



CELLULOSE BINDING TEST

Aim: Verify that the C2 protein binds to cellulose.

Protocol:

- in a 2ml Eppendorf tube, put:
 - 10mg of Avicel cellulose
 - 1ml of the used protein purification buffer: PBS (150mM NaCl, 50mM sodium phosphate) or Buffer A (150mM NaCl, 50mM Tris, pH=7,4)
 - 10µl of BSA (1µg/µl)
 - 100µl of the fraction of the purified protein C2

Use a mixture without C2 protein as a negative control.

- incubate at room temperature for 1 hour, under stirring.
- centrifuge the tube at 13,200rpm for 5min.
- recover the supernatant and store it in a 1.5ml Eppendorf tube.
- wash the pellet with the buffer used before (stir for 5min).
- centrifuge the tube at 13,200rpm for 5min.
- recover the supernatant and store it in another 1.5ml Eppendorf tube.
- denature the pellet with 200µl of Laemli 2x at 95°C for 5min.
- denature 10µl of the two previous supernatants with 10µl of Laemli 2x at 95°C for 5min.
- analyse the three samples on an SDS PAGE protein gel.