

## General protocol: Chromogenic Immunohistochemistry (IHC)

### Required Materials

- ☐ Xylenes
  - Ensure xylenes are handled under sufficient ventilation (fume hood).
- ☐ 100% ethanol
- ☐ 95% ethanol
  - If more 95% ethanol is needed, dilute by adding 41.2mL distilled water to unopened 4L jug of 100% ethanol
- ☐ Distilled water
- ☐ 3% Peroxide Solution
- ☐ Antigen retrieval solution
  - Sodium citrate solution (IHCWorld):
    1. Add 2.94g trisodium citrate to 1000 mL distilled water (milliQ).
    2. Adjust pH to 6.0 or 9.0 (application dependent) using test strips by adding very small amounts of HCl (if too acidic, recover with very tiny amount of NaOH).
    3. Add 0.5mL Tween.
- ☐ Casein blocking buffer
- ☐ Tween 20
- ☐ Unconjugated primary antibodies
- ☐ HRP-conjugated secondary antibodies
- ☐ Humidity chamber
- ☐ PBS (Stock or 10x)
- ☐ DAB kit
  - Carcinogenic. Handle with care & avoid contact.
- ☐ Hematoxylin
- ☐ Bluing buffer (0.1% sodium bicarbonate)
- ☐ Permount slide mounting medium