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 We use this protocol and it's working

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Western Blot (Tank-Blot) V.1

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ABSTRACT

Blotting Buffer:

- 100mL 10x SDS running buffer
- 200mL 100% Methanol
- 700mL H2O

- Fill a tray with blotting buffer
- Transfer SDS gel to the blotting buffer
- Assemble blotting sandwich (take it, soak sponge and place on it, soak filter paper and place on it, place inverted SDS gel on top, without creating bubbles. Add charged PDVF membrane with forceps (charged in 100% ethanol and then soaked in buffer)
- Add soaked filter
- Streak out bubbles
- Add soaked sponge
- Press together and close container
- Place in blotting apparatus and add lotting buffer
- Run for 1h at 100V 350-500mA