Research Square

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Peltogynoids contributing to discoloration in Peltogyne mexicana heartwood

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Abstract

Four new peltogynoid monomers (**3**, **7**, **10**, **11**), a new peltogynoid dimer (**9**), and two new flavanones (**5**, **6**) were isolated from the heartwood of *Peltogyne mexiacana*, along with two known peltogynoids (**1**, **4**) and two known aldehydes (**2**, **8**). The structures of the isolated compounds were determined using NMR and MALDI-TOF MS analysis. The discoloration of the methanol solutions of the isolated peltogynoids and flavanones was examined by exposing them to room light in the air. The methanol solutions of (+)-peltogynol (**1**) and (+)-mopanol (**4**) discolored to reddish and bluish purple, respectively. After discoloration, the *b** values of these compounds decreased significantly from 12.1 and 19.1 to -0.7 and − 1.8, respectively. These precursors of pigment compounds **1** and **4** have a catechol moiety in the B ring, and a hetero-six-membered ring (D ring) connecting the B and C ring of flavan-3,4-diol *via* an oxyethylene bridge, which is similar to the structure of leucoanthocyanidin. These results led to the hypothesis that the metabolized pigment compounds have anthocyanidin-like structures with peltogyne skeleton.

Introduction

Peltogyne mexicana is native to neotropical regions, including Guiana and Brazil, and grows a height of 30–40 m (Janet et al. 2023). The heartwood of *Peltogyne* trees is well known to metabolize characteristic flavonoid, peltogynoids (S. E. Drewes and D. G. Roux 1967). This group of flavonoids has a hetero-six-membered ring (D ring) that connects the B and C rings of flavan-3-ol *via* an oxyethylene bridge (Mathew et al. 2023). In the first isolation report on peltogynoids, (+)-peltogynol was isolated from *P. porphyrocardia Griseb*. ex *Benth*, and its absolute configuration was determined to be 2*R*, 3*S*, 4*R* (G. M. Robinson and R. Robinson 1935; Hassall and Weatherston 1965). Currently, various peltogynoids have been isolated from various species, including *Peltogyne*, *Colophospermum*, and *Cassine* (W. R. Chan et al. 1958; E. Drewes et al. 1991; Daneel et al. 2003). Peltogynoid shows variety of pharmacological activities, such as anti-inflammatory activity of myrtinol E isolated from *Backhousia myrtifolia* (Mathew et al. 2023), cytotoxicity against cancer cells of bougainvinone G isolated from *Bougainvillea spectabilis* (Do et al. 2016), and kinase inhibitory activity of acanilol B isolated from *Acacia nilotica* (Ahmadu et al. 2010).

P. mexicana heartwood, which contains peltogynoids, is well known for its purple discoloration, hence, it is commonly referred to “Purple heart”. The heartwood gradually turns bright purple after its surface is exposed to air and sunlight, while it shows pale yellow immediately after harvesting or injury (Paulina et al. 2019; Janet et al. 2023). Because of this unique purple color and the high durability as wood material (Marcos et al. 2021), P. *mexicana* heartwood has a high market value and is used as high-grade wood material in a wide range of industries. The phenomenon in which the color of heartwood changes after being cut down has been reported for various species used for high-end desks and furniture, including *Caesaplinia sappan* and *Millettia pendula.* Regarding the heartwood of *Caesaplinia sappan*, called sappan wood, brazilin is autoxidized to brazilein and appears red in color (Kim et al. 1997; De Oliveira et al. 2002). The heartwood of *Millettia pendula* turns dark purple after being harvested. This heartwood contains 7,3',4'-trihydroxy-6'-methoxyisoflav-3-ene, which forms quinone methide when oxidized, and

turns dark purple in color (Mitsunaga et al. 1987a, b). As shown in these examples, discoloration in heartwood often results from oxidative reactions (Laver and Arvey 1996; Chang et al. 2000). In *P. mexicana*, the production of red compounds was observed when peltogynoids were heated under acidic conditions (G. M. Robinson and R. Robinson 1935; Brandt and Roux 1979). In another report, peltomexicanin, in which the B ring exhibits a quinone methide form, was isolated as a heartwood pigment compound (Gutiérrez-Macías et al. 2016). However, no research has focused on the precursors to the pigment compounds involved in this discoloration. In this study, isolation of various peltogynoids and other flavonoids from *P. mexicana* heartwood was conducted to identify compounds responsible for heartwood discoloration as precursors.

Materials and methods

4.1 General Experimental procedures

All of color measurement in this study was conducted using Color analyser TES-3250 (TES Electrical Electronic Corp. Taiwan). High-performance liquid chromatography (HPLC) analysis was performed using MD-2018 photodiode array detector (JASCO, Japan) equipped with a Sunniest C18 reversed phase column (5 mm × 250 mm length, ChromaNik Technologies Inc.) at flow rate of 1.0 mL/min, detection wavelength of 190-700 nm, and oven temperature of 35ºC. Regarding the mobile phase, 0.05% TFA aq. (A) and methanol (B) were used as mobile phases. The eluant was applied as a linear gradient elution for 50 min from 20% to 80% of solvent B. Preparative HPLC was conducted using a set of LC-6AD system (SHIMAZU, Japan) equipped with a Inertsil ODS-3 column (5 μm, 20 × 250 mm, GL Sciences, Japan) at flow rate of 9.0 mL/min. Nuclear magnetic resonance (NMR) spectra were recorded on a JEOL JNM ECA-600 (JEOL, Japan), and Bruker Biospin AVANCE III 800 (bruker, USA). Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) spectra were measured using a Shimadzu AXIMA resonance spectrometer equipped with a nitrogen laser (λ=337 nm). Electronic circular Dichroism (ECD) spectra were recorded on a J-820P spectrometer (JASCO, Japan) using 1 mm path precision cells.

4.2 Computational details

For compounds with chiral centers, the absolute configuration was determined by a computational ECD calculation. All calculations were performed by the Gaussian16W program package(M.J. Frisch et al. 2016).

ECD calculation was performed by the TD-SCF method at the WB97XD/6–31+G(d, p) level in methanol as solvent. The final ECD spectra were obtained according to the Boltzmann-calculated contribution of each conformer by SpecDis(T. Bruhn et al. 2017). The calculated experimental data were compared with the ECD spectra.

4.3 Plant materials and extraction

Wood materials of *Peltogyne mexicana* heartwood was obtained from a timber dealer (Yamagataya Industry Co., Ltd., Japan). Heart wood (600 g) was crushed to powder states and extracted with 2 L of methanol, then dried methanol extract was partitioned successively twice with *n*-hexane, diethyl ether, ethyl acetate, and methanol using 2 L each. Each soluble part was evaporated and all of extract procedure was conducted under dark condition.

4.4CIE LAB parameter

The color of methanol extract, each soluble part, and isolated flavonoids were evaluated using CIE LAB parameters (*a**, *b**, *L**). Besides, the values of *h**, , , „ and were calculated from CIE LAB parameters (Supap et al. 2012; Jung and Sato 2013). Discolorations in this study were evaluated by a changing these parameters.

4.5Light exposure behavior of extract and soluble parts

A 5 mg of methanol extract or each soluble part was dissolved with 1.5 ml of methanol and removed into a small vial. The degree of discoloration was evaluated by exposure the vials to a room light (approximately 470 lx) in the air at room temperature for 24 hours. Refer to measuring the color, filter paper chip cut into a circle shape with a diameter of 1.5 cm, then impregnated in methanol extract and each soluble parts part before and after exposure, and the values of *a**, *b**, and *L** were measured using a color anolyser.

4.6 Compounds isolation

All separation procedure procedures were monitored by TLC and HPLC profiles. The part of diethyl ether soluble part (10.0 g) was fractionated on an open silica gel column chromatography (80 mmφ×600 mm) and eluted with ethyl acetate : *n*-hexane (4 : 6, v/v) to obtain four subfractions (EFr.1-4). Subfraction EFr.3 (2 g) was further fractionated on an open silica gel column chromatography (70 mmφ×300 mm) and eluted with methanol : chloroform (3 : 7, v/v) to obtain five subfractions (EFr.3-1 – 3-5). EFr.3-5 (147.3 mg) was subjected to open ODS-silica gel column chromatography (40 mmφ×150 mm) and eluted with 70 % methanol aq. to yield **1** (19.1 mg). **4** (15.6 mg), and **10** (4.0 mg) were purified by preparative HPLC (wavelength at 210 nm) with a gradient elution of methanol : 0.05 % TFA aq. [20 : 80 (v/v) to 80 : 20 (v/v) for 50 min] from EFr.3-4 (47.0 mg). Subfraction EFr.2 (197.1 mg) was further fractionated on an open silica gel column chromatography (60 mmφ×300 mm) and eluted with methanol : chloroform (2 : 8, v/v) to obtain four subfractions (EFr.2-1 – 2-4), then **2** (3.5 mg) was purified by preparative HPLC (wavelength at 210 nm) with a gradient elution of methanol : 0.05 % TFA aq. [20 : 80 (v/v) to 80 : 20 (v/v) for 50 min] from EFr.2-1 (22.5 mg). EFr.2-2 (129.4 mg) was subjected to open silica gel column chromatography (30 mmφ×320 mm) and eluted with methanol : chloroform (1 : 9, v/v) to obtain four subfractions (EFr.2-2-1 – 2-2-4). then **3** (12.5 mg), **5** (2.7 mg), and **6** (3.2 mg) were purified by preparative HPLC (wavelength at 210 nm) with a gradient elution of methanol : 0.05 % TFA aq. [20 : 80 (v/v) to 80 : 20 (v/v) for 50 min] from EFr.2-2-2 (53.2 mg). The part of ethyl acetate soluble part (1.0 g) was fractionated on an open silica gel column chromatography (80 mmφ×600 mm) and eluted with ethyl

acetate : *n*-hexane (2 : 8, v/v) to obtain three subfractions (AFr.1-3). Subfraction AFr.2 (392.9 mg) was further fractionated on an open silica gel column chromatography (35 mmφ×350 mm) and eluted with ethyl acetate : *n*-chloroform (8:2, v/v) to obtain 3 subfractions (AFr.2-1 – 2-3), then **7** (8.7 mg) was purified by preparative HPLC (wavelength at 210 nm) with a gradient elution of methanol : 0.05 % TFA aq. [20 : 80 (v/v) to 80 : 20 (v/v) for 50 min] from EFr.2-3 (23.0 mg). **8** (2.6 mg) was purified by preparative HPLC (wavelength at 210 nm) with a gradient elution of methanol : 0.05 % TFA aq. [20 : 80 (v/v) to 80 : 20 (v/v) for 50 min] from EFr.2-1 (8.1 mg). Subfraction AFr.3 (80.7 mg) was further fractionated on an open silica gel column chromatography (40 mmφ×300 mm) and eluted with methanol : *n*-chloroform (3 : 7, v/v) to obtain 8 subfractions (AFr.3-1 – 3-8), then **9** (4.4 mg) was purified by preparative HPLC (wavelength at 210 nm) with a gradient elution of methanol : 0.05 % TFA aq. [20 : 80 (v/v) to 80 : 20 (v/v) for 50 min] from EFr.3-8 (23.0 mg). The part of ethyl acetate soluble part (3.3 g) was fractionated on an open LH-20 gel column chromatography (50 mmφ×300 mm) and eluted with methanol : chloroform (5 : 5, v/v) to obtain four subfractions (LHFr.1-4). Subfraction LHFr.3 (150.6 mg) was further fractionated on an open ODS-silica gel column chromatography (25 mmφ×210 mm) and 60 % methanol aq. to obtain four subfractions (LHFr.3-1 – 3-4), then **11** (6.6 mg) was purified by preparative HPLC (wavelength at 210 nm) with a gradient elution of methanol : 0.05 % TFA aq. [20 : 80 (v/v) to 80 : 20 (v/v) for 50 min] from LHFr.3-3 (20.4 mg). Structures of isolated compounds were determined by the NMR analysis and the MALDI-TOF MS analysis. For steric compounds, ECD measurement was performed to determine their absolute configuration.

4.7Spectroscopic data

(+)-Peltogynol (**1**)

Pale yellow crystal; ECD (*c* 0.3 mM, methanol) λ_{max} ($\Delta\varepsilon$): 235.0 (+20.41), 289.0 (+4.50); ^{1}H-NMR (ppm, 600 MHz, methanol-d_4) 3.52 (3-H, 1H, *dd*), 4.74 (1”-H, 1H, *d*) 4.76 (4-H, 1H, *d*), 4.77 (2-H, 1H, *d*), 6.35 (8-H, 1H, *d*), 6.49 (6-H, 1H, *dd*), 6.56 (5’-H, 1H, *s*), 7.09 (2’-H, 1H, *s*), 7.35 (5-H, 1H, *d*); ^{13}C-NMR (ppm, 125 MHz, methanol-d_4) 68.3 (C-1”), 69.9 (C-4), 72.7 (C-2), 79.2 (C-3), 103.1 (C-8), 109.9 (C-6), 111.0 (C-5’), 113.4 (C-2’), 118.0 (C-4a), 124.9 (C-1’), 127.4 (C-6’), 130.1 (C-5), 145.0 (C-4’), 145.9 (C-3’), 155.9 (C-8a), 158.7 (C-7); MALDI-TOFMS *m/z* 325.0692 $[M+Na]^+$ (calculated for 325.0688, $C_{16}H_{14}NaO_6$)

2, 4-dihydroxybenzaldehyd (**2**)

White crystal; ^{1}H-NMR (ppm, 600 MHz, acetone-d_6) 6.26 (3-H, 1H, *d*), 6.43 (5-H, 1H, *dd*), 7.48 (6-H, 1H, *d*), 9.67 (7-H, 1H, *s*); ^{13}C-NMR (ppm, 125 MHz, acetone-d_6) 103.2 (C-3), 109.9 (C-5), 116.2 (C-1), 136.7 (C-6), 165.6 (C-2), 167.3 (C-4), 195.5 (C-7); MALDI-TOFMS *m/z* 161.0216 $[M+Na]^+$ (calculated for 161.0215, $C_7H_6NaO_3$)

4*R*-methoxymopanol (**3**)

Pale yellow crystal; ECD (*c* 0.3 mM, methanol) λ_{max} ($\Delta\varepsilon$): 212.0 (+20.96), 227.0 (+5.79); ^{1}H-NMR (ppm, 600 MHz, acetone-d_6) 3.60 (-OCH_3, 3H, *s*), 3.69 (3-H, 1H, *dd*), 4.55 (4-H, 1H, *d*), 4.79 (1”-H, 1H, *d*), 4.80 (2-H, 1H, *d*), 4.92 (1”-H, 1H, *d*), 6.34 (8-H, 1H, *d*), 6.49 (6-H, 1H, *dd*), 6.84 (5’-H, 1H, *d*), 6.99 (6’-H, 1H, *d*), 7.23 (5-H, 1H, *d*); ^{13}C-NMR (ppm, 125 MHz, acetone-d_6) 58.3 (-OCH_3), 65.2 (C-1”), 65.2 (C-1”), 72.6 (C-2), 77.5 (C-3), 78.8 (C-4), 103.3 (C-8), 110.0 (C-6), 114.6 (C-5’), 115.5 (C-4a), 117.8 (C-6’), 123.1 (C-2’), 125.6 (C-1’), 130.4 (C-5), 141.0 (C-3’), 144.3 (C-4’), 156.4 (C-8a), 158.9 (C-7); MALDI-TOFMS *m/z* 339.0842 $[M+Na]^+$ (calculated for 339.0845, $C_{17}H_{16}NaO_6$). All NMR data were shown in Table 1.

(+)-mopanol(**4**)

Yellow crystal; ECD (*c* 0.3 mM, methanol) λ_{max} ($\Delta\varepsilon$): 211.0 (+15.31), 228.0 (+4.51); ^{1}H-NMR (ppm, 600 MHz, acetone-d_6) 3.52 (3-H, 1H, *dd*), 4.74 (1”-H, 1H, *d*), 4.78 (4-H, 1H, *d*), 4.79 (2-H, 1H, *d*), 5.07 (1”-H, 1H, *d*), 6.36 (8-H, 1H, *d*), 6.49 (6-H, 1H, *dd*), 6.84 (5’-H, 1H, *d*), 6.99 (6’-H, 1H, *d*), 7.36 (5-H, 1H, *d*); ^{13}C-NMR (ppm, 125 MHz, acetone-d_6) 65.4 (C-1”), 65.4 (C-1”), 70.0 (C-4), 72.6 (C-2), 78.6 (C-3), 103.1 (C-8), 109.9 (C-6), 114.6 (C-5’), 117.8 (C-4a), 117.8 (C-6’), 123.2 (C-2’), 125.7 (C-1’), 130.0 (C-5), 141.0 (C-3’), 144.3 (C-4’), 155.8 (C-8a), 158.7 (C-7); MALDI-TOFMS *m/z* 325.0689 $[M+Na]^+$ (calculated for 325.0688, $C_{17}H_{16}NaO_6$)

3*R*-methoxy-(+)-butin (**5**)

Colorless crystal;ECD (*c* 0.3 mM, methanol) λ_{max} ($\Delta\varepsilon$): 324.4 (+3.15), 292.6 (-1.69), 268.8 (+1.19), 233.2 (+4.98), 217.2 (+6.92); ^{1}H-NMR (ppm, 600 MHz, methanol-d_4) 3.31 (-OCH_3, 3H, *s*), 3.73 (3-H, 1H, *d*), 5.31 (2-H, 1H, *d*), 6.37 (8-H, 1H, *d*), 6.52 (6-H, 1H, *dd*), 6.75 (5’-H, 1H, *d*), 6.82 (6’-H, 1H, *d*), 7.01 (2’-H, 1H, *d*), 7.72 (5-H, 1H, *d*); ^{13}C-NMR (ppm, 125 MHz, methanol-d_4) 59.1 (-OCH_3), 82.4 (C-2), 82.4 (C-3), 103.6 (C-8), 112.1 (C-6), 113.3 (C-4a), 115.8 (C-5’), 115.9 (C-2’) 120.0 (C-6’) 129.0 (C-1’) 130.3 (C-5), 146.1 (C-4’), 146.6 (C-3’), 165.0 (C-8a), 166.8 (C-7), 190.6 (C-4); MALDI-TOFMS *m/z* 325.0688 $[M+Na]^+$ (calculated for 325.0688, $C_{16}H_{14}NaO_6$). All NMR data were shown in Table 2.

3*S*-methoxy-2*S*-liquiritigenin (**6**)

Pale yellow crystal; ECD (*c* 0.3 mM, methanol) λ_{max} ($\Delta\varepsilon$): 331.4 (+1.31), 302.4 (-1.54), 236.0 (+2.13), 213.6 (+6.28); ^{1}H-NMR (ppm, 600 MHz, methanol-d_4) 3.29 (-OCH_3, 3H, s), 4.22 (3-H, 1H, d), 5.12 (2-H, 1H, d), 6.32 (8-H, 1H, d), 6.52 (6-H, 1H, dd), 6.82 (3'-H, 1H, dd), 6.82 (5'-H, 1H, dd), 7.33 (2'-H, 1H, dd), 7.33 (6'-H, 1H, dd), 7.71 (5-H, 1H, d); ^{13}C-NMR (ppm, 125 MHz, methanol-d_4) 60.6 (-OCH_3), 83.3 (C-3), 84.1 (C-2), 103.6 (C-8), 112.1 (C-6), 114.0 (C-4a), 116.2 (C-3'), 116.2 (C-5'), 129.2 (C-1'), 130.1 (C-5), 130.1 (C-2'), 130.1 (C-6'), 159.2 (C-4'), 164.8 (C-8a), 166.9 (C-7), 193.4 (C-4); MALDI-TOFMS *m/z* 309.0739 $[M+Na]^+$ (calculated for 307.0739, $C_{16}H_{14}NaO_5$). All NMR data were shown in Table 3.

2*S*, 3*S*-peltogynone (**7**)

Colorless crystal; ECD (*c* 0.3 mM, methanol) λ_{max} ($\Delta\varepsilon$): 324.6 (+2.7), 292.8 (-1.5), 270.6 (+1.1), 233.0 (+4.8), 216.8 (+6.7); ^{1}H-NMR (ppm, 600 MHz, methanol-d_4) 4.37 (3-H, 1H, *d*), 4.56 (2-H, 1H, *d*), 4.80 (1″-H, 2H, *d*), 6.46 (8-H, 1H, *d*), 6.50 (3′-H, 1H, *s*), 6.56 (6-H, 1H, *dd*), 7.06 (6′-H, 1H, *s*), 7.75 (5-H, 1H, *d*); ^{13}C-NMR (ppm, 125 MHz, methanol-d_4) 69.4 (C-1″), 76.4 (C-2), 77.8 (C-3), 103.9 (C-8), 111.3 (C-3′), 112.3 (C-6), 113.1 (C-6′), 114.3 (C-4a), 124.1 (C-1′), 126.8 (C-2′), 130.2 (C-5), 146.0 (C-4′), 147.1 (C-5′), 164.5 (C-8a), 166.8 (C-7), 191.3 (C-4); MALDI-TOFMS *m/z* 323.0527 $[M+Na]^+$ (calculated for 323.0531, $C_{16}H_{14}NaO_6$). All NMR data were shown in Table 4.

syringaldehyde (**8**)

White crystal; ^{1}H-NMR (ppm, 600 MHz, methanol-d_4) 3.98 (-OCH_3, 6H, *s*), 7.15 (2, 6-H, 2H, *s*), 9.82 (7-H, 1H, *s*); ^{13}C-NMR (ppm, 125 MHz, methanol-d_4)56.5 (-OCH_3), 106.7 (C-2), 106.7 (C-6), 147.3 (C-1), 164.1 (C-3), 164.1 (C-4), 164.1 (C-5), 190.7 (C-7); MALDI-TOFMS *m/z* 205.0471 $[M+Na]^+$ (calculated for 205.0476, $C_9H_{10}NaO_4$)

(+)-mopanol B-(4β→6‴)-(+)-molanol (**9**)

Colorless crystal; ECD (*c* 0.3 mM, methanol) λ_{max} ($\Delta\varepsilon$): 217.0 (+30.9), 206.4 (-42.3); ^{1}H-NMR (ppm, 600 MHz, acetone-d_6) 3.55 (3‴-H, 1H, *dd*), 4.18 (3-H, 1H, *dd*), 4.72 (4‴-H, 1H, *d*), 4.80 (1⁗-H, 1H, *d*), 4.84 (4-H, 1H, *d*), 4.87 (2‴-H, 1H, *d*), 4.92 (1″-H, 1H, *d*), 4.96 (1″-H, 1H, *d*), 5.00 (2-H, 1H, *d*), 5.02(1⁗-H, 1H, *d*), 6.52 (8‴-H, 1H, *s*), 6.53 (6-H, 1H, *dd*), 6.56 (8-H, 1H, *d*), 6.86 (5‴-H, 1H, *s*), 6.88 (5-H, 1H, *d*), 6.95 (5⁗-H, 1H, *d*), 6.94 (5′-H, 1H, *d*), 7.10 (6⁗-H, 1H, *d*), 7.17 (6′-H, 1H, *d*); ^{13}C-NMR (ppm, 125 MHz, acetone-d_6) 39.6 (C-4), 65.4 (C-1⁗), 65.7 (C-1″), 67.6 (C-2), 69.8 (C-4‴), 72.6 (C-2‴), 76.4 (C-3), 78.6 (C-3‴), 103.5 (C-8), 104.3 (C-8‴), 110.3 (C-6), 114.8 (C-5⁗), 114.9 (C-5′), 115.0 (C-6‴), 117.6 (C-6⁗), 117.8 (C-6′), 118.2 (C-4a‴), 122.7 (C-2′), 123.3 (C-2⁗), 123.8 (C-4a), 125.8 (C-1′), 125.8 (C-1⁗), 132.1 (C-5‴), 132.9 (C-5), 141.0 (C-4′), 141.0 (C-4⁗), 144.3 (C-3′), 144.3 (C-3⁗), 154.5 (C-8a‴), 156.5 (C-8a), 157.3 (C-7⁗), 158.2 (C-7); MALDI-TOFMS *m/z* 609.1369 $[M+Na]^+$ (calculated for 609.1373, $C_{32}H_{26}NaO_{11}$). All NMR data were shown in Table 5.

3-hydroxypeltogynone (**10**)

Colorless crystal; ^{1}H-NMR (ppm, 600 MHz, acetone-d_6) 4.50 (2-H, 1H, *s*), 4.87 (1″-H, 1H, *d*), 4.87 (1″-H, 1H, *d*), 6.36 (8-H, 1H, *d*), 6.56 (6-H, 1H, *dd*), 6.57 (3′-H, 1H, *s*), 6.82 (6′-H, 1H, *s*), 7.48 (5-H, 1H, *d*); ^{13}C-NMR (ppm, 125 MHz, acetone-d_6) 66.0 (C-1″), 68.3 (C-2), 99.4 (C-8), 105.7 (C-3), 111.4 (C-3′), 113.2 (C-4a), 113.2 (C-6), 115.7 (C-6′), 125.7 (C-2′), 125.9 (C-1′), 127.4 (C-5), 145.9 (C-4′), 146.8 (C-5′), 169.6 (C-8a), 174.5 (C-7), 195.3 (C-4); MALDI-TOFMS *m/z* 339.0478 $[M+Na]^+$ (calculated for 339.0481, $C_{16}H_{12}NaO_7$). All NMR data were shown in Table 6.

4-methoxypeltogynol (**11**)

Colorless crystal; ^{1}H-NMR (ppm, 600 MHz, acetone-d_6) 3.57 (-OCH_3, 1H, *s*), 3.70 (3-H, 1H, *d*), 4.54 (4-H, 1H, *dd*), 4.78 (2-H, 1H, *d*), 4.79 (1”-H, 1H, *d*), 4.92 (1”-H, 1H, *d*), 6.36 (8-H, 1H, *d*), 6.49 (6-H, 1H, *dd*), 6.57 (3’-H, 1H, *s*), 7.08 (6’-H, 1H, *s*), 7.22 (5-H, 1H, *dd*); ^{13}C-NMR (ppm, 125 MHz, acetone-d_6); MALDI-TOFMS*m/z* 339.0840 $[M+Na]^+$ (*m/z* 339.0845, Calculated for $C_{17}H_{16}NaO_6$). All NMR data were shown in Table 7.

4.8 Effect of light exposure to isolated compounds

Except for two aldehydes (**2**, **8**), 2 mg of each isolated compound that are peltogynoid s was dissolved in 0.5 ml of methanol and transferred into an NMR tube. Discoloration was evaluated by exposure to room light in the air for 24h. To measure the color, a filter paper chip cut into a circle with a diameter of 1.5 cm was impregnated with each methanol solvent of the isolated compounds, and the values of *a**, *b**, and *L** before and after exposure were measured using a color meter.

Results and discussion

5.1 Light exposure behavior of extract and soluble parts

For the CIE LAB parameters, positive values of *a** indicate redness, while negative values indicate greenness, and positive values of *b** indicate yellowness, while negative values indicate blueness. If the coordinates of (*a*, b**) are (0, 0), it is achromatic (Supap et al. 2012; Jung and Sato 2013). The results of the CIE parameter measurements for the methanol extract and each soluble part are shown in Fig. 1a and Table S1. At 0h of light exposure, the *L** values of all soluble parts, except for the *n*-hexane soluble part, were + 89 to + 95, and the *a** and *b** values were positive, indicating that all of them ranged from orange to brown. After 48 h of light exposure, the *a** values of the methanol extract, ether soluble part, and ethyl acetate soluble parts changed to positive values with $\Delta a^*_{0h\rightarrow 24h}$ of 6.5, 2.6, and 4.3, and the *b** values changed to negative values with $\Delta b^*_{0h\rightarrow 24h}$ of -27.9, -14.5, and − 21.3, respectively. Meanwhile, each parameter of the *n*-hexane and methanol soluble parts remained almost unchanged ($\Delta a^*_{0h\rightarrow 24h}$ = 0.2 and 1.5, $\Delta b^*_{0h\rightarrow 24h}$ = 0.0 and 0.9). Additionally, their hue values were significantly reduced ($\Delta h^*_{0h\rightarrow 24h}$ = -141.7, -100.0, -127.8) and ΔE values were increased ($\Delta E_{0h\rightarrow 24h}$ = 28.6, 15.3, 21.9). These results indicate that the methanol extract, ether soluble part, and ethyl acetate soluble part were discolored purple upon exposure to room light in the air (Fig. 1a). These clearly show that the precursors of the pigment compounds were mainly concentrated in the diethyl ether and ethyl acetate soluble parts.

5.2 Structure determination

1, **2**, **4**, and **8** were identified as (+)-peltogynol (**1**) (S. E. Drewes and D. G. Roux 1966), 2,4-dihydroxybenzaldehyde (**2**) (Eun-Mi et al. 2007), (+)-mopanol (**4**) (S. E. Drewes and D. G. Roux 1966), and syringaldehyde (**8**) (A. Ram Kumar et al. 2021), according to previous literature data. Among them, **2** and

8 were isolated from this species for the first time. All compounds isolated in this study are shown in Fig. 2.

5.2.1 Structure determination of monomers

Compound **3** was isolated as pale-yellow crystals. The MALDI-TOF MS analysis in the positive mode generated an $[M + Na]^+$ ion at *m/z* 339.0842 (calculated for $C_{17}H_{16}NaO_6$, *m/z* 339.0845) indicating the molecular formula $C_{17}H_{16}O_6$ (Fig.S3-1).

The structure was elucidated by analyzing the NMR spectra (Fig.S3-2 to S3-6). The ^{1}H-NMR spectrum showed the presence of five phenyl group protons [δ_H = 7.23 (1H, *d*), 6.99 (1H, *d*), 6.84 (1H, *d*), 6.49 (1H, *dd*), and 6.34 ppm (1H, *d*)], four alkane protons [4.80 (1H, *d*), 4.92 (1H, *d*), 4.79 (1H, *d*), 4.55 (1H, *d*), and 3.69 ppm (1H, *dd*)], and one methoxy proton [3.60 ppm (3H, *s*)]. ^{13}C-NMR spectrum showed seventeen resonances, including twelve phenyl carbons [δ_C = 158.9, 156.4, 144.3, 141.0, 130.4, 125.6, 123.1, 117.8, 115.5, 114.6, 110.0, and 103.3 ppm], four alkane carbons [78.8, 77.5, 72.6, and 65.2 ppm], and one methoxy carbon [58.3 ppm]. For protonated carbon, the correspondences between the protons and carbon were determined based on the HMQC spectrum. COSY spectrum showed a coupled proton system attributed to H-5 (doublet, 7.23 ppm, *J* = 7.8 Hz) and H-6 (double doublet, 6.49 ppm, *J* = 3.0, 8.4 Hz), and H-5′ (doublet, 6.84 ppm, *J* = 8.4 Hz) and H-6′ (doublet, 6.99 ppm, *J* = 8.4 Hz). From the coupling constants and the results of COSY analysis, the five phenyl group protons were separated into a trisubstituted benzene ring consisting of H-5 (doublet, 7.23 ppm, *J* = 7.8 Hz), H-6 (double doublet, 6.49 ppm, *J* = 3.0, 8.4 Hz), and H-8 (doublet, 6.34 ppm, *J* = 3.0 Hz), and a tetrasubstituted benzene ring consisting of H-5′ (doublet, 6.84 ppm, *J* = 8.4 Hz) and H-6′ (doublet, 6.99 ppm, *J* = 8.4 Hz) (A ring and B ring). A continuous spin system of C-2 to C-4 was observed in the COSY correlation and a series of HMBC correlations of H-2/C-1′, C-6′, H-3/C-1″, H-4/C-4a, C-8a, and H-1″/C-1′ were assigned to construct the mopanol skeleton. The position of the methoxy group was determined based on the HMBC correlation of H-4/OCH_3. Thus, the planar structure 3 was determined as 4-methoxymopanol.

The relative configuration of **3** was determined based on the coupling constant of H-2 (doublet, 4.80 ppm, *J* = 11.4 Hz), H-3 (double doublet 3.69 ppm, *J* = 7.8, 9.6 Hz), and H4 (doublet, 4.55 ppm, *J* = 8.4 Hz) as 2,3-*trans*-3,4-*trans*. The absolute configuration of **3** was determined from its ECD spectrum (Fig. S3-7). The experimental ECD spectrum of **3** showed positive Cotton effects at approximately 212 and 227 nm. This result is in good agreement with the calculated ECD spectrum of (2*R*, 3*S*, 4*R*)-**3**. Thus, the absolute configuration of **3** was determined to be 4*R*-methoxymopanol.

Compound **11** was isolated as colorless crystals. The MALDI-TOF MS analysis in positive mode generated an $[M + Na]^+$ ion at *m/z* 339.0840 (calculated for $C_{17}H_{14}NaO_6$, *m/z* 339.0845), indicating the molecular formula $C_{17}H_{14}O_6$ (Fig.S9-1).

The structure was elucidated based on the analysis of the NMR spectra (Fig.S9-2 to S9-6). **11** was determined to be the structural isomer of **3**, differing in the positions of the two hydroxy groups in the B

ring. The ^{1}H-NMR spectrum showed two singlets of H-3’ (singlet, 6.57 ppm) and H-5’ (singlet, 7.08 ppm), while H-3’ and H-5’ of **3** were detected as doublets. A series of HMBC correlation of H-1”/C-3 and H-5’/C-2, and a downfield shift of H-4’ (δ_C = 145.0 ppm) and H-5’ (146.0 ppm) showed that **11** retained a peltogynol skeleton. Thus, the planar structure 8 was determined to be 4-methoxypeltogynol.

The relative configuration of **11** was determined based on coupling constants of H-2 (doublet, 4.78 ppm, *J* = 9.6 Hz), H-3 (double doublet 3.70 ppm, *J* = 8.4, 9.6 Hz), and H-4 (doublet, 4.54 ppm, *J* = 8.4 Hz) as 2,3-*trans*-3,4-*trans*. ECD measurement of **11** was conducted to determine its absolute configuration, however, the Cotton effect was not observed. Therefore, only the planar structure was determined in this study.

Compound **10** was isolated as colorless crystals. The MALDI-TOF MS analysis in the positive mode generated an $[M + Na]^+$ ion at *m/z* 339.0478 (calculated for $C_{16}H_{12}NaO_{7,}$ *m/z* 339.0481), indicating the molecular formula $C_{16}H_{12}O_7$ (Fig.S8-1).

The structure was elucidated based on the analysis of the NMR spectra (Fig.S8-2 to S8-6). Comparison of the spectroscopic data of **10** with **11** showed that **10** retained its mopanol skeleton. Their differences lie in the downfield shift of C-3 (δ_C = 105.7 ppm) and the change to the carbonyl region of C-4 (195.3 ppm) (See Fig.S8-1 to 8 – 5). Thus, the planar structure 10 was determined to be 3-hydroxypeltogynone. The Cotton effect was not observed in the ECD measurement of **10**. Therefore, only the planar structure was determined in this study.

Compound **7** was isolated as colorless crystals. The MALDI-TOF MS analysis in the positive mode generated an $[M + Na]^+$ ion at *m/z* 323.0527 (calculated for $C_{16}H_{12}NaO_{6,}$ *m/z* 323.0531) indicating the molecular formula $C_{16}H_{12}O_6$ (Fig.S6-1).

The structure was elucidated based on the analysis of the NMR spectra (Fig.S6-2 to S6-6). THE NMR spectrum of **7** was similar to that of **10**, differing in the presence of H-3 (doublet, 4.37 ppm, *J* = 11.4 Hz). This indicates that C-3 is a protonated carbon, whereas the C-3 of 10 is hydroxylated. Thus, the planar structure of **7** was determined to be peltogynone.

The relative configuration of **7** was determined based on the coupling constants of H-2 (doublet, 4.56 ppm, *J* = 11.4 Hz) and H-3 (doublet, 4.37 ppm, *J* = 11.4 Hz) as 2,3-*trans*. The absolute configuration of **7** was determined from its ECD spectrum (Fig. S6-7). The experimental ECD spectrum of **7** showed a positive Cotton effect at approximately 325, 271, 233, and 217 nm, and a negative Cotton effect at approximately 293 nm. This result is in good agreement with the calculated ECD spectrum of (2*S*, 3*S*)-**7**. Thus, the absolute configuration of **7** was determined to be 2*S*, 3*S*-peltogynone.

Compound **6** was isolated as pale yellow crystals. The MALDI-TOF MS analysis in the positive mode generated an $[M + Na]^+$ ion at *m/z* 309.0739 (calculated for $C_{16}H_{14}NaO_{5,}$ *m/z* 309.0739), indicating the molecular formula $C_{16}H_{14}O_5$ (Fig.S5-1).

The structure was elucidated based on the analysis of the NMR spectra (Fig.S5-2 to S5-6). The ^{1}H-NMR spectrum of methanol-d_4 showed the presence of seven phenyl group protons [δ_H = 7.33 (1H, *dd*), 7.33 (1H, *dd*), 7.71 (1H, *d*), 6.82 (1H, *dd*), 6.82 (1H, *dd*), 6.52 (1H, *dd*), and 6.32 ppm (1H, *d*),], two alkane protons [5.12 (1H, *d*) and 4.22 ppm (1H, *d*)], and one methoxy proton [3.29 ppm (3H, *s*)]. ^{13}C-NMR spectrum revealed the presence of sixteen carbon signals, including twelve phenyl carbons [δ_C = 166.9, 164.8, 159.2, 130.1, 130.1, 130.1, 129.2, 116.2, 116.2, 114.0, 112.1, and 103.6 ppm], two alkane carbons [84.1 and 83.3 ppm], one carbonyl carbon [193.4 ppm], and one methoxy carbon [60.6 ppm]. A similar structure of A ring and carbonyl carbon at C-4 as **7** was confirmed by the AMX system of protons H-5 (doublet, 7.71 ppm, *J* = 9.6), H-6 (doublet, 6.52 ppm, *J* = 1.8, 9.6), H-8 (doublet, 6.32 ppm, *J* = 2.4), and the COSY correlation of H-5/C-6.

The two double doublets with an integrated value of 2, H-2’ and H-6’ (double doublet, 7.33 ppm, *J* = 1.8, 7.2 Hz), and H-3’ and H-5’ (double doublet, 6.82 ppm, *J* = 1.8, 6.6 Hz) indicated that the B ring was a disubstituted benzene. In addition, the phenol moiety was confirmed by the downfield shift of C-4’ (159.2 ppm) and a series of HMBC correlation of H-2/C-6’, H-2’/C-2, and H-3’/C-4’. The position of the methoxy group was determined based on the HMBC correlation of H-3/OCH_3. Thus, the planar structure of **6** was determined to be 3-methoxyliquiritigenin.

The relative configuration of **6** was determined based on the coupling constants of H-2 (doublet, 5.12 ppm, *J* = 11.4 Hz) and H-3 (doublet, 4.22 ppm, *J* = 10.2 Hz) as 2,3-*trans*. The absolute configuration of **6** was determined from its ECD spectrum (Fig. S5-7). The experimental ECD spectrum of **6** showed a positive Cotton effect at approximately 331, 236, and 214 nm and a negative Cotton effect at approximately 302 nm. This result is in good agreement with the calculated ECD spectra of (2*S*, 3*R*)-**6**. Thus, the absolute configuration of **6** was determined to be 3*S*-methoxy-2*S*-liquiritigenin.

Compound **5** was isolated as colorless crystals. The MALDI-TOF MS analysis in the positive mode generated an $[M + Na]^+$ ion at *m/z* 325.0688 (calculated for $C_{16}H_{14}NaO_6$, *m/z* 325.0688), indicating the molecular formula $C_{16}H_{14}O_6$ (Fig.S4-1).

The structure was elucidated based on the analysis of the NMR spectra (Fig.S4-2 to S4-6). The comparison of the spectroscopic data of **5** and **6** differed in terms of the coupling system in the B ring, H-2’ (doublet, 7.01 ppm, *J* = 2.4 Hz), H-5’ (doublet, 6.75 ppm, *J* = 7.8 Hz), and H-6’ (double doublet, 6.82 ppm, *J* = 1.8, 7.8 Hz). The catechol moiety was confirmed as the B ring by two downfield shifted carbon C-3’ (146.6 ppm) and C-4’ (146.1 ppm) and a series of HMBC correlation of H-2/C-2’, C-6’, H-5’/C-3’, and H-6’/C-4’. Thus, the planar structure of **5** was determined to be 3*R*-methoxybutin.

The relative configuration of **5** was determined based on the coupling constants of H-2 (doublet, 5.31 ppm, *J* = 1.8 Hz) and H-3 (doublet, 3.73 ppm, *J* = 1.8 Hz) as 2,3-*cis*. The absolute configuration of **5** was determined from its ECD spectrum (Fig. S4-7). The experimental ECD spectrum of **5** showed positive Cotton effects at approximately 324, 269, 233, and 217 nm, and a negative Cotton effect at

approximately 293 nm. This result is in good agreement with the calculated ECD spectrum of (2*S*, 3*R*)-**5**. Thus, the absolute configuration of **6** was determined to be 3*R*-methoxy-(+)-butin.

5.2.2 Structure determination of dimer

Compound **9** was isolated as colorless crystals. The MALDI-TOF MS analysis in the positive mode generated an $[M + Na]^+$ ion at *m/z* 609.1369 (calculated for $C_{32}H_{26}NaO_{11,}$ *m/z* 609.1373), indicating the molecular formula $C_{32}H_{26}NaO_{11}$ (Fig.S7-1).

The structure was elucidated based on the analysis of the NMR spectra (Fig.S7-2 to S7-6). The ^{1}H-NMR spectrum showed the presence of nine phenyl group protons [δ_H = 7.17 (1H, *d*), 7.10 (1H, *d*), 6.95 (1H, *d*), 6.94 (1H, *d*), 6.88 (1H, *d*), 6.86 (1H, *s*), 6.56 (1H, *d*), 6.53 (1H, *dd*), and 6.52 ppm (1H, *s*)] and ten alkane protons [5.02 (1H, *d*), 5.00 (1H, *d*), 4.96 (1H, *d*), 4.92 (1H, *d*), 4.87 (1H, *d*), 4.84 (1H, *d*), 4.80 (1H, *d*), 4.71 (1H, br *d*), 4.18 (1H, *dd*), and 3.55 ppm (1H, *dd*)]. The ^{13}C-NMR spectrum showed thirty two resonances, including twenty four phenyl carbons [δ_C = 158.2, 157.3, 156.5, 154.5, 144.3, 144.3, 141.0, 141.0, 132.9, 132.1, 125.9, 125.9, 123.8, 123.3, 122.7, 118.2, 117.8, 117.6, 115.0, 114.9, 114.8, 110.3, 104.3, and 103.5 ppm] and eight alkane carbons [78.6, 76.4, 72.6, 69.8, 67.6, 65.7, 65.4, and 39.6 ppm]. For protonated carbon, the correspondences between the protons and the carbons were determined based on the HMQC spectrum. The COSY spectrum exhibited a coupled proton systems attributed to H-5 (doublet, 6.88 ppm, *J* = 8.4 Hz) and H-6 (double doublet, 6.53 ppm, *J* = 2.5, 8.2 Hz), H-5’ (doublet, 6.83 ppm, *J* = 8.4 Hz) and H-6’ (doublet, 6.99 ppm, *J* = 8.4 Hz), and H-5”” (doublet, 6.95 ppm, *J* = 8.4 Hz) and H-6”” (doublet, 7.10 ppm, *J* = 8.8 Hz), and two of three coupling protons of H-2 (doublet, 5.00 ppm, *J* = 9.9 Hz), H-3 (double doublet, 4.18 ppm, *J* = 5.9, 9.8 Hz), H-4 (doublet, 4.84 ppm, *J* = 5.9 Hz), and H-2”’ (doublet, 4.87 ppm, *J* = 10.0 Hz), H-3”’ (double doublet, 3.55 ppm, *J* = 8.6, 9.9 Hz), and H-4”’ (doublet, 4.72 ppm, *J* = 8.4 Hz). These data indicate that **9** is a flavonoid dimer.

One of the mopanol units was confirmed by a series of HMBC of H-3””/C-1””, H-5”’/C-4”’, H-6””/C-2”’, and H-1””/C-1”” with a difference of absence of H-6”’. Another mopanol unit was determined by the HMBC correlations of H-3/C-1’, H-4/C-4a, H-1”’/C-3, C-2’, and C-3’ without a hydroxy group at C-4. The binding mode of these two molecules of mopanol was determined by the HMBC correlations of H-4/C-5”’, C-6”’, and C-7”’ to be C-4 (39.6 ppm) to C-6”’ (110.3 ppm). Thus, the planar structure of **9** was determined to be mopanol-(4, 6)-mopanol.

The relative configuration of each mopanol units in **9** was determined based on the coupling constants. The relative configuration of one of the mopanol units (composed of C-1 to C-1”) was determined to be 2,3-*trans*-3,4-*cis* based on the coupling constants of H-2 (doublet, 5.00 ppm, *J* = 9.9 Hz), H-3 (double doublet, 4.18 ppm, *J* = 5.9, 9.8 Hz), and H-4 (doublet, 4.84 ppm, *J* = 5.9 Hz). Another mopanol moiety (composed of C-1”’ to C-1””) was determined to be 2,3-*trans*-3,4-*trans* based on the coupling constants of H-2”’ (doublet, 4.87 ppm, *J* = 10.0 Hz), H-3”’ (double doublet, 3.55 ppm, *J* = 8.6, 9.9 Hz), and H-4”’ (doublet, 4.72 ppm, *J* = 8.4 Hz). The absolute configuration of **9** was determined from its ECD spectrum (Fig. S7-7). The experimental ECD spectrum of **9** shows a positive Cotton effect at approximately 217 nm

and a negative cotton effect at approximately 206 nm. This result is in good agreement with the calculated ECD spectrum of (2*R*, 3*S*, 4*S*, 2‴*R*, 3‴*S*, 4‴*R*)-**9**. Thus, the absolute configuration of **9** was determined to be (+)-mopanol B-(4β→6‴)-(+)-mopanol. A dimer composed of peltogynoids was isolated and structurally determined for the first time in this study.

5.3 Effect of light exposure to isolated compounds

Before being exposed to room light in the air, **5**, **7**, **9**, **10**, and **11** were colorless, and **1**, **3**, **4**, and **6** ranged from pale yellow to yellow. After these methanol solutions were exposed to room light and air for 24h, only (+)-peltogynol (**1**) and (+)-mopanol (**4**) showed purple discoloration (Fig. 1b). The color changes of all flavonoids were confirmed using CIE LAB parameters (Table S1). Immediately after isolation, all compounds showed positive *a** and *b** values, hile only the *b** values of (+)-peltogynol (**1**) and (+)-mopanol (4) changed to negative values (-0.7 and − 1.8, respectively) after light exposure. The h^* values ($\Delta h^*_{0h\rightarrow 24h}$) of (+)-peltogynol (**1**) and (+)-mopanol (**4**) changed to -54.9 and − 75.8, respectively, while those of other compounds remained between − 0.7 and 5.6. These results suggested that (+)-peltogynol (**1**) and (+)-mopanol (**4**) are precursors to pigment compounds among isolated compounds in this study. These compounds have a hetero-six-membered ring (D ring) and a hydroxyl group at C-4 in *R*-configuration. In addition, C-2, C-3, and C-4 were chiral carbons with 2*R*, 3*S*, 4*R* configuration. These structural characteristics are similar to the structure of leucoanthocyanidins. Leucoanthocyanidins are easily oxidized by organic acids or air and undergo structural changes to anthocyanidins *via* the elimination of the 4-hydroxyl group (Clark-Lewis and Williams 1967). Scince (+)-peltogynol (**1**) and (+)-mopanol (**4**) both have a hydroxyl group at the 4-hydroxyl group, it is possible that the similar structural change could occur to generate anthocyanidins. The newly isolated compound **9** was thought to be a proanthocyanidin generated by an intermediate that formed a bond at C-4 with (+)-mopanol at C-6, providing evidence for this mechanism.

Conclusion

In this study, five new peltogynoids and two new flavanones were isolated with four known compounds from the heartwood of *P. mexicana*. This study aimed to identify compounds related to heartwood discoloration as precursors to pigment compounds. Among the isolated compounds, (+)-peltogynol (**1**) and (+)-mopanol (**4**) contributed to the discoloration to reddish and blueish purple, respectively, upon exposure to room light in the air. In terms of their CIE LAB parameters, the *b** values of these two compounds decreased and their *a** values increased significantly. These results showed that discoloration was accelerated by exposure to room light in the air for both the methanol extract and the isolated compounds. These two compounds possess a sub-structure similar to that of flavan-3,4-*trans*-diol. These structural features provide insights into the mechanism of discoloration in *P. Mexicana*, and the heartwood contains anthocyanin-like pigment compounds. However, the isolation of dimer compound **9** is believed to contribute greatly to the elucidation of the structure of the pigment compound. In other words, it is speculated that peltogynol and mopanol are converted to anthocyanidins

through quinone methides by air oxidation in the presence of light, and that the unstable structures react to form dimers, oligomers, and polymers to form pigment structures.

Declarations

Author Contribution

TY conducted experiment and wrote the manuscript. KY and TM supervised the study and reviewed the manuscript.

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References

1. A. Ram Kumar, S. Selvaraj, K.S. Jayaprakash, et al (2021) Multi-spectroscopic (FT-IR, FT-Raman, 1H NMR and 13C NMR) investigations on syringaldehyde. J Mol Struct 1229:129490. https://doi.org/10.1016/J.MOLSTRUC.2020.129490
2. Ahmadu A, Abdulkarim A, Grougnet R, et al (2010) Two New Peltogynoids from Acacia nilotica Delile with Kinase Inhibitory Activity. Planta Med 76:458–460. https://doi.org/10.1055/S-0029-1186226
3. Brandt E V., Roux DG (1979) Metabolites from the purple heartwood of Mimosoideae I, Acacia peuce F. Muell: the first natural 2,3-cis-peltogynoids. J Chem Soc Perkin 1 777–780. https://doi.org/10.1039/P19790000777
4. Chang ST, Wang SY, Cheng SS (2000) Environmental effects on the color of sugi (Cryptomeria japonica D. Don) heartwood. Journal of Wood Science 46:390–394. https://doi.org/10.1007/BF00776402/METRICS
5. Clark-Lewis JW, Williams LR (1967) Flavan derivatives. XVII. Epimerization of the benzylic 4-hydroxyl group in flavan-3,4-diols and the formation of 4-alkyl ethers by solvolysis. Aust J Chem 20:2151–2167. https://doi.org/10.1071/CH9672151
6. Daneel F, Jannie P.J. M, Desmond S (2003) Phytochemistry of the mopane, Colophospermum mopane. Phytochemistry 64:31–51. https://doi.org/10.1016/S0031-9422(03)00152-3
7. de Oliveira LFC, Edwards HGM, Velozo ES, Nesbitt M (2002) Vibrational spectroscopic study of brazilin and brazilein, the main constituents of brazilwood from Brazil. Vib Spectrosc 28:243–249. https://doi.org/10.1016/S0924-2031(01)00138-2
8. Do LTM, Aree T, Siripong P, et al (2016) Bougainvinones A-H, Peltogynoids from the Stem Bark of Purple Bougainvillea spectabilis and Their Cytotoxic Activity. J Nat Prod 79:939–945.

https://doi.org/10.1021/ACS.JNATPROD.5B00996/SUPPL_FILE/NP5B00996_SI_003.PDF

9. E. Drewes S, Mashimbye MJ, Field JS, Ramesar N (1991) 11,11-dimethyl-1,3,8,10-tetrahydroxy-9-methoxypeltogynan and three pentacyclic triterpenes from Cassine transvaalensis. Phytochemistry 30:3490–3493. https://doi.org/10.1016/0031-9422(91)83243-E
10. Eun-Mi A, Sang-Jae P, Won-Cheol C, et al (2007) Antioxidant Activity of Isolated Compounds from the Heartwoods of Rhus verniciflua. J Korean SocAppl biol Chem 50:358–361
11. G. M. Robinson, R. Robinson (1935) Leuco-anthocyanins and leuco-anthocyanidins. Part I. The isolation of peltogynol and its molecular structure. J Chem Soc 744–752. https://doi.org/10.1039/JR9350000744
12. Gutiérrez-Macías P, Peralta-Cruz J, Borja-de-la-Rosa A, et al (2016) Peltomexicanin, a Peltogynoid Quinone Methide from Peltogyne Mexicana Martínez Purple Heartwood. Molecules 2016, Vol 21, Page 186 21:186. https://doi.org/10.3390/MOLECULES21020186
13. Hassall CH, Weatherston J (1965) 512. The absolute configuration of the leucoanthocyanidin, peltogynol. Journal of the Chemical Society (Resumed) 2844–2849. https://doi.org/10.1039/JR9650002844
14. Janet V-A, Marlene B-M, Roberto CA-N, et al (2023) Demographic, spatial, and ecological aspects of Peltogyne mexicana - a threatened species – Under different extraction conditions in southern México. Trees, Forests and People 14:100427. https://doi.org/10.1016/J.TFP.2023.100427
15. Jung H, Sato T (2013) Comparison between the Color Properties of Whiteness Index and Yellowness Index on the CIELAB. Textile Coloration and Finishing 25:241–246. https://doi.org/10.5764/TCF.2013.25.4.241
16. Kim DS, Baek NI, Oh SR, et al (1997) NMR assignment of brazilein. Phytochemistry 46:177–178. https://doi.org/10.1016/S0031-9422(96)00874-6
17. Laver ML, Arvey SW (1996) Chemical brown staining of Douglas-fir wood: Light and oxygen susceptibility of extractives. For Prod J 46:96–101
18. Marcos C de MP, Luc APS, Thierry D, Carlito Calil J (2021) Doweled cross laminated timber: Experimental and analytical study. Constr Build Mater 273:121820. https://doi.org/10.1016/J.CONBUILDMAT.2020.121820
19. Mathew S, Zhang K, Zhou X, et al (2023) Myrtinols A–F: New Anti-Inflammatory Peltogynoid Flavonoid Derivatives from the Leaves of Australian Indigenous Plant Backhousia myrtifolia. Molecules 28:2160. https://doi.org/10.3390/MOLECULES28052160/S1
20. Mitsunaga T, Kondo R, Imamura H (1987a) The chemistry of the color of wood. IV. The phenolic consistuent contributes to the coloration of murasakitagayasan (Millettia sp.) heartwood. Mokuzai Gakkaishi Journal of the Japan Wood Research Society 33:239–245
21. Mitsunaga T, Kondo R, Imamura H (1987b) The chemistry of the color of wood. III. The isoflavonoids of the heartwood of murasakitagayasan (Millettia sp.). Mokuzai Gakkaishi Journal of the Japan Wood Research Society 33(3): 234-238 33:234–238
22. M.J. Frisch, G.W. Trucks, H.B. Schlegel, et al (2016) Gaussian 16W, Revision C.01

23. Paulina G-M, Cinthya GG-Z, Leticia G-S, Cynthia O-P (2019) Purple pigment from Peltogyne mexicana heartwood as a potential colorant for food. J Food Sci Technol 56:3225–3238
24. S. E. Drewes, D. G. Roux (1967) Isolation of mopanin from Colophospermum mopane and interrelation of flavonoid components of Peltogyne spp. Journal of the Chemical Society C: Organic 1407–1410. https://doi.org/10.1039/J39670001407
25. S. E. Drewes, D. G. Roux (1966) Stereochemistry and biogenesis of mopanols and peltogynols and associated flavanoids from Colophospermum mopane. Journal of the Chemical Society C: Organic 1644–1653. https://doi.org/10.1039/J39660001644
26. Supap N, Anuchita M, Sirirat D (2012) Application of Functional Colorant Prepared from Black Rice Bran in Yogurt. APCBEE Procedia 2:62–67. https://doi.org/10.1016/J.APCBEE.2012.06.012
27. T. Bruhn, A. Schaumlöffel, Y. Hemberger, G. Pescitelli (2017) SpecDis Version 1.71
28. W. R. Chan, W. G. C. Forsyth, C. H. Hassall (1958) The constitution of the leucoanthocyanidin, peltogynol. Journal of the Chemical Society (Resumed) 3174–3179. https://doi.org/10.1039/JR9580003174

Tables

Table 1 ^{1}H and ^{13}C chemical shifts and 2D NMR correlation of **3** in acetone-d_6 (δ in ppm, J in Hz)

Position	δ_H (ppm) (J in Hz)	δ_C (ppm)	COSY	HMBC
2	4.80 *d* 1H J = 11.4	72.6	3	1’, 2’
3	3.69 *dd* 1H J = 7.8, 9.6	77.5	2, 4	1”, 2, 4
4	4.55 *d* 1H J = 8.4	78.8	3	-OCH_3, 3, 4a, 8a
4a		115.5		
5	7.23 *d* 1H J = 7.8	130.4	6	4, 7, 8a
6	6.49 *dd* 1H J = 3.0, 8.4	110.0	5	4a, 7, 8
7		158.9		
8	6.34 *d* 1H J = 3.0	103.3		4a, 6, 7, 8a
8a		156.4		
1'		125.6		
2'		123.1		
3'		141.0		
4'		144.3		
5'	6.84 d 1H J=8.4	114.6	6’	1’, 3’, 4’
6'	6.99 d 1H J=8.4	117.8	5’	2, 5’, 2’, 4’
1”	4.79 *d* 1H J = 4.8	65.2	1”	3, 1’, 2’
	4.92 *d* 1H J = 4.8		1”	3, 1’, 2’, 3’
-OCH_3	3.60 *s* 3H	58.3		4

Table 2 ^{1}H and ^{13}C chemical shifts and 2D NMR correlation of **5** in methanol-d_4 (δ in ppm, J in Hz)

Position	δ_H (ppm) (J in Hz)	δ_C (ppm)	COSY	HMBC
2	5.31 *d* 1H J = 1.8	82.4	3	1', 2', 6'
3	3.73 *d* 1H J = 1.8	82.4	2	-OCH3
4		190.6		
4a		113.3		
5	7.72 *d* 1H J = 9.0	130.3	6	4, 7
6	6.52 *dd* 1H J = 2.4, 9.0	112.1	5	4a
7		166.8		
8	6.37 *d* 1H J = 2.4	103.6		8a
8a		165.0		
1'		129.0		
2'	7.01 *d* 1H J = 2.4	115.9		2, 3', 6'
3'		146.6		
4'		146.1		
5'	6.75 *d* 1H J = 7.8	115.8	6'	1', 4'
6'	6.82 *dd* 1H J = 1.8, 7.8	120.0	5'	2, 4', 5'
-OCH_3	3.31 *s* 3H	59.1		3

Table 3 ^{1}H and ^{13}C chemical shifts and 2D NMR correlation of **6** in methanol-d_4 (δ in ppm, J in Hz)

Position	δ_H (ppm) (*J* in Hz)	δ_C (ppm)	COSY	HMBC
2	5.12 *d* 1H *J* = 11.4	84.1	3	3, 6'
3	4.22 *d* 1H *J* = 10.2	83.3	2	2, -OCH_3
4		193.4		
4a		114.0		
5	7.71 *d* 1H *J* = 9.6	130.1	6	4, 7, 8a
6	6.52 *dd* 1H *J* = 1.8, 9.0	112.1	5	4a, 8
7		166.9		
8	6.32 *d* 1H *J* = 2.4	103.6		6, 8a
8a		164.8		
1'		129.2		
2'	7.33 *dd* 1H *J* = 1.8, 7.2	130.1	3'	2, 4'
3'	6.82 *dd* 1H *J* = 1.8, 6.6	116.2	2'	1', 4', 5'
4'		159.2		
5'	6.82 *dd* 1H *J* = 1.8, 6.6	116.2	6'	3', 4'
6'	7.33 *dd* 1H *J* = 1.8, 7.2	130.1	5'	2, 4'
-OCH_3	3.29 *s* 3H	60.6		3

Table 4 ^{1}H and ^{13}C chemical shifts and 2D NMR correlation of **7** in methanol-d_4 (δ in ppm, J in Hz)

Position	δ_H (ppm) (J in Hz)	δ_C (ppm)	COSY	HMBC
2	4.56 *d* 1H J = 11.4	76.4	3	3, 4, 1'
3	4.37 *d* 1H J = 11.4	77.8	2	4, 1', 1”
4		191.3		
4a		114.3		
5	7.75 *d* 1H J = 9.0	130.2	6	4, 7, 8a
6	6.56 *dd* 1H J = 2.4, 9.0	112.3	5	4a, 8
7		166.8		
8	6.46 *d* 1H J = 2.4	103.9		6, 7, 8a
8a		164.5		
1'		124.1		
2'		126.8		
3'	6.50 *s* 1H	111.3		1', 4', 1”
4'		146.0		
5'		147.1		
6'	7.06 *s* 1H	113.1		2, 2', 5'
1”	4.80 *d* 2H J = 14.4	69.4		3, 1', 2', 3'

Table 5 is available in the Supplementary Files section.

Table 6 ^{1}H and ^{13}C chemical shifts and 2D NMR correlation of **10** in acetone-d_6 (δ in ppm, J in Hz)

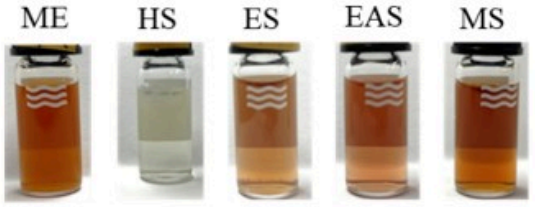
Position	δ_H (ppm) (J in Hz)	δ_C (ppm)	COSY	HMBC
2	4.50 *s* 1H	68.3		3, 1', 6'
3		105.7		
4		195.3		
4a		113.2		
5	7.48 *d* 1H J = 7.0	127.4	6	4, 7, 8a
6	6.56 *dd* 1H J = 2.0, 7.0	113.2	5	4a, 8
7		174.5		
8	6.36 *d* 1H J = 2.0	99.4		7, 4a, 8a
8a		169.6		
1'		125.9		
2'		125.7		
3'	6.57 *s* 1H	111.4		2', 4', 1”
4'		145.9		
5'		146.8		
6'	6.82 *s* 1H	115.7		1', 2', 5'
1”	4.87 *d* 1H J = 14.0	66.0		3, 2'
	4.87 *d* 1H J = 14.0			

Table 7 ^{1}H and ^{13}C chemical shifts and 2D NMR correlation of **11** in acetone-d_6 (δ in ppm, J in Hz)

Position	δ_H (ppm) (J in Hz)	δ_C (ppm)	COSY	HMBC
2	4.78 *d* 1H *J* = 9.6	72.7	3	3, 1’, 2’, 3’
3	3.70 *dd* 1H *J* = 8.4, 9.6	78.1	2, 4	1”, 2, 4
4	4.54 *dd* 1H *J* = 8.4	78.8	3	-OCH_3, 3, 4a, 8a
4a		115.5		
5	7.22 *dd* 1H *J* = 8.4	130.4	6	4, 7, 8a
6	6.49 *dd* 1H *J* = 2.4, 8.4	110.0	5	4a
7		158.9		
8	6.36 *d* 1H *J* = 2.4	103.3		6, 8a
8a		156.3		
1'		127.2		
2'		124.6		
3'	6.57 *s* 1H	111.0		2’, 5’
4'		145.0		
5'		146.0		
6'	7.08 *s* 1H	113.5		2, 1’, 4’
1"	4.79 *d* 1H *J* = 10.2	68.1	1”	2’
	4.92 *d* 1H *J* = 10.2			
-OCH_3	3.57 *s* 1H	58.3		4

Figures

a



	ME	HS	ES	EAS	MS
Light exposure 0h					
L^*	91.5	95.2	89.1	94.6	90.6
a^*	1.8	1.6	4.9	2.9	8.4
b^*	14.1	0.6	6.3	12.3	18.4
Light exposure 24h					
L^*	90.7	95.3	93.2	91.7	88.8
a^*	8.3	1.8	7.4	7.3	9.9
b^*	-13.8	0.6	-8.2	-9.0	19.4

b



	1		4	
	0h	24h	0h	24h
L^*	90.4	83.1	85	85.4
a^*	9.3	16.5	8.0	12.1
b^*	12.1	-0.7	19.1	-1.8

Figure 1

Discoloration of extract, soluble parts, and isolated compounds by light and air exposure (a) Discoloration of methanol extract and each soluble part (ME: methanol extract, HS: Hexane soluble part, ES: ether soluble part, EAS: ethyl acetate soluble part, MS: methanol soluble part) (b) Discoloration of (+)-peltogynol (**1**) and (+)-mopanol (**4**)

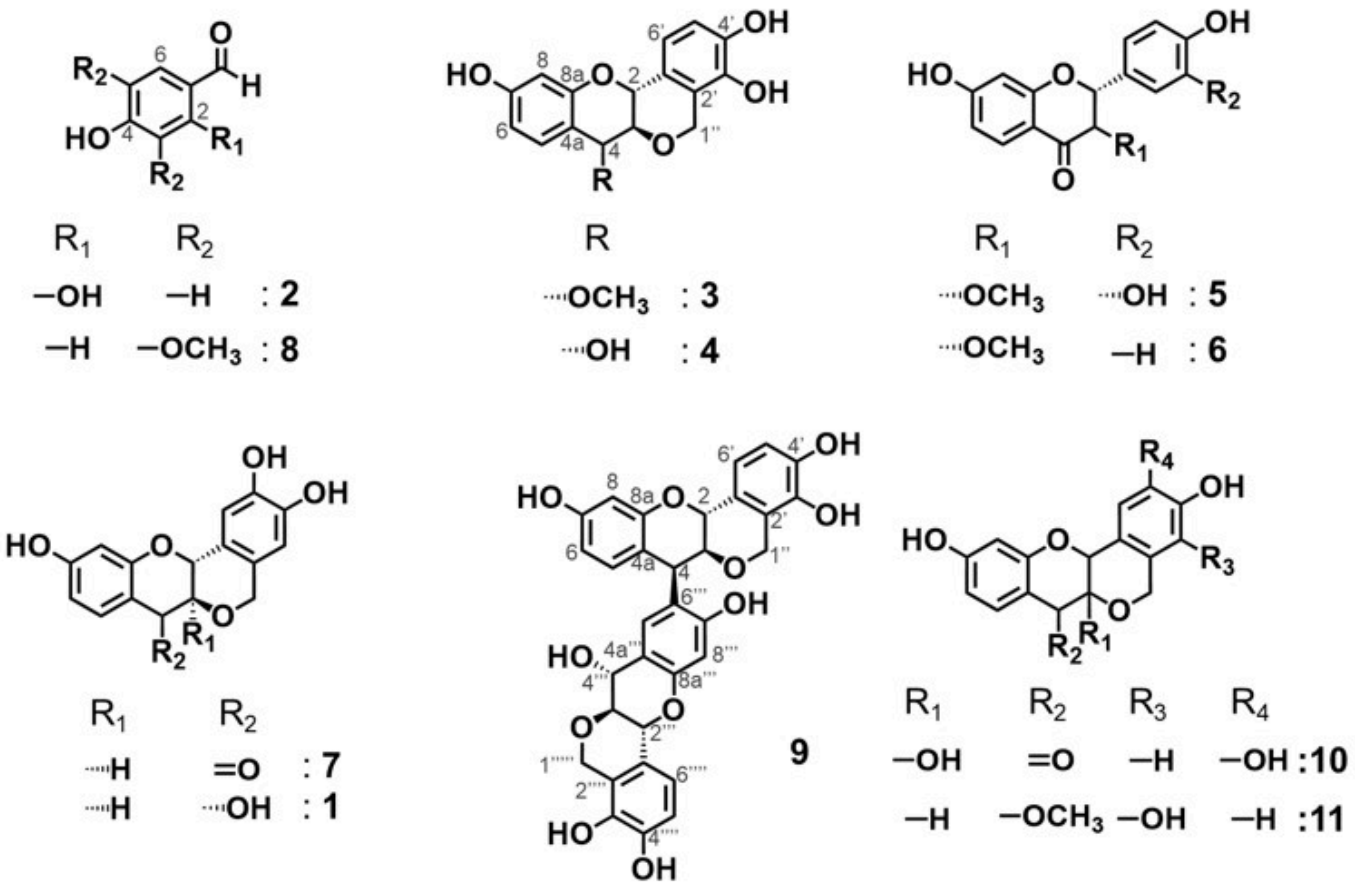


Figure 2

isolated compounds from *P. mexicana* heartwood

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Supportinginformation1.docx
- Table5.docx