

## Uroplakin II

### Concentrated and Prediluted Monoclonal Antibody

Control Number: 902-3051-091117

<b>Catalog Number:</b>	<b>ACR 3051 A, C</b>	<b>APR 3051 AA</b>
<b>Description:</b>	0.1, 1.0 ml, concentrated	6.0 ml, prediluted
<b>Dilution:</b>	1:50-1:100	Ready-to-use
<b>Diluent:</b>	Van Gogh Yellow	N/A

#### Intended Use:

For Research Use Only. Not for use in diagnostic procedures.

#### Summary and Explanation:

Uroplakin II is a 15 kDa protein component of urothelial plaques, which enhance the permeability barrier of the urothelium (1). Studies have shown Uroplakin II mRNA was expressed in bladder cancer tissues and peripheral blood of patients with primary and metastatic urothelial carcinoma of the bladder (2-4). A new mouse monoclonal Uroplakin II antibody [BC21] was developed and exhibited an increased staining sensitivity (46/59, 78%) when compared to Uroplakin III [AU1] (19/56, 34%) in cases of urothelial carcinoma of the bladder (see Performance Characteristics). With the exception of bladder and ureter, Uroplakin II [BC21] was found to be highly specific when evaluated in various normal and neoplastic tissues, including prostate cancer and renal cell carcinoma (see Performance Characteristics). Uroplakin II [BC21] is a highly specific antibody that may be useful in identifying tumors of urothelial origin.

U.S. Patent 9,429,577 and patents pending.

#### Principle of Procedure:

Antigen detection in tissues and cells is a multi-step immunohistochemical process. The initial step binds the primary antibody to its specific epitope. After labeling the antigen with a primary antibody, a secondary antibody is added to bind to the primary antibody. An enzyme label is then added to bind to the secondary antibody; this detection of the bound antibody is evidenced by a colorimetric reaction.

**Source:** Mouse monoclonal

**Species Reactivity:** Human; others not tested

**Clone:** BC21

**Isotype:** IgG1/kappa

**Total Protein Concentration:** ~10 mg/ml. Call for lot specific Ig concentration

**Epitope/Antigen:** Residues 36-50 of human Uroplakin II

**Cellular Localization:** Cytoplasmic and membrane

**Positive Tissue Control:** Normal bladder or urothelial carcinoma of the bladder

#### Known Applications:

Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

**Supplied As:** Buffer with protein carrier and preservative

#### Storage and Stability:

Store at 2°C to 8°C. Do not use after expiration date printed on vial. If reagents are stored under conditions other than those specified in the package insert, they must be verified by the user. Diluted reagents should be used promptly; any remaining reagent should be stored at 2°C to 8°C.

#### Staining Protocol Recommendations:

##### Peroxide Block:

Block for 5 minutes with Biocare's Peroxidized 1.

**Pretreatment:** Perform heat retrieval using Biocare's Reveal Decloaker. Refer to the Reveal Decloaker product data sheet for specific instructions.

**Protein Block (Optional):** Incubate for 5-10 minutes at RT with Biocare's Background Punisher.

**Primary Antibody:** Incubate for 30 minutes at RT.

**Probe:** Incubate for 10 minutes at RT with a secondary probe.

**Polymer:** Incubate for 10-20 minutes at RT with a tertiary polymer.

#### Staining Protocol Recommendations Cont'd:

##### Chromogen:

Incubate for 5 minutes at RT with Biocare's DAB – OR – Incubate for 5-7 minutes at RT with Biocare's Warp Red

##### Counterstain:

Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha's Bluing Solution for 1 minute. Rinse with deionized water.

##### Technical Note:

This antibody has been standardized with Biocare's MACH 4 detection system. Use TBS buffer for washing steps.

##### Performance Characteristics:

Sensitivity and specificity testing of Uroplakin II [BC21] was performed by staining with MACH 4 Universal HRP-Polymer Detection. See Tables 1 and 2 for expected results.

##### Limitations:

This product is provided for Research Use Only (RUO) and is not for use in diagnostic procedures. Suitability for specific applications may vary and it is the responsibility of the end user to determine the appropriate application for its use.

##### Precautions:

1. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous materials according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaN<sub>3</sub>) used as a preservative is toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing. (Center for Disease Control, 1976, National Institute of Occupational Safety and Health, 1976) (5)
2. Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come in contact with sensitive areas, wash with copious amounts of water. (6)
3. Microbial contamination of reagents may result in an increase in nonspecific staining.
4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.
5. Do not use reagent after the expiration date printed on the vial.
6. The SDS is available upon request and is located at <http://biocare.net>.

##### Technical Support:

Contact Biocare's Technical Support at 1-800-542-2002 for questions regarding this product.

##### References:

1. Wu XR, *et al.* Uroplakins in urothelial biology, function, and disease. *Kidney Int.* 2009 Jun; 75(11):1153-65.
2. Wu X, *et al.* Uroplakin II as a promising marker for molecular diagnosis of nodal metastases from bladder cancer: comparison with cytokeratin 20. *J Urol.* 2005 Dec; 174(6):2138-42.
3. Lu JJ, *et al.* Detection of circulating cancer cells by reverse transcription-polymerase chain reaction for uroplakin II in peripheral blood of patients with urothelial cancer. *Clin Cancer Res.* 2000 Aug; 6(8):3166-71.
4. Li SM, *et al.* Detection of circulating uroplakin-positive cells in patients with transitional cell carcinoma of the bladder. *J Urol.* 1999 Sep; 162(3 Pt 1):931-5.
5. Center for Disease Control Manual. Guide: Safety Management, NO. CDC-22, Atlanta, GA. April 30, 1976 "Decontamination of Laboratory Sink Drains to Remove Azide Salts."
6. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline-Fourth Edition CLSI document M29-A4 Wayne, PA 2014.

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**Table 1:** Specificity of mouse monoclonal antibody Uroplakin II [BC21] was determined by testing formalin-fixed, paraffin-embedded normal tissues.

Tissue	Number Positive/ Total Tissues
Adrenal gland	0/3
Bladder	5/7
Bone marrow	0/1
Eye	0/2
Breast	0/3
Cerebellum	0/3
Cerebral cortex	0/3
Fallopian tube	0/3
GI-Esophagus	0/3
GI-Stomach	0/3
GI-Small intestine	0/3
GI-Colon	0/3
GI-Rectum	0/3
Heart	0/3
Kidney	0/16
Liver	0/5
Lung	0/3
Ovary	0/3
Pancreas	0/5
Parathyroid	0/1
Pituitary gland	0/2
Placenta	0/3
Prostate	0/5
Skin	0/2
Spinal cord	0/2
Spleen	0/2
Striated muscle	0/4
Testis	0/3
Thymus	0/3
Thyroid	0/4
Tonsil	0/3
Ureter	3/3
Uterus-cervix	0/3
Uterus-endometrium	0/3
Tongue	0/1
Epiglottis	0/1
Blood vessel and adipose tissue	0/1

**Table 2:** Sensitivity of mouse monoclonal antibody Uroplakin II [BC21] was determined by testing formalin-fixed, paraffin-embedded neoplastic tissues.

Tissue Types	Number Positive/Number of Cases
Bladder cancer	46/59*
Prostate cancer	1/88**
Lung cancer	0/20
Kidney cancer (various phenotypes)	3/75***
Colon cancer	0/63
Brain cancer	0/13
Lymphoma	0/25
Melanoma	0/19
Ovarian cancer	0/11
Seminoma	0/14
Breast cancer	0/74
Adrenal gland cancer	0/2
Thyroid cancer	0/2
Pancreas cancer (various phenotypes)	0/10
Head & neck cancer (various phenotypes)	0/10
Soft tissue cancer (various phenotypes)	0/10
Liver cancer (various phenotype)	0/10
Cervix cancer (various phenotypes)	0/10

\* For comparison, Uroplakin III [Clone AU1] stained 19/56 cases of bladder cancer.  
 \*\* 1 positive case, which may be metastatic bladder cancer that has spread to prostate  
 \*\*\* 3 positive cases, which are transitional cell carcinomas from upper ureters