

COMMENTARY

When it comes to magic mushrooms, less is more

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Luminous Mind, Inc. Announces the Issuance of a U.S. Patent No. 12,697,362 for Non-Hallucinogenic Psychedelic Fungi



Psilocybe mushroom showing blue psilocin oxidation

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Psilocybin-producing fungi, commonly known as “magic mushrooms,” are a polyphyletic group of fungi that enzymatically synthesize and thus contain psilocybin,

which upon ingestion, is rapidly converted to its metabolite psilocin that is the compound responsible for the “psychedelic” effects of magic mushrooms. For most of human history, we have consumed these mushrooms as entheogens or medicines and cultivated or encouraged wild growth or those that produced the most psychoactive psilocybin or the brightest blue oxidized psilocin. Against this tradition, we hold that the specialized metabolism of *Psilocybe* mushrooms and their cousins provides dozens of biologically active molecules apart from the two hallucinogens in whole live mushrooms and in extracts. Further, many of the amazing compounds that would be consumed in fungal hallucinogen production, instead become available to increase valuable and novel bioactive compounds in the edited mushroom. We offer a workbench of innovative methods, gene editing and gene silencing techniques that leave natural genetic variants of endogenous sequences from the same species.

Motivation

Our invention makes it possible to produce a stable fertile and true breeding mushroom (sporocarp and monokaryon and dikaryon mycelia) of *Psilocybe cubensis* by genome editing without producing either of the hallucinogenic compounds psilocybin or psilocin. This paves the way for a nutraceutical product that can potentially be a large market opportunity - analogous to CBD oil - which does not contain significant THC. We also anticipate widespread use of non-hallucinogenic mushroom products in functional foods, nootropics, legal microdoses and therapeutics.

We are most excited to use edited mushrooms to enable the genetic and biochemical analysis of specialized fungal metabolism of unique natural drug-like molecules. For example, the remaining enzymes of the inactivated psilocin pathway generate abundant indoleamine derivatives including 4-hydroxytryptophan, 4-hydroxytryptamine, beta-carbolines such as harmaline and their metabolic products. All mycelial and mushroom metabolites may be targeted by our genetic workbench approaches starting with these indoleamine derivatives. In this way, we can create a sustainable and medically appropriate source of mushroom-derived compounds that can be tested for their safety and biological activity for incorporation into cosmetics,

nutraceuticals or food supplements with no hallucinogenic potential at all.

Different people react very differently to hallucinogenic compounds of "magic" mushrooms owing to their personal metabolism, size and physical and mental health state. While purified psilocybin and psilocin can be obtained and doses calculated by indication, weight or surface area of the consumer and experience of the therapist, the equivalent dose of whole mushroom depends on growth conditions, storage, variety and extraction (for example tea). Notably, where psilocybin and psilocin are sanctioned to be used for therapy, many therapists and consumers prefer to use whole mushroom products rather than isolated active compounds. Standardizing the psilocybin content of whole mushroom extracts is both a therapeutic and a regulatory necessity. Indeed, published and community evidence indicates that a defined ensemble of bioactive molecules should be available in clinical trials for safety and efficacy. Our edited mushroom strains are eminently suitable to optimize and calibrate these whole mushroom metabolites for combination with purified therapeutic psilocybin or psilocin.

Non-GMO

At this time, cleanly gene-edited fungi, in which no foreign nucleic acids or transgenes are present, are not a regulated article according to the U.S. Department of Agriculture (USDA), for example non-browning strains of the white button mushroom *Agaricus bisporus*. Thus, among the advantages of our invention we anticipate provision of non-hallucinogenic psychedelic fungi that are not regulated as genetically engineered (GE) organisms or as genetically modified organisms (GMOs). While we wholeheartedly support and gratefully acknowledge the widespread use of transgenic fungi including yeasts to generate pharmaceuticals and innovative and sustainable sources of flavors and foods, we also believe there is a need to offer craft mushroom products with disclosure of a full genome sequence compatible with the needs and ethics of the most dedicated mycophile. When we use genome editing techniques to create novelty and diversity, we have a responsibility that it be compatible with the range of available natural genetic variation, mobile element propagation, epigenomics, natural genome editing processes and interspecific hybridization mechanisms.

Our technology is extensible to other strains and species

Many hallucinogenic mushrooms have a long history of human coevolution as entheogens and foraged medicines and some have traveled in a literal diaspora out of Africa with humans and prospered worldwide, fertilized by domestic cattle. We recognize the work of mycophiles and psychedelic pioneers who have curated over 200 distinctive *Psilocybe cubensis* strain names for genetic variants. Names like Moby Dick, Golden Teacher and South African Transkei refer to traits like albino pigmentation, helmet-like cap, rusty spore color, or to the region of discovery. This diversity is real: the *P. cubensis* species has several rather different genomes reflecting several regions of origin, but only a few visible shape and color markers caused by single natural or induced mutations that can occur on any genome background. Our methods can also be used on most of the over 100 *Psilocybe* species found worldwide. We have found 11 other genera of basidiomycete fungi that have related genes for a psilocin synthetic pathway and the methods we detail for inactivating and modulating the levels of the enzymes they encode can be broadly applied.

Complete gene editing in a mycelial rather than cellular system

Unlike most plant and animal cells, fungi can move nuclei of one, two or even more genomes around within their strands of mycelial hyphae. This presents an obstacle to most precise gene editing technologies since unmodified and differently modified genomes can offer conflicting instructions to the growth, development and metabolism of the resulting mycelium or mushroom. For this reason, some of our technical advances use periods of selection for and against specific gene alterations using selectable marker genes made from variants of the genome's own essential metabolic pathway genes. Once the desired edits have been established, these markers can be removed before growing a crop of mushrooms or filling a bioreactor with mycelium.

Editing regulatory elements that control gene function

Many of the gene edits we have envisaged achieve complete removal of a single enzyme by rendering a single gene incapable of encoding its respective protein. However, genes come with adjacent sequences responsible for expressing or silencing the gene in response to growth and differentiation of the fungus, in response to mating, symbiotic interaction with plants, damage by microbes, insects and nematodes, responding to nutrients, daylength and circadian rhythms, temperature and water. By editing these specific sequences, the dependency of gene expression upon the environmental stimulus is weakened or disconnected, allowing specific cultivation conditions to optimize the metabolism of the fungi for our uses.

From wild foraged to domesticated mushrooms

The best known and most widespread genetic mechanism in human domestication of crops such as rice is to break the dependence of flowering and seed production upon daylength. In contrast, apple genetics and cross-pollination for fruit production are still restricted by varieties that have compatible flowering times at a specific latitude. The intricate dependencies of most edible and medicinal fungi upon environmental cues makes cultivation or controlled growth of most species too complex for current agricultural practices. This complexity is one of the fascinations of wild mushrooms, and a strong message that we need to try harder to understand the needs of fungi and the ecosystems they interconnect. How a mushroom reacts to a genetic nudge could provide the first clues.

About Luminous Mind Inc.

Luminous Mind Inc., with its focus on Central Nervous System (CNS) disorders, applies a rigorous therapeutic discovery and development process that includes the repurposing of existing, abandoned, and developmental compounds for new indications. Groundbreaking scientific insights in neuroscience, genetics, systems biology, and brain circuitry, together with powerful new tools drive its discovery efforts. For additional information, please visit www.LuminousMindInc.com.

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