SUPPLEMENTARY MATERIALS

Degradation of a leather-dye by the combination of depolymerised wood-chip biochar adsorption and solid-state fermentation with *Trametes villosa* SCS-10

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# LIST OF TABLES AND FIGURES

Table 1.Physical and chemical characteristics of the leather-dye AB161.

Table 2. Analyse of Variance (ANOVA) of the obtained model for dye removal.

Table 3. Analyse of Variance (ANOVA) and Duncan's multiple range test for dye removal by adsorption.

Table 4. Analyse of Variance (ANOVA) and Duncan's multiple range test for the desorption assays.

Table 5. Analyse of Variance (ANOVA) and Duncan's multiple range test for fungal growth.

Table 6. Analyse of Variance (ANOVA) and Duncan's multiple range test for the laccase activity.

Figure 1.Calibration curve: absorbance *vs.* concentration of AB161.

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| Table 1 Physical and chemical characteristics of the leather-dye AB161 | | | | |
| Dye (CI)a | CAS number a | λMax (nm) b | MW (g mol-1) c | Chemical structure |
| Acid Blue 161 (C.I. 15706) | 12392-64-2 | 578 | 394.40 | C20H13N2O5SNaCrx |
| a Colour Index International Classification, b CAS: Chemical Abstracts Service number; c Wavelength of maximum absorbance; d Molecular weight | | | | |

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| Table 2. Analyse of Variance (ANOVA) of the obtained model for dye removal. | | | | | | | |
| Source | Sum of Squares | df | Mean square | F value | p-value | Significance |
| Model | 4081.32 | 9 | 453.48 | 1729.90 | < 0.0001 | \* |
| X1 – pH | 147.92 | 1 | 147.92 | 564.27 | < 0.0001 | \* |
| X2 – Dye concentration | 3026.42 | 1 | 3026.42 | 11544.93 | < 0.0001 | \* |
| X3 – Sorbent dosage | 38.72 | 1 | 38.72 | 147.71 | < 0.0001 | \* |
| X1X2 | 54.02 | 1 | 54.02 | 206.08 | < 0.0001 | \* |
| X1X3 | 12.60 | 1 | 12.60 | 48.07 | 0.0002 | – |
| X2X3 | 1.82 | 1 | 1.82 | 6.95 | 0.0336 | – |
| X12 | 44.13 | 1 | 44.13 | 168.35 | < 0.0001 | \* |
| X22 | 646.11 | 1 | 646.11 | 2464.71 | < 0.0001 | \* |
| X32 | 58.82 | 1 | 58.82 | 224.37 | < 0.0001 | \* |
| Residual | 1.83 | 7 | 0.26 |  |  | – |
| Lack of Fit | 1.49 | 3 | 0.50 | 5.86 | 0.0602 |  |
| Pure Error | 0.34 | 4 | 0.09 |  |  |  |
| Cor Total | 4083.16 | 16 |  |  |  |  |
| (\*) Significant; (–) Not significant. | | | | | | |

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| Table 3. ANOVA and Duncan's multiple range test for dye removal by adsorption | | | | | | | | | |
| Treatments: (4) AC, KWB, NWB, WB | | | | | | | | | |
| Source | | Sum of Squares | | | df | Mean Square | | F Value | Pr > F |
| Model | | 0.11627833 | | | 3 | 0.03875944 | | 484.63 | <.0001 |
| Error | | 0.00063982 | | | 8 | 0.00007998 | |  |  |
| Corrected Total | | 0.11691816 | | | 11 |  | |  |  |
| R-Square | | 0.994528 | | |  |  | | | |
|  | | | | | | | | | |
| Source | Type | | | | df | Mean Square | | F Value | Pr > F |
| Treatments | Type I: 0.11627833 | | | | 3 | 0.03875944 | | 484.63 | <.0001 |
| Type III: 0.11627833 | | | | 3 | 0.03875944 | | 484.63 | <.0001 |
|  | | | | | | | | | |
| Level of treatment | | | N | Mean (Response) | | | Std Dev | | |
| AC | | | 3 | 0.47925463 | | | 0.01493833 | | |
| KWB | | | 3 | 0.43886977 | | | 0.00367603 | | |
| NWB | | | 3 | 0.30493719 | | | 0.00793799 | | |
| WB | | | 3 | 0.23599899 | | | 0.00449805 | | |
| Alpha | | | 0.05 |  | | | | | |
| Error Degrees of Freedom | | | 8 |
| Error Mean Square | | | 0.00008 |

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| Table 4. ANOVA and Duncan's multiple range test for desorption assays | | | | | | | | |
| Treatments: (7) CH3COOH, Ethanol, HCl, Methanol, NaCl, NaOH and Water | | | | | | | | |
| Source | Sum of Squares | | | df | Mean Square | | F Value | Pr > F |
| Model | 0.61609128 | | | 6 | 0.10268188 | | 151.34 | <.0001 |
| Error | 0.00949899 | | | 14 | 0.00067850 | |  |  |
| Corrected Total | 0.62559027 | | | 20 |  | |  |  |
| R-Square | 0.984816 | | |  |  | | | |
|  | | | | | | | | |
| Source | Type | | | df | Mean Square | | F Value | Pr > F |
| Treatments | Type I: 0.61609128 | | | 6 | 0.10268188 | | 151.34 | <.0001 |
| Type III: 0.61609128 | | | 6 | 0.10268188 | | 151.34 | <.0001 |
|  | | | | | | | | |
| Level of treatment | | N | Mean (Response) | | | Std Dev | | |
| CH3COOH | | 6 | 0.04198430 | | | 0.00990222 | | |
| Ethanol | | 6 | 0.49294566 | | | 0.04789024 | | |
| HCl | | 6 | 0.02586378 | | | 0.00246508 | | |
| Methaol | | 6 | 0.37836050 | | | 0.04005863 | | |
| NaCl | | 6 | 0.18794901 | | | 0.00924390 | | |
| NaOH | | 6 | 0.21207961 | | | 0.02543235 | | |
| Water | | 6 | 0.02003271 | | | 0.00386553 | | |
| Alpha | | 0.05 |  | | | | | |
| Error Degrees of Freedom | | 14 |
| Error Mean Square | | 0.000678 |

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| Table 5. ANOVA and Duncan's multiple range test for fungal growth  Treatments: (4) T1, T2, T3 and T4 | | | | | | | | |
| Source | Sum of Squares | | | df | Mean Square | | F Value | Pr > F |
| Model | 1.00992710 | | | 3 | 0.33664237 | | 4122.42 | <.0001 |
| Error | 0.00065329 | | | 8 | 0.00008166 | |  |  |
| Corrected Total | 1.01058039 | | | 11 |  | |  |  |
| R-Square | 0.999354 | | |  |  | | | |
|  | | | | | | | | |
| Source | Type | | | df | Mean Square | | F Value | Pr > F |
| Treatments | Type I: 1.00992710 | | | 3 | 0.33664237 | | 4122.42 | <.0001 |
| Type III: 1.00992710 | | | 3 | 0.33664237 | | 4122.42 | <.0001 |
|  | | | | | | | | |
| Level of treatment | | N | Mean (Response) | | | Std Dev | | |
| T1 | | 3 | 0.90134962 | | | 0.00397649 | | |
| T2 | | 3 | 0.86761648 | | | 0.00760943 | | |
| T3 | | 3 | 0.52659507 | | | 0.01222805 | | |
| T4 | | 3 | 0.18704728 | | | 0.01016880 | | |
| Alpha | | 0.1 |  | | | | | |
| Error Degrees of Freedom | | 8 |
| Error Mean Square | | 0.000082 |

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| Table 6. ANOVA and Duncan's multiple range test for laccase activity.  Treatments: (4) T1, T2, T3 and T4 | | | | | | | | |
| Source | Sum of Squares | | | df | Mean Square | | F Value | Pr > F |
| Model | 469462.4228 | | | 3 | 156487.4743 | | 80.30 | <.0001 |
| Error | 15590.3292 | | | 8 | 1948.7912 | |  |  |
| Corrected Total | 485052.7521 | | | 11 |  | |  |  |
| R-Square | 0.967858 | | |  |  | | | |
|  | | | | | | | | |
| Source | Type | | | df | Mean Square | | F Value | Pr > F |
| Treatments | Type I: 469462.4228 | | | 3 | 156487.4743 | | 80.30 | <.0001 |
| Type III: 469462.4228 | | | 3 | 156487.4743 | | 80.30 | <.0001 |
|  | | | | | | | | |
| Level of treatment | | N | Mean (Response) | | | Std Dev | | |
| T1 | | 3 | 621.851852 | | | 62.2751098 | | |
| T2 | | 3 | 758.703704 | | | 51.7005066 | | |
| T3 | | 3 | 466.296296 | | | 16.4835621 | | |
| T4 | | 3 | 226.296296 | | | 31.1821280 | | |
| Alpha | | 0.1 |  | | | | | |
| Error Degrees of Freedom | | 8 |
| Error Mean Square | | 1948.791 |

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| **A close up of a white wall  Description automatically generated** |
| Figure 1.Calibration curve: absorbance *vs.* concentration of AB161 |

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