HANDLEIDING
DIGESTION REAGENTS

product code: R013R.0000
R014R.0000

SNIJDEN EN PLAKKEN VAN PARAFFINE COUPES
PROTEOLYTISCHE VOORBEHANDELING

Beginnen met *HYBRIDISATIE PROCEDURE* en verd er gaan met stringency wash:

1. Incubeer elk preparaat met PanWash 15 min. bij 37°C hete plaat (wit); 5-6 druppels met uitzondering van de positieve controle

Verder gaan met:

DETECTIE EN TEGENKLEURINGSPROCEDURE

VIAL - LABEL

PANWASH GC% 50%

METODICA D'USO
DIGESTION REAGENTS

product code: R013R.0000
R014R.0000

PRETRATTAMENTO DELLE SEZIONI IN PARAFFINA
TRATTAMENTO PROTEOLITICO

Start with *PROCEDIMENTO DI IBRIDAZIONE* and proceed with stringency wash:

1. Aggiungere 5-6 gocce di PanWash 15 min. su blocco riscaldante a 37°C (bianco) su ogni sezione tranne sul controllo positivo

Continue with:

PROCEDIMENTO DI DETEZIONE E COLORAZIONE

Purchase does not include the right to exploit this product commercially and any commercial use without the explicit authorization of PanPath BV is prohibited.

DATA SHEET-V4 WASH BUFFERS

Product **PanWash (GC < 50%); PanWash (GC = 50%)**
Code **R013R.0000, R014R.0000**

Technical specifications

Cat. No.	Description	Contents	Format	Appearance	pH
R013R.0000	PanWash: stringency wash solution for probes with GC % < 50% (PANWASH GC < 50%)	15 mL (50 tests) SSC; clear vial, white cap; contains formamid	Ready to use	Clear liquid	7.0±0.3
R014R.0000	PanWash: stringency wash solution for probes with GC % = 50% (PANWASH = 50%)	15 mL, (50 tests) SSC; clear vial, white cap; contains formamid	Ready to use	Clear liquid	7.0±0.3

Application : - PanWash: stringency wash solution to eliminate non-specific bound probe for use in *in situ* hybridisation (ISH)
R013R.0000 for probes with GC % < 50%
R014R.0000 for probes with GC % = 50%
- TBS buffer: rinsing solution for use in *in situ* hybridisation (ISH)
Storage : refrigerated (2-8 °C) protected from light; do not freeze
Stability : until expiry date printed on label
Precautions : - homogenise solutions before use
- harmful; avoid contact with eyes and skin; do not swallow

Related products
Please contact your local supplier for further information.

Limitations of Procedure

Product Wash buffers

- The REMBRANDT® Wash buffers are solely applicable in the stringency- and rinsing procedure steps in ISH.
- Appropriate medical decisions are only possible if the medical traceability is ensured. The product is intended for professional use as an aid in the diagnosis by *in situ* hybridisation.
- Either human tissue sections or human cytological preparations may be used. Samples must be fixed in buffered formalin or alcohol. Sections should be cut at 4 um thickness, glued to the glass slides with a bio-adhesive (e.g. organosilane), dried at room temperature, subsequently dried at 37 °C overnight, complete deparaffinisation in xylene and alcohol series and air dried. Cytological specimen should be prepared as required by the user, fixed with cytological fixation agent, rinsed in distilled water prior to the ISH procedure and air dried.
- Many factors can influence the performance of the ISH procedure. Failure in detection can be due to i.e. improper sampling, handling, the time lapse between tissue removal and fixation, the size of the tissue specimen in the fixation medium, the fixation time, processing fixed tissue, the thickness of the section, the bio-adhesive on the slide, deparaffinisation procedure, incubation times, incubation temperatures, all other reagents (i.e. deparaffinisation reagents, proteolytic treatment, probes etc.) used in the procedure and interpretation of results.
- The performance of the ISH procedure is also affected by the sensitivity of the method and the DNA or RNA target load; in case the limit of the sensitivity is reached or when the target DNA or RNA load is too low, a false negative reaction may be the result.
- The clinical interpretation of the results should not be established on the basis of a single test result. Moreover, diagnosis should also take the clinical history, symptoms, as well as morphological data into consideration. Negative results therefore do not rule out any possibility of a positive specimen.
- The REMBRANDT® test results are not to be relied on in case the sampling, sampling method, quality, sample preparation, other reagents used, controls and procedure followed is not optimal.
- Therapeutic considerations based on the result of this test alone should not been taken. Positive results should be verified by other traditional diagnostic methods such as but not limited to clinical history, symptoms, as well as morphological data.
- The medical profession should be aware of risks and factors influencing the intensity, the absence or presence of probe signals which can not be foreseen when applying this test.
- The user should carefully consider the risk and use of sample material for this test in case the sample material does not contain sufficient or representative test material.
- Laboratory personnel performing the test should be knowledgeable and be able to interpret the test results.

Interpretation of the results

First, check the negative and positive controls that have been incubated with the test slides simultaneously:
- The negative control should be really negative, i.e. not show any localised colour precipitations. If the negative control could be interpreted as being positive, discard the results since no conclusions can be drawn.
- The positive control should show colour precipitations in conformity with the localisation of the target DNA or RNA. The colour should show the proper shade and must be clearly visible in the preferential cell/ tissue type and correspond to the target localisation.

In the test slides, start under low power magnification and focus on localisation and colour to see whether:
- The positivity (colour precipitation) observed is localised in the cell type preferred by the target.
- The colour has the right shade (no endogenous or formalin pigment).

Use high power magnification to see whether:
- The positive staining texture (granular, etc), demarcation and localisation are conform the positive control staining pattern.

Product in combination with other devices

The REMBRANDT® Wash buffers are intended for stand-alone usage. The in vitro diagnostic is intended to be used in combination with standard formalin fixed, paraffin embedded tissue blocks, standard tissue freezing, tissue sectioning (microtome), standard cytological preparation methods, hot plate(s), stove(s), incubation device(s), water bath(s), temperature and incubation time control(s), other needed reagents (but not supplied with this reagent) for ISH and a microscope. The combination has been tested and validated. Since the standard formalin fixed, paraffin embedded tissue blocks, standard tissue freezing, tissue sectioning (microtome), standard cytological preparation methods, hot plate(s), stove(s), incubation device(s), water bath(s), temperature controls, incubation time control(s) and other needed reagents such as but not limited to proteolytic reagents, labelled probes, detection reagents and a microscope is not combined with the device as a product, conformity with the essential requirements is not applicable. Assay validation criteria are mentioned in *'Interpretation of the results'* and are also depending on the target; since the Wash buffers can be used for all ISH applications and the results also depend on the target load, the validation criteria may vary.

References

1. Keller, G.H. and Manak, M.M., DNA probes, Stockton Press (1989). ISBN 0-935859-63-2.

REFERENCE GUIDE
DIGESTION REAGENTS
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PRETREATMENT OF PARAFFIN SECTIONS
PROTEOLYTIC TREATMENT

Start with *HYBRIDIZATION PROCEDURE* and proceed with stringency wash:

1. Apply 5-6 drops of PanWash (white) 15 min. on 37°C heating block to each specimen except the positive control

Continue with:

DETECTION AND STAINING PROCEDURE

VIAL - LABEL

PANWASH GC%...50%

ANLEITUNG
DIGESTIONS REAGENTS
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HERSTELLUNG VON PARAFFINSCHNITTEN
PROTEOLYTISCHE BEHANDLUNG

Anfangen mit *HYBRIDISIERUNGSPROZEDUR* und verfolge mit stringency wash:

1. 5-6 Tropfen PanWash (Weiss) zu 15 Min. bei 37°C Heizplatte jedem Präparat geben(mit Ausnahme der Positivkontrolle)

Verfolge mit:

DETEKTIONS-UND FÄRBEPROZEDUR

GUIDE RÉFÉRENCE
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PRE-TRAITEMENT DES SECTIONS PARAFFINEES
TRAITEMENT PROTEOLYTIQUE

Commencer avec *PROTOCOLE D'HYBRIDATION* et continuer avec stringency wash :

1. Ajoutez 5-6 gouttes de PanWash (blanc) 15 min. 37°C bloc chauffant à chaque échantillon à l'exception du contrôle positif

Continuer avec:

PROTOCOLE DE DETECTION ET DE COLORATION

VIAL - LABEL

PANWASH GC%...50%

GUIA DE REFERENCIA
DIGESTION REAGENTS
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PRETRATAMIENTO DE LOS CORTES DE PARAFINA
TRATAMIENTO PROTEOLITICO

Commenzar con *PROTOCOLO DE HIBRIDACION* más continuar con stringency wash:

1. Añadir 5-6 gotas de Pan Wash (vial blanco) a cada muestra 15 min. en un termobloque a 37ºC.

Continuar más:

PROTOCOLO DE DETECCION Y TINCION