
Appendix A: Media and Reagent**1. Potato Dextrose Agar (PDA)**

39g of PDA was suspended in 1 L of distilled water and autoclaved for 15 min. at 15 psi, 121 °C. Subsequently, the medium were cooled to 45 °C – 50 °C and dispensed into Petri dishes. Final pH should be 5.6 ± 0.2 at 25 °C.

2. Yeast – Peptone – Glucose agar (YPG)

10 g of yeast, 20 g of peptone, 20 g of glucose and 40 g of Bacto agar were suspended in 1 L of distilled water and autoclaved for 15 minutes at 15 psi, 121 °C. Subsequently, the medium were cooled to 45 °C – 50 °C and dispensed into Petri dishes.

3. Sabouraud Dextrose Agar (SDA)

65 g SDA and 15 g Bacto agar were suspended in 1 L of distilled water and autoclaved for 15 minutes at 15 psi, 121 °C. Subsequently, the medium were cooled to 45 °C – 50 °C and dispensed into Petri dishes.

4. Glucose – yeast – malt – peptone agar medium (GYMP)

0.5 g of magnesium sulphate ($\text{MgSO}_4 \cdot \text{TH}_2\text{O}$), 0.46 g potassium dihydrogen orthophosphate (KH_2PO_4), 1.0 g di – potassium hydrogen (K_2HPO_4), 20 g glucose, 2.0 g peptone, 2.0 g yeast extract, 2.0 g malt extract and 18.0g of Agar were suspended in 1 liter of distilled water. Then, the solution was transferred into conical flasks and the top was covered with cotton bud and alluminium foil. The solution was autoclaved for 15 minutes at 15 psi, 121 °C. Subsequently, the medium were cooled to 45 °C – 50 °C and dispensed into Petri dishes.

5. Malt extract agar (MEA)

20 g of malt extract and 20 g of Bacto agar were suspended in 1L of distilled water and autoclaved for 15 minutes at 15 psi, 121 °C. Subsequently, the medium were cooled to 45 °C – 50 °C and dispensed into Petri dishes

Appendix B: Preparation for Lignin Peroxidase enzymatic activity.

Preparation of sodium tartarate buffer solution (0.1 M), pH = 3.49, T = 23.9⁰C.

Sodium Tartarate	11.5 g
Distilled water	200.0 ml

The pH was adjusted with 1 N tartaric acid solution:

Tartaric acid	7.51 g
Distilled water	500.0 ml

The standard (stock solution):

3.9 mg of 2, 3 – dimethoxybenzaldehyde (powder) {C₉H₁₀O₃} were dissolved in 20 ml of sodium tartarate buffer solution. It was sparingly soluble in distilled water. Final concentration of stock solution was 0.195 mg / ml.

The Reagent Blank:

H ₂ O ₂	0.2 ml
Veratryl alcohol	0.2 ml
Sodium tartarate buffer	2.6 ml

The assay mixture consisted of

H ₂ O ₂	0.2 ml
Veratryl alcohol	0.2 ml
Filtrate (enzyme)	0.2 ml
Sodium tartarate buffer	2.4 ml

One unit enzyme activity was expressed as the amount of enzyme required for an increase in absorbance of 1.0 μ / min. peroxidase activity can also be assayed with L – 3, 4 – dihydroxy phenyalanine (L – DOPA; Sigma) and 2, 4 – dicholorophenol (2, 4 – DCP; Sigma). This was measured using spectrophotometer.