

## Preparation of a bag with solid substrates and inoculation with grain spawn for fructification

This protocol explains how to prepare a bag to grow a large amount of mycelium with the goal of fructification. This medium can be prepared without using an autoclave but it should be made with proper thick gloves for protection that need to be cleaned while working with spiritus.

**Materials (sufficient for 4 bags of 2.5 kg):** see below, bucket that can hold 20 L (for mixing), pot that can fit 10 L (for wheat bran boiling), jar (1 kg) with clean grain spawn, spiritus, sturdy plastic bags, thick gloves, distilled water (15 L)

component	amount	For bags 10x14x45cm
wheat bran	850 g	116.7 g
sawdust	3250 g	446.4 g
wood chips (beechwood)	1600 g	219.7 g
gypsum	125 g	17.2 g
water	4.1 L	0.56 L
well-grown preculture of oyster mushroom	1kg (solid substrate) or liquid	1 or 2 100 g jars of grain spawn

### Part 1: Soak And Prepare Bag Substrate (24 hours)

- Soak the beechwood wood chips for 24 h in water (**does not count for the water used above!**)
- Drain the wood chips properly (use a sieve/ a pot with holes)
- Bring water (4L) to boil in a large pot and pour over the wheat bran, stir to mix, let cool down a little
- Mix the sawdust and gypsum in a large vessel (10 – 20 L capacity)
- Add the drained beechwood chips to the gypsum/sawdust and mix
- Add the wheat bran (be cautious when hot) and mix well to obtain a uniform mix. Allow to cool down to room temperature (at least below 35°C)



## Part 2: Mix In Mycelium

- Clean your gloves with spiritus and break down the oyster mushroom preculture into small pieces (size of your thumb)
- Mix well into the substrate you prepared in the previous section
- Fill around 3 kg of the mix in plastic bags (thick walled/sturdy), and close the bag with a selfmade cloth-clothing



## Part 3: Incubate

- Incubate for 4 – 8 weeks in the dark

## Part 4: Cold Shock & Fruiting

- Once fully myceliated, remove the plastic layer from the tub and brush over with your (clean) fist on the surface to damage the mycelium
- Do an additional two slits on the side
- Place the plastic bag for 2 days in a colder place (fridge/basement/cellar) to induce fruiting
- Pull it out at room temperature and keep it wet by spraying water

