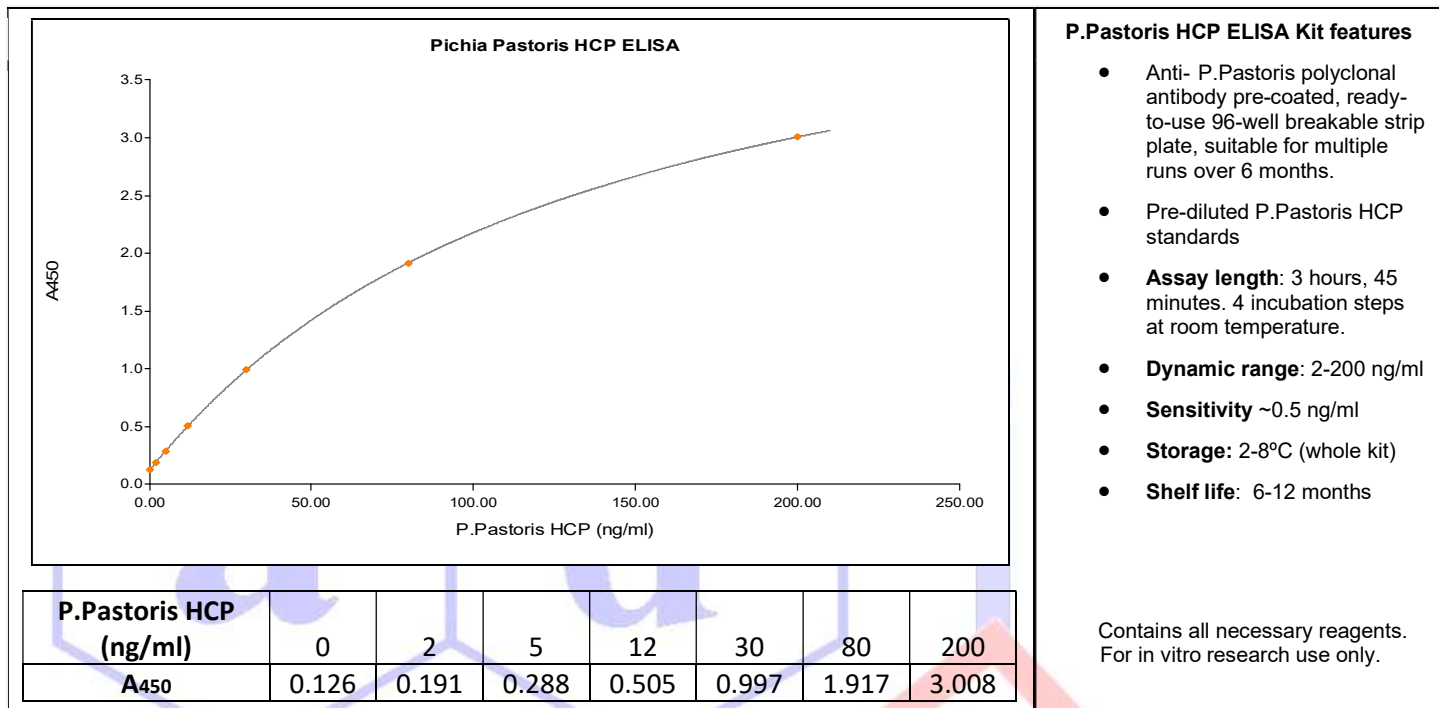


**Pichia Pastoris HCP ELISA Kit Cat# 750-155-PIC**

The Pichia Pastoris HCP ELISA Kit is a highly sensitive sandwich ELISA for the measurement of P.Pastoris contaminants in biological buffers and other appropriately qualified matrices



**Assay Procedure:** Allow all reagents to reach room temperature. Arrange and label required number of strips.

- Step 1.** Pipette 100 ul of samples and calibrators into wells and incubate for 2 hours at room temperature.
- Step 2.** Wash the wells 3X with 300 ul of wash buffer for each well
- Step 3.** Add 100 ul of biotinylated detection antibody to each well and incubate for 1 hour at room temperature
- Step 4.** Wash the wells 3X with 300 ul of wash buffer for each well
- Step 5.** Pipette 100 ul of Streptavidin-HRP conjugate to each well and incubate for 30 minutes at room temperature
- Step 6.** Wash the wells 3X with 300 ul of wash buffer for each well
- Step 7.** Add 100 ul of TMB Substrate solution to all wells, mix gently, and incubate at room temperature for 15 minutes.
- Step 8.** Pipette 100 ul of stop solution into each well and mix gently. Measure at 450 nm w/ 630 nm as a reference filter if available.

**Performance Characteristics**

**Sensitivity:** ~0.5 ng/ml  
**Precision:** Intra-assay: <5%      Inter-assay: <10%

**General Information**

A large number of genes have been cloned and expressed in various host cells (E. coli, yeast, baculovirus, NSO, Sp2/0, HEK, CHO cells). The translated recombinant proteins may remain within the cell, requiring host cell disruption for release, and/or may be secreted into the culture medium. The target recombinant proteins would then be purified from unwanted host cell protein (HCP), often with the aid of a tag (e.g., His, GST, MBP). While traces of HCP (which are often present in the purified material) may not represent a major problem for recombinants that are used for in vitro or research use applications, an increasing number of recombinant proteins are developed for therapeutic purposes (insulin, erythropoietin, GM-CSF or humanized antibodies such Rituximab & Xolair), where the presence of HCP is potentially toxic or allergic, may create other health hazards, or otherwise affect the efficacy of the drug. In these cases, detecting residual HCP and establishing minimum acceptable levels is required. Of two typical and powerful methods used for HCP characterization, Western Blot can reveal the number, size and relative concentrations of HCPs, while ELISA can provide ultra-sensitive detection and quantification using an easy, rapid assay that accommodates large numbers of samples and replicates. The ADI Pichia Pastoris HCP ELISA relies on polyclonal antibodies from rabbits immunized with a full repertoire of intra- and extra-cellular HCPs -- antibodies with multivalent specificities for a wide array of cellular and extra-cellular proteins. The P.Pastoris HCP ELISA provides a broad-range, sensitive tool to conveniently and efficiently screen for the several potential contaminants that may accompany the recombinant protein during processing.