#### Product specification and information

# IRM 922 - Mouse monoclonal antibody Hb5-11 (Clone 10-011)

1. Name of materialMouse monoclonal antibody Hb5-11 (Clone 10-011)2. IRM NumberIRM 9223. Organization preparing the materialIcosagen AS (former Quattromed), Estonia

4. Antibody Specifications

# Table 1 Specification for Hev b 5 monoclonal antibody Hb5-11

Property	ASTM Designation	Limits/Targets
Animal	D-7427, D 5900	Mouse
Antigen	D-7427, D 5900	rMBP-Hevb5
Adjuvant	D-7427, D 5900	Freund's Complete and Incomplete
Subclass		IgG2b
Presentation		Purified MAb in PBS, pH 7,4
Concentration		2 mg/mL
Titer	D-7427, D 5900	Minimum 1/3,000
SDS PAGE Coomassie stain profile	D-7427, D 5900	Two bands detectable at molecular weight approximately 50 kDa and 25 kDa
Capture ELISA reactivity	D-7427, D 5900	Comparative analysis of extracts from a minimum of 5 Natural Rubber products

### 5. Specific conditions under which the IRM is to be stored

The material should be stored as aliquots at  $-20^{\circ}$ C to  $-80^{\circ}$ C. Multiple freeze-thaw cycles should be avoided as this may result in product degradation. The shelf life of the product has not been specifically determined, however, the shelf life has been well established for other mouse monoclonal antibodies. This reagent should have a minimum shelf life 10 years provided that it is stored at  $-20^{\circ}$ C to  $-80^{\circ}$ C. To ensure Mab quality, the antibody should be tested for its reactivity in the IEMA assay every 24 month to ensure that degradation has not occurred.

#### 6. Specific steps before taking IRM into use

Antibody should be conjugated with horseraddish peroxidase (HRP) before using in IEMA assay. Conjugation method is based on antibody modification with S-acetyl-mercapto-succinic anhydride (SAMSA) and HRP modification with Sulfosuccinimidyl-4-(N-maleimidomethyl)cyclohexane-1-carboxylate (Sulfo-SMCC). In this type of conjugation, SAMSA is reacted first with the antibody resulting the generation of free thiol groups, Then excess reagent removed. To ensure success and optimal degree of forming crosslinks, it is important to know that a molecule has the proper degree of thiolation. In the next step Sulfo-SMCC is added to HRP, excess crosslinking reagent removed and the modified antibody is added. This two-step reaction scheme results in formation of specific antibody-enzyme conjugates. Crosslinked antibody-enzyme conjugate is separated from non-crosslinked material and used as detection antibody in IEMA assay.

# 7. Other information

to verify the specificity and immunoreactivity of the Hev b 5 protein (IRM 923), anti-Hev b 5 capture antibody (IRM 921) and anti-Hev b 5 detection antibody (IRM 922) that are used together to perform the Hev b 5 IEMA.



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