

Supplementary information

Tryptophan, a noncanonical melanin precursor: New L-tryptophan based melanin production by *Rubrivivax benzoatilyticus* JA2

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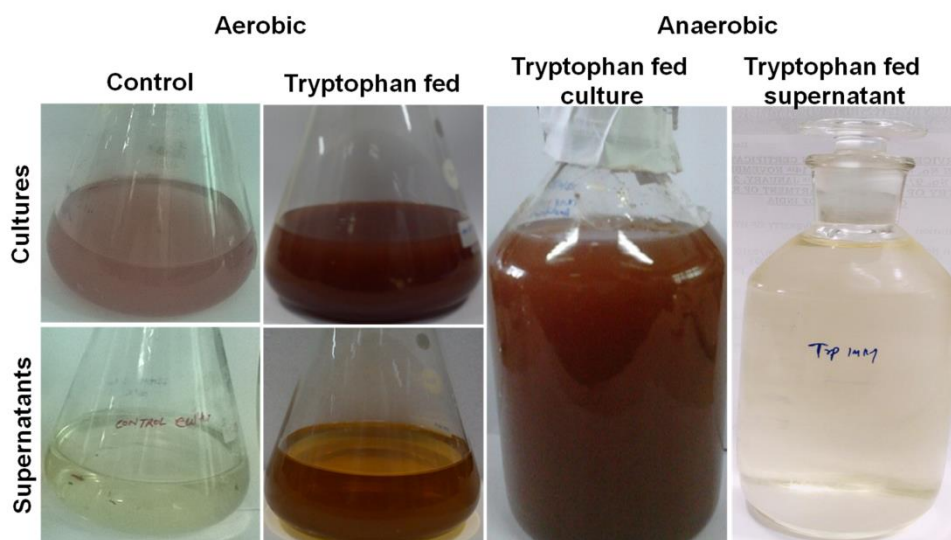
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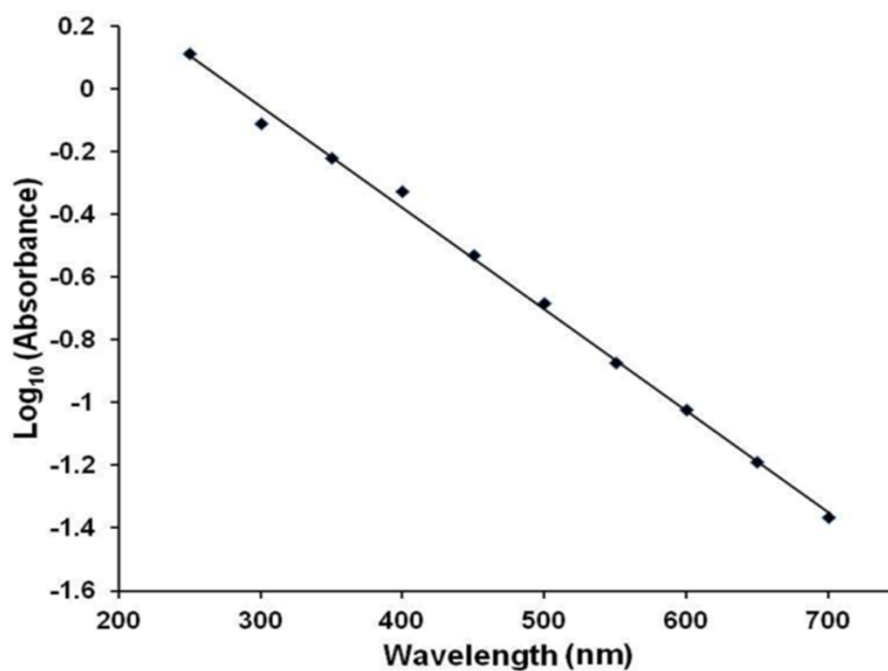
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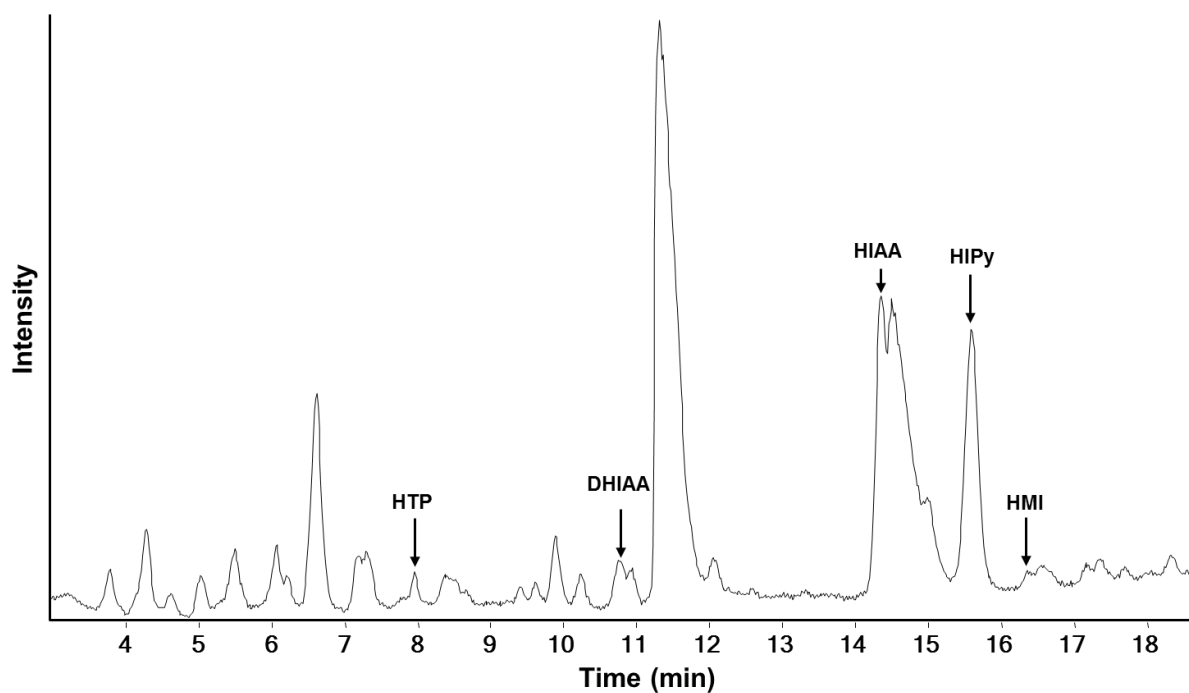
Supplementary information Figures



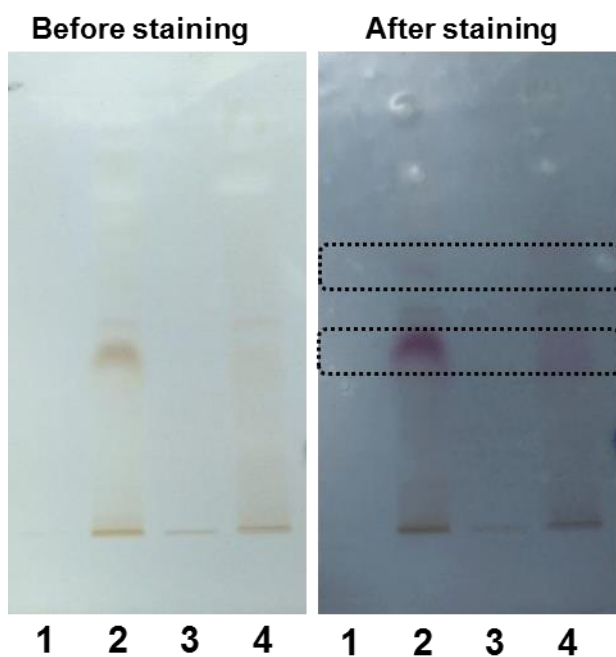
Supporting Information Figure. S1 :Brown pigment production by strain JA2. Control (without tryptophan) and tryptophan-amended cultures and respective supernatants of strain JA2 under anaerobic and aerobic growth conditions



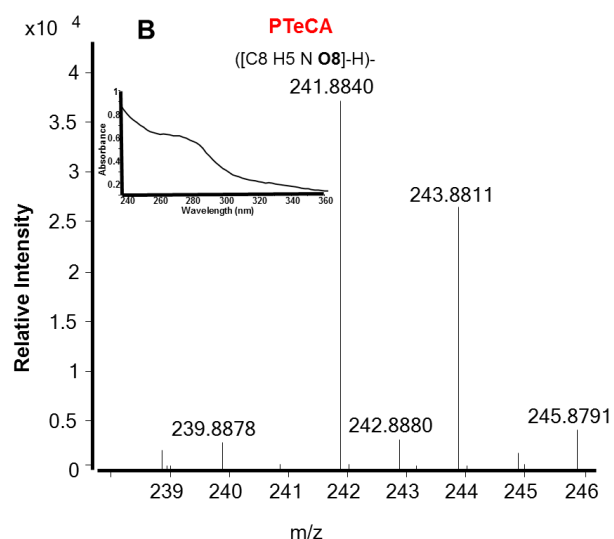
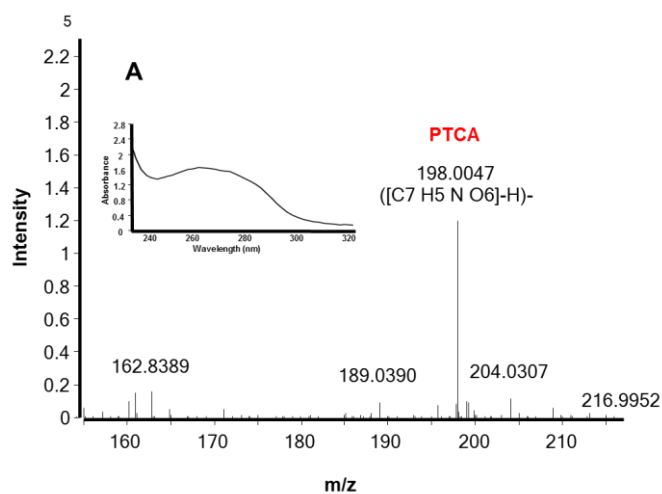
Supporting Information Figure. S2: Linear regression coefficient curve of absorbance vs wavelength of purified brown pigment produced by strain JA2



Supporting Information Figure. S3: Total Ion Chromatogram of methanolic extract of tryptophan-amended aerobic culture supernatant of strain JA2. HTP, 5-Hydroxytryptophan; DHIAA, 5,6-dihydroxyindole-3-acetic acid; HIAA, 5-hydroxyindole-3-acetic acid; HIPy, 5-hydroxyindole-3-pyruvic acid; HMI, 5-hydroxymethyl indole.



Supporting Information Figure. S4: TLC analysis of hydrolyzed brown pigment products of tryptophan and hydroxytryptophan. Lane-1 and 3: un-hydrolyzed Trp, OH-trp fraction, Lane 2 and 4 hydrolyzed products of Trp and OH-Trp. Rectangular boxes showing the indole positive staining (purple bands)



Supporting information Figure S5: Mass spectrum of PTCA (A) and PTeCA showing molecular ion mass of 198 and 241 [M-1] and their corresponding UV spectrum as insert. PTCA, pyrrole-2,3,5-tricarboxylic acid; PTeCA, pyrrole-2,3,4,5-tetracarboxylic acid.

Supporting Information table. S1: Physicochemical properties of purified brown pigment

	Chemical nature	Result
1.	Solubility Organic solvent (a) Ethanol (b) Chloroform (c) Acetone (d) Benzene (e) Ethyl acetate Alkaline solution 1M NaOH /KOH Buffers 50 mM phosphate buffer pH 7 50 mM Sodium bicarbonate buffer pH 10 50 mM dipotassium hydrogen phosphate buffer pH 12 Water	Insoluble Insoluble Insoluble Insoluble Insoluble Insoluble Soluble Insoluble Partially soluble Partially soluble Insoluble
(2)	Colour	Dark brown
(3)	Reaction with 3M HCl	Precipitated
(4)	Reaction with oxidizing agent such as H_2O_2 and NaOCl	Decolorized (black to colorless)
(5)	Reaction of polyphenol with $FeCl_3$	Formed flocculent brown precipitate
(6)	Reaction with $Na_2S_2O_4$ and potassium ferricyanide	Decolorized first turned brown with addition potassium ferricyanide
(7)	Reaction with ammonical silver nitrate	Formed a grey coloured silver precipitate (reduced) lining on the test tube