University of Melbourne iGEM 2014 Lab Procedure

Procedure	Name:	Coomassie Staining		
	Version:	2		
	Description:	How to perform a Coomassie stain on a SDS-PAGE membrane. Visualising Proteins Video		
	Trigger:	Use this procedure after having performed SDS-PAGE.		
Last updated	Name:	Elizabeth Brookes	Date:	19.07.14
You will need	Time:	3 hours		
	PPE:	Gloves Lab coat		
	Equipment:	Container with Lid Rocking Platform		
	Materials:	Coomassie Stain (Cheng Lab recipe):		

	Quantity required for 500 mL stain solution
Coomassie Blue R: 0.25% (0.25 g/100 mL)	=500 mL /100* 0.25 = **1.25 g Coomassie**
Acetic Acid: 20%	=500 mL*0.2=**100 mL**
Methanol: 10%	=500 mL*0.10=**50 mL**
MilliQ Water	=500-100-50=**350 mL**

Destain Solution (Cheng Lab reciple):

	Quantity required for 500 mL destain solution
10% Acetic Acid.	=500*0.1=50 mL
20% MeOH	=100 mL
70% MilliQ Water	=350 mL

Step 1	Place the SDS-PAGE membrane to be stained in a container with a lid. ***Note: If you need to handle the gel make sure to only touch the bottom of the gel, not the top of the gel. Use gloves and handle with care.***
Step 2	Add enough Coomassie Stain to cover the membrane (~60mL) and leave the solution to rock on a rocking platform for 40 minutes.(N.b. can leave it for somewhat longer than 40 minutes but not longer than 1 hr) ***Note: Ensure that you have labelled the container appropriately so that it is not confused with any other Coomassie Stains on the rocking platform.***
Step 3	After the 40 minutes, carefully pour out the Coomassie Stain solution and rinse the gel twice with Milli-Q Water. ***Note: Be careful not to pour the membrane out into the sink along with the solution.***

Step 4	After having rinsed the membrane, cover the membrane with at least 1cm of Destain Solution above the gel. Leave the gel in the solution until the background staining disappears (the stain not bound to the protein is removed).
Step 5	Once the gel has been destained it may be stored in distilled water at 4℃.
Version history	*V2: Changing recipe to match Cheng lab one, as their stains work beautifully and ours have sucked. - Sean*