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Preparing 10 L of M9 buffer for nematode culture V.3

Gurdon Institute media kitchen¹¹Cancer Research UK / Wellcome Gurdon Institute

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Works for me

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ABSTRACT

Prepare 10 litres of M9 solution for collecting and washing *C. elegans*.

MATERIALS

NAME ▾

CATALOG # ▾

VENDOR ▾

double distilled water (ddH₂O)

Di-Sodium Hydrogen Orthophosphate Dihydrate Certified AR for Analysis Fisher Chemical

S/4450/53

Sodium chloride meets analytical specification of Ph.Eur Fisher Chemical

S/3160/65

Fisher Scientific

Magnesium Sulfate Heptahydrate Certified AR for Analysis Fisher Chemical

M/1050/53

Fisher Scientific

Potassium Dihydrogen Orthophosphate Certified AR for Analysis Fisher Chemical

P/4800/53

Fisher Scientific

SYCHEM autoclave

View

Syschem

SAFETY WARNINGS

This protocol makes use of an autoclave. Make sure you know how to use it properly before starting the protocol.

20m

1

Ingredients	Quantity	
Na ₂ HPO ₄		60g
KH ₂ PO ₄		30g
NaCl		50g
Double distilled H ₂ O	up to 10L	
Add 1M MgSO ₄ after autoclaving @ 100ul/100ml		

M9_buffer_10L.xls

15m

2

1	Dissolve ingredients in 8L double distilled H ₂ O
2	Adjust volume to 10L using double distilled H ₂ O
3	Measure or use pump to dispense aliquots accurately.
4	Label, date and autoclave.
5	After autoclaving, add 100ul 1M MgSO ₄ per 100ml. (ie. 100ul to 100ml and 200ul to 200ml bottles).

NB	1M MgSO ₄ at 1ul/1ml in the M9 buffer gives a final concentration of 1mM
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3 The autoclave is SYCHEM VS Series. The media cycle is 15 minutes at 121 degrees centigrade. It is then cooled down in the machine for 3 hours. 3h 15m

4 After autoclaving, add 100 µl 1M MgSO₄ per 100 ml. (ie. 100 µl to 100 ml and 200 µl to 200 ml bottles). 10m

NB: 1M MgSO₄ at 1 µl/1ml in the M9 buffer gives a final concentration of 1 mM



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