

Protocol for fluorescent TUNEL assay + immunofluorescence labeling

(Gelatin or OCT sections)

2 x 5' in PBS at \pm 40-45°C

Microwave treatment : in 200 ml citric acid 10 mM pH6 (required for the TUNEL procedure) :

- prewarm the solution for 5' in the MW (until it boils)
- MW treatment of the sections for 5' (might ebullition)

Add 80 ml water to cool down the solution

2' on ice in Sodium citrate 0.1% + TritonX-100 0.1% (cool down the solution before treatment)

Incubate in TUNEL mix for 60' at 37°C (\pm max. 150 μ l solution/slide) :

- 1/10 TUNEL solution 1 (blue cap)
- 9/10 TUNEL solution 2 (purple cap)

(wash in PBS)

Bloc 30' at RT

Proceed with immunofluorescence as usual