

Supplementary Information

Optimization and kinetic modeling of *Trametes maxima* HPLC-32 laccase and application in recalcitrant dye degradation.

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Fig. S1.

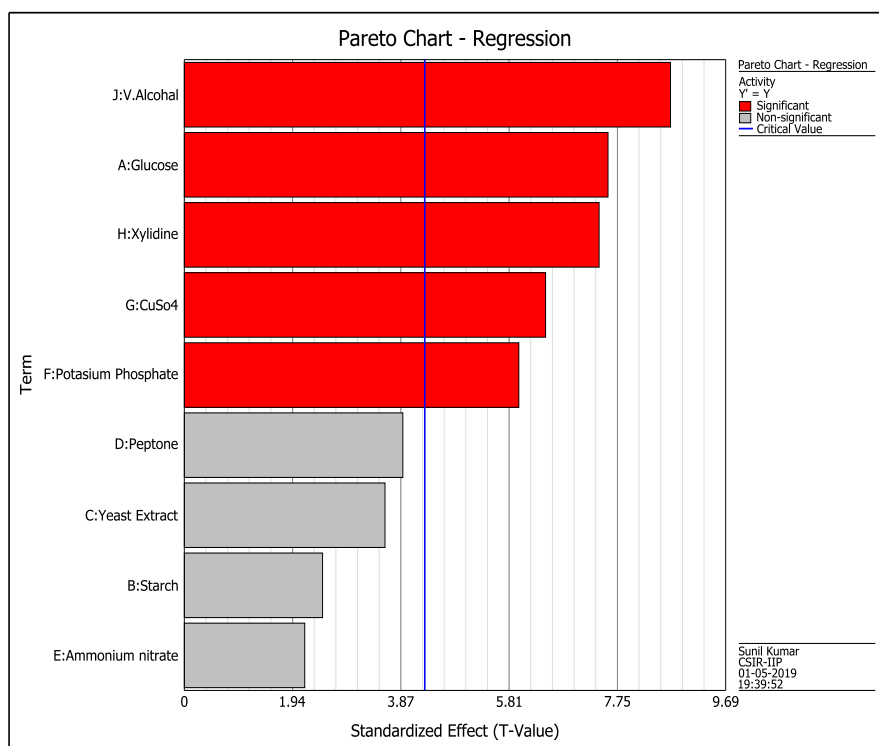


Fig. S1. Pareto chart showing the effect of various factors on laccase production. Significant factors are in red while the non-significant factors are in gray. The critical value is represented in blue.

Fig. S2.

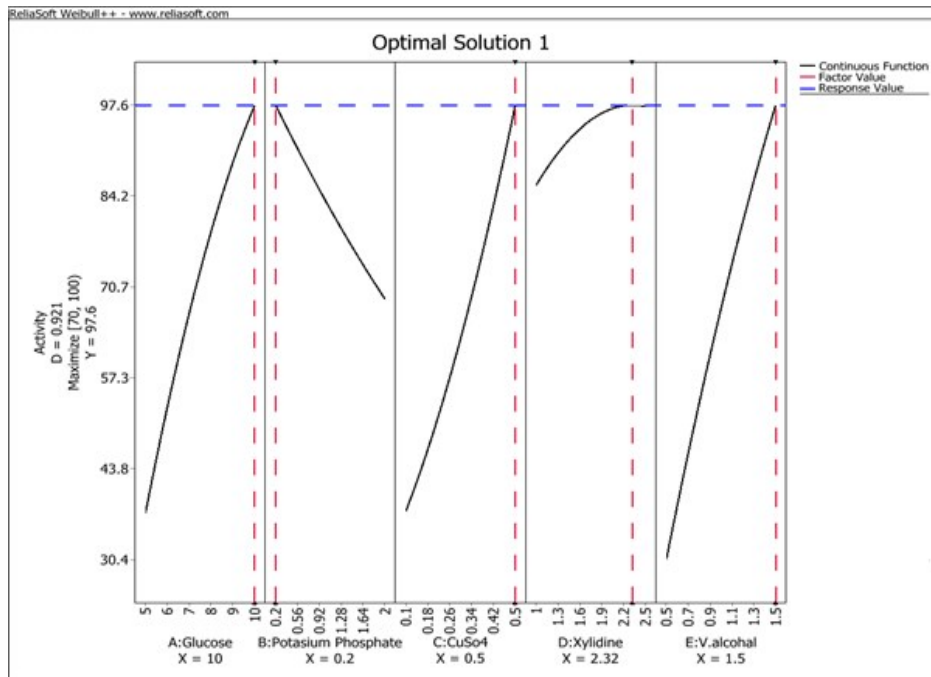


Fig. S2. Optimized conditions for the laccase production predicted by reliasoft DOE.

Table S1 Assigned concentration of variables at different levels in Plackett–Burman Design (PBD) for Screening of factors.

S.No.	Variables with designation	Lower level (-)	Higher level (+)
1	Glucose (A)	5 (g L ⁻¹)	10 (g L ⁻¹)
2	Starch (B)	5 (g L ⁻¹)	10 (g L ⁻¹)
3	Yeast extract (C)	2.5 (g L ⁻¹)	5 (g L ⁻¹)
4	Peptone (D)	2.5 (g L ⁻¹)	5 (g L ⁻¹)
5	Ammonium nitrate (E)	0.2 (g L ⁻¹)	2 (g L ⁻¹)
6	Potassium Phosphate (F)	0.2 (g L ⁻¹)	2 (g L ⁻¹)
7	CuSO ₄ (G)	0.1 (mM)	0.5 (mM)
8	Xylidine (H)	1 (mM)	2.5 (mM)
9	V. Alcohol (I)	0.5 (mM)	1.5 (mM)

Table S2 Concentration of variables used in Box –Behnken Design (BBD) for optimization.

S.N.	Variable	Low	Center point	High
1	Glucose (g L ⁻¹)	5.0	7.5	10
2	Potassium phosphate (g L ⁻¹)	0.2	1.1	2.0
3	CuSO ₄ (mM)	0.1	0.3	0.5
4	Xylidine (mM)	1.0	1.75	2.5
5	Veratryl alcohol (mM)	0.5	1.0	1.50

Table S3 Effect of compounds on laccase production individually and in combination.

	Compounds	Laccase activity (IU mL⁻¹)
Carbon source	Glucose	1.01±0.32
	Sucrose	0.34±0.15
	Maltose	0.57±0.14
	Lactose	0.37±0.12
	Starch	0.61±0.11
	Glucose + Sucrose	1.23±0.13
	Glucose + Maltose	1.32±0.10
	Glucose + Lactose	0.99±0.21
	Glucose + Starch	1.54±0.28
Nitrogen source	Ammonium Nitrate	1.21±0.31
	Sodium Nitrate	0.54±0.11
	Yeast Extract	1.15±0.30
	Peptone	1.01±0.29
Inducer	No inducer	0.34±0.01
	CuSO ₄	3.88±1.17
	Xylidin	2.72±0.47
	Veratryl Alcohol	2.55±0.70
	CuSO ₄ + Xylidin+ Veratryl Alcohol	7.77±0.78

