YES-HU plate

Preparation of HU (Hydroxyurea) Stock Solution; 1 M

Hydroxyurea … MW: 76.05 @ 4 C
Dissolve 0.76 g in 10 ml water. Aliquot in 1.5 ml tubes. Keep –20 C.

Preparation of YES-HU plates

Make 1 L of YES with 20g agar in a 2 L plastic flask with a big stir bar. Autoclave 45 min. Stir slowly until it gets cool enough to add the drug, like you can touch it with bare hands (~60˚C). Add HU Stock Solution and mix thoroughly. Then pour into Petri dishes before it starts solidify. 1 L makes 2 sleeves (40) of plates. Let plates dry for 2 days, put them back in sleeves and store at 4˚C room. The drug in plate last for a year.

2 mM HU plate: 2.0 ml / 1 L
5 mM HU plate: 5.0 ml / 1 L

YES-CPT plate

Preparation of CPT (Camptothecin) Stock Solution; 10 mM

Camptothecin … MW: 348.36 @ 4 C
Dissolve 34.8 mg CPT in 10 ml DMSO. Aliquot in 1.5 ml tubes. Keep –20 C.

Preparation of YES-CPT plates

Make 1 L of YES with 20g agar in a 2 L plastic flask with a big stir bar. Autoclave 45 min. Stir slowly until it gets cool enough to add the drug, like you can touch it with bare hands (~60˚C). Add CPT Stock Solution and mix thoroughly. Then pour into Petri dishes before it starts solidify. 1 L makes 2 sleeves (40) of plates. Let plates dry for 2 days, put them back in sleeves and store at 4˚C room. The drug in plate last for a year.

2 μM CPT plate: 0.2 ml / 1 L
5 μM CPT plate: 0.5 ml / 1 L

YES-G418 plate

Preparation of G418 Stock Solution; 50 mg/ml

Geneticin concentration… 733 μg/mg (Check the label. Each lot is different.)
Dissolve 2.046 g into 30 ml of 1M Tris-HCl (pH8.0), and filtrate with a 0.22 μm disk filter on a 50 ml syringe.

Preparation of YES-G418 plate

Make 1 L of YES with 20g agar in a 2 L plastic flask with a big stir bar. Autoclave 45 min. Stir slowly until it gets cool enough to add the drug, like you can touch it with bare hands (~60˚C). Add 3 ml G418 Stock Solution and mix thoroughly. Then pour into Petri dishes before it starts solidify. 1 L makes 2 sleeves (40) of plates. Let plates dry for 2 days, put them back in sleeves and store at 4˚C room. The drug in plate last for a year.

150 mg/l G418 plate: 3 ml / 1 L for 40 plates (2 sleeves)

YES-NAT plate

Preparation of clonNAT Stock Solution; 200 mg/ml (8000x)

clonNAT (Nourseothricin Sulfate) [Werner BioAgents]… powder @4˚C
Dissolve 200 mg into 1 ml of sterilized H_2O, and filtrate with 0.22 μm filter.
Aliquot 100 μl in 1.5 ml tube, and store at -20˚C.

Preparation of YES-clonNAT plate

Make 500 ml of YES with 20g agar in a 1 L flask with a big stir bar. Autoclave 45 min. Stir slowly until it gets cool enough to add the drug, like you can touch it with bare hands (~60˚C). Add clonNAT Stock Solution and mix thoroughly. Then pour into Petri dishes before it starts solidify. 500 ml makes 1 sleeve (20) of plates. Let plates dry for 2 days, put them back in sleeves and store at 4˚C room. The drug in plate last for I don't know.

25 μg/l NAT plate: 125 μl / 500 ml for 20 plates

YES-HYG plate

Preparation of Hygromycin B Stock Solution; 50 mg/ml in PBS (500x)

Hygromycin B [Roche]… 1 g (in 20 ml PBS solution, sterile) @4˚C
Aliquot in 1.5 ml tubes, and store at 4˚C.

Preparation of YES-HYG plate

Make 1 L of YES with 20g agar in a 2 L plastic flask with a big stir bar. Autoclave 45 min. Stir slowly until it gets cool enough to add the drug, like you can touch it with bare hands (~60˚C). Add 3 ml Hygromycin B Stock Solution and mix thoroughly. Then pour into Petri dishes before it starts solidify. 1 L makes 2 sleeves (40) of plates. Let plates dry for 2 days, put them back in sleeves and store at 4˚C room. The drug in plate last for 6 months.

100 mg/l HYG plate: 2 ml / 1 L for 40 plates