

# SSI ENTERIC MEDIUM



STATENS  
SERUM  
INSTITUT

prevention and control  
of infectious diseases  
and congenital disorders

- *Detects all Enterobacteria*
- *Direct identification*
- *Rapid diagnosis*
- *Cost saving*



PRODUCT INFORMATION

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## Introduction

The SSI Enteric Medium is an indicator medium that combines growth differentiating and selective properties without loss of detection. It can be used in routine laboratories for detection of all commonly found enteric pathogens except *Campylobacter*. The medium has been used for more than 20 years in routine laboratories in Denmark.

## Constituents

Pancreatic digest of casein	Yeast extract
Trisodium citrate	L-phenylalanine
L-tryptophan	Calcium dichloride
Magnesium dichloride	Glucose
Sodium thiosulphate	Ferric citrate
Sodium deoxycholate	Neutral red
Sodium glycerophosphate	Lactose
Sodium pyruvate	Agar
Sodium dodecylbenzenesulphonate	

## Preparation

Mix water, medium and 5 N NaOH in the ratio stated on the box. Bring to the boil with frequent agitation and allow simmering gently to dissolve the agar completely. Cool to about 45°C, before dispensing into petri dishes (e.g. 25 ml in 9 cm or 60 ml in 14 cm dishes).

Preparation in an media preparator: Add deionized water, NaOH and medium in the bowl and agitate manually with a whisk. Start the cycle after setting the temperature to 100°C for one minute. Cool to 45°C and pour in petri dishes.

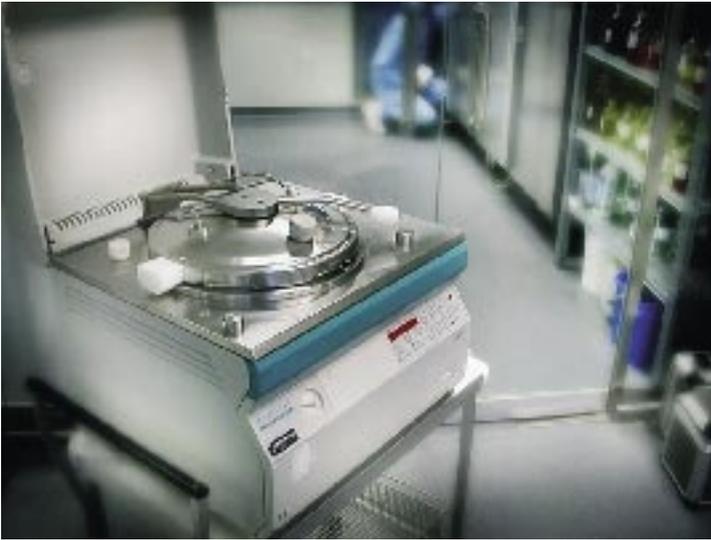
pH 24 hour after production should be 8.1 +/- 0.2.

Store petri dishes at 2-8°C and use within 8 weeks. Before use, dry the plates at room temperature for at least 24 hours protected from daylight.

**Do not autoclave at 121°C!**

## Storage and shelf life

Store the dehydrated medium below 25°C and use before the expiry date indicated on the label.



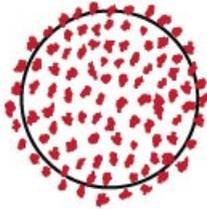
## Quality control

Bacteria	Growth	H <sub>2</sub> S	Phenylalanine	Lactose	Rough
<i>S. Enteritidis</i> ATCC 13076	+++	+	-	-	-
<i>P. mirabilis</i> ATCC 12453	+++	+	+	-	-
<i>E. coli</i> ATCC 25922	+++	-	-	+	-
<i>S. sonnei</i> ATCC 25931	+++	-	-	-	+
<i>E. faecalis</i> ATCC 29212	-	-	-	-	-



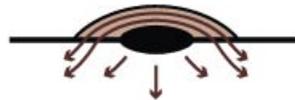
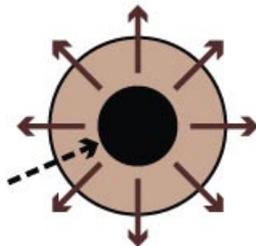
### H<sub>2</sub>S reaction

H<sub>2</sub>S reaction is based on the combined action of sodium thiosulphate, glucose, sodium pyruvate and ferric citrate. H<sub>2</sub>S positive organisms (all with few exceptions are lactose negative) give a distinct ferric sulphide precipitate located centrally and deeply in the anaerobic parts of the colony. The black center is well developed after 20 hours of incubation also in *Salmonella Typhi*.



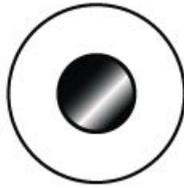
### Lactose fermentation

Lactose fermentation is manifested by acid production and made visible using the indicator combination sodium deoxycholate and neutral red. The acid production is confined to the colony and its immediate surroundings because of the buffering action of deoxycholate resulting in conversion to deoxycholic acid. Deoxycholic acid precipitates and becomes moderately colored by neutral red. The confinement of the reaction to the colony makes recognition of adjacent lactose negative colonies possible.



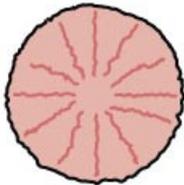
### Phenylalanine deaminase reaction

Phenylalanine deaminase reaction is the result of L-phenylalanin conversion to phenylpyruvate, which with ferric citrate is visible as a brown, diffusible pigment e.g. *Proteus spp.*



### Metallic sheen reaction

Metallic sheen reaction presumably depends on a special concentration of calcium deoxycholate on top of the black center in  $H_2S$  producing *Salmonella* (except in *Salmonella* Typhi). The reaction is almost diagnostic for *Salmonella* bacteria.



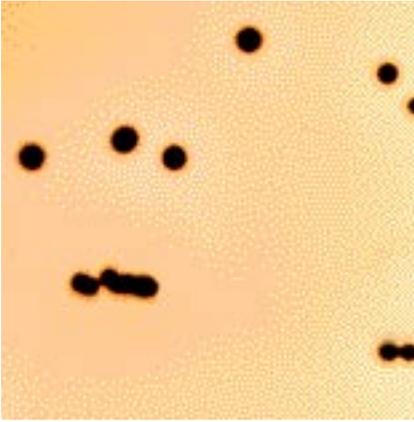
### Rough transformation

Rough transformation in *Shigella sonnei* is a well known phenomenon. By adjusting the concentrations of divalent ions ( $Mg^{2+}$  and  $Ca^{2+}$ ) to rather high levels, a rough transformation is regularly induced in *Shigella sonnei* affecting the surface and the edge of the colonies. The increased spreading of the colonies makes recognition possible also when located among colonies of other bacteria.



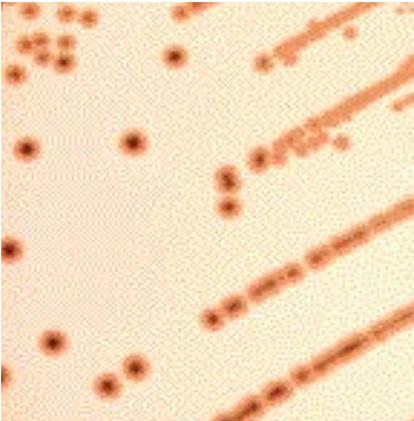
### Indole reaction

It is possible to perform the indole reaction by placing a piece of filter paper wetted with Ehrlich's paradimethylaminobenzaldehyd reagent in the lid of the inverted petri dish at the time of incubation of the plate. The paper will turn red if indole positive colonies grow on the agar plate. It is important to use filter paper with an acid pH.



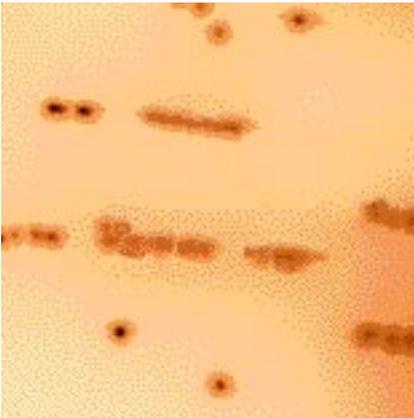
### **S. Enteritidis**

Typical colonies of zoonotic *Salmonella* (in this case *S. Enteritidis*) showing a pale, narrow edge, convex colonies with a black centre ( $H_2S$  production). The metallic sheen reaction is impossible to reproduce on a photo.



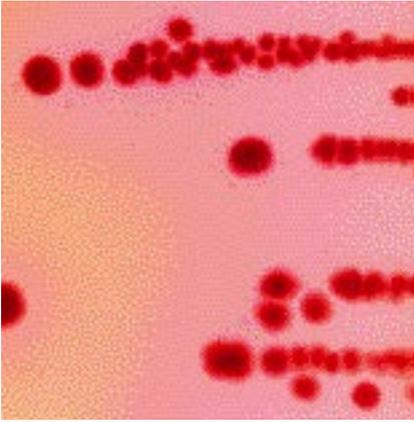
### **S. Typhi**

Typical colonies of *S. Typhi*. Compared to the zoonotic *Salmonella*, the  $H_2S$  production is much less pronounced, the pale edge is broad, and the colonies are less convex.



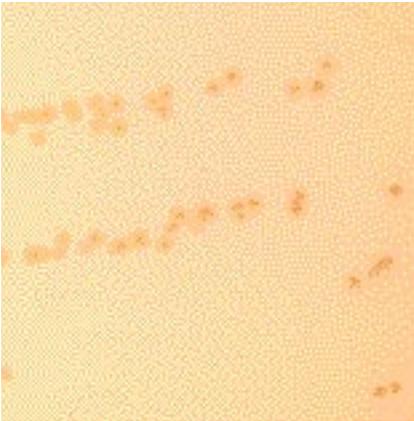
### **P. vulgaris**

A pure culture of *Proteus vulgaris*. The colonies are flat, pale with a dark grey center ( $H_2S$  production). The phenylalanine deaminase reaction is seen as a brown discoloration of the medium below the colonies. Swarming of *Proteus* strains is inhibited. *Proteus* never produce a metallic sheen reaction.



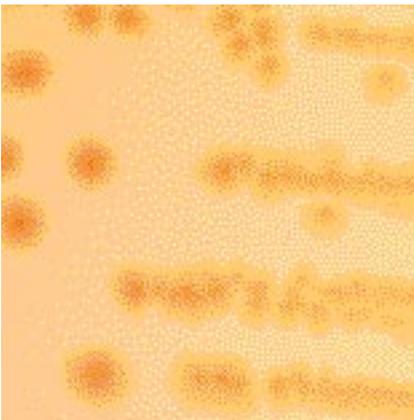
***E. coli***

Typical red colonies of a pure culture of lactose fermenting *E. coli*.



***Y. enterocolitica***

A pure culture of *Yersinia enterocolitica*. Small, round, convex, pale colonies like "pearls on a string".



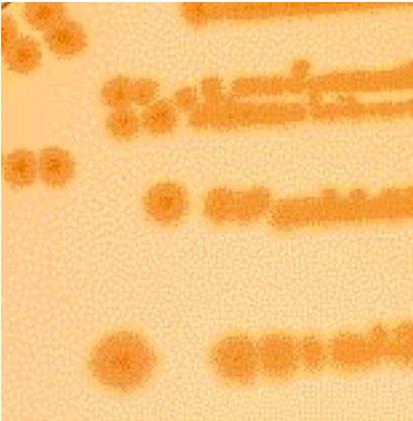
***Sh. sonnei***

Large pale rough colonies of *Shigella sonnei*.



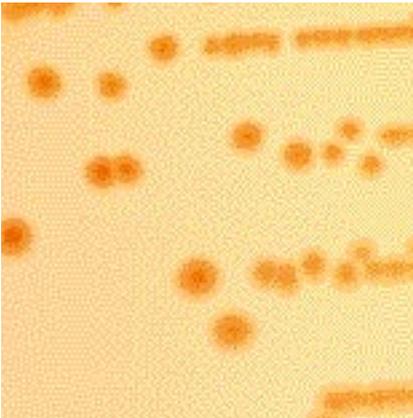
### ***V. cholera***

A pure culture of lactose negative *Vibrio cholera*, reddish brown translucent round colonies rather flat with curved edges. The translucence is impossible to reproduce on a photo.



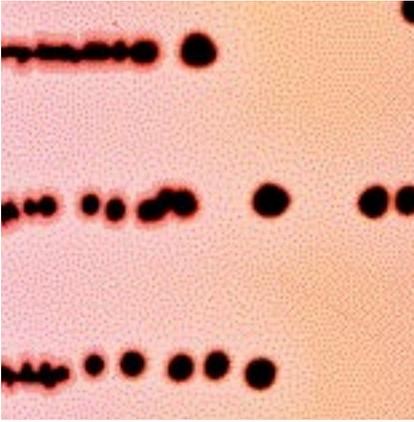
### ***Aeromonas* spp.**

A pure culture of lactose negative *Aeromonas* spp., reddish brown opaque round rather flat colonies with curved edges.



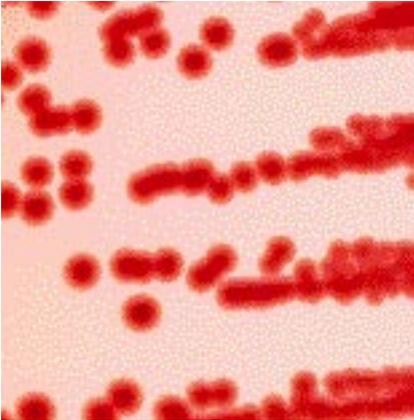
### ***Plesiomonas* spp.**

A pure culture of lactose negative *Plesiomonas* spp., reddish brown opaque round rather flat colonies with curved edges.



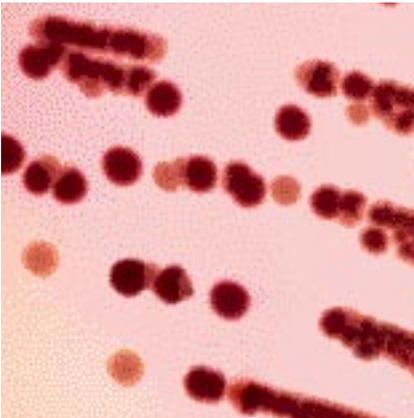
### ***Citrobacter* spp.**

H<sub>2</sub>S producing *Citrobacter*. Large, dark grey center and pink edge. *Citrobacter* never produce a metallic sheen reaction.



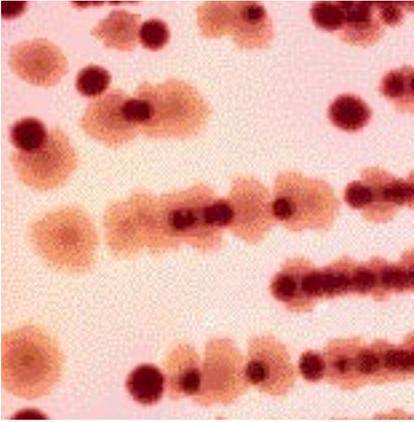
### ***Klebsiella* spp.**

Typical red colonies of a pure culture of lactose fermenting *Klebsiella* spp.



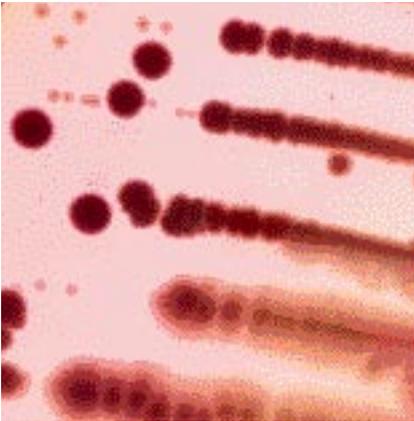
### ***Mixed culture***

A mixed culture of *E. coli* and *Shigella* spp.



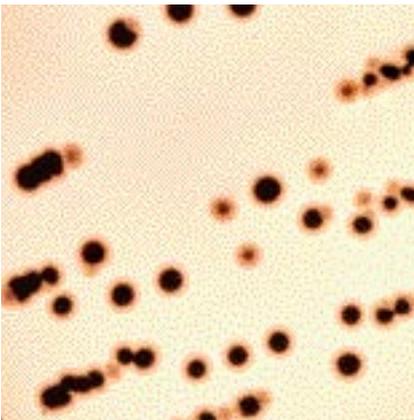
### Mixed culture

A mixed culture of *E. coli* and *Shigella sonnei*.



### Mixed culture

A mixed culture of *E. coli* and *Yersinia* spp.



### Mixed culture

A mixed culture of *Salmonella* Enteritidis and *Salmonella* Typhi.

H <sub>2</sub> S	Lactose	Phenylalanine	Indole	Possible identity
+	-	-	-	<i>Salmonella</i> spp. (incl. <i>arizonae</i> ), lactose neg. <i>Citrobacter freundii</i>
+	-	-	+	<i>E. coli</i> (H <sub>2</sub> S pos. variety), <i>Edwardsiella tarda</i>
+	+	-	-	<i>Citrobacter freundii</i> , lactose pos. <i>Salmonella</i> spp.
+	+	-	+	<i>E. coli</i> (H <sub>2</sub> S pos. variety)
+	-	+	-	<i>Proteus mirabilis</i>
+	-	+	+	<i>Proteus vulgaris</i>
-	-	+	+	<i>Morganella</i> spp., <i>Providencia</i> spp.
-	-	-	-	<i>Shigella</i> spp., <i>Yersinia</i> spp., <i>S. Paratyphi A</i>
-	-	-	+	<i>Shigella</i> spp., <i>Vibrio</i> spp., <i>Aeromonas</i> spp., <i>E. coli</i> , <i>Plesiomonas</i> spp.
-	+	-	-	<i>Klebsiella pneumoniae</i> , <i>Enterobacter</i> spp.
-	+	-	+	<i>E. coli</i> , <i>Aeromonas</i> spp., <i>Klebsiella oxytoca</i>

## References

1. Blom, M., Meyer, AA., Gerner-Smidt, P., Gaarslev, K. & Espersen, F. 1999 Evaluation of Statens Serum Institut Enteric Medium for Detection of Enteric Pathogens. *J. Clin. Microbiol.* 37(7): 2312-2316

2. D. Blue-Hnidy, C.M. Kaufman, B. Barrett, S.D. Allen Comparison of the SSI Enteric Medium with commonly used selective and differential media for isolation of Enteric pathogens. Poster C-300, ASM 2004

## Available products

SSI Enteric Medium, 500 g Article No. 34121

SSI Enteric plate, 9 cm Article No. 724

SSI Enteric plate, 14 cm Article No. 22880

## Additional products for detection of Enteric pathogens

*E. coli* antisera

*Salmonella* antisera

## Ordering and Information

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