

Product Specification Sheet

Timeless (Tim1) Antibodies

Cat. TIM11/12-S	Rabbit Anti-Mouse/Human Tim1 antiserum	SIZE: 100 ul
Cat. TIM11/12-A	Rabbit Anti-Mouse/Human Tim1 IgG (aff pure)	SIZE: 100 ug
Cat. TIM11/12-P	Mouse/Human Tim1 Control/blocking peptide	SIZE: 100 ug

Several endogenous factors have been linked to rhythmicity or circadian behavior of living organisms. In *Drosophila*, the genes *period* (**dPer**) and *timeless* (*tim*), and in *Neurospora* *frequency* (*freq*), have been proposed to be responsible for their circadian rhythm. In mammals, the suprachiasmatic nuclei (SCN) of the anterior hypothalamus serves as a master circadian clock with a measurable circadian rhythm. In *drosophila*, the RNA and protein products of *per* and *tim* oscillate and may be involved in rhythm regulation. The levels of *per* and *tim* is positively regulated by two basic helix-loop-helix (bHLH)/PAS proteins, dCLOCK and dBMAL1, which heterodimerize and bind to E box enhancers. Translated *per* and *tim* are phosphorylated, form heterodimers, and then translocate to nucleus. Once in the nucleus, PER and TIM function as negative regulators of their own transcription. Recently *drosophila* homolog of mammalian TIM have been cloned and characterized (1). Human Timeless 1 (hTIM1; 1208 aa, chromosome 12q12-13) and mouse TIM1 (1197 aa, chromosome 10D3) share 83% identity. Tim1 is weakly expressed in SCN, and in several peripheral tissues (eyes, spleen and testis). Unlike *drosophila*, mouse Tim1 do not oscillate in the SCN or modulated by light exposure. Human TIM1 and mPER1 specifically inhibit CLOCK-BMAL1-induced transactivation of mPER1 promoter.

Function: Involved in the circadian rhythm autoregulatory loop. Negatively regulates CLOCK-NPAS2/BMAL1-induced transactivation of PER1 possibly via translocation of PER1 into the nucleus. May also play an important role in epithelial cell morphogenesis and formation of branching tubules.

Subcellular Location: Nucleus.

Similarity: Belongs to the timeless family.

Protein name Protein timeless homolog

Synonym mTim

Gene name Name: Timeless; Synonyms: Tim1, Timeless1

Source of Antigen and Antibodies

Antigen	A mixture of 14 aa peptide sequence (protein accession #Q9R1X4, refs 1) (designated TIM11-P) near the N-terminus of mouse TIM1 and 14 aa peptide sequence (designated TIM12-P), control/blocking peptide) conjugated to KLH; conjugated to KLH; Epitope location ~ C-terminus
Ab Host/type	Rabbit, Polyclonal unpurified antiserum (#TIM11/12-S) and IgG, purified over antigen-agarose (Cat # TIM11/12-A)
2-Ab	Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available).
-ve control	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

Form & Storage of Antibodies/Peptide Control

Antiserum (unpurified)

100ul solution lyophilized powder
Supplied in Buffer: 0.05% azide

Reconstitute powder in 100 ul PBS

Affinity pure IgG

100 ug/100ul solution lyophilized powder
Supplied in **Buffer:** PBS+0.1% BSA
Reconstitute powder in PBS at 1mg/ml

Control/blocking peptide

100 ug/100 ul solution lyophilized powder
Supplied in Buffer: PBS pH 7.5,
Reconstitute powder in PBS at 1 mg/ml.

Storage

Short-term: unopened, undiluted liquid vials at 20°C and powder at 4°C or -20°C..

Long-term: at -20°C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at -20°C or below.

Shipping: 4°C for solutions and room temp for powder

Recommended Usage

Western Blotting (1:1K-5K for neat serum and 1-10 ug/ml for affinity pure using ECL technique).

ELISA (1:10K-1:100K; using 50-100 ng of control peptide/well).

Histochemistry: Not tested. We recommend the use of affinity purified antibody at 2-20 ug/ml.

Specificity & Cross-reactivity

Mouse TIM11-P sequence is 100% conserved in rat Tim1 and 85% homologous with *drosophila* and human TIM1. Human TIM12-P sequence is 57% homologous with mouse TIM1. Since antibodies have been produced to a mixture of mouse and human TIM1, antibodies should react with mouse, rat, human, and *drosophila* TIM1. Antibody cross-reactivity with TIM1 from other species is not known. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity

General References: Sangoram AM (1998) Neuron 21, 1101-1113; Zylka MJ (1998) Neuron 21, 1115-1122; Koike N (1998) FEBS Lett. 441, 427-431.

(2) Citations of ADI's Antibodies (see web site for updated list)

Mix E, 2004, Neuroimmunol. 151, 158-170, WB

*This product is for *in vitro* research use only.

Related material available from ADI

TIM11/12-S-A-P 70926J