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(71) Applicant *(for all designated States except US):* BEECHAM GROUP P.L.C. [GB/GB]; Beecham House, Great West Road, Brentford, Middlesex TW8 9BD (GB).

(72) Inventor; and
(75) Inventor/Applicant *(for US only) :* MILNER, Peter, Henry [GB/GB]; 20 Goose Green Close, Horsham, Sussex (GB).

(74) Agents: HESKETH, Alan; Beecham Pharmaceuticals, Great Burgh, Yew Tree Bottom Road, Epsom, Surrey KT18 5XQ (GB) et al.

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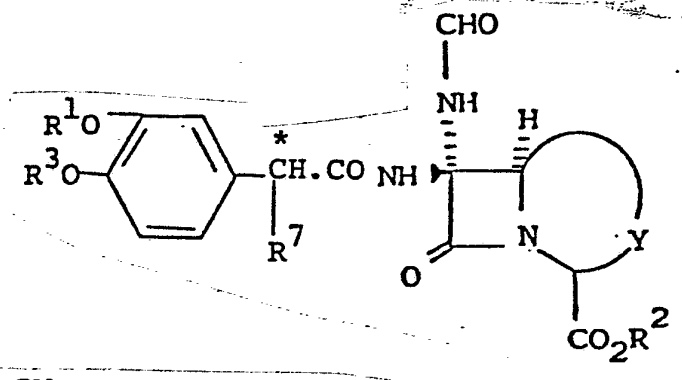
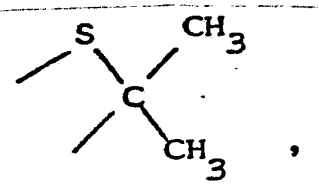


(I)



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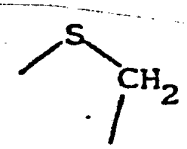

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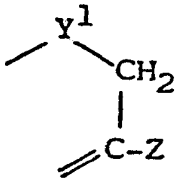

(II)

(57) Abstract

A compound of formula (I) or a salt thereof, wherein R^1 and R^3 are independently an *in vivo* hydrolysable group, or R^1 and R^3 together form an *in vivo* hydrolysable group, provided that R^1 and R^3 are not both C_{1-6} alkylcarbonyl; R^7 is a hydroxyl, carboxylic acid group or lower alkyl or phenyl, tolyl or indanyl ester thereof, amino or a substituted amino group; R^2 is hydrogen or a readily removable carboxyl protecting group; and Y is, formula (II), wherein Y^1 is oxigen, sulphur or $-CH_2-$ and Z represents hydrogen, halogen, or an organic group such as C_{1-4} alkoxy, $-CH_2Q$ or $-CH=CH-Q$ wherein Q represents hydrogen, halogen, hydroxy, mercapto, cyano, carboxy, carbamoyloxy, carboxylic ester, C^{1-4} alkyloxy, acyloxy, aryl, a heterocyclyl group bonded via carbon, a heterocyclylthio group or a nitrogen containing heterocyclic group bonded via nitrogen.

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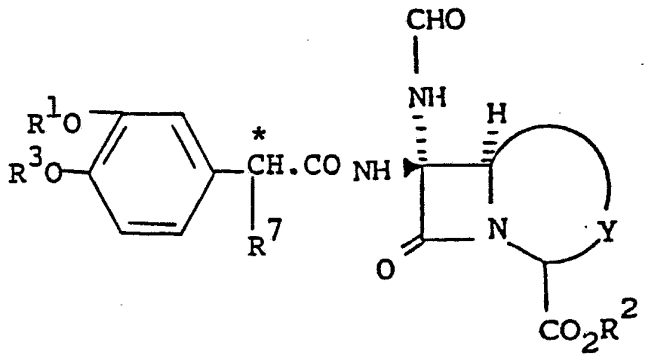
β-LACTAM ANTIBACTERIAL AGENTS

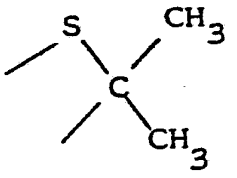
This invention relates to a class of novel β-lactam derivatives, which have antibacterial activity and are of value in the treatment of infections in animals especially mammals including man caused by a wide range of organisms, particularly Gram-negative organisms. The invention also relates to a process for the preparation of such compounds, intermediates for use in the preparation of the compounds and to pharmaceutical compositions comprising the antibacterially active compounds.

According to the present invention there is provided a compound of formula (I) or a salt thereof.



(I)

wherein R^1 and R^3 are independently an in vivo hydrolysable group, or R^1 and R^3 together form an in vivo hydrolysable group, provided that R^1 and R^3 are not both C_{1-6} alkylcarbonyl; R^7 is a hydroxyl, carboxylic acid group or lower alkyl or phenyl, tolyl or indanyl ester thereof, amino or a substituted amino group; R^2 is hydrogen or a readily removable carboxyl protecting group; and Y is:



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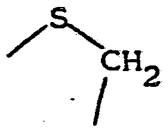

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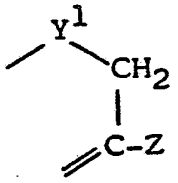


;

wherein Y^1 is oxygen, sulphur or $-CH_2-$ and Z represents hydrogen, halogen, or an organic group such as C_{1-4} alkoxy, $-CH_2Q$ or $-CH=CH-Q$ wherein Q represents hydrogen, halogen, hydroxy, mercapto, cyano, carboxy, carbamoyloxy, carboxylic ester, C_{1-4} alkyloxy, acyloxy, aryl, a heterocyclyl group bonded via carbon, a heterocyclylthio group or a nitrogen containing heterocyclic group bonded via nitrogen.

When used herein the term "halogen" unless otherwise defined is suitably fluorine, chlorine, bromine, and iodide, preferably chlorine and bromine.

When used herein the term "carboxylic ester" unless otherwise defined suitably includes C_{1-6} alkyl esters.

When used herein the term "acyloxy" unless otherwise defined suitably includes C_{1-6} alkylcarbonyloxy groups.

When used herein the term "aryl" unless otherwise defined suitably includes phenyl and naphthyl, preferably phenyl, optionally substituted with up to five halogen, C_{1-6} alkyl, C_{1-6} alkoxy, halo(C_{1-6}) alkyl, hydroxy, amino, carboxy, C_{1-6} alkoxycarbonyl, or C_{1-6} alkoxycarbonyl-(C_{1-6})-alkyl groups.

When used herein the term "heterocyclyl" unless otherwise defined suitably includes single or fused rings comprising up to four hetero atoms in the ring selected from oxygen, nitrogen and sulphur and optionally substituted with up to three halogen, C_{1-6} alkyl, C_{1-6} alkoxy, halo-(C_{1-6})-alkyl, hydroxy, amino, carboxy, C_{1-6} alkoxycarbonyl, C_{1-6} alkoxycarbonyl(C_{1-6}) alkyl, aryl or oxo groups.

The group R^1 and R^3 may represent, for example C_{1-6} alkoxycarbonyl, such as methoxycarbonyl or ethoxycarbonyl. Preferably R^1 and R^2 are the same group.

When R^1 and R^3 are joined they may represent C_{1-6} alkylene, in particular methylene or ethylene. When R^1 and R^3 together represent methylene, the substituent on the phenyl ring becomes $-O.CH_2.O-$.

The compounds of the present invention may contain both an amino group and/or a carboxyl group and may, therefore, exist as the zwitterion or may form salts with suitable acids or bases.

The formamido group can exist in two preferred conformations, those wherein the hydrogen atoms of the -NH-CHO are, cis- or trans-, of which the cis-conformation normally predominates.

Suitably Y is $-S-C(CH_3)_2-$, $-S-CH_2-$,
$-S-CH_2-C(CH_2Q')=$; or
$-O-CH_2-C(CH_2Q')=$,
wherein Q' represents hydrogen, halogen, hydroxy, mercapto, cyano, carboxy, carboxylic ester, C_{1-4} alkyloxy, acyloxy or heterocyclylthio group.

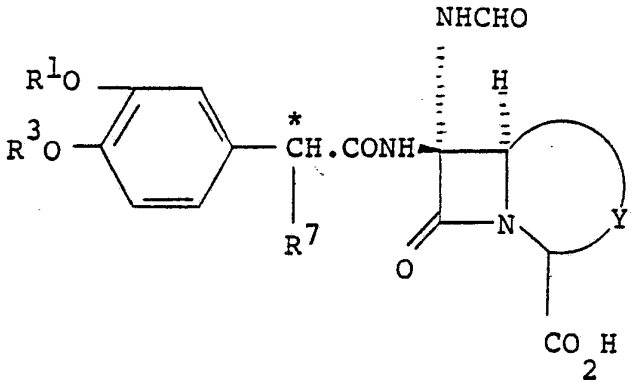
Preferred values for Y in the compounds of formula (I) are $-S-C(CH_3)_2-$ and $-S-CH_2-C(CH_2Q)=$, ie when the compound of formula (I) is a derivative of a penicillin and cephalosporin.

A particularly preferred value for Y is $-S-C(CH_3)_2-$.

A further preferred value for Y is $-S-CH_2-CZ=$ wherein Z is as hereinbefore defined.

Those compounds of the formula (I) wherein R^2 is a readily removable carboxyl protecting group or a non-pharmaceutically acceptable salt are primarily useful as intermediates in the preparation of compounds of the formula (I) wherein R^2 is a free carboxyl group or a pharmaceutically acceptable salt thereof. Also included within the readily removable carboxyl protecting groups R^2 are pharmaceutically acceptable in vivo hydrolysable ester groups.

From the forgoing it will be realised that suitable antibacterially active compounds are those of formula (II) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof.

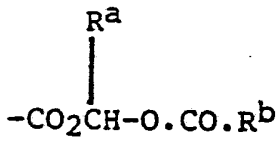


(II)

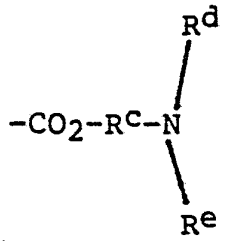
wherein R^1, R^3, Y and R^7 are as defined with respect to formula (I).

Since the β-lactam antibiotic compounds of the present invention are intended for use in pharmaceutical compositions it will readily be understood that they are each provided in substantially pure form, for example at least 50% pure, more suitably at least 75% pure and preferably at least 95% pure (% are on a wt/wt basis). Impure preparations of the compounds may be used for preparing the more pure forms used in the pharmaceutial compositions. Although the purity of intermediate compounds of the present invention is less critical it will readily be understood that the substantially pure form is preferred as for the β-lactam antibiotic compounds. Preferably, whenever possible, the compounds of the present invention are obtained in crystalline form.

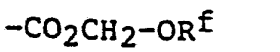
Examples of suitable pharmaceutically acceptable <u>in vivo</u> hydrolysable ester groups include those which break down readily in the human body to leave the parent acid or its salt. Suitable ester groups of this type include those of part formula (i), (ii) and (iii):



(i)



(ii)



(iii)

wherin R^a is hydrogen, methyl, or phenyl, R^b is C_{1-6} alkyl, C_{1-6} alkoxy or phenyl; or R^a and R^b together form a 1,2-phenylene group optionally substituted by one or two methoxy groups; R^c represents C_{1-6} alkylene optionally substituted with a methyl or ethyl group - R^d and R^e independently represent C_{1-6} alkyl; R^f represents C_{1-6} alkyl. Examples of suitable in vivo hydrolysable ester group include for example acyloxyalkyl groups such as acetoxymethyl, pivaloyloxymethyl, α-acetoxyethyl and α-pivaloyloxyethyl groups; alkoxycarbonyloxyalkyl groups, such as ethoxycarbonyloxymethyl and α-ethoxycarbonyloxyethyl; dialkylaminoalkyl especially di-loweralkylamino alkyl groups such as dimethylaminomethyl, dimethylaminoethyl, diethylaminomethyl or diethylaminoethyl; lactone groups such as phthalidyl and dimethoxyphthalidyl; and esters linked to a second β-lactam antibiotic or to a β-lactamase inhibitor.

Suitable readily removable carboxyl protecting groups for the group $-CO_2R^2$ in formula (I) include ester derivatives of the carboxylic acid. The derivative is preferably one which may readily be cleaved.

Suitable ester-forming carboxyl-protecting groups are those which may be removed under conventional conditions. Such groups for R^2 include benzyl, p-methoxybenzyl, benzoylmethyl, p-nitrobenzyl, 4-pyridylmethyl, 2,2,2-trichloroethyl, 2,2,2-tribromoethyl, t-butyl, t-amyl, allyl, diphenylmethyl, triphenylmethyl, adamantyl, 2-benzyloxyphenyl, 4-methylthiophenyl, tetrahydrofur-2-yl,

tetrahydropyran-2-yl, pentachlorophenyl, acetonyl, p-toluenesulphonylethyl, methoxymethyl, a silyl, stannyl or phosphorus-containing group, an oxime radical of formula -N=CHR° where R° is aryl or heterocyclic, or an in vivo hydrolysable ester radical such as defined above.

The carboxyl group may be regenerated from any of the above esters by usual methods appropriate to the particular R^2 group, for example, acid - and base - catalysed hydrolysis, or by enzymically -catalysed hydrolysis, or by hydrogenolysis.

Suitable pharmaceutically acceptable salts of the carboxy group of the compound of formula (I) include metal salts eg aluminium, alkali metal salts such as sodium or potassium, alkaline earth metal salts such as calcium or magnesium and ammonium or substituted ammonium salts, for example those with lower alkylamines such as triethylamine, hydroxy-lower alkylamines such as 2-hydroxyethylamine, bis-(2-hydroxyethyl)-amine or tris-(2-hydroxyethyl)-amine, cycloalkylamines such as dicyclohexylamine, or with procaine, dibenzylamine, N,N-dibenzylethylenediamine, 1-ephenamine, N-ethylpiperidine, N-benzyl-β-phenethylamine, dehydroabietylamine, N,N'-bisdehydroabietylamine, ethylenediamine, or bases of the pyridine type such as pyridine, collidine or quinoline, or other amines which have been used to form salts with known penicillins and cephalosporins. Other suitable salts include the lithium and silver salt.

Some of the compounds of this invention may be crystallised or recrystallised from solvents containing water. In such cases water of hydration may be formed. This invention includes within its scope stoichiometric hydrates as well as compounds containing variable amounts of water that may be produced by processes such as lyophilisation.

Suitable values for Q in the compounds of the formula (I) include the acetoxy, heterocyclylthio group, and nitrogen containing heterocyclic group bonded via nitrogen.

More suitably Q and Q' represent the acetoxy or heterocyclylthio group.

The heterocyclylthio group may suitably be represented by the formula:

- S - Het

wherein "Het" is a five or six membered heterocyclic ring containing from 1 to 4 atoms selected from N, O, and S unsubstituted or substituted with one or two groups selected from C_{1-6} alkyl, C_{1-6} alkoxy, hydroxyalkyl, C_{1-6} alkenyl, alkoxyalkyl, carboxyalkyl, sulphonylalkyl, carbamoylalkyl, trifluoromethyl, hydroxy, halogen, oxo, (subst)aminoalkyl, and carboxyalkyl or two substituents may be linked to form the residue of a heterocyclic or carbocyclic ring.

Examples of the group "Het" include unsubstituted and substituted imidazolyl, triazolyl, tetrazolyl, thiazolyl, thiadiazolyl, thiatriazolyl, oxazolyl, triazinyl and oxadiazolyl.

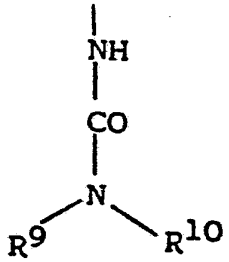
Suitable groups "Het" include unsubstituted and substituted 1, 2, 3-triazolyl; 1, 2, 4-triazolyl; tetrazolyl; oxazolyl; thiazolyl; 1, 3, 4-oxadiazolyl; 1, 3, 4-thiadiazolyl, or 1, 2, 4-thiadiazolyl. Preferably the heterocyclylthio group is 1-methyl-1H-tetrazol-5-ylthio, 2-methyl-1,3,4-thiadiazol-5-ylthio, 1-carboxymethyl-1H-tetrazol-5-ylthio or 6-hydroxy-2-methyl-5-oxo-2H-1,2,4-triazin-3-ylthio.

The nitrogen containing heterocyclic group bonded via nitrogen is suitably a pyridinium group unsubstituted or substituted with one or two groups selected from C_{1-6} alkyl, C_{1-6} alkoxy, hydroxyalkyl, C_{1-6} alkenyl, alkoxyalkyl, carboxyalkyl, sulphonylalkyl, carbamoylmethyl, carbamoyl, trifluoromethyl, hydroxy, halogen, oxo, and aminoalkyl or two substituents may be linked to form the residue of a carbocyclic ring.

Preferably R^7 is a substituted amino group.

More preferably the substituted amino group R^7 is a ureido, acylamino or acylureido group.

One suitable group R^7 is of formula (III):



(III)

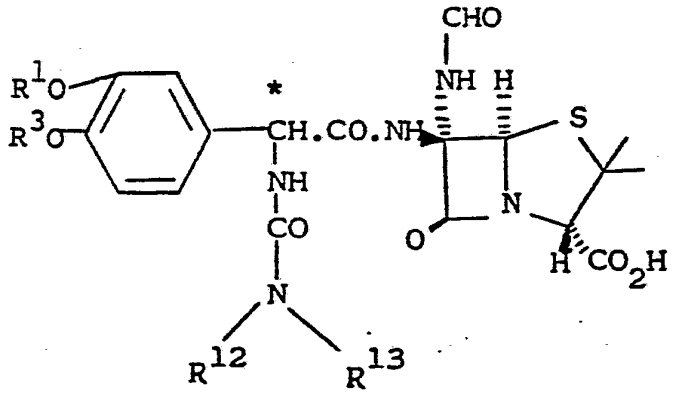
wherein R^9 is hydrogen or a C_{1-6} alkyl group and R^{10} is an optionally substituted 5- or 6- membered heterocyclic group containing one or two nitrogen heteroatoms; or R^9 and R^{10} together with the nitrogen atom to which they are attached form an optionally substituted five- or six-membered heterocyclic group containing one or two nitrogen heteroatoms.

Suitably R^9 is hydrogen.

Suitable substituents for the 5- or 6- membered heterocyclic group of R^{10} or R^9 and R^{10} together include the optionally substituted alkyl, alkenyl, alkynyl, cycloalkyl or cycloalkenyl group; optionally substituted phenyl, oxo; the hydroxy group optionally substituted by alkyl, alkenyl, cycloalkyl, phenyl, pyridyl, pyrimidyl or benzyl; the optionally substituted mercapto group, the alkylsulphonyl group; the substituted imino group; or the amino group optionally substituted by an alkyl, alkenyl, cycloalkyl, phenyl, substituted phenyl or benzyl group. Alternatively two substituents on the ring may form the residue of a further carbocyclic or heterocyclic ring.

The carbon atom marked * in formulae herein is asymmetric so that the compounds may exist as two optically active diastereoisomers. In general that prepared from the D-side chain exhibits the highest antibacterial activity and accordingly the D compound or the DL mixtures are preferred, with the D compound being particularly preferred.

Preferred compounds within formula (I) are the penicillin derivatives of formula (IV) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:



(IV)

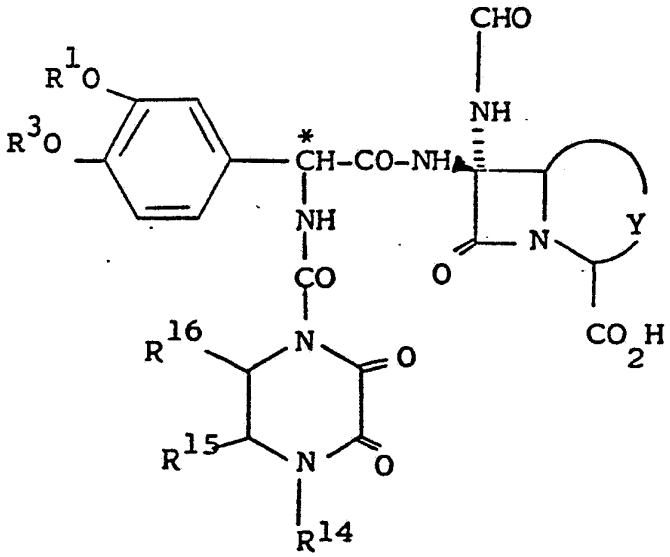
wherein R^1 and R^3 are as defined with respect to formula (I); R^{12} is hydrogen or C_{1-6} alkyl and R^{13} is an optionally substituted five- or six-membered heterocyclic group containing one or two nitrogen heteroatoms; or R^{12} and R^{13} together with the nitrogen atom to which they are attached form an optionally substituted five- or six-membered heterocyclic group containing one or two nitrogen heteroatoms.

Suitable substituents for the five- or six-membered heterocyclic group of R^{13} or R^{12} and R^{13} together include the alkyl, alkenyl, alkynyl, cycloalkyl or cycloalkenyl group, optionally substituted phenyl, oxo, the hydroxy group optionally substituted by alkyl, alkenyl, cycloalkyl, phenyl, pyridyl, pyrimidyl or benzyl, the optionally substituted mercapto group, the alkylsulphonyl group,

the substituted imino group, or the amino group optionally substituted by an alkyl, alkenyl, cycloalkyl, phenyl, substituted phenyl or benzyl group. Alternatively two substituents on the ring may form the residue of a further carbocyclic or heterocyclic ring.

Preferably R^{12} is hydrogen.

One particularly preferred sub-group within the present invention provides a compound of formula (V) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:



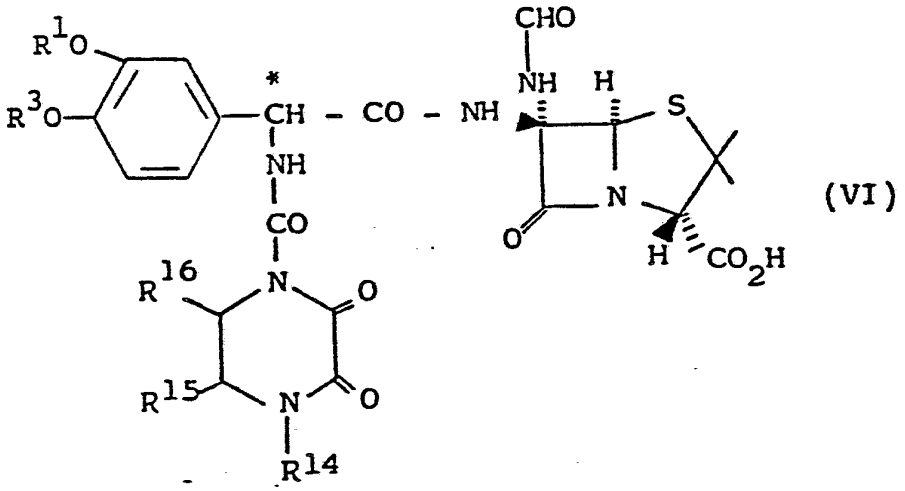
(V)

wherein R^1, R^3 and Y are as defined with respect to formula (I) and R^{14} represents hydrogen, C_{1-6} alkyl, substituted alkyl, aryl, or aralkyl; R^{15} and R^{16} are the same or different and represent hydrogen, C_{1-6} alkyl, substituted alkyl, halogen, amino, hydroxy or C_{1-6} alkoxy or R^{15} and R^{16} form the residue of 5- or 6-membered carbocyclic or heterocyclic ring.

Suitable values for Y in the compounds of formula (V) are $-S-C(CH_3)_2-$ and $-S-CH_2-C(CH_2Q)=$ wherein Q is as hereinbefore defined.

Preferably Y in the compounds of formula (V) is $-S-C(CH_3)_2-$ or $-S-CH_2-C(CH_2Q')=$ wherein Q' is as hereinbefore defined.

Preferred compounds within formula (V) are the penicillin derivatives of formula (VI) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:

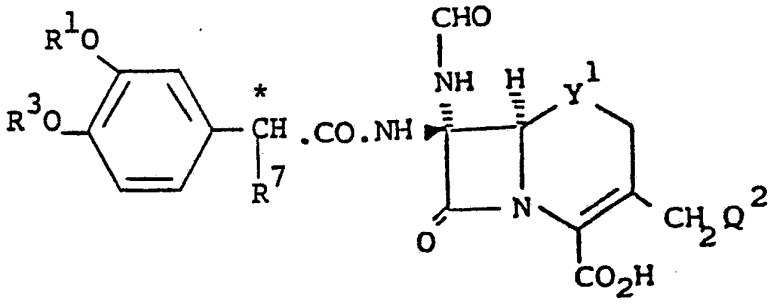


(VI)

wherein R^1, R^3, R^{14}, R^{15} and R^{16} are as hereinbefore defined.

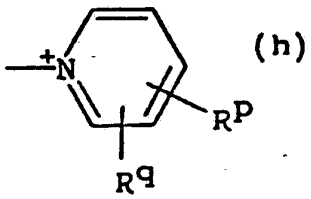
Suitable C_{1-6} alkyl groups for the groups R^{14}, R^{15} and R^{16} in formula (V) and formula (VI) include methyl, ethyl, n- and iso-propyl, n, sec-, iso- and tert-butyl. Preferably R^{14} is ethyl. Preferably R^{15} and R^{16} are hydrogen.

A further preferred subgroup of compounds within the present invention are the compounds of formula (VII) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:



(VII)

wherein R^1, R^3 and R^7 are as hereinbefore defined; Y^1 is oxygen or sulphur; and Q^2 represents acetyloxy, a group -SHet, wherein Het is as hereinbefore defined, or Q^2 represents a subgroup of formula (h):



(h)

wherein R^q and R^p may be the same or different and each represents hydrogen, C_{1-6} alkyl, C_{1-6} alkoxy, hydroxyalkyl, C_{1-6} alkenyl, alkoxyalkyl, carboxyalkyl, sulphonylalkyl, carbamoylalkyl, carbamoyl, trifluoromethyl, hydroxy, halogen, and aminoalkyl. or R^q and R^p together from the residue of a carbocyclic ring.

Suitable groups 'Het' within formula (VII) include substituted and unsubstituted 1,2,3-triazolyl; 1,2,4-triazolyl; tetrazolyl; oxazolyl; thiazolyl; 1,3,4-oxadiazolyl; 1,2,4-triazinyl; 1,3,4-thiadiazolyl or 1,2,4-thiadiazolyl. Preferably the groups 'SHet' is 1-methyl-1H-tetrazol-5-ylthio,

2-methyl-1,3,4-thiadiazol-5-ylthio, 1-carboxymethyl-1H-tetrazol-5-ylthio or 6-hydroxy-2-methyl-5-oxo-2H-1,2,4-triazin-3-ylthio.

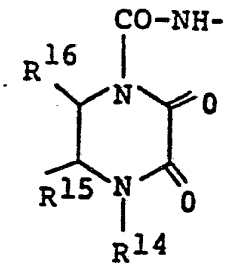
Suitably R^q represents hydrogen.

Suitably R^p represents hydrogen, sulphonylalkyl or carbamoyl, preferably the substituent R^p is in the 4-position.

Suitably Y^1 is sulphur.

Suitably Y^1 is oxygen.

Preferably R^7 within formula (VII) is a subgroup of formula (j):



(j)

wherein R^{14}, R^{15} and R^{16} are as hereinbefore defined with reference to formula (V).

Specific compounds within this invention include the following and pharmaceutically acceptable salts and in-vivo hydrolysable esters thereof:

6β-[D-2-[3,4-bis(ethoxycarbonyloxy)phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]acetamido]-6α-formamidopenicillanic acid; and

6α-formamido-6β-[D-2-[3,4-(methylenedioxy)phenyl]-2-(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino)acetamido] penicillanic acid.

The antibiotic compounds according to the invention may be formulated for administration in any convenient way for use in human or veterinary medicine, according to techniques and procedures per se known in the art with reference to other antibiotics, and the invention therefore includes within its scope a pharmaceutical composition comprising an antibiotic compound according to the present invention such as, for example a compound of formula (II) above together with a pharmaceutically acceptable carrier or excipient.

The compositions may be formulated for administration by any suitable route, such as oral or parenteral, or by topical application. The compositions may be in the form of tablets, capsules, powders, granules, lozenges, creams or liquid preparations, such as oral or sterile parenteral solutions or suspensions.

Tablets and capsules for oral administration may be in unit dose presentation form, and may contain conventional excipients such as binding agents, for example syrup, acacia, gelatin, sorbitol, tragacanth,or polyvinylpyrollidone; fillers, for example lactose, sugar, maize-starch, calcium phosphate, sorbitol or glycine, tabletting lubricants, for example magnesium stearate, talc, polyethylene glycol or silica; disintegrants, for example potato starch; or acceptable wetting agents such as sodium lauryl sulphate. The tablets may be coated according to methods well known in normal pharmaceutical practice. Oral liquid preparations may be in the form of, for example, aqueous or oily suspensions, solutions, emulsions, syrups or elixirs, or may be presented as a dry product for reconstitution with water or other suitable vehicle

before use. Such liquid preparations may contain conventional additives such as suspending agents, for example sorbitol, methyl cellulose, glucose syrup, gelatin, hydroxyethylcellulose, carboxymethyl cellulose, aluminium stearate gel or hydrogenated edible fats, emulsifying agents, for example lecithin, sorbitan monooleate, or acacia; non-aqueous vehicles (which may include edible oils), for example almond oil, oily esters, glycerine, propylene glycol, or ethyl alcohol; preservatives, for example methyl or propyl p-hydroxybenzoate or sorbic acid, and if desired conventional flavouring or colouring agents.

Suppositories will contain conventional suppository base, eg cocoa-butter or other glyceride.

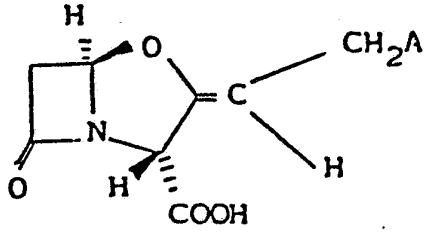
For parenteral administration, fluid unit dosage forms are prepared utilising the compound and a sterile vehicle, water being preferred. The compound, depending on the vehicle and concentration used, can be either suspended or dissolved in the vehicle. In preparing solutions the compound can be dissolved in water for injection and filter sterilised before filling into a suitable vial or ampoule and sealing. Advantageously, agents such as local anaesthetic, preservative and buffering agents can be dissolved in the vehicle. To enhance the stability, the composition can be frozen after filling into the vial and the water removed under vacuum. The dry lyophilised powder is then sealed in the vial and an accompanying vial of water for injection may be supplied to reconstitute the liquid prior to use. Parenteral suspensions are prepared in substantially the same manner except that the compound is suspended in the vehicle instead of

being dissolved and sterilisation cannot be accomplished by filtration. The compound can be sterilised by exposure to ethylene oxide before suspending in the sterile vehicle. Advantageously, a surfactant or wetting agent is included in the composition to facilitate uniform distribution of the compound.

The composition may contain from 0.1% by weight, preferably from 10-60% by weight, of the active material, depending on the method of administration. Where the compositions comprise dosage units, each unit will preferably contain from 50-500 mg of the active ingredient. The dosage as employed for adult human treatment will preferably range from 100 to 10000 mg per day, for instance 1500 mg per day depending on the route and frequency of administration. Such a dosage corresponds to 1.5 to 50 mg/kg per day.

The antibiotic compound according to the present inventio may be the sole therapeutic agent in the compositions of the invention or a combination with other antibiotics and/or β-lactamase inhibitor may be employed.

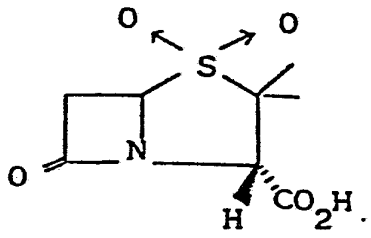
Advantageously the compositions also comprise a compound of formula (VIII) or a pharmaceutically acceptable salt or ester thereof:



(VIII)

wherein A is hydroxyl, substituted hydroxyl, thiol, substituted thiol, amino, mono- or di-hydrocarbyl substituted amino, or mono- or di-acylamino.

A further advantageous composition comprises an antibiotic compound according to the invention together with a β-lactamase inhibitor of formula (IX) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:



(IX)

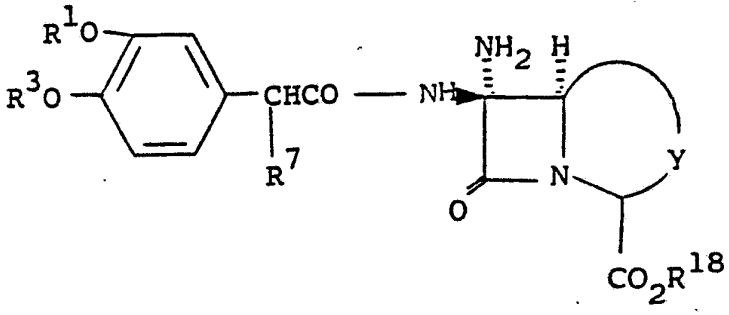
Further suitable β-lactamase inhibitors include 6β-bromopenicillanic acid and salts and in vivo hydrolysable esters and 6β-iodopenicillanic acid and salts and in vivo hydrolysable esters thereof.

Such compositions of this invention comprising a β-lactamase inhibitor are formulated in conventional manner.

The present invention also includes a method of treating bacterial infections in humans and animals which comprises the administration of a therapeutically effective amount of an antibiotic compound of this invention.

The antibiotic compounds of the present invention are active against a broad range of gram positive and gram negative bacteria, in particular they are useful for treatment of respiratory tract and urinary tract infections in humans and mastitis in cattle. A particular advantage of the antibacterially active compounds of this invention is their stability to β-lactamase enzymes and they are therefore effective against β-lactamase producing organisms.

The present invention further provides a process for the preparation of a compound of formula (I) which process comprises formylating a compound of formula (X):



(X)

where any reactive groups may be protected; R^1, R^3, R^7 and Y are as defined with respect to formula (I); R^{18} is a readily removable carboxy protecting group; and thereafter, if necessary, carrying out one or more of the following steps:

(i) converting a group R^{18} to a group R^2;

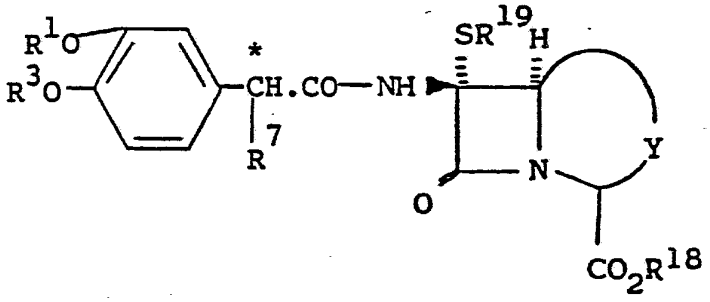
(ii) converting one group Z into a different group Z;

(iii) converting the product into a salt.

Suitable formylating agents include mixed anhydrides such as formic acetic anhydride. The reaction may suitably be carried out in a temperature in the range -50°C to 30°C in aprotic solvent such as, for example, dichloromethane, chloroform, dimethylformamide, tetrahydrofuran, hexamethylphosphoramide, or dimethylsulphoxide, in the presence of a tertiary base. A preferred tertiary base employed in the reaction is a base of the pyridine type, such as pyridine, lutidine or picoline.

A process for preparing compounds within formula (X) is disclosed in US Patent No. 3,962,214 and in UK Patent No. 1348984.

Compounds of the formula (X) may be prepared by the reaction of a corresponding compound of the formula (XI):



(XI)

wherein Y, R^1, R^3, R^7 and R^{18} are as hereinbefore defined, and R^{19} is C_{1-6} alkyl, aryl or benzyl; with anhydrous ammonia, an ammonium salt or an amine of the formula (XII):

$$R^{20}-NH_2 \qquad (XII)$$

wherein R^{20} is a removable protecting group such as benzyl; in the presence of a metal ion such as mercury, silver, thallium, lead or copper and thereafter if necessary removing any protecting group to form the compound of formula (X).

Suitable examples of the alkyl group for R^{19} include C_{1-6} alkyl groups such as methyl, ethyl, n-, or iso-propyl, and n-, sec-; iso-, or tert-butyl groups.

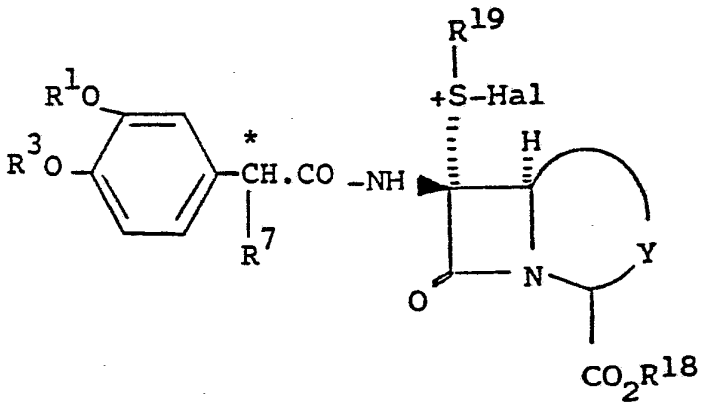
A preferred alkyl group for R^{19} is methyl.

Suitable examples of the aryl group R^{19} include phenyl, optionally substituted with C_{1-6} alkyl, C_{1-6} alkoxy, halogen, or nitro. Preferred aryl groups for R^{19} include phenyl, o-, m- or p-methylphenyl, o-, m- or p-nitrophenyl, in particular p-methylphenyl.

Suitable solvents in which the reaction may be performed include for example, diethylether, tetrahydrofuran, dimethylformamide, methanol and hexamethylphosphoramide. The reactions are generally carried out under an inert atmosphere and at moderate to low temperatures ie in the range -100°C to 30°C. The course of the reaction may be followed by conventional methods such as thin layer chromatography and terminated when an optimum quantity of product is present in the reaction mixture.

The preferred metal ion for use in the above process is the mercuric ion, aptly in the form of mercuric acetate.

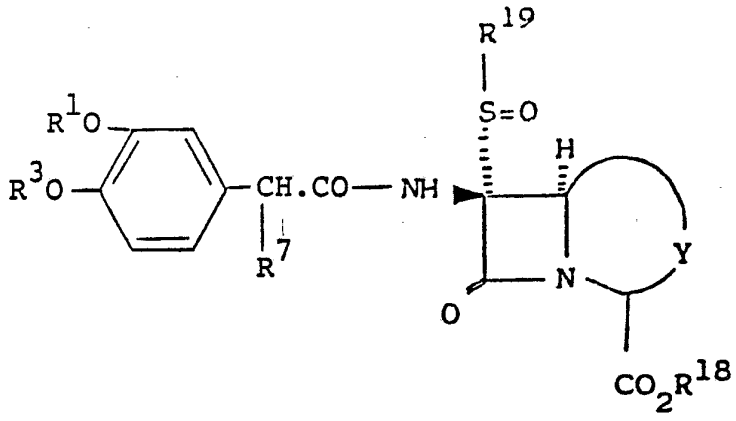
Alternatively compounds of the formula (X) may be prepared by the reaction of a corresponding compound of the formula (XIII):



(XIII)

wherein R^1, R^3, R^7, R^{18} and R^{19} are as hereinbefore defined; Hal is chloro or bromo, with anhydrous ammonia, an ammonium salt or an amine of formula (XII) as hereinbefore defined and thereafter if necessary removing any protecting group to form the compound of formula (X). The compounds of the formula (XIII) may be prepared by the reaction of a compound of the formula (XI) as hereinbefore defined, with a halogenating agent such as chlorine or bromine in an inert solvent, for example dichloromethane, at a depressed temperature such as -80°C to -30°C.

A further method of preparation of the compounds of the formula (X) comprises the reaction of a compound of the formula (XIV):



(XIV)

wherein R^1, R^3, R^7, R^{18} and R^{19} are as hereinbefore defined; with anhydrous ammonia, an ammonium salt or an amine of the formula (XII) as hereinbefore defined and thereafter if necessary removing any protecting group to form the compound of the formula (X).

Suitably such a reaction is performed at a non-extreme temperature for example 0°C - 60°C, normally 10°C - 40°C and preferably ambient. The reaction is conveniently performed in an aprotic solvent such as tetrahydrofuran or dioxan.

It will be appreciated that the processes for preparation of a compound of formula (X) described hereinbefore proceed via an imine intermediate; other processes proceeding via such an intermediate are also included herein.

The compounds of the formula (XIV) may be prepared by the oxidation of a compound of the formula (XI) as hereinbefore defined. Such oxidation may conveniently performed in conventional manner, for example using a per-acid such as peracetic acid or m-chloroperbenzoic

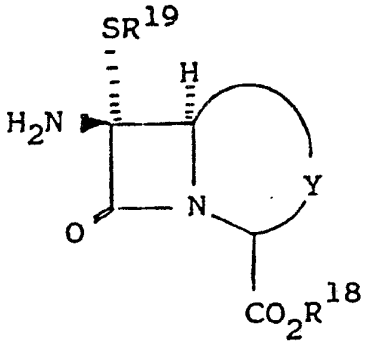
acid, suitably at an ambient or depressed temperature. Suitable solvents for such a sulphoxidation include ethylacetate, chloroform, dichloromethane, dioxan and tetrahydrofuran.

Examples of suitable protecting groups for the group R^{20} include those known in the art as being cleavable to provide the -NH-. Mention may be made of silyl groups such as trimethylsilyl, tertiarybutyl-dimethylsilyl, and tri-isopropylsilyl. A preferred protecting group is (p-methoxymethoxy)phenyl which is removable by cerium ammonium nitrate. Other protecting groups of interest include those cleavable by methanolysis such as $-C(CO_2R)=O$ (This moiety may be derived from groups of the type $-C(CO_2R)=C(CH_3)_2$). Further suitable protecting groups include 4-nitro-benzyl and 2,4-dimethoxybenzyl which is removable with potassium persulphate.

The oxidation of a compound of the formula (XI) which contains sulphur atoms in addition to that shown in the formula may oxidise the additional sulphur atoms and accordingly it may be necessary to reduce the thus formed sulphoxide or sulphone to the corresponding sulphide.

Preferably Y in the compound of formula (XIII) is $-O-CH_2-CZ=$ wherein Z is as hereinbefore defined.

The starting material for the above processes ie compound of formula (XI) above may be prepared by acylation, under conventional conditions of the compound (XV):



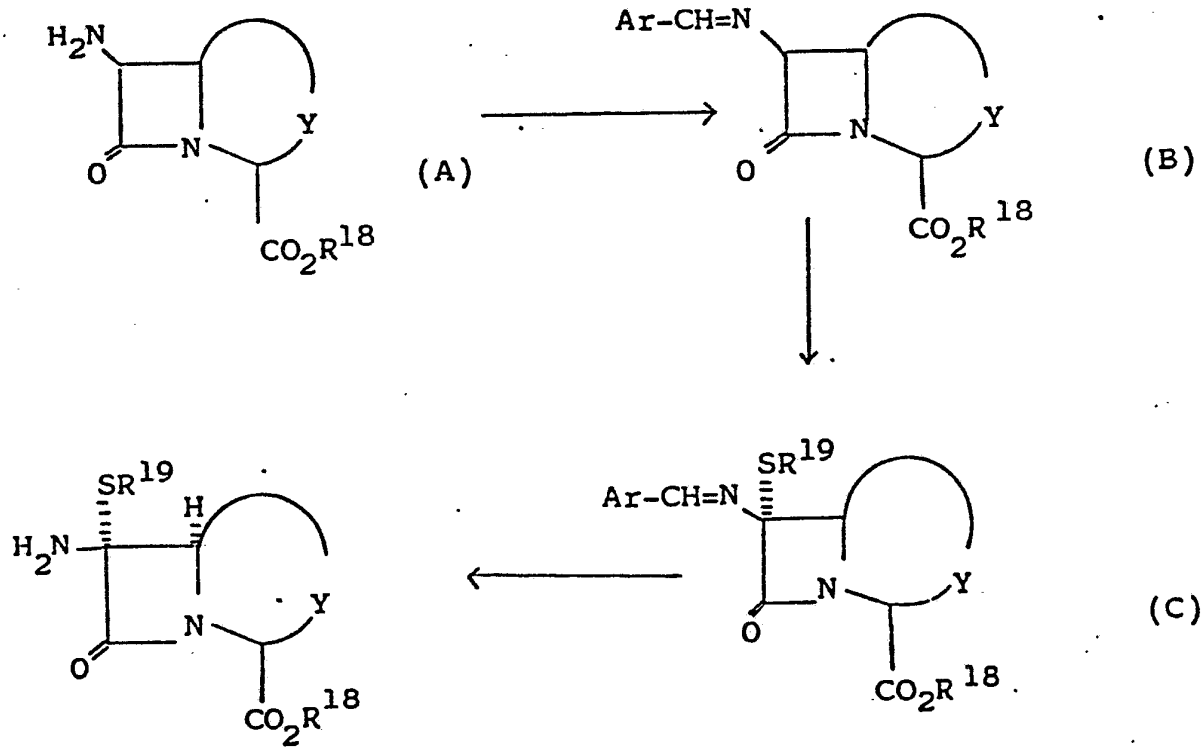
(XV)

wherein R^{18}, R^{19} and Y are as defined hereinbefore.

Compounds of the formula (XV) may be prepared by methods known or analogous to those known for the preparation of 7α-substituted-thio cephalosporins and 6α-substituted-thio penicillins.

Compounds of formula (XV) may suitably be prepared from a Schiff's base derivative as outlined in Scheme 1.

Scheme 1



The compound of formula (XV) is prepared by reacting the amino compound (A) with an aldehyde of formula Ar-CHO wherein Ar is an aryl group to form the Schiff base (B). The Schiff base (B) is reacted with a base to form an anion which is treated with a thiosulphonate of formula:

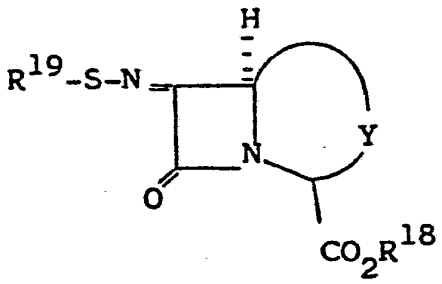
$$R^{19}S.SO_2R^{19}$$

or a sulphenyl chloride of formula:

$$R^{19}SCl$$

wherein R^{19} is as hereinbefore defined to give the compound of formula (C). Acidic hydrolysis of the Schiff base gives the β-amino compound of formula (XV).

Compounds of formula (XV) may also be prepared by reacting a thiooxime compound of formula (XVI):

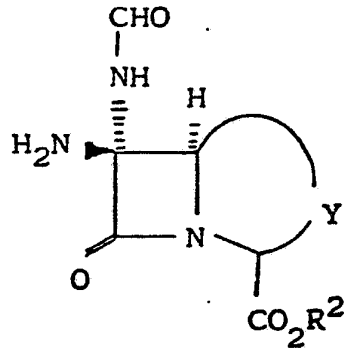


(XVI)

where R^{18} and R^{19} are as defined hereinbefore above with a tri(alkyl)phosphine or tri(aryl)phosphine, followed by treatment with an acid catalyst such as silica gel. The process is as described in US Patent No. 4,119,778 and in J. Amer. Chem. Soc., 1980, 102, 1690.

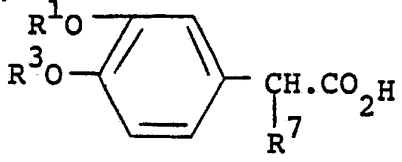
Compounds within formula (XV) and (XVI) may also be prepared by the process disclosed in US Patent No. 3,962,214 or an appropriate modification thereof.

The compounds of formula (I) may also be prepared by reacting a compound of formula (XVII):



(XVII)

wherein the amino group is optionally substituted with a group which permits acylation to take place and R^2 is as hereinbefore defined with reference to formula (I) above, with an N-acylating derivative of an acid of formula (XVIII):



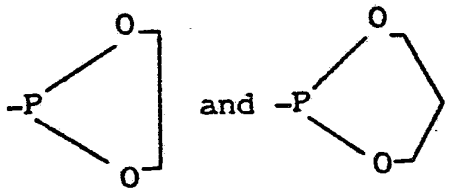
(XVIII)

wherein R^1, R^3 and R^7 are as defined with respect to formula (I) and wherein any reactive groups therein may be protected; and thereafter, if necessary, carrying out

one or more of the following steps:

i) removing any carboxyl-protecting group R^2;
ii) removing any protecting groups on the side-chain group;
iii) further derivatising the side chain group;
iv) converting one group Z to a different group Z;
v) converting the product into a salt or in vivo hydrolysable ester thereof.

Suitable groups which permit acylation to take place and which are optionally present on the amino group of the starting material of the formula (XVII) include N-silyl, N-stannyl and N-phosphorus groups, for example trialkylsilyl groups such as trimethylsilyl, trialkyltin groups such as tri-n-butyltin, groups of formula $-P.R^aR^b$ wherein R^a is an alkyl, haloalkyl, aryl, aralkyl, alkoxy, haloalkyl, aryl, aralkyl, alkoxy, haloalkoxy, aryloxy, aralkyloxy or dialkylamino group, R^b is the same as R^a or is halogen or R^a and R^b together form a ring; suitable such phosphorus groups being $-P(OC_2H_5)_2$, $-P(C_2H_5)_2$,



The carboxyl group may be regenerated from any of the above esters by usual methods appropriate to the particular R^2 group, for example, acid - and base - catalysed hydrolysis, or by enzymically - catalysed hydrolysis, or by hydrogenolysis.

Suitable carboxyl-protecting derivatives for the group $-CO_2R^2$ in formula (XVII) include salts and ester derivatives of the carboxylic acid as described hereinbefore with reference to formula (I).

A reactive N-acylating derivative of the acid (XVIII) is employed in the above process. The choice of reactive derivative will of course be influenced by the chemical nature of the substituents of the acid.

Suitable N-acylating derivatives include an acid halide, preferably the acid chloride or bromide. Acylation with an acid halide may be affected in the presence of an acid binding agent for example, tertiary amine (such as triethylamine, pyridine or dimethylaniline), an inorganic base (such as calcium carbonate or sodium bicarbonate) or an oxirane, which binds hydrogen halide liberated in the acylation reaction. The oxirane is preferably a (C_{1-6})-1,2-alkylene oxide - such as ethylene oxide or propylene oxide. The acylation reaction using an acid halide may be carried out at a temperature in the range -50°C to +50°C, preferably -20°C to +20°C, in aqueous or non-aqueous media such as water, acetone, tetrahydrofuran, ethyl acetate, dimethylacetamide, dimethylformamide, acetonitrile, dichloromethane, 1,2-dichloroethane, or mixtures thereof. Alternatively, the reaction may be carried out in an unstable emulsion of water-immiscible solvent, especially an aliphatic ester or ketone, such as methyl isobutyl ketone or butyl acetate.

The acid halide may be prepared by reacting the acid (XVIII) or a salt thereof with a halogenating (eg chlorinating or brominating) agent such as phosphorus pentachloride, thionyl chloride or oxalyl chloride.

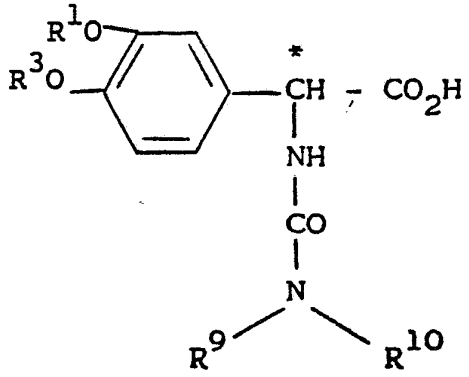
Alternatively, the N-acylating derivative of the acid (XVIII) may be a symmetrical or mixed anhydride. Suitable mixed anhydrides are alkoxyformic anhydrides, or anhydrides with, for example, carbonic acid monoesters, trimethyl acetic acid, thioacetic acid, diphenylacetic acid, benzoic acid, phosphorus acids (such as phosphoric or phosphorous acids) or aliphatic or aromatic sulphonic acids (such as p-toluenesulphonic acid). When a symmetrical anhydride is employed, the reaction may be carried out in the presence of 2,6-lutidine as catalyst.

Alternative N-acylating derivatives of acid (XVIII) are the acid azide, or activated esters such as esters with 2-mercaptopyridine, cyanomethanol, p-nitrophenol, 2,4-dinitrophenol, thiophenol, halophenols, including pentachlorophenol, monomethoxyphenol, N-hydroxy succinimide, or 8-hydroxyquinoline; or amides such as N-acylsaccharins, N-acylthiazolidin-2-thione or N-acylphthalimides; or an alkylidene iminoester prepared by reaction of the acid (XVIII) with an oxime.

Other reactive N-acylating derivatives of the acid (XVIII) include the reactive intermediates formed by reaction in situ with a condensing agent such as a carbodiimide, for example, N,N'-diethyl-, dipropyl- or diisopropylcarbodiimide, N,N'-di-cyclohexyl-carbodiimide, or N-ethyl-N'-[3-(dimethylamino)propyl]-carbodiimide; a suitable carbonyl compound, for example, N,N'-carbonyldiimidazole or N,N'-carbonyldi-triazole; an isoxazolinium salt, for example, N-ethyl-5-phenylisoxazolinium-3-sulphonate or N-t-butyl-5-methylisoxazolinium perchlorate; or an N-alkoxycarbonyl 2-alkoxy-1,2-dihydroquinoline, such as N-ethoxycarbonyl

2-ethoxy-1,2-dihydroquinoline. Other condensing agents include Lewis acids (for example BBr_3 - C_6H_6); or a phosphoric acid condensing agent such as diethylphosphorylcyanide. The condensation reaction is preferably carried out in an organic reaction medium, for example, methylene chloride, dimethylformamide, acetonitrile, alcohol, benzene, dioxan or tetrahydrofuran.

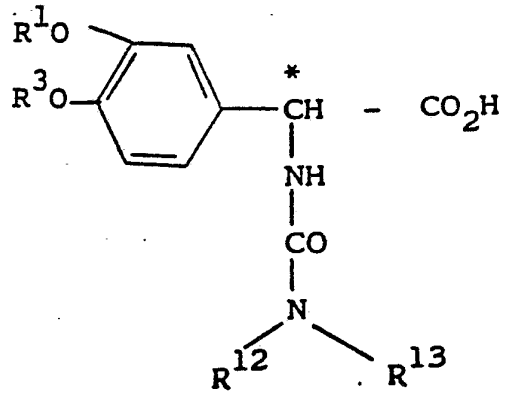
Aptly the acid of formula (XVIII) is an acid of formula (XIX):



(XIX)

wherein R^1, R^3, R^9 and R^{10} are as hereinbefore defined; thereby affording a compound having a group R^7 of formula (III) as hereinbefore defined.

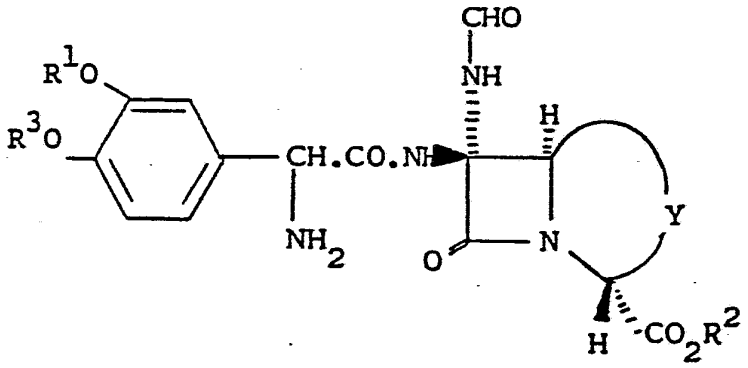
Aptly Y in formula (XVII) is $-S-C(CH_3)_2-$ and the acid of formula (XVIII) is an acid of formula (XX):



(XX)

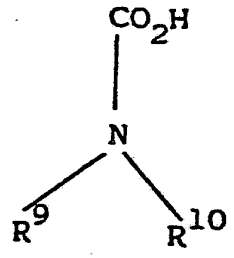
wherein R^1, R^3, R^{12} and R^{13} are as hereinbefore defined; thereby affording a compound of formula (IV) as hereinbefore defined.

The compounds of formula (III) may also suitably be prepared by reacting a compound of formula (XXI):



(XXI)

wherein R^1, R^3, R^2, R^8 and Y are as hereinbefore defined and the α-amino group is optionally substituted with a group which permits acylation to take place, and any reactive groups may be protected, with an N-acylating derivative of an acid of formula (XXII):



(XXII)

wherein R^9 and R^{10} are as hereinbefore defined and wherein any reactive groups may be protected; and thereafter, if necessary, carrying out one or more of the following steps:

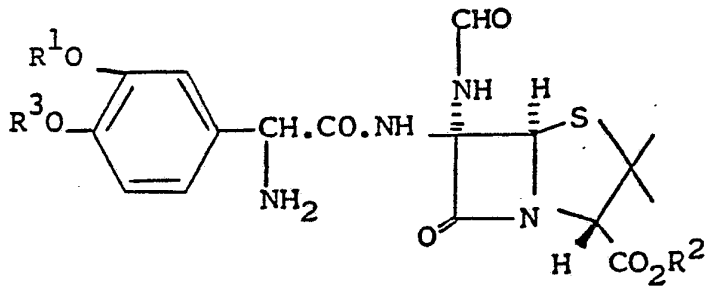
i) removing any carboxyl-protecting group R^2;

ii) removing any protecting groups on the side-chain group;

iii) converting one group Z to a different group Z;

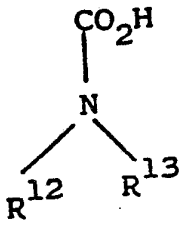
iv) converting the product into a salt or in vivo hydrolysable ester thereof.

The compounds of formula (IV) as hereinbefore defined are aptly prepared by reacting a compound of formula (XXIII):



(XXIII)

wherein the ∝-amino group is optionally substituted with a group which permits acylation to take place and any reactive groups may be protected, and R^1, R^2 and R^3 are as hereinbefore defined with an N-acylating derivative of an acid of formula (XXIV):



(XXIV)

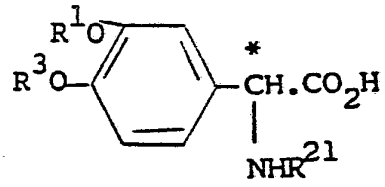
wherein R^{12} and R^{13} are as defined with respect to formula (IV) above and any reactive groups may be protected; and thereafter, if necessary, carrying out one or more of the following steps:

i) removing any carboxyl-protecting group R^2;

ii) removing any protecting groups on the side-chain group;

iii) converting the product into a salt or in vivo hydrolysable ester thereof.

The compounds of formula (XXI) herein may be prepared by reacting a compound of formula (XVII) with an N-acylating derivative of an acid of formula (XXV):

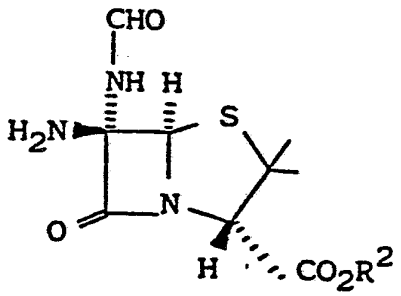


(XXV)

wherein R^{21} is an amino-protecting group and thereafter removing protecting group R^{21}.

Suitable amino protecting groups R^{21} include alkoxycarbonyl groups such as, for example, 4-nitrobenzyloxycarbonyl and trichloroethyloxycarbonyl.

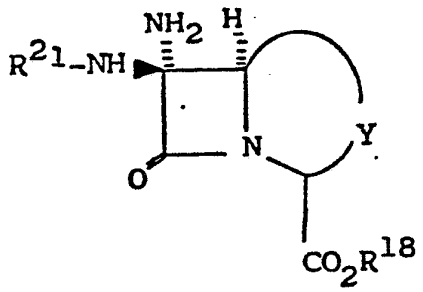
The compounds of formula (XXIII) herein which are inter alia intermediates for the compounds of formula (IV) as hereinbefore defined may be prepared by reacting a compound of formula (XXVI):



(XXVI)

wherein R^2 is as defined hereinbefore with an N-acylating derivative of an acid of formula (XXV) as hereinbefore defined.

The intermediate compound of formula (XVII) as hereinbefore defined may suitably be prepared by formylating a compound of formula (XXX):



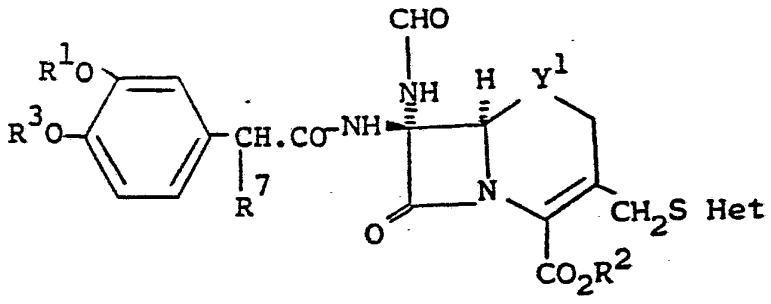
(XXX)

wherein R^{18}, R^{21} and Y are as hereinbefore defined and thereafter removing the protecting group R^{21} and if necessary, converting a group R^{18} to a group R^2.

Suitable formylating agents and reaction conditions are as hereinbefore defined.

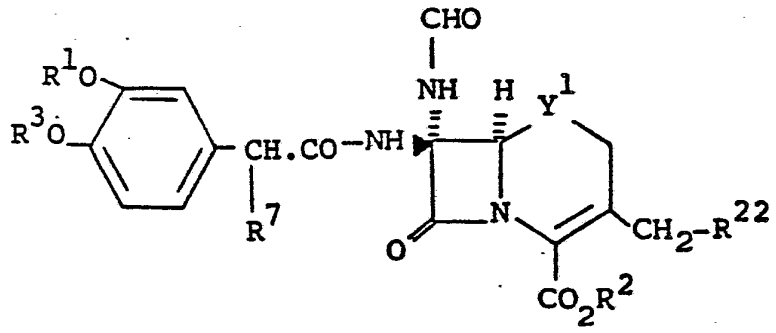
When Y in the compound of formula (XXX) is $-S-C(CH_3)_2-$ the process produces the compound of formula (XXVI) herein.

The sub-group of compounds within the present invention of formula (XXXI):



(XXXI)

wherein Y^1 and 'Het' are as defined hereinbefore with reference to formula (VII) and R^1, R^2, R^3 and R^7 are as defined hereinbefore with reference to formula (I) may suitably be prepared by reacting a compound of formula (XXXII):



(XXXII)

wherein Y^1, R^1 and R^2, R^3 and R^7 are as defined hereinbefore and wherein any reactive groups may be protected and R^{22} is a leaving group; with a thiol of formula:

HetSH

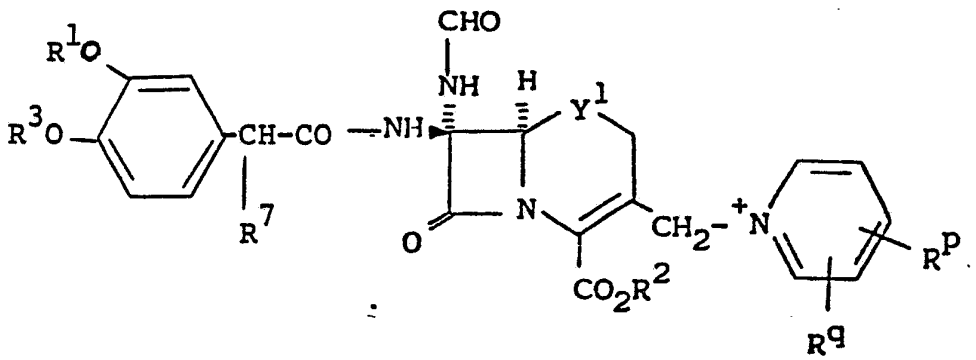
with the proviso that when R^{22} is an acyloxy group $-CO_2R^2$ must be in the free acid form or a salt thereof.

Suitable leaving groups R^{22} include halogen such as iodide or bromide or an acyloxy groups such as, for example the acetyloxy group.

The thiol HetSH may be reacted as the free compound or a salt with an alkali metal such as sodium or potassium. This reaction is desirably conducted in a solvent. For example, use can be made of water, or organic solvents inert to the starting compounds, such as dimethylformamide, dimethylacetamide, dioxane, acetone, alcohol, 1,2-dichloroethane, acetonitrile, dimethylsulfoxide or tetrahydrofuran, or mixtures thereof. The reaction temperature and time depend, among other factors, upon the starting compounds and solvent to be employed but generally the reaction is carried out at a selected temperature within the range of 0 to 100°C for a selected time of a few hours to several days. The reaction is desirably conducted between pH 3 and 7.

To prevent oxidation of the thio compounds it is advantageous to carry out the reaction in an inert gaseous atmosphere, eg nitrogen gas.

The subgroup of compounds within the present invention of formula (XXXIII):



(XXXIII)

wherein R^1, R^2, R^3, R^7, R^p, R^q and Y^1 are as defined hereinbefore may suitably be prepared by reacting a compound of formula (XXXII) as hereinbefore defined with the appropriately substituted pyridine.

Suitably the reaction with the pyridine is carried out in a polar solvent such as water, and in the presence of a catalyst such as an alkali metal thiocyanate or an alkali metal halide such as, for example sodium iodide.

The antibiotic compounds of the present invention are active against a wide range of gram negative and gram positive organisms including E.coli such as, for example ESS, JT4, JT425 and NCTC 10418; Pseudomonas Spp. such as Ps.aeruginosa for example 10662 and Dalgleish; Serratia marcescens US32; Klebsiella aerogenes A; Enterobacter cloacae N1; P.mirabilis such as, for example C977 and 889; P.morganii; P.rettgeri; B.subtilis; Staph aureus such as, for example Oxford and Russell; N.catarrhalis 1502; Strep faecalis I; β-Haemolytic Strep CN10. The MIC data included in the following examples is representative of the activity of the compounds of the present invention.

The following Examples illustrate the preparation and use of the compounds of the present invention.

Example 1

6β-[D-2-[3,4-bis(ethoxycarbonyloxy)phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]acetamido]-6α-formamidopenicillanic acid, sodium salt.

(a) DL-2-[3,4-bis(ethoxycarbonyloxy)phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]acetic acid.

DL-2-(3,4-Dihydroxyphenyl)-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino] acetic acid (3.51g;10mMole) in water (50ml) was dissolved by addition of 10% aqueous sodium hydroxide to pH 7.5. Acetone (50ml) was added and the mixture treated at room temperature with ethyl chloroformate (2.39g;22mMole). The pH was maintained throughout between 7.0 and 7.5 by addition of saturated, aqueous sodium hydrogen carbonate solution when necessary. After 1.5 h, the pH was steady at 7.5 and 5M. hydrochloric acid was added to give pH 6.5. The acetone was removed in vacuo and the residue was washed with ether (50ml) and covered with a layer of ethyl acetate (50ml). The pH of the mixture was adjusted to pH 2 by addition of 5M. hydrochloric acid and the phases were separated. The aqueous phase was extracted with ethyl acetate (50ml), the extracts combined, washed with water (50ml), saturated brine (25ml), dried over anhydrous magnesium sulphate and evaporated to dryness in vacuo to give the title compound (4.43g, 89%) as a white foam; ν_{max} ($CHCl_3$) 3600 - 2400, 1760, 1710, 1682, 1250, and 1210 cm^{-1}; δ [$(CD_3)_2CO$] 1.00 - 1.57 (9H, m, CH_3s), 3.20-4.60 (10H, m, CH_2's), 5.66 (1H, d, J 7Hz, CH), 7.56 (3H, br s, aromatics), 9.00 (1H, br s, CO_2H), 10.15 (1H, d, J 7Hz, NH).

(b) Benzyl 6β-[D-2-[3,4-bis(ethoxycarbonyloxy) phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]-6α-formamidopenicillanate.

Benzyl 6α-formamido-6β-(2,2,2-trichloroethoxycarbonylamino) penicillanate (1.31g, 2.5mMole) was dissolved in tetrahydrofuran (100ml) at room temperature and treated with M. potassium dihydrogen phosphate (25ml) followed by acid washed zinc metal (5g). The resulting pH of the mixture was 3.5 and was maintained below 4.5 by addition of 5M. hydrochloric acid when necessary. When thin layer chromatography (silica gel, ethyl acetate) indicated that no starting material remained (usually 1 Hour) the mixture was filtered and the insolubles were washed with ethyl acetate (100ml) and water (50ml). The phases in the filtrate were separated, the aqueous phase further extracted with ethyl acetate (100ml), the extracts combined, washed with water (50ml), saturated brine (25ml), dried over anhydrous magnesium sulphate and evaporated to dryness in vacuo to yield benzyl 6β-amino-6α-formamidopenicillanate as a yellow foam. This was redissolved in tetrahydrofuran (10ml) and treated with N, N' -dicyclohexylcarbodiimide (0.6g, 2.9mMole), then a solution of DL-2-[3,4-bis(ethoxycarbonyloxy)phenyl] -2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino] acetic acid (1.24g; 2.5mMole) in tetrahydrofuran (10ml) was added over 0.5h. It was stirred at room temperature for 18h then filtered and evaporated to dryness in vacuo. Chromatography on silica gel 60 (< 230 mesh ASTM), eluting with ethyl acetate gave the title compound plus its L-diastereoisomer(total 1.36g, 66% as a white foam, from which the D-diastereoisomer was separated by crystallisation from ethyl acetate /ethanol as a white solid; $[\alpha]_D^{20}$ + 140.0° (c 1.04, $CHCl_3$); ν_{max}(KBr) 3280, 1770, 1710sh, 1675, 1500, 1368, 1250 , 1185cm^{-1}; δ [$(CD_3)_2CO$] 0.98 and 1.23 (6H, 2s, 2- CH_3'S), 1.16 (3H, t, J 7Hz, $NCH_2\underline{C}H_3$), 1.30 (6H, t, J 8Hz, $2\underline{C}H_3CH_2OCO_2$'S), 3.48 (2H, q, J 7Hz, $NC\underline{H}_2CH_3$),

3.60 - 3.88 and 3.89 - 4.11 (4H, 2m, NCH_2CH_2N), 4.26 (4H, q, J 8Hz, $2CH_3CH_2OCO_2$'S), 4.39 (1H, s, 3-H), 5.18 (2H, s, CH_2Ph), 5.58 (1H, s, 5-H), 5.64 (1H, d, J 7Hz, NCHCO), 7.24 - 7.66 (8H, m, aromatics), 8.16(1H, s, CHO), 8.23 and 8.88 (2H, 2br s,2NH's), 10.05 (1H, d, J 7Hz. NH CHCO).

(c) 6β-[D-2-[3,4-bis(ethoxycarbonyloxy)phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]acetamido]-6α-formamidopenicillanic acid, sodium salt.

Benzyl 6β-[D-2-[3,4-bis(ethoxycarbonyloxy)phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]acetamido]-6α-formamidopenicillanate (570mg; 0.7mMole) was hydrogenated over 10% palladium on charcoal in tetrahydrofuran (25ml) until complete (ca. 1h). Then the catalyst was removed by filtration and washed with tetrahydrofuran (10ml). The filtrate was treated with 1.87M. sodium 2-ethylhexanoate in 4-methylpentan-2-one (0.37ml; 0.7mMole), and ether (100ml) was added. The precipitate was collected by filtration, washed with ether and dried in vacuo; (470mg, 90%); $\nu_{max.}$ (KBr) 3700 - 2600, 1770, 1712, 1678, 1608, 1503, 1392, 1370, 1255 and 1190cm^{-1}; δ[D_2O] 0.89 and 1.26 [6H, 2s, 2-CH_3's) 1.17 (3H, t, J8Hz, $NCH_2\underline{CH_3}$), 1.32 (6H, t, J8Hz, $2\underline{CH_3}CH_2O$'s), 3.49 (2H, q, J8Hz $N\underline{CH_2}CH_3$), $\underline{CH_2}N$,), 3.98 (2H, q, J 8Hz,$NCH_2\underline{CH_2}N$), 4.06 (1H, s, 3-H), 4.13 (4H, q, J8Hz, $2CH_3\underline{CH_2}O$'s), 5.49 (1H, s, 5-H), 5.59 (1H, s, CH), 7.37 - 7.58 (3H, m, aromatics), 8.12 (1H,s, CHO); MIC against Proteus mirabilis 889 is 0.05 μg/ml

Example 2

Sodium 6α-formamido-6β-[D-2-[3,4-(methylenedioxy)phenyl]-2-(2,3-dioxo-4-ethylpiperazin-1-ylcarbonylamino)acetamido] penicillanate.

(a) Benzyl 6α-methylthio-6β-[2-[3,4-(methylen edioxy) phenyl]-2-(4-nitrobenzyloxycarbonylamino)acetamido] penicillanate.

A solution of benzyl 6β-amino-6α-(methylthio)penicillanate (3.52g, 10mmol) and N,N'-dicyclohexylcarbodiimide (2.27g, 11mmol) in dichloromethane (25ml) was stirred and cooled to 0-5°C and treated dropwise with a solution of 2-[3,4-(methylenedioxy)phenyl]-2-(4-nitrobenzyloxycarbonylamino) acetic acid (3.74g, 10mmol) in acetone (40ml). The reaction mixture was then stirred at room temperature for 5.5 hour, filtered, evaporated in vacuo, and the residue dissolved in dichloromethane (60ml). This solution was washed with dilute hydrochloric acid (30ml), sodium bicarbonate solution (1M,30ml) and brine (30ml), dried and evaporated in vacuo to give a yellow foam. This was chromatographed on silica gel 60 (<230 mesh ASTM), using 20-25% ethyl acetate in cyclohexane as eluant, to give the title compound (4.22g, 60%); δ[$(CD_3)_2CO$] 1.20-1.60 (6H,m,2xCH_3), 2.00 and 2.27(3H,2s,SCH_3), 4.33 and 4.40 (1H,2s,3-H), 5.10 (4H,s,2xCH_2), 5.30 - 5.60(2H,m, 5-H and α-H), 5.83(2H,s,O-CH_2-O), 6.53-7.16 (3H,m,aromatics), 7.23(5H,s,benzylester aromatics), 7.34 and 7.50 (2H,2s, part AA'BB', PNB aromatics), 7.63(1H,s,amide), 7.92 and 8.08(2H,2s, part AA'BB' PNB aromatics), 8.47 (1H,d,amide).

(b) Benzyl 6α-formamido-6β-[2-(3,4-methylen edioxyphenyl) -2-(4-nitrobenzyloxycarbonylamino)acetamido]penicillanate.

A solution of benzyl 6α-methylthio-6β-[2-(3,4-methylene-dioxyphenyl)-2-(4-nitrobenzyloxycarbonylamino)acetamido] penicillanate (4.03g, 5.7mmol) in DMF (25ml) at -40°C under argon, was treated with mercuric acetate (1.82g, 5.7mmol), followed immediately by a solution of anhydrous ammonia (0.116g, 6.8mmol) in DMF (4.7ml). The mixture was allowed to warm to room temperature, and then stirred for 1 hour. After filtration, the solution was diluted with ethyl acetate (100ml), washed with water and brine, dried over magnesium sulphate, filtered and evaporated in vacuo to give an orange foam (3.8g, 98%). This foam was dissolved in dry dichloromethane (40ml), cooled to 0°C, and treated sequentially with pyridine (4.5ml, 56mmol) and acetic formic anhydride (2.2ml, 28mmol). The reaction mixture was stirred at room temperature for 1¼ hour before being washed with dilute hydrochloric acid (2x50ml), water (50ml) and brine (50ml). It was dried over magnesium sulphate, filtered and evaporated in vacuo to give an orange foam (3.5g). This was chromatographed on silica gel 60 (230 mesh ASTM), using 40-50% ethyl acetate in cyclohexane as eluant, to afford the title compound as its two separate isomeric forms; L-isomer (1.069g, 27%) and D-isomer (0.966g, 24%); δ ($(CD_3)_2CO$) for D-isomer, 1.00-1.40 (6H,m,2xCH_3), 4.43(1H,s,3-H), 5.10 and 5.15(4H,2s, 2xCH_2), 5.40-5.73(2H,m,5-H and α-H), 5.87 (2H, s, O-CH_2-O), 6.53-7.16(3H,m,aromatics), 7.27 (5H,s, benzyl ester aromatics), 7.36 and 7.52 (2H,2s,part AA'BB' DNB aromatics), 7.65(1H,s,amide), 8.78(1H,broad, s, amide).

(c) 6α-formamido-6β-[D-2-[3,4-(methylenedioxy)phenyl]-2-aminoacetamido]penicillanic acid.

Benzyl 6α-formamido-6β-[D-2-[3,4-(methylenedioxy)phenyl]-2-

(4-nitrobenzyloxycarbonylamino)acetamido]penicillanate (0.960g, 1.36mmol) was dissolved in THF (25ml) and water added until the solution almost went cloudy, then 10% palladium on charcoal (0.96g) was added under an inert atmosphere. This mixture was hydrogenated for 2 hour, the catalyst filtered off, and the THF removed in vacuo. The aqueous solution was then washed with ethyl acetate (2x25ml) and ether (25ml) and freeze-dried to give the title compound as an off-white powder (0.457g, 77%); ν_{max}(KBr disc) 3410, 3191,2985, 1772, 1684, 1609, 1501, 1251cm^{-1}; δ (D_2O) 0.96 and 1.35(6H,2s,2xCH_3), 4.18(1H,s, 3-H), 5.08(1H,s,α-H), 5.63(1H,s,5-H), 6.01(2H,s,O-CH_2-O), 6.85-7.20 (3H,m,aromatics), 8.14(1H,s,CHO).

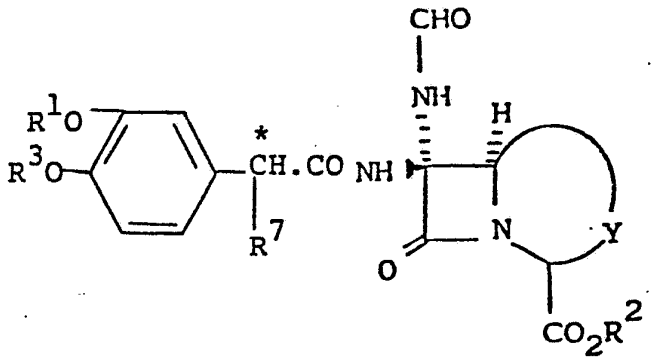
(d) Sodium 6α-formamido-6β-[D-2-[3,4-(methylenedioxy) phenyl]-2-(2,3-dioxo-4-ethylpiperazin-1-ylcarbonylamino) acetamido]penicillanate.

A solution of 6α-formamido-6β-[D-2-[3,4-(methylenedioxy) phenyl]-2-aminoacetamido]penicillanic acid (0.397g, 0.91 mmol) in water (25ml) was treated with a solution of 2,3-dioxo-4-ethylpiperazin-1-ylcarbonyl chloride (0.223g, 1.09 mmol) in acetone (8ml), while maintaining the pH at 7-7.2. The solution was then stirred at this pH for 1 hour, washed with ethyl acetate(20ml), acidified to pH 2.5, and extracted with ethyl acetate (4x30ml). The combined organic extracts were dried over magnesium sulphate, filtered and evaporated in vacuo to give the free acid as a pale pink solid (0.439g). This was dissolved in acetone (4ml) and treated with sodium 2-ethylhexanoate in 4-methylpentan-2-one (1.85N, 0.38ml). The sodium salt which precipitated was filtered off, washed with acetone then diethyl ether, and then dried in vacuo to yield the title compound as a white powder (0.436g, 77%); $\nu_{max.}$ (KBr disc) 3431, 1771, 1711, 1676, 1609, 1501, 1487, 1397, 1368, 1244, 1190cm^{-1}; δ (D_2O) 0.96 and 1.30(6H,2s, gem dimethyl),

1.20 (3H,t,J7.5Hz,CH_3), 3.51(2H,q,J7.5Hz,CH_2), 3.67 and 3.97 (4H,2m, 2xCH_2), 4.17(1H,s,3-H), 5.36(1H,s,α-H), 5.59 (1H,s,5-H), 5.97(2H,d,J1Hz,O-CH_2-O), 6.88(1H,d,J7.5Hz, aromatic 6-H), 6.96-7.05 (2H,m,aromatic 2 and 5-H), 8.12 (1H,s,CHO) (Found: MH^+, 627.1509. $C_{25}H_{28}N_6NaO_{10}S$ requires m/z 627.1485). MIC against Proteus mirabilis 889 is 1.0 µg/ml.

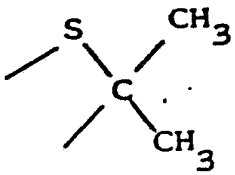
CLAIMS:

1 A compound of formula (I) or a salt thereof.



(I)

wherein R^1 and R^3 are independently an in vivo hydrolysable group, or R^1 and R^3 together form an in vivo hydrolysable group, provided that R^1 and R^3 are not both C_{1-6} alkylcarbonyl; R^7 is a hydroxyl, carboxylic acid group or lower alkyl or phenyl, tolyl or indanyl ester thereof, amino or a substituted amino group; R^2 is hydrogen or a readily removable carboxyl protecting group; and Y is:



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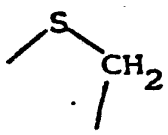
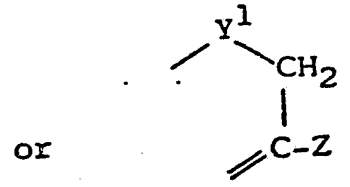
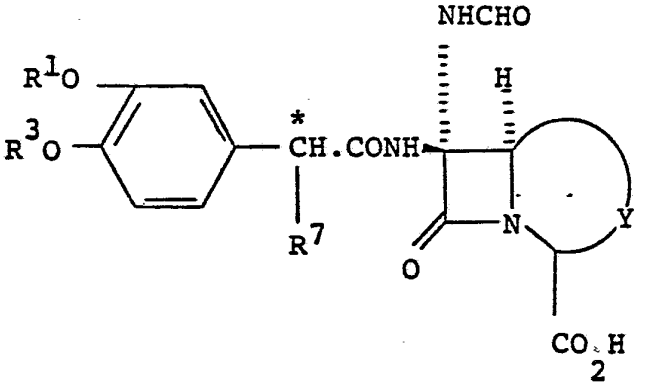




or ;

wherein Y^1 is oxygen, sulphur or $-CH_2-$ and Z represents hydrogen, halogen, or an organic group such as C_{1-4} alkoxy, $-CH_2Q$ or $-CH=CH-Q$ wherein Q represents hydrogen, halogen, hydroxy, mercapto, cyano, carboxy, carbamoyloxy, carboxylic ester, C_{1-4} alkyloxy, acyloxy, aryl, a heterocyclyl group bonded via carbon, a heterocyclylthio group or a nitrogen containing heterocyclic group bonded via nitrogen.

2. A compound as claimed in claim 1 of formula (II) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:



(II)

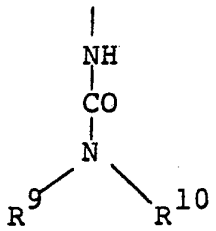
wherein R^1, R^3, Y and R^7 are as defined with respect to formula (I).

3. A compound as claimed in claim 1 or claim 2 wherein R^1 and R^2 are both the same and represent C_{1-6} alkoxycarbonyl.

4. A compound as claimed in claim 1 or claim 2 wherein R^1 and R^2 are joined and together represent C_{1-6} alkylene.

5. A compound as claimed in any one of claims 1 to 4 wherein Y is $-S-C(CH_3)_2-$.

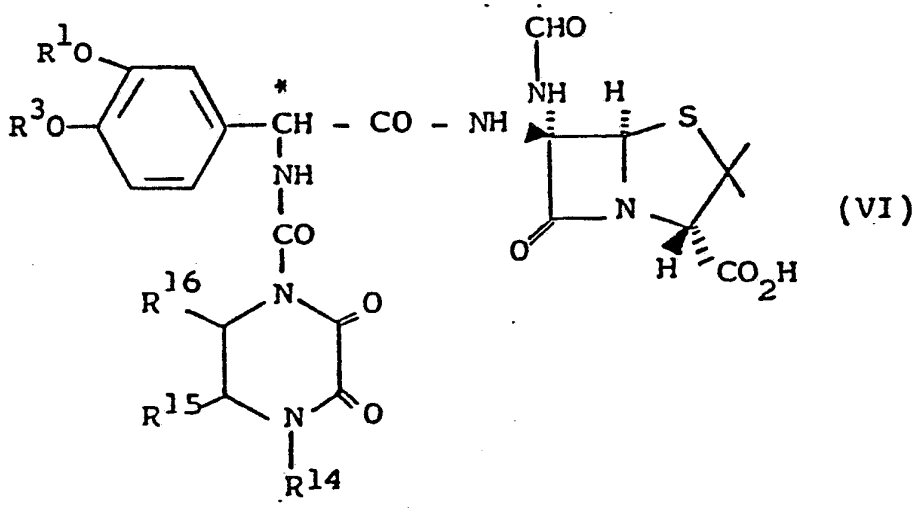
6. A compound as claimed in any one of claims 1 to 5 wherein R^7 is of formula (III):



(III)

wherein R^9 is hydrogen or a C_{1-6} alkyl group and R^{10} is an optionally substituted 5- or 6- membered heterocyclic group containing one or two nitrogen heteroatoms; or R^9 and R^{10} together with the nitrogen atom to which they are attached form an optionally substituted five- or six- membered heterocyclic group containing one or two nitrogen heteroatoms.

7. A compound as claimed in any one of claims 1 to 6 of formula (VI) or a pharmaceutically acceptable salt or <u>in vivo</u> hydrolysable ester thereof:



(VI)

wherein R^1 and R^3 are as defined with respect to formula (I); R^{14} represents hydrogen, C_{1-6} alkyl, substituted alkyl, aryl, or aralkyl; R^{15} and R^{16} are the same or different and represent hydrogen, C_{1-6} alkyl, substituted alkyl, halogen, amino, hydroxy or C_{1-6} alkoxy; or R^{15} and R^{16} form the residue of 5- or 6- membered carbocyclic or heterocyclic ring.

8. A compound as claimed in claim 1 selected from the following or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:

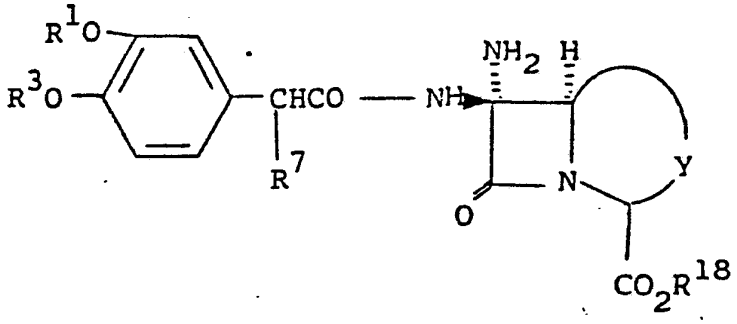
6β-[D-2-[3,4-bis(ethoxycarbonyloxy)phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]acetamido]-6α-formamidopenicillanic acid; and

6α-formamido-6β-[D-2-[3,4-(methylenedioxy)phenyl]-2-[(2,3-dioxo-4-ethylpiperazin-1-yl)carbonylamino]-acetamido]penicillanic acid.

9. A pharmaceutical composition comprising a compound of formula (II) as claimed in claim 2 together with a pharmaceutically acceptable carrier or excipient.

10. A process for the preparation of a compound as claimed in claim 1 which process comprises:

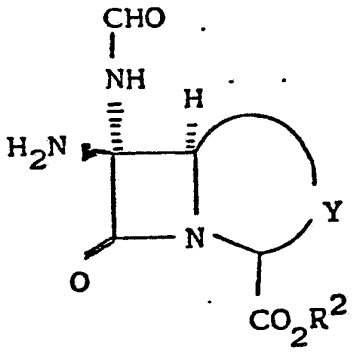
a) formylating a compound of formula (X):



(X)

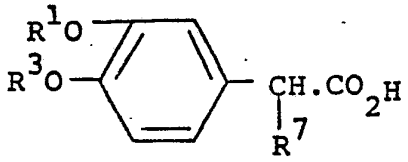
wherein R^1, R^3, R^7 and Y are as defined with respect to formula (I), wherein any reactive groups may be protected; and R^{18} is a readily removable carboxy protecting group; or

b) reacting a compound of formula (XVII):



(XVII)

wherein the amino group is optionally substituted with a group which permits acylation to take place and R^2 is as defined with reference to formula (I) above, with an N-acylating derivative of an acid of formula (XVIII):



(XVIII)

wherein R^1, R^3 and R^7 are as defined with respect to formula (I) and wherein any reactive groups therein may be protected;
and after either of processes a) or b) carrying out, if necessary, one or more of the following steps:

i) converting a group R^{18} to a group R^2;
ii) converting one group Z into a different group Z;
iii) converting the product into a salt.

INTERNATIONAL SEARCH REPORT

International Application No PCT/GB 84/00010

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) [2]

According to International Patent Classification (IPC) or to both National Classification and IPC

IPC[3]: C 07 D 499/68; 501/20; 498/04; 471/04;// A 61 K 31/43; C 07 D 241/08 (C 07 D 498/04, 265/00, 205/00) ./.

II. FIELDS SEARCHED

Minimum Documentation Searched [4]

Classification System	Classification Symbols
IPC[3]	C 07 D 499/00; C 07 D 501/00; C 07 D 498/00; C 07 D 471/00; A 61 K 31/00

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched [5]

III. DOCUMENTS CONSIDERED TO BE RELEVANT [14]

Category *	Citation of Document, [16] with indication, where appropriate, of the relevant passages [17]	Relevant to Claim No. [18]
A	EP, A, 0066373 (BEECHAM) 8 December 1982 see claims	1,9,10
P,A	EP, A, 0071395 (BEECHAM) 9 February 1983 see claims	1,9,10

* Special categories of cited documents: [15]

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

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"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step

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IV. CERTIFICATION

Date of the Actual Completion of the International Search [3]	Date of Mailing of this International Search Report [3]
18th April 1984	17 MAI 1984
International Searching Authority [1]	**Signature of Authorized Officer [20]**
EUROPEAN PATENT OFFICE	G.L.M. Kruydenberg

INTERNATIONAL SEARCH REPORT

International Application No PCT/GB 84/00010 -2-

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) [3]

According to International Patent Classification (IPC) or to both National Classification and IPC

IPC[3]: (C 07 D 471/04; C 07 D 221/00; C 07 D 205/00)

II. FIELDS SEARCHED

Minimum Documentation Searched [4]

Classification System	Classification Symbols
IPC[3]	

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched [5]

III. DOCUMENTS CONSIDERED TO BE RELEVANT [14]

Category *	Citation of Document, [16] with indication, where appropriate, of the relevant passages [17]	Relevant to Claim No. [18]

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IV. CERTIFICATION

Date of the Actual Completion of the International Search [2]	Date of Mailing of this International Search Report [2]
International Searching Authority [1]	**Signature of Authorized Officer** [20]
EUROPEAN PATENT OFFICE	G.L.M. Kruydenberg

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON

INTERNATIONAL APPLICATION NO. PCT/GB 84/00010 (SA 6436)

This Annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The members are as contained in the European Patent Office EDP file on 10/05/84

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Patent document cited in search report	Publication date	Patent family member(s)		Publication date
EP-A- 0066373	08/12/82	JP-A-	57200393	08/12/82
		AU-A-	8400282	25/11/82
		GB-A-	2119363	16/11/83
EP-A- 0071395	09/02/83	JP-A-	58038288	05/03/83
		GB-A-	2107307	27/04/83