Stain-Free Analysis in GeneSys - Quick Guide

Start GeneSys

1. Having run your stain-free gel it will need to be activated via the UV-transilluminator
2. Select-> **Gel-> Stain Free**
3. Select "Stain-free gel (13.3 x 8.7 cm)" or customise your gel size
4. In the dye selection select 'Criterion Stain-Free'. This will automatically set-up a 5 minute UV exposure time. However, if a lesser time is required then this action can be completed via the manual capture mode
5. Position the stain-free gel on the UV transilluminator, ensure the Iris, Zoom and the Focus are set at optimal, then select the 'Next' green arrow

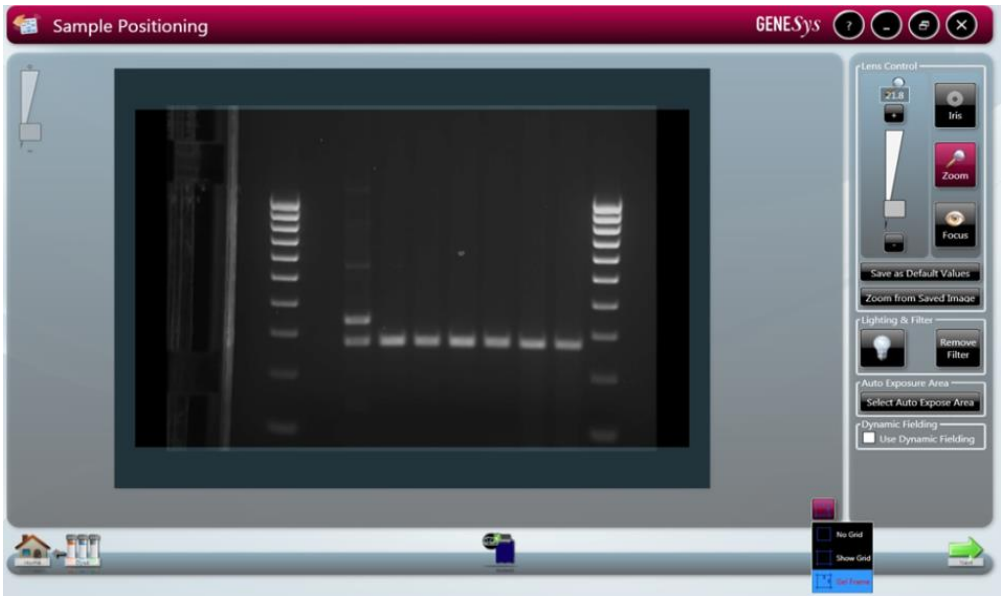
UV activation of the stain-free gel

1. The stain-free gel will be UV activated for 5 minutes

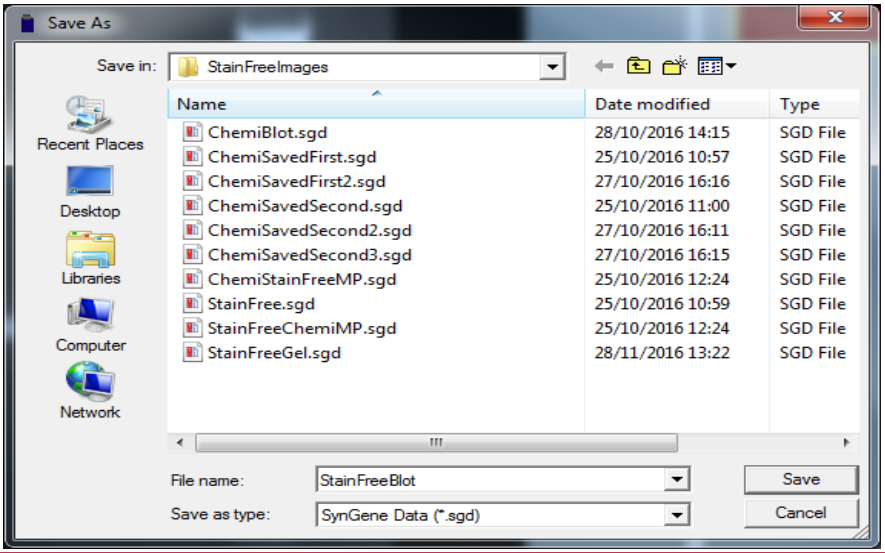
2. Save the protocol if required
3. Save the captured image in an SGD format

Transfer proteins to membrane/blot

1. Upon membrane/blot transfer, it can now be imaged via the UV-transilluminator
2. Select -> **Blots -> Stain-Free Blot**
3. Select the blot size or customise your blot size
4. Select the 'Criterion Stain-Free' (Membrane) in the Dye Selection
5. Position the blot on the UV-transilluminator
6. Use the "Gel Frame" to align your blot. We recommend you align the top-left of the frame with the top-left of your blot
7. Ensure the Iris, Zoom and the Focus are set at optimal, then select the 'Next' green arrow to capture the image of the blot



7. Save the protocol if required
8. Save the blot image in an SGD format, to be used in the future for normalisation studies



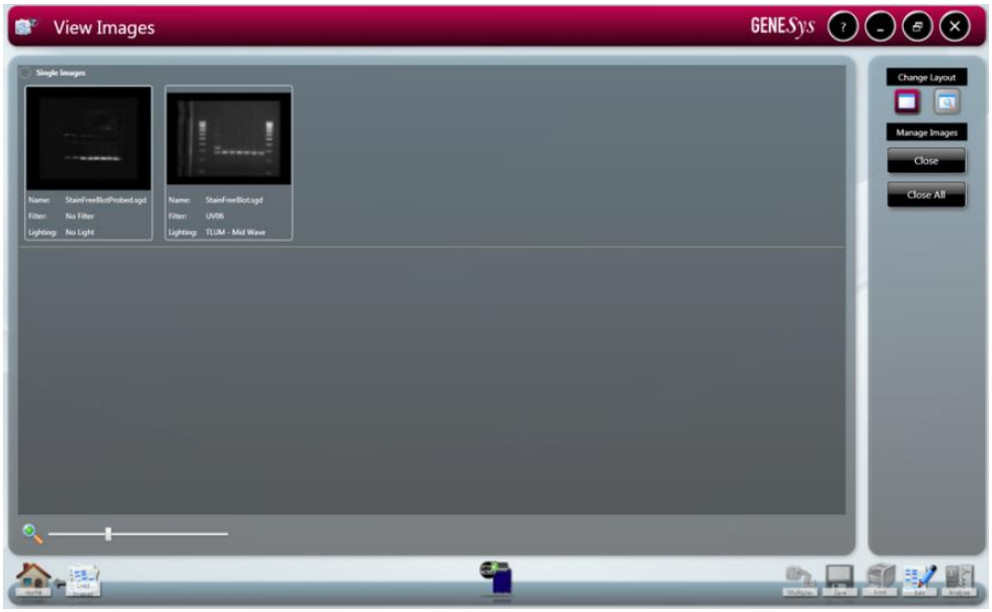
Probe the membrane/blot for proteins of interest

1. Probe the blot for your protein of interest if you are performing a stain-free normalisation experiment
2. To capture the probed image: Select **-> Blots -> select your detection chemistry, ie, chemiluminescence**
3. 'Select from file' to open previously saved blot image to permit the zoom parameter to be the same. The Iris and Focus can still be adjusted
4. Use the "Gel frame" to align your blot; align the top-left of the frame with the top-left of your blot
5. **NB** if you **did not** select "Select from file" you can select "Zoom from Saved Image" to use the same zoom parameter
6. Then select the 'Next' green arrow to capture the image of the blot
7. Save the image in an SGD format

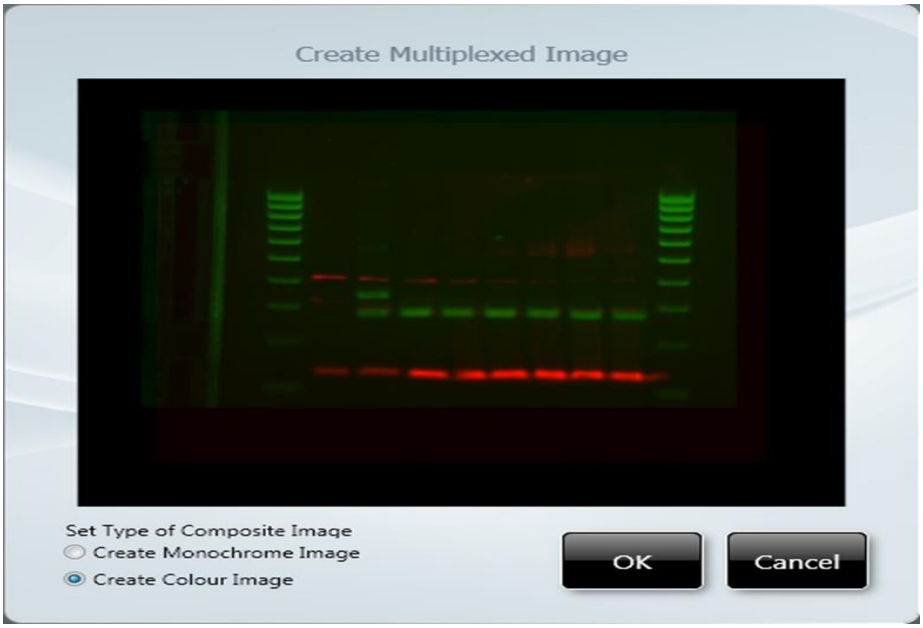
NB if capturing chemi images in auto mode; for Chemi Blot (series), binning may be defaulted to 3x3, you should set this to 'No Binning'. This will allow you to multiplex your image, if binning was not used previously.

Multiplex the membrane/blot images together

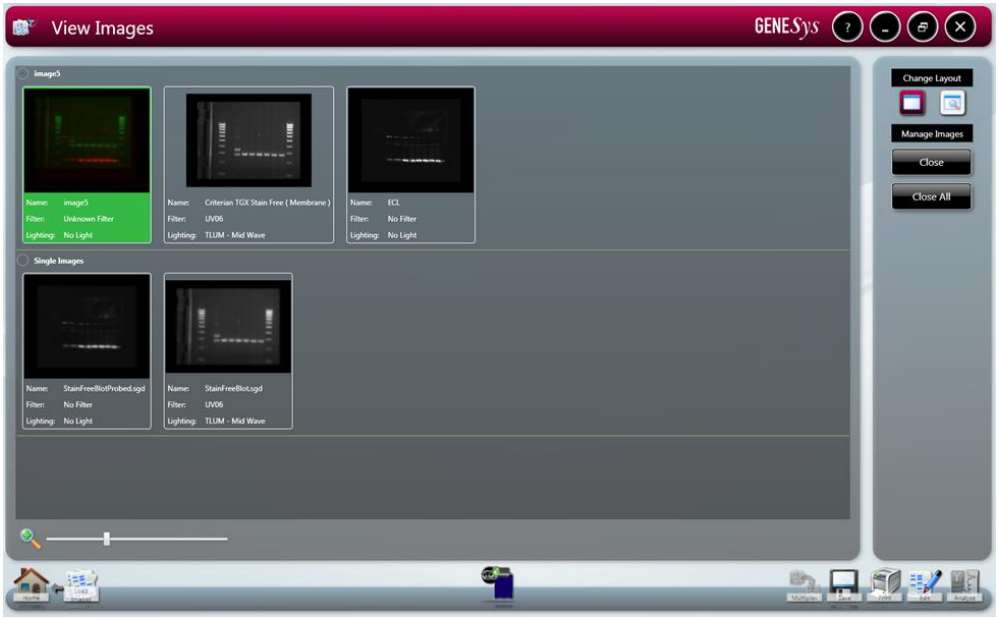
1. At this stage you will have two or more images permitting you to perform a stain-free normalisation experiment in GeneTools
2. Multiplexing the membrane/blot images together; go to view images



3. Select the two or more images required to form a multiplex and then press the 'Multiplex button'



4. Select "Create colour image" for the composite which makes the identification of the channels easier and has no influence on the data to be analysed. Press OK. The new multiplex image will appear in the view images screen



5. Select the composite image, press the Save button and save as a single file by selecting 'Together'

6. Save the multiplex file in an SGD format
7. The file is now ready to be analysed in GeneTools

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