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L.C.S.M. (Lin's Cupric Sulfate Medium) Instructions

Derived from Siebel Institute of Technology

Propose: Used for detection of primarily non-*saccharomyces* wild yeast.

1. For samples containing high populations of yeast cells (such as pitching yeast or yeast in storage tank):

    a. Determine the cell concentration using a hemocytometer. Dilute the yeast slurry to approximately 5 million cells/ml. Inoculate 0.2 ml diluted sample containing approximately 1 million yeast cells onto L.C.S.M. Disperse the inoculum over the surface of the medium, using a sterile cell spreader. Incubate plates aerobically at room temperature. Culture will grow faster if incubated at 82 ^{o}F. Examine plates after 4 to 6 days. Distinct colonies developed on the medium may be considered as wild yeasts.

2. For samples containing low populations of yeast cells (such as unpasteurized beer, draft beer):

    a. Filter sample through a membrane filter followed by 300 ml sterile water to wash and remove any extraneous material from the filter. Then, transfer the membrane filter from the filtering apparatus to the surface of L.C.S.M. in the petri dish. Incubate plates aerobically at room temperature for 4 to 6 days; culture will grow faster if incubated at 82 ^{o}F. Distinct colonies that develop on the membrane filter may be considered as wild yeasts.