

Datasheet for 611-144-003

## Rabbit IgG Fc Antibody DyLight™ 680 Conjugated

### Overview

<b>Description:</b>	Goat Anti-Rabbit IgG Fc Antibody DyLight™ 680 Conjugated - 611-144-003
<b>Item No.:</b>	611-144-003
<b>Size:</b>	100 µg
<b>Applications:</b>	WB
<b>Reactivity:</b>	Rabbit
<b>Host Species:</b>	Goat

### Product Details

<b>Background:</b>	Anti-Rabbit IgG F(c) DyLight generated in goat is a proteolytic fragment of immunoglobulin G (IgG) obtained by limited digestion with the enzyme papain under controlled conditions of temperature, time and pH. Receptors bind the Fc portion of rabbit IgG and often this fragment is removed from immunoglobulins to minimize receptor binding and lower background reactivity.
<b>Synonyms:</b>	Goat Anti Rabbit IgG F(c) DyLight 680™ Conjugated Antibody, Goat Anti-Rabbit IgG Fc Fragment Antibody DyLight 680™ conjugation, Goat Anti Rabbit IgG Fc Antibody DyLight 680™ conjugated
<b>Host Species:</b>	Goat
<b>Specificity:</b>	IgG Fc
<b>Conjugate:</b>	DyLight™ 680
<b>Clonality:</b>	Polyclonal
<b>Format:</b>	IgG
<b>F/P Ratio:</b>	2.8

### Target Details

<b>Reactivity:</b>	Rabbit
<b>Immunogen:</b>	Rabbit IgG F(c) fragment

**Purity/Specificity:** This product was prepared from monospecific antiserum by immunoaffinity chromatography using Rabbit IgG coupled to agarose beads followed by solid phase adsorption(s) to remove any unwanted reactivities. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Goat Serum, Rabbit IgG, Rabbit IgG F(c) and Rabbit Serum. No reaction was observed against Rabbit IgG F(ab). This antibody will react with heavy chains of Rabbit IgG. Minimal reactivity is expected against other Rabbit immunoglobulins.

## Application Details

**Suggested Applications:** WB (Based on references)

**Application Note:** The emission spectra for this DyLight™ conjugate match the principle output wavelengths of most common fluorescence instrumentation. This product is designed for immunofluorescence microscopy, fluorescence based plate assays (FLISA) and fluorescent western blotting. This product is also suitable for multiplex analysis, including multicolor imaging, utilizing various commercial platforms.

**Assay Dilutions:** All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.

**FLISA:** >1:20,000

**IF:** >1:5,000

**WB:** >1:10,000

## Formulation

**Physical State:** Lyophilized

**Concentration:** 1.0 mg/mL by UV absorbance at 280 nm

**Buffer:** 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

**Preservative:** 0.01% (w/v) Sodium Azide

**Stabilizer:** 10 mg/mL Bovine Serum Albumin (BSA) - Immunoglobulin and Protease free

**Reconstitution Volume:** 100 µL

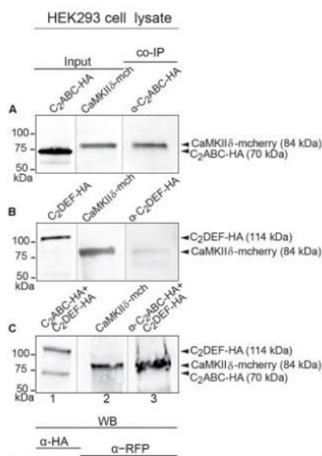
**Reconstitution Buffer:** Restore with deionized water (or equivalent)

## Shipping & Handling

**Shipping Condition:** Ambient

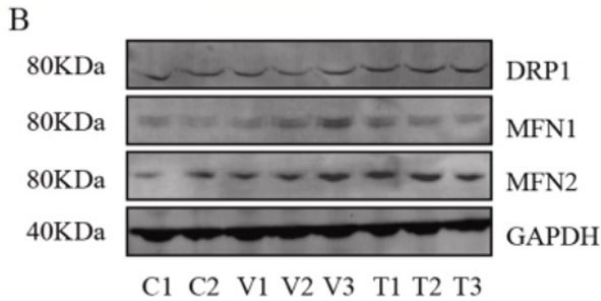
<b>Storage Condition:</b>	Store vial at 4° C prior to restoration. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
<b>Expiration:</b>	Expiration date is one (1) year from date of receipt.

## Images



### Western Blot

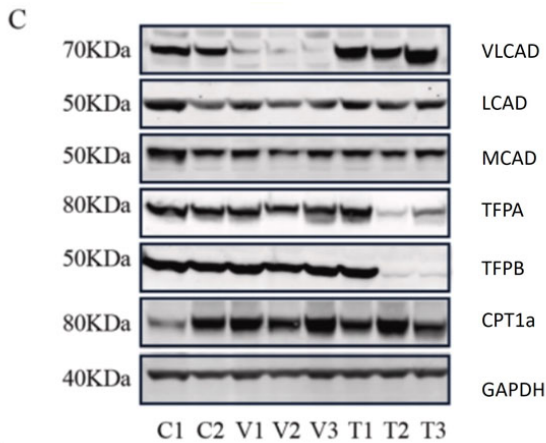
Immunoprecipitation and western blot show interaction of otoferlin with CaMKIIδ. (A–C) Two HA-tagged mouse otoferlin fragments, C2ABC (aa 1–632 in NP\_001093865; 70 kDa) and C2DEF (aa 933–1920; 114 kDa) were co-transfected with mcherry-tagged mouse CaMKIIδ into HEK293 cells. Transfections were performed either with otoferlin C2ABC and CaMKIIδ (A, Input Lane 1 and 2), otoferlin C2DEF and CaMKIIδ (B, Input Lane 1 and 2) or in the presence of both C2ABC and C2DEF fragments and CaMKIIδ (C, Input Lane 1 and 2). Co-immunoprecipitations of C2ABC-HA and C2DEF-HA were conducted from HEK293 cell lysates using anti-HA antibodies (p/n 600-401-384). CaMKIIδ-mcherry was detected in the eluate using an anti-RFP (red fluorescent protein) antibody (p/n 200-301-379) (A–C, Lane 3), indicating that CaMKIIδ co-precipitated with recombinant otoferlin fragments. Secondary anti-rabbit Dylight680 (p/n 611-144-003) and anti-mouse Dylight800 antibodies (610-145-003) (1:10,000). FIGURE 5. PMID: 29046633.



#### Western Blot

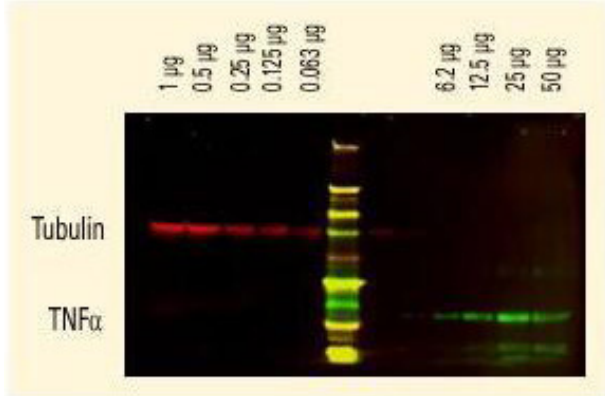
Assessment of mitochondrial fusion and fission.

B. Representative western blots (original blots are shown in supplementary Fig. S10) and quantification of MFN1/2 and DRP1. No significant changes in the relative levels of proteins that facilitate mitochondrial fusion (MFN1/2) and fission (DRP1) between non-disease (control) and mutant primary fibroblasts. Data are depicted as mean  $\pm$  SD,  $n = 3$ . The primary antibodies used as follows: MFN1 1:400, MFN2 (1:400, DRP1 1:100 and GAPDH 1:30,000 dilutions overnight at 4 °C. The membranes were then incubated with fluorescent conjugated secondary antibodies for 1 h; DyLight 800 conjugated goat Anti-Rabbit IgG (611-145-002), Antibody DyLight 680 conjugated Anti-Rabbit IgG made in goat (611-144-003), DyLight 800 conjugated goat Anti-Mouse IgG (610-145-002), and DyLight 680 conjugated donkey Anti-Mouse IgG (610-744-124). Fig 3. PMID: 33725513.



#### Western Blot

C. Representative western blots, original blots are shown in (supplementary Fig S8-9). And densitometric quantification of relative protein levels from western blots. Data are depicted as mean  $\pm$  SD,  $n = 3$ , \*\* $P < 0.01$ , \*\*\* $P < 0.001$  and \*\*\*\* $P < 0.0001$  by one-way ANOVA. Intracellular transport, activation, mitochondrial transport,  $\beta$ -oxidation, carnitine shuttle, and auxiliary proteins. The primary antibodies used as follows: VLCAD 1:1000, MCAD 1:1000, LCAD 1:1000, TFPa 1:500, TFPb 1:3000, CPT1 $\alpha$  1:1000, and GAPDH 1:30,000 dilutions overnight at 4 °C. The membranes were then incubated with fluorescent conjugated secondary antibodies for 1 h; DyLight 800 conjugated goat Anti-Rabbit IgG (611-145-002), DyLight 680 conjugated goat Anti-Rabbit IgG (611-144-003), DyLight 800 conjugated goat Anti-Mouse IgG (610-145-002), and DyLight 680 conjugated donkey Anti-Mouse IgG (610-744-124). Fig 1. PMID: 33725513.









### Western Blot

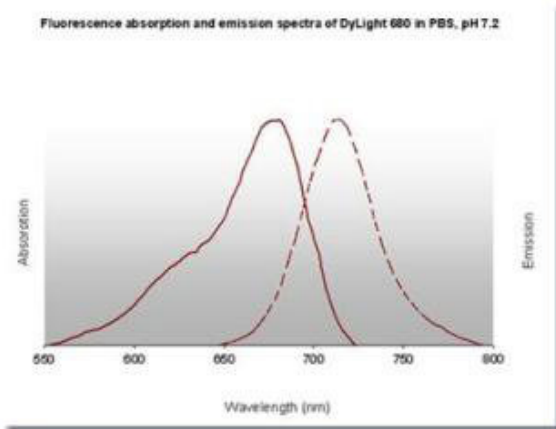
DyLight™ dyes can be used for two-color Western Blot detection with low background and high signal. Anti-tubulin was detected using a DyLight™ 680 conjugate. Anti-TNF $\alpha$  was detected using a DyLight™ 800 conjugate. The image was captured using the Odyssey® Infrared Imaging System developed by LI-COR.

### Diagram

Properties of DyLight™ Conjugates.

Emission	Color	DyLight™ Dye	Ex/Em (nm)	$\epsilon$ ( $M^{-1} cm^{-1}$ )	Similar Dyes
Blue		405	400/420	30,000	Alexa™ 405, Cascade Blue
Green		488	493/518	70,000	Alexa™ 488, Cy2®, FITC
Yellow		549	550/568	150,000	Alexa™ 546, Alexa 555, Cy3®, TRITC
Red		649	646/674	250,000	Alexa™ 647, Cy5®
Near Infrared		680	682/715	140,000	Alexa™ 680, Cy5.5®, IRDye™ 700
Infrared		800	770/794	270,000	IRDye™ 800

### Diagram



## References

- Raimo S et al. Mitochondrial morphology, bioenergetics and proteomic responses in fatty acid oxidation disorders. *Redox Biol.* (2021)
- Meese et al. Activity-Dependent Phosphorylation by CaMKII $\delta$  Alters the Ca<sup>2+</sup> Affinity of the Multi-C2-Domain Protein Otoferlin. *Frontiers in Synaptic Neuroscience* (2017)

## Disclaimer

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