

BLACK MEXICAN SWEET CELL CULTURE AND MAINTENANCE PROTOCOL

M237 Media (1L)

1. 900 mL ddH₂O
2. Add the following
 - 4.3 g MS Salt
 - 30 g Sucrose
3. Adjust pH to 5.6 with 1M KOH
4. Adjust volume to 1L
5. Distribute 50 ml media into 250 mL flasks. Cover the flasks.
6. Autoclave for 20 min

Let the media cool down (approx. 60°C), and add 1 mL MS vitamins (Sigma-Aldrich, cat# M3900-50ML) + 0.5ml 2,4-D (2mg/ml, filter-sterilized). For plates add 0.7% of agar before autoclaving.

Suspension Cell Culture

The suspension cell culture was maintained on a rotary shaker (120 rpm) at 24C in dark conditions.

Every 3 weeks, transfer cells into fresh M237 media (1:10 v/v) for subculture.