

# Novocastra™ Lyophilized Rabbit Polyclonal Antibody

## *Helicobacter pylori*

### Product Code: NCL-HPp

<b>Intended Use</b>	FOR RESEARCH USE ONLY.
<b>Specificity</b>	Reactive with heat stable, somatic antigens of the whole <i>Helicobacter pylori</i> ( <i>H.pylori</i> ) organism.
<b>Antigen Used for Immunizations</b>	Heat-treated cells of <i>Helicobacter pylori</i> strain CH-20 429.
<b>Preparation</b>	Lyophilized immunoglobulin fraction purified from rabbit serum and diluted in PBS with 1% BSA containing 15 mM sodium azide. Reconstitute with the volume of sterile distilled water indicated on the vial label.
<b>Effective on Frozen Tissue</b>	Not evaluated.
<b>Effective on Paraffin Wax Embedded Tissue</b>	Yes
<b>Recommendations on Use</b>	Immunohistochemistry: Typical working dilution 1:20–1:40. Trypsin digestion of paraffin sections is recommended. 60 minutes primary antibody incubation at 25 °C. Standard ABC technique. Western Blotting: Not evaluated.
<b>Positive Controls</b>	Immunohistochemistry: <i>Helicobacter pylori</i> gastric mucosa.
<b>Staining Pattern</b>	Staining of bacteria on the surface of infected mucosa.
<b>Storage and Stability</b>	Store unopened lyophilized antibody at 4 °C. Under these conditions, there is no significant loss in product performance up to the expiry date indicated on the vial label. The reconstituted antibody is stable for at least two months when stored at 4 °C. For long term storage, it is recommended that aliquots of the antibody are frozen at -20 °C (frost-free freezers are not recommended). Repeated freezing and thawing must be avoided. Prepare working dilutions on the day of use.
<b>General Overview</b>	<i>Helicobacter pylori</i> (previously known as <i>Campylobacter pylori</i> ) is a motile spiral shaped gram-negative bacillus that can colonise the human gastric mucosa. Infection with <i>Helicobacter pylori</i> is very common affecting about half of the world's population. The prevalence of the bacterium in the gastric mucosa increases with age. It is thought to survive the acid conditions in the stomach by colonising regions deep in the mucus layer near the epithelial surface where physiological pH is present. Also, <i>H. pylori</i> produces factors that modify its microenvironment and enhance its ability to survive. One of these factors is a protease that renders gastric mucus impermeable to acid and another is a potent urease which yields ammonia with consequent buffering of acid.
<b>General References</b>	Marzio L, Angelucci D, Grossi L, et al.. The American Journal of Gastroenterology. 93 (2): 223–226 (1998). Nagashima R, Maeda K, Yuda F, et al.. Virchows Arch. 431: 235–239 (1997). Yabuki N, Sasano H, Tobita M, et al.. American Journal of Pathology. 151 (3): 821–829 (1997). Ashton-Key M, Diss T C and Isaacson P G. Journal of Clinical Pathology. 49: 107–111 (1996).



# Instructions for Use

## Trypsin Digestion for Immunohistochemical Demonstration on Paraffin Sections

1. Preheat the following to 37 °C using a water bath:
  - (i) 200 mL of TBS
  - (ii) 200 mL of distilled water.
2. Dissolve 0.2 g Trypsin 250 and 0.2 g Calcium chloride in the 200 mL of TBS.
3. Once the Trypsin solution is at 37 °C, pH to 7.8 with 1 M sodium hydroxide.
4. Place rehydrated paraffin sections in the distilled water to preheat the sections to 37 °C for a minimum of 5 minutes.
5. Incubate sections in Trypsin solution at 37 °C. The time required will depend on the antibody and tissue, however, 30 minutes is usually sufficient.
6. Rinse sections in running tap water.
7. Proceed with immunohistochemistry protocol.

### Reagents Required but not Supplied

50 mM Tris-buffered saline

Trypsin 250: Difco order code 0152-13 (available from Becton Dickinson).

Calcium chloride

1 M Sodium Hydroxide

*\* Trypsin containing chymotrypsin should always be used. The enzyme activities can vary from a supplier and between batches. Such variations may affect the incubation time required.*