Rabbit monoclonal antibody against

Cytokeratin 8 (QR112)

In Vitro Diagnostic Use (IVD)

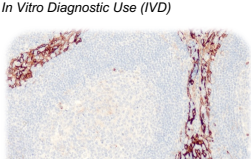

Figure 1 Tonsil stained with anti-Cytokeratin 8 (QR112)

Product identification

C-C040-025	25 µl	Concentrate
C-C040-01	0.1 ml	Concentrate
C-C040-05	0.5 ml	Concentrate
C-C040-10	1 ml	Concentrate
P-C040-30	3 ml	Ready-to-use
P-C040-70	7 ml	Ready-to-use
P-C040-150	15 ml	Ready-to-use

Intended use

Anti-human antibody for *in vitro* diagnostic use. The primary antibody is intended for the qualitative detection of associated antigens as listed in the section 'Summary and explanation'. It is intended to be used within an immunohistochemistry (IHC) procedure on formalin-fixed, paraffin-embedded (FFPE) tissue sections followed by light microscopy visualization to aid tumor diagnosis. The antibody may be used manually or with any automated staining platform.
Authorized and skilled personnel may only use the product. The clinical interpretation of any test results should be evaluated within the context of the patient's medical history and other diagnostic laboratory test results. A qualified pathologist must perform evaluation.

Summary and explanation

Keratins are cytoplasmic intermediate filament proteins expressed by epithelial cells. Cytokeratin 8 is a type II keratin that is expressed in early embryogenesis. It is found in simple epithelia in respiratory, gastrointestinal and reproductive tract as well as thyroid. This antibody detects most non-squamous epithelial tumors and adenocarcinomas of the breast, ovary, gastrointestinal tract, thyroid, pancreas, bile duct and salivary glands. It is often co-expressed with cytokeratin 18, and the major keratin pair in simple-type epithelia, as found in the liver, pancreas, and intestine.

Principle of the procedure

The stated primary antibody is suitable for immunohistochemical staining of FFPE tissue sections based on specific antigen-antibody reaction. Using a detection system linked to horseradish peroxidase (HRP) or alkaline phosphatase (AP) the antigen visualization is performed via specific binding of the primary antibody. Secondary antibody is binding to the primary antibody, and the enzyme complex labels this complex. The enzymatic activation of the chromogen results in a visible reaction product at the antigen site. Each step is incubated for a precise time and temperature and requires interposed washing steps. The specimen may then be counterstained. Results are interpreted using a light microscope.

Materials provided

Primary antibody	Anti-Cytokeratin 8 (QR112)
Host	Rabbit
Subclass	IgG
Immunogen	Synthetic peptide of human Cytokeratin 8
Antibody concentrate	Concentrated antibody in TRIS (pH 7.4) with < 1 % sera (bovine, donkey) and < 0.1 % sodium azide
Recommended working dilution range	1:100 – 1:200
Ready-to-use antibody	Prediluted antibody in TRIS (pH 7.4) with < 1 % sera (bovine, donkey) and < 0.1 % sodium azide

Product label shows the specific lot number. Each individual lot is compared and adjusted to a reference lot to ensure a consistent immunohistochemical staining performance from lot-to-lot.

Prediluted antibody is ready-to-use and optimized for staining. No further dilution, reconstitution, mixing, or titration is needed.
Antibody concentrate is optimized for dilution within dilution range using Q Diluent for IHC (Cat. No. AD-001-xxxx). Indicated dilution range should be considered as recommendation and depends on different facts (tissue, fixation, incubation conditions, etc.). Optimum dilution to be determined in user's own system.

Materials required but not provided

- Positive and negative controls
- Microscope slides (positively charged) and cover slips
- Staining jars
- Timer
- Xylene or xylene alternative, e.g. Q Dewax Solution (Cat. No. DW-001-xxxx)
- Ethanol
- Deionized or distilled water
- Heating equipment for tissue pretreatment step
- Antibody diluent, e.g. Q Diluent for IHC (Cat. No. AD-001-xxxx)
- Antigen retrieval reagent, e.g. Q Retrieval Low pH 6.0 (Cat. No. AR-001-0120) or Q Retrieval High pH 9.0 (Cat. No. AR-002-0120)
- Detection system, e.g. PolyQ Stain kits and appropriate chromogen
- Wash buffer: TBS (Cat. No. BU-006-xxxx) or TBS-Tween20 (Cat. No. BU-007-xxxx)
- Blocking reagent
- Hematoxylin
- Mounting medium
- Light microscope

Storage and handling

Store at 2 – 8 °C.

When stored correctly, the antibody is stable up to the expiration date indicated on the vial. This also applies to the shelf life after opening or after dilution of the concentrate by the end user. Do not use after expiration date.
To ensure proper reagent delivery and stability of the antibody, replace the dispenser cap after every use and immediately place the bottle cool in an upright position.

Specimen preparation

Routinely processed, FFPE tissues are suitable for use with this primary antibody. The recommended tissue fixative is 10 % neutral buffered formalin. Variable results may occur as a result of prolonged fixation or special processes such as decalcification of bone marrow preparations. Thickness of tissue sections, which should be placed on positively charged slides, should be 2 – 5 µm. Pretreatment of deparaffinized tissue with heat-induced epitope retrieval (HIER) is recommended. Slides should be stained as soon as possible, as antigenicity of cut tissue sections may diminish over time.
The optimum pretreatment protocol must be determined in user's own system.

Warnings and precautions

1. Authorized and skilled personnel may only use the product.
2. There are no estimated health risks, if the product is used as directed. MSDS is available on request.
3. Product contains sodium azide as preservative. Pure sodium azide is toxic. The concentration of sodium azide in this reagent is < 0.1 % which is not classified as hazardous.
4. As with any product derived from biological sources, proper handling procedures should be used.
5. Do not use reagents after expiration date.
6. Take reasonable precautions when handling reagents. Use protective clothing and gloves.
7. All hazardous materials should be disposed according to guidelines for hazardous waste disposal. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions.
8. Avoid microbial contamination of reagents as it may cause incorrect results.

Staining procedure

Primary antibody has been optimized for use in combination with PolyQ Stain detection systems. The following data are recommendations. Due to variation in tissue fixation and processing, as well as general lab instrument and environmental conditions, it may be necessary to adjust incubation times. The optimum protocol must be determined in user's own system.

Antigen retrieval: HIER; Boil tissue sections in Q Retrieval for approximately 20 min followed by cooling at room temperature (RT) for approximately 20 min.

Incubation of primary antibody for 30 – 60 min at RT.

Staining protocol: Follow the procedure described in the instructions of the used detection system.

Quality control procedures

Positive tissue control
A positive tissue control must be run with every staining procedure performed for monitoring the correct performance of processed tissues and test reagents. Known positive tissue controls should not be utilized as an aid in determining a specific diagnosis of patient sample. If the positive tissue controls fail to demonstrate appropriate positive staining, results with the test specimens must be considered invalid.

Example for positive tissue control:
- Liver

Negative tissue control
Negative tissue controls provide an indication of non-specific background staining. If specific staining occurs in the negative tissue control sites, results with the patient specimens must be considered invalid.

The variety of cell types present in most tissue sections offers internal negative control sites. Therefore, the same tissue used for the positive tissue control may be used as the negative tissue control.

Discrepancies
If quality control results do not meet specifications, patient results are invalid. Identify and correct the problem (see section "Troubleshooting"), then repeat the entire procedure with the patient samples.

Negative control reagent
A negative control reagent is used in place of the primary antibody to evaluate non-specific staining. Host species and incubation time should be similar to primary antibody.

Interpretation of results

The immunostaining procedure causes a colored reaction product to precipitate at the antigen sites localized by the primary antibody.

Cellular localization: Cytoplasmic.

A qualified pathologist experienced in immunohistochemistry procedures must evaluate positive and negative tissue controls before interpreting patient specimens.
Positive staining intensity should be assessed within the context of any background staining of the negative reagent control.

Note: A negative result means that the antigen in question was not detected, not that the antigen is absent in the cells or tissue assayed. A panel of antibodies may be used to verify the results. Additionally, the morphology of each tissue sample should be examined utilizing a hematoxylin and eosin stained section. A qualified pathologist must interpret the patient's morphologic findings and pertinent clinical data.

Performance characteristics

The antibody passed all analytical performance tests. The antibody is highly specific and highly sensitive. The trueness of the method is confirmed, as the results of the product to be evaluated and the reference antibody/equivalent device match completely. The method has a high level of precision - repeatability within run, reproducibility between run and reproducibility from lot to lot are confirmed. The trueness and precision result in a high level of accuracy of measurement for the method.

Comparison with sources of clinical performance data shows that the antibody stains normal tissues as well as neoplastic tissues as indicated in the literature.

Limitations

1. For *in vitro* diagnostic use.
2. For laboratory use only.
3. This reagent is "for professional use only" as immunohistochemistry is a multiple step process that requires specialized training in the selection of the appropriate reagents, tissues, fixation and processing, preparation of the immunohistochemistry slide, choice of detection system, and interpretation of the staining results.
4. Tissue staining is dependent on the handling, processing and storage of the tissue prior to staining. Improper fixation, freezing, thawing, washing, drying, heating, sectioning, or contamination with other tissues or fluids may produce artifacts, antibody

trapping, or incorrect results. Optimal performance requires adequate specimen quality as well as appropriate sample preparation.

5. Excessive or incomplete counterstaining may compromise proper interpretation of results.
6. False positive results may be seen because of non-immunological binding of proteins or substrate reaction products. They may also be caused by pseudo peroxidase activity (erythrocytes), endogenous biotin (example: liver, brain, kidney) or endogenous peroxidase activity (cytochrome C).
7. When used in blocking steps, normal sera from the same animal source as the secondary antisera may cause false negative or false positive results because of the effect of autoantibodies or natural antibodies.
8. Tissues from persons infected with hepatitis B virus and containing hepatitis B surface antigen may exhibit nonspecific staining with HRP.
9. Unexpected results may occur due to biological variability of antigen expression in neoplasms or other pathological tissues.
10. The clinical interpretation of any test results should be evaluated within the context of the patient's medical history and other diagnostic laboratory test results. Staining must be performed in a certified, licensed laboratory under the supervision of a qualified pathologist who is responsible for evaluation and assuring the adequacy of positive and negative controls. Manufacturer is not liable for incorrect results due to visual evaluation.
11. Prediluted antibodies are ready-to-use and optimized for staining. Further dilution may lead to incorrect results.
12. After successful validation users may dilute antibody concentrates according to requirements. Appropriate controls must be employed and documented.
13. The performance of the product was established using the procedures provided in this package insert only and modifications to these procedures may lead to changes in efficiency. Non-application as prescribed in this data sheet leads to loss of all liability. Any changes in product, composition, implementation, as well as use in combination with any reagents other than recommended herein is not allowed; users are responsible themselves for those changes and have to perform prior validation.
14. Application in combination with diagnostic devices, e.g. an automated staining platform, requires prior validation before staining patient specimen.
15. We do not take responsibility for any possible damage including personal injury, time or effort on economic loss caused by this product. Our warranty is limited to the price paid for the product.

Troubleshooting

1. Only intact cells should be used for interpretation of staining results, as degenerated cells show non-specific staining.
2. If no staining occurs, control application order of reagents. Follow all indications given in the instructions for use.
3. Do not allow the sections to dry out.
4. If weak staining occurs, pay attention during staining steps to freshly prepared chromogen, incubation times and temperatures, as well as accurate draining off of reagents.
5. Avoid surplus background staining by optimal removal of paraffin, washing of slides and dilution of primary antibody. If excessive background staining occurs, high levels of endogenous biotin may be present (unless a biotin-free detection system is being used). A biotin blocking step should be included.
6. Sodium azide inactivates HRP, which may lead to false results. Wash sections in sodium azide free buffer.
7. Contact quartett customer service in case of any uncertainties.

Literature

[1] Franke, W W et al. Diversity of cytokeratins. Differentiation specific expression of cytokeratin polypeptides in epithelial cells and tissues. Journal of molecular biology vol. 153,4 (1981): 933-59.

[2] Angus, B et al. NCL-5D3: a new monoclonal antibody recognizing low molecular weight cytokeratins effective for immunohistochemistry using fixed paraffin-embedded tissue. The Journal of pathology vol. 153,4 (1987): 377-84.

Distributor

quartett Biotechnologie GmbH
Am Mühlenberg 4, 14476 Potsdam, Germany
Tel: +49 (0)30 765 925-0 • Fax: +49 (0)30 765 925-55
service@quartett.com • www.quartett.com

Manufacturer

biocyc Biotechnologie GmbH & Co. KG
Am Mühlenberg 4, 14476 Potsdam, Germany
cert. by TÜV Rheinland LGA Products GmbH
ISO 13485 & ISO 9001
Tel: +49 (0)331 967 826-00

In the event that the user experiences any technical or performance-related issues with the product, please consult the manufacturer or a competent authority.

Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the member state in which the user and/or the patient is established.

Summary of safety and performance (SSP) can be found in EUDAMED when the related module is available.

Date of publication or revision

2023-11-01
Change(s) made: Section 'Distributor'

Explanation of symbols

REF	Bestellnummer Catalog number		Verwendbar bis Use by
LOT	Chargenbezeichnung Batch code		Temperaturbegrenzung Temperature limitation
IVD	In Vitro Diagnostika In vitro diagnostic agent		Gebrauchsanweisung beachten Consult instructions for use
	Hersteller Manufacturer		