

Wallerstein Labs Differential (WLD) Agar

Intended Use

Wallerstein Labs Differential (WLD) agar is a medium used to select for and differentiate between bacteria found in beer and the brewery while simultaneously inhibiting the growth of brewing yeast.

Summary and Explanation

WLD agar was designed by Green and Gray while researching methods to isolate common organisms found in fermenting media. They had already developed a nutrient medium for the cultivation of organisms commonly found in fermenting beer which contained an acid color indicator (WL Nutrient Agar). The aim was then to alter this medium to inhibit the growth of brewing yeast (*S. cerevisiae*) while allowing for the growth of possible spoilage organisms. By adding cycloheximide to the medium, Green and Gray were able to prevent the growth of most common brewing yeast.

Principles

Yeast extract provides a source of nutrients and amino acids for microbial growth. Pancreatic digest of casein is added as an additional source of nitrogen and amino acids. Dextrose provides a carbohydrate nutrient source. Monopotassium phosphate is included as a medium buffer. Potassium chloride, calcium chloride, ferric chloride, manganese sulfate and magnesium sulfate are added for osmotic balance, essential nutrients and buffering. Bromocresol green is included as an acid color indicator. Cycloheximide (actidione) is added to prevent the growth of yeast and mold. Agar acts as a solidifying agent.

Physical Appearance

WLD agar appears in dehydrated form as a homogenous, free flowing powder with a blue green tint.

When prepared, WLD appears as a homogenous clear solid agar without apparent particulate. The media appears blue-green in color prior to inoculation and incubation.

Storage and Shelf Life

Stored dehydrated media in a cool, dry area not exceeding 30° C until expiration date listed on bottle. Store lid tightly between use. Media is highly hygroscopic and will readily absorb moisture. Discard media if media is not free flowing and if premature solidification has occurred.

Do not hold prepared media above 40° C in excess of 4 hours to prevent deterioration of nutrients.

Store prepared petri dishes between 2-8° C for up to 14 days. It is recommended that petri dishes be stored in a seal container to prevent the loss of moisture leading to desiccation.

Precautions and Safety Information

This product contains low concentrations of cycloheximide, a known toxin and carcinogen. Do not ingest and prevent extended contact with the medium in dehydrated or prepared form. Avoid exposure to inhalation of dehydrated powder. When preparing medium, it is recommended that proper safety apparel be worn at all times, including gloves and a dust mask where applicable.

Refer to the Safety Data Sheet for specific hazards, preparation and disposal instructions.

Sterilize all biohazardous waste prior to disposal.

Directions for Preparation

1. Suspend 80 grams of the dehydrated powder in 1 Liter of purified or deionized water. Mix thoroughly.
2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
3. Autoclave at 121° C for 15 minutes.
4. Cool to approximately 45° C.
5. While still warm, pour the medium into sterile petri dishes.
6. Cool the prepared plates until solidified. Store until use.

Final pH 5.5 ± 0.2 at 25° C

Directions for Use

1. Inoculate the prepared WLD plates with desired sample.
2. Incubate aerobically or anaerobically for at least 48 hours at 30-35° C
3. Interpret growth for beer spoilage organisms.
 - a. Appearance of a pale beige halo around colonies indicates an acid pH change. Strong acid production may lead to de-coloration of all the prepared medium

Cultural Response

Organism	Recovery
<i>Lactobacillus brevis</i>	Good
<i>Enterobacter</i> spp.	Good
<i>Saccharomyces cerevisiae</i>	None

References

1. Green, S.R. and P.P. Gray. 1950. Paper read at American Society of Brewing Chemists Meeting. Wallerstein Lab. Commun.; 12:43.
2. Green, S.R. and P.P. Gray. 1950. A differential procedure applicable to bacteriological investigation in brewing. Wallerstein Lab. Commun.; 13:357.

Availability

Weber Scientific LMDA Medium, 500 grams

Cat. No. 3118-20

Visit weberscientific.com or contact info@weberscientific.com for availability and pricing info.