

# **BSK-H Medium Base**

# **Technical Data**

# **M1668**

# Intended use

For the cultivation of Borrelia burgdorferi.

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Composition**	
Ingredients	g / L
Bovine Serum Albumin	50.000
Part B	-
L-Cysteine.HCl.H <sub>2</sub> O	0.260
L-Cystine	0.020
L-Glutamic acid	0.075
Glycine	0.050
L-Histidine HCl. H <sub>2</sub> O	0.020
Trans-4-Hydroxy-L-Proline	0.010
L-Isoleucine	0.020
L-Leucine	0.060
L-Lysine.HCl	0.070
L-Methionine	0.015
L-Phenylalanine	0.025
L-Proline	0.040
L-Serine	0.025
L-Threonine	0.030
L-Tryptophan	0.010
L-Tyrosine	0.040
L-Valine	0.025
N-Acetyl-D-Glucosamine	0.400
L-Ascorbic acid	0.050
PABA	0.00005
D-Biotin	0.00001
Choline chloride	0.0005
Citric acid.3Na.2H <sub>2</sub> O	0.700
Coenzyme A	0.0025
Cocarboxylase	0.001
2'-Deoxyadenosine	0.010
2'-Deoxyguanosine	0.010
2'-Deoxycytidine.HCl	0.0116
Flavin Adenine Dinucleotide.2Na	0.000106
Folic acid	0.00001
myo-Inositol	0.00005
5-Methyldeoxycytidine	0.0001
Nicotinamide adenine dinucleotide	0.007
Nicotinamide adeninedinucleotide phosphate	0.001
Niacinamide	0.000025
Nicotinic acid	0.000025
D-Pantothenic Acid-Hemicalcium	0.00001
Pyridoxal.HCl	0.000025
Pyridoxine.HCl	0.000025
Pyruvic Acid.Na	0.800
Riboflavin	0.00001
Thiamine.HCl	0.00001
Thymidine	0.010
Uridine-5-Triphosphate.Na	0.001
Calcium Chloride (anhydrous)	0.200
Magnesium Sulphate (anhydrous)	0.09769
Potassium Chloride	0.400
Sodium Acetate (anhydrous)	0.050
Please refer disclaimer Overleaf.	

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Sodium Bicarbonate	2.200	
Sodium Chloride	6.800	
Sodium Phosphate monobasic (anhydrous)	0.122	
D-Glucose	6.000	
Phenol Red. Na	0.02124	
Glutathione	0.010	
D-Glucuronic acid.Na	0.00388	
Cholesterol	0.0002	
Tween 80	0.005	
HEPES	6.000	
Peptone, special	5.000	
Yeast Extract	2.000	
L-Alanine	0.025	
L-Arginine	0.05787	
L-Asparatic acid	0.030	
Final pH ( at 25°C)	7.6±0.2	
**Formula adjusted, standardized to suit performance parameters		

# Directions

Suspend 25.0 grams of Bovine Serum albumin in 450 ml distilled water. Mix well to dissolve the medium completely. To this add 15.9 grams of the medium. Add distilled water to make the final volume to 500 ml. Mix thoroughly to get a clear solution. Sterilize by filtration. **DO NOT AUTOCLAVE.** Aseptically add entire contents of one vial of APR Selective Supplement (FD179) and 30 ml of BSK-H Supplement (FD180). Mix well and dispense as desired.

# **Principle And Interpretation**

BSK-H Medium is a modification of BSK-II medium developed by Pollack et al. (1) for the cultivation of *Borrelia burgdorferi*. *Borrelia* species are relatively slow-growing and their nutritional requirements appear to be complex. The study of *Borrelia* was greatly facilitated by the development of a culture medium by Kelly (2) that supported the growth of Spirochaetes. Stoenner enriched the basic formulation of Kelly with the addition of yeast extract tissue culture medium (3). Subsequent modifications by Barbour (4) resulted in BSK (Barbour-Stoenner-Kelly) medium, which facilitated isolation of *Borrelia* form a variety of tissue. This medium is complex mixture of different amino acids, vitamins and growth factors which are required for the growth of *Borrelia* and *Spirochaete*, it is enriched with bovine albumin and rabbit serum. Peptone, special serves as nitrogen source while glucose as energy source. Cholesterol incorporated in the medium acts as source of lipid. The success of in vitro culture of *Borrelia* is usually dependent on the quality of the animal serum or albumin used in media preparation (5). HEPES provides buffering capacity to the medium while different salts of Magnesium, sodium, calcium and potassium maintain the ionic balance in the medium.

#### Type of specimen

Clinical samples -skin scrappings

#### **Specimen Collection and Handling:**

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (6,7). After use, contaminated materials must be sterilized by autoclaving before discarding.

#### **Warning and Precautions**

In Vitro diagnostic Use only. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

#### Limitations

1. Since the medium is nutritionally rich, it is more likely to get contaminated.

- 2.Proper filter sterilization should be carried out
- 3. Medium contains heat sensitive components, hence should not be boiled or heated.
- 4. Further isolation and biochemical tests must be performed for confirmation.

# Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the when stored at recommended temperature.

#### **Quality Control**

#### Appearance

Light yellow to pink homogeneous free flowing powder

#### Colour and Clarity of prepared medium

Light orange coloured, clear transparent liquid forms in tubes.

#### Reaction

Reaction of 3.18% w/v aqueous solution of the medium along with Bovine Serum albumin at 25°C. pH : 7.6±0.2

#### pН

7.40-7.80

#### **Microbial Load**

Maximum Limit

#### Cultural Response

M1668: Cultural characteristics observed after 1-4 weeks incubation at 30-35°C after addition of APR Selective Supplement (FD179) and of BSK-H Supplement (FD180).

Organism	Growth	
Cultural Response		
Escherichia coli ATCC	Inhibited	
25922 (00013*)		
Borrelia burgdorferi ATCC	Fair to good	
35210		
Staphylococcus aureus	Inhibited	
subsp. aureus ATCC		
25923 (00034*)		
Key : *Corresponding WDCM numbers.		

### **Storage and Shelf Life**

Store at 2 - 8°C in tightly closed container. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

#### Reference

- 1.Pollack R.J. et al, 1993, J. Clin. Microbiol., 31:1251-5.
- 2.Kelly, R., 1971, Science, 173:443-444.
- 3.Stoenner, H.G. et al, 1982, J. Exp. Med., 156:1297-1311.
- 4.Barbour, A.G., 1984, J. Biol. Med. , 57:521-525.
- 5.Calister, S.M, et al., 1990, J. Clin. Microbiol., 28:363-365.
- 6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.

7.Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015). Manual of Clinical Microbiology, 11th Edition. Vol. 1.



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