化学応用分野 Division of Natural Products Chemistry

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◇研究目的及び概要 Research projects

Ⅰ）伝統薬物から栄養飢餓状態で殺細胞作用を有する物質の探索

国立がんセンターとの共同研究で，膵臓癌 PANC-1 細胞株を用い，低栄養状態（IMEM）で PANC-1 に対する殺細胞活性を示し，通常培地（DMEM）では細胞の成育に活性を示さないような薬物を天然資源より探索している。これまでに，伝統薬物600種のエキス中，36エキスに PANC-1 細胞株に対する選択的な細胞毒性があることを見いだした。さらに36の伝統薬物のうち，特に強い活性を示した伝統薬物エキス 6 種を選び，これらについて現在研究を進めている。

Ⅱ）天然薬物から酵母 Ca^{2+} シグナル伝達阻害物質の探索と医薬への応用

広島大学で開発したポジティブスクリーニング法を用いて，新規医薬シーズ開発を目的に研究を行っている。中国及び東南アジア産生薬によるスクリーニングを行い，これまで約1000サンプルの試験を終え，25検体に阻害活性を見い出している。

Ⅲ）インドネシア産薬用植物から抗マラリア活性物質の探索

現代のマラリア流行地は，ほぼ熱帯・亜熱帯に限定され，それら地域では多剤耐性マラリアに有効な新しい抗マラリア薬が必要とされている。我々は東南アジア等で抗マラリア薬として用いられている薬用植物エキスについて多剤耐性マラリアに対する活性をスクリーニングを行い，活性を示した薬用植物中の活性物質を単離・構造解析を行っている。現在，インドネシア産薬用植物 *Caesalpinia crista* について抗マラリア作用を検討し，その活性物質の構造を解析した。

Ⅳ）天然薬物の薬物代謝酵素阻害に関する研究

和漢薬を始めとする天然薬物は，通常，合成医薬品と併用されている現状では，天然薬物が“薬物代謝酵素（シトクロームP450，CYP）”に及ぼす影響（薬物間相互作用）を系統的に検証しておく事が，天然薬物の有効利用の上で必要とされている。我々は，漢方生薬78種及びインドネシアジャムウ生薬30種について CYP3A4 及び CYP2D6 阻害活性を検索し，漢方生薬“五味子”およびジャムウ生薬“*Zingiber aromaticum*”の活性成分を明らかにした。

Ⅴ）骨粗鬆症に有効な天然薬物成分の開発研究

中医学において骨粗鬆症に類似の疾患（骨痿や骨痺）の治療に補腎剤や強筋骨剤が使用されている事に注目し，使用されている漢薬30種について抗骨粗鬆症活性をスクリーニングした。その結果，強い活性を示した漢薬“メンヒセン（*Dioscorea spongiosa* の根茎）”の成分の解明を行い，得られた成分の pQCT 装置による抗骨粗鬆症活性と合わせて，活性本体がステロイド配糖体である事を明らかにした。

また，紅豆杉の活性成分のリグナンが，抗骨粗鬆症活性がある事も判明した。

Ⅵ）ベトナム産生薬の Xanthine Oxidase 阻害活性物質の研究

痛風治療薬の開発を目的に，ベトナム産生薬（98種）について Xanthine Oxidase 阻害作用を指標にスクリーニングした。阻害作用の強かった *Caesalpinia sappan* 中の活性物質の構造を決定した。

Ⅶ）NO 産生阻害活性成分の検索

東南アジア各地で採集した *Orthosiphon stamineus* について，地理的な成分比較ならびに NO 阻害活性物質の構造を解析した。

上記の研究課題によって得られた本年度の成果（原著及び学会報告）は下記の通りである。

◇原著論文 Original papers

1）**Awale S., Tezuka Y., Kobayashi M., Ueda J., and Kadota S.: Neoorthosiphonone A; A Nitric Oxide (NO) Inhibitory Diterpene with New Carbon Skeleton From *Orthosiphon stamineus*. *Tetrahedron Lett.*, 45, 1359-1362 (2004).**

Abstract: From the aerial part of *Orthosiphon stamineus* from Hainan island of China, a diterpene named neoorthosiphonone A (**1**), having a novel carbon framework, has been isolated. Neoorthosiphonone A (**1**) possessed a unique unprecedented structural feature of eight membered ring C in its structure, which may be biogenetically derived from its isopimarane precursor, orthosiphonone A, through the insertion of vinyl group into the C_{13}-C_{14} bond. Neoorthosiphonone A (**1**) displayed potent inhibitory activity on the nitric oxide production in LPS-activated macrophage-like J774.1 cells with an IC_{50} value of 7.08 μM, more potent than the positive control L-NMMA.

2）**Nguyen N. T., Banskota A. H., Tezuka Y., Tran Q. L., Nobukawa T., Kurashige Y., Sasahara M., and Kadota S.: Hepatoprotective Effect of Taxiresinol and (7'*R*)-7'-Hydroxylariciresinol on D-Galactosamine and Lipopolysaccharide-Induced Liver Injury in Mice. *Planta Med.*, 70, 29-33 (2004).**

Abstract: The hepatoprotective effect of taxiresinol (**1**) and (7'*R*)-7'-hydroxylariciresinol (**2**), two tetrahydrofuran-type lignans isolated from the wood of *Taxus yunnanensis,* were investigated on D-galactosamine (D-GalN)/lipopolisaccharide (LPS)-induced hepatic liver injury in mice. Pre-administration of **1** or **2** at doses of 50 and 10 mg/kg (*i.p.*) at 12 and 1 h before D-GalN/LPS injection significantly inhibited hepatocyte DNA fragmentation and apoptotic body formation. Pre-treatment of these two lignans further suppressed hepatic necrosis which occur at later stage of D-GalN/LPS intoxication as demonstrated by the significant and dose-dependent reduction in serum glutamic pyruvic transaminase (sGPT) and serum glutamic oxaloacetic transaminase (sGOT) at 8 h after intoxication. The elevation of serum tumor necrosis factor-alpha (TNF-α) level by D-GalN/LPS toxication was significantly inhibited by **1** or **2** at doses of 50 and 10 mg/kg. Moreover, both of these lignans significantly protected hepatocytes from D-GalN/TNF-α-induced cell death in primary cultured mouse hepatocytes. These results suggested that **1** and **2** had protected the hepatocytes from apoptosis via an inhibition of TNF-α production by activated macrophages and a direct inhibition of apoptosis induced by TNF-α in D-GalN/LPS-treated mice.

3）**Iwata H., Usia T., Tezuka Y., Kadota S., Hiratsuka A., and Watabe T.: Inhibition of Human Liver Microsomal CYP3A4 and CYP2D6 with Extract from 78 Herbal Medicines. *J. Trad. Med.*, 21, 42-50 (2004).**

Abstract: The inhibitory effects of 78 herbal extracts on cytochrome P450 3A4 (CYP3A4) and P450 2D6 (CYP2D6) activity were investigated using human liver microsomes. The incubation mixture contained a methanol soluble fraction prepared from the powder of each herbal water extract (equivalent to 1.65 mg of extract powder per mL). Thirty-one herbal extracts inhibited over 50% of human liver microsomal erythromycin N-demethylation, a marker reaction of CYP3A4 activity. Among the 31 herbal extracts, 8 of them (Angelica Dahurica Root, Cassia Bark, Clove, Incised Notopterygium Rhizome, Moutan Bark, Rhubarb, Sappan Wood, Schisandra Fruit) inhibited N-demethylation by over 90%. Among the herbal extracts examined, the strongest inhibition of CYP3A4 was noted with Sappan Wood, which had an IC_{50} value of 43 μg/mL. Rhubarb, Schisandra Fruit, Incised Notopterygium Rhizome, and Angelica Dahurica Root had IC_{50} values of 77, 127, 144, and 185 μg/mL, respectively. Further, 28 of the herbal extracts inhibited over 50% of human liver microsomal dextromethorphan *O*-demethylation, which is a marker of CYP2D6 activity. Among the 28 herbal extracts, 13 (Cassia Bark, Clove, Coptis Rhizome, Ephedra Herb, Gambir Plant, Incised Notopterygium Rhizome, Magnolia Bark, Moutan Bark, Phellodendron Bark, Rhubarb, Sappan Wood, Sinomenium Stem, Zanthoxylum Fruit) inhibited *O*-demethylation by over 90%. The strongest inhibition of CYP2D6 was noted with Phellodendron Bark, which had an IC_{50} value of 4 μg/mL. Coptis Rhizome,

Sinomenium Stem, Sappan Wood, and Rhubarb showed IC_{50} values of 14, 40, 52, and 64 μg/mL, respectively. These results indicate that many herbal extracts have an inhibitory effect on CYP3A4 and CYP2D6.

4) **Yin J., Tezuka Y., Kouda K., Tran Q. L., Miyahara T., Chen Y., and Kadota S.: Antiosteoporotic Activity of the Water Extract of *Dioscorea spongiosa*. *Biol. Pharm. Bull.*, 27, 583-586 (2004).**

Abstract: After 60 MeOH and water extracts of natural crude drugs were screened for their ability to stimulate osteoblast proliferation, four MeOH extracts (*Cynomorium songaricum, Drynaria fortunei, Licium chinense, Rehmannia glutinosa*) and seven water extracts (*Cornus officinalis, Dendrobium nobile, Dioscorea spongiosa, Drynaria fortunei, Eucommia ulmoides, Lycium chinensis, Viscum coloratum*) showed that potent activities were evaluated for inhibition of osteoclast formation. The results indicated that the water extract of *D. spongiosa* not only showed the strongist stimulation of osteoblast proliferation but also possesed potent inhibitory activity against osteoclast formation, whereas it showed lower cytotoxicity in osteoblast and bone marrow cells. A further *in vivo* experiment determined the antiosteoporotic activity of this extract, in which it inhibited the decrease in cancellous bone mineral content, cancellous bone mineral density, and cortical bone mineral content of the proximal tibia in ovariectomized rats.

5) **Yin J., Kouda K., Tezuka Y., Tran Q. L., Miyahara T., Chen Y., and Kadota S.: New Diarylheptanoids from the Rhizomes of *Dioscorea spongiosa* and Their Antiosteoporotic Activity. *Planta Med.*, 70, 54-58 (2004).**

Abstract: Bioassay-guided fractionation of the water extract of the rhizomes of *Dioscorea spongiosa* led to the isolation and identification of new diarylheptanoids, diospongins A-C, together with three known lignans. Their structures, including absolute stereochemistry, were determined by analyses of NMR data, chemical conversions and CD spectrum. The isolated compounds, except for diospongin A, exerted potent inhibitory activities on bone resorption induced by parathyroid hormone in a bone organ culture system.

6) **Nakano H., Ogura K., Takahashi E., Harada T., Nishiyama T., Muro K., Hiratsuka A., Kadota S., and Watabe T.: Regioselective Monosulfation and Disulfation of the Phytoestrogens Daidzein and Genistein by Human Liver Sulfotransferases. *Drug Metab. Pharmacokinet.*, 19, 216-226 (2004).**

Abstract: Regioselective sulfation of the phytoestrogens daidzein (DZ, 7,4'-dihydroxyisoflavone) and genistein (GS, 5,7,4'-trihydroxyisoflavone) was investigated using human liver cytosol and purified recombinant human sulfotransferase (SULT) isoforms, SULT1A1, SULT1A3, SULT2A1, and SULT1E1. 7-Position-preferential sulfation of DZ and GS was observed in human hepatic cytosols from 3 male and 3 female subjects. Average ratios for 7- to 4'-sulfate formation were 4.5:1 from DZ and 8.4:1 from GS in these human liver cytosols. Apparent K(m) values for the 7- and 4'-sulfation of DZ and GS by these cytosols were similar and in a range from 0.46 to 0.66 microM. All recombinant human SULTs had activity for 7- and 4'-sulfation of these phytoestrogens except for 7-sulfating activity of SULT1A3. SULT1A1 and SULT1E1 exhibited much higher catalytic efficiency, k(cat)/K(m), for 7- and 4'-sulfation of these substrates than did the other two, SULT1A3 and SULT2A1. SULT1A1 showed K(m) values of 0.47 and 0.52 microM for the mono-sulfation of DZ and GS, respectively, which were very similar to those of human cytosol. The observed k(cat)/K(m) indicated that SULT1A1 catalyzed 7-sulfation of DZ and GS at rates 4.4- and 8.8-fold higher, respectively, than such 4'-sulfation. However, with SULT1E1, catalytic efficiency was very similar for the sulfation of both positions. These data strongly suggest that SULT1A1 plays a major role in monosulfation of the phytoestrogens and determines the regioselectivity of sulfation in human hepatic cytosol. A kinetic study for 7,4'-disulfate formation of DZ and GS from their 7- and 4'-monosulfates indicated that SULT1E1

most efficiently catalyzed both reactions among human SULTs.

7) **Nguyen M. T. T., Awale S., Tezuka Y., Chang C.-H., and Kadota S.: Staminane- and Isopimarane-type Diterpenes from *Orthosiphon stamineus* of Taiwan and Their Nitric Oxide Inhibitory Activity. *J. Nat. Prod.*, 67, 654-658 (2004).**

Abstract: From the MeOH extract of Taiwanese *Orthosiphon stamineus,* two new staminane-type diterpenes, staminols C (**1**) and D (**2**), and three new isopimarane-type diterpenes, orthosiphonones C (**3**) and D (**4**) and 14-deoxo-14-*O*-acetylorthosiphol Y (**5**), have been isolated together with 16 known diterpenes, orthosiphols A, B, D, K, M, N, O, X, and Y, norortho siphonolide A, neoorthosiphol B, orthosiphonone A, secoorthosiphols B and C, 3-*O*-deacetylorthosiphol I, and 2-*O*-deacetylorthosiphol J. Their structures were determined based on the spectroscopic data. All the newly isolated diterpenes exhibited dose-dependent inhibition of nitric oxide (NO) production in lipopolysaccharide (LPS)-activated macrophage-like J774.1 cells, and 2-*O*-deacetylorthosiphonone A showed the most potent activity with an IC_{50} value of 35.0 μM, comparable to that of positive control N^G-monomethyl-L-arginine (L-NMMA; IC_{50}, 35.7 μM).

8) **Yin J., Tezuka Y., Kouda K., Tran Q. L., Miyahara T., Chen Y., and Kadota S.: *In vivo* Antiosteoporotic Activity of a Fraction of *Dioscorea spongiosa* and Its Constituent, Methyl Protodioscin. *Planta Med.*, 70, 220-226 (2004).**

Abstract: The antiosteoporotic activity of the 90% EtOH fraction of the water extract of rhizomes of *Dioscorea spongiosa* and methylprotodioscin, its major constituent, were examined in the model of postmenopausal bone loss using ovariectomized (OVX) rats or mice. After 6 weeks treatment, the proximal tibia of rats or mice and the distal femora of mice were scanned by peripheral quantitative computed tomography (pQCT). Both the 90% EtOH fraction (100 mg/kg/d) and methylprotodioscin (50 mg/kg/d) significantly inhibited bone loss in bone mineral content (BMC) and bone mineral density (BMD) in total, cancellous and cortical bones, and the decrease in bone strength indexes induced by OVX, without side effect on the uterus.

9) **Banskota A. H., Nguyen N. T., Tezuka Y., Tran Q. L., Nobukawa T., Kurashige Y., Sasahara M., and Kadota S.: Secoisolariciresinol and isotaxiresinol inhibit tumor necrosis factor-α-dependent hepatic apoptosis in mice. *Life Sci.*, 74, 2781-2792 (2004).**

Abstract: The effect of secoisolariciresinol (**1**) and isotaxiresinol (**2**), two major lignans isolated from the wood of *Taxus yunnanensis,* were investigated on tumor necrosis factor-α (TNF-α)-dependent hepatic apoptosis induced by D-galactosamine (D-GalN)/lipopolysaccharide (LPS) in mice. Co-administration of D-GalN (700 mg/kg) and LPS (10 μg/kg) resulted typical hepatic apoptosis characterized by DNA fragmentation and apoptotic body formation in mice. The serum glutamic pyruvic transaminase (sGPT) and glutamic oxaloacetic transaminase (sGOT) were also raised at 8 h after D-GalN/LPS intoxication due to severe necrosis of the hepatocytes. Pre-administration of **1** or **2** (50, 10 mg/kg, *i.p.*) at 12 and 1 h before D-GalN/LPS intoxication significantly reduced DNA fragmentation and prevented the emergence of chromatin condensation, apoptotic body formation and hepatitis of the mice. TNF-α secreted from LPS-activated macrophages is an important mediator for hepatocyte apoptosis in this model. Pretreatment of **1** or **2** significantly inhibited the elevation of serum TNF-α level. In a separate experiment, both lignans showed significant and dose-dependent hepatocyte protective effect towards D-GalN/TNF-α-induced cell death in primary cultured mouse hepatocytes. These results indicated that **1** and **2** protect D-GalN/LPS-induced hepatic injury by inhibiting hepatocyte apoptosis through blocking TNF-α production from activated macrophages and a direct inhibition of apoptosis induced by TNF-α.

10) **Usia T., Iwata H., Hiratsuka A., Watabe T., Kadota S., and Tezuka Y.: Sesquiterpenes and Flavonol Glycosides From *Zingiber aromaticum* and Their CYP3A4 and CYP2D6 Inhibitory Activities. *J. Nat. Prod.*, 67, 1079-1083 (2004).**

Abstract: Three new sesquiterpenes, (2*R*,3*S*,5*R*)-2,3-epoxy-6,9-humuladien-5-ol-8-one (**1**), (2*R*,3*S*,5*R*)-2,3-epoxy-6,9-humuladien-5-ol-8-one (**2**), and (5*R*)-2,6,9-humulatrien-5-ol-8-one (**3**), and two new flavonol glycosides, kaempferol-3-*O*-(2,3-di-*O*-acetyl-α-L-rhamnopyranoside) (**4**) and kaempferol-3-*O*-(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranoside) (**5**), were isolated from the EtOAc-soluble fraction of the water extract of *Zingiber aromaticum,* along with 13 known compounds (**6-18**). The structures of the isolated compounds were elucidated on the basis of spectroscopic and chemical analyses. The isolated compounds were tested for their inhibitory activity on the metabolism mediated by CYP3A4 or CYP2D6 using [*N*-methyl-^{14}C]erythromycin or [*O*-methyl-^{14}C]dextromethorphan as a substrate, respectively. Kaempferol-3-*O*-(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranoside) (**5**) showed the most potent inhibitory activity (IC_{50}, 14.4 μM) on the metabolism mediated by CYP3A4 and kaempferol-3-*O*-methylether (**14**) inhibited CYP2D6 most potently (IC_{50}, 4.63 μM).

11) **Nguyen M. T. T., Awale S., Tezuka Y., Tran Q. L., Watanabe H., and Kadota S.: Xanthine Oxidase Inhibitory Activity of Vietnamese Medicinal Plants. *Biol. Pharm. Bill.*, 27, 1414-1421 (2004).**

Abstract: Among 288 extracts, prepared from 96 medicinal plants used in Vietnamese traditional medicine to treat gout and related symptoms, 188 demonstrated xanthine oxidase (XO) inhibitory activity at 100 μg/ml, with 46 having greater than 50% inhibition. At 50 μg/ml, 168 of the extracts were active, with 21 possessing more than 50% inhibition. At 25 μg/ml, 146 extracts exhibited inhibitory activity, with 8 showing over 50 % inhibition, while 126 extracts presented activity at 10 μg/ml, with 2 having greater than 50% inhibition. The MeOH extracts of *Artemisia vulgaris, Caesalpinia sappan* (collected at the Seven-Mountain area), *Blumea balsamifera* (collected in Lam Dong province), *Chrysanthemum sinense* and MeOH-H_2O extract of *Tetracera scandens* (Khanh Hoa province) exhibited strong XO inhibitory activity with IC_{50} values less than 20 μg/ml. The most active extract was the MeOH extract of the flower of C. sinense with an IC_{50} value of 5.1 μg/ml. Activity-guided fractionation of the MeOH extract led to the isolation of caffeic acid (**1**), luteolin (**2**), eriodictyol (**3**), and 1,5-di-*O*-caffeoylquinic acid (**4**). All these compounds showed significant XO inhibitory activity in a concentration-dependent manner, and the activity of **2** was more potent (IC_{50} 1.3 μM) than the clinically used drug, allopurinol (IC_{50} 2.5 μM).

12) **Nguyen M. T. T., Awale S., Tezuka Y., Tran Q. L., and Kadota S.: Neosappanone A, a Xanthine Oxidase (XO) Inhibitory Dimeric Methanodibenzoxocinone with New Carbon Skeleton from *Caesalpinia sappan. Tetrahedron Lett.*, 45, 8519-8522 (2004).**

Abstract: A novel dimeric methanodibenzoxocinone, named neosappanone A (**1**), possessing a unique unprecedented novel carbon framework, has been isolated from the heartwood of *Caesalpinia sappan* L. of Vietnam, and its structure was elucidated on the basis of spectroscopic analysis. Neosappanone A (**1**) competitively inhibited xanthine oxidase in a concentration-dependent manner (IC_{50}, 29.7 μM; *Ki*, 16.3 μM).

13) **Iwata H., Tezuka Y., Kadota S., Hiratsuka A., and Watabe T.: Identification of Potent CYP3A4 Inhibitors in Schizandra Fruit Extract. *Drug Metab. Disp.*, 32, 1351-1358 (2004).**

Abstract: Schisandra fruit, a Schisandraceae family herb, is used as a component in Kampo medicines (developed from Chinese medicines, but established in Japan). It can act as a sedative and antitussive, improve hepatic function, and give a general tonic effect. An extract of Schisandra fruit has been shown with a potent inhibitory effect on human liver microsomal erythromycin *N*-demethylation activity mediated by cytochrome P450 3A4 (CYP3A4). The present study was conducted to identify Schisandra fruit components having inhibitory effects on CYP3A4 by

surveying the effect on human liver microsomal erythromycin *N*-demethylation activity. Known components of Schisandra fruit, gomisins B, C, G, and N and γ-shizandrin, showed inhibitory effects on *N*-demethylation activity. Among these components, gomisin C displayed the most potent and competitive inhibitory effect with a *K*i value of 0.049 μM. Furthermore, the inhibitory effect of gomisin C was stronger than that of ketoconazole (*K*i = 0.070 μM), a known potent CYP3A4 inhibitor. Gomisin C, however, inhibited CYP1A2-, CYP2C9-, CYP2C19-, and CYP2D6-dependent activities only to a limited extent (IC_{50} values > 10 μM). Moreover, gomisin C inactivated human liver microsomal erythromycin N-demethylation activity in a time- and concentration-dependent manner. The inactivation kinetic parameters k_{inact} and K_I were 0.092 min^{-1} and 0.399 μM, respectively. The human liver microsomal erythromycin *N*-demethylation activity inactivated by gomisin C did not recover on dialysis of the microsomes. Spectral scanning of CYP3A4 with gomisin C yielded an absorbance at 455 nm suggesting gomisin C inactivated the CYP via the formation of a metabolite intermediate complex. This pattern is consistent with the metabolism of the methylenedioxy substituent in gomisin C. These results indicate that gomisin C is a mechanism-based inhibitor that not only competitively inhibits but irreversibly inactivates CYP3A4.

14) **Iwata H., Tezuka Y., Kadota S., Hiratsuka A., and Watabe T.: Metabolism-Dependent Inhibition of CYP3A4 and CYP2D6 by Extracts from 26 Herbal Medicines. *J. Trad. Med.*, 21, 281-286 (2004).**

Abstract: A total of 26 herbal medicines were examined for their inhibitory effects on cytochrome P450 3A4 (CYP3A4) and 2D6 (CYP2D6). A methanol extract of each herbal medicine was prepared and then preincubated with human liver microsomes in the presence of an NADPH-generating system. Residual microsomal CYP3A4 and CYP2D6 activity was then determined by measuring the *N*-demethylation of erythromycin and the *O*-demethylation of dextromethorphan, respectively. Of the 26 herbal medicines tested, 16 were found to decrease the residual CYP3A4 activity in a preincubation time-dependent manner. The extract of Evodia Fruit caused the most dramatic decrease in residual CYP3A4 activity (i.e. 22.3% residual activity after 30 min preincubation). A substantial decrease in residual CYP3A4 activity was also observed from extracts of Sappan Wood, Incised Notopterygium Rhizome, Schisandra Fruit, Great Burdock Achene, Angelica Dahurica Root and Rhubarb (residual activity of 40.6, 41.2, 53.4, 47.1, 53.4 and 59.2% after 30 min preincubation, respectively). These results are comparable to those using troleandmycin, a known irreversible inhibitor of CYP3A4, which gave a residual activity of 49.4% under identical conditions. We found 5 herbal medicines that showed a preincubation time-dependent inhibition of CYP2D6. The extract of Incised Notopterygium Rhizome caused the most dramatic decrease in residual CYP2D6 activity (i.e. 61.9% residual activity after 30 min preincubation). These results suggest that extracts of herbal medicines contain metabolism-dependent inhibitors of CYP, especially CYP3A4.

15) **Kalauni S. K., Awale S., Tezuka Y., Banskota A. H., Linn T. Z., and Kadota S.: Cassane- and Norcassane-type Diterpenes of *Caesalpinia crista* from Myanmar. *J. Nat. Prod.*, *67*, 1859-1863 (2004).**

Abstract: From the CH_2Cl_2 extract of seed kernels of *Caesalpinia crista* from Myanmar, five new cassane-type diterpenes, caesalpinins MA-ME(**1-5**), and three new norcassane-type diterpenes, norcaesalpinins MA-MC (**6-8**), have been isolated, together with 12 known cassane-type diterpenes, 14(17)-dehydrocaesalmin F, caesaldekarin e, caesalmin B, caesalmin C, caesalmin E, 2-acetoxy-3-deacetoxycaesaldekarin e, 2-acetoxycaesaldekarin e, caesalpinin C, 7-acetoxybonducellpin C, caesalpinin E, norcaesalpinin B, and 6-acetoxy-3-deacetoxycaesaldekarin e. The structures of the isolated compounds were elucidated by analysis of their spectroscopic data.

◇総説　Review papers

1）門田重利，手塚康弘：プロポリス成分 CAPE およびその類縁体の癌転移抑制活性に関する研究.

ミツバチ科学 (*Honeybee Science*), 25, 107-112 (2004).

◇学会報告　Scientific presentation (*: 招待講演)

＊1) Shigetoshi Kadota, Arjun H. Banskota, Yasuhiro Tezuka, Nhan Trung Nguyen and Takahiro Nobukawa: Chemical Constituents and Biological Activities of the Wood of *Taxus yunnanensis*. IUPAC International Conference on Biodiversity and Natural Products: Chemistry and Medical Application, 2004, 1, New Delhi, India.

2) Suresh Awale, 手塚康弘, 上田純也, 門田重利：Study on the bioactive constituents from Brazilian medicinal plant *Tabebuia avellanedae* "Taheebo". 日本薬学会第124年会, 2004, 3, 大阪.

3) Surya K. Kalauni, Arjun H. Banskota, 手塚康弘, 門田重利：New Cassane- and Norcassane-type Diterpenes of *Caesalpinia crista* from Myanmar. 日本薬学会第124年会, 2004, 3, 大阪.

4) Thein Zaw Linn, 手塚康弘, Suresh Awale, Arjun H. Banskota, Faisal Attamimi, 門田重利：New Cassane- and 17-norcassane-type diterpenes of *Caesalpinia crista* from Indonesia. 日本薬学会第124年会, 2004, 3, 大阪.

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＊2）門田重利：Recent Studies on the Natural Medicine. 2004, 3/13, エジプト, ナショナル・リサーチセンター.
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◇海外調査　Field work

1）門田重利, Suresh Awale, 上田純也, Mai Thanh Thi Nguyen, Subehan：文部科学省科学研究費, 基盤研究B（2）「ベトナム, ミャンマーおよびインドネシアにおける伝統医学と天然薬物資源の調査研究」, 2004, 10/25-12/4, ベトナム, ミャンマー, インドネシア.

◇共同研究　Co-oparative researches

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1）江角浩安：国立がんセンター研究所支所,「がん生物学に基づく新しい治療法に関する研究」, 2003, 4～
2）宮川都吉：広島大学大学院・先端物質科学研究科,「酵母 Ca^{2+} シグナルによる細胞周期制御に関する総合的研究」, 2004, 4～
3）平塚　明：東京薬科大学,「和漢薬が関与する薬物相互作用に関する研究」, 2002, 4～
4）信川高寛：金沢医科大学,「紅豆杉の生物活性物質の探索」, 2004, 4～

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1）Dejair Message, Alfredo A. G. Fuertas：ブラジル・ヴィソーサ大学,「プロポリスの品質評価に関する研究」, 1996, 10～
2）殷　軍：中国・瀋陽薬科大学,「漢方方剤の抗骨粗鬆症活性成分に関する研究」, 2004, 10～
3）李　建新：中国・南京大学化学工学院・薬物化学研究所,「抗骨粗鬆症に有効な薬物の開発研究」, 2004, 4～
4）Tran Le Quan：ベトナム・国立ホーチミン市大学,「ベトナム産薬用植物の科学的評価に関する研究」, 2003, 4～

◇研究費取得状況　Acquisition of research funds

1）文部科学省科学研究費, 基盤研究B（2）（代表：門田重利）「ベトナム, ミャンマーおよびインドネシアにおける伝統医学と天然薬物資源の調査研究」
2）厚生労働省がん研究助成（分担：門田重利）「がん生物学に基づく新しい治療法に関する研究」
3）平成16年度研究拠点形成費補助金（COE）（フェロー：手塚康弘）「東洋の知に立脚した個の医療の創生」
4）平成16年度上原記念生命科学財団研究助成金（代表：門田重利）「漢方薬と合成医薬品の薬物相互作用」

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大学院後期1年：史　麗穎（2004，4～）
大学院後期2年：Surya Kant Kalauni
大学院後期3年：Tepy Usia，岩田　宏，Mai Thanh Thi Nguyen
外国人研究生：李　峰（2004，12/23～2005，3/31）
外国人特別研究学生：李　霞（日本国際教育協会，瀋陽薬科大学大学院，2004，5/8～2005，5/7）
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◇学位（修士，博士）取得者　Academic degrees and theses

修士論文（2004年3月）：

Thein Zaw Linn : Chemical Constituents of the Seed Kernels of *Caesalpinia crista*

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