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8. Anexos

8.1. MW (Modified Whitten's Medium)

MW	Código Sigma	mM	Quantidade em 1L
NaCl	S9625	100	5,84 g
KCl	P3911	4,78	0,357 g
$CaCl_2$	4225	2,5	0,275 g
KH_2PO_4	P0662	1,19	0,207 g
$MgSO_4$ x 7 H_2O	M2393	1,19	0,293
$NaHCO_3$	S6014	25,07	2,106 g
Piruvato de Sódio	P2256	1	0,022 g
Lactato de Sódio	L1375	4,8	3,68 mL
Glicose	G8270	5,5	0,991 g
HEPES	H3375	22	5,236 g
BSA	A9647	-	7 g
Água Destilada	-	-	Completar para 1000 mL

(Modificado de Whitten and Biggers, 1968; por McPartlin *et al*., 2008)

8.2. FR5 Modificado

Solução 1: FR1				
Água Destilada	100 mL	250 mL	500 mL	1000 mL
Glicose	5 g	12,5 g	25 g	50 g
Lactose	0,3 g	0,75 g	1,5 g	3 g
Rafinose	0,3 g	0,75 g	1,5 g	3 g
Citrato de Sódio	0,05 g	0,125 g	0,25 g	0,5 g
Citrato de Potássio	0,082 g	0,205 g	0,41 g	0,82 g
HEPES	1,42 g	3,55 g	7,1 g	14,2 g
300 – 310 mOsm / pH 7,2 – 7,4				
Cada 100 mL dessa solução faz 250 mL do FR5				
Solução 2: Leite em pó - Glicose				
Água Destilada	100 mL	250 mL	500 mL	1000 mL
Leite em pó desnatado	14,4 g	36 g	72 g	144 g
Glicose	0,8 g	2 g	4 g	8 g
300 – 310 mOsm / pH 7,2 – 7,4				
Solução 3: FR4				
Adicionar volumes iguas da solução 1 e solução 2				
Adicionar 3% de gema de ovo ao volume final				
Centrifugar a 7000 g por 30 minutos				
Filtrar o sobrenadante em filtros de papel 8 microns a vácuo				
Solução 4: FR5				
Adicionar Crioprotetor (5% glicerol)				