Anti-tuberculosis Activity of Quassinoids

Shakila RAHMAN,[a] Narihiko FUKAMIYA,[a] Masayoshi OKANO,*[,a] Kiyoshi TAGAHARA,[b] and Kuo-Hsiung LEE[c]

Department of Interdisciplinary Studies on Natural Environment, Faculty of Integrated Arts and Sciences, Hiroshima University,[a] 1–7–1 Kagamiyama, Higashi-Hiroshima 739, Japan, Faculty of Pharmaceutical Sciences, Kobe Pharmaceutical University,[b] 1–19–4 Motoyamakita-machi, Higashinada-ku, Kobe 658, Japan, and Division of Medicinal Chemistry and Natural Products, School of Pharmacy, University of North Carolina,[c] Chapel Hill, N.C. 27599–7360, U.S.A. Received April 10, 1997; accepted May 19, 1997

***In vitro* evaluation of anti-tuberculosis activity was conducted for fifty-six quassinoids isolated in our laboratory from Simaroubaceous plants, *Ailanthus altissima* (=Aa, 10 compounds), *Brucea antidysenterica* (=Ba, 16 compounds), *Picrasma ailanthoides* (=Pa, 14 compounds), and *Brucea javanica* (=Bj, 16 compounds). Of the compounds tested, shinjulactone K (1), ailanthone (2), shinjudilactone (3), and dehydrobruceantin (4) were the most potent. Although the activities were very low (0—19%), the resulting data provided a picture of structure–activity relationships.**

Key words anti-tuberculosis activity; quassinoid; structure–activity relationship

Many quassinoids isolated from the Simaroubaceous plants have biological activities,[1] such as antitumor, anti-inflammatory, antimalarial, amoebicidal, antifeedant, insecticidal, and herbicidal effects.

Recently, we have isolated eighteen new natural cytotoxic antitumor quassinoids[2] and we also prepared thirty-three new cytotoxic antitumor quassinoid derivatives.[3] We have also reported the inhibitory activities of forty-five natural quassinoids on 12-*O*-tetradecanoylphorbol-13-acetate (TPA)-induced Epstein–Barr virus early antigen (EBV-EA) activation and, by implication, their potential as antitumor-promoting agents.[4] Furthermore, we reported the inhibitory activity of twenty-seven natural quassinoids on HIV replication in H9 lymphocytic cells and the responses were very positive.[5] We have also reported quassinoids with potent antifeedant and insecticidal activity against the diamondback moth (*Plutella xylostella*).[6] Thus, the quassinoids have versatile biological activities and display a wide spectrum of structure–activity relationships.

Recently, the number of tuberculosis patients has been increasing globally. Antibiotics[7] such as rifampin, isoniazid, streptomycin, and combinations of other drugs are used for the treatment of tuberculosis, but resistance leading to tolerance is emerging and new treatments are required. TAACF (the tuberculosis antimicrobial acquisition and coordinating facility)[8] has set up a new drug screening program to discover novel agents for the treatment of tuberculosis. Recently, we sent fifty-six quassinoids[2a,2b,2h,2j,9–13] to TAACF for screening as anti-tuberculosis agents. In this paper, we report the results of anti-tuberculosis screening of these quassinoids.

Results and Discussion

Eighteen compounds (**1**—**18**) of the fifty-six (**1**—**56**) quassinoids examined showed anti-tuberculosis activity. Their structures are displayed in Fig. 1 and their activities are shown in Table 1. Rifampin was used as the positive control drug. The compound numbers also indicate the order of their activities. Although the activities of the quassinoids tested were very low, a structure–activity analysis may provide clues for the developement of more active compounds, and structural diversity is very important for the drug discovery effort.

On the basis of the anti-tuberculosis data and the structures of the eighteen quassinoids, the following structure–activity relationships appear to hold: (1) the carbonyl group in ring A may play an important role in the emergence of the activity; (2) some quassinoids with a side chain at C-15 are active and an aliphatic side chain is preferred to an aromatic one; (3) acetylation of the aliphatic side chain decreases the activity; (4) a hemiacetal group in ring D is preferred rather than a lactone group; and (5) a sugar moiety decreases the activity.

Experimental

Materials Most quassinoids used for testing were obtained from Simaroubaceous plants, *Ailanthus altissima* (=Aa), *Brucea anti-*

Table 1. Inhibitory Effects of Quassinoids as Anti-tuberculosis Agents

No.	Compound	% Inhibition at 12.5 μg/ml	Plant[a)]
1	Shinjulactone-K	19	Aa
2	Ailanthone	17	Aa
3	Shinjudilactone	15	Aa
4	Dehydrobruceantin	15	Ba
5	Nigakihemiacetal-D	12	Pa
6	Amarolide	12	Aa
7	Bruceantin	9	Ba
8	Nigakilactone-L	9	Pa
9	Dehydrobruceantarin	8	Ba
10	Neoquassin	8	Pa
11	Nigakihemiacetal-A	8	Pa
12	Bruceoside-D	7	Bj
13	Quassin	7	Pa
14	Nigakilactone-H	5	Pa
15	Nigakilactone-E	5	Pa
16	Picrasin-A	4	Pa
17	Bruceanol-F	3	Ba
18	Dehydrobruceantinol	1	Ba

The test organism was *Mycobacterium tuberculosis*, strain H37Rv, and the concentration of test compounds was 12.5 μg/ml. The lowest concentration inhibiting 99% of the inoculum (MIC) was >12.5 μg/ml. The MIC of rifampin was 0.031 μg/ml, (97% inhibition). *a)* Aa = *Ailanthus altissima*; Ba = *Brucea antidysenterica*; Pa = *Pirasma ailanthoides*; Bj = *Brucea javanica*.

* To whom correspondence should be addressed.

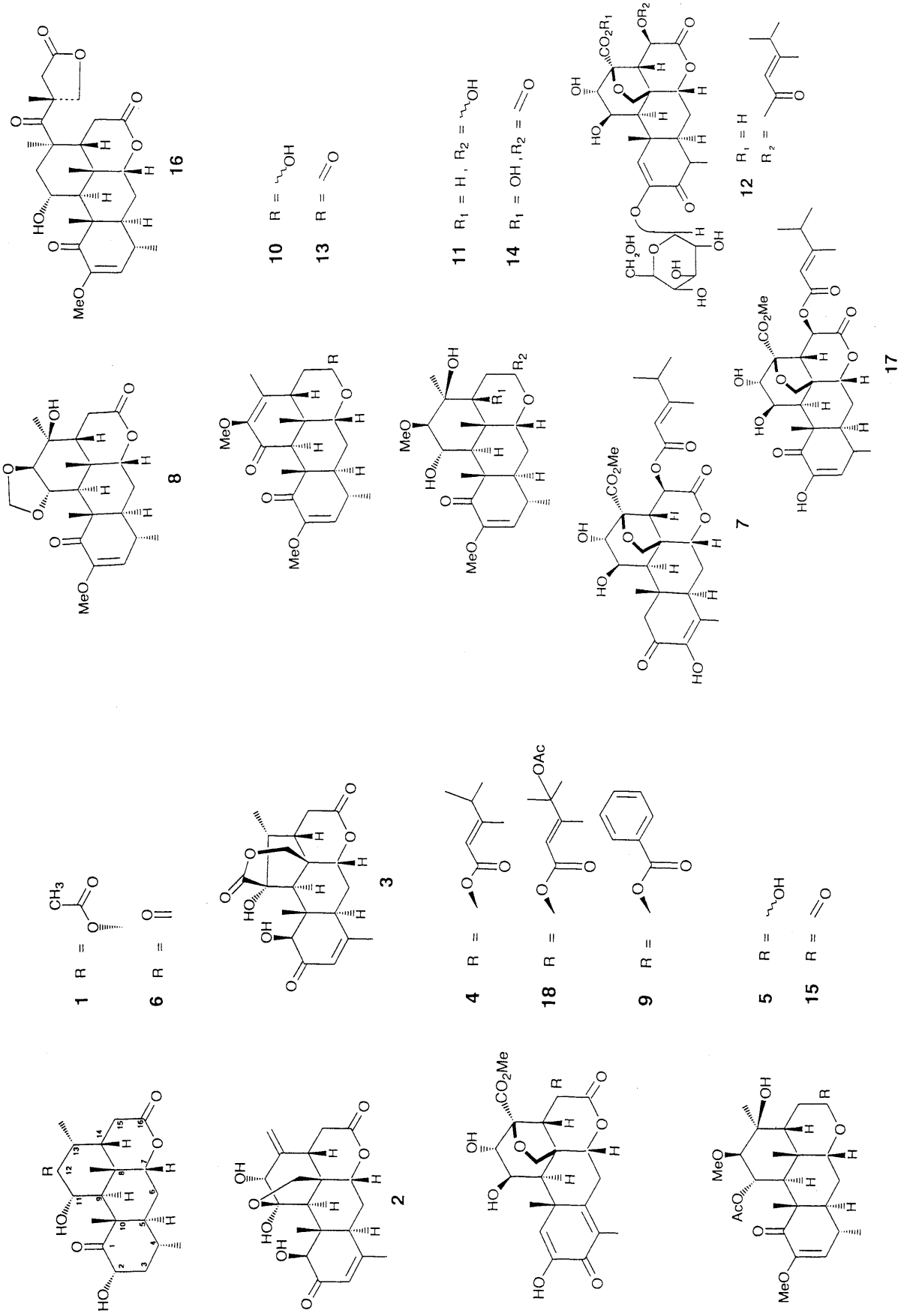

Fig. 1. Structures of Quassinoids with Anti-tuberculosis Activity

dysenterica (=Ba), *Picrasma ailanthoides* (=Pa), and *Brucea javanica* (=Bj), as follows. Shinjulactone-K (**1**), ailanthone (**2**), shinjudilactone (**3**), amarolide (**6**), $\Delta^{13(18)}$-dehydroglaucarubinone (**51**), ailantinol-A (**52**), shinjulactone-A (**53**), shinjulactone-C (**54**), amaloride 11-acetate (**55**) and shinjulactone-B (**56**) were isolated from Aa.[9] Dehydrobruceantin (**4**), bruceantin (**7**), dehydrobruceantarin (**9**), bruceanol-F (**17**), dehydrobruceantinol (**18**), yadanzioside-M (**31**), isobrucein-B (**35**), bruceanol-D (**36**), bruceanol-B (**37**), bruceanol-A (**38**), bruceolide (**39**), bruceanol-E (**40**), bruceanol-H (**41**), yadanzioside-N (**42**), bruceantinoside-C (**43**) and bruceantinoside-B (**44**) were isolated from Ba.[2a,2b,2h,2j,10] Isobrucein-B-hy (**34**) was prepared by alkaline hydrolysis of **35**.[11] Nigakihemiacetal-D (**5**), nigakilactone-L (**8**), neoquassin (**10**), nigakihemiacetal-A (**11**), quassin (**13**), nigakilactone-H (**14**), nigakilactone-E (**15**), picrasin-A (**16**), picrasin-B (**45**), nigakilactone-F (**46**), picrasinol-C (**47**), picrasin-D (**48**), picrasin-E (**49**) and picrasinol-D (**50**) were obtained from Pa.[12] Bruceoside-D (**12**), brucein-D (**19**), brucein-E (**20**), yadanzioside-E (**21**), yadanzioside-B (**22**), bruceoside-B (**23**), yadanzioside-L (**24**), yadanzioside-F (**25**), yadanzioside-A (**26**), bruceoside-A (**27**), bruceantinoside-A (**28**), yadanzioside-C (**29**), yadanzioside-G (**30**), bruceoside-E (**32**) and bruceoside-F (**33**) were obtained from Bj.[13]

***In Vitro* Evaluation of Anti-tuberculosis Activity** This primary screening of quassinoids was conducted at 12.5 μg/ml against *Mycobacterium tuberculosis* H37Rv in BACTEC 12B medium using the BACTEC 460 radiometric system by the TAACF at the Southern Research Institute, U.S.A.

Acknowledgements The authors thank Dr. Joseph A. Maddry, Manager of the TAACF, for his help in screening of quassinoids as anti-tuberculosis agents.

References and Notes

1) Okano M., Fukamiya N., Lee K. H., "Studies in Natural Products Chemistry, Vol. 7, Structure and Chemistry (Part A)," ed. by Atta-ur-Rahman, Elsevier, Amsterdam, 1990, pp. 369—404.
2) *a*) Okano M., Lee K. H., Hall I. H., Boettner F. E., *J. Nat. Prod.*, **44**, 470—474 (1981); *b*) Okano M., Fukamiya N., Aratani T., Ju-ichi M., Lee K. H., *ibid.*, **48**, 972—975 (1985); *c*) Fukamiya N., Okano M., Tagahara K., Aratani T., Muramoto Y., Lee K. H., *ibid.*, **50**, 1075—1079 (1987); *d*) Fukamiya N., Okano M., Tagahara K., Aratani T., Lee K. H., *ibid.*, **51**, 349—352 (1988); *e*) Toyota T., Fukamiya N., Okano M., Tagahara K., Chang J. J., Lee K. H., *ibid.*, **53**, 1526—1532 (1990); *f*) Matsuzaki T., Fukamiya N., Okano M., Fujita T., Tagahara K., Lee K. H., *ibid.*, **54**, 844—848 (1991); *g*) Fukamiya N., Okano M., Miyamoto M., Tagahara K., Lee K. H., *ibid.*, **55**, 468—475 (1992); *h*) Imamura K., Fukamiya N., Okano M., Tagahara K., Lee K. H., *ibid.*, **56**, 2091—2097 (1993); *i*) Ohnishi S., Fukamiya N., Okano M., Tagahara K., Lee K. H., *ibid.*, **58**, 1032—1038 (1995); *j*) Imamura K., Fukamiya N., Nakamura M., Okano M., Tagahara K., Lee K. H., *ibid.*, **58**, 1915—1919 (1995).
3) *a*) Lee K. H., Okano M., Hall I. H., Brent D. A., Soltmann B., *J. Pharm. Sci.*, **71**, 338—345 (1982); *b*) Imamura K., Fukamiya N., Okano M., Tagahara K., Lee K. H., *Bull. Chem. Soc. Jpn.*, **65**, 2684—2689 (1992).
4) Okano M., Fukamiya N., Tagahara K., Tokuda H., Ishikawa A., Nishino H., Lee K. H., *Cancer Lett.*, **94**, 139—146 (1995).
5) Okano M., Fukamiya N., Tagahara K., Cosentino M., Lee T. T. Y., Natschke S. M., Lee K. H., *Bio-Org. & Med. Chem. Lett.*, **6**, 701—706 (1996).
6) *a*) Daido M., Fukamiya N., Okano M., Tagahara K., Hatakoshi M., Yamazaki H., *Biosci. Biotech. Biochem.*, **57**, 244—246 (1993); *b*) Daido M., Ohno N., Imamura K., Fukamiya N., Hatakoshi M., Yamazaki H., Tagahara K., Lee K. H., Okano M., *ibid.*, **59**, 974—979 (1995).
7) The official statement of the American Thoracic Society,"Treatment of tuberculosis and other mycobacterial diseases," *Amer. Rev. Respir., Dis.*, **127**, 790—796 (1982).
8) This program was supported by the U. S. National Institute of Allergy and Infectious Diseases (NIAID).
9) Kubota K., Fukamiya N., Hamada T., Okano M., Tagahara K., Lee K. H., *J. Nat. Prod.*, **59**, 683—686 (1996).
10) Okano M., Fukamiya N., Toyota T., Tagahara K., Lee K. H., *J. Nat. Prod.*, **52**, 398—401 (1989).
11) Rahman S., Fukamiya N., Ohno N., Tokuda H., Nishino H., Tagahara K., Lee K. H., Okano M., *Chem. Pharm. Bull.*, **45**, 675—677 (1997).
12) *a*) Daido M., Fukamiya N., Okano M., Tagahara K., *J. Nat. Prod.*, **55**, 1643—1647 (1992); *b*) Daido M., Fukamiya N., Okano M., Tagahara K., *ibid.*, **58**, 605—608 (1995).
13) *a*) Fukamiya N., Okano M., Miyamoto M., Tagahara K., Lee K. H., *J. Nat. Prod.*, **55**, 468—475 (1992); *b*) Ohnishi S., Fukamiya N., Okano M., Tagahara K., Lee K. H., *ibid.*, **58**, 1032—1038 (1995).