

Technical Data Sheet

Biotin Mouse Anti-Human CD178

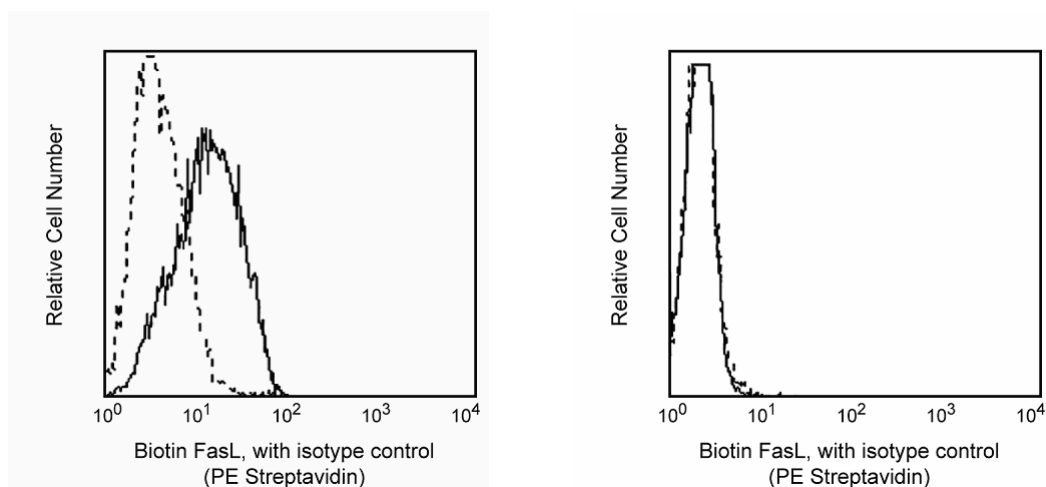
Product Information

Material Number:	556373
Alternate Name:	Fas Ligand; CD95 Ligand
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	NOK-1
Immunogen:	Mouse T lymphoma cells (L5178Y) expressing human FasL
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

Fas (CD95; APO-1) is a 45 kDa cell surface protein that mediates apoptosis when cross-linked with agonistic anti-Fas antibodies or by Fas ligand (FasL; CD178). Fas belongs to the TNF (Tumor Necrosis Factor)/NGF (Nerve Growth Factor) receptor family, and is expressed in various tissues and cells including the thymus, liver, ovary and lung. CD178 (FasL), a member of the TNF cytokine family, induces apoptosis by binding to Fas, its cell-surface receptor. FasL may exist as either membrane bound or soluble forms and is expressed by activated T and NK cells. FasL may also be constitutively expressed in some immunologically privileged sites, e.g., eye and testis. Fas and FasL play an important role in the induction of apoptosis, and thus regulate a variety of immunological responses.

The NOK-1 antibody clone has been reported to recognize human FasL, recognizing both the membrane bound (FasL) and soluble (sFasL) forms. It is reported that the epitope for NOK-1 has been mapped to the COOH-terminus of FasL, at the region implicated in Fas binding. FasL and sFasL have been reported to migrate at reduced molecular weights of 40 and 26 kDa, respectively. However, the molecular weights observed in a particular sample may vary according to FasL and sFasL glycosylation and breakdown patterns as described in the literature. The NOK-1 antibody clone is not recommended for the western blot application.



Flow cytometric analysis of human Fas Ligand (FasL). The mouse T cell lymphoma cell line, L5187Y was transfected with human FasL cDNA and treated with a metalloproteinase inhibitor, KB8301 (left panel). KB8301 blocks FasL cleavage resulting in high levels of cell surface FasL. As expected, FasL was not detected in the parental L5187Y cells after KB8301 treatment (right panel). The data was generated using a 2-step procedure with either biotin-labeled, isotype-matched control antibody (dashed line) or biotin-labeled (NOK-1, Cat. No. 556373) (solid line) followed by Streptavidin-PE.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. The antibody was conjugated with biotin under optimum conditions, and unreacted biotin was removed. Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

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Application Notes

Application

Flow cytometry	Routinely Tested
Western blot	Not Recommended

Recommended Assay Procedure:

Biotin-conjugated NOK-1 (Cat. No. 556376) is recommended for this application (titrate between 0.06 to 1 µg/1x10⁶ cells). Biotin-conjugated NOK-1 is also available in a 100 test size (Cat. No. 556374), and has been optimally titrated for use in flow cytometry. NOK-1 has also been used for immunoprecipitation (see purified format, Cat. No. 556372), and to neutralize the cytotoxic activity of FasL. The NA/LE™ format of NOK-1 (Cat. No. 556371) should be used for all functional assays. NOK-1 and a related human FasL clone, NOK-2 (purified, Cat. No. 556376; BD NA/LE™, Cat. No. 556375) may give different profiles in neutralization assays. It is thought that NOK-1 and NOK-2 recognize different FasL epitopes. Neither NOK-1 and NOK-2 are suggested for western blot analysis. Clone G247-4 (Cat. No. 556387) is suggested for western blot.

Suggested Companion Products

Catalog Number	Name	Size	Clone
555747	Biotin Mouse IgG1 κ Isotype Control	100 tests	MOPC-21

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References

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