

## The semi-solid medium by Garda for identification of flagellar antigens of *Salmonella*

### Description

The semi-solid medium by Garda is a medical device for *in vitro* use, which is used for identification of flagellar antigens of *Salmonella*. It is a ready-to-use medium for multiplication of bacterial culture of *Salmonella*. This medium allows for maximum development of flagella, what in consequence allows for serological identification of flagellar antigens. It can also be used for inhibition of dominant phase of flagella (strongly developed phase) and induction of poorly exposed flagellar antigens (underdeveloped phase). Flagellar antigens can be identified in serological tests in *Salmonella* strains isolated on bacteriological media from samples taken from humans or from other materials.

### Application

The semi-solid medium by Garda contains components that allow for expression of the maximum amount of flagella, what allows for complete serological identification of *Salmonella* strains. The medium is designed to test flagellar antigens through slide agglutination assay.

The semi-solid medium by Garda allows for obtaining *Salmonella* strains with well-developed flagella. This is expressed in bacterial growth over the entire Petri dish's surface. This medium is used in IMMUNOLAB and it has not happened yet that smooth *Salmonella* strain with flagella would not cover most of the plate surface. This usually allows to determine the both phases of flagellar antigens of biphasic strains. In order to inhibit strongly developed phase and induce underdeveloped phase, you can add to the medium a few drops of antiserum (6-8 drops per plate of  $\varnothing=5\text{cm}$ ) that gives a strong agglutination reaction with a tested strain.

### Composition

The composition of semi-solid medium by Garda for 500 ml.

Agar 2,5 g

Enriched broth 10,0 g

Sodium deoxycholate 0,15 g

pH after dissolution  $7.4 \pm 0,2$

### Properties

Identification of *Salmonella* is based on determination of the antigenic structure of the tested strain. The test is based on the detection of somatic antigens "O" and flagellar antigens "H" in a slide agglutination assay with specific antisera. Identification of flagellar antigens is possible only with the use of enriched semi-solid medium, such as designed semi-solid medium by Garda.

### Medium Preparation

Package contents suspend in 500 ml of distilled water, mix and sterilize in an autoclave at a temperature of 121°C for 20 minutes. After cooling down of the medium to approx. 50°C pour in the Petri dish - diameter of 5 cm (approx. 10 ml for 1 plate). After the medium is cooled down and solidified inoculate *Salmonella* as detailed below. Petri dishes with solidified medium can only be used on the day of their preparation. **ATTENTION! – PETRI DISHES WITH SOLIDIFIED MEDIUM MUST NOT BE DRIED.**

### Performance of the test

After isolation of tested strain of bacteria, it is the best to choose one smooth colony from agar medium and inoculate it in the middle of the plate (using f.e. wire loop) on semi-solid medium by Garda already prepared on Petri dish. After  $24 \pm 2$  hours of incubation at  $37 \pm 1^\circ\text{C}$ , bacteria should cover most of the plate's surface. For flagellar antigens identification collect bacteria from the plates periphery and mix with a drop of selected antiserum on a glass slide. Within 30 seconds to 1 minute after a slight swinging there should occur clumps/cloudlets of bacteria joined by antibodies on the surface of the drop. [Interpretation of results is described in the instruction for use of Set of Antiserum for the identification of *Salmonella* produced by the Research and Development Department of *Salmonella* Center "IMMUNOLAB" Ltd.]

In order to inhibit strongly developed phase add to the Petri dish with semi-solid medium by Garda (temp. approx. 50°C) few drops of antiserum (6-8 drops per plate of  $\varnothing=5\text{cm}$ ) that gives a strong agglutination reaction with a tested strain. Carefully mix serum with medium through plate cradling. After cooling down and solidifying of medium inoculate bacteria in the middle of the plate. Continue as mentioned above. If there is still only dominant phase detected, repeat the whole procedure. If bacteria strain grows only in the place where it was inoculated, it should be recognized as a monophasic strain.

### The content of the package unit

1 package contains 12,65 g of the medium and is designed to prepare 500 ml of the semi-solid medium by Garda.

### Storage conditions and precautions

The medium should be stored at room temperature in a dry and dark place. DO NOT INSERT INTO REFRIGERATOR AND DO NOT FREEZE!

Do not use after the expiry date stated on the package.

The product is intended only for laboratory use and is not suitable for consumption.

The medium is not injurious to health.

### Manufacturer

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