

## NB600-450 Protocol

### Immunohistochemistry Protocol for Collagen I Antibody (NB600-450)

[[URL:[https://www.novusbio.com/products/collagen-i-alpha-1-antibody-col-1\\_nb600...](https://www.novusbio.com/products/collagen-i-alpha-1-antibody-col-1_nb600...)]][[Caption:Collagen I Antibody]]

#### Sample

Human Tissue sections (Normal Skin and Basal Cell Carcinoma/Solar Elastosis)

#### Application

Immunohistochemistry (Formalin-fixed paraffin-embedded sections)

#### Blocking step:

None

#### Antigen retrieval step:

Heat mediated

#### Incubation time:

30 minute(s)

#### Dilution: 1/800

#### Additional notes

The tissue was FFPE (formalin fixed, paraffin embedded tissue sectioned at 3-4 microns). The antigen retrieval method was a heat-induced antigen retrieval (HEIR) using a vegetable steamer. Sections were incubated in the steamer for 20 minutes in 1 mM EDTA buffer, pH 8.0. Sections were then rinsed in H<sub>2</sub>O and TBS buffer prior to starting the staining protocol.

The staining appeared to be very specific and robust, with very little to no non-specific background staining.

#### Staining Summary & Notes:

1mM EDTA Buffer, pH 8.0, HIER for 20 minutes in steamer

Rinse with TBS containing 0.05% Tween 20

10 minute blocking in 3% Peroxide

Rinse with TBS containing 0.05% Tween 20

30 minutes in Primary Antibody @ RT

Rinse with TBS containing 0.05% Tween 20

30 minutes in Secondary Antibody @ RT

Rinse with TBS containing 0.05% Tween 20

10 minutes in DAB+ (DakoCytomation) @ RT

Water Rinse

Counterstain for 6 minutes in Mayer's Hematoxylin (DakoCytomation) @ RT

#### Secondary antibody

##### Name:

DakoCytomation Envision+ Anti-Mouse/HRP (RTU)

Conjugation: HRP

Dilution: 1/1