Preparation of 100 ml germination medium

The medium consists of inorganic salts and vitamins of MS medium, 30 g/L sucrose, 100 mg/L myo-inositol and 8 g/L agar Using:

- **powdered MS medium (salts and vitamins) 4.4 g/L**
- **Sucrose**
- **Myo-inositol stock solution (10 mg/ml).**
- **Agar**

The pH is 5.8

Calculations

MS Powder

4.4 g ⟶ 1000 ml

X g ⟶ 100 ml

$$X = \frac{4.4 * 100}{1000} = 0.44 \text{ g}$$

Sucrose

30 g ⟶ 1000 ml

X g ⟶ 100 ml

$$X = \frac{30 * 100}{1000} = 3 \text{ g}$$

Myo-inositol stock conc. = 10 mg/ml = 10 000 mg/L

Stock — NV = N'V' — Medium

10 000 * V = 100 * 100

$$V = \frac{100 * 100}{10\,000} = 1 \text{ ml}$$

Agar

8 g ⟶ 1000 ml

X g ⟶ 100 ml

$$X = \frac{8 * 100}{1000} = 0.8\ g$$

Steps:

In 250 ml clean beaker, put 50 ml distilled water and dissolve:

0.44 g powdered MS medium (weigh using 3 digits electric balance)

3 g sucrose

1 ml myo-inositol stock solution (10 mg/ml) (using 1 ml glass Pipette or 1000 µl micropipette)

Up to 90 ml with distilled water (using 100 ml measuring cylinder)

Adjust pH to 5.8 using KOH and HCl

Up to 100 ml with distilled water (using 100 ml measuring cylinder)

Add 0.8 g Agar and boil with continuous stirring till disappearance of Agar

Divide into 2 jars

Autoclave for 20 minutes at 121 ^{o}C.

Results for seed germination

$$\text{Contamination\%} = \frac{\text{No. of Contaminated Cultures}}{\text{Total No. of Cultures}} X100$$

$$\text{Gernination\%} = \frac{\text{No. of germinating seeds}}{\text{Total No. of seeds}} X100$$

Seven-day-old canola seedling

Comment: