

**Novel rapid- immunohistochemistry using an alternating current electric field for  
intraoperative diagnosis of sentinel lymph nodes in breast cancer**

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**Abstract**

Axillary lymph node status and pathological diagnosis of sentinel lymph nodes (SLNs) is a prognostic factor that influences management of postoperative therapy. Recent reports indicate that one-step nucleic acid amplification and hematoxylin and eosin (HE)-stained frozen sections are effective for intraoperative diagnosis of SLNs. In the present study, we report a rapid-immunohistochemical staining (R-IHC) method that enables intraoperative detection of SLN metastases within 16 min using an anti-cytokeratin antibody.

With this R-IHC system, we apply a high-voltage, low-frequency AC electric field to lymph node sections while they are incubating with the antibodies. The antibodies are mixed within microdroplets and the opportunity for contact between the antibody and antigen is increased. This greatly reduces the time required for the antigen-antibody reaction. This is the first report on SLN diagnosis using R-IHC in patients with breast cancer. We prospectively examined 160 dissected SLNs from 108 breast cancer patients who underwent surgery at our institute. The dissected SLNs were sectioned and conventionally stained with HE or immunohistochemically labeled with anti-cytokeratin antibody using R-IHC procedures. Intraoperative R-IHC analyses were completed within 16 min, after which diagnoses were made by two pathologists. The total time required for intraoperative diagnosis was about 20 min. In this study series, R-IHC detected four metastatic SLNs that were undetected using conventional HE staining (4/20, 20.0%). Compared with subsequent permanent diagnosis, R-IHC offered 95.2% sensitivity and 100% specificity. These findings indicate R-IHC is a clinically applicable technique that enables precise and quick intraoperative detection of micro- and macrometastasis in breast cancer.