HIF1a | ***Validation File***

TARGET HIF1a
CLONE NAME SIMA343B
DESCRIPTION rat monoclonal
ANTIGEN USED mouse HIS-HIF1a (fragment 530-825)
ISOTYPE IgG2a
SPECIES REACTIVITY mouse and human
LOCALIZATION nuclei
POSITIVE CONTROL placenta
STORAGE BUFFER Tissue culture supernatant: 0.02% sodium azide
Purified antibody: PBS plus 1%BSA and 0.02% sodium azide. MAb concentration: 1mg/ml
STORAGE Aliquot and store at 4C. Do not freeze

Recommended Inconclusive Not Recommended Not Tested

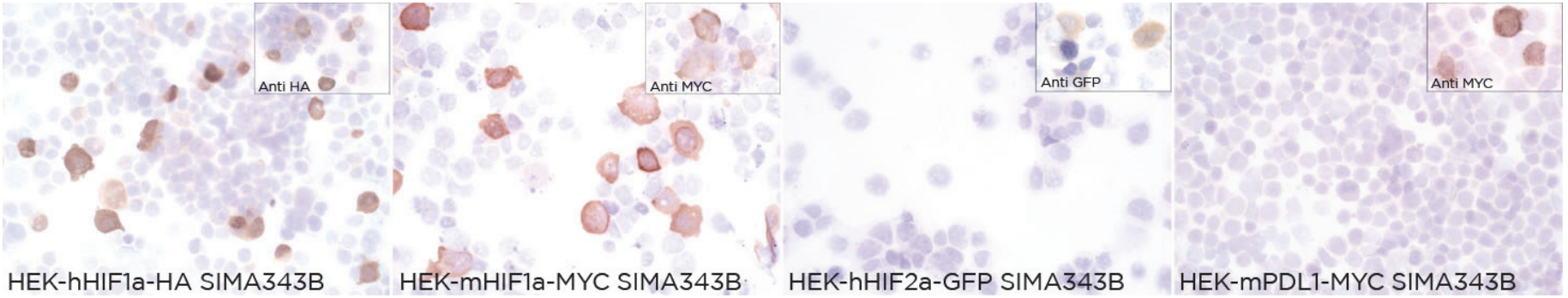
APPLICATIONS

| ICC | ***Immunocytochemistry***

SIMA343B mAb is able to detect human and mouse HIF1a protein in immunocytochemistry

DILUTION no dilution (neat supernatant)
1:200 purified antibody (1mg/ml)

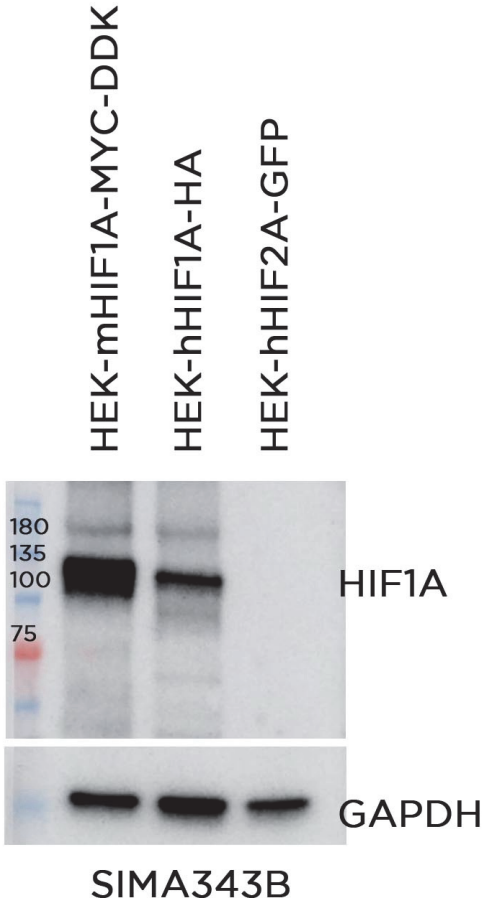
To confirm that SIMA343B mAb recognizes human and mouse HIF1a protein, immunocytochemistry on frozen cytospins preparation of human and mouse HIF1a and hHIF2a expressed in HEK293T was performed. Anti-MYC, anti-HA and anti GFP Abs were used as positive controls. Cytospin preparation of mouse PDL1 protein was used as negative control.



| WB | ***Western Blotting***

SIMA343B mAb is able to detect human Hif1a protein by WB (over-expression).

DILUTION no dilution (neat supernatant)
1:200 purified antibody (1mg/ml)



Predicted molecular weight: ***93kDa***
Observed molecular weight: ***>100kDa***

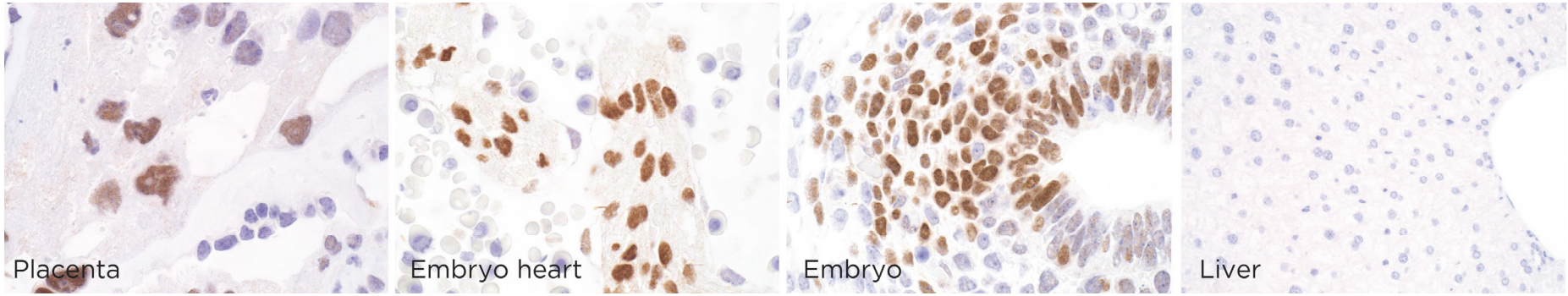
LANES

Lane 1 HEK-mouse HIF1A-MYC-DDK (10ug) (+)
Lane 2 HEK-human HIF1A-HA (10ug) (+)
Lane 3 HEK-human HIF2a-GFP (10ug) (-)

| M-IHC-P | ***Immunohistochemistry (paraffin)***

SIMA343B mAb is able to detect mouse HIF1a protein in mouse paraffin tissues

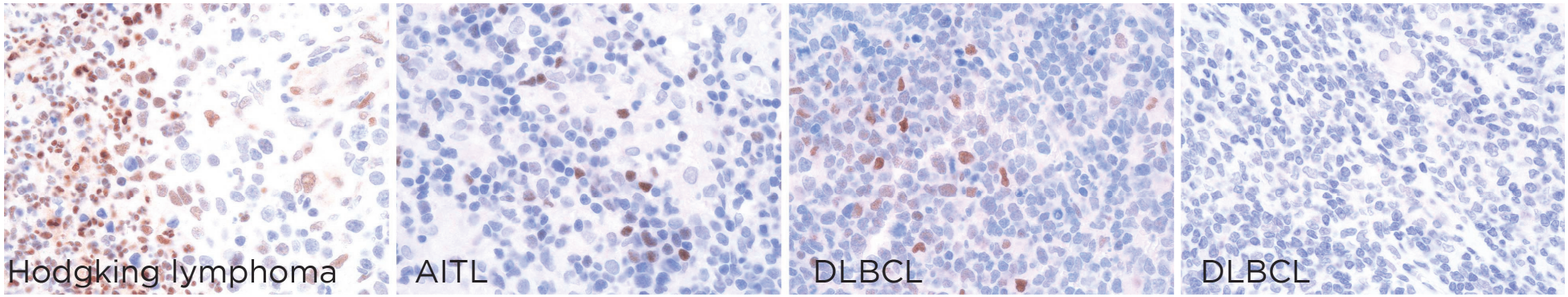
TISSUE SAMPLE mouse placenta, embryo heart, embryo and liver
DILUTION 1:30 (supernatant) and 1:1500 (purified)
ANT. RETRIEVAL High pH9 95ºC
Ab INCUBATION 20 minutes, Rabbit anti-rat Vector + Novolink Polymer
DETECTION SYSTEM Autostainer Link® (Dako, Agilent)



|H-IHC-P | ***Immunohistochemistry (paraffin)***

SIMA343B antibody can be used to detect human HIF1a protein in human paraffin tissues.

TISSUE SAMPLE human Hodgkin lymphoma, AITL and DLBCL tissues.
D***ILUTION*** no dilution (neat supernatant)
1:200 purified antibody (1mg/ml)
ANT. RETRIEVAL 20 minutes ER2 (Tris-EDTA)
DETECTION SYSTEM Novolink kit (BondMax Leica)



| IP | ***Immunoprecipitation*** Not tested

| IHC-F | ***Immunohistochemistry (frozen)*** Not tested

| IF | ***Immunofluorescence (paraffin)*** Not working

| FC | ***Flow Cytometry*** Not tested

REFERENCES

Metformin reduces macrophage HIF1 -dependent proinflammatory signaling to restore brown adipocyte function in vitro. Nuria Pescador, Vera Francisco, Patricia Vázquez, Eva María Esquinas, Cristina González-Páramos, M Pilar Valdecantos, Irma García-Martínez, Andrés A Urrutia, Laura Ruiz, Carmen Escalona-Garrido, Marc Foretz, Benoit Viollet, Miguel Ángel Fernández-Moreno, Alfonso L Calle-Pascual, María Jesús Obregón, Julián Aragonés and Ángela M Valverde. Redox Biol. 2021 Oct 19;48:102171. DOI: 10.1016/j.redox.2021.102171. doi: 10.1016/j.redox.2021.102171.