There is a great medical need for an injectable antifungal agent with a broad spectrum for the treatment of severe deep mycoses of hospitalized patients. Currently, injectable antifungal azoles, fluconazole (FLCZ), fosfluconazole and voriconazole are available for parenteral use, but they have limitations in terms of antifungal spectra and safety, respectively. Most of the azoles under development have a broader spectrum but cannot be administered parenterally because of low water-solubility.

Previously, we identified CS-758 which has a broad antifungal spectrum covering Aspergillus spp., FLCZ-resistant Candida spp. and has a good safety profile, including low drug–drug interaction. Since the water solubility of CS-758 was, however, too low for a parenteral formulation, we conducted a study on a prodrug of CS-758 which should have sufficient water solubility and efficient bioconversion.

CS-758 was selected as a candidate for clinical trials, but since its water-solubility was insufficient for an injectable formulation, phosphoryl ester prodrugs were designed. In this study, the synthesis and evaluation of these injectable prodrugs are described. Phosphoryl ester 17h was soluble in water, and was stable in both water and in a solid state. 17h was converted to CS-758 in human liver microsome and was also converted to CS-758 in rats after intravenous (i.v.) administration with good conversion speed and efficiency. 17h (i.v.) reduced the viable cell counts in kidneys in a murine hematogenous Candida albicans infection model and in lungs in a murine pulmonary Aspergillus fumigatus infection model, wherein the effects were comparable to or slightly superior to that of CS-758 (per os).

Key words water-soluble prodrug; phosphate ester; antifungal azole; CS-758
rapid and spontaneous intra-molecular cyclization to release CS-758 and a lactone.\textsuperscript{20}

In this paper, we describe the design and synthesis of water-soluble prodrugs of CS-758, and their physicochemical and biological properties.

First, we designed and synthesized the prodrug 2. We chose a 4-hydroxybutyrate moiety as the self-cleavable linker and a phosphoric acid ester moiety as the solubilizing part because phosphoric acid ester has good water solubility and dephosphorylation is a familiar enzyme response. 4-Hydroxybutyrate ester afforded by dephosphorylation was expected to afford a lactone and the parent drug CS-758. The synthesis of compound 2 is shown in Chart 2.

Sodium 4-hydroxybutyrate 4 was reacted with 4-methoxybenzyl chloride to afford 4-methoxybenzyl ester 5. As the ester 5 was unstable, 5 was used for the next step without further purification. 5 was phosphorylated by using the procedure developed by Fraser–Reid\textsuperscript{21} to give the phosphoryl ester 6. The 4-methoxybenzyl (PMB) group of 6 was removed using trifluoroacetic acid (TFA) and anisole to obtain the carboxylic acid 7, which was treated with oxalyl chloride in CH$_2$Cl$_2$ to afford the corresponding acid chloride 8. Though the tertiary hydroxy group of CS-758 was almost unreactive with acid chlorides because of its steric hindrance, the sodium alkoxide form of CS-758, which was generated from CS-758 and NaH in tetrahydrofuran (THF), could be reacted with acid chloride 8 at room temperature to afford the desired ester 9 in 63\% yield. Removal of two allyl groups of 9 was accomplished by the use of tetrakis(triphenylphosphine)palladium and pyrrolidine in CH$_2$Cl$_2$ to afford the desired compound as a pyrrolidinium salt. The salt was purified with a C-18 reverse phase column, and then the counter ion was exchanged using Dowex 50W×8 (Na form) to give sodium salt 2.

The pharmacokinetic (PK) profile after intravenous (i.v.) bolus administration of 2 to rats at a dose of 2 mg (CS-758 equivalent)/kg is shown in Fig. 3. As expected, compound 2 disappeared from plasma within 1 h and rapid formation of dephosphorylated intermediate alcohol was observed by HPLC identification. Though the intermediate alcohol disappeared from plasma rapidly and formation of CS-758 was

\textbf{Chart 1. General Concept of Cascade-Type Prodrugs}

\textbf{Chart 2. Synthesis of 2}

\textbf{Chart 3. Synthesis of 3}

\textbf{Fig. 3. Plasma Level of CS-758 and Intermediate Alcohol after i.v. Administration of 2 to Rats at a Dose of 2 mg (CS-758 Equivalent)/kg (Average of Three Rats)
observed, the conversion efficiency was 3.4% and there are some room for improvement.

We assumed that conformational restriction of linker part might hasten intra-molecular cyclization of the intermediate alcohol and high conversion efficiency might be achieved. On the basis of this supposition, we designed compound 3, which employs a 2-(hydroxymethyl)benzoyl moiety as the self-cleavable linker and a phosphoric acid ester moiety as the solubilizing part.

Compound 3 was synthesized in a similar manner to 2. The phthalide ring in 10 was opened by treatment with 1 eq of NaOH to afford sodium salt 11. The sodium salt 11 was treated with 4-methoxybenzyl chloride to give the unstable hydroxy ester 12. The ester 12 was treated with diallyl disopropylphosphoramidite in the presence of tert-butyl hydroperoxide to give the corresponding phosphate 13. The 4-methoxybenzyl group of 13 was removed by TFA and the afforded carboxylic acid 14 was treated with oxalyl chloride to give acid chloride 15. The oxide anion prepared from CS-758 was esterified with tert-butyl)tin hydride.

The PK profile after i.v. bolus administration of 3 to rats at a dose of 2 mg (CS-758 equivalent)/kg is shown in Fig. 4. The conversion rate of intermediate alcohols to drug CS-758 was improved. In the case of 3, maximum drug concentration time (T_{max}) is 0.5 h, whereas T_{max} was 1.3 h in the case of 1. The conversion efficiency (BA)^22) of compound 3 to CS-758 was 28.7%.

Aiming to further improve the conversion efficiency, we decided to continue modifying compound 3, as shown in Fig. 5. We expected that certain steric effects or electric effects in the linker part might improve the conversion efficacy and conversion speed. In compounds 17a–i, substituents were introduced to the benzene ring of the linker part and in compounds 17j and k, the linker benzene ring was replaced by a naphthalene or furanyl ring. These compounds were synthesized as follows.

Compounds 17a–d, h and i were synthesized in a similar manner starting from corresponding phthalide derivatives (Chart 4).

Compounds 17e–g were synthesized as depicted in Chart 5. Diols 25e–g were reacted with 1 eq of tert-butylchlorodi-methylsilane (TBSCI) in THF at 0°C to afford isomers 26e–g preferentially. The structure of 26e–g was determined by measuring the nuclear Overhauser effect (NOE) in the 1H-NMR spectrum. Phosphorylation of 26e–g with diallyl disopropylphosphoramidite and successive oxidation with tert-butyl hydroperoxide gave phosphate esters 27e–g. The silyl group of 27e–g was removed with tetrabutylammonium fluoride in THF and the following oxidization with Jones reagent afforded carboxylic acids 29e–g. Acid chlorides 30e–g were reacted with the sodium alkoxide form of CS-758 to give 31e–g. Removal of the two allyl groups in 31e–g was accomplished by use of bis(triphenylphosphine)dichloropalladium and tri(n-butyl)tin hydride in dichloromethane. The sodium salt 17e–g was prepared by treatment with sodium hydrogen carbonate and purified by C-18 reverse-phase column chromatography. 17j and 17k were synthesized in a manner similar to the synthesis of

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Table 1. Conversion of Compounds 3, and 17a–k to CS-758 in Human Plasma and Liver Micosome

<table>
<thead>
<tr>
<th>Compd.</th>
<th>Human plasma(^a)</th>
<th>Human liver micosome(^a)</th>
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<td>30 min</td>
<td>120 min</td>
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<tr>
<td>3</td>
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<td>17k</td>
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\(^a\)−, no detection or very low formation of CS-758 (conversion yield: <20%); ±low formation of CS-758 (conversion yield 20–40%); +, middle formation of CS-758 (conversion yield: 40–70%); ++, high formation of CS-758 (conversion yield: >70%).

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Fig. 4. Plasma Level of CS-758 after i.v. Administration of 3 to Rats at a Dose of 2 mg (CS-758 Equivalent)/kg (Average of Three Rats)

Fig. 5. Structural Formulas of Compounds 17a–k
Compounds 17a–k were incubated with human plasma or with human liver microsome and the conversion of these compounds to CS-758 was determined. The results are summarized in Table 1. Compounds 17a–d, which had a F atom on the benzene ring, 17a and 17d were superior. The steric effect of the F atoms which are situated in α-position or ψ-position on the benzene ring (position number shown in Chart 4) might hasten second spontaneous cleavage of the –O–CH2–Ar–C(O)– linker. Compounds 17e–g, which have a substituent on the ψ-position of benzene ring gave favorable results, but the conversion speed and conversion efficiency of these compounds were inferior to 17d. In these compounds, 17h and 17i which had CN group at β-position or γ-position on the benzene ring afforded best results. The electron-withdrawing group on the benzene ring might hasten cyclization of the –O–CH2–Ar–C(=O)– group. Also a compound with a furan ring in the linker part (17k) and a compound with a naphthalene ring in the linker part (17j) showed good results.

The disappearance of 17h and 17i, and the formation of CS-758 after incubation with human liver microsome are shown in Figs. 6 and 7, respectively. These compounds were converted to CS-758 within 1 h with excellent conversion efficacy.

The in vivo conversion of 17h and 17i to CS-758 upon i.v. administration to rats (2 mg of CS-758 equivalent/kg) is shown in Figs. 8 and 9, respectively. When 17h was administered to rats, CS-758 was quickly formed (Tmax: 0.7 h) and a high concentration of CS-758 was observed (Cmax: 0.512 µg/ml). CS-758 was eliminated slowly (t1/2: 5.7 h) and the BA of CS-758 was almost quantitative (94.4%). Interestingly, when 17i was administered to rats, 17i was eliminated from plasma within 1 h. But the conversion efficiency of 17i was far less than that of 17h. The reason for this large difference between 17h and 17i in the conversion to CS-758 was undefined.

The in vivo efficacies of the prodrug 17h (i.v. administration) and CS-758 (oral administration) were evaluated in a hematogenous C. albicans infection model in mice (Fig. 10) and in a pulmonary A. fumigatus infection model in mice (Fig. 11). In the hematogenous C. albicans infection model, i.v.-administered prodrug 17h exhibited high activity which was almost equal to or slightly better than that of orally administered CS-758. In a pulmonary aspergillosis model in mice, the efficacy of i.v-administered 17h was superior to that of orally administered CS-758.

Furthermore compound 17h had good water solubility (>30 mg/ml in water) and sufficient stability in solution at around neutral conditions. 17h was also stable in solid state as an amorphous salt.

In summary, we developed a widely applicable prodrug technique for the solubilization of compounds having a hydroxy group. We identified the phosphoryl ester prodrug 17h, which showed potent antifungal activity against both hematogenous candidasis and pulmonary aspergillosis in
mice via i.v. administration. Because there are few injectable antifungal azole agents with a broad spectrum, 17h could be a promising drug for the treatment of fungal infections.

Experimental

1H-NMR spectra were recorded either on a Varian Mercury 400 (400 MHz) or a Varian Mercury 500 (500 MHz) spectrometer using tetramethylsilane as an internal standard. MS and high-resolution MS (HR-MS) were recorded either on a JEOL HX-100, a SX-102A or a JMS-AX-505H mass spectrometer. Melting points were determined using a Yanagimoto micro melting point apparatus and are uncorrected. IR spectra were recorded on a JASCO FT-IR 8900 spectrometer. Optical rotations were measured on a JASCO P-1030 polarimeter.

4-Methoxybenzyl 4-[(Bis(allyloxy)phosphoryl)oxy]butanoate (6) To a suspension of a sodium 4-hydroxybutyrate (630 mg, 5.00 mmol) in N,N-dimethylformamide (DMF) (3.5 ml) was added 4-methoxybenzyl chloride (783 mg, 5.00 mmol), and the mixture was heated at 80 °C for 3 h. Water and EtOAc were added to the mixture and the organic layer was separated. The organic layer was dried over anhydrous magnesium sulfate, and the solvent was distilled off under reduced pressure to give a crude product of 5 as an oil. The crude oil of 5 was dissolved in CH2Cl2 (5 ml), and tetrazole (700 mg, 10 mmol) and diallyl diisopropylphosphoramidite (1.5 g, 6.1 mmol) were added at 0 °C. The mixture was stirred at room temperature for 30 min, and methanol (0.1 ml) was added to the mixture. The mixture was stirred for 5 min at room temperature and tert-butyl hydroperoxide (ca. 5 M nonane solution, 1.5 ml, ca. 7.5 mmol) was added to the solution at 0 °C followed by stirring the mixture at room temperature for 30 min. Saturated NaHCO3 solution and an aqueous solution of sodium thiosulfate were added to the mixture, and the mixture was stirred for 10 min. The product was extracted with EtOAc and the organic layer was washed with brine, dried over anhydrous magnesium sulfate and the solvent was removed under reduced pressure to give an oily residue. The residue was subjected to chromatography on silica gel (30 g) column (EtOAc : hexane = 2 : 3—1 : 1) to give 6 (1.55 g, 81% yield) as a colorless oil. 1H-NMR (400 MHz, CDCl3) δ: 2.01 (2H, quint, J = 7 Hz), 2.47 (2H, t, J = 7 Hz), 3.81 (3H, s), 4.10 (2H, q, J = 7 Hz), 4.50—4.55 (4H, m), 5.06 (2H, s), 5.25 (2H, br d, J = 10 Hz), 5.36 (2H, br d, J = 17 Hz), 5.93 (2H, ddt, J = 17, 10, 5 Hz), 6.84 (2H, d, J = 9 Hz), 7.29 (2H, d, J = 9 Hz). IR (neat) cm⁻¹: 1731, 1613, 1516, 1464, 1254. MS m/z (FAB):
was used for the next step without further purification. 1H-NMR (400 MHz, D 2O) 3.55 (M d, J 12 Hz), 3.46 (1H, t, J 11 Hz), 3.37 (2H, q, J 7 Hz), 2.35—2.50 (2H, m), 2.87 (1H, m), 3.43 (1H, t, J 11 Hz), 5.35 (2H, s), 5.38 (2H, d, J 11 Hz), 5.32 (2H, d, J 11 Hz), 4.15—4.25 (2H, m), 4.55—4.60 (4H, m), 5.00 (1H, d, J 4 Hz), 5.27 (2H, d, J 11 Hz), 5.35 (2H, s), 5.38 (2H, d, J 17 Hz), 5.85 (1H, dd, J 15, 4 Hz), 5.90—6.00 (2H, m), 6.58 (1H, d, J 17 Hz), 6.85—6.95 (3H, m), 7.30—7.45 (3H, m), 7.57 (1H, t, J 8 Hz), 7.90 (1H, s), 7.92 (1H, s). IR (KBr) cm⁻¹: 2233, 1741, 1615, 1600, 1504. MS m/z (FAB): 789 (M⁺+1).

4-[(Bis(allyloxy)phosphoryl)oxy]butanoic Acid (7) To a mixture of 6 (700 mg, 1.82 mmol) and anisole (0.7 ml) was added TFA (3 ml) at room temperature. The mixture was stirred at room temperature for 15 min, then diluted with toluene (5 ml), and the solvent was distilled off under reduced pressure to give crude product of 7 as a pale yellow oil. The crude oil of 7 was used for the next step without further purification. 1H-NMR (400 MHz, CDCl3) δ: 2.03 (2H, quint, J 15, 10, 5 Hz), 11.29 (1H, br s).

(2R,3R)-3-[(trans-2-[(1E,3E)-4-(4-Cyano-2-fluorophenyl)buta-1,3-dien-1-yl]-1,3-dioxan-5-yl)sulfanyl]-2-(2,4-difluorophenyl)-1-(1H-1,2,4-triazol-1-yl)butan-2-yl 4-[(Bis(allyloxy)phosphoryl)oxy]butanoate (9) DMF (0.05 ml) and oxalyl chloride (350 mg, 2.76 mmol) were added to the solution of 7 in CH2Cl2 (3.5 ml) and the mixture was stirred at room temperature for 1 h, then toluene was added, and the mixture was concentrated under reduced pressure to afford 8 as a crude material. Sodium hydride (55% dispersion in mineral oil; 80 mg, 1.83 mmol) was added to the solution of 8 in CH2Cl2 (3.5 ml) and the mixture was stirred at room temperature for 3 h. The obtained suspended mixture was cooled to 0 °C, and the crude material of 8 was added to the mixture. The mixture was stirred at room temperature for 30 min. After cooling, the mixture was partitioned between EtOAc and an aqueous solution of ammonium chloride, and the organic layer was washed with brine, dried over anhydrous magnesium sulfate and the solvent was distilled off under reduced pressure. The oily residue was purified by chromatography on silica gel (30 g) (EtOAc : methanol = 1: 0—10: 1) to afford 9 (862 mg, 63% yield) as a pale yellow amorphous solid. 1H-NMR (400 MHz, CDCl3) δ: 1.35 (3H, dd, J = 7, 2 Hz), 1.90—2.10 (2H, m), 2.46 (1H, dt, J = 17, 7 Hz), 2.57 (1H, dt, J = 17, 7 Hz), 3.04 (1H, tt, J = 11, 5 Hz), 3.52 (2H, t, J = 11 Hz), 3.90 (1H, q, J = 7 Hz), 4.12 (2H, q, J = 7 Hz) 4.15—4.25 (2H, m), 4.55—4.60 (4H, m), 5.00 (1H, d, J = 4 Hz), 5.27 (2H, d, J = 11 Hz), 5.35 (2H, s), 5.38 (2H, d, J = 17 Hz), 5.85 (1H, dd, J = 15, 4 Hz), 5.90—6.00 (2H, m), 6.58 (1H, dd, J = 16, 11 Hz), 6.74 (1H, d, J = 15 Hz), 6.85—6.95 (3H, m), 7.30—7.45 (3H, m), 7.57 (1H, t, J = 8 Hz), 7.90 (1H, s), 7.92 (1H, s). IR (KBr) cm⁻¹: 2233, 1741, 1615, 1600, 1504. MS m/z (FAB): 789 (M⁺+1).

Disodium 4-[(2R,3R)-3-[(trans-2-[(1E,3E)-4-(4-Cyano-2-fluorophenyl)buta-1,3-dien-1-yl]-1,3-dioxan-5-yl)sulfanyl]-2-(2,4-difluorophenyl)-1-(1H-1,2,4-triazol-1-yl)butan-2-yl]oxy]-4-oxobutyl Phosphate (2) Pyrrolidine (644 mg, 9.06 mmol) was added to the solution of 9 (350 mg, 0.453 mmol), tetrakis(triphenylphosphine)palladium (5 mg, 0.0043 mmol), and triphenylphosphine (5 mg, 0.019 mmol) in CH2Cl2 (3 ml) at room temperature under N2 atmosphere. After the reaction mixture was stirred for 1 h, it was diluted with toluene and the solvent was distilled off under reduced pressure. The residue was purified by reverse phase column chromatography using Cosmosil 75 C18-PREP (water : methanol = 1: 0—4: 6). The concentrated eluates were passed through an ion exchange column of Dowex 50W×8 (Na form). The collected fractions were concentrated under reduced pressure and lyophilized to afford 5 (233 mg, 64% yield) as an amorphous colorless solid. 1H-NMR (400 MHz, D2O) δ: 1.13 (3H, d, J = 7, 16 Hz), 2.37—2.50 (2H, m), 2.87 (1H, m), 3.43 (1H, t, J = 12 Hz), 3.46 (1H, t, J = 12 Hz), 3.55—3.65 (3H, m), 3.95—4.05 (2H, m), 4.12 (2H, q, J = 7 Hz), 4.15—4.25 (2H, m).
(3H, m), 7.20—7.30 (1H, m), 7.30—7.45 (3H, m), 7.50—7.65 (FAB): 801 (M + 1). 4-Methoxybenzyl 2-[[(allyl)oxo]phosphoryl(oxo)methyl]benzoate (13) An aqueous solution (4.8 ml) of sodium hydroxide (0.56 g, 10 mmol) was added to a solution of 2-fluorobenzil (1.34 g, 10 mmol) in THF (30 ml). After the reaction mixture was stirred at room temperature for 3 h, the solution was concentrated under reduced pressure, and the residue was dried using a vacuum pump to give 11 as a crude material. 4-Methoxybenzyl chloride (2.04 g, 13 mmol) was added to the solution of the crude material of 11 in DMF (30 ml), and, then the mixture was stirred at 80°C for 1 h. After cooling the mixture, a saturated NH4Cl solution was added to the mixture, and the product was extracted with EtOAc. The organic layer was washed with water and then with brine, and the solvent was distilled off under reduced pressure to afford crude product of 12 as the residue. The residue was dissolved in CH2Cl2 (70 ml), and tetraceto (1.4 g, 20 mmol) and diisopropylphosphamide (2.45 g, 10 mmol) were added at 0°C, then the mixture was stirred at room temperature for 30 min, and methanol (0.1 ml) was added to the mixture. The mixture was stirred for 5 min further, and tert-butyl hydroperoxide (ca. 5 M nonane solution, 4.6 ml, ca. 23 mmol) was added at 0°C followed by stirring the mixture at room temperature for 30 min. Saturated NaHCO3 solution and an aqueous solution of sodium thiosulfate were added to the mixture, and the mixture was stirred for 10 min and then partitioned between EtOAc and water. The organic layer was washed successively with saturated NaHCO3 solution, saturated NH4Cl solution, and an aqueous solution of sodium chloride, and then dried over anhydrous MgSO4, and the solvent was distilled off under reduced pressure to give an oily residue. The residue was chromatographed on silica gel (EtOAc:hexane = 3:1—1.2) to give 13. 1H NMR (400 MHz, CDCl3): 6.85—6.90 (3H, m), 7.20—7.30 (1H, m), 7.30—7.45 (3H, m), 7.50—7.65 (FAB): 837 (M + 1). With water and then with brine, and the product was extracted with EtOAc. The organic layer was washed with water and then with brine, and the solvent was distilled off under reduced pressure to afford crude product of 12 as the residue. The residue was dissolved in CH2Cl2 (70 ml), and tetraceto (1.4 g, 20 mmol) and diisopropylphosphamide (2.45 g, 10 mmol) were added at 0°C, then the mixture was stirred at room temperature for 30 min, and methanol (0.1 ml) was added to the mixture. The mixture was stirred for 5 min further, and tert-butyl hydroperoxide (ca. 5 M nonane solution, 4.6 ml, ca. 23 mmol) was added at 0°C followed by stirring the mixture at room temperature for 30 min. 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Disodium 2-[[Bis(allyloxy)phosphoryl]methyl]-5-fluorobenzonic Acid (22c) In a similar manner to 22c was obtained in 21c in quantitative yield as a colorless oil. The crude product of 22c was used for the next step without further purification.

(2R,3R)-3-[[trans-2-[(1E,3E)-4-(4-Cyano-2-fluorophenyl)buta-1,3-dien-1-yl]-1,3-dioxan-5-yl]sulfanyl]-2-(2,4-difluorophenyl)-1-(1H,1,2,4-triazol-1-y1)-butan-2-yl (22d) In a similar manner to 22d was obtained from 21d in quantitative yield as a colorless oil.

Disodium 4-[[trans-2-[(1E,2E)-4-(4-Cyano-2-fluorophenyl)buta-1,3-dien-1-yl]-1,3-dioxan-5-yl]sulfanyl]-2-(2,4-difluoropheny]-1-(1H,1,2,4-triazol-1-y1)-butan-2-yl (22e) In a similar manner to 22e was obtained from 21e in quantitative yield as a colorless oil.
The image contains a page from a scientific document discussing experimental procedures and results. The text is too detailed and technical to be summarized accurately without context or additional information. It appears to be related to chemical synthesis and characterization, including the use of solvents, chromatography, and spectroscopic analyses. The page includes equations and specific chemical compounds. Without more context, it is not possible to provide a meaningful summary in the natural text format.
(1H, ddd, J = 11, 5, 2 Hz), 4.19 (1H, ddd, J = 11, 5, 2 Hz), 4.42—4.45 (4H, m), 4.96 (1H, d, J = 5 Hz), 5.19 (2H, br, J = 10 Hz), 5.30 (2H, br, J = 18 Hz), 5.43—5.56 (4H, m), 5.83 (1H, dd, J = 16, 5 Hz), 5.82—5.92 (2H, m), 6.55 (1H, dd, J = 16, 11 Hz), 6.73 (1H, d, J = 16 Hz), 6.86—6.89 (3H, m), 7.30—7.35 (2H, m), 7.39—7.43 (3H, m), 7.48 (1H, td, J = 9, 6 Hz), 7.57 (1H, t, J = 8 Hz), 7.94 (1H, s), 8.00 (1H, s). IR (CHCl₃) cm⁻¹: 2233, 1731, 1504, 1462, 1277, 1141, 1059, 1018, 991. MS m/z (FAB): 851 (M⁺)

Disodium 2-[(Butyl(dimethyl)silyloxy)methyl]-2-(4-fluorophenyl)-2,4-di- fluorophenyl]-1-(1H,1,2,4-triazol-1-yl)-butan-2-yl-oxy]carbonyl]-6-methylbenzyl Phosphonic acid (17f) In a similar manner to 17a, 17f was obtained from 31f in 57% yield as a colorless solid. ¹H-NMR (400 MHz, CD₃OD): δ: 0.87 (9H, s), 3.47—3.48 (4H, m), 5.16 (2H, br, J = 10 Hz), 5.24 (2H, dq, J = 18, 1 Hz), 5.25 (2H, s), 5.80 (2H, J = 10 Hz), 7.07—7.16 (3H, m), 7.47—7.48 (2H, m), 7.61 (1H, d, J = 7, 1 Hz), 7.89 (1H, d, J = 7, 1 Hz), 7.92 (1H, d, J = 7, 1 Hz). IR (CHCl₃) cm⁻¹: 3603, 1732, 1720, 1208, 990. MS m/z (FAB): 463 (M⁺)
lar manner to 28e, 41 was obtained from 40 in 76% yield as a colorless oil. 1H-NMR (400 MHz, CDCl3)  δ: 3.168 (1H, t-like, J = ca. 6 Hz), 4.523 (4H, td, J = 7, 1.2 Hz), 4.580 (2H, d, J = 6.6 Hz), 5.050 (2H, d, J = 9.5 Hz), 5.255 (2H, dd-like, J = 10, ca. 1.2 Hz), 5.350 (2H, dd-like, J = 17, ca. 1.2 Hz), 5.917 (2H, dd, J = 17, 10, 6 Hz), 7.414 (1H, s), 7.483 (1H, s). IR (CHCl3) cm⁻¹: 3401, 1602, 1544, 1422, 1267, 1022. MS m/z (FAB): 289 (M⁺+1).

**Diallyl (4-Formyl-3-furyl)methyl Phosphate (42)** To a solution of 41 (113 mg, 0.39 mmol) in CH2Cl2 (2 ml), activated manganese dioxide (0.52 g, 3401, 1602, 1554, 1462, 1424, 1426, 1267, 1022. MS m/z (FAB): 289 (M⁺+1).

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References and Notes
22) Bioavailability (BA) was calculated by the followiing. BA (%) = [(area under the curve (AUC) of CS-758 after iv administration of the prodrug]/(AUC of CS-758 after iv administration of CS-758 dissolved in polyethylene glycol solution)]×100.