Antitumor Agents. 282. 2′-(*R*)-*O*-Acetylglaucarubinone, a Quassinoid from *Odyendyea gabonensis* As a Potential Anti-Breast and Anti-Ovarian Cancer Agent

Yoshihide Usami,[†,⊥] Kyoko Nakagawa-Goto,[†,⊥] Jing-Yu Lang,[‡] Yoon Kim,[§] Chin-Yu Lai,[†] Masuo Goto,[¯] Nobuko Sakurai,[†] Masahiko Taniguchi,[†] Toshiyuki Akiyama,[†] Susan L. Morris-Natschke,[†] Kenneth F. Bastow,[∥] Gordon Cragg,[▽] David J. Newman,[▽] Mihoyo Fujitake,[o] Koichi Takeya,[□] Mien-Chie Hung,[‡] Eva Y.-H. P. Lee,[§] and Kuo-Hsiung Lee*[,†,^]

Natural Products Research Laboratories, Eshelman School of Pharmacy, University of North Carolina, Chapel Hill, North Carolina 27599-7568, The University of Texas M. D. Anderson Cancer Center, Houston, Texas 77030, Biological Chemistry and Developmental and Cell Biology, College of Medicine, University of California, Irvine, California 92697, Cell and Developmental Biology, School of Medicine, University of North Carolina, Chapel Hill, North Carolina 27599-7090, Division of Medicinal Chemistry & Natural Products, Eshelman School of Pharmacy, University of North Carolina, Chapel Hill, North Carolina 27599-7568, Frederick Cancer Research and Development Center, National Cancer Institute, P.O. Box B, Frederick, Merryland 21702-1201, Osaka University of Pharmaceutical Sciences, Takatsuki, Osaka 569-1094, Japan, Tokyo University of Pharmacy and Life Science, Hachioji, Tokyo 192-0355, Japan, and Chinese Medicine Research and Development Center, China Medical University and Hospital, Taichung, Taiwan, Republic of China

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A new quassinoid, designated 2′-(*R*)-*O*-acetylglaucarubinone (**1**), and seven known quassinoids (**2**–**8**) were isolated, using bioactivity-guided separation, from the bark of *Odyendyea gabonensis* (Pierre) Engler [syn. *Quassia gabonensis* Pierre]. The structure of **1** was determined by spectroscopic analysis and by semisynthesis from glaucarubolone. Complete ^{1}H and ^{13}C NMR assignments of compounds **1**–**8** were also established from detailed analysis of two-dimensional NMR spectra, and the reported configurations in odyendene (**7**) and odyendane (**8**) were corrected. Compound **1** showed potent cytotoxicity against multiple cancer cell lines. Further investigation using various types of breast and ovarian cancer cell lines suggested that **1** does not target the estrogen receptor or progesterone receptor. When tested against mammary epithelial proliferation in vivo using a *Brca1/p53*-deficient mice model, **1** also caused significant reduction in mammary duct branching.

Quassinoids are known as the bitter principles of Simaroubaceous plants.[1−3] They are highly oxygenated triterpenes and possess a wide spectrum of in vitro and in vivo biological activities, including antitumor, antimalarial, antiviral, anti-inflammatory, antifeedant, insecticidal, amoebicidal, antiulcer, and herbicidal effects.[1] In particular, the quassinoid bruceantin was brought to a phase II clinical trial as an anticancer drug candidate;[4] however, lack of significant efficacy in treating human cancer led to termination of its clinical development in the early 1980s.[5] From the initial studies on various quassinoids, the mechanism of action was attributed to the inhibition of site-specific protein synthesis by prevention of ribosomal peptidyl transferase activity, leading to termination of chain elongation.[6] However, other postulated mechanisms for inhibition of cancer cell growth include, but are not limited to, inhibition of plasma membrane NADH oxidase activity,[7] down-regulation of c-*myc* oncogene,[8] and mitochondrial membrane depolarization with caspase-3 activation.[9]

The stem bark of *Odyendyea gabonensis* (Pierre) Engler [syn. *Quassia gabonensis* Pierre (Simaroubaceae)] is a source of quassinoids, and seven quassinoids, 2′-(*S*)-*O*-acetylglaucarubinone (**2**), glaucarubinone (**3**), ailanthinone (**4**), 2′-(*R*)-*O*-acetylglaucarubin (**5**), excelsin (**6**), odyendene (**7**), and odyendane (**8**), were previously isolated.[10−12] We have reinvestigated the stem bark of this plant due to its potent selective cytotoxicity against breast cancer cell lines in our prior studies aimed at discovering antitumor agents from higher plants. Bioactivity-directed fractionation of this plant extract led to the isolation and characterization of a new quassinoid, 2′-(*R*)-*O*-acetylglaucarubinone (**1**), as an active principle, along with **2**–**8**. We describe herein the isolation and structure determination of **1** by semisynthesis. The isolated quassinoids were evaluated in vitro against human tumor cell replication (DU145 prostate cancer, A549 human lung carcinoma, KB human epidermoid carcinoma of the nasopharynx, and KB-V multi-drug-resistant expressing P-glycoprotein). Compound **1** was also further investigated for in vitro cytotoxic activity against multiple breast cancer cell lines. We also describe the effect of **1** on mammary epithelial proliferation in vivo using a *Brca1/p53*-deficient mice model.

During this investigation, detailed analyses of ^{1}H and ^{13}C NMR spectra of quassinoids **1**–**8** were conducted and led to the revision of some previously reported data of **2**–**6**, as well as the correction of configurations in odyendene (**7**) and odyendane (**8**).[12] For the convenience of comparison and discussion, 2′-acetylglaucarubinone (**2**) is referred to as 2′-(*S*)-*O*-acetylglaucarubinone and its 2′-epimer (**1**) as 2′-(*R*)-*O*-acetylglaucarubinone.

Results and Discussion

O. gabonensis was collected in Gabon in 1991 by NCI. The active MeOH/CH_2Cl_2 (1:1) extract of the bark was fractionated into hexane- and EtOAc-soluble fractions, as well as an insoluble residue. The active EtOAc-soluble fraction was subjected to silica gel column chromatography (CC), (hexane/EtOAc gradient, then MeOH), followed by reversed-phase HPLC, to give the new compound **1** and the known quassinoids **2**–**8**.

Compound **1** was isolated as a colorless, amorphous solid, and its HREIMS indicated a molecular formula of $C_{27}H_{36}O_{11}$, which was identical with that of **2**. The close similarity in the ^{1}H and ^{13}C NMR spectra (Tables 1 and 2, respectively) of **1** and **2**, including

* To whom correspondence should be addressed. Tel: 919-962-0066. Fax: 919-966-3893. E-mail: khlee@unc.edu.
† Natural Products Research Laboratories, University of North Carolina.
⊥ Equal Contribution.
‡ The University of Texas M. D. Anderson Cancer Center.
§ Biological Chemistry and Developmental and Cell Biology, University of California.
¯ Cell and Developmental Biology, School of Medicine, University of North Carolina.
∥ Division of Medicinal Chemistry & Natural Products, University of North Carolina.
▽ Frederick Cancer Research and Development Center.
o Osaka University of Pharmaceutical Sciences.
□ Tokyo University of Pharmacy and Life Science.
^ China Medical University and Hospital.

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