Monoclonal Mouse Anti-Human Cytokeratin 19
Clone BA17
Code No. M 0772
Lot 089. Edition 15.11.01

Intended use
For in vitro diagnostic use.
DAKO Monoclonal Mouse Anti-Human Cytokeratin 19, Clone BA17, is intended for use in immuno- cytochemistry. The antibody labels epithelial cells in those epithelia expressing the cytokeratin 19-protein, and hence the antibody is a useful tool for the differentiation and identification of epithelial tumours, and in particularly, it may be useful in identifying ductal and glandular neoplastic epithelia (1). Differential identification is aided by the results from a panel of antibodies. Interpretation must be made within the context of the patient’s clinical history and other diagnostic tests by a qualified pathologist.

Introduction
Cytokeratin 19 belongs to the intermediate filaments, which create a cytoskeleton in almost all cells. In contrast to other intermediate filaments, cytokeratins (CKs) are made up of a highly complex multigene family of polypeptides, with molecular masses ranging from 40 to 68 kDa. Until now, 20 distinct CK polypeptides (2), have been revealed in various human epithelial cells (3). They can be divided into an acidic (type I) and a neutral-basic (type II) subfamily. CK19 belongs to the acidic type of cytokeratins, and is a low molecular mass cytokeratin, at 40 kDa, typically expressed in simple epithelia, transitional epithelium as well as a few complex epithelia, e.g. basal and myoepithelial cells, but not in stratified squamous epithelia (3).

Reagent provided
Monoclonal mouse antibody provided in liquid form as cell culture supernatant dialysed against 0.05 mol/L Tris/HCl, pH 7.2, and containing 15 mmol/L NaN3. Package size is 1 mL.
Clone: BA17 (4). Isotype: IgG1, kappa.
Mouse IgG concentration: 80 mg/L. Total protein concentration: 13.9 g/L.

Immunogen
Detergent-insoluble extract of organoids prepared from the human mammary gland (4).

Specificity
In Western blotting of intermediate filament preparations from breast cancer cells (BT20) and from organoids of human mammary epithelium, the antibody labels a single band of 40 kDa corresponding to cytokeratin 19 (4).
In immunocytochemistry, on human cultured cell lines, the epitope recognized by the antibody is less detectable compared with fixed, paraffin-embedded tissue sections (1, 5). The antibody labels BT20, BT474, MCF-7, T47D, MDA MB-231, MDA MB-157, CAMA-1, ZR75.1, PMc42, milk cells, SVK14, HT29, T24 and PC/AA. No labelling was observed in cell lines FR2, FR5, HBL-100, N13F, A431, HeLa, L1168, U-2-OS, U-118-MG, LT, MRC5 and LEP (1).

Precautions
1. For in vitro diagnostic use.
2. The NaN3 used as a preservative is toxic if ingested. NaN3 may react with lead and copper plumbing to form highly explosive metal compounds. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing.

Storage
Store at 2-8 °C. Do not use after expiration date stamped on vial. If reagents are stored under any conditions other than those specified, the user must verify the conditions. There are no obvious signs to indicate instability of this product. Therefore, positive and negative controls should be run simultaneously with patient specimens. If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the antibody is suspected, contact DAKO Technical Services.

Specimen preparation
Paraffin sections: The antibody can be used for on paraffin-embedded tissue sections fixed in formalin or methacarn (5, 6). Pre-treatment of tissues with proteinase K or heat-induced epitope retrieval is required. For heat-induced epitope retrieval, the following solutions were found efficient: 10 mmol/L Tris buffer, 1 mmol/L EDTA, pH 9.0; or DAKO Target Retrieval Solution, High pH, code No. S3308. 10 mmol/L citrate buffer, pH 6.0, and DAKO Target Retrieval Solution, code No. S 1700, were found less efficient. The tissue sections should not dry out during the treatment or during the following immunocytochemical staining procedure.
Frozen sections and cell preparations: The antibody can be used on methanol/acetone fixed frozen sections (4).

Staining procedure
Dilution: DAKO Monoclonal Mouse Anti-Human Cytokeratin 19, code No. M 0772, may be used at a dilution range of 1:50-1:100 when applied on formalin-fixed, paraffin-embedded sections of human breast carcinoma and using 5 minutes proteolytic epitope retrieval with proteinase K, and 30 minutes incubation at room temperature with the primary antibody. Optimal conditions may vary depending on specimen and preparation method, and should be determined by each individual laboratory. As negative control, DAKO Mouse IgG1, code No. X 0931, diluted to the same mouse IgG concentration as the primary antibody, is recommended.
Visualization: DAKO LSAB®+/HRP kit, code No. K 0679, and the DAKO EnVision™+/HRP kits, code Nos. K 4004 and K 4006, are recommended. For frozen sections and cell preparations, the DAKO APAAP kit, code No. K 0670, is a good alternative if endogenous peroxidase staining is a concern. Follow the procedure enclosed with the selected visualization kit.

Automation: The antibody is well-suited for immunocytochemical staining using automated platforms, such as the DAKO Autostainer.

Performance characteristics

Cells labelled by the antibody display a cytoplasmic staining pattern.

Normal tissues: The following cells and tissues are labelled by the antibody: hair follicles, secretory cells of sweat glands, Merkel cells, luminal epithelial cells of breast ducts, surface mucosa and glands of endometrium and endocervix, exocervix, ovary surface mesothelium, Fallopian tube epithelium, cyto- and syncytiotrophoblast cells, amnion, umbilical cord surface epithelium, luminal- and basal cells of prostate, testes rete epithelium, ductuli efferentes, epididymal tubules, Bowman’s capsule, proximal-, distal- and collecting tubules of the kidney, urothel, bile duct- and gall bladder epithelium, squamous epithelium-, taste buds-, secretory glandular cells and glandular ducts of tongue, squamous epithelium- and submucosal glands of esophagus, surface mucosa- and glands of stomach, surface mucosa- and crypts of small- and large intestine, pancreas ducts, secretory- and duct cells of salivary glands, thyroid epithelium, surface mucosa- and glands of trachea, bronchial mucosa and -glands, alveoli, pleura-mesothelium, Hassal’s corpuscles and thymus epithelial cells.

No labelling was observed in interfollicular epidermis, sebaceous glands, myoepithelium of sweat glands and breast, corpus luteum, corpus albicans, Leydig cells, seminiferous tubules, hepatocytes, pancreatic acini, Langerhans islets, adrenal gland, smooth muscle, striated muscle, myocard, cartilage, blood vessels, brain tissue, cerebellum, stroma of all organs, spleen and lymph nodes (6), nor in frozen tissues of normal epidermal keratinocytes (7).

Abnormal tissues: In human breast tumours the antibody labelled: 20/20 (5) and 8/8 (1) fibroadenomas, 18/18 (5) and 5/5 (1) fibrocystic diseases, 7/7 cystosarcoma phylloides, 96/96 (5) and 12/12 (1) infiltrating ductal carcinomas, 14/14 (5) and 5/5 (1) infiltrating lobular carcinomas respectively, 2/2 medullary carcinomas, 21/21 (5) and 11/11 (1) metastases, 1/1 male invasive carcinoma (1), 4/4 Paget’s diseases and 7/7 pure in situ carcinomas, 4/4 Paget’s diseases and 6/6 special types of carcinomas (5). In addition, the antibody labelled 1/1 thyroid adenoma, 5/5 colon-, 2/2 gastric- and 2/2 lung adenocarcinomas, 2/2 ovarian- and 4/4 urinary bladder carcinomas, 2/2 teratomas, 4/4 embryonal carcinomas and 8/8 tumours of more than one histological type in testicular tissue (1). In epidermal tumours, the antibody labelled 3/8 squamous- and 6/20 basal cell carcinomas, and 3/5 keratoacanthomas (7). No labelling was observed in 4/4 squamous cell, and 2/2 basal cell carcinomas of skin, 7/7 sarcomas, 6/6 malignant melanomas, 7/7 lymphomas, 5/5 seminomas (1), 5/5 Bowen’s disease and 6/6 clear cell acanthomas (7).

References