Eastern Health Pathology Immunohistochemistry Laboratory strives to provide antibody testing that is effective and appropriate for the various types of specimens.

**POLICY**

It is important that all new antibodies be validated prior to being
offered for clinical use, in order to document the specificity and sensitivity of the antibody being tested.

Scope

Laboratory technologists working in the IHC lab

Purpose

To ensure that optimal staining is achieved for IHC procedures.

Procedure

Validation Procedure:

1. Obtain a hard copy of the manufacturer’s protocol for performance of the antibody test, and as well a copy of the currently used protocol if such protocol exists.

2. Enter protocol in the Ventana benchmark immunostainer. This protocol should be the currently used protocol if any, as well as the manufacturer’s recommended protocol, both with and without antigen retrieval. Antigen retrieval should be that recommended by the manufacturer, if any. If no antigen retrieval is recommended by the manufacturer, mildest antigen retrieval should be performed first. As well, protocols should use both the ultraview and lview detection kits. This will result in a total of eight (8) slides: Four (4) using the currently run protocol, one (1) having the antigen retrieval, the other one not being subjected to antigen retrieval and each of these stained with both the ultraview and lview detection kits. Four (4) similar slides will be created using the manufacturer’s protocol. These new protocols should be printed out in hard copy.

3. Enter data into the antibody validation document.
4. Preliminary validation slides should be reviewed by the Medical Director and Technical Advisor to determine adequacy of staining. These results should be entered in the validation sheet.

If an appropriate antibody protocol is found using this procedure, the validation process then proceeds to the testing of multiple knowns. If, however, adequate staining is not obtained, antigen retrieval, antibody incubation time, and detection kit can be varied at the discretion of the Technical Advisor and Medical Director, as necessary.

5. Once protocol has been determined as adequate, this protocol must be performed against ten (10) cases of known antigenicity. These are reviewed by the Medical Director and the Technical Advisor, results recorded in the antibody validation sheet, and when the results are satisfactory, the antigen can be released for clinical use.

Supporting Documents


Linkages

410-JPCO-520
PRC-PAT(IHC) - Antibody Protocol
### Definitions & Acronyms

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### Policy History

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