

Commonly used *Streptomyces* media

Medium	Ingredients and notes
<p>Solid Minimal Medium (Kieser <i>et al.</i>, 2000))</p>	<p>0.5 g or 1 g L-asparagine or (NH₄)₂SO₄ 0.5 g K₂HPO₄ 0.2 g MgSO₄*7H₂O 0.01 g FeSO₄ *7 H₂O 10 g Agar</p> <p>Add 1000 ml dH₂O, pH to 7.0 with 5M NaOH, Make up 1% carbon (see below) and add after autoclaving.</p>
<p>Solid Minimal Medium (Karandikar <i>et al.</i>, 1997)</p>	<p>1.7 g NaNO₃ 1 g K₂HPO₄ 0.5 g KCl 0.5 g MgSO₄*7 H₂O 0.01 g FeSO₄*7 H₂O 2.5 mL/L Trace element solution * 15 g Agar</p> <p>Add 800 ml dH₂O, pH to 7.0 with 5M NaOH, add 200 ml of 50 g/l glucose after autoclaving.</p>
<p>Hobbs Minimal Medium (Hobbs <i>et al.</i>, 1989)</p>	<p>5 g NaCl 5 g Na₂SO₄ 4.5 g NaNO₃ 1.2 g TRIS 2 g MgSO₄* 7 H₂O 0.01 g ZnSO₄* 7 H₂O Add 1000 ml dH₂O, pH was adjusted to 7.2</p> <p>added after autoclaving:</p> <p>1ml K₂HPO₄ (20 g/L stock) 1 mL/L trace element solution *</p> <p>either glucose or Tween40 were used as carbon source (0.33 mol carbon) 20 µL of Sigma Antifoam A</p>

<p>Mannitol Soya Flour Medium</p> <p>(Hobbs <i>et al.</i>, 1989)</p>	<p>16 g Mannitol</p> <p>16 g Soya Flour</p> <p>16 g Agar</p> <p>Add 1000 ml tap water</p> <p>Make in 250 ml batches and autoclave in 500 ml Duran bottles as the medium bubbles out</p>
<p>R2 (Solid)</p> <p>(Kieser <i>et al.</i>, 2000)</p>	<p>103 g Sucrose</p> <p>0.25 g K₂SO₄</p> <p>5 g MgCl₂ *6 H₂O</p> <p>10 g glucose</p> <p>0.1 g casein hydrolysate</p> <p>2 mL/L trace element solution **</p> <p>5.73 g TES buffer</p> <p>22 g agar</p> <p>Add 1000 ml dH₂O (heat to dissolve the components and aliquot in to 200 ml batches for autoclaving)</p> <p>additional components added after autoclaving (per 100 mL)</p> <p>1 mL 0.5% KH₂PO₄</p> <p>0.4 mL 3.68% CaCl₂*2 H₂O</p> <p>1.5 mL 20 % L-Proline</p> <p>0.7 mL 1 M NaOH</p>

<p>R5 (Solid) (Kieser <i>et al.</i>, 2000)</p>	<p>103g sucrose 0.25 g K₂SO₄ 10.12 g MgCl₂*6 H₂O 10 g glucose 0.1 g casein hydrolysate 2 mL/L trace element solution** 5 g yeast extract 5.73 g TES buffer 27.5 g agar</p> <p>Add 1000 ml dH₂O (heat to dissolve the components and aliquot in to 320 ml batches for autoclaving). Additional components added after autoclaving (per 80 mL)</p> <p>1 mL 0.5% KH₂PO₄ 0.4 mL 5 M CaCl₂*2 H₂O 1.5 mL 20% L-Proline 0.7 mL of 1 M NaOH</p>
<p>2xYT (Liquid) (Kieser <i>et al.</i>, 2000)</p>	<p>16 g tryptone 10 g yeast extract 5 g NaCl Add 1000 ml dH₂O</p>
<p>YEME (Liquid and solid) (Hoskisson <i>et al.</i>, 2000; Kieser <i>et al.</i>, 2000)</p>	<p>5 g peptone 3 g yeast extract 3 g malt extract 10 g glucose</p> <p>OPTIONAL: 340 g sucrose (Can be added for more dispersed growth).</p> <p>Add 1000 ml dH₂O – Adjust pH to 7.2 with 5 M NaOH. Make in 250 ml batches and autoclave in 500 ml Duran bottles as the medium can bubble out.</p> <p>A solid version of this can be made by omitting the sucrose and adding 20 g of agar per litre. Again, make in 250 ml batches and autoclave in 500 ml Duran bottles as the medium bubbles out.</p>

* Trace element solution of Hobbs et al., (1989) consists of (g/100 mL; Filter sterilised): 0.1 ZnSO₄*7 H₂O, 0.1 FeSO₄ *7 H₂O, 0.1 MnCl₂ * 4 H₂O, 0.1 CaCl₂ * 6 H₂O and 0.1 NaCl, can be stored at 4° C for 2–4 weeks. It is diluted 1:10 before added to the medium

** Trace element solution was composed of: 8.78 g/L FeCl₃, 2.04 g/L ZnSO₄, 1.02 g/L MnCl₂*4 H₂O, 0.43 g/L CuSO₄*2 H₂O, 0.42 g/L NaI, 0.31 g/L H₃BO₃, 0.24 g/L CaCl₂*6 H₂O and 0.24 g/L Na₂MoO₄*2 H₂O

Carbon sources added to minimal medium for screening

Carbon source	Stock/pH	mL to 1L medium (1 %)
Glucose	50	20
Mannitol	25	40
Tween80	20	50
Pyruvate	20/ pH 7.2	50
Na-Acetate	20/ pH 7.2	50
Na-Citrate	20/ pH 7.2	50
Malic acid	20/ pH 7.2	50
PEP	20	50
α-KG	20	50
GlcNac	20	50

References

- Hobbs, G., Frazer, C., Gardner, D. J., Cullum, J. & Oliver, S. (1989).** Dispersed growth of *Streptomyces* in liquid culture. *Applied Microbiology and Biotechnology* **31**.
- Hoskisson, P. A., Hobbs, G. & Sharples, G. P. (2000).** Response of *Micromonospora echinospora* (NCIMB 12744) spores to heat treatment with evidence of a heat activation phenomenon. *Letters in Applied Microbiology* **30**, 114–117.
- Karandikar, A., Sharples, G. P. & Hobbs, G. (1997).** Differentiation of *Streptomyces coelicolor* A3 (2) under nitrate-limited conditions. *Microbiology (Reading, Engl)* **143**, 3581–3590. Soc General Microbiol.
- Kieser, T., Bibb, M. J., Buttner, M. J., Chater, K. F. & Hopwood, D. A. (2000).** *Practical Streptomyces Genetics*. John Innes Foundation.