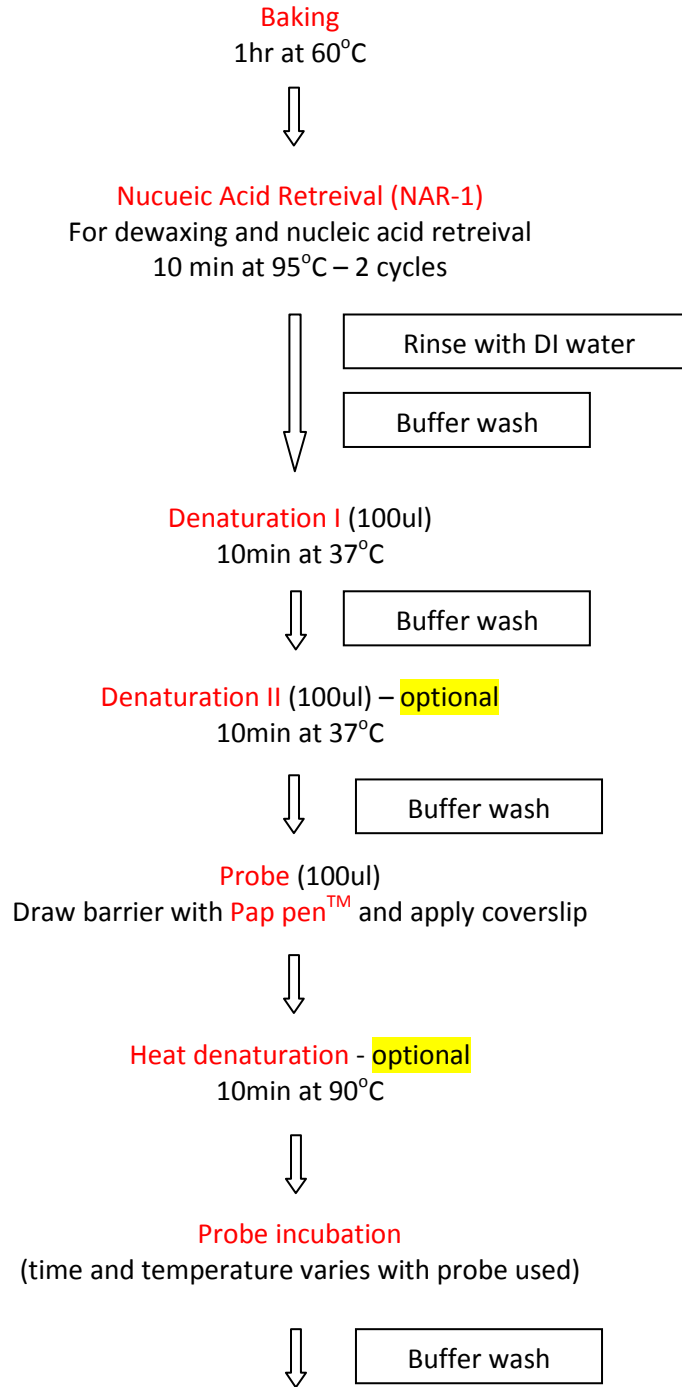


# *In Situ* hybridization (ISH) Protocol

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For probes that require Nucleic Acid retrieval (DNA probes)



Wash A (100ul)  
10min (temp varies with probe)



Buffer wash

Wash B (100ul)  
10min (temp varies with probe)



Buffer wash

Peroxide Block II (100ul)  
10min at room temp



Buffer wash

Power Block™ (100ul)  
10min at room temp



Anti-forescein/Anti-Digoxigenin  
20 min at room temp



Buffer wash

Super Enhancer™ (100ul)  
20min at room temp



Buffer wash

Polymer HRP (100ul)  
30min at room temp



Buffer wash

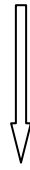
DAB (To be freshly prepared)  
1 drop of liquid DAB chromogen in 1ml of Stable DAB buffer  
Add 100ul to the tissue and incubate for 2-3 min at room temp



Buffer wash

Rinse with DI water

Hematoxylin counter stain (100ul)  
3 min at room temp



Buffer wash

Rinse with DI water

### Clearing and Mounting

70% alcohol - 5min

90% alcohol - 5min

100% Alcohol - 5min

Xylene I - 10min

Xylene II - 10min