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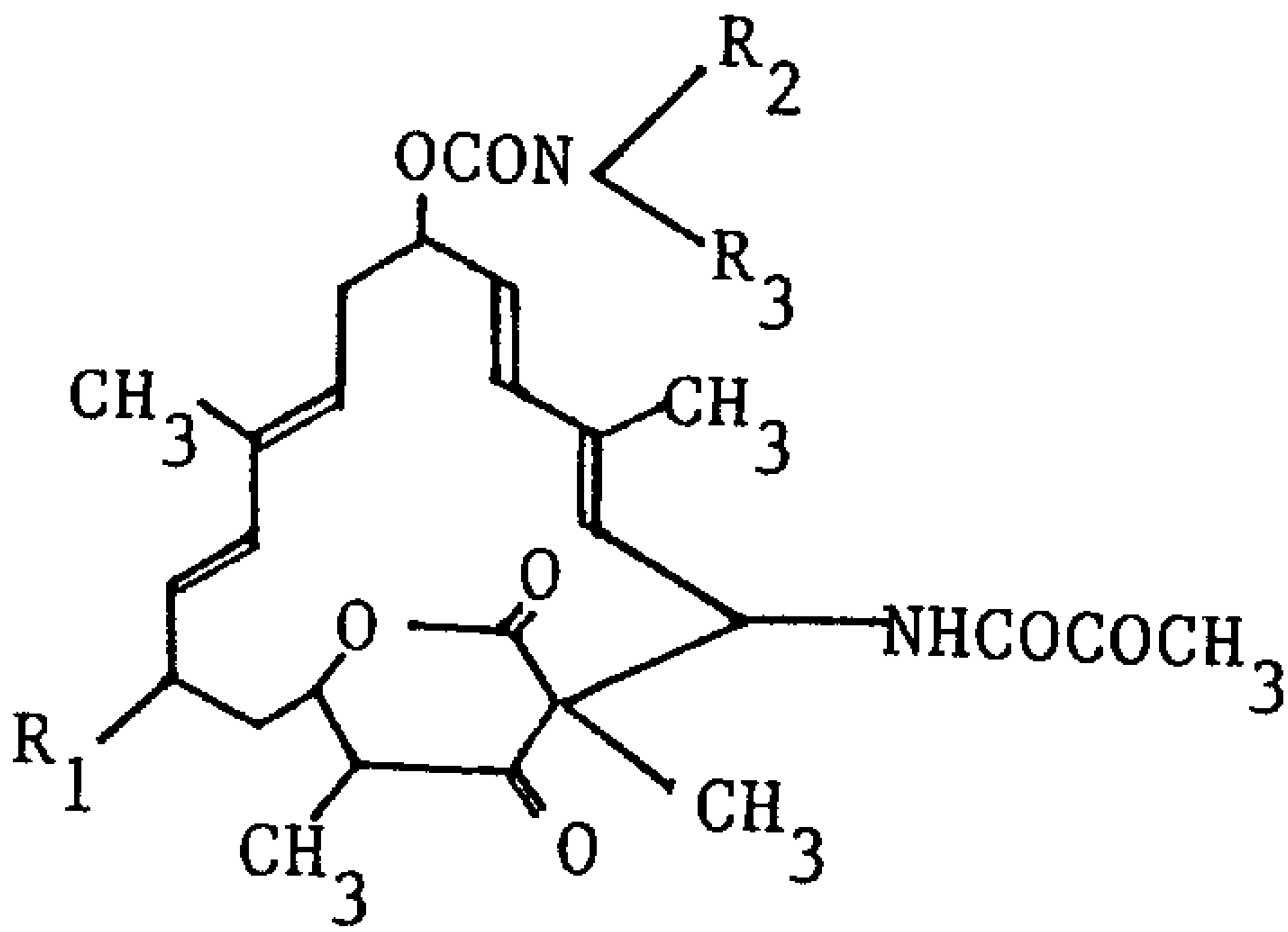
(30) 1989/07/25 (191818/1989) JP
(54) **PROCEDE DE PREPARATION DE DERIVES DE CARBAMATE DE LANKACIDINE**
(54) **PROCESS FOR PREPARING LANKACIDINE CARBAMATE DERIVATIVES**



(57) A process for preparing a lankacidine carbamate derivative of the formula (see above formula) or a salt thereof, by the reaction of a lankacidine 8-substituted carbonate with an amine, wherein R_1 is a hydroxy group or an alkanoyloxy group; and R_2 and R_3 are each a hydrogen atom, an optionally substituted lower alkyl group, a cycloalkyl group or a phenyl group; or R_2 and R_3 together with an adjacent nitrogen atom to which they bond form an optionally substituted heterocyclic group. The product is useful as anti-microbial agent.

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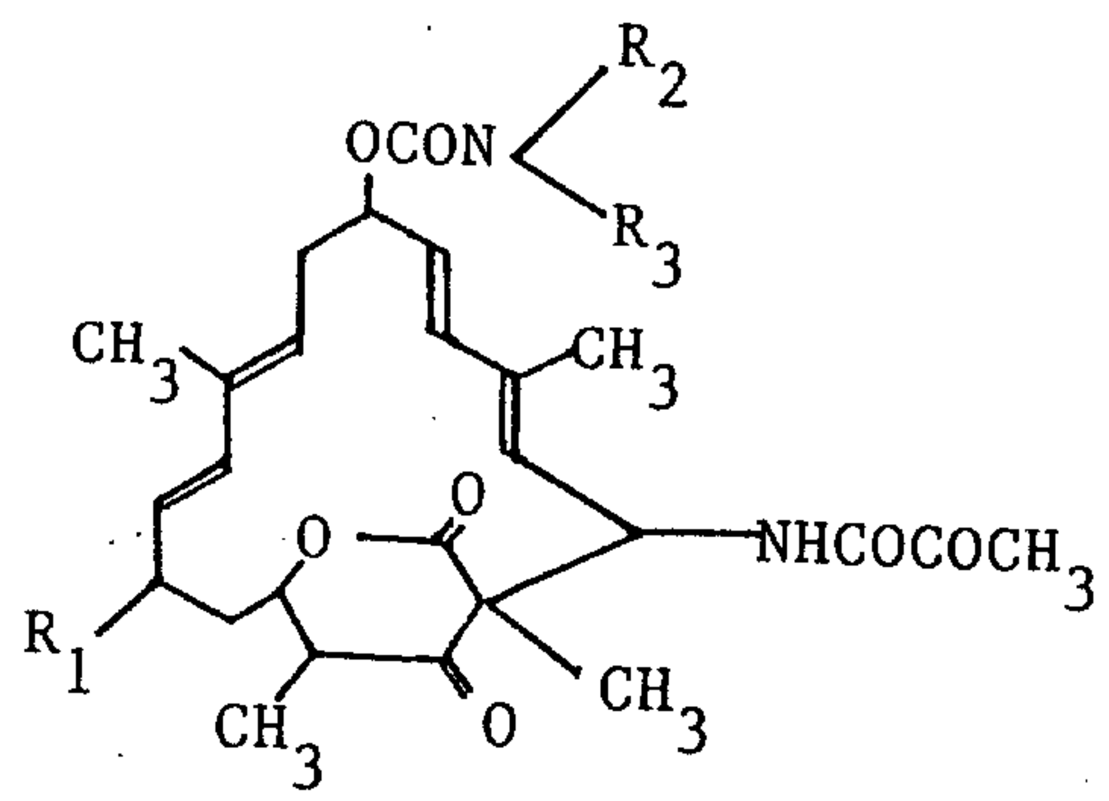
ABSTRACT OF THE DISCLOSURE

A process for preparing a lankacidine carbamate derivative of the formula



or a salt thereof, by the reaction of a lankacidine 8-substituted carbonate with an amine, wherein R_1 is a hydroxy group or an alkanoyloxy group; and R_2 and R_3 are each a hydrogen atom, an optionally substituted lower alkyl group, a cycloalkyl group or a phenyl group; or R_2 and R_3 together with an adjacent nitrogen atom to which they bond form an optionally substituted heterocyclic group. The product is useful as anti-microbial agent.

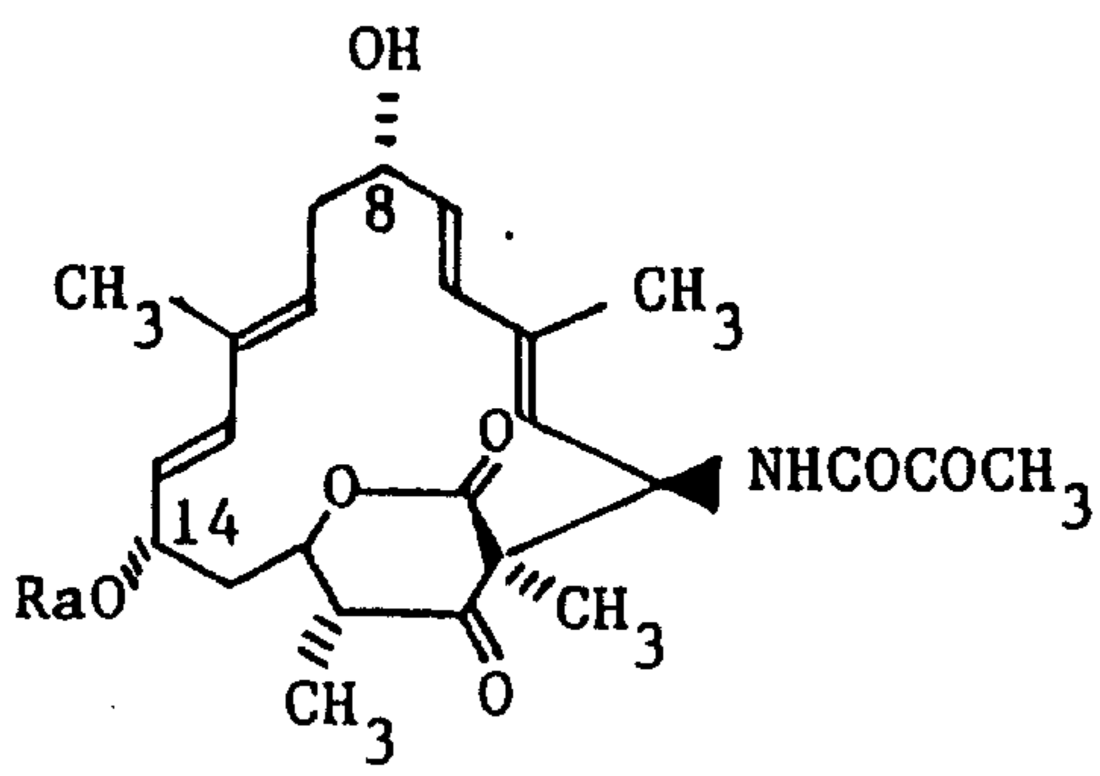
BACKGROUND OF THE INVENTION

1. Field of the Invention

The present invention relates to a process for preparing lankacidine carbamate derivatives having antimicrobial activities.

2. Prior Art

Lankacidines are antibiotics which are produced and accumulated by cultivating a Streptomyces strain and have the structure represented by the formula:



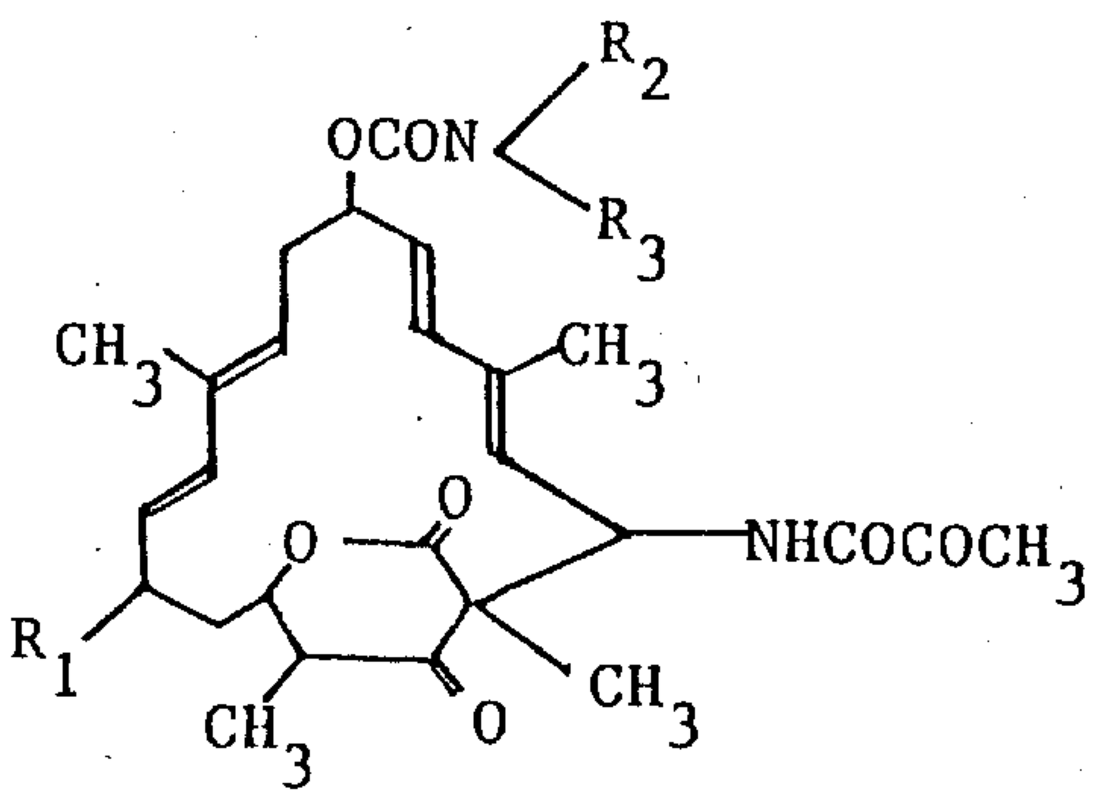
In the above formula, where Ra is $COCH_3$, the compound is lankacidine A, and where Ra is H, it is lankacidine C. Known derivatives of the lankacidines are, for example, those having an ester group at the 8 and/or 14 positions of lankacidine C [see Kagaku & Seibutsu (Chemistry and Organism, Vol. 15, pp. 337 - 342 (1977)): Journal of the Takeda Research Laboratories, Vol. 41, pp. 81 - 113 (1982)], those having an acyloxy group or the like at 8 and/or 14 positions thereof [see The Journal of Antibiotics, Vol. 26, pp 647 (1973)], and those having 8-substituted alkyl ester, 8-carbonate ester and 8-substituted carbamate (Japanese Published Unexamined Patent Application No. 240687/1987). Japanese Published Unexamined Patent Application No. 240687/1987 also discloses processes for preparing lankacidine A carbamate in which lankacidine A is reacted with isocyanates, and pentachlorophenoxy- or 2,4,5-trichlorophenoxycarbonate of lankacidine A is reacted with amines. These known processes are, however, required to use high toxic isocyanates or expensive polyhalogenophenoxycarbonylchloride, while their yields are not satisfactory.

SUMMARY OF THE INVENTION

The present invention is to provide an industrially advantageous process for preparing

8-lankacidine carbamate derivatives having excellent antimicrobial activities.

Thus, the present invention provides a process for preparing a compound represented by the formula [III]:

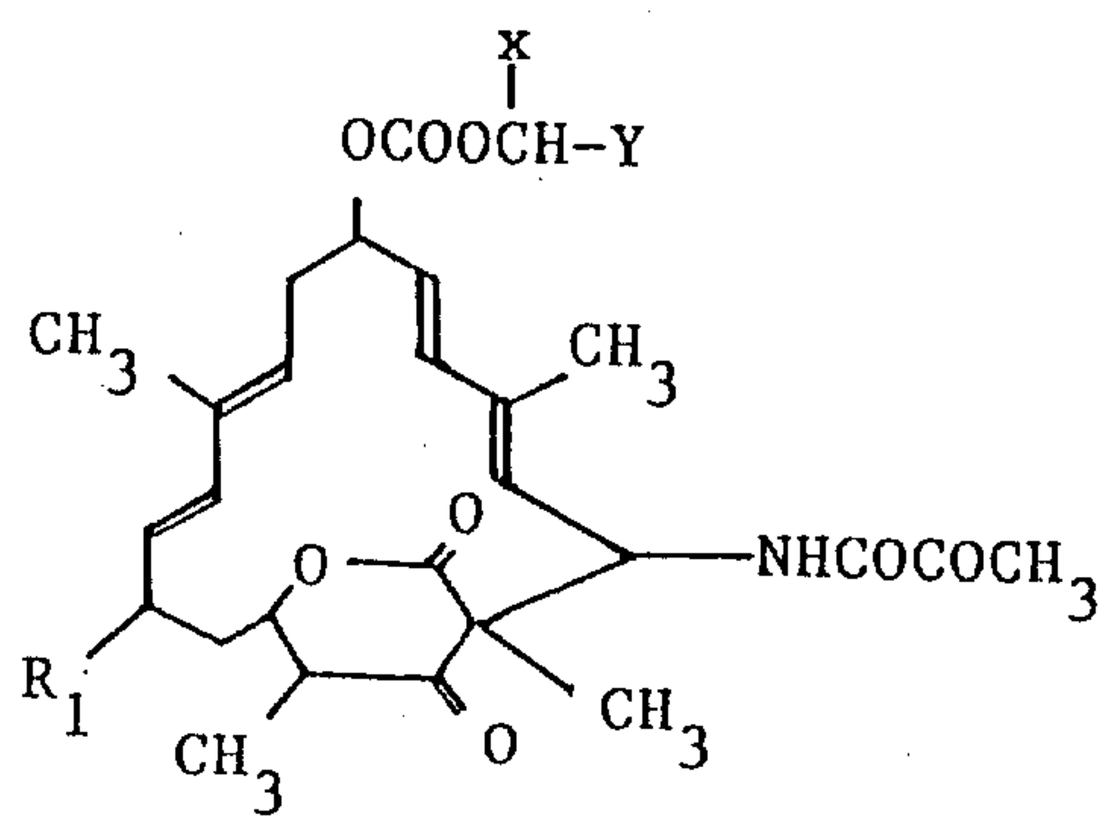


[III]

wherein R_1 is a hydroxy group or an alkanoyloxy group; and R_2 and R_3 are each a hydrogen atom, an optionally substituted lower alkyl group, a cycloalkyl group or a phenyl group; or R_2 and R_3 together with the adjacent nitrogen atom to which they bond form an optionally substituted heterocyclic group,

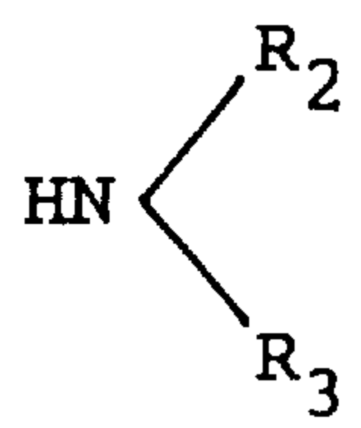
or a salt thereof,

comprising reacting a compound represented by the formula [I]:



[I]

wherein R_1 has the same meaning as above; X is a halogen atom; and Y is a hydrogen atom, a lower alkyl group or a trihalogenoalkyl group,
with a compound represented by the formula [II]:



[II]

wherein R_2 and R_3 have the same meaning as above.

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENT

The term "lower alkyl group" used in the above formula is meant by a straight or branched chain alkyl group having 1-6 carbon atoms. Examples thereof are methyl, ethyl, n-propyl, i-propyl, n-butyl, tert-butyl, n-pentyl and n-hexyl.

The term "cycloalkyl group" used here is meant by a cycloalkyl group having 3-6 carbon atoms. Examples thereof are cyclopropyl, cyclobutyl, cyclopentyl and cyclohexyl.

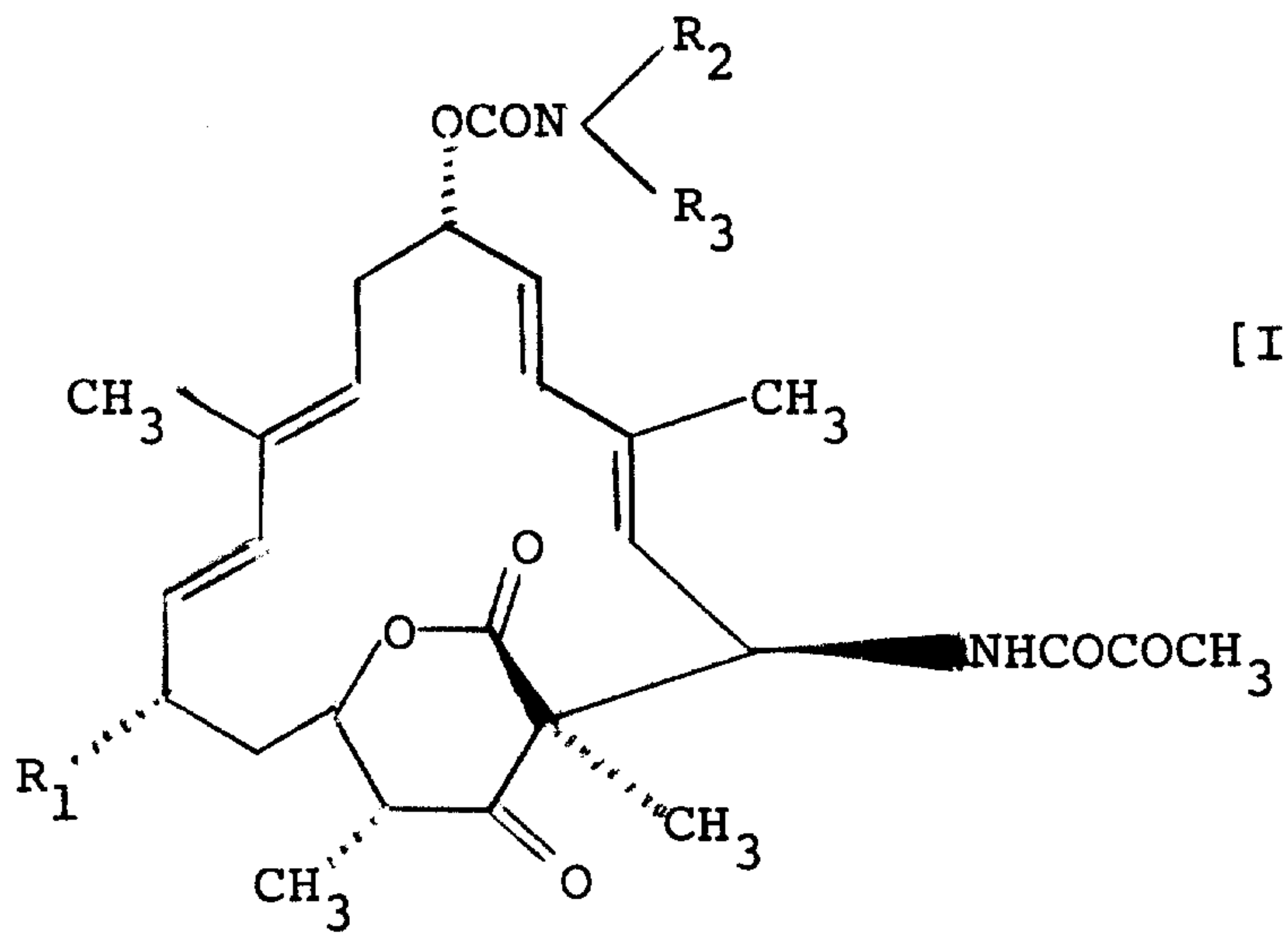
The term "alkanoyloxy group" is meant by an alkanoyloxy having 2-6 carbon atoms. Examples thereof are acetyloxy, propionyloxy, butyryloxy, valeryloxy and hexanoyloxy. Examples of substituents for the optionally substituted lower alkyl group represented by R_2 or R_3 are a hydroxy, an amino, a mono-lower alkylamino (e.g., methylamino or ethylamino), a lower alkoxy (e.g., methoxy or ethoxy), a halogen (e.g., chlorine or bromine) and a heterocycle (e.g., pyridyl).

In the case that R_2 and R_3 together with the adjacent nitrogen atom to which they bond form a heterocyclic group, the heterocyclic group is meant by a 4 - 7 membered ring having at least one nitrogen atom, and optionally an oxygen atom and/or an sulfur atom. Usually preferred is a 5 or 6 membered ring. Examples of the 5 or 6 membered rings are pyrrolidino, piperidino, morpholino and piperazino. Each of these 5 or 6 membered rings may be substituted by an optionally substituted lower alkyl (e.g., methyl, ethyl or hydroxy ethyl), a substituted phenyl (e.g., chlorophenyl) or a heterocycle (e.g., pyridyl). As Y in the formula [I], desirable is methyl, ethyl, propyl, butyl, trichloromethyl or tribromomethyl group, and particularly preferable is methyl or trichloromethyl group. As the halogen atom, desirable is chlorine, bromine or iodine.

According to the present invention, the compound of the formula [III] can be prepared by reacting a compound of the formula [I] with an amine of the formula [II]. The reaction is suitably conducted in an organic solvent optionally coexisted with water. Examples of the organic solvents are dichloromethane, chloroform, 1,2-dichloroethane, tetrahydrofuran, 1,4-dioxane, acetonitrile, ethyl acetate and methyl acetate. The amount of the amine [II] used in this reaction is suitably approximately 1 - 10 mol equivalents, to the compound [I]. The reaction time and reaction temperature vary within approximately 0 - 100°C and approximately 30 minutes to 24 hours, respectively, depending on the sort of the amine used.

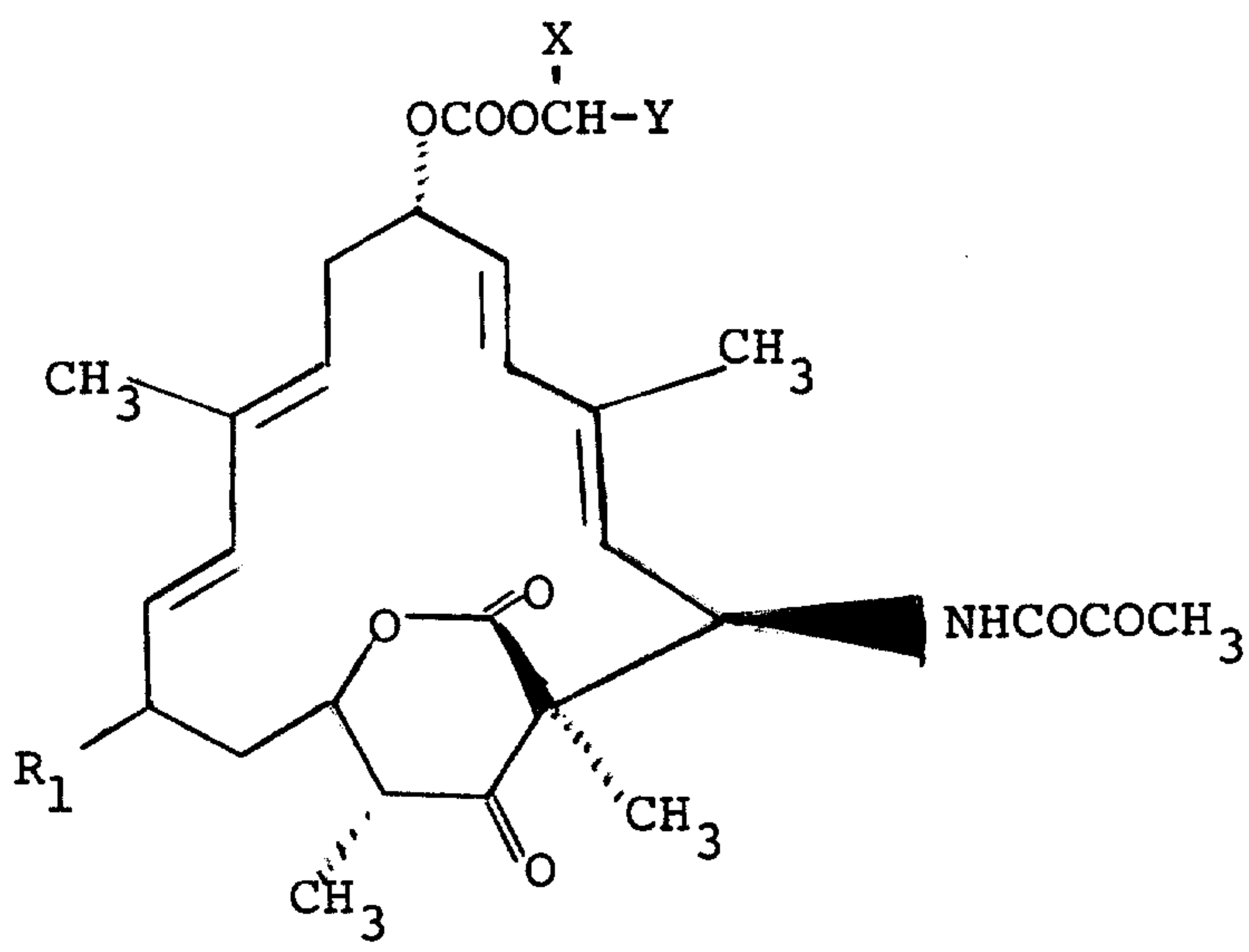
The objective compounds [III] thus obtained can be isolated and purified by a well known technique such as concentration, solvent extraction, chromatography, crystallization or recrystallization. In case where the compound has a basic group such as an amino group or a substituted amino group as R_2 and R_3, the basic group may form an acid-addition salt. Examples of such acid-addition salts are the hydrochloride, hydrobromide, hydroiodide, nitrate, sulfate, phosphate, acetate, benzoate, maleate, fumarate, succinate, tartrate, citrate, methanesulfonate, benzenesulfonate and the like.

Among the compounds [III], preferred are those of the formula:



[III']

(wherein the symbols have the same meanings as above) which may be produced using starting compounds of the formula:



[I']

The resultant compounds [III] exhibit a potent antimicrobial activity on Gram-positive bacteria. As well, they exhibit an antimicrobial activity on some sorts of Gram-negative bacteria. Also, they exhibit a potent antimicrobial activity on not only macrolide-resistant Staphylococcus aureus but also methicillin- and cephem-resistant Staphylococcus aureus (MRSA). Further, they possess an antimicrobial activity even on mycoplasma and swine-dysentery bacillus while showing low toxicity.

As described above, the compounds [III] possess an excellent antimicrobial activity besides low toxicity. Therefore, they can be used as an antimicrobial agent for curing microbism of animals such as chickens, sheep, dogs, cats, rabbits, pigs, bovines, horses, monkeys and human beings, or for curing mycoplasma infectious diseases thereof. They can also be used as a feed additive for preventing microbism or promoting growth of the animals.

A daily dose of the compound [III] or its salt differs depending on its administration manner, the kind of animals to be administrated and the administration purpose, and is usually about 0.001 - 1000 mg/Kg, preferably about 0.1 - 300 mg/Kg.

The compound [III] or its pharmaceutically acceptable salt can be orally administered in the form of preparations such as tablets, granules, capsules and drops which can be prepared in admixture of carriers, diluents and other conventional agents in accordance with conventional techniques; and alternatively can be parenterally administered in the form of injections which can be prepared by conventional means or together with sterile carriers.

The above oral preparation, for example, tablets can be prepared by using a binder (e.g., hydroxypropylcellulose, hydroxypropylmethylcellulose, macrogol or the like), a disintegrator (e.g., starch, calcium carboxymethylcellulose or the like), a diluent (e.g., lactose, starch or the like) or a lubricant (e.g., magnesium stearate, talc or the like) can be appropriately mixed.

Further, the above parenteral preparation, for example, injection can be prepared by appropriately mixing with an isotonization agent (e.g., glucose, D-sorbitol, D-mannitol, sodium chloride or the like), a preservative (e.g., benzyl alcohol, chlorobutanol, methyl para-hydroxybenzoate, propyl para-hydroxybenzoate or the like), a buffer (e.g., phosphate buffer solution, sodium acetate buffer solution or the like).

Hereinafter, the invention will be more fully described in conjunction with Reference Example and Examples, to which the invention are not limited.

Elution in column chromatography in the Reference Example and Examples was conducted while monitoring with TLC (Thin Layer Chromatography). In the TLC monitoring, the TLC plate used was $60F_{254}$ manufactured by Merck Co., the developing solvent was the same one as used for eluting in the column chromatography, and the detection was conducted with a UV detector. The silica gel for the column was Kieselgel* 60 (230 - 400 mesh) manufactured by Merck Co.

NMR spectra were measured using tetramethylsilane as an internal or external standard in a spectrometer of XL-100A (100MHz), EM360 (60MHz), EM390 (90MHz) or T_{60} (60MHz) type, and all s values are expressed in ppm. The value shown in ( ) for a mixed solvent is a mixing ratio in volume of constituent solvents. The symbols in the Reference Example and Examples mean as follows:

s : singlet
d : doublet
t : triplet
q : quartet
ABq : AB type quartet

*Trade-mark

dd : double doublet

m : multiplet

br. : broad

J : coupling constant

Reference Example 1

Lankacidine A 8-(1-chloroethyl)carbonate

Lankacidine A (50 g) was dissolved in 950 ml of dichloromethane, to which a solution of 19 g of 1-chloroethyl chloroformate [see Synsesis, 627 (1986)] in 40 ml of dichloromethane was added under ice cooling. To the mixture was dropwise added 10.3 g of pyridine. Then, the mixture was stirred at room temperature for an hour, mixed with 2 ml of 1- chloroethyl chloroformate and 1.5 ml of pyridine sequentially and stirred for an hour. The reaction mixture was washed with 1N-HCl, water and then dilute sodium bicarbonate solution, and dried over $MgSO_4$. The mixture was evaporated under reduced pressure to remove its solvent, and the resulting residue was crystallized from 500 ml of a mixture of ether-hexane (1 : 1) to obtain 53.7 g of the title compound.

NMR (90 MHz, $CDCl_3$)δ :

1.32 (d, 3H, J=7Hz), 1.40 (s, 3H), 1.57 (s, 3H), 1.82 (d, 3H, J=6Hz), 1.93 (s, 3H), 2.05 (s, 3H), 2.2-2.7 (m, 5H), 2.47 (s, 3H), 4.42 (dt, 1H, J= 3 & 12Hz), 4.47 (d, 1H, J = 11Hz), 4.8-5.2 (m, 1H), 5.3-6.1 (m, 6H), 6.30 (d, 1H, J = 15Hz), 6.40 (q, 1H, J = 6Hz), 8.10 (d, 1H, J = 11Hz)

Example 1

Lankacidine A 8-(4-methylpiperazino)carboxylate

Lankacidine A 8-(1-chloroethyl)carbonate (6 g) was dissolved in 40 ml of dichloromethane, to which a solution of 6 g of N-methylpiperazine in 20 ml of dichloromethane was dropwise added under ice cooling. After stirring for 40 min., the reaction mixture was washed with 1N-HCl to remove excess amine, subsequently with a dilute sodium bicarbonate solution and water and dried over $MgSO_4$. The mixture was evaporated under reduced pressure to remove the solvent. The resulting residue was subjected to silica gel column chromatography (silica gel: 300 g) eluting with chloroform-methanol (30 : 1). The fractions including the objective compound were collected and concentrated. The resulting oily substance was then crystallized from a mixture of ether-hexane (1 : 1) to obtain 4.7 g of the title compound.

Melting point: 203 - 207°C (decomp.)

Elemental analysis for $C_{33}H_{45}N_3O_9 \cdot \frac{1}{2}H_2O$:

Calcd.: C, 61.83; H, 7.33; N, 6.51

Found: C, 61.71; H, 7.25; N, 6.79

NMR (90 MHz, $CDCl_3$) δ :

1.32 (d, 3H, J=7Hz), 1.38 (s, 3H), 1.55 (s, 3H), 1.92 (s, 3H), 2.03 (s, 3H), 2.1-2.7 (m, 9H), 2.30 (s, 3H), 2.46 (s, 3H), 3.4-3.7 (m, 4H), 4.43 (d.d, 1H, J = 3 & 12Hz), 4.72 (d, 1H, J = 11Hz), 4.2-5.2 (m, 1H), 5.2-5.9 (m, 6H), 6.32 (d, 1H, J = 15Hz), 8.08 (d, 1H, J = 11Hz)

Example 2

Lankacidine A 8-chloromethylcarbonate (300 mg, prepared by the process disclosed in Japanese Published Unexamined Patent Application No. 240687/1987) was dissolved in 10 ml of tetrahydrofuran, to which 100 mg of N-methylpiperazine was added at room temperature. After stirring for 3 hours, the reaction mixture was mixed with 50 ml of ethylacetate and washed with 60 ml of saturated aqueous sodium chloride solution. The resulting organic layer was dried over $MgSO_4$ and concentrated under reduced pressure. The residue was subjected to silica gel column chromatography (silica gel : 60 g) eluting with chloroform-methanol (30 : 1). The fractions including the objective compound were collected and concentrated. The residue was then crystallized from a mixture of ether-hexane (1 : 1) to obtain 180 mg of the compound identical to that prepared in Example 1.

Example 3

Lankacidine A 8-iodomethylcarbonate (342 mg, prepared by the process disclosed in Japanese Published Unexamined Patent Application No. 240687/1987) was dissolved in 10 ml of tetrahydrofuran, to which 120 mg of N-methylpiperazine was added at room temperature. After stirring for 30 min. at room temperature, the reaction mixture was mixed with 50 ml of ethylacetate and washed with 60 ml of saturated aqueous sodium chloride solution. The resulting organic layer was dried over $MgSO_4$ and evaporated to remove the solvent. The residue was subjected to silica gel column chromatography (silica gel: 60 g) eluting with chloroform-methanol (30 : 1). The fractions including the objective compound were collected and concentrated. The residue was then crystallized from a mixture of ether-hexane (1 : 1) to obtain 193 mg of the compound identical to that prepared in Example 1.

Examples 4 - 13

Lankacidine A 8-(1-chloroethyl)carbonate was reacted with various amines by the method as described in Example 1 to obtain the compounds shown in Table 1.

Table 1

Lankacidine A 8-carbonate Derivatives

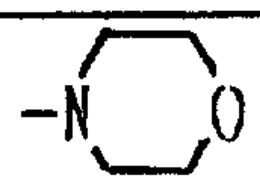
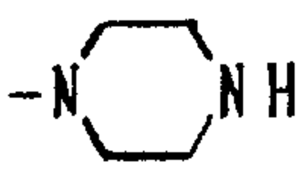
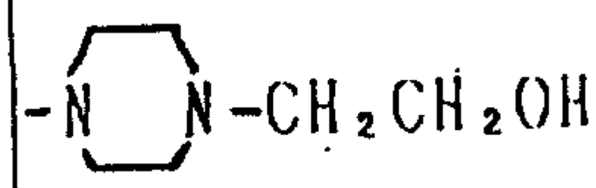
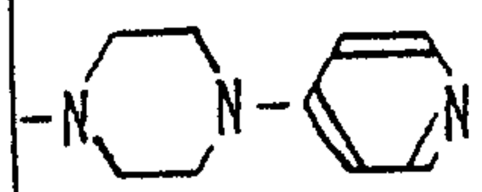
Example No.	$-N\langle{}^{R^2}_{R^3}$	NMR: 11-Me, $CO\underline{CH_3}$ 8-H, etc.	mp (℃)	yield (%)
4	$-NHCH_3$	1.53(s), 2.44(s), 4.98(m), 2.77(d, J=5Hz, $NH\underline{CH_3}$)	–	51
5	$-NH(CH_2)_2CH_3$	1.53(s), 2.43(s), 4.98(m), 0.89(t, J=7Hz, $-CH_2CH_2\underline{CH_3}$)	–	75
6	$-NHC_6H_5$	1.55(s), 2.44(s), 5.05(m), 6.9~7.5(m, $-C_6H_5$)	231-232	62
7	$-NHCH\langle{}^{CH_3}_{CH_3}$	1.56(s), 2.43(s), 4.95(m), 1.13(d, J=7Hz, $-CH(\underline{CH_3})_2$)	198-200	90
8	$-NHCH_2-$ 	1.53(s), 2.44(s), 5.01(m), 4.47(d, J=5Hz, $NH\underline{CH_2}-$)	192-194	77
9	$-NHCH_2-$ 	1.53(s), 2.45(s), 5.01(m), 4.36(d, J=6Hz, $NH\underline{CH_2}-$)	222-224	78
10		1.56(s), 2.46(s), 5.02(m)	223-225	74
11		1.54(s), 2.45(s), 4.97(m) ~2.3(br. piperizine)	–	70

Table 1 (continued)

Example No.	$-N<^{R^2}_{R^3}$	NMR: 11-Me, $COCH_3$, 8-H, etc.	mp (℃)	yield (%)
12		1.55(s), 2.45(s), 4.99(m), 3.62(t, J=6Hz, CH_2CH_2OH)	173-175	82
13		1.54(s), 2.43(s), 5.01(m), 6.55-6.75 & 8.15-8.14(pyridine)	175-177	71

Example 14

Lankacidine C 8-(4-methylpiperazino)carboxylate

Lankacidine C 8-iodomethylcarbonate (300 mg, prepared by the process disclosed in Japanese Published Unexamined Patent Application No. 240687/1987) was dissolved in 10 ml of tetrahydrofuran, to which 110 mg of N-methylpiperazine was added at room temperature. After stirring for 30 min. at room temperature, the reaction mixture was mixed with 50 ml of ethylacetate and washed with 60 ml of saturated aqueous sodium chloride solution. The resulting organic layer was dried over $MgSO_4$ and evaporated under reduced pressure to remove the solvent. The residue was subjected to silica gel column chromatography (silica gel: 60 g) eluting with chloroform-methanol (30 : 1). The fractions including the objective compound were collected and concentrated under reduced pressure to obtain 230 mg of the title compound.

Melting point: 201 - 203°C

Elemental analysis for $C_{31}H_{43}N_3O_8 \cdot 3/2H_2O$:

Calcd.: C, 60.77; H, 7.57; N, 6.86

Found: C, 60.82; H, 7.30; N, 6.67

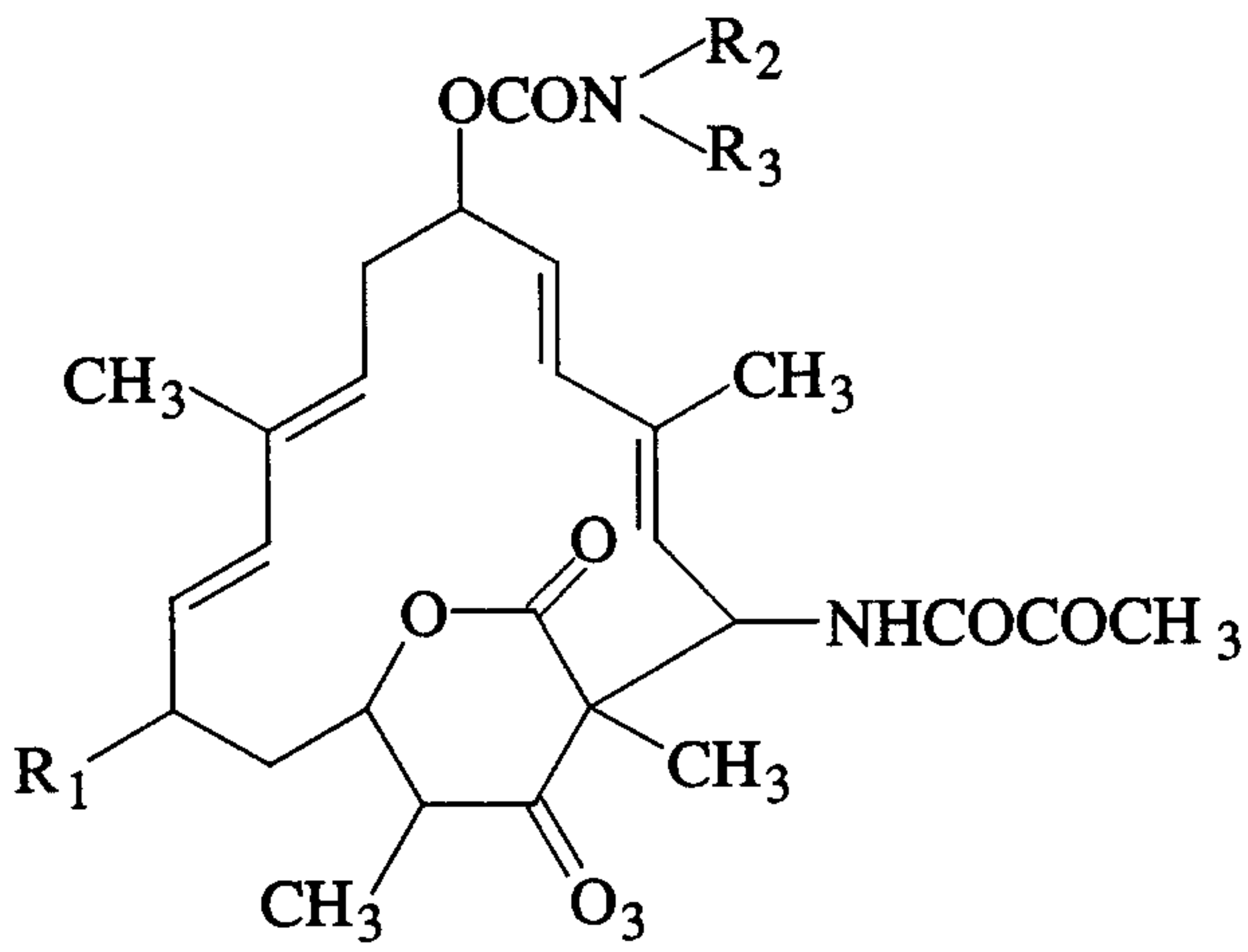
NMR (90 MHz, $CDCl_3$) δ :

1.25 (d, 3H, J=6Hz), 1.38 (s, 3H), 1.54 (s, 3H), 1.92 (s, 3H), 2.1-2.7 (m, 9H), 2.46 (s, 3H), 3.4-3.7 (m, 4H), 4.2-4.6 (m, 2H), 4.70 (d, 1H, J = 11Hz), 4.8-6.0 (m, 6H), 6.17 (d, 1H, J=15Hz), 8.07 (d, 1H, J = 11Hz)

According to the present invention, 8-lankacidine carbamate derivatives having excellent antimicrobial activities can be efficiently prepared using stable reactants of low prices.

THE EMBODIMENTS OF THE INVENTION IN WHICH AN EXCLUSIVE PROPERTY OR PRIVILEGE IS CLAIMED ARE DEFINED AS FOLLOWS:

1. A process for preparing a compound represented by the formula [III]:



[III]

(wherein:

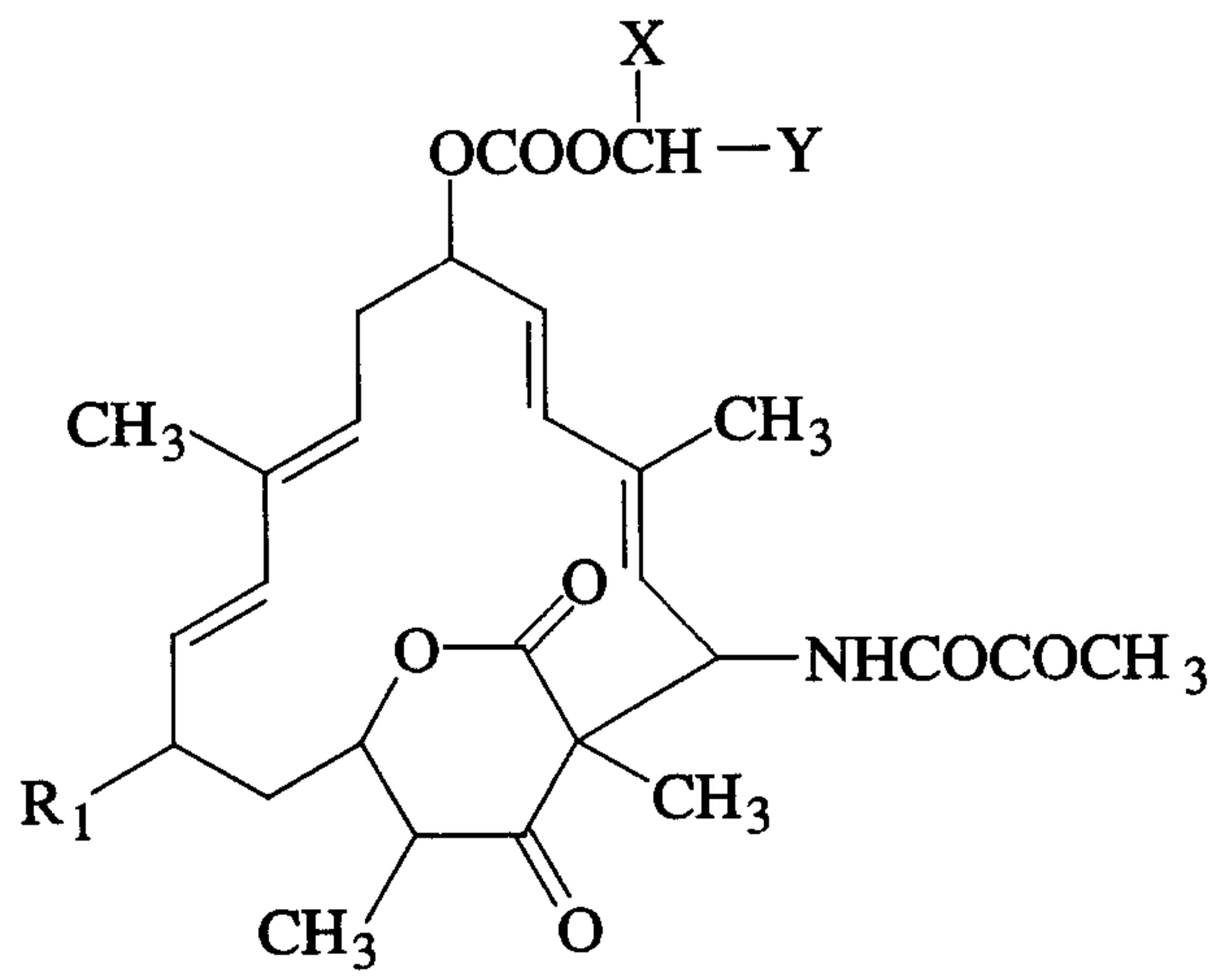
R_1 is a hydroxyl group or an alkanoyloxy group having 2-6 carbon atoms; and

R_2 and R_3 are each (1) a hydrogen atom, (2) a lower alkyl group which may optionally be substituted by hydroxyl, amino, mono-lower alkylamino, lower alkoxy, halogen or pyridyl, (3) a cycloalkyl group having 3-6 carbon atoms or (4) a phenyl group; or

R_2 and R_3 together with the adjacent nitrogen atom to which they are bonded form a 4-7 ring member heterocyclic group which has at least one nitrogen atom and may optionally have an oxygen atom or a sulfur atom or both of them and which may optionally by substituted by methyl, ethyl, hydroxyethyl, chlorophenyl or pyridyl) or a salt thereof, which comprises

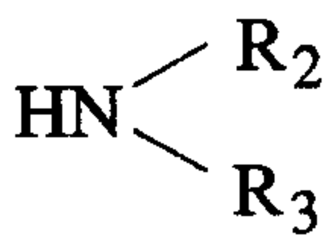
B

reacting a compound represented by the formula [I]:



[I]

(wherein R_1 has the same meaning as above; X is a halogen atom; and Y is a hydrogen atom, a lower alkyl group or a trihalogenoalkyl group), with a compound represented by the formula [II]:



[II]

(wherein R_2 and R_3 have the same meaning above).

2. The process of claim 1 in which the reaction is conducted in an organic solvent selected from the class consisting of dichloromethane, chloroform, 1,2-dichloroethane, tetrahydrofuran, 1,4-dioxane, acetonitrile, ethyl acetate and methyl acetate.

B

3. The process of claim 1 or 2, wherein R_1 is a hydroxyl or acetoxy group, and either one of R_2 and R_3 is a hydrogen atom and the other is a lower alkyl group optionally substituted by pyridyl.

4. The process of claim 3 in which the lower alkyl group in the lower alkyl group optionally substituted by pyridyl is methyl, ethyl, n-propyl, i-propyl, n-butyl, i-butyl, tert-butyl, n-pentyl or n-hexyl.

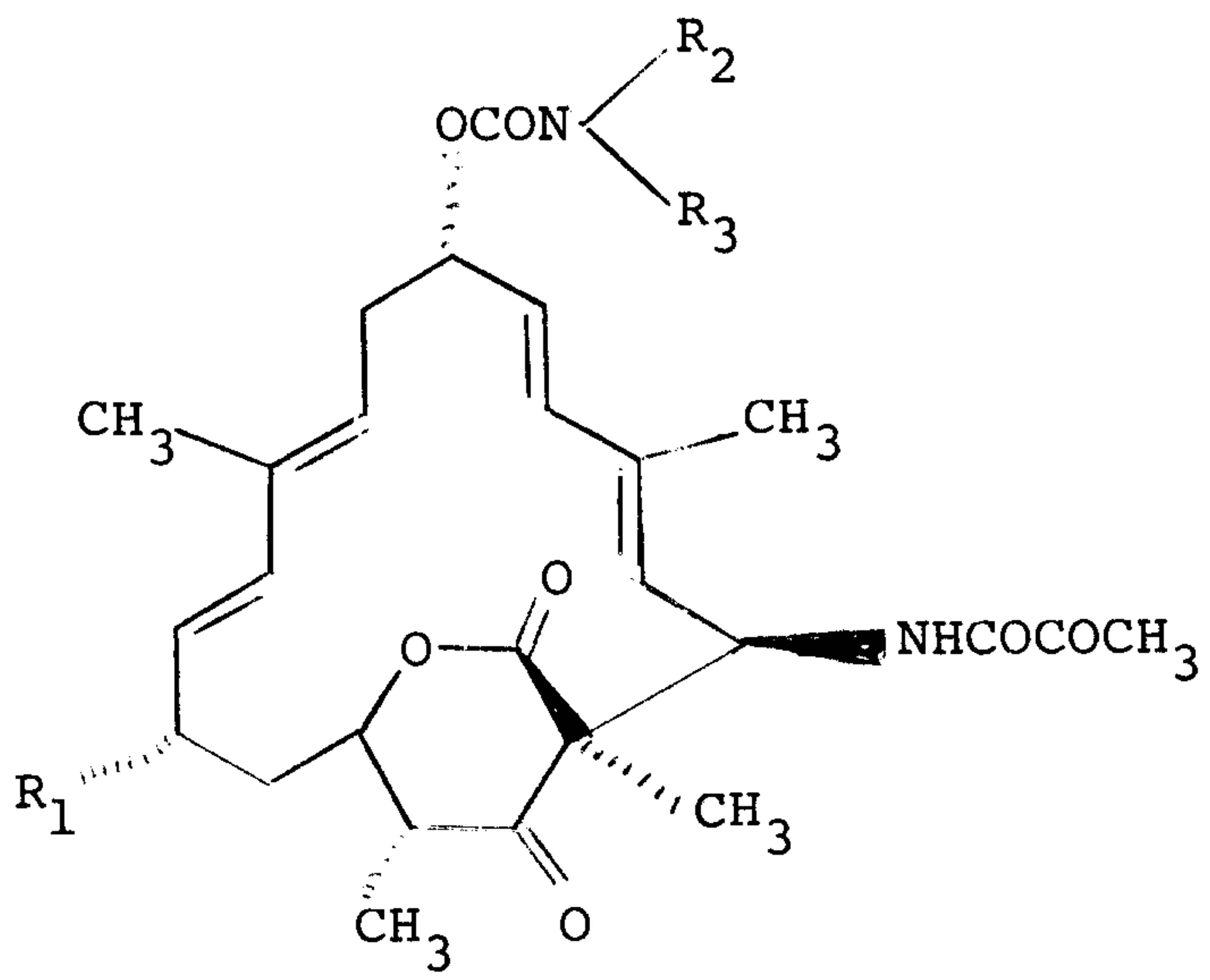
5. The process of claim 1 or 2, wherein R_1 is a hydroxyl or acetoxy group, and either one of R_2 and R_3 is a hydrogen atom and the other is a phenyl group.

6. The process of claim 1, wherein R_1 is a hydroxyl or acetoxy group, and R_2 and R_3 together with the adjacent nitrogen atom to which they are bonded are a 5 or 6 membered heterocyclic group which has at least one nitrogen atom and may optionally have an oxygen atom, a sulfur atom or both of them and which may optionally be substituted by hydroxyethyl or pyridyl.

7. The process of claim 6 in which the 5 or 6 membered heterocyclic group in the 5 or 6 membered heterocyclic group optionally substituted by hydroxyethyl or pyridyl is pyrrolidino, piperidino, morpholino or piperazino.

B

8. A process for preparing a lankacidine carbamate compound represented by the formula:



[III']

[wherein:

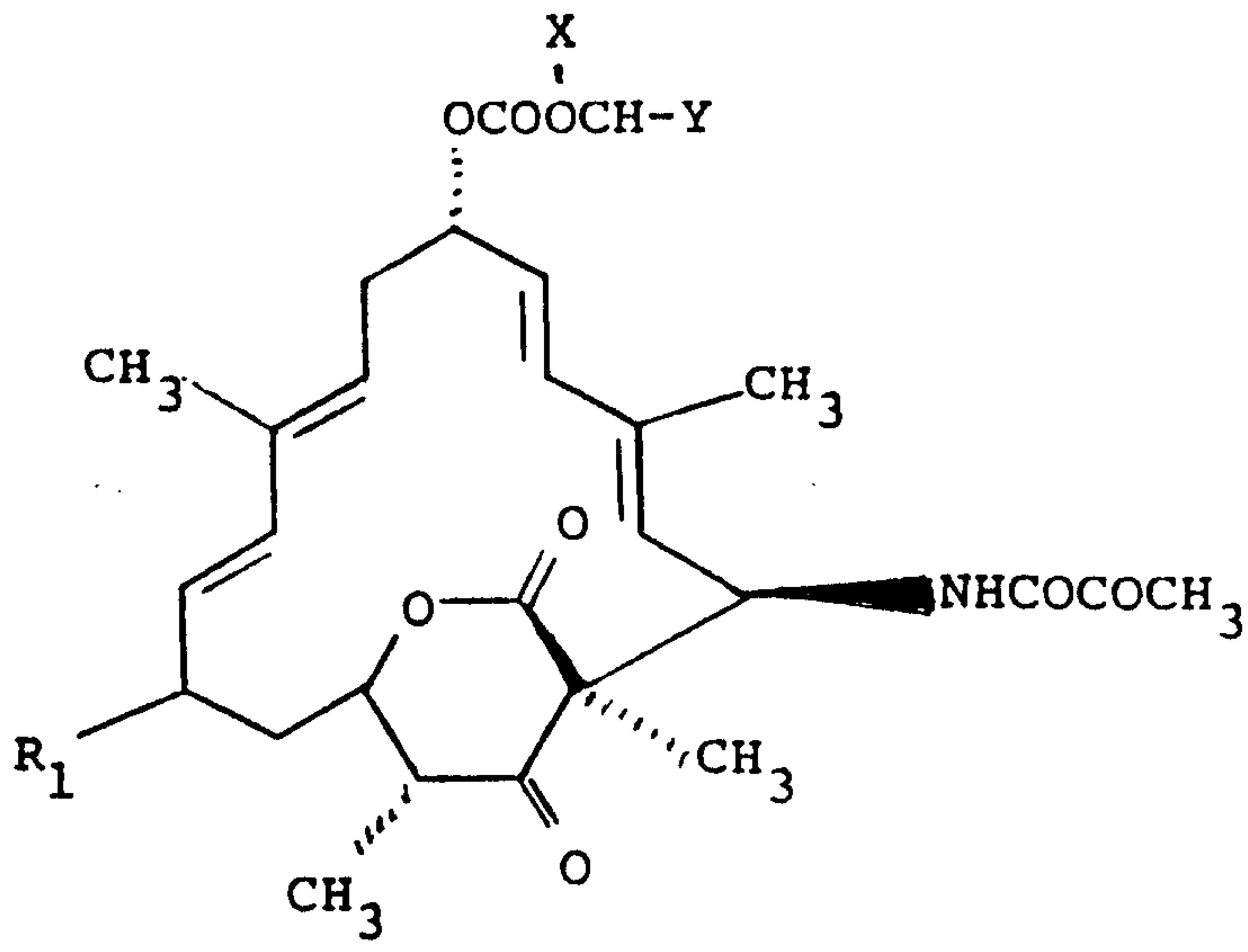
R_1 is a hydroxyl group or a C_{2-6}alkanoyloxy group; and

R_2 and R_3 are each a hydrogen atom, a C_{1-6}alkyl group (which may have a substituent selected from the class consisting of hydroxy, amino, mono-lower alkylamino, lower alkoxy, halogen and pyridyl), C_{3-6}cycloalkyl or phenyl, or

R_2 and R_3 together with the adjacent nitrogen atom to which they are attached form a heterocyclic group which is a member selected from the class consisting of pyrrolidino, piperidino, morpholino and piperazino and may have a substituent selected from the class consisting of lower alkyl, hydroxy-lower alkyl, chlorophenyl and pyridyl]

or a salt thereof, which process comprises:

reacting a compound of the formula:



[I']

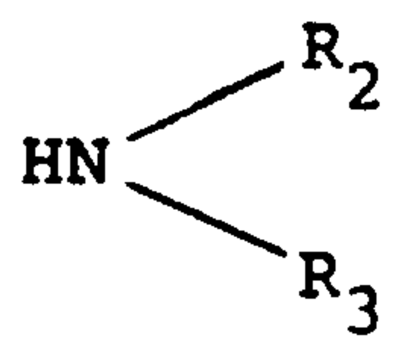
(wherein:

R_1 has the same meanings as above;

X is a halogen atom; and

Y is a hydrogen atom, a lower alkyl group or a trihalogenoalkyl group)

with a compound of the formula:



[II]

(wherein R_2 and R_3 have the same meanings as above).

9. The process of claim 8, wherein

X is chlorine, bromine or iodine; and

Y is hydrogen, lower alkyl, trichloromethyl or tribromomethyl.

D

10. The process of any one of claims 1 to 9, wherein the reaction is conducted using the compound of the formula [II] in an amount of 1-10 mol equivalents relative to the compound of the formula [I] or [I'] at a temperature of 0-100°C for a period of 30 minutes to 24 hours.

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PATENT AGENTS

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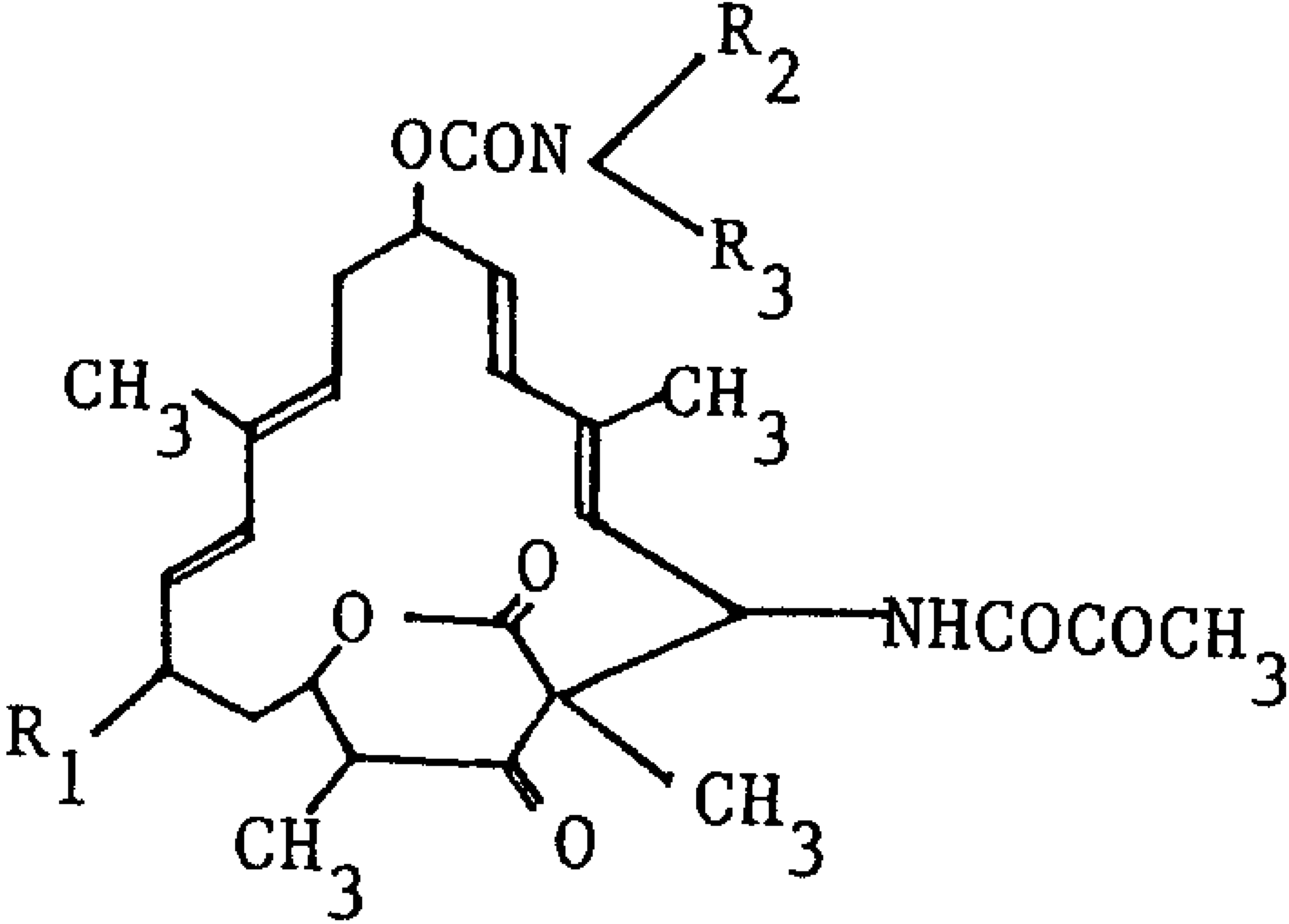
R2
OCON
R3
CH3
CH3
O
O
NHCOCOCH3
R1
CH3
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