# INSTRUCTIONS FOR USE

# EGG YOLK AGAR (EYA)

#### **PRODUCTS**

AS-511 Egg Yolk Agar (EYA)

1 plate / pkg

#### INTENDED PURPOSE

TruPRAS™ Anaerobic Culture Media is intended for the transport, preservation, or cultivation of a wide variety of microorganisms from specimens to aid in the isolation of bacteria for in vitro diagnostic and / or research / general laboratory purposes.

#### INTENDED USERS

Scientists, laboratory, and healthcare professionals trained in anaerobic microbiology techniques working in areas such as clinical, research, industrial, pharmaceutical and veterinary applications.

## **FORMULATION\***

EYA Agar is an enriched, non-selective, and differential medium that supports the growth of a wide range of anaerobic bacteria, including *Clostridium* species and other obligate anaerobes. It is used to detect lecithinase, lipase, and proteolytic activity through characteristic colony reactions. The nutritive base contains casein, yeast extract, dextrose, and tryptophan to support robust bacterial growth. Hemin and vitamin K₁ are added to enhance the growth of fastidious anaerobes. The addition of an egg yolk suspension makes the medium differential by enabling the detection of specific enzymatic activities. Lecithinase activity is indicated by the formation of an opaque, insoluble precipitate surrounding the colonies, caused by the degradation of lecithin. Lipase-positive organisms produce an iridescent sheen ("oil on water") due to the hydrolysis of free fats in the egg yolk. Proteolytic activity is observed as clearing of the medium surrounding the colonies. This medium is prepared, dispensed, and packaged under oxygen-free conditions using TruPRAS™ Technology to prevent the formation of oxidized products prior to use. This product is supplied ready to use, with no pre-reduction step required.

Pancreatic digest of casein	20.00	g
Yeast extract	5.00	g
Sodium phosphate dibasic	5.00	g
Sodium chloride	2.50	g
Dextrose	2.00	g
Agar	20.00	g
L-Tryptophan	0.20	g
Polysorbate 80	1.00	mL
Magnesium sulfate heptahydrate	0.01	g
Sodium pyruvate	0.50	g
L-Cystine	0.40	g
Hemin	5.00	mg
Vitamin K <sub>1</sub>	10.00	mg
Sodium hydroxide	0.16	g
Egg yolk suspension	100.00	mL
DI Water	1.00	L

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



Final pH:  $7.2 \pm 0.3$  at  $25^{\circ}$ C Final weight:  $16.0 \text{ g} \pm 1.6 \text{ g}$ 

#### **PRECAUTIONS**

For <u>IN VITRO DIAGNOSTIC USE</u> only. Utilize approved biohazard precautions and aseptic technique when using this product. This product is for use by properly trained and qualified personnel only. Sterilize all biohazard waste prior to disposal. This product is manufactured as a single use device.

Report serious incidents that occur in direct relation to this product to <u>tech@biolog.com</u>. As necessary, report serious incidents to the regulatory authority in which the user is established.

This product may contain components of animal origin. All components of animal origin have been sourced from Bovine Spongiform Encephalopathy- (BSE-) free and Transmissible Spongiform Encephalopathy- (TSE-) free countries. Certified knowledge of the origin of animal derived components does not guarantee the absence of transmissible pathogenic agents. It is recommended that Universal Precautions be observed.

When working with anaerobic culture media, the potential for ergonomic hazards may exist due to repetitive motions, awkward postures, improper bench/chair heights or poor lighting. Although it is beyond the scope and provision of products by Anaerobe Systems, it should be recognized and mitigated by the end user in the laboratory environment.

#### STORAGE AND SHELF LIFE

**Storage:** Upon receipt, store at room temperature  $(15 - 25^{\circ}C)$  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 the 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. The selection of specimens for culture is made by physicians or scientists collecting the sample.

**Methods for Use:** EYA agar should be inoculated with a pure culture of the organism to evaluate for lecithinase and/or lipase activity. Streak the plate to obtain isolated colonies and immediately place it into an anaerobic atmosphere. Incubate at 35 – 37°C for 18 to 48 hours. Extended incubation may be necessary for the recovery of slow-growing anaerobes or for the detection of lipase activity. For detailed procedures on processing anaerobic cultures, refer to the appropriate 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.



## INTERPRETATION OF RESULTS

EYA agar should support robust growth of obligate anaerobes commonly isolated from a specimen. In addition, the medium supports characteristic lecithinase activity produced by some *Clostridium* species, as well as lipase activity observed in certain *Fusobacterium* and *Clostridium* isolates.

#### **LIMITATIONS**

EYA agar does not provide complete information for the definitive identification of bacterial isolates. Additional test procedures and media are required for full identification. Consult appropriate reference materials for further guidance.

## **QUALITY CONTROL**

The following organisms are routinely used for quality control testing at Anaerobe Systems using the specifications outlined in the CLSI document M22-A3: Quality Control for Commercially Prepared Microbiological Culture Media.

Organism Tested	ATCC® #	Results	Special Reaction
Bacteroides fragilis	25285	Growth	
Prevotella melaninogenica	25845	Growth	
Fusobacterium necrophorum	25286	Growth	Lipase positive
Fusobacterium nucleatum	25586	Growth	
Clostridium perfringens	13124	Growth	Lecithinase positive
Peptostreptococcus anaerobius	27337	Growth	
Staphylococcus aureus	25923	Growth	Lipase positive
Cutibacterium acnes	6919	Growth	
Clostridioides difficile	9689	Growth	

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

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

Organism	ATCC® #	Expected	Special Reactions
		Results	
Bacteroides fragilis	25285	Growth	
Fusobacterium necrophorum	25286	Growth	Lipase positive
Clostridium perfringens	13124	Growth	Lecithinase positive
Clostridioides difficile	9689	Growth	

Physical Appearance: EYA agar should appear opaque and light yellow in color.

 $\mbox{ATCC}^{\mbox{\tiny 0}}$  is a registered trademark of American Type Culture Collection.

# **REFERENCES**

- 1. CLSI. Principles and Procedures for Detection of Anaerobes in Clinical Specimens; Approved Guideline. CLSI document M56-A. Clinical and Laboratory Standards Institute; 2014
- 2. Leber AL, Burnham CA, eds. *Clinical Microbiology Procedures Handbook*. 5th ed. 4 vols. Washington, DC: ASM Press; 2023.



- Carroll KC, Pfaller MA, eds. Manual of Clinical Microbiology. 13th ed. 4 vols. Hoboken, NJ: Wiley-Blackwell; 2023.
- 4. Jousimies-Somer HR, Sutter VL, eds. *Wadsworth-KTL Anaerobic Bacteriology Manual*. 6th ed. Belmont, CA: Star Publishing Company; 2002.
- 5. CLSI. Quality Control for Commercially Prepared Microbiological Culture Media; Approved Standard- Third Edition. CLSI document M22-A3. Wayne, PA: Clinical and Laboratory Standards Institute; 2004.
- 6. U.S. Department of Agriculture, Animal and Plant Health Inspection Service. *Animal Health Status of Regions*. Published March 12, 2025. <a href="https://www.aphis.usda.gov/regionalization-evaluation-services/region-health-status">https://www.aphis.usda.gov/regionalization-evaluation-services/region-health-status</a>
- 7. European Commission. Note for guidance on minimising the risk of transmitting animal spongiform encephalopathy agents via human and veterinary medicinal products (EMA/410/01 Rev. 3). Published March 5, 2011. <a href="https://op.europa.eu/en/publication-detail/-/publication/3392e464-ba89-4ae4-955c-a07f617c8e06/language-en">https://op.europa.eu/en/publication-detail/-/publication/3392e464-ba89-4ae4-955c-a07f617c8e06/language-en</a>

## **GLOSSARY OF SYMBOLS**

SYMBOL	TITLE	DESCRIPTION	STANDARD	REF#
REF	Catalog number	Indicates the manufacturer's catalog number so that the medical device can be identified.	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labelling, and information to be supplied	5.1.1
LOT	Lot number/ Batch code	Indicates the manufacturer's batch code so that the batch or lot can be identified.	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labeling, and information to be supplied	5.1.5
	Use-by date	Indicates the date after which the medical device is not to be used.	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labelling, and information to be supplied	5.1.4
XX REP	Authorized Representative	Indicates the Authorized Representative in the identified country or jurisdiction.	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labelling, and information to be supplied	5.1.2
<b>(2)</b>	Do not re-use/ Single use only	Indicates a medical device that is intended for one single use only.	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labelling, and information to be supplied	5.4.2
[]i	Consult instructions for use or consult electronic instructions for use	Indicates the need for the user to consult the instructions for use.	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labelling, and information to be supplied	5.4.3
*	Temperature limit	Indicates the temperature limits to which the medical device can be safely exposed.	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labelling, and information to be supplied	5.3.7
IVD	In vitro diagnostic medical device	Indicates that a medical device is intended to be used as an in vitro diagnostic medical device	ISO 15223-1 Medical devices – Symbols to be used with medical device labels, labelling, and information to be supplied	5.5.1
CE	CE Mark European Conformity	Designates that the product labeled Is authorized for sale in European countries.	EU IVDR (EU) 2017/746	



#### AUTHORIZED REPRESENTATIVE INFORMATION



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**REVISION 2** 

Additions: Intended Use, Intended Users, Animal Origin Statement, Ergonomics Precautions, Serious Incident Report Contact Information, Glossary of Symbols Changes: Title change from Product Insert to Instructions for Use. Room temperature from 20 – 25°C to 15 – 25°C. References updated. Contact information. Deletions: None