



# Tissue Microarray (TMA)

## Labelled Strept-Avidin – Biotin Method (LSAB)

### GENERAL NOTE

This Product Application Information is based on standard application protocols, modified on the basis of our own lab experience.

As handling in numerous labs is quite different oligene can not guarantee for the results using this protocol, as well as for mistakes in the protocol. As it does not contain any safety instruction, the operator is responsible to inform about and to follow the safety instructions of the supplier of any material involved.

### HANDLING NOTE

Be aware, that increased mechanical stress during TMA slide treatment may cause dislodging of spots! All wash solutions and reagents should be applied gently to the slide.

## Protocol labelled strept-avidin – biotin method (LSAB)

### Pre-treatment of TMA slide:

- Incubate the slide over night at 37-58°C
- Remove paraffin wax carefully (standard protocols of descending alcohol sequence) and rehydrate

### deparaffination

- 3 x 10 min XyloI
- 2 x 5 min. 100% Ethanol (RT)
- 2 x 5 min. 96% Ethanol (add DEPC-H<sub>2</sub>O)
- 2 x 5 min. 70% Ethanol (add DEPC- H<sub>2</sub>O)
- Aqua dest.

Unmask if necessary (check datasheet of your application kit / antibody specifications)

### implementation

- incubate slide 5 min. at RT in TBS
- swab excrescent wetness and cover the test area with optimal diluted prime antibody
- incubate 30 min. at RT in wet chamber
- wash 3 x 5 min. with TBS
- swab excrescent wetness and cover the test area with optimal diluted biotinylated secondary antibody
- incubate 20 min. at RT in wet chamber
- wash 3 x 5 min. with TBS
- swab excrescent wetness and cover the test area with optimal diluted AP- or HRP-conjugated Streptavidin
- incubate 20 min at RT in wet chamber
- wash 3 x 5 min. with TBS
- apply substrate depending on enzyme conjugation substrate (i.e. DAB or Fuch sine)
- affect substrate 10 min.
- rinse slide with H<sub>2</sub>O
- nucleus staining with Hämatoxylin according to Mayer 5 min.
- blue 5 min. in warm tap water
- seal with an aqueous mounting medium (i.e. glycerine-gelatine)

## For Research Use Only

Not intended for use in diagnostic or therapeutic procedures.



**notice:**

- important: do not let slides run dry!
- decide the blocking options of background depending on used tissue and enzyme-substrate mixture
- using additional kits please follow the company advices

**Storage**

- Store at +4°C in the dark

Orders and Technical Information

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