Mouse Monoclonal Antibody (CAL2) Against All *CALRETICULIN* (*CALR*) Mutations

Product Information

Catalog No.: **DIA-CAL-250** (250µl)
DIA-CAL-100 (100µl)

Clone: CAL2

Isotype: Mouse IgG2a

Immunogen: C-neoterminus of mutated CALR.

Specificity: Human CALR protein expressed by all types of Exon 9 *CALR* mutations (deletion/insertion in 19p 13.3-13.2 of)

Application: Immunohistochemistry (IHC) for B5-fixed or formalin-fixed paraffin-embedded (FFPE) tissue with or without EDTA-decalcification

Physical state: Lyophilized powder

Reagent provided: Antibody purified from culture supernatant in PBS with 2% BSA, 0.05% NaN3, pH 7.4.

Storage and stability: Unopened vials for at least 1 year at -20°C; As reconstituted liquid several weeks at 2-8 °C; For long term storage freeze at -20°C or -80°C.
Avoid repeated freeze/thaw cycles.

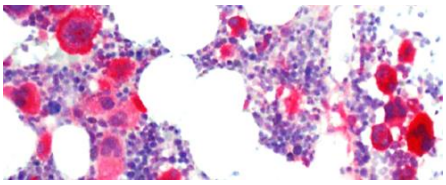
Instructions for use: Reconstitute DIA-CAL-250 with 250 µl and DIA-CAL-100 with 100 µl sterile distilled water followed by gentle shaking for 10 min.
Detection of CALRmut with clone CAL2, Heat-induced epitope retrieval (HIER):
I.
Formalin-fixed and EDTA-decalcified BM:
Citrate-buffer pH6.0, 10min pressure cooker or 30-60min microwave at 98-100°C
II.
B5-fixed and EDTA-decalcified BM:
EDTA-buffer pH8.0, 10min pressure cooker or 30-60min microwave at 98-100°C
III.
B5-fixed and acid-decalcified BM:
EDTA-buffer pH8.0, 10min pressure cooker or EDTA-TRIS-buffer pH9.0, 60min microwave at 98-100°C
After HIER follow standard IHC protocols. The CAL2 antibody IHC is suited for using automated platforms.

Dilution: 1:20-1:40 for IHC

General recommendation: Validation of antibody performance/protocol is the responsibility of the end user. Positive/negative controls should be run simultaneously with tissue specimen.

Practical implementation: CAL2 labels the megakaryocytes in myeloproliferative neoplasms (essential thrombocythaemia (ET) and primary myelofibrosis (PMF)) with *CALR* mutation and enables to distinguish ET and PMF with *CALR* mutation from polycythemia vera (PV), from *CALR* mutation negative ET and PMF and from reactive bone marrow.

Staining pattern: Cytoplasmic staining of megakaryocytes harboring *CALR* mutation. The CAL2 IHC assay indicates absence of *CALR* mutation when all megakaryocytes remain unlabeled.
Formalin-fixed BM:
Simple and fast analysis by a specific and intense staining of megakaryocytes and a variable amount of cells with small nucleus
B5-fixed BM:
Simple and fast analysis by a specific and intense staining of megakaryocytes. For analysis include only megakaryocytes. Sometimes weak non-specific staining of granulopoietic or erythropoietic cells



Positive control: Megakaryocytes from *CALR* mutated PMNs

Negative control: Megakaryocytes of reactive bone marrow specimens or *JAK2* mutated PV

Safety notes: The reconstituted liquid contains 0.05% sodium azide as a preservative. Avoid skin and eye contact, inhalation and ingestion.

References:

1.
Stein, H et al. A new monoclonal antibody (CAL2) detects CALRETICULIN mutations in formalin-fixed and paraffin embedded bone marrow sections. Leukemia 2015, accepted for publication.
2.
Nangalia J et al. Somatic CALR Mutations in Myeloproliferative Neoplasms with Nonmutated JAK2. *N Engl J Med* 369(25): 2391-2405, 2013.
3.
Klampfl T et al. Somatic Mutations of Calreticulin in Myeloproliferative Neoplasms. *N Engl J Med* 369(25): 2379-2390, 2013.

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