



## BRETT-SD™

## Brettanomyces/Dekkera Selective &amp; Differential Agar Plates

Product # Brett SD-6 (6/Pack)  
Brett SD-48 (48/Case)

**Intended Use**

For the detection and cell counts of Dekkera/ Brettanomyces in juice or wine samples - this media is highly selective has a new proprietary spectrum of selection optimized for Brettanomyces in wine samples. The differentially media promotes relatively fast growth of this slow-growing organism. It is enriched with growth factors and specific nutrients including carbon sources specific for Brett; the media inhibits bacteria, other yeast & mold. Brettanomyces tends to settle to the bottom of barrels/tanks and bottom samples should (or mixed wine samples) are recommended.

**Procedure**

- Remove** parafilm™ from the plate(s). Apply 50 uL of the wine using a sterile (or clean) pipette and spread with sterile cell spreader -- OR Concentrate wine by filtering through a 0.45-µm pore membrane and place the filter on the surface of the agar. Avoid trapping air bubbles between the membrane and agar.
- Cover plate** and seal in supplied self-sealing bag. Incubate plates at 25 °C–30 °C (or at room temperature) for several days. Plate orientation is not important; e.g. they may be stored upright in a file box for easy reference.
- Examine** the plates after 1–3 days, recording color change and phenolic odor. Examine over 12 days, recording the number of colonies.



- Consider positive** if:
  - A phenolic medicinal or leathery odor is detected when opening the plate, generally in 24 hours. *Suggestion: Compare odor to an unused plate, and to the Le Wine Brettanomyces Sensory Set™ (sold separately).*
  - Color of agar changes from blue to yellow within 1–12 days
  - Colony growth is detected between 3–12 days; colonies appear cheesy, whitish-yellow

**Statistically Positive Results** - 3 to 10 distinct colonies are considered positive. *If you observe 1 or 2 colonies, repeated test in triplicate to verify quantitation of Brett – or concentrate wine by microfiltration, place the 0.45u membrane on the agar and retest (see 1 above.)*

- Interpretation - Cell Counts (Wine as Sample)**  
Multiply observed colonies by 20 for colony forming units (cfu)/mL (*note: 5-Drops = 50uL*).  
**Detection Limit:** 3 colonies/50uL = 60 cfu/mL of (unconcentrated) wine.

**Interpretation of Concentrated Samples:** *If testing microfilter concentrated wine, divide the colonies observed by the volume of wine tested to obtain cfu/mL. e.g. Detection Limit for 1 Liter of microfiltered wine is 3 colonies/L.*

**TNTC:** Record that your sample is TNTC (Too Numerous To Count) i.e. contains over 10,000 cfu/mL, if:

- Agar color changes
- Penolic odor development is rapid (1–3 days)
- A haze develops over the entire plate.

*Brettanomyces* at high levels produce phenolics that taint the wine.

Possible action to take for TNTC results:

- Immediately filter or otherwise sanitize the wine.
- Repeat Easy Blue testing with 1:10 serial dilutions to determine the exact cell concentration and confirm the extent of Brettanomyces contamination.
- Retest after remediation to make sure that it was effective.

**Precautions**

- Minimize the time that the top is off the dish. Work in a draft-free environment.
- Avoid directly touching the agar medium
- Avoid cross contamination in the winery - when the test has been completed applied either 10% bleach or 70% alcohol to the plate and beads prior to disposal.

**Storage**

Store refrigerated between 2–8°C.

**Related Unitech Products**

- BioSart 100 Monitors** (inquire re: additional Unitech Scientific 0.45u membrane products) for wine concentration by filtration
- Uni-Barrel Pipette** - Curved barrel racking tube with pipette bulb for controlled collecting (and releasing) of 15-20mL bottom samples.
- Centrifuge** e.g. Microfuge Unitech *PN: LX-100*, \$180) 1.5mL centrifuge tube capability

Manufactured by Le Wine Lab, Irvine CA

**Distributed by: Unitech Scientific LLC**

12026 Centralia Road Suite H,  
Hawaiian Gardens, CA 90716

Tel: 562-924-5150 Fax: 562-809-3140

[info@unitechscientific.com](mailto:info@unitechscientific.com)