

***CORDYCEPS MILITARIS* MUSHROOM CULTIVATION**

WHAT IS CORDYCEPS MILITARIS?

Cordyceps militaris is an entomopathogenic fungus known for its numerous medicinal properties. It belongs to the same genus as the highly prized *Cordyceps sinensis*, but is more readily cultivated on an industrial scale. This fungus has a wide range of potential therapeutic applications. *Cordyceps militaris* is a rich source of various bioactive compounds that contribute to its medicinal properties. One of the most studied compounds is cordycepin, a nucleoside analog with potent anti-inflammatory, antioxidant, and anti-tumor activities. The fungus is also abundant in polysaccharides, such as galactomannan, which exhibit immunomodulatory, anti-tumor, and anti-fatigue effects.

Ergosterol, a sterol compound found in fungal cell membranes, and its derivatives present in *Cordyceps militaris* have been reported to possess antioxidant, anti-inflammatory, and anticancer properties. Additionally, the fungus contains adenosine, a nucleoside with anti-inflammatory, neuroprotective, and cardioprotective effects, as well as cordycepin triphosphate (cordycepin₃P), a derivative of cordycepin that induces apoptosis in cancer cells. Mannitol, a sugar alcohol found in *Cordyceps militaris*, is associated with antioxidant, anti-inflammatory, and potential benefits for diabetes management. The fungus is also a rich source of the antioxidant enzyme superoxide dismutase (SOD), which neutralizes harmful superoxide radicals and protects cells from oxidative stress. Moreover, it contains various phenolic compounds, such as gallic acid, catechin, and protocatechuic acid, contributing to its antioxidant and anti-inflammatory activities.

Carotenoids like β -carotene, lutein, and zeaxanthin, potent antioxidants with potential anti-inflammatory and anticancer properties, are also present in *Cordyceps militaris*. Additionally, the fungus contains various fatty acids, including oleic acid, linoleic acid, and palmitic acid, which play roles in cellular functions and may contribute to its therapeutic effects. This diverse array of bioactive compounds, along with others present in *Cordyceps militaris*, is believed to be responsible for the fungus's broad range of medicinal properties and potential therapeutic applications. However, the specific composition and concentrations of these compounds can vary depending on factors such as the strain, cultivation conditions, and extraction methods used.

IMPORTANCE OF CORDYCEPS MILITARIS CULTIVATION

Harvesting *Cordyceps militaris* from its natural habitat can be challenging and unsustainable. Cultivation offers a reliable and eco-friendly way to produce this valuable fungus on a larger scale, ensuring a consistent supply for medicinal and research purposes. Additionally, cultivated strains can be optimized for desired traits, such as increased production of bioactive compounds. Some of the key medicinal properties of *Cordyceps militaris* are listed below,

1. Anti-inflammatory Effects:

Cordyceps militaris contains various bioactive compounds, such as cordycepin and polysaccharides, that have been shown to possess potent anti-inflammatory properties. These

compounds can inhibit the production of inflammatory mediators like nitric oxide, prostaglandins, and cytokines, thereby reducing inflammation in the body.

2. Antioxidant Activity:

The fungus is rich in antioxidants, including phenolic compounds, carotenoids, and superoxide dismutase (SOD). These antioxidants help neutralize harmful free radicals and protect cells from oxidative stress, which is linked to various chronic diseases and aging processes.

3. Immunomodulatory Effects:

Cordyceps militaris has been found to modulate the immune system by stimulating the activity of various immune cells, such as macrophages, natural killer cells, and T-cells. This immunomodulatory effect can enhance the body's defense mechanisms against pathogens and potentially support the treatment of autoimmune disorders.

4. Anti-tumor and Anti-cancer Properties:

Several studies have demonstrated the potential of *Cordyceps militaris* in inhibiting the growth and proliferation of various cancer cell lines, including lung, breast, and colon cancer cells. This anti-tumor activity is attributed to its ability to induce apoptosis (programmed cell death) in cancer cells and suppress angiogenesis (formation of new blood vessels that feed tumor growth).

5. Hypoglycemic and Anti-diabetic Effects:

Cordyceps militaris has been shown to have hypoglycemic (blood sugar-lowering) effects and may be beneficial for managing diabetes. It can improve insulin sensitivity, reduce oxidative stress in diabetic conditions, and inhibit the enzymes responsible for breaking down carbohydrates, thereby regulating blood sugar levels.

6. Liver and Kidney Protection:

The fungus exhibits protective effects against liver and kidney damage caused by various toxins or diseases. Its antioxidant and anti-inflammatory properties contribute to this protective action, helping to maintain the health and function of these vital organs.

7. Cardiovascular Benefits:

Cordyceps militaris may have beneficial effects on cardiovascular health by reducing cholesterol levels, improving blood circulation, and protecting against oxidative stress and inflammation in the cardiovascular system.

8. Neuroprotective and Anti-aging Effects:

Some studies suggest that *Cordyceps militaris* may have neuroprotective properties and could potentially help in managing age-related neurodegenerative diseases like Alzheimer's and

Parkinson's. It may also exhibit anti-aging effects by reducing oxidative stress and promoting longevity.

STANDARD OPERATING PROCEDURE

MATERIALS NEEDED:

1. *Cordyceps militaris* culture/liquid spawn
2. Substrate (usually a mixture of rice, grains, or other suitable materials)
3. Sterilization equipment (pressure cooker or autoclave)
4. Petriplate and Glass jars containers
5. pH meter and pH adjusting solutions
6. Incubation chamber or room
7. Laminar Air flow Chamber
8. Tray dryer
9. Humidity and temperature control equipment
10. Light source (for inducing fruiting)
11. Humidifier
12. AC
13. Shaker
14. Incubator
15. Refrigerator
16. Weighing Balance
17. Clean working area

PROCEDURE:

1. Preparation of Substrate:

- Prepare a suitable grain-based brown rice substrate
- Soak the substrate for 20 minutes in water

2. Sterilization:

- Fill the jar with substrate and media
- Cover the hole with micro filter tape for air circulation.
- Sterilize the substrate in an autoclave or pressure cooker at around 121°C (250°F) for 1-2 hours to kill competing microorganisms.
- Let the substrate cool down to room temperature before inoculation.

3. Inoculation and Culture Development:

- In a laminar flow hood, transfer *Cordyceps militaris* spores onto the prepared substrate using a sterile inoculation loop, needle, or pipette.
- Seal the jar.
- Incubate the cultures in a controlled environment at around 25-28°C (77-82°F) with high humidity (90-95%) and low light intensity for 2-4 weeks, depending on the growth rate.

4. Mycelium Expansion:

- Observe the growth of the mycelium and check for any signs of contamination.
- Once the mycelium has fully colonized the substrate, transfer a piece of the colonized mycelium to a fresh substrate in a larger container using a sterile scalpel or knife.

5. Transfer and Fruiting Initiation:

- The mycelium will reach its maximum growth potential
- Create a conducive environment for fruiting by decreasing the temperature slightly (around 18-22°C or 64-72°F) and increasing humidity to near saturation (95% or higher).
- Expose the culture to indirect light for 12 hours per day to induce photoperiodism.

6. Fruiting and Harvesting:

- Watch for the formation of fruiting bodies (ascocarps) from the mycelium. They will appear as small orange bumps that will gradually elongate and develop into caterpillar-like structures.

- Maintain high humidity and indirect light to encourage fruiting body development.
- Harvest mature fruiting bodies carefully by cutting them at the base with a sterile scissors or knife. They will be ready when they have reached their full size and color, usually after 2-3 weeks.

7. Spore Collection (Optional):

- If you want to collect spores for future use or to start new cultures, you can let some of the mature fruiting bodies release spores onto a clean surface, such as a piece of paper or glass.
- Collect spores by scraping them off with a sterile spatula or brush. Store them in a sealed container in a cool and dry place.

8. Cleaning and Maintenance:

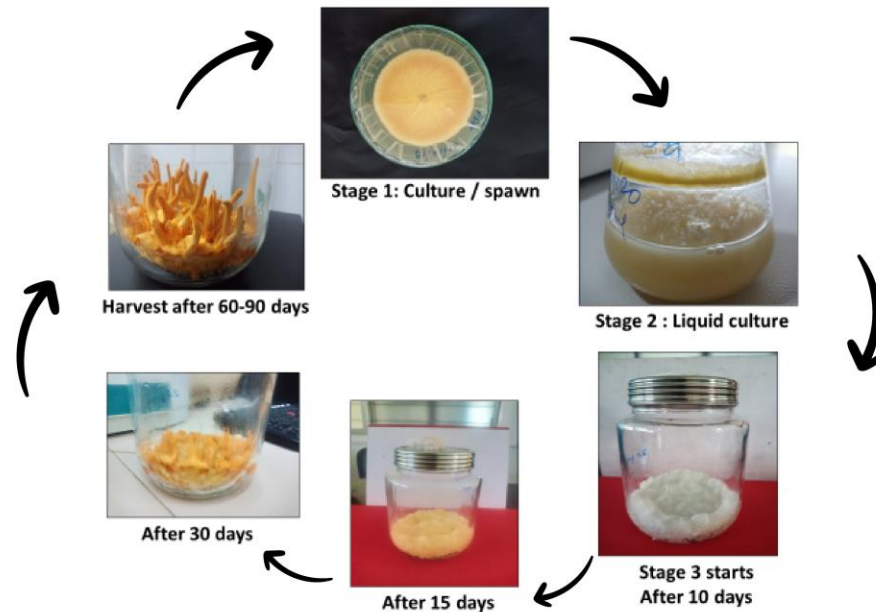
- Regularly clean the laboratory environment and equipment with disinfectants to prevent contamination.
- Maintain aseptic techniques when handling cultures and substrates.
- Dispose of used substrates and cultures properly according to safety regulations.

9. Scaling Up (Optional):

- If you want to scale up your production, you can use larger containers or growing systems, such as bags, trays, bottles, jars, or boxes.
- You can also experiment with different substrates, temperatures, humidities, light intensities, and inoculation methods to optimize your yield and quality.

Remember that *Cordyceps militaris* cultivation can be complex and may require optimization based on your specific laboratory conditions and available resources.

CORDYCEPS MILITARIS PROJECT MODEL



MARKETING STRATEGIES FOR *CORDYCEPS MILITARIS* MUSHROOM CULTIVATION

- Educate trainees on the health benefits of cordyceps and how to effectively communicate these to customers since the medicinal qualities are a key selling point.
- Assist trainees in identifying target buyers such as health food stores, herbal supplement companies, traditional medicine practitioners, and pharmacies. This encourages partnerships that aggregate supply to meet the higher volume needs of wholesalers, manufacturers, etc. This expands market access.
- Advise trainees to obtain organic, good agricultural practices (GAP), or other certifications to add value and appeal to certain health-conscious buyers.
- Promote trainee's products through marketing materials, social media, and advertising to raise awareness among consumer groups. Help develop their brand identity.

- Suggest trainees offer samples and discounts to health influencers, supplement retailers, etc. to generate word-of-mouth buzz about the products' benefits.
- Facilitate trainee's attendance at relevant trade shows and events to network, promote their products, and identify leads.
- Recommend novel but value-added preparations like cordyceps powders or tinctures for niche markets. Innovation can boost interest.
- Helping trainees form strategic partnerships and relationships with buyers in the healthcare sector.

FAQ FOR *CORDYCEPS MILITARIS* CULTIVATION

1. What is *Cordyceps militaris*?

It is one of the most important traditional Chinese medicines of fungus that belongs to the class Ascomycetes.

2. Where it can be cultivated?

The cultivation of Cordyceps is done in a temperature and humidity-regulated laboratory

3. How much cordycepin is in Cordyceps?

The cordycepin concentration was 0.97% in the fruiting body and 0.36% in the corpus. There were differences in adenosine and cordycepin contents between the fruiting body and the corpus of *C. militaris*. The adenosine and cordycepin concentration in the fruiting body was approximately 3 fold higher than in the corpus.

4. How much time does it take to cultivate Cordyceps?

It takes around 60 – 80 days for the full cropping

5. What do you need to grow Cordyceps?

Cordyceps naturally grow on brown rice and broth substrate will be a nutrient-rich substitute. The dry ingredients include nutritional yeast or soy peptone, potato starch, magnesium sulfate, crushed multivitamins, and gypsum.

6. At what temperature does Cordyceps grow?

The highest fruiting bodies were formed at 25°C. Low temperatures of as low as 15°C also produced fruiting bodies, however at high temperatures, 30°C and above, no fruiting bodies were formed. Mycelia do not grow at higher temperatures.

7. Can Cordyceps infect people?

No. There is zero correlation with that fungus in humans.

8. Can Cordyceps be grown easily?

The Cordyceps mushrooms don't grow like other mushrooms. Nearly all *Cordyceps militaris* growers start with a supplemented rice substrate in glass jars under controlled condition.

9. What is the yield of Cordyceps?

The ability to successfully grow cordyceps in glass jars would greatly increase the economics of this crop. The highest average dry yield was 12 grams per jar.

10. How many types of Cordyceps are there?

Cordyceps, comprising over 400 species, was historically classified in the Clavicipitaceae, based on cylindrical asci, thickened ascus apices, and filiform ascospores, which often disarticulate into part-spores.

11. What is Cordyceps best for?

Cordyceps is used to treat coughs, chronic bronchitis, respiratory disorders, kidney disorders, nighttime urination, male sexual problems, anemia, irregular heartbeat, high cholesterol, liver disorders, dizziness, weakness, ringing in the ears, unwanted weight loss, and opium addiction.

12. What is the best grain for cordyceps?

Brown rice is considered the best grain substrate for cordyceps.

13. Can you eat Cordyceps fungus?

Cordyceps mushrooms are safe to eat. The fungus may interact with some medicines, such as blood thinners and certain immunosuppressants. However, you should consult your healthcare professional before taking a cordyceps supplement.

14. What are the benefits of this mushroom?

Cordyceps might improve immunity by stimulating cells and specific chemicals in the immune system. It might also help fight cancer cells and shrink tumor size, particularly with lung or skin cancers. The active principles of *C. militaris* are beneficial to act as anti-inflammatory, anti-

oxidant/anti-aging, anti-tumor/anti-cancer/anti-leukemic, anti-proliferative, anti-metastatic, immunomodulatory, anti-microbial, anti-bacterial, anti-viral, anti-fungal, anti-protozoal, insecticidal, larvicidal, anti-fibrotic, steroidogenic, hypoglycemic, hypolipidemic, anti-angiogenetic, anti-diabetic, anti-HIV, anti-malarial, anti-fatigue, neuroprotective, liver-protective, reno-protective as well as pneumo-protective, let alone their other synergistic activities.