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日本木材学会中国・四国支部
第 22 回 研 究 発 表 会

研究発表要旨集
(高知，2010年９月)

Abstract of the 22nd Meeting of the Chugoku-Shikoku Branch
of the Japan Wood Research Society
(Kochi, September 2010)

会　期
2010年9月13日(月)－14日(火)
Date: September 13-14, 2010

会　場
公立学校共済組合　高知会館
Venue: Kochi Hall, Kochi, Japan

主　催
日本木材学会中国・四国支部
Organized by the Chugoku-Shikoku Branch, the Japan Wood Research Society

共　催
森林バイオマス利用学会
日本木材加工技術協会中国支部
Co-organized by the Forest Biomass Utilization Society, Japan
and the Chugoku Branch, Wood Technological Association of Japan

後　援
高知県
Supported by Kochi Prefecture

研究・技術発表プログラム

【口頭発表　A会場：平安　2010年9月13日　9：30～12：10】

9：30-10：30　座長：伊藤和貴

A01　9:30-9:45　綿布を基材とする高吸水性セルロースグラフト共重合体
（島大総理工）○安田美菜子，吉延匡弘，和久芳春

A02　9:45-10:00　Quality, chemical compositions and antifungal activity of *Melaleuca leucadendron* Linn. leaf oils from Java, Indonesia
(Kochi University)　○ Rini Pujiarti, Yoshito Ohtani and Hideaki Ichiura

A03　10:00-10:15　Berberine, a main secondary metabolite isolated from Tali Kuning (*Tinospora dissitiflora* Diels.), an indigenous medical plants from Manokwari, Papua, Indonesia
(Kochi University) ○Wahyudi, Yoshito Ohtani and Hideaki Ichiura

A04　10:15-10:30　ハンゲショウにおける9, 9’ −デオキシ型リグナンとネオリグナンの探索
（香川大学農学部）　○傳田健二、牧野啓輔、片山健至、鈴木利貞

10：30-10：40　休憩

10：40-11：25　座長：市浦英明

A05　10:40-10:55　ハンゲショウにおける9,9'-デオキシ-8-O-4'-ネオリグナンの光学活性と生合成
（香川大学農学部）○牧野啓輔、片山健至、鈴木利貞

A06　10:55-11:10　熱帯産早生樹*Acacia mangium*樹皮からの抽出物のDPPHラジカル消去活性およびフェルロイルオキシ脂肪酸の単離
（香川大農）○鈴木大元、鈴木利貞、片山健至、（ボゴール農業大）Syamsul Falah

A07　11:10-11:25　コルクガシ外樹皮の塩基性メタノリシス：β-O-4’エノールエーテル型フェルラ酸メチル二量体の単離
（香川大農）○伊藤彰伸、國方美晴、鈴木利貞、片山健至

11：25-12：10　座長：吉延匡弘

A08　11:25-11:40　DFRC法によるトチュウ材リグニンの構造解析：γ-メチルグアイアシルクマロン型二量体の単離
（香川大学農学部）　立永梨恵、疇地修子、片山健至、鈴木利貞

A09　11:40-11:55　イネ突然変異体lsi1におけるリグニン分析
（香川大学農学部）○山畑 梓、鈴木利貞、片山健至、（岡山大資源植物研）武田 真

A10　11:55-12:10　Screening of asphaltene-degrading fungi from nature
(愛媛大学農学部）○ Dede Heri Yuli Yanto, Kazutaka Itoh, Sanro Tachibana

【口頭発表　B 会場：飛鳥　2010年9月13日　9：30〜12：10】

9：30-10：00　座長：川上敬介

B01　9:30-9:45　島根県産木材の腐朽と残存強度との関係 I 一腐朽に伴う部分圧縮強さの変化一
(島根県中山間地研セ）〇後藤崇志、冨川康之、中山茂生、（島大総合理工・文化財調査コンサルタント(株)）古野　毅

B02　9:45-10:00　木材の薬剤注入性とコーンカロリーメータ試験機を用いた発熱性試験
（広総研林技セ）〇花ヶ崎　裕洋、川元　満夫

10：00-10：30　座長：加藤定信

B03　10:00-10:15　高温セット正角材の内部割れと曲げ強度の関係
（広総研林技セ）〇吉村太一、藤田和彦、川元満夫、花ヶ崎裕洋

B04　10:15-10:30　徳島県産スギ梁材の高温低湿処理・減圧乾燥の可能性について
（徳島農技セ）　〇金磯牧夫、（九大農）　藤本登留、（（株）大島造船所）　永富裕和

10：30-10：40　休憩

10：40-11：25　座長：杉森正敏

B05　10:40-10:55　縄文時代中期から後期にかけての三瓶小豆原埋没林の古植生
（島根大学・文化財調査コンサルタント(株)）〇 古野　毅
（文化財調査コンサルタント(株)）　渡辺　正巳

B06　10:55-11:10　最近の一般住宅室内における温湿度環境
（岡山県農総セ森林研）〇野上英孝、小川　裕、金田利之

B07　11:10-11:25　天井裏に炭を敷設したRC造集合住宅におけるアメニティ測定
（島大院総理）〇安野　恭代、（島大総理）中井　毅尚

11：25-12：10　座長：鈴木利貞

B08　11:25-11:40　Isolation of potential anti-diabetic compounds from *Eugenia polyantha*
(愛媛大学農学部）〇 Raden Arthur Ario Lelono, Kazutaka Itoh, Sanro Tachibana

B09　11:40-11:55　界面重合法を活用した剥離紙不要粘着紙の開発
（高知大院・農）〇坂本　世悟、市浦　英明、大谷　慶人

B10　11:55-12:10　イオン液体の活用による製紙スラッジ完全再資源化技術の開発
（高知大院・農）〇中谷　拓宏、市浦　英明、大谷　慶人

A03

Berberine, a main secondary metabolite isolated from Tali kuning (*Tinospora dissitiflora* Diels), an indigenous medicinal plant from Manokwari, Papua, Indonesia

(Kochi University) ○ Wahyudi, Yoshito Ohtani and Hideaki Ichiura

Introduction

Tali kuning (*Tinospora dissitiflora* Diels) is a lying, climbing liana, with yellow stem, belonging to family Menispermaceae. The indigenous people of Papua are using this plant extracts for herbal medicine used for either preventing or combating malaria symptoms. This practice is still the main option, when the malaria drugs and modern health treatments are difficult to be achieved due to geographical and transportation reasons. Malaria symptoms are very complicated, ranging from higher in fever but feeling cold, the tongue feels very bitter taste, and could not swallow food into the stomach due to feeling vomiting. With respect to the malaria herbage, the bioactivity of Tali kuning (Tinospora dissitiflora Diels) mainly from Manokwari, best to our knowledge has not been investigated yet. This paper describes the isolation, and structural elucidation of main secondary metabolites isolated from Tali kuning (*Tinospora dissitiflora* Diels).

Material and methods

Sample preparation and collection

Tali kuning (*Tinospora dissitiflora* Diels) was collected from Assay Village, North Manokwari, January 2009. Mature stem of 12 cm diameter was harvested, dried at room temperature for a week, and powdered with hammer mill and passed the 40-mesh sieve. The wood meal is then stored at sealed plastic until used.

Chemicals and Tools

All chemicals (methanol, benzene, ethyl acetate, and formic acid) are analytical grade (Wako pure Chemical Industries, Ltd, Osaka, Japan), Column Chromatography (CC) (24/40, Vidrex P), Silica gel 60 (70-230 mesh) (Naclai tesque, Inc, Tokyo, Japan), Thin Layer Chromatography Plates (TLC) 5 x 10 cm (Whatman International Ltd, Maidstone,England), Preparative Thin layer Chromatography plate (PLC) 20 x 20 cm Merck (Merck KGaA, Damstadt, Germany), Microcap 5μL (Drummond, Scientific, Bromoll USA).

Extraction and partition

One kg of Tali kuning wood meal (moisture content 15%) was extracted with methanol for 48 hours, filtered by a glass funnel filter (25G-3), earning of 5000 ml of methanolic extract. This extract is evaporated and concentrated into 140 ml, and dried to give 9.91g. This dried extract was then portioned with hexane, chloroform, acetone and methanol.

Silica Gel Column Chromatography (CC) and Preparative Thin Layer Chromatography (PLC)

Silica gel column chromatography (CC) using solvent system benzene: ethyl acetate: formic acid (5:4:1), preparative thin layer chromatography (PLC) using system solvent benzen:ethyl acetate :formic acid (5:4:2) were used to isolate and purify the chloroform fraction. Targeted compounds are having yellow band on the TLC plate and detected under UV light. A further CC with solvent system benzene: methanol (2:3) was employed. Retention time (Rf) was used for marking targeted compound. Eleven compounds were isolated from Tali kuning (TK), namely TK1(15.9mg), TK2 (8mg), TK3 (23 mg), TK4 (360mg), TK5 (3mg), TK6 (2.9mg), and TK 7 (60mg), TK8 (439mg), TK9(190mg), TK10(3 mg) and TK11 (44mg). These isolated compounds were subjected to the mass spectrometry (MS), ultra violet (UV) spectrometry and NMR analysis.

Result and discussion

Ten isolated TKs (except TK5) were analyzed with NMR, and eight (8) TK spectra (^{1}H-NMR, ^{13}C-NMR) were confirmed, ranging from TK1, TK2, TK4, TK6, TK7, TK8, TK9, and TK11. The spectra (UV, ^{1}-H-NMR, ^{13}-C-NMR, MS) of 8 TK compounds were identified by comparing with those spectra data available on the published literatures.

The UV spectra of 8 isolated compounds reveal that all compounds have a similar pattern, having λ max at 222, 226, 234, 236, 264, and 347 nm. These UV spectra agree with those reported by Min *et al.*,(2006), Grycova *et al.*,(2007), and Shamma *et al.*, (1968) suggesting that these signals are of berberine. Unexpectedly, all 7 TKs (except TK6) recorded from LC-EI MS/MS spectra have the molecular ion peak at 336 m/z, and high resolution EI-MS at 335.95 m/z confirmed the molecular formula of berberine ($C_{20}H_{17}NO_4$). Furthermore, NMR spectra of 7 TKs (except TK6) strongly indicate the presence of two methoxyl groups (-OCH_3) at C-9, C-10, and a dioxymethylene (O-CH_2-O-) connected C2 to C3. These spectra also confirm that 7 TKs are having Quartenary Protoberberine Alkaloid (QPA) Skeleton, as presented by Figure 1.

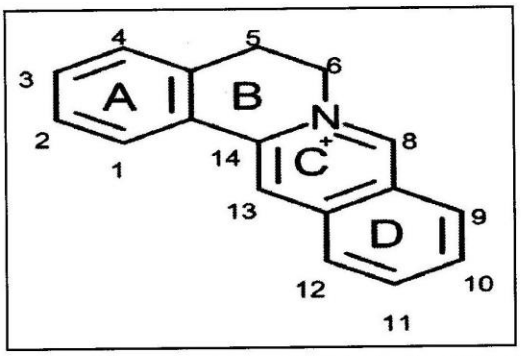

Figure1. Skeleton of Quaternary Protoberberine Alkaloid.

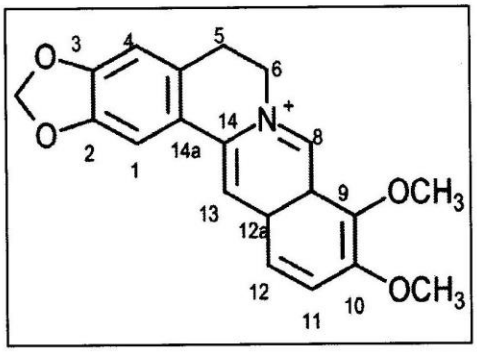

Figure 2. Elucidated structure of TK 7 (Berberine)

Figure 1 shows that the skeleton of quaternary protoberberine alkaloid is 5,6-dihydro quinolizine ($C_{17}H_{14}N^{+}$), having four rings, A, B, C, and D respectively. Usually substituents are present at positions 2,3,9,10 or 2,3,10, and 11, Grycova et al., (2007). The typical substituents are hydroxyl groups, methoxyl groups, and dioxymethylene groups. These workers also reported that the QPA cations are relatively planar species. Their planarity is distributed only in the partially saturated ring B, which is in a twisted half-chair conformation mainly due to atom C-5 and C-6, whereas the rings A, C and D are relatively plane (flat).

The 8 isolated TKs have predominantly yellow color, except for TK 7, which is brown. Grycova et al., (2007) reported that QPA are characterized by bright color, ranging from yellow to orange. These color appearances depend on many variables, such as method of crystallization, purification, both, or solvent pH.

TK7 was obtained as brown crystal, and the spectra (UV,^{1}-HNMR,^{13}-C-NMR,MS) correspond to the protoberberine spectra reported from the literature by Min et al.,(2006), and Grycova et al.,(2007). MS spectra reveal the ion peak at (M+H) m/z: 336 (100), 337 (80), having a molecular formula of $C_{20}H_{17}NO_4$, corresponding to berberine. NMR spectra reveal that these spectra are according to the berberine spectra on the literature, and can be detailed as follows: ^{1}H-NMR and 1^{3}-C-NMR spectra are as follows: ^{1}H-NMR : (400 MHz, CD_3OH) : ∂ 3.26 (2H, t, j=6.4 Hz, H-5), 4.11 (3H, s,-OCH3), 4.20 (3H-OCH3), 4.92 (2H, t, J=6.4 H-6), 6.10 (2H, s, O-CH2-O), 6.96 (1H, s, H-4), 7.65 (1H, s, H-1), 7.99 (1H, d, J=8.8 Hz, H-12), 8.11 (1H,d, J=8.8 Hz, H-11), 8.68 (1H, s, H-13), 9.76 (1H, s, H-8). ^{13}C-NMR (100.4 MHz, CD_3OH) : ∂ 28.19 (C -5), 57.18 (C-6), 57.65 (C-10-O-CH_3), 62.53 (C-9-O-CH_3), 103.68 (C-O-CH_2-O), 106.53 (C-1), 109.38 (C-4), 121.49 (C-14a), 121.86 (C-13), 123.33 (C-8a), 124.48 (C-12), 128.11 (C-11), 131.89 (C-4a), 135.18 (C-12a), 139.70 (C-13), 145.78 (C-8), 146.39 (C-9), 149.94 (C-2) 152.02 (C-10), 152.19 (C-3). TK 7 is the berberine having an elucidated structure as presented by Figure 2.

Conclusion

The main secondary metabolite of Tali Kuning (*Tinospora dissitiflora* Dields) is protoberberine (having protoberberine skeleton) and it is reported the first time isolated from this plant.
The work on the other secondary metabolites and the biological performances of these plant metabolites are in progress.

References

Grycova.L, Dostal.J,and Marek.R(2007). Quaternary Protoberberine Alkaloid "Review". Phytochemistry (68) pp. 150-1175;

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Shamma.M, Hillman.M.J,and Jones,C.D (1968). The Ultraviolet Spectra of Protoberberine pp: 779-784