

Bactident® Aminopeptidase

For the detection of L-alanine aminopeptidase in microorganisms

Mode of Action

L-alanine aminopeptidase is an enzyme which is localized in the cell envelope of bacteria and which is found in relevant activities almost exclusively in Gram-negative microorganisms.

This enzyme splits off the amino acid L-alanine from various substrates.

In the case of these test strips, the substrate L-alanine-4-nitroanilide is split into 4-nitroaniline and the amino acid L-alanine in the presence of alanine aminopeptidase. The presence of L-alanine aminopeptidase, is indicated by the yellow colouration of the 4-nitroaniline.

The results of the investigations performed so far indicate that there is a very good correlation between the amino-peptidase reaction and the Gram-behaviour of the microorganisms.

Typical Composition

The reaction zone of a test strip contains:

L-alanine-4-nitroanilide 0.5µmol; buffering agents.

Preparation

Suspend a thickly grown individual colony (about 2 mm Ø) in 0.2ml of distilled water to give an opalescent mixture.

Note:

Only bacterial colonies without strong intrinsic colourations should be used for the aminopeptidase test. We recommend that a control test with an aminopeptidase-positive bacterium (e.g. *E.coli*) and an aminopeptidase-negative bacterium (e.g. *Staphylococcus aureus*) should always be carried out at the same time as the main test.

Stability

See expiring date.

Only remove the amount of strips needed at the time! Do not touch the reaction zone of the test strips. Close receptacle firmly immediately after use. Please store at the specified temperature.

Safe Removal

The test strip is to be removed safely after use like bacteria containing material. This may be done by burning, autoclaving or by placing into a 5 to 6% disinfectant solution - for at least 6 hours.

Experimental Procedure

1. Using an inoculation loop, remove an individual, thickly-grown colony from the nutrient medium.
2. In a small test tube, suspend the bacterial mass in 0.2ml of distilled water.
3. Insert the aminopeptidase test strip into the test tube such that the reaction zone is completely immersed in the bacteria suspension.
4. Incubate the test tube in a water bath (or incubation cupboard) for 10 to a maximum of 30 minutes* at 37°C.
5. Read off the reaction by comparison with the colour scale.

Note

- * A clear yellow colouration of the bacteria suspension can be seen after only 10 minutes in the case of most aminopeptidase-positive microorganisms; if no yellow colouration appears within this time, the incubation should be extended to a maximum of 30 minutes so that the weakly aminopeptidase-positive strains can be recognized or the absence of Gram-negative microorganisms can be confirmed (see table for exceptions).

Aminopeptidase-positive Strains*

all Gram-negative microorganisms

The suspension of bacteria turns yellow if L-alanine aminopeptidase-positive organisms are present.

Exceptions: *Bacteroides vulgatus*, *Bacteroides fragilis*, *Camphylobacter* species, *Veillonella parvula*

Aminopeptidase-negative Strains*

all Gram-positive microorganisms

- * acc. to the results of investigations performed so far.

Ordering Information

Product	Merck Cat. No.	Pack contents
Bactident® Amino-peptidase	1.13301.0001	50 test strips