

Product Specification Sheet

**Timeless (Tim1) Antibodies**

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

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

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