# Synthesis of Extremely Simplified Compounds Possessing the Key Pharmacophore Units of Taxol, Phenylisoserine and Oxetane Moieties

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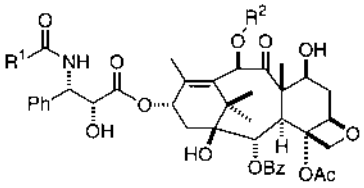
**Straight chain compounds having a phenylisoserine unit and an oxetane ring at the $\alpha$- and $\omega$- position, respectively as extremely simplified analogues of taxol were prepared. None of these compounds showed promising tubulin inhibitory activity.**

**Key words** taxol; oxetane; phenylisoserine; biological activity; tubulin

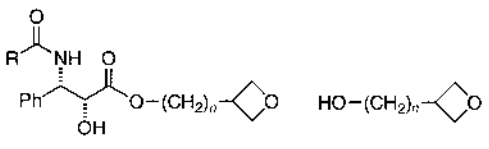
Taxol (**1**),[1] a diterpenoid isolated from *taxus brevifolia* NUTT.,[2] is clinically used in cancer chemotherapy. The mechanism of action of taxol (**1**) involves stabilization of microtubules, thereby causing inhibition of depolymerization back to tubulin.[3] Extensive studies on structure–activity relationship of taxol derivatives have revealed that a phenylisoserine moiety at C-13 and the oxetane ring are indispensable to the biological activity.[4] It is also reported that the C-2 benzoate is important for activity,[5—8] but is tolerant of subtle change.[9] A-nortaxol[10] and C-nortaxol[11] analogues have been shown to be less active both in a cytotoxicity test and a tubulin-assembly assay.

Although ample data for biological activity have been accumulated on taxol derivatives, the more fundamental question of whether the taxane skeleton is indeed necessary for the activity has not been answered. Based on our working-hypothesis that the role of the taxane skeleton is to keep the two active sites, the phenylisoserine moiety and the oxetane ring, in a suitable position to bind to the microtubule polymer, we designed compounds **3**—**8** as the simplest model compounds possessing the two moieties indispensable to the biological activity of taxol (**1**). The distance between C-13 and C-4 was determined to be 4.51 Å in the most stable conformation of taxol (**1**) calculated by MacroModel (Fig. 1).[12,13] The starting structure was generated from the crystal structure coordinates of taxotere (**2**).[14—16] The most stable conformations of **3**—**8** obtained by molecular mechanics (MM) calculations are shown in Fig. 2. The methylene chain adopts an extended conformation which aligns parallel to the benzoyloxy group. The distance between the carbon bearing the phenylisoserine unit and that bearing the oxetane is also shown in Fig. 2. The distance observed with **5** is closest to that of taxol (**1**). The straight chain analogues are highly flexible, but may easily to change conformation to accommodate the binding site on microtubules.

The synthesis of target molecule **8** is outlined in Chart 1. The iodide **17** was obtained from monobenzyl ether **16** by mesylation followed by treatment with potassium iodide in refluxing acetone in 83% overall yield. Alkylation of diethyl malonate with **17** gave **18**, which was directly reduced with $LiAlH_4$ to afford diol **19** in 43% overall yield. Monotosylate **20** was converted into oxetane derivative **21** on treatment with sodium hydride in dimethylformamide (DMF) in 43% yield. Reductive removal of the benzyl group of **21** afforded **15** in 51% yield. Attachment of a phenylisoserine unit was successfully achieved by means of the $\beta$-lactam synthon method.[17] Thus, reaction of oxetane **15** with $\beta$-lactam **22** in the presence of sodium hydride, followed by removal of the benzyl protecting group gave the desired compound **8** in 41% yield. Other target molecules **3**—**7** were synthesized from the corresponding $\alpha,\omega$-diols by a reaction sequence similar to that for **8**, although the overall yield was low. In general, in-




**1** : $R^1$ = Ph, $R^2$ = Ac (taxol)
**2** : $R^1$ = O$^t$Bu, $R^2$ = H (taxotere)




**3** : $n$ = 2, R = Ph
**4** : $n$ = 3, R = Ph
**5** : $n$ = 4, R = Ph
**6** : $n$ = 5, R = Ph
**7** : $n$ = 6, R = Ph
**8** : $n$ = 7, R = Ph
**9** : $n$ = 5, R = O$^t$Bu

**10** : $n$ = 2
**11** : $n$ = 3
**12** : $n$ = 4
**13** : $n$ = 5
**14** : $n$ = 6
**15** : $n$ = 7

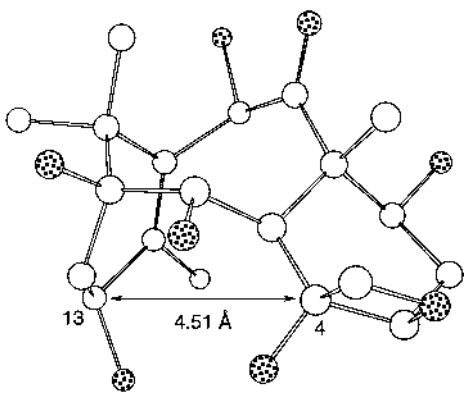



Fig. 1. Most Stable Structure of Taxol (**1**) Calculated by Molecular Mechanics (MM) with the MacroModel/MM2 (Version 4.0) Force Field

Hydrogen atoms and all of the acyl groups were omitted for clarity.

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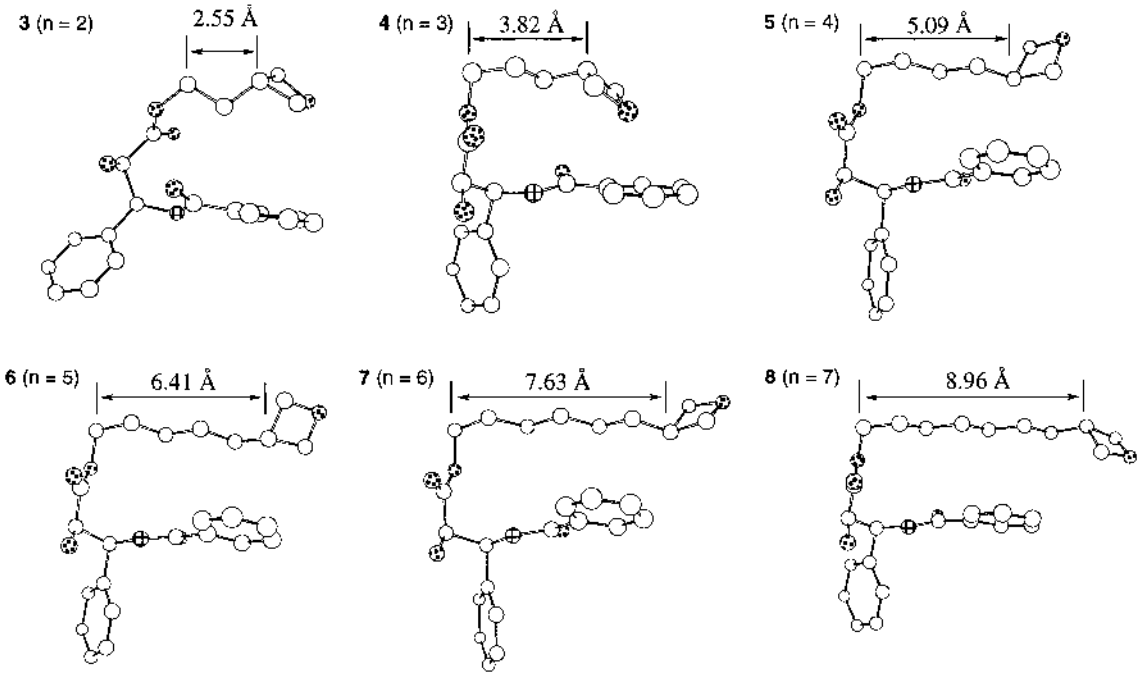


Fig. 2. Most Stable Conformation fo **3**—**8** Calculated by MM with the MacroModel/MM2 (Version 4.0) Force Field

Hydrogen atoms were omitted for clarity.

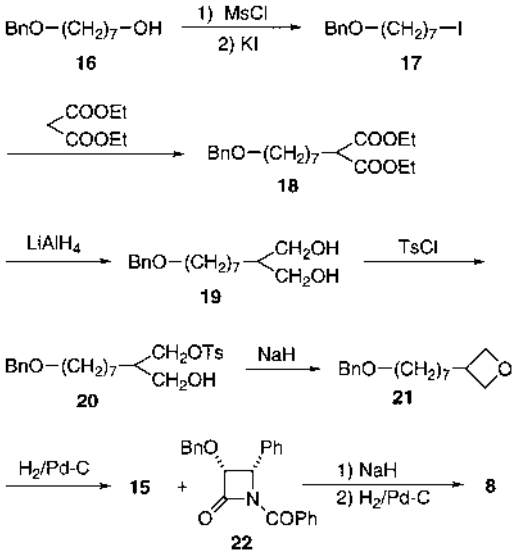


Chart 1. Synthesis of **8**

termediates were used without full characterization before the attachment of the phenylisoserine moiety. The results of combustion analysis and $^1$H-NMR spectral data of the final products **3**—**9** and the corresponding parent alcohols are given in Table 1 and Table 2, respectively.

Compounds **3**—**9** were assayed for the ability to interfere with the exchange of tubulin subunits between microtubules and the free tubulin pool using a sensitive method of screening for tubulin inhibitors that we have successfully applied to the nerve growth factor/PC12 cell system.[18] None of these compounds were found to possess any tubulin inhibitory activity (data not shown), although compounds **6**—**8** and **9** were slightly cytotoxic.

**Experimental**

$^1$H-NMR spectra were recorded on a Varian Gemini 200 (200 MHz) spectrometer and chemical shifts are given in $\delta$ (ppm) values with tetramethylsilane as the internal standard. Melting points were measured using a Yanagimoto micro melting point apparatus and are uncorrected. Preparative TLC and TLC analyses were performed on commercial glass plates bearing 0.5- and 0.25-mm layer of Merck Kiesel-gel 60 $F_{254}$, respectively. Column chromatography on silica gel was performed with Wakogel C-200 or Nacalai Tesque silica gel 60 (150—325 mesh).

**1-Benzyloxy-7-iodoheptane (17)** A mixture of 1,7-heptanediol monobenzyl ether (20.9 g, 94 mmol), pyridine (12 ml, 149 mmol), and methanesulfonyl chloride (MsCl) (8 ml, 103 mmol) in $CH_2Cl_2$ (50 ml) was stirred for 3 h at room temperature. After removal of volatile material under reduced pressure, the mixture was partitioned between 1 N HCl and AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and concentrated to give the corresponding mesylate (28.0 g, 99%) as a colorless oil. A part of mesylate (7.3 g, 24 mmol) was refluxed with NaI (11.0 g, 73 mmol) in acetone (100 ml) for 5 h. The solvent was removed under reduced pressure to give a residue which was partitioned between aq. $Na_2SO_3$ and AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and evaporated to give **17** (6.8 g, 84%). $^1$H-NMR ($CDCl_3$) $\delta$: 7.42—7.26 (5H), 4.50 (2H, s), 3.46 (2H, t, $J$=6.5 Hz), 3.18 (2H, t, $J$=7.0 Hz), 1.81 (2H, m), 1.62 (2H, m), 1.48—1.26 (6H).

**2-(7-Benzyloxyheptyl)-1,3-propanediol (19)** Ethyl malonate (4.5 ml, 30 mmol) was added to a suspension of NaH (60% oil dispersion, 1.2 g, 30 mmol) in tetrahydrofuran (THF) (30 ml) at 0 °C and the mixture was stirred at room temperature for 30 min. A solution of **17** (6.5 g, 20 mmol) in a small amount of THF was added dropwise to the mixture and the whole refluxed for 5 h. After addition of water, the mixture was concentrated under reduced pressure and extracted with AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and evaporated to give **18** in quantitative yield, which was used for the next step without further purification.

To a suspension of LiAlH4 (2.1 g, 55 mmol) in THF (50 ml), the diester **18** (6.8 g, 19 mmol) in THF (10 ml) was added and the mixture was refluxed for 5 h. After the addition of AcOEt, conc. HCl was added until a the clear solution was obtained, followed by extraction with AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and evaporated to give a residue which was purified by silica gel column chromatography. Elution with hexane–AcOEt (3 : 1) gave **19** as a colorless oil (2.3 g, 43%). $^1$H-NMR ($CDCl_3$) $\delta$: 7.32—7.42 (5H), 4.53 (2H, s), 3.82 (2H, dd, $J$=10.6, 3.9 Hz), 3.65 (2H, dd, $J$=10.5, 7.5 Hz), 3.50 (2H, t, $J$=6.6 Hz) 2.56 (2H, s), 1.86—1.55 (3H), 1.50—1.16 (10H).

Table 1. Physical Data for **3**—**15**

| Compound | Yield (%) | mp (°C) | Formula | m.w. | Analysis | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Calcd | | | Found | | |
| | | | | | C | H | N | C | H | N |
| **3** | 58[a] | 87—88[b] | $C_{21}H_{23}NO_5$ | 369.4 | 68.28 | 6.28 | 3.79 | 68.36 | 6.25 | 3.77 |
| **4** | 14[a] | 91—92[b] | $C_{22}H_{25}NO_5$ | 383.4 | 68.91 | 6.57 | 3.65 | 68.66 | 6.57 | 3.57 |
| **5** | 40[a] | 112[b] | $C_{23}H_{27}NO_5$ | 397.5 | 69.50 | 6.85 | 3.52 | 69.63 | 6.88 | 3.53 |
| **6** | 27[a] | 105—106[b] | $C_{24}H_{29}NO_5$ | 411.5 | 70.05 | 7.10 | 3.40 | 69.92 | 7.07 | 3.43 |
| **7** | 33[a] | 105[b] | $C_{25}H_{31}NO_5$ | 425.5 | 70.57 | 7.34 | 3.29 | 70.55 | 7.40 | 3.34 |
| **8** | 41[a] | 60—62[b] | $C_{26}H_{33}NO_5$ | 439.6 | 71.05 | 7.57 | 3.19 | 71.07 | 7.53 | 3.22 |
| **9** | 20[a] | 70—72[b] | $C_{22}H_{33}NO_6$ | 407.5 | 64.84 | 8.16 | 3.44 | 64.74 | 8.29 | 3.31 |
| **10** | 1[d] | | $C_5H_{10}O_2$ | 102.1 | 58.80 | 9.87 | | | | |
| **11** | 2[d] | | $C_6H_{12}O_2$ | 116.2 | 62.04 | 10.41 | | | | |
| **12** | 6[d] | | $C_7H_{14}O_2$ | 130.2 | 64.58 | 10.84 | | | | |
| **13** | 9[d] | | $C_8H_{16}O_2$ | 144.2 | 66.63 | 11.18 | | 66.26 | 11.28 | |
| **14** | 2[d] | | $C_9H_{18}O_2$ | 158.2 | 68.31 | 11.46 | | 68.01 | 11.45 | |
| **15** | 6[d] | | $C_{10}H_{20}O_2$ | 172.3 | 69.72 | 11.70 | | 69.83 | 11.73 | |

*a*) Overall yield from the corresponding oxetane. *b*) Recrystallized from AcOEt. *c*) Recrystallized from hexane–AcOEt. *d*) Overall yield from the starting monobenzyl ether.

Table 2. $^1$H-NMR Spectral Data for **3**—**15**

| Compound | Spectral data |
|---|---|
| **3** | 7.77 (2H, dd, *J*=6.3, 2.0 Hz), 7.76—7.27 (8H), 6.96 (1H, d, *J*=8.8 Hz), 5.74 (1H, *J*=9.3, 1.9 Hz), 4.78 (2H, q, *J*=6.9 Hz), 4.62 (1H, dd, *J*=3.9, 2.1 Hz), 4.39 (2H, m), 4.19 (2H, t, *J*=6.5 Hz), 3.35 (1H, d, *J*=4.0 Hz), 3.08 (1H, hep., *J*=7.5 Hz), 2.08 (2H, q, *J*=6.4 Hz) |
| **4** | 7.77 (2H, d, *J*=7.4 Hz), 7.60—7.30 (8H), 7.02 (1H, d, *J*=9.2), 5.76 (1H, dd, *J*=9.2, 2.0 Hz), 4.82—4.64 (3H), 4.37—4.05 (4H), 3.40 (1H, br ), 2.92 (1H, m), 1.88—1.42 (4H) |
| **5** | 7.80 (2H, d, *J*=8.2 Hz), 7.60—7.25 (8H), 7.03 (1H, d, *J*=9.7 Hz), 5.78 (1H, d, *J*=9.2 Hz), 4.81—4.64 (3H), 4.39—4.14 (4H), 3.35 (1H, br s), 2.91 (1H, m), 1.82—1.53 (4H), 1.35—1.17 (2H) |
| **6** | 7.77 (2H, d, *J*=8.1 Hz), 7.53—7.30 (8H), 7.01 (1H, d, *J*=8.8 Hz), 5.75 (1H, d, *J*=9.1 Hz), 4.75 (2H, dd, *J*=7.8, 5.9 Hz), 4.63 (1H, br s), 4.32 (2H, t, *J*=6.2 Hz), 4.23 (2H, m), 3.33 (1H, d, *J*=3.7 Hz), 2.99 (1H, hep., *J*=7.8 Hz), 1.70—1.54 (4H), 1.39—1.12 (4H) |
| **7** | 7.80 (2H, d, *J*=7.4 Hz), 7.54—7.28 (8H), 7.02 (1H, d, *J*=8.8 Hz), 5.75 (1H, dd, *J*=9.3, 1.4 Hz), 4.77 (2H, dd, *J*=8.5, 6.3 Hz), 4.64 (1H, dt, *J*=2.9, 1.4 Hz), 4.35 (2H, t, *J*=6.0 Hz), 4.22 (2H, m), 3.36 (1H, d, *J*=3.7 Hz), 1.75—1.57 (4H), 1.41—1.14 (6H) |
| **8** | 7.78 (2H, d, *J*=7.9), 7.56—7.28 (8H), 7.14 (1H, d, *J*=8.8 Hz), 5.75 (1H, d, *J*=9.1 Hz), 4.77 (2H, dd, *J*=7.8, 6.1 Hz), 4.63 (1H, s), 4.36 (2H, t, *J*=5.9 Hz), 4.21 (2H, m), 3.64 (1H, br s), 2.93 (1H, m), 1.71—1.53 (4H), 1.41—1.09 (8H) |
| **9** | 7.37—7.28 (5H), 5.40 (1H, d, *J*=9.2 Hz), 5.21 (1H, d, *J*=9.2 Hz), 4.77 (2H, dd, *J*=8.0, 6.0 Hz), 4.46 (1H, br s), 4.37 (2H, t, *J*=6.0 Hz), 4.25—4.18 (2H), 3.17 (1H, d, *J*=2.8 Hz), 2.97 (1H, m), 1.75—1.58 (4H), 1.41 (9H, s), 1.37—1.22 (4H) |
| **10** | 4.83 (2H, dd, *J*=7.9, 6.0 Hz), 4.46 (2H, t, *J*=6.1 Hz), 3.62 (1H, t, *J*=6.4 Hz), 3.15 (1H, hep., *J*=7.5 Hz), 1.95 (2H, q, *J*=7.5 Hz) |
| **11** | 4.82 (2H, dd, *J*=7.8, 5.9 Hz), 4.41 (2H, t, *J*=6.1 Hz), 3.63 (2H, t, *J*=6.4 Hz), 3.02 (1H, m), 2.39 (1H, s), 1.84—1.70 (2H), 1.56—1.42 (2H) |
| **12** | 4.79 (2H, dd, *J*=7.8, 6.0 Hz), 4.38 (2H, t, *J*=5.9 Hz), 3.62 (2H, t, *J*=6.6 Hz), 2.97 (1H, m), 2.12 (1H, br s), 1.77—1.48 (4H), 1.36—1.24 (2H) |
| **13** | 4.81 (2H, dd, *J*=7.8, 5.8 Hz), 4.40 (2H, t, *J*=6.1 Hz), 3.64 (2H, t, *J*=6.6 Hz), 2.97 (1H, m), 1.77—1.52 (5H), 1.45—1.18 (4H) |
| **14** | 4.78 (2H, dd, *J*=7.8, 5.8 Hz), 4.37 (2H, t, *J*=6.0 Hz), 3.63 (2H, t, *J*=6.6 Hz), 2.96 (1H, m), 1.73—1.46 (5H), 1.46—1.15 (6H) |
| **15** | 4.78 (2H, dd, *J*=7.8, 5.9 Hz), 4.37 (2H, t, *J*=6.1 Hz), 3.63 (2H, t, *J*=6.6 Hz), 2.96 (1H, m), 1.83 (1H, br s), 1.72—1.47 (4H), 1.44—1.10 (8H) |

**9-Benzyloxy-2-*p*-toluensulfonyloxymethyl)-1-nonanol (20)** A mixture of **19** (6.7 g, 24 mmol), pyridine (5.8 ml, 72 mmol), and *p*-toluenesulfonyl chloride (TsCl) (5.0 g, 26 mmol) in $CH_2Cl_2$ (100 ml) was stirred at room temperature for 8 h. After evaporation of the solvent under reduced pressure, the residue was partitioned between 1 N HCl and AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and concentrated *in vacuo*. Column chromatography of the residue gave **20** as a colorless oil (6.8 g, 65%). $^1$H-NMR ($CDCl_3$) $\delta$: 7.83 (1H, d, *J*=7.9 Hz), 7.48—7.30 (7H), 4.53 (2H, s), 4.09 (2H, m), 3.62 (2H, m), 3.48 (2H, t, *J*=6.6 Hz), 2.48 (3H, s), 1.81 (1H, m), 1.71—1.55 (3H), 1.43—1.13 (10H).

**3-(7-Benzyloxyheptyl)oxetane (21)** To a mixture of NaH (60% oil dispersion, 1.8 g, 48 mmol) in DMF (80 ml) was added **20** (6.8 g, 16 mmol) in DMF (10 ml) and the mixture was stirred at room temperature for 3 h. After addition of water, the reaction mixture was concentrated *in vacuo* and extracted with AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and evaporated to give a residue which was chromatographed over silica gel (hexane : AcOEt=3 : 1) to afford **21** as a colorless oil (1.8 g, 43%). $^1$H-NMR ($CDCl_3$) $\delta$: 7.44—7.25 (5H), 4.78 (2H, dd, *J*=7.8, 5.8 Hz), 4.51 (2H, s), 4.37 (2H, t, *J*=6.1 Hz), 3.46 (2H, t, *J*=6.6 Hz), 2.95 ($^1$H, hep. *J*=7.7 Hz), 1.77—1.50 (4H), 1.47—1.07 (8H).

**3-Oxetaneheptanol (15)** A mixture of **21** (1.8 g, 6.9 mmol) and 10% palladium on charcoal (300 mg) in EtOH (10 ml) was stirred under $H_2$ for 4 d. Usual work-up followed by column chromatography on silica gel (hexane : AcOEt=3 : 1) gave **15** as a colorless oil (610 mg, 51%). Physical data, including $^1$H-NMR spectrum, are given in Tables 1 and 2.

**Compound 8** A mixture of **15** (91 mg, 0.53 mmol) and NaH (60% oil dispersion, 500 mg, 12.5 mmol) was stirred at 0 °C for 10 min. and $\beta$-lactam **22** (236 mg, 0.66 mmol) in THF (2 ml) was then added to the mixture. After stirring for 45 min. a mixture of AcOH (1 ml), water (1 ml), and THF (10 ml) was added and the mixture was concentrated *in vacuo* to give a residue that was then partitioned between 1 N HCl and AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and evaporated to give a residue. Column chromatography over silica gel (hexane : AcOEt=2 : 1) afforded the desired ester (210 mg, 60%). This product was directly hydrogenated over 10% palladium on charcoal in EtOH gave **8** (118 mg, 68%) after purification by column chromatography over silica gel (hexane : AcOEt=3 : 1). Physical data, including $^1$H-NMR spectrum, are given in Tables 1 and 2.

**References and Notes**


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