MycoMate® liquid Culture Vial

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*TIP:
For best results utilize MycoMate® Liquid Culture Vials with MycoMate® and SporeMate® brand products. Success relies on the integrity of each product used in combination with MycoMate® Liquid Culture vials.

INSTRUCTIONS Liquid Growth Medium For Growth of Edible Mushroom Mycelium

1 INCLUDED:

- Mycomate® Liquid Culture Vial
- Sterile alcohol swabs
- Sterile syringe and 18 gauge needle

2 REQUIRED:

- Gas flame (a butane "torch-style" lighter works well)
- SporeMate® Edible Spore Suspension Vial or spore syringe, Mycomate® Liquid Culture Vial (containing mycelium), Mycomate® Cloning Kit, or other suitable inoculant
- Mycomate® Edible Mushroom or sclerotia kit, or other media or substrate suitable for growth (if desired end result is mushrooms)
- Clean hands and area to work

3 Inoculation:

Injection and colonization of liquid culture:

1. If stored under refrigeration, remove liquid growth media vial from refrigeration 24hrs before use and store at room temperature.
2. Wash hands thoroughly (antibacterial soap is optional) and dry with clean towel (i.e. paper towels).
3. Remove vial from bag and place on clean table in clean area, free from drafts.
4. Swab injection site of Liquid Culture vial and any other intended injection site (i.e. spore vial or colonized Liquid Culture vial). Utilize a fresh alcohol swab for each product to reduce cross contamination. Do not breath directly on injection sites.
5. Remove syringe and needle from bag. Remove the plastic shield protecting the needle (twist clockwise then pull). Do not touch the tip of the needle to anything else or it must be re-sterilized*. If utilizing syringe and needle from another supplier then it must first be sterilized.
   *Needles may be sterilized with a flame. You want tip of needle to become red but must be careful not to overheat and melt the area where the needle is attached to the plastic. Always let needle cool before injecting it. (See picture a)
6. If spore or colonized Liquid Culture vial is utilized, shake vial rigorously to mix up suspension. Hold vial at angle, inject with syringe, and draw up desired quantity of suspension. (See picture b)
7. Inject several drops of culture suspension (i.e. colonized liquid culture vial) or several drops to 1ml of spore suspension (depending upon density of spore solution) into Liquid Culture vial. Withdraw the needle very slowly or the pressure may cause some media to squirt out (which is ok). (See picture c)
8. Clean injection site of Liquid Culture vial with fresh alcohol swab, let dry, and store upright in airtight plastic bag.
9. Shake vial rigorously (10 seconds) once per day and store out of sunlight. Incubate species of fungi at appropriate temperature (typically 23-27º C). A lower temperature slows down spore germination and colonization time, however too high a temperature (above 27º C) creates an increased risk of contamination. (See picture d)
   Note: Temperature requirements are species-dependent.
10. Growth is similar with both spores and liquid culture, however growth from spores takes longer than from liquid culture.
    Spore Suspension: Spores typically germinate within 24 to 72 hours and within 3 to 7 days you should observe colonization of liquid media growing within the lower two thirds of the vial.
    Note: MycoMate® Liquid Culture Vials require very few spores for growth.
    Liquid Culture: Growth is apparent within 24 hours when transferring mycelium from a colonized vial of Liquid Culture into fresh Liquid Culture vials. Fresh vials are often colonized within several days (species, strain, and temperature-dependent).
    Observation is best when very carefully picking up the vial without agitating (shaking) and holding it up to the light. Sediment from the media will swirl if you are not careful making it more difficult to identify growth (this is ok, it will settle again). White strands of mycelium (fungi) will float towards surface as you pick vial up. There should be a small cloud around the mycelium but the entire vial should not be cloudy. The area surrounding cloud of mycelium should be fairly transparent. If vial is picked up slowly, yet completely cloudy then it's possibly contaminated. Let vial sit undisturbed for several hours and check again to be certain it was not sediment on bottom clouding the vial. Vial may be utilized as soon as there is healthy growth or after one to two weeks when vial shows greater colonization.
    Note: When one to two-thirds of vial has been colonized it should be utilized or stored under refrigeration for later use (see below).
11. When colonization is complete the vial may be used to inoculate other MycoMate® Liquid Culture Vials (repeat steps1 through 10), MycoMate® Edible Mushroom and Sclerotia Kits (Repeat steps 1 through 6), or other uses mentioned below. Additionally, follow directions of other products used in combination with your liquid culture.
    MycoMate® Liquid Culture makes preservation, mass production, and trading cultures between associates simple.

4 storage:

MycoMate® Liquid Culture vials may be utilized for both short and long-term storage of cultures, including but not limited to multi-spore, isolated, and cloned strains. One vial of liquid culture can be used to make thousands more. Once the vial is inoculated, and mycelium colonizes 30% to 65% of the liquid media, it may be stored under refrigeration (2-8º C) for many months (or longer) without detrimental effects. Store upright in two or three airtight plastic bags. Agitation (shaking) of cultures is not required during this period.
Save valuable time: When liquid culture is required, simply remove vial from refrigerator and use.

5 experimenting:

Cloning:
MycoMate® Liquid Culture vials may be utilized with MycoMate® Cloning Kits to clone both mushrooms and sclerotia. Multi-spore cultures are derived from a group of parents and strains that are not all compatible with one another and not all capable of reproduction (fruiting). Clones are developed from the tissue culture of a single parent (mushroom or sclerotium) as opposed to a multi-spore culture (i.e. spore syringe). Clones are considered pure strains (monocultures) and can often produce significantly higher yields with greater predictability. MycoMate® Cloning Kits make cloning easy. Alternatively, standard cloning techniques may be employed. However, we recommend reviewing appropriate literature prior to doing this on your own.

Hybrids:
MycoMate® Liquid Culture Vials may be utilized experimentally for the creation of hybrids. In essence hybrids can be formed between sub-strains of a single strain or between different strains of the same species.

Grain:
MycoMate® Liquid Culture allows you to inoculate much larger quantities of grain-based media than spores alone will allow. Colonization time and contaminations are significantly decreased which results in improved fruiting of mushrooms.

Bulk substrates
MycoMate® Liquid Culture Vials may be utilized to inoculate bulk substrates (i.e. pasteurized straw, dung, sawdust, woodchips, and newspaper) with success. End-results are experience and species-dependent and procedures should be researched prior to experimentation.

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Note to scientists: These vials may be utilized in non-sterile environments (i.e. in the field) during the course of research with a high degree of sterility and predictability. However, contamination levels are statistically reduced in relationship to the cleanliness of the environment in which inoculations are conducted.

Looking for a pure strain (monoculture)?
Try the MycoMate® Cloning Kit.

More info @ www.mycomate.com and www.sporemate.com

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