

Product Information

MONOCLONAL ANTI-INVOLUCRIN CLONE SY5 Mouse Ascites Fluid

Product No. I 9018

Product Description

Monoclonal Anti-Involucrin (mouse IgG1 isotype) is derived from the SY5 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with purified human involucrin.¹ The isotype is determined using the Sigma ImmunoType™ Kit (Product Code ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Product Code ISO-2).

Monoclonal Anti-Involucrin detects involucrin in a range of primate and non-primate mammals using ELISA, immunoblotting, immunoprecipitation and immunohistochemical staining of cultured cells and tissue sections. In immunoblots of cultured human keratinocytes, the antibody stains a 120 kDa band, under denaturing and reducing conditions.¹ However, in other systems the antibody stains the involucrin protein in a variety of sizes: 170 kDa in human breast adenocarcinoma cell line MCF-7, a doublet of approx. 115 and 150 kDa in gorilla and owl monkey,¹ 66 kDa in dog,¹ and a doublet at approx 105 kDa in pig.¹ The antibody does not react with mouse epidermis, thus enabling the use of the antibody for study of human xenografts in nude mice.¹ The product may be used for immunohistological staining of frozen and formalin-fixed, paraffin-embedded tissue sections. It stains the upper spinous and granular layers in human skin and the cytoplasm of suprabasal terminally differentiated keratinocytes in stratified colonies.¹

The skin serves as a vital barrier that protects the internal environment from external influences. As cells start to migrate out of the basal cell layer of the epidermis towards the surface of the skin, they lose the ability to divide, and progress towards a terminal differentiation stage.² During this process, keratinocytes go through marked morphological and structural changes. At the same time, the synthesis of important differentiation-dependent structural and catalytic proteins is initiated, which include keratins, filaggrin, transglutaminase, loricrin and involucrin. Involucrin is a soluble cytoplasmic protein precursor of the epidermal cornified envelope, that becomes cross-

linked by transglutaminase during envelope assembly. Involucrin is expressed in a range of stratified squamous epithelia, including the cornea, which lacks a distinct cornified layer.¹ The involucrin molecule has a polymorphic form; in gel permeation chromatography under non-denaturing condition it has a size of approx. 350 kDa.² However, in immunoblotting, under denaturing and reducing conditions, a 120-170 kDa protein is reported for human keratinocytes,¹⁻³ while a variety of sizes in the range of 66-150 kDa is reported for other species.¹ Involucrin is a marker of keratinocyte terminal differentiation and is expressed in the suprabasal layers of stratified squamous epithelium.^{3,4} It is first expressed in the upper spinous layers *in vivo* and immediately above the basal layer *in vitro*. It is cross-linked in the cornified layer during the construction of the cornified envelope, of which it is a major component. Epithelia that do not possess a stratum corneum continue to express involucrin. This suggests that involucrin may have an additional function other than the formation of the cornified envelope, a theory that is supported by the observation that even in cornified epithelia not all the involucrin synthesized by the keratinocyte is incorporated into the cornified envelope. Involucrin is normally expressed *in vivo* when the cells have lost the ability to divide.³ In pathological conditions, involucrin expression may be altered; in psoriasis and other benign epidermal hyperplasias involucrin expression begins closer to the basal layer than normal expression. It is increased abnormally in squamous cell carcinomas and premalignant lesions and is reduced in severe dysplasias of the larynx and cervix.^{1,5} Antibodies reacting specifically with involucrin may be used to detect involucrin both biochemically and histologically in a range of species in order to further study the expression, function and evolution of the protein.

Monoclonal Anti-Involucrin may be used for the localization of involucrin using various immunochemical assays such as ELISA, immunoblotting, immunocytochemistry, immunohistochemistry, and immunoprecipitation.

Reagents

The product is provided as ascites fluid with 0.1% sodium azide as a preservative.

Precautions and Disclaimer

Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

Product Profile

Minimum 1:100

The minimum antibody titer of 1:100 was determined by indirect immunofluorescent labeling of formalin-fixed, paraffin-embedded sections of human skin.

In order to obtain best results in different techniques or preparations, it is recommended that each individual user determine their optimal working dilutions by titration assay.

Storage/Stability

For continuous use, store at 2-8 °C for a maximum of one month. For extended storage, the solution may be frozen in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

References

1. Hudson, D., et al., Hybridoma, **11**, 367 (1992).
2. Van Duijnhoven, et al., Arch. Dermatol. Res., **284**, 167 (1992).
3. Griffin, E., and Harris, H., J. Cell Sci., **102**, 799 (1992).
4. Carroll, J., et al., Proc. Natl. Acad. Sci. (USA), **90**, 10270 (1993).
5. Rice, R., et al., "Involucrin: Biochemistry and Immunohistochemistry" in: Advances in Immunohistochemistry, Delellis, R. (ed.), pp. 111, Masson Publish., New York (1984).

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