## SIEBEL LABORATORY MEDIA TECHNICAL DATA SHEET

# LWYM Lin's Wild Yeast Medium

Siebel Institute LWYM is used for the detection and quantitative determination of wild yeast populations in brewing culture yeast.

Approximately 1 million culture yeast is plated on LWYM. While the growth of culture yeast is suppressed by the crystal violet, wild yeast will develop as larger, distinct colonies. While this medium is designed to encourage the growth of Saccharomyces wild yeasts primarily, a number of non-Saccharomyces yeast will also be able to grow on this medium.

#### 1. PREPARATION OF LWYM PLATES

- A. This medium is prepared from two separate containers, using the following ingredients:
  - 1. LWYM dehydrated powder
  - 2. Crystal Violet solution

The crystal violet concentration has been adjusted to give optimum conditions for wild yeast growth and culture yeast inhibition. It is absolutely essential to use the crystal violet solution, of the same lot number, that is provided with the LWYM dehydrated powder.

- B. Suspend 4.4 grams of L.W.Y.M. in 100 ml of distilled water in a 500 ml Erlenmeyer flask and add 1.0 ml of crystal violet solution
- C. Heat to boiling to dissolve the medium. Swirl the flask frequently to avoid caking or scorching.
- D. Autoclave at 121 °C (15 lbs. Pressure) for 15 minutes to sterilize the medium. Remove promptly from the autoclave.
- E. Transfer the flask to a water bath at 45 °C as soon as possible after sterilization. Once cooled, pour about 15 ml of medium into 6 sterile petri dishes and leave them undisturbed so that they can solidify.

Poured plates can be stored in inverted position at 4°C for 24 to 48 hours before use, but should be used within 5 days of preparation because the medium will inhibit the growth of wild yeast if allowed to stand for extended periods of time.



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#### 2. DETECTION OF WILD YEAST - Samples with high yeast counts

- A. For samples containing high populations of yeast cells (such as pitching yeast or yeast in storage tank), the cell concentration needs to be determined using a hemocytometer.
- B. Dilute the yeast slurry to have approximately 5 million cells /ml to start with.
- C. Inoculate 0.2 ml diluted sample containing approximately 1 million yeast cells onto a LWYM plate.
- D. Disperse the inoculum evenly over the surface of the medium, using a sterile spreader. Spread the sample near, but not onto the edges of the plate.
- E. Incubate the plates upside down under aerobic conditions at 28-30 °C.
- F. Examine plates after 4 to 6 days. Distinct colonies developed on the medium may be considered wild yeast.\*\*\*

### 3. DETECTION OF WILD YEAST - Samples with low yeast counts

- A. For samples containing low populations of yeast cells (such as unpasteurized beer, draft beer), filter the sample through a non-cellulose membrane filter (e.g. Nuclepore N40 CPR 04700 or equivalent), followed by the filtration of 300 ml sterile water to wash and remove any extraneous material from the filter.
- B. Transfer the membrane filter from the filtering apparatus to the surface of a LWYM plate avoiding the formation of air bubbles.
- C. Incubate the plates aerobically at 28-30 °C.
- D. Examine the plates after 4 to 6 days. Distinct colonies that develop on the membrane filter may be considered wild yeast. \*\*\*
- E. Report the number of wild yeast per unit of starting material: pitching yeast, process beer, or finished product.

\*\*\* Some strains of culture yeast may show slight growth on LWYM, which is why only distinct colonies are considered as wild yeast. Moreover, some fast growing wild yeasts (e.g. S. willianus or Candida mycoderma) will enhance the growth of culture yeast. Therefore, some colonies of culture yeast may show slight growth in the surrounding of wild yeast colonies.

### 4. ORDERING INFORMATION

Product: LWYM Media

Product Code: M9200 Packaging Size: 200g

For prices and online ordering, please visit our website at <a href="www.siebelinstitute.com/products/">www.siebelinstitute.com/products/</a>
You can also send your order by email to <a href="mailto:brewingcanada@siebelinstitute.com">brewingcanada@siebelinstitute.com</a>

#### 4. CONTACT INFORMATION

For questions regarding this product or how our laboratory media can support your quality assurance/control programs, please contact us at <a href="mailto:lab.media@siebelinstitute.com">lab.media@siebelinstitute.com</a> and a member of our technical support team will follow up on your request.

