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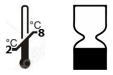
Salmonella H Antisera
Salmonella H for phase inversion Antisera

Salmonella antisera are produced for serological identification of *Salmonella*, based on agglutination method. These polyclonal antibodies are prepared by immunizing rabbits with the standard strains. For high specificity; the non-specific agglutinins have been removed by absorption.

Preservative: 0.1% Sodium azide
Storage condition : 2-8 °C / See Exp. Date on package



On storage, some antisera may become slightly turbid; this does not necessarily indicate deterioration and antisera may be clarified by centrifugation or filtration before use. Gross turbidity indicates contamination and such antisera should be discarded.

Complete identifications of *Salmonella* requires cultural isolation, biochemical characterization and serotyping. However well defined the serology, the use of serological procedures do not supersede cultural isolation and biochemical characterization.

Procedure for H Antisera

1. Pick the colony from Nutrient Agar or Trypticase Soy Agar then spot inoculate to the center surface of Swarm agar. Incubate at 37°C for 18 – 24 hrs. If present the motile strain of *Salmonella* will show spread zone of strain on the surface of Swarm agar.
2. Put a drop of normal saline onto the control area and a drop of antiserum onto the test area of the clean glass slide.
3. Using platinum wire, transfer a portion of loopful of colony from Swarm agar onto the drop of antiserum or normal saline , then mix the cultures and antiserum or cultures and normal saline well. Tilt the glass slide back and forth for one minute.
4. If agglutination is found with Polyvalent H (phase 1&2) antiserum and non-agglutination with normal saline, further test with others H Polyvalent antiserum as described below.

Cat. No.	Description	Factors Present
AS121	Polyvalent H (phase 1&2)	a b c d i e,h e,n,x e,n,z_{15} f,g g,m g,p g,q g,s,t g,z_{51} m,t k l,v l,w l,z_{13} l,z_{40} r y z z_6 z_{10} z_{29} z_{35} z_{36} z_{38} z_{39} z_{41} z_{42} z_{44} z_{60} z_4,z_{23} z_4,z_{24} z_4,z_{32} 1,2 1,5 1,6 1,7
AS122	HMA	a b c d i z_{10} z_{29}
AS123	HMB	E complex G complex
AS124	HMC	k y z r L complex Z_4 complex
AS125	HMD	z_{35} z_{36} z_{38} z_{39} z_{41} z_{42} z_{44} z_{60}
AS126	HME	z_{52} z_{53} z_{54} z_{55} z_{57} z_{61}
AS127	HMF	1,2 1,5 1,6 1,7 z_6
AS128	H : 1 complex	1,2 1,5 1,6 1,7
AS129	H : E complex	e,h e,n,x e,n,z_{15}
AS130	H : G complex	f,g f,g,s f,g,t g,m g,m,s g,m,s,t g,m,t g,p g,p,s g,p,u g,q g,s,t g,z_{51} g,t g,m,q m,t
AS131	H : L complex	l,v l,w l,z_{13} l,z_{28} l,z_{40}
AS132	H : Z_4 complex	z_4,z_{23} z_4,z_{24} z_4,z_{32}

If agglutination occurs to the polyvalent antisera, then test further slide agglutinatin with the specific H-factor antisera present in that set.

Salmonella H-phase and H-factor Antisera as following list.

Cat. No.	Description	Cat. No.	Description
AS141	H : a	AS205	H : 5
AS142	H : b	AS206	H : 6
AS143	H : c	AS207	H : 7
AS144	H : d	AS208	H : z_{13}
AS145	H : i	AS209	H : z_{15}
AS146	H : k	AS210	H : z_{23}
AS147	H : r	AS211	H : z_{24}
AS148	H : y	AS212	H : z_{28}
AS149	H : z	AS213	H : z_{32}
AS150	H : z_6	AS214	H : z_{51}
...	...	...	...

Procedure for H – antigen test (Sven Gard Technique)

Transfer the culture from NA to Swarm agar by spot-inoculation at the center (one loopful of culture taken from an agar medium), incubate at 37°C for 18-24 hrs. Motile strains will spread on the surface of Swarm agar.

Use these strains to test for H – antigen step by step.

1. Add one drop of NSS (control) and one drop of H – antiserum separately to each test area on the glass slide.

2. Using the loop, tale a loopful of culture from the edge of swarm agar, carefully mix with NSS (control) and H – antiserum separately

3. Rock the glass slide gently for 30 seconds to 1 minutes. Observe the reaction, should not be agglutinate with the NSS (control).

4. Start from polyvalent H (phase 1&2) antiserum for the first step to reduce your work time. Always use polyvalent H (phase 1&2) antiserum, HMA ,HMB and HMC antiserum.
4.1 If positive with polyvalent H (phase 1&2) antiserum and HMA antiserum further test for specific antisera such as : H : a , H : b, H : c , H : d , H : i , H : z_{10} , H : z_{29}
4.2 If positive with polyvalent H (phase 1&2) antiserum and HMB antiserum further test for specific antisera H : E complex and H : G complex (If positive with H :G complex antiserum, you must test with H:f, H:g, H:s, H:t, H:m, H:p, H:q , ….. or positive with H:E complex antiserum, you must test with H:h , H:n,x , H:x , H:z_{15})
4.3 If positive with polyvalent H (phase 1&2) antiserum and HMC antiserum further test for specific antisera H:k ,H: y, H:z , H: L complex ,H: Z_4 complex , H: r (If positive with H: L complex antiserum ,you must test with H : v , H:w, H:z_{13} , H:z_{28} or positive with H: Z_4 complex , you must test with H : z_{23} , H : z_{24} , H : z_{32})
4.4 If positive with polyvalent H (phase 1&2) antiserum and HMF antiserum, test for specific antisera such as H:2, H:5, H:6, H:7 and H:z_6

5. Absorbing for H – antigen, by drop H for phase inversion antiserum, should be 1:800 or 1:1,600 titer, which specified to be found for 0.09 ml. in small petri dish, pour melted swarm agar, shake well to mix homogeneous. Allow swarm agar to cool, inoculate culture from first plate swarm agar and incubate at 37°C for 18 – 24 hrs. Specific antibody in swarm agar will be combined with antigen, antigen which isn't specified will spread, further test for another phase antigen.

6. Test for another phase H – antigen, using the same procedure.

7. Absorbing for both 2 phase H-antigen, by a drop for specified H for phase inversion antiserum into another one petri dish and mix with the swarm agar, shake vigorously, and allow agar to cool.
Transfer culture from the second petri dish to the last petridish, incubate at 37°C for 18-24 hrs. Observe the result if there isn't any spread strain in swarm agar, report 2 phase antigen which found.

8. Diagnosis by using Antigenic formulas table of the *Salmonella* serovar 2007, WHO Collaborating Center for Reference and Research on *Salmonella*, Institute Pasteur, France.

Salmonella for phase inversion Antisera as following list.

Cat. No.	Description	Cat. No.	Description
AS231	H : a for phase inversion	...	...
AS232	H : b for phase inversion	AS281	H : 1,6 for phase inversion
AS233	H : c for phase inversion	AS282	H : 1,7 for phase inversion
AS234	H : d for phase inversion	AS283	H : Rz_{40} for phase inversion
AS235	H : e,h for phase inversion	AS284	H : Rz_{45} for phase inversion

Swarm agar Formular (Gard plate)

Tryptic soy broth …… 1,000 ml.
Sodium desoxycholate ……..…….. 0.3 g.
Agar "Difco" …….. …… 6.5 g.
Final pH 7.4

Sterilized by steam pressure at 110 °C (10 lbs./inch^2) for 20 minutes.

Remark : Percentage of Agar may vary to gel strength of each specification.

Reference:

1. Edward,P.R. and Ewing. W.H.,1986, Identification of Enterobacteriaceae, 4th Edition,Burgess Company, Minnesota.
2. Kauffmann, F., Classification of Bacteria , 1975, Munksgarrd ,Copenhagen.
3. Michel Y. Popoff , 2001, Guidelines for The Preparation Of Salmonella Antisera , 8th edition., WHO Collarboraing Centre for Reference and Research on Salmonella, Institute Pasteur, Paris, France.
4. Michel Y. Popoff ,2007 Antigenic formulas of the Salmonella Serovars, 9th edition, WHO Collarboraing Centre for Reference and Research on Salmonella, Institute Pasteur, Paris, France.
5. Manual for the laboratory Identification and Antimicrobial Susceptibility Testing of Bacterial Pathogens of Public Health Importance in the Developing World , 2003 , Center for Disease Control and Prevention : National Center for Infectious Diseases and WHO : Department of Communicable Disease Surveillance and Response.

Serotest

Salmonella H-Polyvalent Antisera as following list.

Cat. No.	Description	Factors Present
AS121	Polyvalent H (phase 1&2)	a b c d i e,h e,n,x e,n,z_{15} f,g g,m g,p g,q g,s,t g,z_{51} m,t k l,v l,w l,z_{13} l,z_{40} r y z z_6 z_{10} z_{29} z_{35} z_{36} z_{38} z_{39} z_{41} z_{42} z_{44} z_{60} z_4,z_{23} z_4,z_{24} z_4,z_{32} 1,2 1,5 1,6 1,7
AS122	HMA	a b c d i z_{10} z_{29}
AS123	HMB	E complex G complex
AS124	HMC	k y z r L complex Z_4 complex
AS125	HMD	z_{35} z_{36} z_{38} z_{39} z_{41} z_{42} z_{44} z_{60}
AS126	HME	z_{52} z_{53} z_{54} z_{55} z_{57} z_{61}
AS127	HMF	1,2 1,5 1,6 1,7 z_6
AS128	H : 1 complex	1,2 1,5 1,6 1,7
AS129	H : E complex	e,h e,n,x e,n,z_{15}
AS130	H : G complex	f,g f,g,s f,g,t g,m g,m,s g,m,s,t g,m,t g,p g,p,s g,p,u g,q g,s,t g,z_{51} g,t g,m,q m,t
AS131	H : L complex	l,v l,w l,z_{13} l,z_{28} l,z_{40}
AS132	H : Z_4 complex	z_4,z_{23} z_4,z_{24} z_4,z_{32}

Salmonella H-phase and H-factor Antisera as following list.

Cat. No.	Description
AS 141	Salmonella H : a Antiserum
AS 142	Salmonella H : b Antiserum
AS 143	Salmonella H : c Antiserum
AS 144	Salmonella H : d Antiserum
AS 145	Salmonella H : i Antiserum
AS 146	Salmonella H : k Antiserum
AS 147	Salmonella H : r Antiserum
AS 148	Salmonella H : y Antiserum
AS 149	Salmonella H : z Antiserum
AS 150	Salmonella H : z_6 Antiserum
AS 151	Salmonella H : z_{10} Antiserum
AS 153	Salmonella H : z_{29} Antiserum
AS 154	Salmonella H : z_{35} Antiserum
AS 155	Salmonella H : z_{36} Antiserum
AS 156	Salmonella H : z_{38} Antiserum
AS 157	Salmonella H : z_{39} Antiserum
AS 158	Salmonella H : z_{41} Antiserum
AS 159	Salmonella H : z_{42} Antiserum
AS 160	Salmonella H : z_{44} Antiserum
AS 161	Salmonella H : z_{52} Antiserum
AS 162	Salmonella H : z_{53} Antiserum
AS 163	Salmonella H : z_{54} Antiserum
AS 164	Salmonella H : z_{55} Antiserum
AS 165	Salmonella H : z_{57} Antiserum
AS 167	Salmonella H : z_{60} Antiserum
AS 168	Salmonella H : z_{61} Antiserum
AS 191	Salmonella H : f Antiserum
AS 192	Salmonella H : g Antiserum
AS 193	Salmonella H : h Antiserum
AS 194	Salmonella H : m Antiserum
AS 195	Salmonella H : n,x Antiserum
AS 196	Salmonella H : p Antiserum
AS 197	Salmonella H : q Antiserum
AS 198	Salmonella H : s Antiserum
AS 199	Salmonella H : t Antiserum
AS 200	Salmonella H : u Antiserum
AS 201	Salmonella H : v Antiserum
AS 202	Salmonella H : w Antiserum
AS 203	Salmonella H : x Antiserum
AS 204	Salmonella H : 2 Antiserum
AS 205	Salmonella H : 5 Antiserum
AS 206	Salmonella H : 6 Antiserum
AS 207	Salmonella H : 7 Antiserum
AS 208	Salmonella H : z13 Antiserum
AS 209	Salmonella H : z15 Antiserum
AS 210	Salmonella H : z23 Antiserum
AS 211	Salmonella H : z24 Antiserum
AS 212	Salmonella H : z28 Antiserum
AS 213	Salmonella H : z32 Antiserum
AS 214	Salmonella H : z51 Antiserum
AS 152	Salmonella H : Rz27 Antiserum
AS 221	Salmonella H : Rz40 Antiserum
AS 222	Salmonella H : Rz45 Antiserum
AS 166	Salmonella H : Rz59 Antiserum
AS 169	Salmonella H : Rz66 Antiserum

Salmonella for phase inversion Antisera as following list.

Cat. No.	Description
AS 231	Salmonella H : a for phase inversion Antiserum
AS 232	Salmonella H : b for phase inversion Antiserum
AS 233	Salmonella H : c for phase inversion Antiserum
AS 234	Salmonella H : d for phase inversion Antiserum
AS 235	Salmonella H : e,h for phase inversion Antiserum
AS 236	Salmonella H : e,n,x for phase inversion Antiserum
AS 237	Salmonella H : e,n,z_{15} for phase inversion Antiserum
AS 238	Salmonella H : f,g for phase inversion Antiserum
AS 239	Salmonella H : g,m for phase inversion Antiserum
AS 240	Salmonella H : g,m,s for phase inversion Antiserum
AS 241	Salmonella H : g,p for phase inversion Antiserum
AS 242	Salmonella H : g,p,u for phase inversion Antiserum
AS 243	Salmonella H : g,q for phase inversion Antiserum
AS 244	Salmonella H : g,s,t for phase inversion Antiserum
AS 245	Salmonella H : g,z_{51} for phase inversion Antiserum
AS 246	Salmonella H : i for phase inversion Antiserum
AS 247	Salmonella H : k for phase inversion Antiserum
AS 248	Salmonella H : l,v for phase inversion Antiserum
AS 249	Salmonella H : l,w for phase inversion Antiserum
AS 250	Salmonella H : l,z_{13} for phase inversion Antiserum
AS 251	Salmonella H : l,z_{28} for phase inversion Antiserum
AS 252	Salmonella H : m,t for phase inversion Antiserum
AS 253	Salmonella H : r for phase inversion Antiserum
AS 254	Salmonella H : y for phase inversion Antiserum
AS 255	Salmonella H : z for phase inversion Antiserum
AS 256	Salmonella H : z_4,z_{23} for phase inversion Antiserum
AS 257	Salmonella H : z_4,z_{24} for phase inversion Antiserum
AS 258	Salmonella H : z_4,z_{32} for phase inversion Antiserum
AS 259	Salmonella H : z_6 for phase inversion Antiserum
AS 260	Salmonella H : z_{10} for phase inversion Antiserum
AS 261	Salmonella H : Rz_{27} for phase inversion Antiserum
AS 262	Salmonella H : z_{29} for phase inversion Antiserum
AS 263	Salmonella H : z_{35} for phase inversion Antiserum
AS 264	Salmonella H : z_{36} for phase inversion Antiserum
AS 265	Salmonella H : z_{38} for phase inversion Antiserum
AS 266	Salmonella H : z_{39} for phase inversion Antiserum
AS 267	Salmonella H : z_{41} for phase inversion Antiserum
AS 268	Salmonella H : z_{42} for phase inversion Antiserum
AS 269	Salmonella H : z_{44} for phase inversion Antiserum
AS 270	Salmonella H : z_{52} for phase inversion Antiserum
AS 271	Salmonella H : z_{53} for phase inversion Antiserum
AS 272	Salmonella H : z_{54} for phase inversion Antiserum
AS 273	Salmonella H : z_{55} for phase inversion Antiserum
AS 274	Salmonella H : z_{57} for phase inversion Antiserum
AS 275	Salmonella H : Rz_{59} for phase inversion Antiserum
AS 276	Salmonella H : z_{60} for phase inversion Antiserum
AS 277	Salmonella H : z_{61} for phase inversion Antiserum
AS 278	Salmonella H : Rz_{66} for phase inversion Antiserum
AS 279	Salmonella H : 1,2 for phase inversion Antiserum
AS 280	Salmonella H : 1,5 for phase inversion Antiserum
AS 281	Salmonella H : 1,6 for phase inversion Antiserum
AS 282	Salmonella H : 1,7 for phase inversion Antiserum
AS 283	Salmonella H : Rz_{40} for phase inversion Antiserum
AS 284	Salmonella H : Rz_{45} for phase inversion Antiserum

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