

## Product Insert

### BRUCELLA BLOOD AGAR (BRU)

#### Products

AS-111	Brucella Blood Agar (BRU)	1 plate / pkg
AS-141	Brucella Blood Agar (BRU)	4 plates / pkg
AS-614	Brucella Blood Agar (BRU) – 150 mm plates	1 plate / pkg

The following products contain BRU as one of the multiple components:

AS-302	BRU Mono / BBE-LKV Biplate	1 plate each / pkg
AS-322	BRU Mono / BBE-PEA Biplate	1 plate each / pkg
AS-303	BRU Mono / LKV Mono / PEA Mono	1 plate each / pkg
AS-323	BRU Mono / PEA Mono / BBE-LKV Biplate	1 plate each / pkg
AS-444	BRU Mono / PEA Mono / LKV Mono / BBE Mono	1 plate each / pkg

#### Intended Use

Brucella Blood agar (BRU) is intended for the isolation, quantitation, and partial identification of obligate anaerobic bacteria from clinical specimens. This media will also support the growth of aerobic and microaerophilic bacteria if incubated appropriately. BRU is also suitable for use in antibiotic differential disk examination and spot biochemical testing.

#### Summary

BRU agar is an enriched non-selective media that supports the growth of fastidious microorganisms. BRU agar contains casein, peptones, yeast extract, and dextrose as the nutrient base medium. It is supplemented with vitamin K<sub>1</sub> and hemin to facilitate the recovery and pigment production of *Prevotella melaninogenica*, and other fastidious anaerobes. Sheep blood has been added for the growth factors required by some anaerobic bacteria, and allows the observation of hemolytic reactions as seen by the double zone  $\beta$ -hemolysis of *Clostridium perfringens*. This media is prepared, dispensed, and packaged under oxygen-free conditions to prevent the formation of oxidized products prior to use.

#### Formulation\*

Pancreatic Digest of Casein	10.00	g
Soy Peptone	3.00	g
Meat Peptone	10.00	g
Dextrose	1.00	g
Yeast Extract	2.00	g
Sodium Chloride	5.00	g
Sodium Bisulfite	0.10	g
L-Tryptophan	0.20	g
Calcium Lactate	0.50	g
Sodium Acetate	0.50	g
Ascorbic Acid	0.10	g
Hemin (0.1% solution)	5.00	mL
Vitamin K <sub>1</sub> (1.0% solution)	1.00	mL
L-Cystine	0.40	g
Sodium Hydroxide (4.0% solution)	4.00	mL
Agar	15.00	g
Sheep Blood	45.50	mL
DI Water	1.00	L

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

Final pH:  $7.1 \pm 0.4$  at  $25^{\circ}\text{C}$   
Final weight:  $16.0\text{ g} \pm 1.6\text{ g}$

(Final weight:  $68.0\text{ g} \pm 3.4\text{ g}$ ; Fill depth:  $4.0\text{ mm} \pm 0.5\text{ mm}$  for AS-614 only.)

## Precautions

For *IN VITRO DIAGNOSTIC USE* only. Utilize approved biohazard precautions and aseptic technique when using this product. This product is for use only by properly-trained and qualified personnel. Sterilize all biohazard waste prior to disposal.

## Storage and Shelf Life

**Storage:** Upon receipt, store at room temperature in original package until used. Avoid overheating or freezing. Do not use media if there are signs of deterioration (shrinking, cracking, or discoloration due to oxidation of media) or contamination. The expiration date applies to the product in its original packaging and stored as directed. Do not use product past the expiration date shown on the label.

**Shelf Life:** 90 days from date of manufacture.

## Procedure

**Specimen Collection:** Protect specimens for anaerobic culture from oxygen during collection, transportation, and processing. Consult appropriate references for detailed instructions concerning collection and transportation of anaerobes.

**Methods for Use:** BRU should be inoculated directly with clinical specimen or from a broth that has been inoculated from a clinical specimen. Streak plates with inoculum to obtain isolated colonies and immediately place into an anaerobic atmosphere, incubating at  $35\text{-}37^{\circ}\text{C}$  for 18-48 hours. Extended periods of incubation may be required to recover some anaerobes. Detailed instructions for processing anaerobic cultures can be found in the listed references.

## Materials Required, But Not Provided

Standard microbiological supplies and equipment such as loops, saline blanks, slides, staining supplies, microscope, incinerator / autoclave, incubators, anaerobic chamber / anaerobic jars, disinfectant, other culture media, and serological / biochemical reagents.

## Interpretations of Results

BRU agar should support good growth of obligate anaerobes and other fastidious microorganisms found in clinical infections. In addition, this media should support typical pigment production by *Prevotella melaninogenica* and typical double zone of  $\beta$ -hemolysis around colonies of *Clostridium perfringens*.

## Limitations

BRU agar will not provide complete information for identification of bacterial isolates. Additional test procedures and media are required for complete identification. In some cases, BRU agar may be overgrown with swarming *Proteus* spp. or *Clostridium* spp. It is recommended that selective media such as Brucella Laked Blood Agar with Kanamycin and Vancomycin (LKV, catalog #: AS-112) and/or Brucella Blood Agar with Phenylethyl Alcohol (PEA, catalog #: AS-113) also be inoculated from clinical specimens to prevent such overgrowth and thus provide isolated colonies. Consult reference materials for additional information.

## Quality Control

The following organisms are routinely used for quality control testing (nutritive capacity) at Anaerobe Systems.

Organism Tested for BRU	ATCC #	Results	Time	Special Reaction
Bacteroides fragilis*	25285	Growth	24 hrs	
Prevotella melaninogenica*	25845	Growth	24 – 48 hrs	Pigment <sup>†</sup> (tan color)
Fusobacterium necrophorum	25286	Growth	24 hrs	
Fusobacterium nucleatum*	25586	Growth	24 hrs	
Clostridium perfringens*	13124	Growth	24 hrs	Double Zone of $\beta$ -hemolysis
Peptostreptococcus anaerobius*	27337	Growth	24 hrs	
Staphylococcus aureus or Enterococcus faecalis	25923 29212	Growth	24 hrs	
Escherichia coli	25922	Growth	24 hrs	
Proteus mirabilis	12453	Growth	24 hrs	
Propionibacterium acnes or Clostridium difficile	6919 9689	Growth	24 – 48 hrs 24 hrs	

\* Organisms specified by CLSI for Quality Control testing of Anaerobic Blood Agars.

<sup>†</sup> Pigment production may require more than 48 hours of incubation

**User Quality Control:** The final determination to the extent and quantity of user laboratory quality control must be determined by the end user.

If sterility testing is to be performed on this product, a representative sample of the lot(s) should be incubated anaerobically and aerobically for 48 – 96 hours.

If the nutritive capacity of this media is to be tested for performance, it is recommended that the following ATCC organisms be evaluated for growth.

Organism	ATCC #	Expected Results	Special Reactions
B. fragilis	25285	24 hrs	
P. melaninogenica	25845	24-48 hrs	Pigment <sup>†</sup>
F. necrophorum	25286	24 hrs	
C. perfringens	13124	24 hrs	Double zone of $\beta$ -hemolysis
P. anaerobius	27337	24 hrs	

<sup>†</sup> Pigment production may require more than 48 hours of incubation

**Physical Appearance:** BRU should appear opaque burgundy red in color. For AS-614, the media is in a 150 mm x 15 mm plate with a fill depth of 4.0 mm  $\pm$  0.5 mm. All other catalog numbers for BRU come in standard sized 100 mm x 15 mm petri plates.

## References

1. Dowell, V. R., Jr., G. L. Lombard, F. S. Thompson and A. Y. Armfield. 1977. *Media for the Isolation, Characterization and Identification of Obligately Anaerobic Bacteria*. USDHHS, CDC. Atlanta, GA 30333.
2. Engelkirk, P. G., Duben-Engelkirk, J. and Dowell, V. R. 1992. *Principles and Practices of Clinical Anaerobic Bacteriology*. Star Publishing Co., Belmont, CA 94002.
3. Holdeman, L. V., F. P. Cato and W. E. C. Moore. 1987. *Anaerobe Laboratory Manual*. Virginia Polytechnic Institute and State University. Blacksburg, VA 24061
4. Jousimeis-Somer, H. R., Summanen, P., Citron, D. M., Baron, E. J., Wexler, H. M. and S. M. Finegold. 2002. *Wadsworth – KYL Anaerobic Bacteriology Manual*. Star Publishing Co., Belmont, CA 94002.
5. CLSI. *Quality Control for Commercially Prepared Microbiological Culture Media; Approved Standard- Third Edition*. (2004). CLSI document M22-A3. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.

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