



## Raising *Drosophila melanogaster* in the laboratory

### Preparation of growth medium for *D. melanogaster*

#### General Notes

The following growth medium is used in the “Insect Molecular Biology” laboratory of the Institute of Biosciences and BioResources“ of the CNR of Naples.

It is a medium that is compact enough to resist the liquefaction produced by intense larval activity. Once put into containers, they can be stored for 3-4 days at room temperature (RT); however, for weaker strains or newly started crosses, it is recommended to use fresh or, at most, 1-2 day-old medium. If stored at 4°C, this medium will last for a week, but it may exude liquid when brought to RT. To reduce this problem, containers should be left at RT for 24 hours before storage at 4°C.

#### Recipe

The following amounts yield approximately 1.3 liters of medium, enough to fill about 60 3cm diameter or 130 2cm diameter containers.

Tap water	1.1 liter
Brewer's yeast	100 grams
Precooked cornmeal	90 grams
Sugar	100 grams
Agar	3-4 grams
p-hydroxymethyl benzoate	2.5 grams
ethanol 95%	30 milliliters

#### Cooking process

The cooking process is fundamental to producing high quality growth medium. Poor growth medium is often simply a medium that is too dry. Be aware that seemingly small variations in the cooking process may result in a substantial difference in the quality of the growth medium if they lead to increased water loss. Some parameters which must be given careful attention are: the ratio of the quantity of medium to the volume of the pot, using an uncovered pot, the necessity of long cooking times, an especially dry work environment.

- Put the yeast in the tap water and bring it to a boil.
- Separately, measure the cornmeal, sugar and agar and mix them together.
- After boiling for 5-10 minutes, enough time to deactivate the yeast, pour the previously prepared mix into the water/yeast.
- Once it has begun to boil again, cook for about 10 minutes, stirring continuously.
- Turn off the heat source.



- Mix in the p-hydroxymethyl benzoate, previously dissolved in the 95% ethanol.
- Put the medium into containers, filling them about  $\frac{1}{4}$  full, using an appropriate device or a plastic cylinder.
- Allow the medium to dry for an adequate period, to be calculated experimentally, in front of a fan to avoid flies entering and contaminating the containers.
- Plug with hydrophobic cotton wool.

## Notes on the ingredients

### Cornmeal

Not every brand can be used; different brands should be tested before purchasing the laboratory stock. In our lab, we use “Polenta Valsugana”.

### Agar

The quantity of agar must be determined experimentally, depending on the brand or batch delivered.

### Yeast

Powdered or granular yeast can be used. In our lab, we use cakes of brewer's yeast which are immediately frozen after purchase for long-term storage.