgenic mushrooms, researchers attached a gene that confers resistance to hygromycin, an antibiotic, to circular pieces of bacterial DNA called plasmids, which have the ability to multiply within a bacterium known as *Agrobacterium*.

The hygromycin resistance gene is a marker gene to help sort out the transgenic mushroom cells from the non-transgenic cells, Dr. Romaine explained. "What we are doing is taking a gene, as for example a drug gene, that is not part of the mushroom, and camouflaging it with regulatory elements from a mushroom gene. We then patch these genetic elements in the plasmid and insert it back into the bacterium," he added.

The researchers then snipped small pieces off the mushroom's gill tissue and added it to a flask containing the altered bacterium.

Over the course of several days, as the bacterium goes through its lifecycle, it transfers a portion of its plasmid out of its cell right into the mushroom cell, and integrates the introduced gene into the chromosome of the mushroom.

Next, the researchers exposed the mushroom cells to hygromycin. The antibiotic kills all the normal cells, separating out those that have been genetically altered for resistance.

The test demonstrates that if a second gene, insulin for example, were to be patched in the plasmid, that gene would be expressed as well.

"There is a high probability that if the mushroom cell has the hygromycin resistance gene, it will also have the partner gene," Dr. Romaine added.

The degree of gene expression ultimately depends on where exactly the imported gene lands in the mushroom chromosome, among a complexity

of other factors, but researchers point out that the process of producing biopharmaceuticals is potentially faster and cheaper with mushrooms than conventional technologies. Unlike plants that have long growth cycles, "with mushrooms, we can use commercial technology to convert the vegetative tissue from mushroom strains stored in the freezer into vegetative seed. A crop from which drugs may be extracted could be ready in weeks," Dr. Romaine said. A mushroom-based biofactory also would not require expensive infrastructure set up by major drug companies, he added.

Source: Penn State

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