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A Condensed, Scalable Synthesis of Racemic Koningic Acid

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KEYWORDS Koningic acid, heptelidic acid, glycolysis inhibitor, total synthesis

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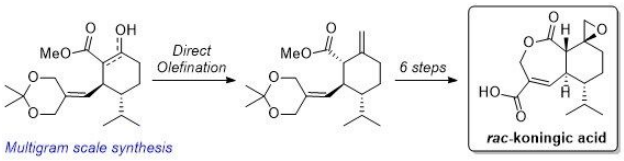
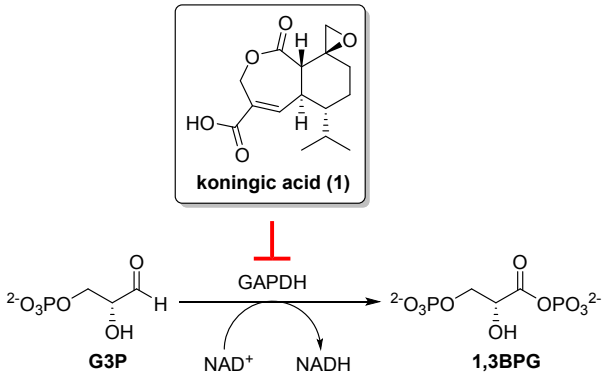


The natural product koningic acid (KA) is a selective covalent inhibitor of glyceraldehyde-3-phosphate dehydrogenase (GAPDH), a critical node in the glycolysis pathway. While KA is available commercially, sources are limited and its cost becomes rapidly prohibitive beyond the milligram scale. Additionally, a practical and flexible synthetic route to KA and analogs remains to be developed. Here we detail a new route that is operationally safer, scalable and offers a five step reduction in the previously reported longest linear sequence.

Glycolysis is a cornerstone metabolic pathway for cellular energy supply that involves the conversion of glucose into pyruvate, ATP and NADH. The pathway becomes critically important when cells are unable to produce energy by other means, such as in a hypoxic environment (*i.e.* anaerobic glycolysis) or when the energy demand surges (*e.g.* immune cell activation, rapid cell proliferation) under normoxic conditions (*i.e.* aerobic glycolysis).[1] The latter phenomenon, known as the Warburg effect, enables cell proliferation (*e.g.* cancer and immune cells) by generating energy and essential biosynthetic building blocks from glycolytic intermediates to permit growth and duplication of cellular components during division.[2]

As part of our effort to gain a high-resolution understanding of the role metabolism plays in immune cell function, we became interested in opportunities to pharmacologically interrogate glycolysis. One such opportunity is the natural product koningic acid (KA, **1**, **Figure 1**), a potent, selective and well-characterized covalent inhibitor of glyceraldehyde-3-phosphate dehydrogenase (GAPDH).[3] This enzyme catalyzes the conversion of glyceraldehyde-3-phosphate (G3P) to D-glycerate 1,3-bisphosphate (1,3BPG) (**Figure 1**) and resides at the interface between the ATP-consuming (-2 ATP) and the ATP-producing (+4 ATP) phases of glycolysis. Taken together, the critical role of GAPDH and the strength of KA as a chemical tool provided a unique opportunity to study the role of glycolysis in immune cells.[4]

Figure 1. GAPDH inhibitor koningic acid (**1**).



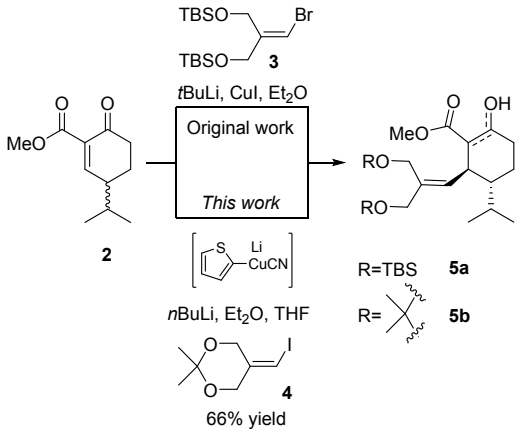
While KA is commercially available via microbial fermentation from *Trichoderma* fungus, sources are limited and the cost of multi-milligram quantities becomes rapidly prohibitive. These factors limit the ability to pharmacologically interrogate GAPDH as an important node of glycolysis as well as to access synthetically-derived KA analogs or tools (*e.g.* inactive controls, molecular probes, etc.). Given these limitations, we initiated an effort aimed at developing a synthesis that offered rapid, reliable access to useful quantities of KA and its analogs.

In 1988, Danishefsky reported the first total synthesis of racemic KA (*rac*-KA).[5] In 1997, Urban adapted Danishefsky's synthesis into a chiral auxiliary-based, asymmetric formal synthesis of the natural product.[6] To first access KA, we used Danishefsky's synthesis of *rac*-KA followed by chiral separation into its natural and non-natural enantiomers and were able to secure milligram quantities (data not shown). As we progressed through the synthesis, however, multiple opportunities were identified for improvement. Herein, we report a condensed and scalable synthesis of racemic KA.

A key step in Danishefsky's synthesis is a conjugate addition reaction that brings together 14 of the 15 carbons present in KA (**Scheme 1**). While the chemistry worked well in our hands, we sought improvements in several areas. First, we found that the Michael acceptor **2** could readily undergo an irreversible tautomerization to a corresponding dienol upon storage. Since the dienol

is completely unreactive under the conjugate addition conditions, we found it imperative to use the material right away in the next step. Second, the protected vinyl bromide **3** was produced in 56% yield over five steps and required storage at -20 °C to minimize spontaneous TBS deprotection, which was occasionally observed over time. As such, we saw an opportunity to reduce the number of steps while increasing the overall yield and practicality of the synthesis. Third, we were interested in finding an alternative to the use of *t*BuLi as a lithiation agent in order to improve the overall safety profile of the synthetic route upon scale up.

Scheme 1. Circumventing the use of *t*BuLi in the synthesis.



Inspired by the work of Urban,[6] we turned our attention to vinyl iodide **4**, which was found to offer multiple advantages. First, **4** was readily available on multi-gram scale in 3 steps from commercially available starting materials. Second, the acetonide protecting group was stable upon long-term storage (*vs* TBS, see above). Third, it was hypothesized that a more reactive vinyl iodide (*vs* bromide) could enable the substitution of *t*BuLi with a safer alternative. Gratifyingly, lithium-halogen exchange with *n*BuLi was successful and the subsequent transmetallation with copper enabled the desired 1,4-addition to enone ester **2** to afford **5b** as a keto/enol mixture. While having

a mixture is inconsequential to the next step (see below), it did complicate the purification procedure upon scale up. Indeed, when conducted on a relatively small scale (*e.g.* 1-2 g), purification by flash chromatography was uneventful and provided pure material **5b** in 66% yield. However, upon scaling the reaction to over 20 g of the Michael acceptor **2**, purification by flash chromatography was complicated by the ability of **5b** to tautomerize on silica gel. To circumvent this issue, crystallization conditions were developed for isolation of **5b**, albeit with a decreased 27% yield. Interestingly, the keto tautomer preferentially crystallized from the mixture and the isolated material contained only trace amounts of the enol form. Overall, the development of this conjugate addition procedure alleviated the scale limitations imposed by the original use of *t*BuLi as well as long term stability concerns for TBS-protected intermediates. Moreover, initial crystallization work provided a promising starting point for future efforts to streamline purification.

The next critical component of the synthesis of KA involves the introduction of the final carbon that will host the epoxide functionality. Originally, this was achieved via a 4-step sequence that featured nickel cross-coupling and allylsilane protodesilylation reactions (**Scheme 2**). We envisioned that, theoretically, a Wittig reaction could directly provide us with the desired exocyclic alkene **9**. Though we initially suspected that the acidity of the hydrogen atom residing between the two carbonyls might simply quench the Wittig reagent, a number of similar cases were previously reported in the literature.[7] Initial investigation into the Wittig olefination led to two key observations (**Table 1**). First, when preparing the Wittig reagent *in situ*, the conversion of the olefination reaction is strongly dependent on the nature of the cationic counterion (**Table 1**, entries 1-4).[8] Additionally, the nature of the reaction product (*i.e.* desired **9** vs. isomerized products **10**) is significantly impacted by the number of equivalents of the Wittig reagents (**Table 1**, entry 5).

Based on the latter observation, it was postulated that use of the free ylide would be an efficient way to prevent any excess basic species in the reaction. We were pleased to find that upon using the free ylide, the reaction conversion could be increased to 89% while maintaining the integrity of the resulting double bond (**Table 1**, entries 6-9). Interestingly, the results with the free ylide demonstrate that the cationic counterion, while impactful to the reaction outcome, is not necessary to achieve high conversion. Finally, it is noteworthy that the excess ylide and corresponding phosphine oxide byproduct could be removed by precipitation in heptane at 0 °C, thereby allowing for simpler subsequent purification by silica gel chromatography. Under the optimal conditions, olefin **9** could be obtained in 82% yield, including a minor diastereomer observed by NMR (12:1 ratio favoring **9**).

Scheme 2. Introduction of the final carbon atom of KA.

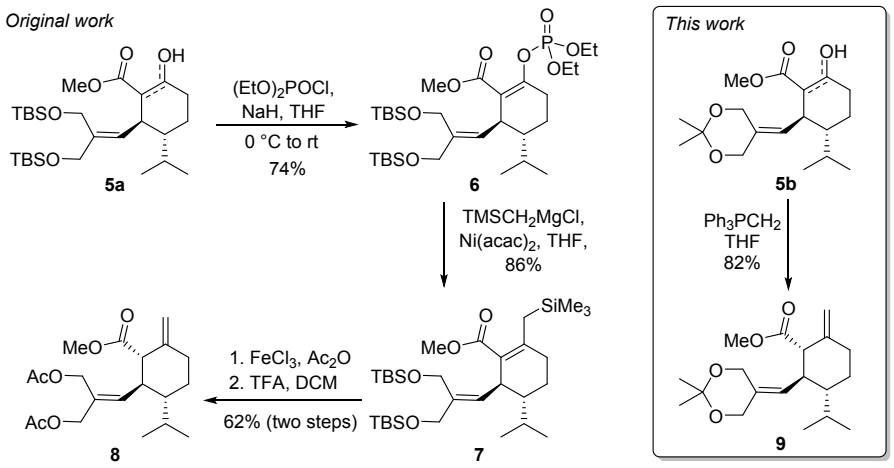
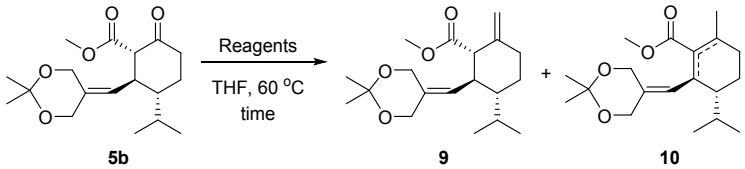


Table 1. Optimization of Wittig reaction conditions.

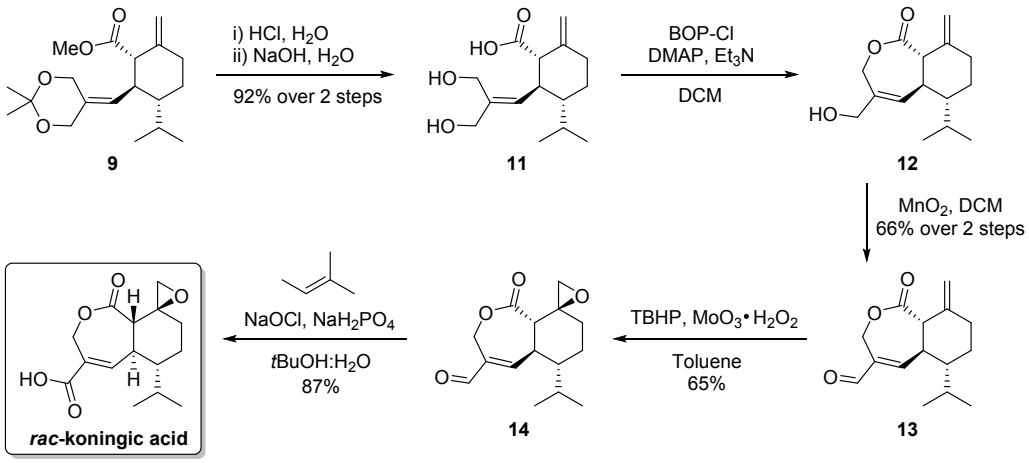


Entry	Base	PPh_3MeBr	PPh_3CH_2	Time	Conversion	Product

	(equivalents)	(equivalents)	(equivalents)			
1	LiHMDS (1.9)	2.0	-	24 h	0%	-
2	NaHMDS (1.9)	2.0	-	24 h	0%	-
3	KHMDS (1.9)	2.0	-	24 h	100%	**10**
4	KO*t*Bu (1.9)	2.0	-	24 h	100%	**10**
5	KO*t*Bu (1.05)	1.1	-	24 h	80%	**9**
6	-	-	1.0	20 h	75%	**9**
7	-	-	1.1	20 h	87%	**9**
8	-	-	1.2	20 h	89%	**9**
9	-	-	1.5	20 h	100%	**10**

With the exocyclic double bond in place, we proceeded to complete the total synthesis of *rac*-KA (**Scheme 3**). First, we developed a one-pot, two-step hydrolysis protocol that allowed for the synthesis of the diol acid **11** in 92% yield. Using the lactonization conditions previously reported by Danishefsky, followed by a simple extraction procedure and the removal of any remaining triethylammonium salts by precipitation with acetone, **12** was obtained in sufficient purity to be progressed to the next step. We then elected to use MnO_2[6] for the allylic oxidation. These reaction conditions provided the aldehyde **13** in 66% yield over two steps, results that were found to be more reproducible than when using Jones oxidation conditions.[5] Likewise, we also found that conducting the subsequent epoxidation reaction using $MoO_3.H_2O_2$[9] versus $Mo(CO)_6$[5] as a catalyst provided a more reproducible 65% yield of KA precursor **14.** Finally, generation of the carboxylic acid using Pinnick oxidation conditions[10] followed by crystallization afforded pure *rac*-KA in 87% yield.

Scheme 3. Completion of the synthesis of racemic KA



In summary, a shortened synthesis of racemic KA has been developed which builds on the foundational work of both the Danishefsky and Urban groups. This new route offers a 5-step reduction in the longest linear sequence (7.8% overall yield) and alleviates the scale limitations imposed by the original use of *t*BuLi for the key conjugate addition step. We believe the improved practicality of this new route will allow for an improved understanding of structure-activity relationships around koningic acid and will provide new avenues to interrogate the role of glycolysis in both immune and non-immune cell function.

Experimental Section

General Information

Unless otherwise noted, all commercial-grade reagents and substrates were ordered from Sigma Aldrich, TCI America, and Combi-blocks. These chemicals were used without further purification. Anhydrous solvents were used as received. Reactions were heated using an oil bath. Column chromatography was performed using Teledyne ISCO Combiflash Rf+ with Redisep Rf gold silica gel normal phase columns or Biotage SNAP C18 RP columns. Thin-layer

chromatography (TLC) was performed using glass plates pre-coated with 0.25 mm of silica gel 60 F254. ^{1}H NMR spectra were recorded on either a 300 or 400 MHz NMR spectrometer. Chemical shifts (δ values) for ^{1}H NMR spectra are reported in parts per million (ppm) downfield, and the coupling constants (*J* values) are in Hz. ^{13}C NMR spectra were recorded on a 100 MHz NMR spectrometer with complete proton decoupling. LCMS data were acquired on an Agilent single quadrupole LCMS series 6100.

*Methyl 3-isopropyl-6-oxocyclohex-1-ene-1-carboxylate (**2**)*

2 was prepared according to the previously described methods.[5]

*Synthesis of 5-(iodomethylene)-2,2-dimethyl-1,3-dioxane **(4)***

(iodomethyl)triphenylphosphonium iodide[11]

A solution of triphenylphosphine (146 g, 556 mmol) and diiodomethane (60.2 mL, 750 mmol) in toluene (487 mL) in a flask equipped with a reflux condenser was heated at 100 °C for 20 h under an argon atmosphere (protected from light by aluminum foil). The reaction mixture was cooled to 0 °C. The resulting pale white solid was filtered, washed with cold toluene and CH_2Cl_2 and then dried under vacuum to give (iodomethyl)triphenylphosphonium iodide (287 g, 542 mmol, 97.6% yield) as a pale white solid. LCMS m/z: $[M\text{-}I]^+$ Calcd for $C_{19}H_{17}IP$ 403.0; Found 403.0. ^{1}H NMR (DMSO-d_6, 400 MHz): δ_H 7.91-7.74 (15H, m), 5.03 (2H, d, J = 8.7 Hz).

2,2-dimethyl-1,3-dioxan-5-one[12]

Tris(hydroxylmethyl)aminomethane hydrochloride (100 g, 634 mmol) and 4-methylbenzene-1-sulfonic acid monohydrate (5.42 g, 28.5 mmol) were added to dry dimethylformamide (257 mL). 2,2-dimethoxypropane (85.2 mL, 697 mmol) was added and the mixture was stirred at rt for 60 h. Triethylamine (14.8 mL, 107 mmol) was added and the reaction mixture was concentrated under reduced pressure. The resulting crude gel was sonicated in MeOH (1 L). The solvent was removed to give a white solid mixture. The residue was suspended in ethyl acetate (860 mL) and triethylamine (143 mL, 1.03 mol) and the mixture was stirred at rt for 10 min. The precipitate was filtered off and the solvents were removed under reduced pressure to give crude (5-amino-2,2-dimethyl-1,3-dioxan-5-yl)methanol as a yellow oil (97.68 g, 545 mmol product (86.0% yield) calculated by ^{1}H NMR). No mass found by LCMS. ^{1}H NMR ($CDCl_3$, 400 MHz): δ_H 3.79 (2H, d, J = 11.8 Hz), 3.55 (2H, d, J = 11.7 Hz), 3.50 (2H, s), 2.46 (3H, br s), 1.44 (3H, s), 1.41 (3H, s).

A mixture of sodium periodate (151 g, 708 mmol) in water (1.22 L) was added over 90 min to the crude (5-amino-2,2-dimethyl-1,3-dioxan-5-yl)methanol (97.68 g, 545 mmol) in water (700 mL) and methanol (180 mL) at 0 °C. The mixture was stirred for 1.5 h at 0 °C. The white suspension was filtered off and the solution was extracted with CH_2Cl_2 (x7). The combined organic layers were washed with a sodium bicarbonate solution (5%; x2), dried over sodium sulfate and evaporated under reduced pressure (keeping the temperature below 20 °C). The crude was purified by vacuum distillation at 15 mbar using an oil bath at 110 °C, with fractions collected at 55-60 °C. Portion A (beginning), 8.2 g, colorless oil, contained 7.19 g pure product 2,2-dimethyl-1,3-dioxan-5-one, 12% w/w DMF; portion B, 38.95 g, colorless oil, contained 36.88 g pure product, 5% w/w DMF. Total amount of product 2,2-dimethyl-1,3-dioxan-5-one was 44.07 g (339 mmol, 53% yield

in two steps). No mass found by LCMS. R_f = 0.4 (20% EtOAc/hexane). ^{1}H NMR ($CDCl_3$, 400 MHz): δ_H 4.15 (4H, s), 1.45 (6H, s).

5-(iodomethylene)-2,2-dimethyl-1,3-dioxane ***(4)***

To a suspension of (iodomethyl)triphenylphosphonium iodide (164 g, 311 mmol) in tetrahydrofuran (725 mL) at -23 °C was slowly added sodium bis(trimethylsilyl)amide (311 mL, 311 mmol) (1M in THF). After stirring for 15 min, 2,2-dimethyl-1,3-dioxan-5-one (38.95 g, 283 mmol) was added. The reaction mixture was allowed to warm to rt and was stirred for 1 h. Water (100 mL) was added to the mixture, the suspension was filtered and the filtrate was extracted with MTBE (3 × 800 mL). The combined organic layers were dried over anhydrous sodium sulfate and concentrated under reduced pressure to obtain the crude product. 250 g silica gel and 300 mL DCM were added to the crude residue. The mixture was dried under reduced pressure to give a free-flowing powder. Filter paper was placed over a sand core funnel (dm ~ 13 cm), to which silica gel (~ 1 cm) was added. The silica gel was washed with hexanes under vacuum. The crude dry powder was then loaded in the funnel. A filter paper was put on the top of the powder. This solid was washed with hexanes (1500 mL) under vacuum. The filtrate was collected in 4 portions. Product was detected via TLC in the first three portions. Each portion was concentrated and ^{1}H NMR showed clean desired product. The three portions were combined to give **4** (56.9 g, 223 mmol, 79.1% yield) as a brown oil containing 2% mol/mol of triphenylphosphine oxide. HRMS (ESI) m/z: $[M+Na]^+$ Calcd for $C_7H_{11}IO_2Na$ 276.9701; Found 276.9686. R_f = 0.5 (20% EtOAc/hexane). IR (Nujol) ν_{max}: 2986, 2846, 1632, 1445, 1380, 1370, 1277, 1236, 1216, 1194, 1120, 1083, 1035, 1017, 828, 765, 731, 680, 644, 541, 516 cm^{-1}. ^{1}H NMR ($CDCl_3$, 400 MHz): δ_H 5.98 (1H, m), 4.27-

4.26 (2H, m), 4.22 (2H, s), 1.39 (6H, s). ^{13}C NMR ($CDCl_3$, 100 MHz): δ_C 145.0, 99.7, 71.8, 66.3, 64.3, 24.0 (x2).

*Methyl (2SR,3RS)-2-((2,2-dimethyl-1,3-dioxan-5-ylidene)methyl)-3-isopropyl-6-oxocyclohexane-1-carboxylate (**5b**)*[6]

A solution of **4** (1.96 g, 7.73 mmol) in diethyl ether (30 mL) was cooled to -78 °C. *n*-Butyllithium (3.7 mL, 9.26 mmol, 2.5 M solution in hexane) was added and the mixture was stirred for 45 minutes at -78 °C. Subsequently, this mixture at -78 °C was cannulated dropwise into a stirred solution of lithium 2-thienylcyanocuprate (31.3 mL, 7.84 mmol, 0.25 M solution in 15.3 mL tetrahydrofuran) at - 78 °C. After 1.5 h at this temperature, a solution of **2** (1.4 g, 7.13 mmol) in tetrahydrofuran (14.2 mL) was added and the mixture was stirred at -78 °C overnight. After completion, the mixture was quenched by adding 20% saturated ammonium chloride in 30% ammonium hydroxide (1:4 sat. NH_4Cl:NH_4OH) and the mixture was stirred at rt for 1 h. The mixture was then extracted with MTBE (x2) and the combined organic layers were dried over sodium sulfate, filtered, and concentrated under reduced pressure to give the crude product. The crude material was purified by flash chromatography on silica gel (MTBE/hexane = 0/1 to 3/7) to give **5b** (1.53 g, 7.13 mmol, 66% yield) as a yellow oil, which solidified upon storage. LCMS m/z: $[M+Na]^+$ Calcd for $C_{18}H_{28}O_5Na$ 347.2; Found 347.2. R_f = 0.25-0.45 (20% EtOAC/hexane). ^{1}H NMR ($CDCl_3$, 400 MHz): δ_H 4.91 (1H, d, J = 10.4 Hz), 4.43-4.30 (2H, m), 4.18 (2H, s), 3.72 (3H, s), 3.18 (1H, d, J = 11.7 Hz), 2.87 (1H, q, J = 10.7 Hz), 2.54-2.49 (1H, m), 2.38-2.30 (1H, m), 2.00-1.89 (2H, m), 1.55-1.51 (2H, m), 1.38 (6H, s), 0.95 (3H, d, J = 7.0 Hz), 0.75 (3H, d, J = 6.9 Hz).

Note: Alternatively, the crude mixture (starting from 22 g of **2**) can be filtered over short path silica-gel and eluted with ethyl acetate until all colored residue has come out. The crude material was again concentrated under reduced pressure, diluted with heptane (250 mL), stirred at -30 °C for 30 minutes and filtered. The solid was washed with cold heptane (- 78 °C) to give **5b** (9.84 g, 30.3 mmol, 27.1% yield) as a beige solid. Note: the keto form was major with only traces of the enol form present.

*methyl (1SR,2SR,3RS)-2-((2,2-dimethyl-1,3-dioxan-5-ylidene)methyl)-3-isopropyl-6-methylenecyclohexane-1-carboxylate (**9**)*

A mixture of **5b** (1.29 g, 3.97 mmol) and methylidenetriphenylphosphane[13] (1.09 g, 3.97 mmol) in tetrahydrofuran (26 mL) was heated at 60 °C for 24 h. After completion, the mixture was concentrated under reduced pressure to give the crude product. Purification by flash chromatography on silica gel (EtOAc/hexane = 0/1 to 15/85) gave **9** (1.05 g, 3.25 mmol, 82.0% yield) as a white solid. HRMS (ESI) m/z: $[M+Na]^+$ Calcd for $C_{19}H_{30}O_4Na$ 345.2042; Found 345.2032. R_f = 0.5 (20% EtOAc/hexane). IR (Nujol) ν_{max}: 2992, 2954, 2845, 1729, 1646, 1434, 1379, 1355, 1261, 1238, 1216, 1189, 1150, 1067, 1043, 1015, 969, 910, 830, 728, 651, 513 cm^{-1}. ^1H NMR ($CDCl_3$, 400 MHz): δ_H 4.83 (1H, d, *J* = 10.6 Hz), 4.77 (1H, s), 4.46-4.40 (2H, m), 4.34-4.27 (1H, m), 4.13 (2H, dd, *J* = 18.1, 14.1 Hz), 3.65 (3H, s), 2.82 (1H, d, *J* = 11.0 Hz), 2.44-2.35 (2H, m), 1.99-1.93 (1H, m), 1.83-1.70 (2H, m), 1.37-1.34 (6H, m), 1.22-1.07 (2H, m), 0.85 (3H, d, *J* = 6.9 Hz), 0.67 (3H, d, *J* = 6.9 Hz). Minor peaks corresponding to a diastereomer of **9** were also observed (12:1 ratio favoring **9**). ^{13}C NMR ($CDCl_3$, 100 MHz): δ_C 173.1, 145.0, 135.2, 124.7, 108.0, 99.1, 64.5, 60.2, 56.2, 51.5, 46.8, 42.7, 35.3, 28.1, 25.1, 24.4, 23.8, 21.6, 15.9. Minor peaks corresponding to a diastereomer of **9** were also observed.

Note: Unreacted starting material **5b** could be recovered (220 mg, 0.678 mmol, 17.0 %) from the column chromatography.

(1SR,2SR,3RS)-2-(3-hydroxy-2-(hydroxymethyl)prop-1-en-1-yl)-3-isopropyl-6-methylenecyclohexane-1-carboxylic acid ***(11)***[5]

3 M aqueous hydrochloric acid (5.2 mL, 15.5 mmol) was added to a solution of crude **9** (500 mg, 1.55 mmol) in methanol (1.0 mL) and the mixture was stirred at rt. The starting material oiled out at the beginning, then slowly precipitated over 30-45 minutes. The mixture was stirred until TLC indicated completion (1 h). The reaction mixture was cooled to 0 °C, then 2.5 M aqueous sodium hydroxide (20 mL, 50 mmol) was added dropwise and the reaction mixture was allowed to stir vigorously at rt. After overnight, solid material formed around the stir bar and was broken down by sonication. The mixture was vigorously stirred at rt over 2 more days. After completion (checked by LCMS), the mixture was acidified with 6 N aqueous HCl to pH = 3 at 0 °C, then the solid was filtered via Buchner funnel and washed with water (x3). The collected solid was triturated in *tert*-butyl methyl ether to give **11** (380 mg, 1.41 mmol, 92% yield) as a beige solid. LCMS m/z: $[M-H]^-$ Calcd for $C_{15}H_{24}O_4$ 267.2; Found 267.1. ^1H NMR ($CDCl_3$, 400 MHz): δ_H 5.25 (1H, d, J = 10.7 Hz), 4.83 (1H, s), 4.61 (1H, s), 4.26-4.06 (4H, m), 2.91 (1H, d, J = 10.9 Hz), 2.65 (1H, q, J = 10.7 Hz), 2.48-2.43 (1H, m), 2.12-2.03 (1H, m), 1.93-1.81 (2H, m), 1.37-1.31 (1H, m), 1.21-1.10 (1H, m), 0.91 (3H, d, J = 7.0 Hz), 0.73 (3H, d, J = 6.9 Hz).

(5aSR,6RS,9aSR)-4-(hydroxymethyl)-6-isopropyl-9-methylene-5a,6,7,8,9,9a-hexahydrobenzo[c]oxepin-1(3H)-one ***(12)***[5]

To a round bottom flask containing a solution of **11** (1.20 g, 4.60 mol, 1.00 equiv) in dichloromethane (140 mL) was added triethylamine (1.9 mL, 14.0 mol, 3.00 equiv) and dimethylaminopyridine (DMAP) (28.0 mg, 0.23 mmol, 0.05 equiv). The resulting solution was cooled to 0 °C and bis(2-oxooxazolidin-3-yl)phosphinic chloride (BOP-Cl) (1.20 g, 4.90 mol, 1.05 equiv) was added. The mixture was warmed to 25 °C and stirred for 16 h. After completion, the mixture was adjusted to pH = 2 with 0.5 M aqueous HCl solution and was extracted with dichloromethane (x2). The combined organic layers were washed with saturated aqueous Na_2CO_3 and brine, dried over $MgSO_4$, filtered and concentrated under reduced pressure. Acetone (100 mL) was added (crystalline solid precipitates), the solid was filtered and the filtrate was concentrated under vacuum to yield crude **12** as a green oil, which was used directly in the next step without purification. LCMS m/z: $[M+H]^+$ Calcd for $C_{15}H_{22}O_3$ 251.2; Found 251.1. R_f = 0.8 (100% EtOAc). ^{1}H NMR ($CDCl_3$, 400 MHz): δ_H 5.92 (1H, d, *J* = 3.8 Hz), 5.26 (1H, s), 4.97-4.93 (2H, m), 4.48 (1H, d, *J* = 14.5 Hz), 4.12-4.02 (2H, m), 3.65 (1H, d, *J* = 12.0 Hz), 2.42-2.37 (1H, m), 2.34-2.27 (1H, m), 2.07-1.98 (2H, m), 1.75-1.69 (1H, m), 1.41-1.34 (1H, m), 1.25-1.15 (2H, m), 0.88 (3H, d, *J* = 6.9 Hz), 0.72 (3H, d, *J* = 6.9 Hz).

*(5aSR,6RS,9aSR)-6-isopropyl-9-methylene-1-oxo-1,3,5a,6,7,8,9,9a-octahydrobenzo[c]oxepine-4-carbaldehyde **(13)***[5]

To a round bottom flask containing a solution of crude **12** (1.20 g, 4.60 mmol) in dichloromethane (33.0 mL) was added manganese dioxide (4.20 g, 46.0 mmol, 10 equiv). The resulting heterogenous mixture was heated to reflux for 16 h. The mixture was filtered on a short silica plug and eluted with EtOAc. The resulting yellow solution was concentrated under reduced pressure to give **13** as a pale yellow oil (0.750 g, 3.02 mmol, 66.0% yield over 2 steps). LCMS

m/z: $[M+H]^+$ Calcd for $C_{15}H_{20}O_3$ 249.1; Found 249.1. R_f = 0.2 (20% EtOAc/hexane). ^1H NMR ($CDCl_3$, 400 MHz): δ_H 9.42 (1H, d, *J* = 0.5 Hz), 6.95-6.94 (1H, m), 5.45-5.44 (1H, m), 5.14-5.08 (2H, m), 4.97 (1H, dt, *J* = 15.1, 2.1 Hz), 3.83 (1H, d, *J* = 12.0 Hz), 2.65 (1H, tt, *J* = 11.4, 2.8 Hz), 2.50 (1H, dt, *J* = 13.7, 4.3 Hz), 2.18-2.08 (2H, m), 1.87-1.80 (1H, m), 1.61-1.54 (1H, m), 1.36 (1H, qd, *J* = 12.3, 4.5 Hz), 1.00 (3H, d, *J* = 6.8 Hz), 0.89 (3H, d, *J* = 6.9 Hz).

(5aSR,6RS,9SR,9aSR)-6-isopropyl-1-oxo-1,5a,6,7,8,9a-hexahydro-3H-spiro[benzo[c]oxepine-9,2'-oxirane]-4-carbaldehyde ***(14)***[5]

To a reactor containing molybdenum trioxide[9] (52 mg, 0.36 mmol, 0.20 equiv), 30% aqueous hydrogen peroxide (74 µL, 0.72 mmol, 0.40 equiv) was added and the suspension was heated to 40 °C and stirred for 2 h. The mixture was concentrated under reduced pressure to yield a bright yellow solid. A solution of **13** (450 mg, 1.81 mmol, 1.0 equiv) in toluene (18.0 mL) was added, followed by *tert*-butyl hydroperoxide (5.5 M solution in decane, 0.99 mL, 5.43 mmol, 3.0 equiv). The mixture was heated to 80 °C and stirred for 5 h. The mixture was cooled to 25 °C and dimethyl sulfide (0.53 mL, 7.24 mmol, 4.0 equiv) was added. The mixture was stirred at 25 °C for 30 minutes. The suspension was filtered on a short silica plug and eluted with EtOAc. The resulting yellow solution was concentrated under reduced pressure to give **14** as a pale yellow solid (310 mg, 1.17 mmol, 65% yield). LCMS m/z: $[M+H]^+$ Calcd for $C_{15}H_{20}O_4$ 265.1; Found 265.2. R_f = 0.1 (20% EtOAc/hexane). ^1H NMR ($CDCl_3$, 400 MHz): δ_H 9.41 (1H, d, *J* = 0.4 Hz), 6.94-6.92 (1H, m), 5.12 (1H, d, *J* = 15.3 Hz), 4.91 (1H, dt, *J* = 15.3, 2.3 Hz), 3.82 (1H, dd, *J* = 5.1, 1.5 Hz), 3.55 (1H, d, *J* = 12.1 Hz), 2.77-2.70 (1H, m), 2.62 (1H, d, *J* = 5.1 Hz), 2.23-2.15 (1H, m), 1.98-1.91 (1H, m), 1.86-1.78 (1H, m), 1.67-1.59 (2H, m), 1.53-1.42 (1H, m), 1.03 (3H, d, *J* = 6.9 Hz), 0.94 (3H, d, *J* = 6.9 Hz).

rac-KA[5]

To a solution of **14** (310 mg, 1.17 mmol) in *tert*-butanol (11.7 mL) and 2-methyl-2-butene (9.00 mL) at 0 °C was added a solution of sodium chlorite (1.05 g, 11.7 mmol, 10.0 equiv) and sodium dihydrogen phosphate (1.39 g, 11.7 mmol, 10.0 equiv) in water (4.25 mL). The resulting solution was warmed to 25 °C and stirred for 2 h. After completion, sodium chloride (2.05 g, 35.1 mmol, 30.0 equiv) and acetic acid (1.00 mL) were added and the mixture was extracted with EtOAc (x4) and dried over $MgSO_4$. Heptane (20.0 mL) was gradually added during concentration under vacuum, until the solution turned cloudy. The mixture was heated to boiling (crystalline white solid precipitates). The suspension was filtered and the filter cake was washed with heptane and dried under high vacuum to give *rac*-koningic acid as a white powder (0.285 g, 1.01 mmol, 87.0% yield). LCMS m/z: $[M+H]^+$ Calcd for $C_{15}H_{20}O_5$ 281.1; Found 281.1. R_f = 0.8 (1% AcOH/Et_2O). ^1H NMR (CD_3OD, 400 MHz): δ_H 7.28 (1H, dd, *J* = 4.2, 1.9 Hz), 5.26 (1H, dt, *J* = 14.9, 2.0 Hz), 5.03 (1H, d, *J* = 14.9 Hz), 3.88 (1H, d, *J* = 12.1 Hz), 3.85 (1H, dd, *J* = 5.5, 1.6 Hz), 2.61 (1H, dddd, *J* = 12.1, 10.7, 4.2, 2.1 Hz), 2.54 (1H, d, *J* = 5.5 Hz), 2.14 (1H, pd, *J* = 6.9, 3.3 Hz), 1.96-1.90 (2H, m), 1.70-1.62 (1H, m), 1.52-1.39 (2H, m), 1.01 (3H, d, *J* = 6.9 Hz), 0.93 (3H, d, *J* = 6.9 Hz).

Supporting Information

Copies of ^1H NMR ((iodomethyl)triphenylphosphonium iodide, 2,2-dimethyl-1,3-dioxan-5-one, compounds **4**, **5b**, **9**, **11**, **12**, **13**, **14** and *rac*-koningic acid), ^{13}C NMR (compounds **4** and **9**), and IR (compounds **4** and **9**) spectra can be found in the supporting information.

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