



University of Melbourne iGEM 2014 Lab Procedure

Procedure	Name:	Preparation of Liquid Broth Media		
	Description:	Preparation of LB (liquid broth) from raw materials and milliQ water. Can be heated up and used to make plates when needed. <b>Storage conditions:</b> Store finished LB under 4°C (bar fridge) after autoclaving. <a href="#">Preparation of Media Video</a>		
	Trigger:	When bacterial growth is desired.		
Last updated	Name:	Michael	Date:	23/06/14
You will need	Time:	20 minutes + autoclave		
	PPE:	Standard latex gloves		
	Equipment:	<ul style="list-style-type: none"> <li>➤ 1L/300mL storage bottles</li> <li>➤ 2.d.p scale</li> <li>➤ Autoclave</li> </ul>		
	Materials:	<ul style="list-style-type: none"> <li>➤ MilliQ water</li> <li>➤ 3.0g tryptone               <ul style="list-style-type: none"> <li>- 1.0% per L</li> </ul> </li> <li>➤ 1.5g yeast extract               <ul style="list-style-type: none"> <li>- 0.5% per L</li> </ul> </li> <li>➤ 3.0g sodium chloride NaCl               <ul style="list-style-type: none"> <li>- 1.0% per L</li> </ul> </li> <li>➤ 4.5g bacteriological agar               <ul style="list-style-type: none"> <li>- 1.5% per L</li> </ul> </li> </ul>		
<b>Preparation of liquid broth:</b>				
Step 1	Obtain a 300mL clean glass bottle.			
Step 2	Into the bottle, weigh and add: <ul style="list-style-type: none"> <li>- 3.0g tryptone</li> <li>- 1.5g yeast extract</li> <li>- 3.0g sodium chloride</li> <li>- 4.5g bacteriological agar</li> </ul> Make up solution to 300mL <b>Note:</b> with milliQ water.			
Step 3	Mix contents thoroughly with the magnetic stirrer until most contents have dissolved. <b>Note:</b> this may take a while. Remove the magnetic stirrer once completed.			
Step 4	Tighten the bottle cap loosely and place the bottle into the autoclave tray. Follow the prompts on the autoclave machine and select 'Wet 20' option.  Ready to start work? > Yes > Wet 20  <b>Note:</b> If only autoclaving empty bottles, use Dry 20/Dry 25. For LB preparation, use Wet 20.			
Version history	<i>Describe the changes made in each new version of the protocol here.</i>			