User Manual

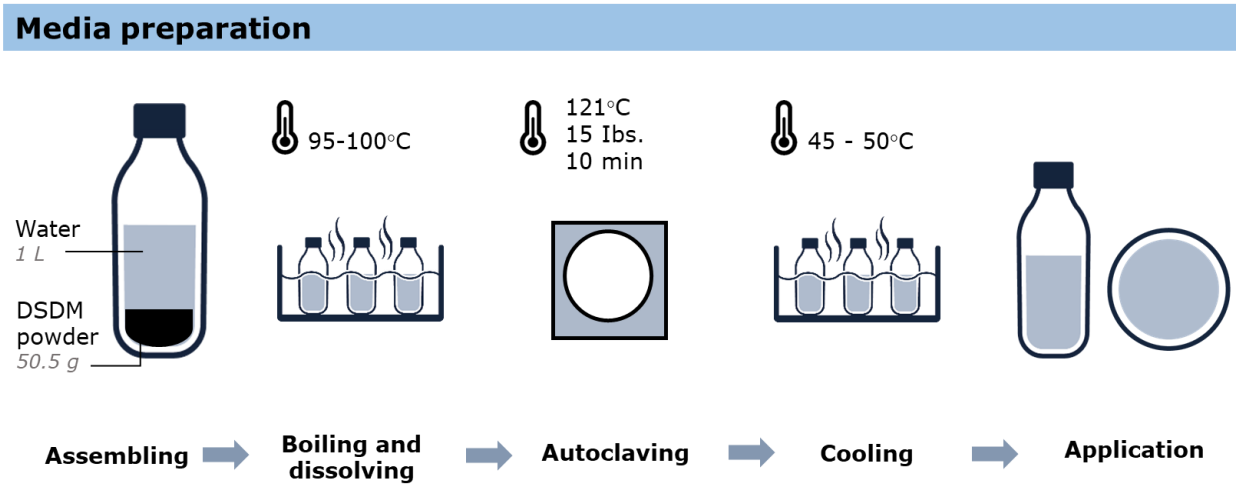
DSDM®
Item No. 9.71231.244

DSDM® (Döhler *Saccharomyces diastaticus* Medium) is a complex nutrient medium (pH 4.5-5.0) for the selective detection of super-attenuating yeast, in particular Diastatic *Saccharomyces cerevisiae* (formally known as *S. cerevisiae* var. *diastaticus*) commonly encountered in a brewery environment. This strain is described as a super-attenuating yeast due to its ability to ferment long chain carbohydrates in beer (dextrins and soluble starch), which are normally not metabolized by culture yeast strains. Diastatic *S. cerevisiae* carries a STA gene encoding for the enzyme glucoamylase which degrades starch and higher wort dextrins to fermentable sugars. This subsequent secondary fermentation leads to formation of excess CO_2 which may cause bottle gushing or even dangerous bursting of beer stored in bottles or cans. Other damage patterns include unwanted alcohols, off-taste, turbidity and sedimentation. DSDM® is applied as a broth or by adding agar as a solid medium. It provides the necessary growth substances for the growth of super-attenuating yeasts, whereas the growth of culture yeasts is largely suppressed. In addition, a colour indicator is incorporated which changes from violet/blue to yellow/greenish when positive.

1. Media Preparation

DSDM® Powder is a ready-to-use mixture for the production of **DSDM® Broth**. Dissolve 50.5 g of DSDM® in 1L distilled water and seal with an appropriate permeable closure. To prepare **DSDM® Agar**, add an additional 20 - 30 g agar-agar (2.0 – 3.0 %) per litre of ready-to-use medium.

Bring the content of the flask to boil until medium is completely dissolved. Swirl the flask frequently to avoid sticking or scorching of media when direct fire is used for heating. Autoclave medium at 121 °C for 10 minutes to sterilize. After autoclaving, cool the medium in a water bath at 45 - 50 °C. The medium can be stored in a dry, dark and cool place at 4 - 8°C for later use.



Note: A deviating temperature and/or autoclaving time can have a negative impact on the quality of the medium. For storage and later use, store the medium in a dark, dry and cool place at 4 – 8 °C.

2. Application

Please work under sterile conditions to avoid secondary contamination of the samples

2.1 Liquid pre-enrichment

For yeast samples with high cell count (e.g. young beer, harvest or propagation yeast) the cell concentration should be determined and diluted to about 5 million cells/ml. Mix 1 ml of yeast suspension with 10 ml of DSDM® broth in a sterile sample container. Ensure that at least 30% headspace is available for aerobic incubation. Additionally, a sterile Durham tube can be used to detect and measure CO2.

Tip:
In particular, for the detection of slow-growing germs or low bacterial counts, a combination of liquid pre-enrichment and subsequent incubation on solid medium is recommended. Therefore, after an incubation for 5 days in broth, transfer 3 mL to a agar plate and incubate it for another 2–5 days.

2.2 Membrane filtration

For filterable samples like water, filtered beer or samples containing low populations of yeast, filter about 100 -150 ml sample through a non-cellulose membrane filter. Then filter about 300ml of sterile water to wash the membrane. Transfer the membrane filter into sealable glass tubes filled with 15 ml of DSDM® broth or for quantitative analysis onto a petri dish with DSDM® agar.

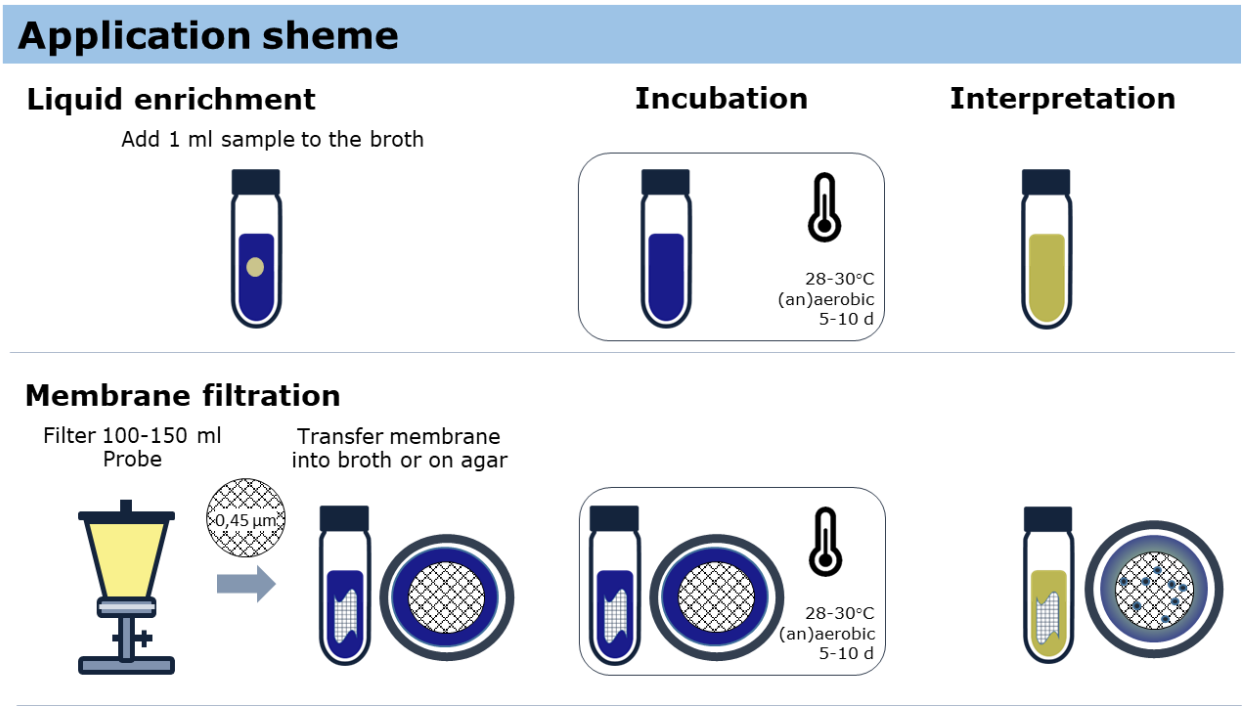
2.3 Incubation

Incubate the sample under aerobic conditions for 5 to 10 days at a temperature of 28-30 °C. An incubation at 36 °C is advantageous while testing for bottom-fermenting yeast as this inhibits competitor yeast. An anaerobic incubation is recommended for specific detection of Diastatic *S. cerevisiae*.

If the detection of beer spoilage yeasts, e.g. Diastatic *Saccharomyces cerevisiae*, is to be carried out, anaerobic incubation is recommended in order to exclude e.g. respiratory yeasts, which also utilize long-chain carbohydrates.

Incubation at higher temperatures or under anaerobic conditions, may require longer incubation times.

Application sheme

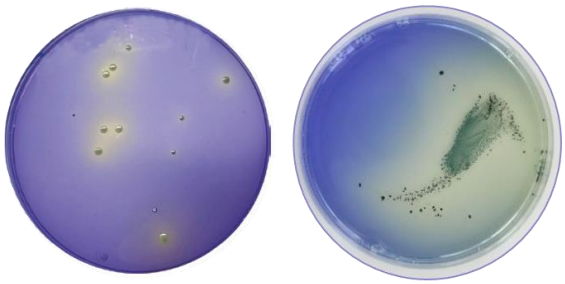


3. Evaluation and Result Interpretation

A positive result is indicated by colony growth and additional change of the color indicator from violet to green/yellow. Depending on the number and type of yeast, growth may be observed earlier. A first evaluation is recommended after 3 days, the final between 5 - 10 days.

A)

B)



A: DSDM®-Broth: Negative (left), Positive (mid), Positive with membrane filter (right)

B: DSDM®-Agar: Spread plated (left) and Streak (right)

Note: Residual growth of culture yeast cannot be excluded 100% which could lead to a slight color change. Strongly fermenting yeasts for achieving a higher final fermentation degree must be considered.

4. Product Information

4.1 Packaging and content

Package/content :	Plastic bottle with a 200 g content
Size :	16 cm x 7 cm x 7 cm
Weight :	0.26 kg

4.2 Storage and shelf life

Store in a dry and dark place at 4 – 8 °C. Shelf life of 730 days (from production date).

4.3 Waste disposal

- For information on dangerous goods or hazardous substances, please refer to the safety data sheet
- Please consider your local waste regulations
- Non inoculated media can be disposed of with normal laboratory waste
- Inoculated and incubated media should be sterilized for 20 min at 121 °C

4.4 Warnings and precautions

Do not overheat or freeze product. Wear protective clothing when handling hot media. This product is for use in microbiological control only and not intended for medical use. More information in SDS.

5. Quality Control

DSDM® was tested for functionality with the following microorganisms.

Microorganism	Growth aerobic
Saccharomyces cerevisiae ale (BRY 96)	negative
Saccharomyces cerevisiae lager (BRY 420)	negative
Saccharomyces pastorianus TUM (34/70)	negative
Diastatic *Saccharomyces cerevisiae*	positive

6. Similar Products

Product	Item No.	Target microorganism
LWYM	9.23552.244	Detection of wild yeast
LCSM	9.23556.244	Detection of non-saccharomyces wild yeast

7. References

[1] Hutzler, M (2009). Entwicklung und Optimierung von Methoden zur Identifizierung und Differenzierung von getrankerelevanten Hefen. Technische Universität München

[2] Meier-Dörnberg, T., Kory, O. I., Jacob, F., Michel, M., & Hutzler, M., (2018). *Saccharomyces cerevisiae* variety diastaticus friend or foe? – spoilage potential and brewing ability of different *Saccharomyces cerevisiae* variety diastaticus yeast isolates by genetic, phenotypic and physiological characterization. 18(4)