

DESCRIPTION

Species Reactivity	Canine
Specificity	Detects canine SCF/c-kit Ligand in ELISAs and Western blots. In sandwich immunoassays, less than 20% cross-reactivity with recombinant feline SCF is observed, less than 8% cross-reactivity with recombinant human SCF is observed, and less than 0.5% cross-reactivity with recombinant mouse SCF is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	<i>E. coli</i> -derived recombinant canine SCF/c-kit Ligand Lys26-Ala190 Accession # Q06220
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Western Blot	0.1 µg/mL	Recombinant Canine SCF/c-kit Ligand (Catalog # 2278-SC)
Immunocytochemistry	5-15 µg/mL	See Below
Canine SCF/c-kit Ligand Sandwich Immunoassay		Reagent
ELISA Capture	0.2-0.8 µg/mL	Canine SCF/c-kit Ligand Antibody (Catalog # AF2278)
ELISA Detection Standard	0.1-0.4 µg/mL	Canine SCF/c-kit Ligand Biotinylated Antibody (Catalog # BAF2278) Recombinant Canine SCF/c-kit Ligand (Catalog # 2278-SC)
Neutralization	Measured by its ability to neutralize SCF/c-kit Ligand-induced proliferation in the TF-1 human erythroleukemic cell line. Kitamura, T. <i>et al.</i> (1989) <i>J. Cell Physiol.</i> 140 :323. The Neutralization Dose (ND ₅₀) is typically 0.1-0.3 µg/mL in the presence of 5 ng/mL Recombinant Canine SCF/c-kit Ligand.	

DATA

Neutralization

Cell Proliferation Induced by SCF/c-kit Ligand and Neutralization by Canine SCF/c-kit Ligand Antibody. Recombinant Canine SCF/c-kit Ligand (Catalog # 2278-SC) stimulates proliferation in the TF-1 human erythroleukemic cell line in a dose-dependent manner (orange line). Proliferation elicited by Recombinant Canine SCF/c-kit Ligand (5 ng/mL) is neutralized (green line) by increasing concentrations of Goat Anti-Canine SCF/c-kit Ligand Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2278). The ND₅₀ is typically 0.1-0.3 µg/mL.

Immunocytochemistry

SCF/c-kit Ligand in Canine PBMCs. SCF/c-kit Ligand was detected in immersion fixed canine peripheral blood mononuclear cells (PBMCs) using Goat Anti-Canine SCF/c-kit Ligand Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2278) at 15 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to plasma membrane. View our protocol for [Fluorescent ICC Staining of Non-adherent Cells](#).

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Canine SCF (stem cell factor; also known as c-kit ligand) is a type I transmembrane (TM) glycoprotein that plays an important role in a number of fetal and adult developmental processes (1-4). It is synthesized as a 274 amino acid (aa) precursor that contains a 25 aa signal sequence, a 190 aa extracellular region, a 23 aa TM segment and a 36 aa cytoplasmic tail (5). Within the extracellular region there are four potential N-linked glycosylation sites, two intrachain disulfide bonds, and four α -helices. Although the predicted molecular weight is 19 kDa, the native molecule is anywhere from 28-40 kDa in size and reflects both N- and O-linked glycosylation (1). Glycosylation is not necessary for bioactivity (6). The transmembrane form of SCF can be cleaved proteolytically, generating a 165 aa soluble form. Circulating SCF exists as both a monomer and nondisulfide-linked homodimer, with monomer predominating (50% to 75%) (6). Both the soluble and TM forms have bioactivity. Their principal targets may be different, however (7). A second, alternate splice short form of SCF has been identified in other species (1). It is membrane bound but lacks the proteolytic cleavage site found in the long form. Thus, it cannot give rise to a soluble molecule. No such isoform has been reported for canine, but it could be assumed to exist. The ratio of long form to short form varies from tissue to tissue (1). Soluble canine SCF shares 88%, 93%, 86%, 83%, 76%, 76%, 86% and 88% aa sequence identity with porcine, feline, bovine, human, mouse, rat, goat and equine SCF, respectively. Cells known to express SCF include endothelial cells, fibroblasts and keratinocytes (1).

References:

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6. Hsu, Y-R. *et al.* (1997) *J. Biol. Chem.* **272**:6406.
7. Kapur, R. *et al.* (1998) *Blood* **91**:879.