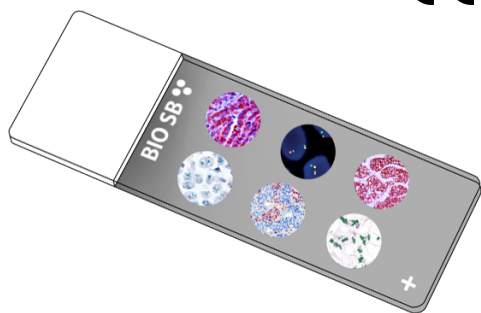


NRAS Control Slides



Intended Use

For In Vitro Diagnostic Use.

Summary and Explanation

NRAS is an enzyme that in humans is encoded by the NRAS gene. The NRAS oncogene is a member of the Ras gene family. It is mapped on chromosome 1. The Ras genes have GTP/GDP binding and GTPase activity, and their normal function may be as G-like regulatory proteins involved in the normal control of cell growth. Mutations which change amino acid residues 12, 13 or 61 activate the potential of N-Ras to transform cultured cells and are implicated in a variety of human tumors.

Tumor mutation profiling has identified the highest incidence of NRAS mutations in melanoma (~30%), acute myeloid leukemia (~15%) and thyroid carcinoma (5-10%). The Q61R is the most common NRAS mutation found in melanoma that is thought to occur due to UV and radiation exposure. The Q61R mutation results in an amino acid substitution at position 61 in NRAS, from a glutamine (Q) to an arginine (R). It is a commonly acquired mutation found in primary melanomas from melanoma-prone families that have known germline CDKN2A mutations, and it is associated with loss of the wild-type NRAS allele. It is also detected in patients with congenital melanocytic nevi, which can develop into malignant melanoma. The NRAS Q61R is the most common oncogenic event in encapsulated thyroid tumors of follicular cell origin with high-grade features. 20% of adrenocortical tumors solely carry this RAS alteration; however, adrenocortical hyperplasia does not, suggesting NRAS Q61R is an oncogenic event driving adrenocortical tumor development.

Presentation

Five slides of NRAS positive tissues, each mounted on Hydrophilic Plus Slides, provided in a plastic mailer.

<i>Catalog No.</i>	<i>Quantity</i>
BSB-9311-CS	5 slides
BSB 2669	5 slides

Storage Store at 20-25°C

Precautions

1. For professional users only. Results should be interpreted by a qualified medical professional.
2. Ensure proper handling procedures are used with this reagent.
3. Always wear personal protective equipment such as a laboratory coat, goggles, and gloves when handling reagents.
4. Dispose of unused solution with copious amounts of water.
5. Follow safety precautions of the heating device used for epitope retrieval (TintoRetriever Pressure Cooker or similar).
8. For additional safety information, refer to Safety Data Sheet for this product.
9. For complete recommendations for handling biological specimens, please refer to the CDC document, "Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories" (see References in this document).

Stability

This product is stable up to the expiration date on the product label.
Do not use after expiration date listed on the package label.

IHC Protocol

1. Subject tissues to heat induced epitope retrieval (HIER) using a suitable retrieval solution such as ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023) or EDTA (BSB 0030-BSB 0033).

2. Any of three heating methods may be used:

a. TintoRetriever Pressure Cooker or Equivalent

Place tissues/slides in a staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA and place on trivet in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high. Incubate for 15 minutes. Open and immediately transfer slides to room temperature.

b. TintoRetriever PT Module or Water Bath Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA at 95°-99° C. Incubate for 30-60 minutes.

c. Conventional Steamer Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA in a steamer, cover and steam for 30-60 minutes.

3. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
4. For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.
5. Wash slides with ImmunoDNA washer or DI water.
6. Continue IHC staining protocol. Wash slides between each step with ImmunoDNA washer solution.

Abbreviated Immunohistochemical Protocol

Step	ImmunoDetector AP/HRP	PolyDetector AP/HRP	PolyDetector Plus HRP
Peroxidase/AP Blocker	5 min.	5 min.	5 min
Primary Antibody	30-60 min.	30-60 min.	30-60 min.
1st Step Detection	10 min.	30-45 min.	15 min.
2nd Step Detection	10 min.	Not Applicable	15 min.
Substrate- Chromogen	5-10 min.	5-10 min.	5-10 min.
Counterstain / Coverslip	Varies	Varies	Varies

Abbreviated IF Protocol

Step	Incubation Time
Rinse slides in IF wash buffer	5 minutes
Drain and wipe excess IF wash buffer off slide	
Conduct remaining steps in the dark	
Apply Antibody	30-60 minutes
Rinse with 3 changes of IF wash buffer	3x15 minutes each
Coverslip with IF mounting medium	

Mounting Protocols

For detailed instructions using biodegradable permanent mounting media such as XyGreen PermaMunter (BSB 0169-0174) or organic solvent based resin such as PermaMunter (BSB 0094-0097), refer to PI0174 or PI0097.

Product Limitations

Due to inherent variability present in immunohistochemical procedures (including fixation time of tissues, dilution factor of antibody, retrieval method utilized, and incubation time), optimal performance should be established through the use of positive and negative controls. Results should be interpreted by a qualified medical professional.

References

1. Eskandarpour M1, et al. Frequency of UV-inducible NRAS mutations in melanomas of patients with germline CDKN2A mutations. J Natl Cancer Inst. 2003 Jun 4;95(11):790-8.
2. Papp T1, et al. Mutational analysis of the N-ras, p53, p16INK4a, CDK4, and MC1R genes in human congenital melanocytic naevi. J Med Genet. 1999 Aug;36(8):610-4.
3. Omholt K1, et al. Screening of N-ras codon 61 mutations in paired primary and metastatic cutaneous melanomas: mutations occur early and persist throughout tumor progression. Clin Cancer Res. 2002; 8(11):3468-74.
4. Nikiforov YE1, et al. Prevalence of mutations of ras and p53 in benign and malignant thyroid tumors from children exposed to radiation after the Chernobyl nuclear accident. Oncogene. 1996 ;13(4):687-93.
5. Rivera M1, et al. Encapsulated thyroid tumors of follicular cell origin with high grade features (high mitotic rate/tumor necrosis): a clinicopathologic and molecular study. Hum Pathol. 2010 ;41(2):172-80.
6. U.S. Department of Health and Human Services: Centers for Disease Control and Prevention. Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories. Supplement / Vol. 61, January 6, 2012. <https://www.cdc.gov/mmwr/pdf/other/su6101.pdf>

Symbol Key / Légende des symboles/Erläuterung der Symbole

EC REP	QAdvis EAR AB Ideon Science Park Scheelevägen 17 SE-223 70 Lund, Sweden	 Storage Temperature Limites de température Zulässiger Temperaturbereich	 Manufacturer Fabricant Hersteller	REF	Catalog Number Référence du catalogue Bestellnummer
IVD	In Vitro Diagnostic Medical Device Dispositif médical de diagnostic in vitro In-Vitro-Diagnostikum	 Read Instructions for Use Consulter les instructions d'utilisation Gebrauchsanweisung beachten	 Expiration Date Utiliser jusqu'à Verwendbar bis	LOT	Lot Number Code du lot Chargenbezeichnung