

Design, synthesis, and biological evaluation of a biyouyanagin compound library

K. C. Nicolaou^{a,b,c,1}, Silvano Sanchini^{a,b}, David Sarlah^{a,b}, Gang Lu^{a,b}, T. Robert Wu^{a,b}, Daniel K. Nomura^{b,d}, Benjamin F. Cravatt^{b,d}, Beatrice Cubitt^e, Juan C. de la Torre^e, Ann J. Hessel^e, and Dennis R. Burton^{e,f,g}

^aDepartment of Chemistry, ^bSkaggs Institute for Chemical Biology, ^cDepartment of Chemical Physiology, ^dDepartment of Immunology and Microbial Science, and ¹International AIDS Vaccine Initiative Neutralising Antibody Center, The Scripps Research Institute, 10550 North Torrey Pines Road, La Jolla, CA 92037; ^eRagon Institute of Massachusetts General Hospital, Massachusetts Institute of Technology and Harvard, Boston, MA 02114; and ^fDepartment of Chemistry and Biochemistry, University of California, 9500 Gilman Drive, La Jolla, CA 92093

Edited by Stuart L. Schreiber, Broad Institute, Cambridge, MA, and approved December 10, 2010 (received for review October 26, 2010)

Modern drug discovery efforts rely, to a large extent, on lead compounds from two classes of small organic molecules; namely, natural products (i.e., secondary metabolites) and designed compounds (i.e., synthetic molecules). In this article, we demonstrate how these two domains of lead compounds can be merged through total synthesis and molecular design of analogs patterned after the targeted natural products, whose promising biological properties provide the motivation. Specifically, the present study targeted the naturally occurring biyouyanagins A and B and their analogs through modular chemical synthesis and led to the discovery of small organic molecules possessing anti-HIV and anti-arenavirus properties.

[2 + 2] photocycloaddition | anti-inflammatory agents | antiviral agents

Nature's medicine cabinet has been steadily expanding and continues to be enriched as chemists discover natural products with important biological properties that modulate the function of disease-related biomolecules. Such natural products have led to an impressive array of medications, either directly or indirectly, by serving as leads for structural modification and optimization (1–4). Indeed, it is estimated that the majority of the currently used drugs are derived through one of these two closely related approaches to drug discovery and development. An equally successful approach to drug discovery is the utilization of compound libraries of small synthetic molecules whose biological screening often uncovers lead compounds that ultimately become clinically approved medications by appropriate optimization achieved through molecular design, chemical synthesis, and biological evaluation. In recent years, the surge for biologics has been gathering momentum (5). These drugs are primarily proteins (e.g., antibodies), and often have a special place in the human pharmacopoeia (6, 7). Despite their niche, however, biologics are currently expensive and require intravenous administration. Their sustainability as drugs for a universal healthcare system is, therefore, questionable (8–10).

For these reasons, the search for small organic molecules (natural or designed) as ligands and lead compounds for drug discovery continues to be highly attractive. These organic molecules also serve as powerful tools in chemical biology, probing biological pathways and physiological effects (11–15). The studies described herein were undertaken as part of our ongoing research in the area of total synthesis of natural products and their analogs for biological evaluation (16–20). Our inspiration in this instance came from the forest in the form of biyouyanagin A, a natural product whose significance was reflected in its novel molecular architecture and important biological properties.

Plants of the *Hypericum* genus (Clusiaceae) have been exploited for a long time as traditional medicines, with *Hypericum perforatum* (St. John's wort) perhaps being the most well-known as a remedy for mild depression (21). Recent investigations of *Hypericum chinense* (biyouyanagi in Japanese) led to the discovery of several bioactive compounds, including biyouyanagins A (22) and B (23), whose structures were originally assigned as 1' and 2' (Fig. 1), respectively. Biyouyanagin A was reported to

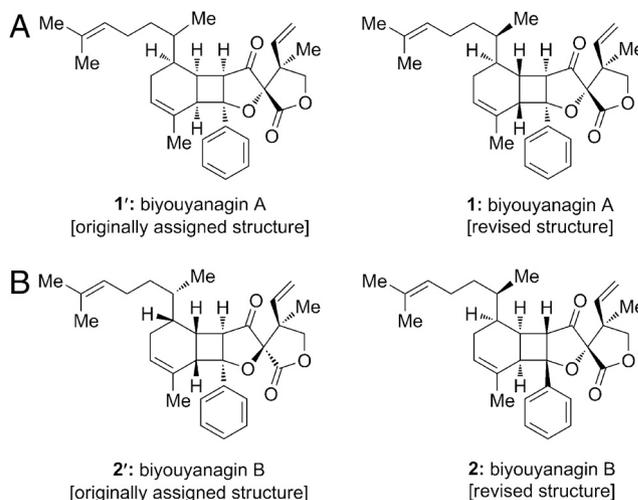


Fig. 1. Originally assigned (1' and 2') and revised (1 and 2) structures of biyouyanagins A and B.

possess selective inhibitory activity against HIV replication in H9 lymphocytes ($EC_{50} = 0.798 \mu\text{g mL}^{-1}$ vs. $EC_{50} > 25 \mu\text{g mL}^{-1}$ against noninfected lymphocytes), demonstrating a good therapeutic index (TI > 31.3) (22). This compound also exhibited potent inhibition of lipopolisaccharide-induced cytokine production at $10 \mu\text{g mL}^{-1}$ [IL-10 = 0.03; IL-12 = 0.02; tumor necrosis factor- α (TNF α) = 0.48] (22).

Inspired by the novel molecular architectures and important biological properties of biyouyanagin A, we initiated a program directed toward its total synthesis (24, 25). As it turned out, the total synthesis of biyouyanagin A led to its structural revision from 1' to 1 (see Fig. 1A) (24, 25). Interestingly, our total synthesis (26) of the subsequently reported biyouyanagin B (23) also led to its structural revision from the originally assigned structure 2' to 2 (see Fig. 1B).

The key step of the synthetic strategy employed for the total synthesis of biyouyanagins A (1) and B (2) involved a [2 + 2] photocycloaddition reaction as shown in Scheme 1 (24–26). Intriguingly, this process also delivered biyouyanagin C (3, a compound not discovered in nature as yet). It is interesting to

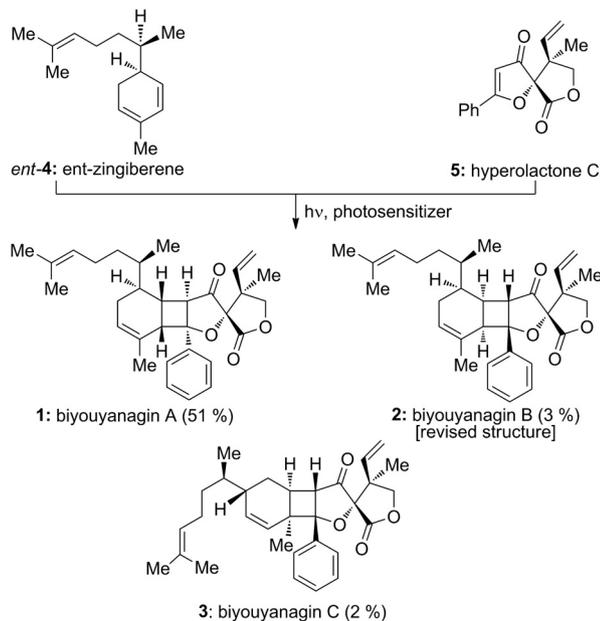
Author contributions: K.C.N., S.S., D.S., B.F.C., J.C.d.I.T., and D.R.B. designed research; S.S., D.S., G.L., T.R.W., D.K.N., B.C., and A.J.H. performed research; K.C.N., S.S., D.S., G.L., T.R.W., D.K.N., B.F.C., B.C., J.C.d.I.T., and A.J.H. analyzed data; S.S., D.S., G.L., T.R.W., D.K.N., and B.C. contributed new reagents/analytic tools; and K.C.N., D.S., B.F.C., J.C.d.I.T., and A.J.H. wrote the paper.

The authors declare no conflict of interest.

This article is a PNAS Direct Submission.

¹To whom correspondence should be addressed. E-mail: kcn@scripps.edu.

This article contains supporting information online at www.pnas.org/lookup/suppl/doi:10.1073/pnas.1015258108/-DCSupplemental.



Scheme 1. Synthesis of biyouyanagins A (1), B (2), and C (3) through [2 + 2] photocycloaddition.

note that hyperlactone C (5) is naturally occurring within the *H. chinense* plant (23, 27). Motivated by the potentially useful biological activities reported for biyouyanagin A and our ability to synthesize the molecular framework of this complex structure, we proceeded to explore the molecular space around the biyouyanagin molecule in search of simpler structures with enhanced biological activities. In this article, we describe our results, including preliminary biological data, on some members of the synthesized compound library.

Results and Discussion

Molecular Design. The design of our compound library was guided by the modular nature of the biyouyanagin structure and the synthetic approach to biyouyanagins A, B, and C as outlined in Scheme 1. Thus, using biyouyanagin A (1) as a lead compound, this modularity led to the design of general structure I (Fig. 2) as the formula representing the targeted focused library. Retrosynthetic disconnection of I through a [2 + 2] photocycloaddition led to olefin module building block II and enone hyperlactone module building block III. Further disconnection of the hyperlactone module III through a palladium-catalyzed cascade reaction revealed propargylic alcohols IV and aryl iodides V as the required fragments (plus carbon monoxide). This analysis defined a three-domain general structure for the library (i.e., I), and inspired a ready access to its members from the three relatively simple fragments II, IV, and V through a practical and robust synthetic route (24–26).

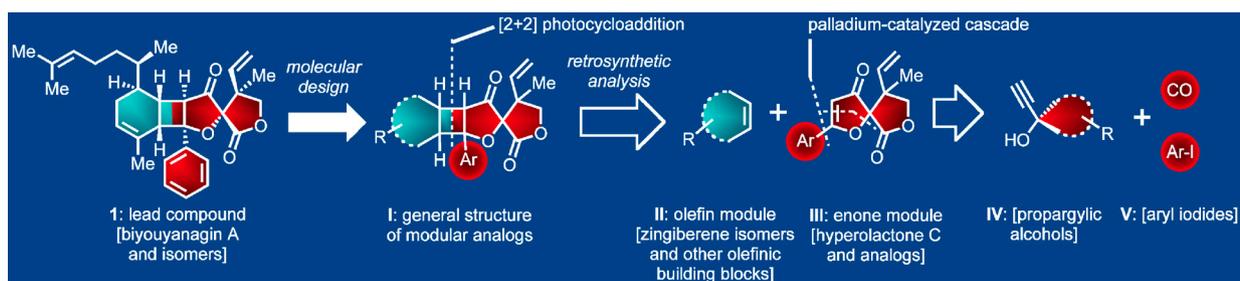


Fig. 2. Modular compound library (I) design and its retrosynthetic analysis.

Chemical Synthesis. The construction of the designed biyouyanagin library was based on our previously streamlined route (24–26) to this structural motif and followed two branches as outlined in Fig. 3. Thus, following along branch a, the requisite hyperlactone C and its stereoisomers (III, Fig. 3; see also square box, Fig. 4, 5, 4-*epi*-5, *ent*-5, 3-*epi*-5) were obtained through a palladium-catalyzed cascade reaction that combined acetylenic alcohols (IV) with aryl iodides (V) and carbon monoxide (Fig. 3). These substrates were subsequently reacted with the four synthetic stereoisomeric zingiberenes (Fig. 3; see also rectangular box, Fig. 4, *ent*-4, 4, *ent*-7-*epi*-4, 7-*epi*-4) in all possible combinations ($4 \times 4 = 16$) under photoirradiation conditions to afford the various biyouyanagins (Fig. 4, 1–3, 6–22). In most of the cases, only one major biyouyanagin isomer was obtained, although in some instances two or even three isomeric products were isolated. In one case, an analog was obtained by spontaneous postphotocycloaddition ring closure (e.g., 46, Fig. 5). Branch b started from similar building blocks (IV, V, and CO, Fig. 3) to form hyperlactone C analogs (VIII, Fig. 3; see also Fig. 5, 23–27) beyond those employed in branch a. It then utilized an array of olefinic building blocks (Fig. 5, *ent*-4, 54–64), other than those used in branch a, as partners in the photocycloaddition step to generate a series of analogs (IX, Fig. 3; see also Fig. 5, 28–45), some of which were elaborated further to produce additional members (X, Fig. 3) of the library (i.e., 52, 53, Fig. 5).

In addition to the biyouyanagin compound library depicted in Figs. 4 and 5, this study also produced a hyperlactone compound library [see structures 5, 4-*epi*-5, *ent*-5, 3-*epi*-5 (Fig. 4), and 23–27, 47–51 (Fig. 5); hyperlactones outside the boxes (i.e., 47–51, Fig. 5) were not utilized in the [2 + 2] photocycloaddition reaction to produce biyouyanagin type compounds] (experimental procedures and selected physical data for all compounds described herein is found in *SI Appendix*). Members of these compound libraries were subjected to biological screening in a variety of antiviral and cytokine production assays as described below.

Biological Screening in an Arenavirus Assay. A number of arenaviruses cause hemorrhagic fever (HF) disease in humans associated with high morbidity and significant mortality (28). Thus, Lassa (LASV) and Junin (JUNV) viruses, the causative agents of Lassa and Argentine HF, respectively, have devastating consequences on public health within their respective endemic regions of West Africa (LASV) and Argentina (JUNV). In addition, evidence indicates that the globally distributed lymphocytic choriomeningitis virus (LCMV) is a neglected human pathogen of clinical significance, especially to immunosuppressed individuals (29, 30). Besides the public health risk, arenaviruses pose a potential biodefense threat, and six of them, including LASV, JUNV, and LCMV, are listed as Category A agents (31). Concerns about arenavirus infections are aggravated by the lack of licensed vaccines; furthermore, current anti-arenavirus therapy is limited to the use of the nucleoside analog ribavirin, which is only partially effective, requires early and intravenous administration for optimal efficacy, and is often associated with significant side effects. The significance of arenaviruses in human

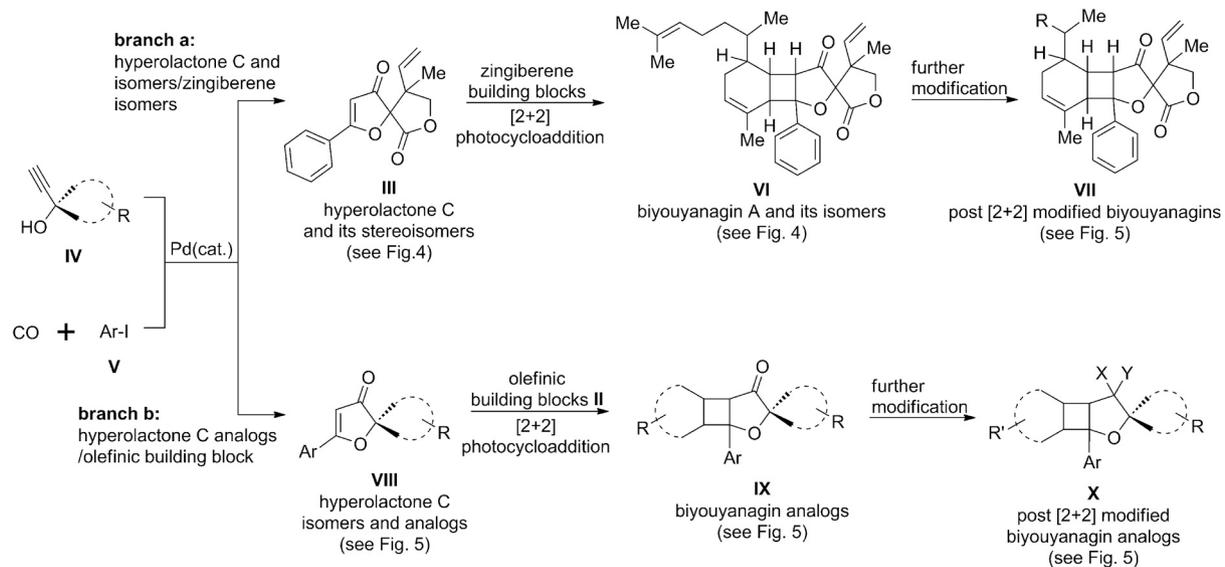


Fig. 3. General strategy for the construction of hyperlactone C and biyouyanagin libraries (see Figs. 4 and 5).

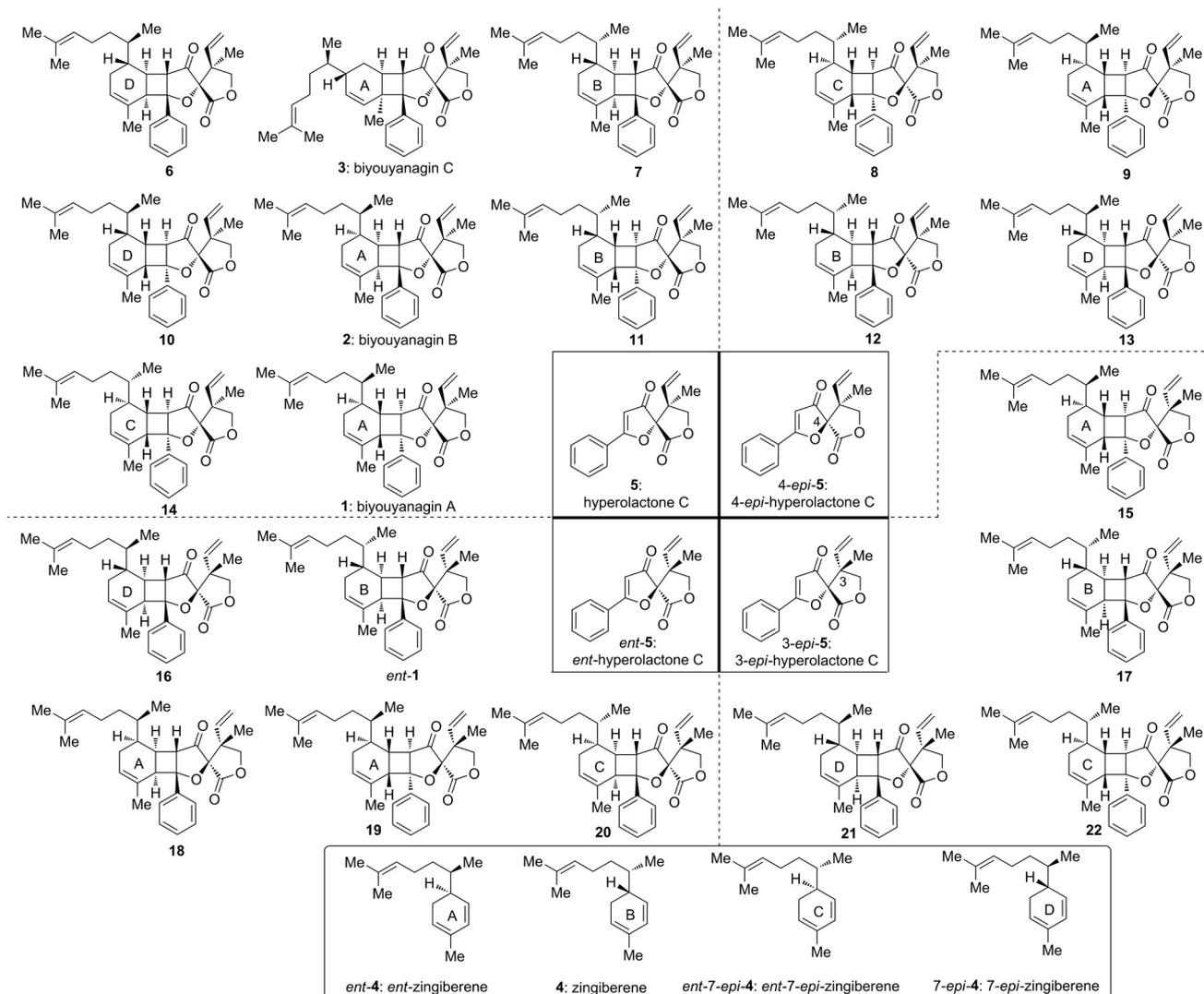


Fig. 4. Biyouyanagin library obtained from isomeric hyperlactone and zingerberene building blocks through [2+2] photocycloaddition (see Fig. 3, branch a).

levels of GFP expression (Fig. 6). Thus, two biyouyanagins [i.e., **1** and **20**, Fig. 4] and two biyouyanagin analogs (i.e., **35** and **33**, Fig. 5) displayed significant activity against LCMV in the middle micromolar range. Interestingly, in contrast to their anti-HIV properties, hyperolactone **C** (**5**, Fig. 4) and its analogs displayed no or only weak activity against the LCMV. For the most active compounds, maximal anti-arenaviral (greater than 99% inhibition of GFP expression) activity was observed at 50 μM , a concentration at which these compounds did not cause noticeable BHK-21 cell toxicity. In addition, none of the active compounds exerted virucidal activity or inhibition of virus binding to cells; rather, they inhibited virus replication and gene expression through a mechanism that remains to be determined.

Biological Evaluation in an HIV Assay. In view of the previous discovery of significant inhibitory activities for a number of biyouyanagin **A** and hyperolactone **C** analogs against HIV-1 replication in MT-2 lymphocytes (22, 25), we subjected members of the compound library to a neutralization assay using the molecular clone HIV-1_{HXB2}. From all the biyouyanagin isomers tested, the naturally occurring biyouyanagin **B** (**2**) and the newly synthesized biyouyanagin **C** (**3**) were found to be the most active in this assay [2: $\text{IC}_{50} = 42.90 \mu\text{M}$; 3: $\text{IC}_{50} = 83.97 \mu\text{M}$, as compared to biyouyanagin **A** (**1**), which exhibited $\text{IC}_{50} = 123.4 \mu\text{M}$]. The most active compound of the entire library of biyouyanagins and hyperolactones, however, was the postphotocycloaddition modified biyouyanagin analog **53**, which exhibited an IC_{50} value of 7.00 μM in the same assay. Fig. 7 graphically displays the results of this assay for compounds **1–3** and **53**. As a lead compound, **53** (IC_{50} 7.0 μM) compares favorably with AZT (IC_{50} 0.056 μM) in terms of potency (see Fig. 7).

LPS-Induced Cytokine Inhibition Assays. Although the body's inflammatory response is elicited as a defense mechanism against various noxious stimuli such as infection or tissue injury, chronic or uncontrolled inflammation can lead to a wide range of pathologies, including sepsis, cancer, arthritis, neurodegenerative dis-

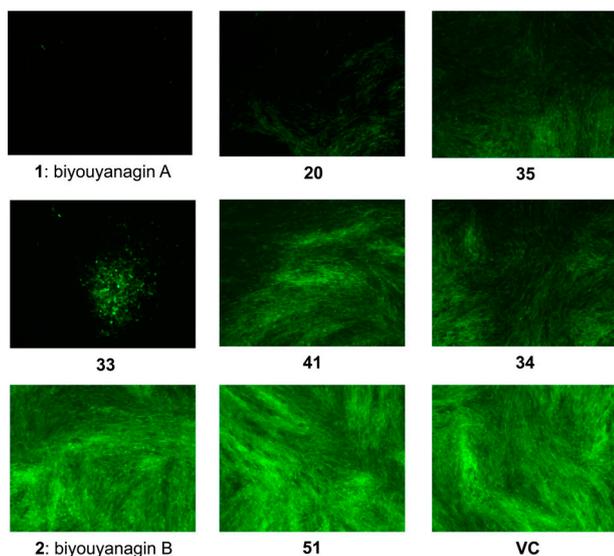


Fig. 6. BHK-21 cells were infected with a recombinant LCMV virus expressing GFP, called r3LCMV-GFP at a multiplicity of infection of 0.1 in the presence of the indicated compound (50 μM). At 36 h postinfection, cells were fixed in 4% paraformaldehyde/PBS, and viral load was assessed based on GFP expression. Compound-associated cell (BHK-21) toxicity was determined by trypan blue. Compounds exhibiting cell toxicity of $\geq 30\%$ (at 50 μM) were not evaluated for their antiviral activity. The figure shows examples of compounds with high (**1** and **20**), medium (**35** and **33**), low (**41** and **34**), or negligible (**2** and **51**) anti-LCMV activity. VC, treatment with vehicle (DMSO) (for further examples of compounds tested, see Table S1).

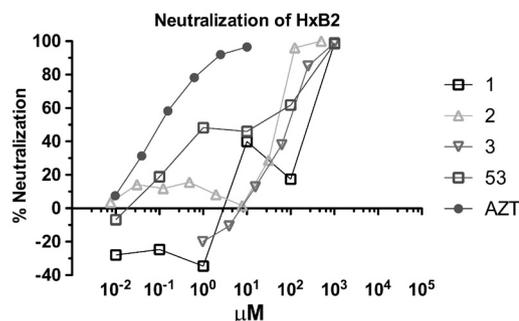


Fig. 7. Neutralization assay using the molecular clone HIV-1_{HXB2}. Pseudoviruses were generated in 293T cells, and neutralization with single-round infectious pseudovirus was performed using TZM-bl cells as targets for infection. AZT (Azidothymidine) was used as a control.

ease, obesity, diabetes, and atherosclerosis (33–36). Despite the availability of many effective anti-inflammatory therapies, including nonsteroidal anti-inflammatory drugs, glucocorticoids, and anti-cytokine agents, several of these medications have significant side effects that discourage their chronic use (37). Therefore, the identification of novel anti-inflammatory agents is crucial for devising new therapies for inflammatory diseases. Here, we show that several members of the biyouyanagin library (e.g., **34–36**, **46** and **53**, Fig. 5) significantly reduce the levels of LPS-stimulated inflammatory IL6 release in the human macrophage cell line THP-1 (Fig. 8A). Furthermore, we show that several of these bioactive compounds (e.g., **34** and **53**, Fig. 5) also elicit selective effects on the production of other anti-inflammatory cytokines, such as IL1 β and TNF α (Fig. 8B), with no effect on IL1 α or IL8 production (see Fig. S1). Compound **53** had the most potent effect across multiple inflammatory LPS-induced cytokines (i.e., IL6, IL1 β , and TNF α), with 90–96% inhibition

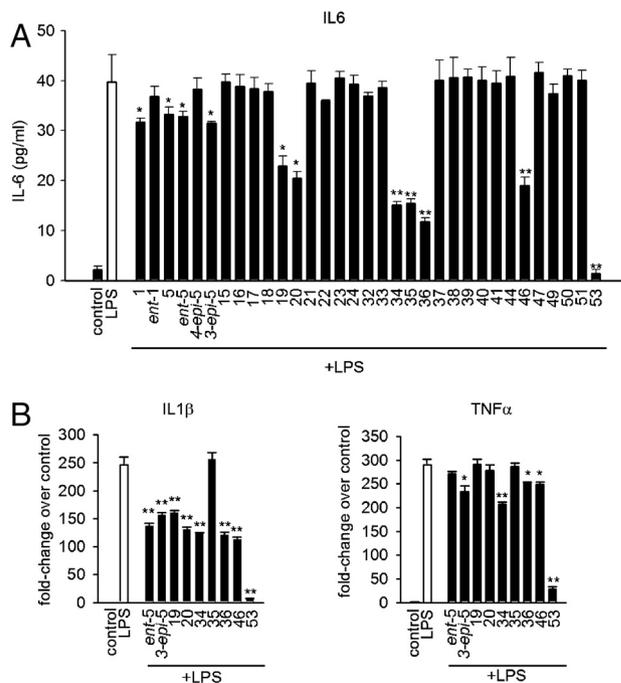


Fig. 8. Anti-inflammatory screen of 33 members of the biyouyanagin library. (A) Human macrophage THP-1 cells were pretreated with each compound (10 μM) for 1 h before stimulating the cells with LPS (2 $\mu\text{g}/\text{mL}$) for 6 h. (B) Several of the compounds that inhibited IL6 release in this screen were further assessed for inhibition of IL1 β and TNF α . Control denotes no compound, and LPS denotes LPS-treatment alone without any compound. IL6 release was assessed. Data are with $n = 3–5/\text{group}$. Significance is expressed as $*p < 0.05$, $**p < 0.01$ compared to LPS alone.

of IL6, IL1 β , and TNF α at 10 μ M. Compounds *ent-5*, *3-epi-5*, **19**, **20**, and **36** also exhibited significant inhibitory activity against LPS-induced cytokine IL1 β production, as shown in Fig. 8B.

Conclusion

Inspired by nature, a series of biyouyanagin-like molecules were designed and synthesized through a modular strategy whose key assembling processes were a palladium-catalyzed cascade sequence and a [2 + 2] photocycloaddition reaction. The synthesized compound libraries were subjected to biological screening, aiming to detect lead compounds for antiviral (i.e., anti-arenavirus and anti-HIV properties) and anti-inflammatory (i.e., LPS-induced cytokine production inhibitory properties) agents. Indeed, these investigations led to the discovery of a number of such compounds. Thus, biyouyanagin A (**1**), biyouyanagin **20**, and biyouyanagin analogs **35** and **33** exhibited significant activity against arenavirus LCMV, apparently through an as yet unknown mechanism of action. Biyouyanagins B (**2**, naturally occurring) and C (**3**, synthetic, not found in nature as yet) showed higher potencies than biyouyanagin A (**1**) in the HIV neutralization assay, and the most potent compound in the series was the newly synthesized biyouyanagin analog **53**, exhibiting IC₅₀ = 7.00 μ M. Several compounds possessing selective inhibitory activity against LPS-induced cytokine production were also identified, including *ent-5*, *3-epi-5*, **19**, **20**, **34–36**, **46**, and **53**. The most potent compound in the cytokine assay proved to be compound **53**, whose inhibitory activity against LPS-induced production of IL6, IL1 β , and TNF α was consistently high, and it showed essentially no effect in the LPS-induced production of IL1 α and IL8. In view of these results, we project that some of these compounds may act as useful probes in biological investigations and serve as path-pointing leads in drug discovery efforts toward antiviral and

anti-inflammatory agents. The higher potency of compound **53** in both the HIV neutralization and cytokine assays is intriguing, especially in light of its biyouyanagin–nucleic acid base hybrid structure. Indeed, the impressive activity of **53**, as compared to the other tested analogs, begs the question as to whether its properties are primarily derived from its dichloronucleobase or its biyouyanagin-like domain or both. Further studies along this line are clearly warranted and should provide answers to this question as well as further optimization of the biological profile of this compound. The success of these studies in identifying compounds with enhanced biological properties underscores the continuing importance of natural products as starting points for chemical biology and drug discovery efforts through rational molecular design and chemical synthesis.

Materials and Methods

The experimental procedures and physical data of the compounds used in this study can be found in the *SI Appendix*, which includes the following sections: *I. Experimental Procedures and Spectroscopic Data for Compounds*, *II. Biological Screening in an Arenavirus Assay*, *III. Biological Screening in an HIV Assay*, and *IV. Assessing Anti-inflammatory Activity of the Biyouyanagin Library*.

ACKNOWLEDGMENTS. We thank Dr. Dee-Hua Huang and Dr. Laura Pasternack for NMR spectroscopic assistance; Dr. Gary Siuzdak and Dr. Raj Chadha for mass spectrometric and X-ray crystallographic assistance, respectively; and Eric Rogers for performing the neutralization assays. Financial support for this work was provided by the National Institutes of Health (USA), The Skaggs Institute for Chemical Biology, the Università degli Studi di Urbino "Carlo Bo" (graduate fellowship to S.S.), the Japan Society for the Promotion of Science (postdoctoral fellowship to G.L.), the Natural Sciences and Engineering Research Council of Canada (postdoctoral fellowship to T.R.W.), and the National Institute on Drug Abuse (award K99DA030908 to D.K.N.).

- Newman DJ, Cragg GM (2007) Natural products as sources of new drugs over the last 25 years. *J Nat Prod* 70:461–477.
- Cragg GM, Newman DJ, Snader KM (1997) Natural products in drug discovery and development. *J Nat Prod* 60:52–60.
- Nicolau KC, Chen JS, Dalby SM (2009) From nature to the laboratory and into the clinic. *Bioorg Med Chem* 17:2290–2303.
- Nicolau KC, Montagnon T (2008) *Molecules that Changed the World* (Wiley-VCH, Weinheim, Germany) p 366.
- Carter PJ (2006) Potent antibody therapeutics by design. *Nat Rev Immunol* 6:343–357.
- Reichert JM, Rosensweig CJ, Faden LB, Dewitz MC (2005) Monoclonal antibody successes in the clinic. *Nat Biotechnol* 23:1073–1078.
- Waldmann TA (2003) Immunotherapy: Past, present and future. *Nat Med* 9:269–277.
- Scott CT (2005) The problem with potency. *Nat Biotechnol* 23:1037–1039.
- Carter P (2001) Improving the efficacy of antibody-based cancer therapies. *Nat Rev Cancer* 1:118–129.
- Glennie MJ, Johnson PW (2000) Clinical trials of antibody therapy. *Immunol Today* 21:403–410.
- Hinterding K, Alonso-Diaz D, Waldmann H (1998) Organic synthesis and biological signal transduction. *Angew Chem Int Edit* 37:688–749.
- Hung DT, Jamison TF, Schreiber SL (1996) Understanding and controlling the cell cycle with natural products. *Chem Biol* 3:623–639.
- Burke MD, Schreiber SL (2003) A planning strategy for diversity-oriented synthesis. *Angew Chem Int Edit* 43:46–58.
- Schreiber SL (2000) Target-oriented and diversity-oriented organic synthesis in drug discovery. *Science* 287:1964–1969.
- Burke MD, Berger EM, Schreiber SL (2003) Generating diverse skeletons of small molecules combinatorially. *Science* 302:613–618.
- Nicolau KC, et al. (2000) Natural product-like combinatorial libraries based on privileged structures. 1. General principles and solid-phase synthesis of benzopyrans. *J Am Chem Soc* 122:9939–9953.
- Nicolau KC, et al. (2009) Total synthesis and biological evaluation of cortistatins A and J and analogues thereof. *J Am Chem Soc* 131:10587–10597.
- Nicolau KC, Roschangar F, Vourloumis D (1998) Chemical biology of epothilones. *Angew Chem Int Edit* 37:2014–2045.
- Nicolau KC (2005) Endeavors in chemical biology and medicinal chemistry. *J Med Chem* 48:5613–5638.
- Cee VJ, Chen DY-K, Lee MR, Nicolau KC (2009) Cortistatin A is a high-affinity ligand of protein kinases ROCK, CDK8, and CDK11. *Angew Chem Int Edit* 47:8952–8957.
- Schulz V (2002) Clinical trials with *Hypericum* extracts in patients with depression—Results, comparisons, conclusions for therapy with antidepressant drugs. *Phytotherapy* 9:468–474.
- Tanaka N, et al. (2005) Biyouyanagin A, an anti-HIV agent from *Hypericum chinense* L. var. *salicifolium*. *Org Lett* 7:2997–2999.
- Tanaka N, et al. (2009) Acylphloroglucinol, biyouyanagiol, biyouyanagin B, and related spiro-lactones from *Hypericum chinense*. *J Nat Prod* 72:1447–1452.
- Nicolau KC, Sarlah D, Shaw DM (2007) Total synthesis and revised structure of biyouyanagin A. *Angew Chem Int Edit* 46:4708–4711.
- Nicolau KC, et al. (2008) Total synthesis, revised structure, and biological evaluation of biyouyanagin A and analogues thereof. *J Am Chem Soc* 130:11114–11121.
- Nicolau KC, Sanchini S, Wu TR, Sarlah D (2010) Total synthesis and structural revision of biyouyanagin B. *Chem Eur J* 16:7678–7682.
- Aramaki Y, Chiba K, Tada M (1995) Spiro-lactones, hyperolactone A–D from *Hypericum chinense*. *Phytochemistry* 38:1419–1421.
- Buchmeier M, Peters CJ, de la Torre JC (2007) *Fields Virology*, ed DM Knipe (Lippincott Williams & Wilkins, Philadelphia), 5th Ed., pp 1792–1827.
- Barton LL, Mets MB, Beauchamp CL (2002) Lymphocytic choriomeningitis virus: Emerging fetal teratogen. *Am J Obstet Gynecol* 187:1715–1716.
- Peters C (2006) Lymphocytic choriomeningitis virus—An old enemy up to new tricks. *New Engl J Med* 345:2208–2211.
- Borio L, et al. (2002) Hemorrhagic fever viruses as biological weapons: Medical and public health management. *J Am Med Assoc* 287:2391–2405.
- Emonet SF, Garidou L, McGavern DB, de la Torre JC (2009) Generation of recombinant lymphocytic choriomeningitis viruses with trisegmented genomes stably expressing two additional genes of interest. *Proc Natl Acad Sci USA* 106:3473–3478.
- Glass CK, Saijo K, Winner B, Marchetto MC, Gage FH (2010) Mechanisms underlying inflammation in neurodegeneration. *Cell* 140:918–934.
- Medzhitov R (2008) Origin and physiological roles of inflammation. *Nature* 454:428–435.
- Grievnikov SI, Greten FR, Karin M (2010) Immunity, inflammation, and cancer. *Cell* 140:883–899.
- Hotamisligil GS (2010) Endoplasmic reticulum stress and the inflammatory basis of metabolic disease. *Cell* 140:900–917.
- Dinarello CA (2010) Anti-inflammatory agents: Present and future. *Cell* 140:935–950.