TT Broth Base, Hajna

Intended Use

TT (Tetrathionate) Broth Base, Hajna is used for enriching *Salmonella* from food and dairy products prior to isolation procedures.

Summary and Explanation

TT Broth Base, Hajna is used as a selective enrichment for the cultivation of *Salmonella* spp. *Salmonella* organisms can be injured in food-processing procedures. These procedures include exposure to low temperatures, sub-marginal heat, drying, radiation, preservatives and sanitizers.[1] Although injured cells may not form colonies on selective media, they can cause disease if ingested.[2] *Salmonella* spp., in particular, cause many types of infections from mild self-limiting gastroenteritis to life-threatening typhoid fever.[3] The most common form of *Salmonella* disease is self-limiting gastroenteritis with fever lasting less than 2 days and diarrhea lasting less than 7 days.[3]

TT Broth Base, Hajna conforms to the formulation of Hajna and Damon.[4] The medium is a modification of the enrichment described by Kauffmann[5] and Knox.[6] Hajna and Damon[4] developed a new broth containing yeast extract, peptone, carbon sources and the selective agents, sodium desoxycholate and brilliant green (replacing bile salts).

TT Broth Base, Hajna is used in testing *Salmonella* in egg processing plants.[7] It is included in procedures for the isolation and identification of *Salmonella* from meat and poultry as well as egg products.[8]

Principles of the Procedure

Peptone provides nitrogen and amino acids. Yeast extract supplies growth factors and vitamins. Dextrose and mannitol are fermentable carbohydrates. Selectivity is accomplished by the combination of sodium thiosulfate and tetrathionate, suppressing coliform organisms.[6] Tetrathionate is formed in the medium by the addition of a solution containing iodine and potassium iodide. Organisms containing the enzyme tetrathionate reductase will proliferate in this medium.

Sodium desoxycholate and brilliant green are selective agents that suppress coliform bacteria and inhibit gram-positive organisms. Sodium chloride maintains the osmotic balance of the medium. Calcium carbonate is a neutralizer that absorbs toxic metabolites.

Formula

Difco™ TT Broth Base, Hajna

Approximate Formula* Per Liter		
Yeast Extract	2.0	g
Tryptose	18.0	g
Dextrose	0.5	g
D-Mannitol	2.5	g
Sodium Desoxycholate	0.5	g
Sodium Chloride	5.0	g
Sodium Thiosulfate	38.0	g
Calcium Carbonate	25.0	g
Brilliant Green	0.01	g

**Adjusted and/or supplemented as required to meet performance criteria.*

User Quality Control

Identity Specifications

Difco™ TT Broth Base, Hajna

Dehydrated Appearance:	Beige to very light green, free-flowing, homogeneous.
Solution:	9.15% solution, partially insoluble in purified water upon boiling. Solution is light green, slightly opalescent with a heavy white precipitate.
Prepared Appearance:	Light green, slightly opalescent with a heavy white precipitate.
Reaction of 9.15% Solution at 25°C:	pH 7.6 ± 0.2 (after addition of the iodine solution)

Cultural Response

Difco™ TT Broth Base, Hajna

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-24 hours. After incubation, plate the inoculated broth onto MacConkey Agar and incubate at 35 ± 2°C for 18-24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	COLONY COLOR ON MACCONKEY AGAR
Escherichia coli	25922	10^2-10^3	None to poor	Pink with bile precipitate
Salmonella enterica subsp. *enterica* serotype Typhimurium	14028	10^2-10^3	Good	Colorless

Directions for Preparation from Dehydrated Product

1. Suspend 91.5 g of the powder in 1 L of purified water. Mix thoroughly.
2. Heat to boiling. DO NOT AUTOCLAVE. Cool to below 50°C.
3. Add 40 mL iodine solution (5 g iodine crystals and 8 g potassium iodide dissolved in 40 mL of purified water) and mix well.
4. Dispense into sterile tubes while keeping suspension well mixed. Do not heat the medium after adding iodine.
5. Test samples of the finished product for performance using stable, typical control cultures.

Procedure

After preparation, add 1-3 g of fecal specimen to each tube (heavy inoculum). Incubate tubes for 12-24 hours at 35 ± 2°C in an aerobic atmosphere.

Expected Results

Growth is indicated by turbidity in the medium. Subculture to selective and differential enteric plating media for further investigations.

References

1. Hartman and Minnich. 1981. J. Food Prot. *44*:385.
2. Sorrells, Speck and Warren. 1970. Appl. Microbiol. *19*:39.
3. Gray. 1995. *In* Murray, Baron, Pfaller, Tenover and Yolken (ed.), Manual of clinical microbiology, 6th ed. American Society for Microbiology, Washington, D.C.
4. Hajna and Damon. 1956. Appl. Microbiol. *4*:341.
5. Kauffman. 1930. Zentralb. Bakteriol. Parasitenkd. Infektionskr. Hyg. Abr. I Orig. *113*:148.
6. Knox, Gell and Pollack. 1942. J. Pathol. Bacteriol. *54*:469.
7. Catalano and Knable. 1994. J. Food Prot. *57*:587.
8. U.S. Department of Agriculture. 1998. Microbiology laboratory guidebook, 3rd ed. Food Safety and Inspection Service, USDA, Washington, D.C.

Availability

Difco™ TT (Tetrathionate) Broth Base, Hajna

USDA

Cat. No.	249120	Dehydrated – 500 g
	249110	Dehydrated – 2 kg