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(72) Inventeurs/Inventors:

(54) Titre : SYNTHESE ET UTILISATION DE FLUORURES D'ACIDES AMINES COMME REACTIFS DANS LE COUPLAGE DE PEPTIDES

(54) Title: SYNTHESIS AND USE OF AMINO ACID FLUORIDES AS PEPTIDE COUPLING REAGENTS

$$BLK - \underset{\displaystyle X}{\underset{|}{AA}} - F \qquad (I)$$

(57) Abrégé/Abstract:
A compound of formula (I) (see formula I) or the acid fluoride salts thereof, wherein BLK is an N-amino protecting group or hydrogen; AA is an amino acid residue and X is H or a protecting group, is useful as a coupling agent in a peptide synthesis.

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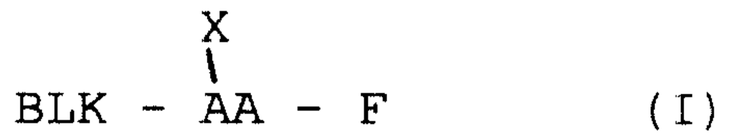
(72) Inventeurs(suite)/Inventors(continued): CARPINO, LOUIS A., US; SADAT-AALAEE, DEAN, US

(73) Propriétaires(suite)/Owners(continued):UNIVERSITY OF MASSACHUSETTS, US

(74) Agent: MCFADDEN, FINCHAM

ABSTRACT

A compound of formula (I)



(I)

or the acid fluoride salts thereof, wherein BLK is an N-amino protecting group or hydrogen; AA is an amino acid residue and X is H or a protecting group, is useful as a coupling agent in a peptide synthesis.

SYNTHESIS AND USE OF AMINO ACID FLUORIDES AS PEPTIDE COUPLING REAGENTS

The present invention relates to novel amino acid fluorides and protected amino acid fluorides and their use in synthetic biochemistry, including peptide syntheses. More particularly, this invention is directed to the N-protected amino acid fluorides, and free amino acid fluorides and the hydrogen fluoride salts thereof, the side chain of which may be unprotected or protected with a blocking group and their use thereof in peptide synthesis.

As more and more polypeptides become of medicinal importance, there is an increasing incentive to improve the methods by which they may be synthesized. In recent years, peptides which have been found to be of possible pharmacological importance include those active against various diseases, such as cancers, diabetes, and plant toxins, etc. Others have shown specific activity as growth promoters or suppressants, antibiotics, insecticides, contraceptives, anti-hypertensives, sleep-inducers, anti-depressants, analgesics, etc. The list is long and varied.

Currently, syntheses of peptides in solution by classic or various repetitive methods or on a solid support (Merrifield) are popular techniques. Solution methods have the advantages of being easily monitored and allowing purification of intermediates, if necessary, at any stage. A major drawback is the relative slow pace of the synthesis with each step being carried out manually.

The major advantage of the Merrifield Method is its easy automation so that unattended, computer-controlled machine syntheses is possible. Unfortunately, this method suffers from an inherent deficiency due to the insoluble nature of

the support on which the synthesis proceeds. Unless each acylation step occurs with 100% efficiency, mixtures will inevitably be built up on the polymer. The longer the chain, the greater will be the contamination due to undesired side reactions. Products produced in such reactions remain to contaminate the desired product when at the end of the cycle it is removed from the polymeric matrix. The properties of these peptides will not differ sufficiently for peptides of greater than about 30-40 residues to make efficient separation feasible.

For very long segments (50 or more amino acids), therefore, current methods are not satisfactory. Often mixtures are obtained of such forbidding complexity that it may be difficult or impossible to isolate the desired peptide.

The problems enumerated hereinabove could be eliminated if the proper derivatives of the underlying amino acids and the proper reaction conditions could be found.

For example, FMOC, (Nα-(9-fluorenylmethyl)oxycarbonyl), protected amino acid chlorides, which are described by Carpino, et al. in J. Org. Chem. 51, 3732 (1986) have been used as acylating agent for stepwise peptide syntheses for both solution and solid phase techniques.

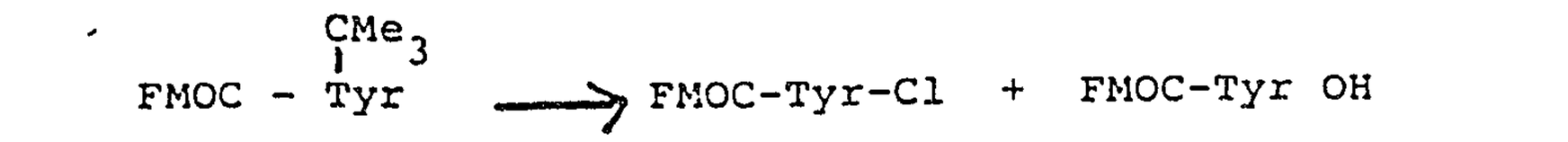
However, the amino acid chlorides have major drawbacks associated therewith. First, the acid chlorides react with trace amounts of water, such as moisture in the air, to give the corresponding amino acid. Therefore, they are not so stable, and as such, they are not a prime candidate for long term storage. Consequently, an objective was to find an amino acid derivative which was stable to moisture.

Moreover, another problem associated with amino acid chlorides is that it has not been possible to date to synthesize amino acid chlorides in which the protecting

groups on the side chains of the amino acids can be removed under extremely mild conditions. As one skilled in the art is well aware, many of the amino acids have functional groups on the side chains which can interfere with peptide formation unless otherwise protected. In peptide synthesis, only the mildest conditions should be used to remove these protecting groups. For example, one of the easiest protecting groups to remove from the side chains containing amino, hydroxyl or carboxyl functions, such as lysine, tyrosine, threonine, serine, aspartic acid, glutamic acid and the like, is t-butyl or t-butyl containing moieties. For example, trifluoracetic acid can easily remove the t-butyl group from a serine side chain; on the other hand, a benzyl protecting group on the side chain can not be removed by said treatment but instead requires a more potent acid such as HF or trifluoromethane-sulfonic acid. Therefore, the conditions for removing the benzyl group from the side chain are much harsher relative to the t-butyl groups. Furthermore, the mild catalytic hydrogenolysis of benzyl groups is not generally applicable to long chain peptides or resin attached peptides.

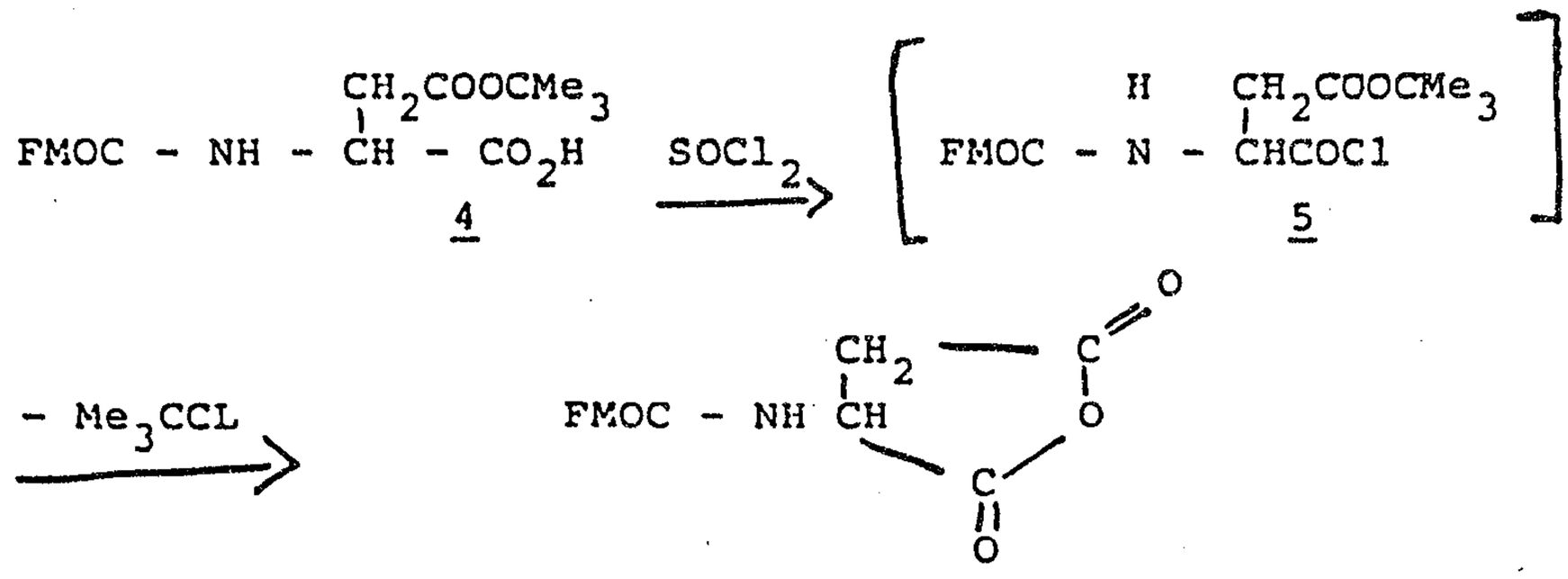
Although benzyl groups on the side chains of N-protected amino acid chloricles, can be prepared, such as FMOC-cysteine-S-benzyl chloride, FMOC-lysine- -carbobenzoxy chloride, FMOC-tyrosine-O-benzyl chloride, FMOC-serine-O-benzyl chloride and FMOC aspartic acid -benzyl ester, these molecules suffer from the disadvantages described hereinabove. Consequently, an investigation was commenced to determine if t-butyl or "t-butyl like" containing groups can be used to protect the side chain of amino acid chlorides. Unfortunately, efforts in this area were unsuccessful. None of the above compounds could be synthesized if the t-butyl group was used in place of the benzyl substitution.

This was not unusual since it is well known that t-butyl-based protecting groups are readily deblocked by hydrogen chloride which is an inevitable by-product of acid chloride formation and/or long term storage (hydrolysis by trace amounts of water). For example, in the case of the FMOC-tyrosine derivative 1, the acid chloride could be obtained, but after several days it was noted to lose the t-butyl group slowly.



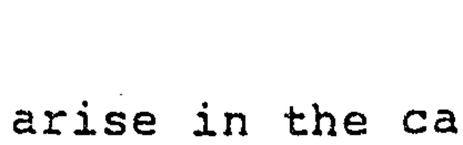
Furthermore, compound 1 as well as the analogous serine and threonine derivatives could be obtained only as oils which could not be crystallized and were therefore difficult, if not impossible, to purify.

In the case of the FMOC aspartic acid derivative 4, treatment with thionyl chloride gave only the aspartic acid anhydride 6, presumably via the unstable acid chloride 5 which undergoes intramolecular loss of t-butyl chloride.



SUBSTITUTE SHEET

Lysine derivative 7 could not be converted to an acid chloride because of the marked sensitivity of the BOC function.



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Similar problems arise in the cases of Arg, His, Asn, Gln, and Trp. The net result of these problems is that only about one half of the commonly occurring amino acids can be converted to stable amino acid chlorides.

Therefore, a search was undertaken to find an amino acid candidate for use in peptide synthesis which is inexpensive, stable to moisture, and which shows great potential for long-term storage. Moreover, it was hoped that a candidate could be found where the protecting groups on the amino acid side chain could be removed under milder conditions then those used to remove the benzyl group. Preferably, it was hoped that a t-butyl containing group or a group as easily removable as t-butyl could be placed on the side chain of these amino acid candidates.

The present invention circumvents the difficulties experienced with respect to the acid chlorides and accomplishes the goals described hereinabove. The compounds of the present invention are effective in coupling with amino acids or peptides to form new peptide bonds. Moreover, the compounds of the present invention are more stable to moisture then the acid chlorides and therefore can be used for long term storage. Furthermore, t-butyl containing protecting groups and other protecting groups can be placed on the side chains of these amino acid compounds and removed under milder conditions than those required for the removal of benzyl groups. Finally, the compounds of the present

invention are potent acylating agents in peptide bond formation.

These compounds are, much to our surprise, the corresponding amino acid fluorides.

The present invention is directed to an amino acid of the formula:

or the hydrogen fluoride salts thereof

wherein

BLK is hydrogen or an N-amino protecting group;

AA is an amino acid residue; and

X is - or a protecting group.

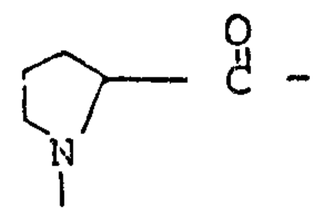
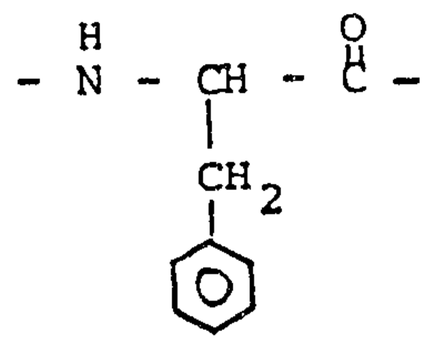
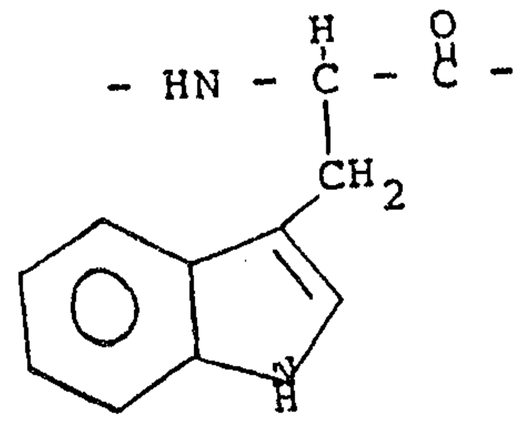
The present invention is also directed to a method for preparing a peptide which comprises reacting the amino acid fluoride described hereinabove with an amino acid or peptide having a free amino group and removing the protecting groups therefrom.

As used herein, the term "amino acid" refers to an organic acid containing both a basic amino group (NH_2) and an acidic carboxyl group (COOH). Therefore, said molecule is amphoteric and exists in aqueous solution as dipole ions. (See, "The Condensed Chemical Dictionary", 10th ed. edited by Gessner G. Hawly, Van Nostrand Reinhold Company, London, Eng. p.48 (1981)). The preferred amino acids are the -amino acids. They include but are not limited to the 25 amino acids that have been established as protein constituents. They must contain at least one carboxyl group and one primary or secondary amino group on the amino acid molecule. They include such proteinogenic amino acids as alanine, valine, leucine, isoleucine, norleucine, proline, hydroxyproline,

phenylalanine, tryptophan, methionine, glycine, serine, threonine, cysteine, cystine, tyrosine, asparagine, glutamine, aspartic acid, glutamic acid, lysine, hydroxylysine, ornithine, arginine, histidine, penicillamine and the like.

An "amino acid residue", as defined herein, is an amino acid minus an amine hydrogen on the amino end of the molecule and the OH group on the carboxy end of the molecule (i.e., it includes the acyl group $-C(=O)-$ on the carboxy end of the molecule). Therefore, unless designated to the contrary, the group "AA" signifies an amino acid residue. For example, the amino acid residues of various amino acids are represented below:

AA Symbol	AA Residue
Gly	$-NH-CH_2-C(=O)-$
Ala	$-NH-CH(CH_3)-C(=O)-$
Leu	$-NH-CH(CH_2-CH(CH_3)_2)-C(=O)-$
Ile	$-NH-CH(CH(CH_3)-CH_2-CH_3)-C(=O)-$

1	Pro	
5	Phe	
10	Trp	
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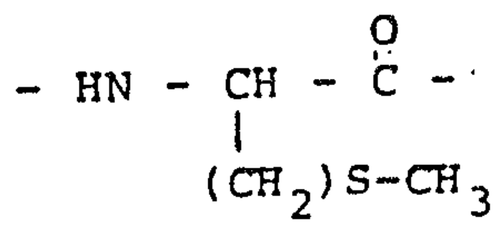
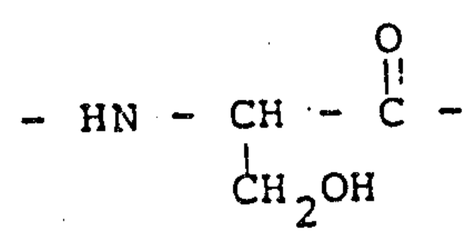
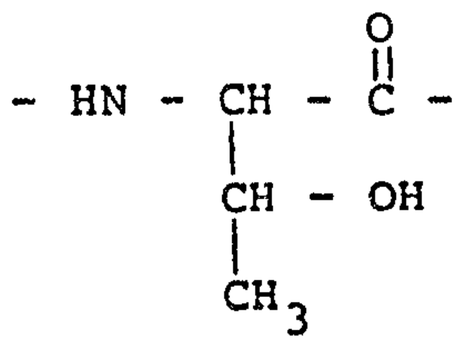
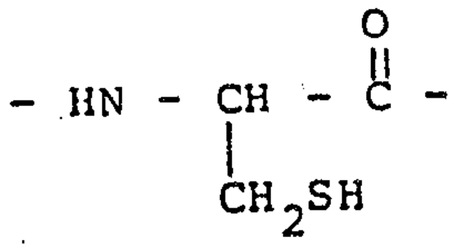
20	Met	
25	Ser	
	Thr	
30	Cys	
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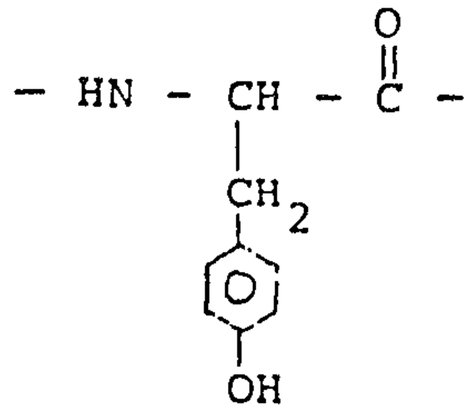
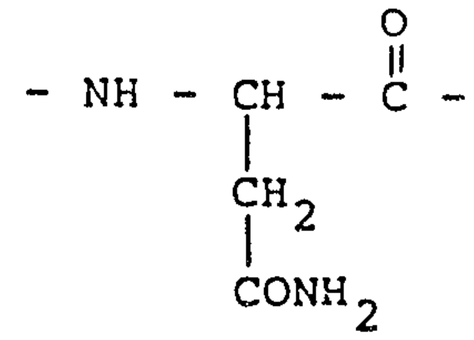
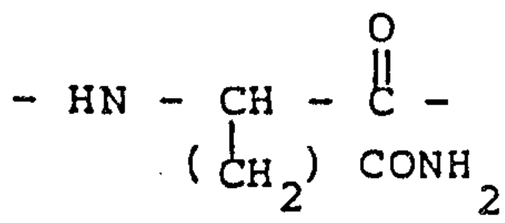
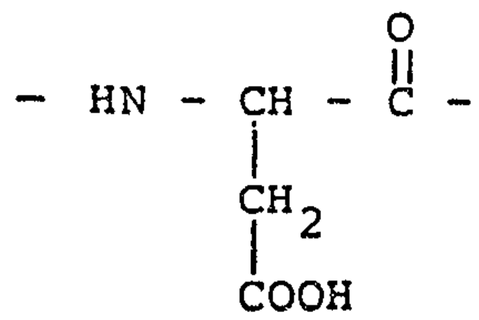
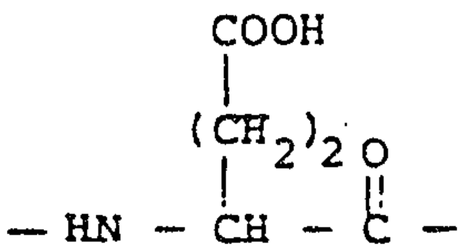
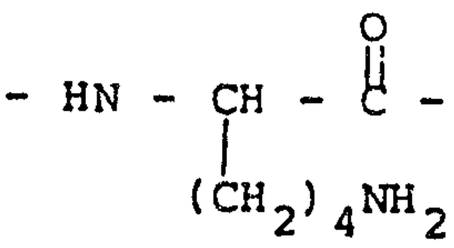
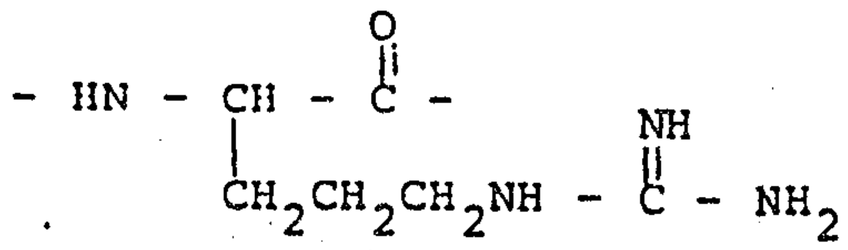
1	Tyr	
5	Asn	
10	Gln	
	Asp	
20	Glu	
25	Lys	
30	Arg	
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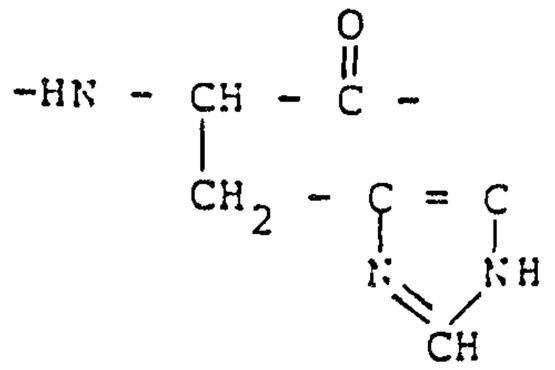
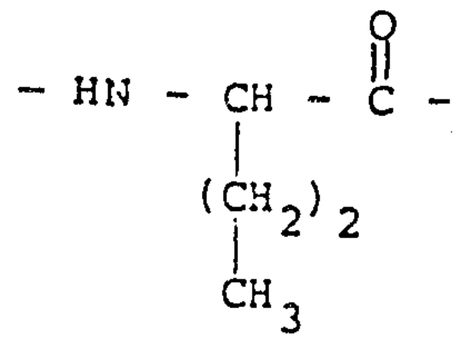
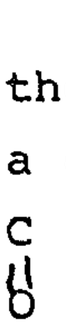
His	
Nor	

Therefore, the symbol "AA-F" refers to an amino acid fluoride, i.e., a compound having a fluoro group attached to the acyl group  of the amino acid. When BLK is hydrogen, then the structure becomes the amino acid fluoride of an amino acid having a free amino group. However, in view of the synthesis of the amino acid fluoride described hereinbelow, the free amino acid fluoride may be isolated as the hydrogen fluoride salt thereof.

It will be apparent to one skilled in the art, shown by exemplification in the table hereinabove that in the course of protein synthesis, it may be necessary to protect certain side chains of the amino acids to prevent unwanted side reactions. For example, it may be necessary to protect the hydroxyl group on the side chain of tyrosine, serine, or threonine in order to prevent these groups from interfering with the desired reactions. This is a common problem in peptide synthesis and many procedures are available for protecting the functional groups on the side chains of the amino acids. Such procedures for protecting various functional groups are known to one skilled in the art and are described in the treatise entitled "The PEPTIDES", Vol. 2,

Edited by E. Gross and J. Meienhoffer, Academic Press, NY, NY, PP. 166-251 (1980), and the book entitled "Reagents for Organic Synthesis", by T.W. Green, John Wiley and Sons, New York, 1981.

For example, when the functional side chain contains an hydroxy group, such as threonine or serine, it can be protected by such groups as methyl, methoxymethyl(MOM), 2-methoxyethoxymethyl(MEM), tetrahydropyranyl, β-trimethylsilylethyl, 4-methoxytetrahydropryanyl, 1-ethoxyethyl, t-butyl, p-methoxybenzyl, p-halobenzyl, o-nitrobenzyl, p-nitrobenzyl, o-chlorobenzyl, adamantyl, diphenylmethyl, triphenylmethyl, cyclohexyl, cyclopentyl, 1-benzyloxycarbonyl, tri-substituted silyl, wherein the substituents are independently aryl, alkyl or aralkyl, 2,2,2-trifluoroethyl, and the like. The preferred groups for the protection of the hydroxyl side chain are adamantyl, t-butyl, 4-methoxybenzyl, cyclopentyl and cyclohexyl.

When the side chain contains a phenol, such as in tyrosine, it may be protected by such groups as methyl, methoxymethyl(MOM), methoxyethoxymethyl(MEM), -trimethylsilylethyl, methylthiomethyl, tetrahydropyranyl, isopropyl, cyclohexyl, cyclopentyl, t-butyl, adamantyl, 4-methoxysilyl, o-nitrobenzyl, 2,4-dinitrophenyl, m-bromobenzyl, 2,6-dichlorobenzyl, trisubstituted-silyl wherein the substituents are independently alkyl, aryl or aralkyl, ethoxycarbonyl, carbamoyl and the like. The most preferred protecting groups are adamantyl, 4-methoxybenzyl, t-butyl, cyclopentyl, and cyclohexyl. An especially preferred protecting group is t-butyl.

A carboxy side chain, such as that found in aspartic acid or glutamic acid, can be protected by the following groups: 1-or 2-adamantyl, methoxymethyl, methythiomethyl, t-butyl, methyl, ethyl, phenyl, tetrahydropyranyl,

cyclopentyl, cyclohexyl, cycloheptyl, 4-picolyl, trisubstituted-silyl wherein the substituents are independently alkyl, aryl or aralykl, N-piperidinyl, N-succinimidoyl, β-trimethylsilylethyl, 4-methoxybenzyl, benzyl, p-bromobenzyl, p-chlorobenzyl, p-nitrobenzyl, phenacyl, N-phthalimidoyl, 4-alkyl-5-oxo-1,3-oxazolidinyl, trisubstituted-stannyl wherein the substituents are independently alkyl, aryl or aralkyl, and the like. The most preferred protecting group is t-butyl.

If the functional group on the side chain is mercapto, e.g., cysteine, such groups as triphenylmethyl, benzyl, 4-methylbenzyl, 3,4-dimethylbenzyl, 4-methoxybenzyl, β-trimethylsilylethyl, p-nitrobenzyl, 4-picolyl, diphenylmethyl, triphenylmethyl, bis(4-methoxyphenyl)methyl, diphenyl-4-pyridylmethyl, 2,4-dinitrophenyl, t-butyl, t-butylthio, adamantyl, isobutoxymethyl, benzylthiomethyl, thiazolidinyl, acetamidomethyl, (ACM) benzamidomethyl, 2-nitro-1-phenylethyl, 2,2-bis(carboethoxy)ethyl, 9-fluorenemethyl, acetyl, benzoyl, and the like can be used to protect said group. In this case, it is preferred that the protecting groups are t-butyl, t-butylthio, 4-methoxybenzyl, and triphenylmethyl.

If the side chain contains an amino group, such as the ε-amino group of lysine and ornithine, the following groups may be used: 9-fluorenylmethyloxycarbonyl, 9-(2-sulfo)fluorenylmethyloxycarbonyl, β-trimethylsilyl-ethyloxycarbonyl, 2-furanylmethyloxycarbonyl, adamantyloxycarbonyl, carbobenzoxy, t-butyloxycarbonyl, t-amyloxycarbonyl, cyclobutyloxycarbonyl, 1-methylcyclobutyloxycarbonyl, cyclopentyloxycarbonyl, cyclohexyloxycarbonyl, 1-methylcyclohexyloxycarbonyl, isobornyloxycarbonyl, benzyl, p-methoxybenzyloxycarbonyl, p-nitrobenzyloxycarbonyl, chlorobenzyloxycarbonyl, isonicotinyloxycarbonyl, p-toluenesulfonylamidocarbonyl, methylsulfonylethyloxycarbonyl,

β,β,β-trichloroethyloxycarbonyl, dithiasuccinoyl, phthaloyl, 4,5-diphenyl-4-oxazoline-2-one, piperidino oxycarbonyl trifluoroacetyl, chloroacetyl, p-toluenesulfonyl and the like. Preferred groups for the protection of this amino side chain are carbobenzoxycarbonyl, t-butyloxycarbonyl, and adamantyloxycarbonyl.

If the amino acid has an imidazole group, such as in histidine, the following groups may be used to protect the side chain: benzyloxymethyl, piperdinylcarbonyl, phenacyl, pivaloyloxymethyl, 1-(alkoxycarbonylamino)- 2,2,2-trifluoroethyl, 1-trifluoromethyl-1-(p-chlorophenoxy-methoxy)-2,2,2-trifluoroethyl, 2,4-dinitrophenyl, toluenesulfonyl, FMOC, triphenylmethyl, t-butyloxycarbonyl, t-butyloxymethyl and the like. The preferred groups are FMOC, t-butyloxycarbonyl, triphenylmethyl, and t-butyloxymethyl.

When the amino acid has a guanidine side chain, such as in arginine, the following protecting groups can be used to protect the ω nitrogen on the guanidine moiety: methoxytrimethylbenzenesulfonyl, pentamethylchromane-sulfonyl, mesitylenesulfonyl, tolunesulfonyl, 2,4,6-tri-methylbenzenesulfonyl, trimethoxybenzensulfonyl, bisadamantyloxylcarbonyl, nitro, tosyl, and the like. The preferred protecting groups are methoxytrimethylsulfonyl, pentaamethylchromanesulfonyl, bisadamantyloxycarbonyl, and mesitylenesulfonyl.

For side chains containing an amide group such as in glutamine and asparagine, the following groups can be used to protect the side chain: dimethoxybenzyhydryl, 9-xanthenyl, 2,4,6-trimethoxybenzyl, and the like.

As used herein, the term "alkyl", when used alone or in combination with other groups refers to a carbon chain containing from 1 to 6 carbon atoms. They may be straight

chains or branched and include such groups as methyl, ethyl, propyl, isopropyl, n-butyl, sec-butyl, isobutyl, t-butyl, n-pentyl, amyl, n-hexyl and the like. The preferred alkyl group contains from 1-3 carbon atoms. The term aryl as used herein refers to an aromatic ring system containing from 6-10 ring carbon atoms and up to a total of 15 carbon atoms. It includes such groups as phenyl, α-naphthyl, β-naphthyl, and the like. The preferred group is phenyl.

Aralkyl groups are aryl groups attached to the main chain through an alkylene bridge. Such groups include benzyl, phenethyl and the like. The preferred aralkyl group is benzyl.

The aryl and aralkyl groups herein may be unsubstituted or may be substituted with an electron donating groups in situations wherein the protecting group is cleaved by acid. An electron donating group as defined herein shall be interpreted as a group that will release or donate electrons more than hydrogen would if it occupied the same position in the molecule. See, J. Marsh, Advance Organic Chemistry, 2nd Ed., McGraw Hill, Inc. (1977). These types of groups are well known in the art. Examples of electron donating groups include alkyl, lower alkoxy, aralkoxy; and the like. These electron donating groups, e.g., alkoxy, may be present on the aryl moiety of the following groups: DMB, TMB, Mtr, Pmc, Bz, Trt, CBZ and the like.

The protecting groups described hereinabove are well known to one skilled in the art. They can be removed under very mild acidic or basic conditions. The preferred protecting groups are those which can be cleaved by acid or base under conditions which are milder than those used to cleave the benzyl group. These include groups which can be cleaved by trifluoroacetic acid at room temperature within one to four hours. The especially preferred protecting groups are groups which can be cleaved by trifluoroacetic acid at room temperature within 1-2 hours.

These protecting groups include such groups as tetrahydropyranyl, β-trimethylsilylethyl, 1-ethoxyethyl, t-butyl, p-methoxybenzyl, 1-adamantyl, diphenylmethyl, triphenylmethyl, trialkylsilyl, (e.g. trimethylsilyl, triethylsilyl, and the like) trialkylstannyl, (e.g. trimethylstannyl, triethylstannyl, and the like), bis (4-methoxyphenyl)methyl, 2-furanylmethyloxycarbonyl, t-amyloxycarbonyl, 1-methylcyclohexyloxycarbonyl, isobornyloxy carbonyl, methoxytrimethylbenzenesulfonyl, pentamethylchromanesulfonyl, 2,4,6-trimethoxybenzyl, 9-xantheneyl and the like.

However, not all of the amino acids have side-chain functional groups. For example, many amino acids have hydrogen, alkyl or aralkyl side chains. These include glycine, alanine, valine leucine, norleucine, phenylalanine, isoleucine and the like. Therefore, these amino acids do not require protecting groups thereon.

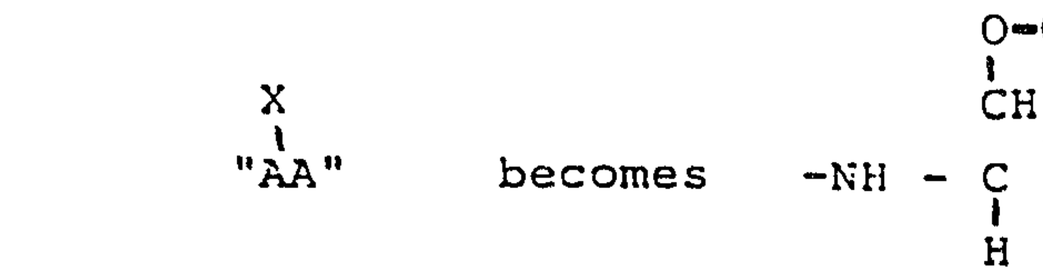
To differentiate between those amino acids having protecting groups and those not having protecting groups thereon,

$$\overset{X}{\overset{|}{AA}}$$

the term AA is used. As used herein, if no protecting group is present on the amino acid side chain, such as, e.g., in alanine, (which doesn't have a functional group and therefore no blocking group is required) "X" is -. Moreover, if the amino acid side chain has a functional group, such as in tyrosine, but is unprotected, then this also is indicated by X being defined as -. In other words, in both instances, when X is -, the side group is unprotected.

Although the term functional group is understood by one skilled in the art, it is defined as a group which could react with the reactants used or products formed under peptide forming condition if not protected by a blocking group. These functional groups include amino, carboxy, hydroxy, guanidino, imidazole, amino and the like.

On the other hand, if X is a blocking group, then this signifies that the functional group on the side chain is protected. For example, if AA is serine and X is t-butyl, then the residue



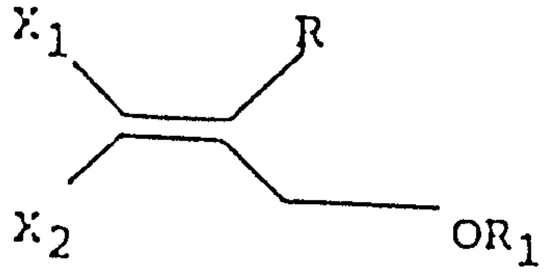
These protecting groups include the protecting groups described hereinabove.

Abbreviations have been used in the specification and claims with respect to these blocking groups and are listed hereinbelow:

Protecting group	Abbreviation
dimethoxybenzhydryl	DMB
2,4,6-trimethoxybenzyl	TMB
2,3,6-trimethyl-4 methoxybenzenesulfonyl	Mtr
9-fluorenylmethyloxycarbonyl	FMOC
t-butoxycarbonyl	BOC
t-butoxymethyl	Bom
pentamethylchromanesulfonyl	Pmc
adamantyl	ada
β-trimethylethyl	TMSE
β-trimethylethyloxycarbonyl	TEOC
t-butyl	t-bu
benzyl	Bz
cyclopentyl	Cp
cyclohexyl	Ch
triphenylmethyl	Trt
benzyloxycarbonyl	Cbz
adamantyloxycarbonyl	Adoc
formyl	CHO
trifluoroacetyl	TFA

The term amino acid protecting group, as used herein, refers to blocking groups which are known in the art and which have been utilized to block the amino (NH_2) group of the amino acid. Blocking groups such as 9-lower alkyl-9-fluorenyloxycarbonyl, 2-chloro-1-indanylmethoxy-carbonyl (CLIMOC) and benz [f] indene-3- methyloxycarbonyl (BIMOC) and dbd-TMOC are discussed in U. S. Patent Nos. 3,835,175, 4,508,657, 3,839,396, 4,581,167, 4,394,519, 4,460,501 and 4,108,846.

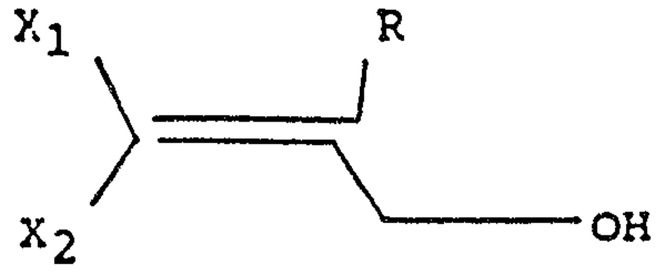
Moreover, other amino protecting groups can be used. These protecting groups are derived from 2-propenyloxy-carbonyl compounds having the general formula I :



I

wherein R is an electron withdrawing group, R_1 is COZ, X_1 and X_2 are independently H, lower alkyl, aryl, aryl lower alkyl or a solid support or R and X_1 taken together with the carbon atoms to which they are attached form a ring containing from 4 to 15 ring carbon atoms and may contain up to 2 heteroatoms, wherein the heteroatoms are O, S or N; and Z is a leaving group.

Compounds or Formula I can be derived from the corresponding alcohols of the formula II:



II

wherein R, X_1, and X_2 are as hereinbefore defined.

These 2-propenyloxycarbonyl compounds of formula I protect the primary or secondary amino group during chemical syntheses thereby protecting said amine from further reaction. Thus, a portion of the organic molecule other than the protected amine is modified by chemical reaction; and, (c) subsequently are easily cleaved from the amino group.

In a preferred embodiment, the protecting group can be rapidly and cleanly deblocked via treatment with a nucleophile. The nucleophile can also act as a scavenger for the protecting group. The preferred nucleophile is a primary or secondary amine.

An electron withdrawing group as defined herein shall be interpreted as a group that will draw electrons to itself more than a hydrogen atom would if it occupied the same position in the molecule. See, J. March, Advanced Organic Chemistry, 3rd Ed., John Wiley & Sons, P.17 (1985). These types of groups are well-known to one skilled in the art.

Examples of electron withdrawing groups include SO_2R_2, SOR_2, $COOR_2$, COR_2, CHO, $CONR_2R_3$, CN, CF_3, NO_2, aryl, pyridyl and the like, wherein R_2 and R_3 are independently lower-alkyl having from 1 to 6 carbon atoms, aryl, aryl lower-alkyl, hetero-aryl or a solid support and the alkyl, aryl or hetero-aryl groups are unsubstituted, or mono- or di-substituted with halides, SO_2R_2, SOR_2, $COOR_2$, COR_2, CHO, CN, CF_3 or NO_2.

As used herein, the terms lower alkyl, when used singly or in combination, refer to alkyl groups containing one to six carbon atoms. They may be straight chain or branched and include such groups as methyl, ethyl, propyl, isopropyl, butyl, sec-butyl, isobutyl, t-butyl, pentyl, isopentyl, neopentyl, hexyl and the like. The preferred alkyl groups contain one to four carbon atoms.

The term aryl, when used alone or in combination, refer to an aromatic ring containing six to ten ring carbon atoms. The aryl group includes phenyl, and 1 or 2-naphthyl. The preferred aryl group is phenyl.

The most preferred aralkyl group is benzyl.

The term hetero-aryl when used alone or in combination refers to an hetero-aromatic ring having from four to nine carbon ring atoms and one or two hetero ring atoms. The heterocyclic rings may contain sulfur, nitrogen or oxygen. The hetero-aryl groups include quinolyl, isoquinolyl, furyl, thienyl, or pyridyl.

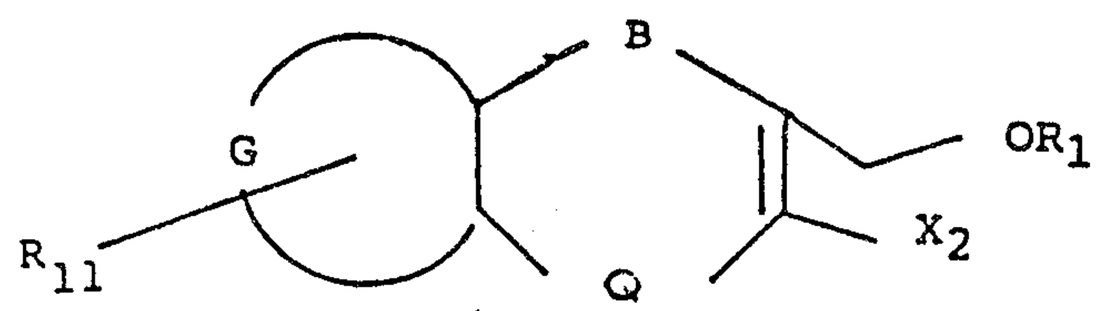
The preferred heteroaryl groups are 2- or 4-pyridyl.

The preferred R groups are SO_2R_2, SOR_2, $COOR_2$, COR_2, $CONR_2R_3$, aryl, or pyridyl, wherein R_2 and R_3 are as defined hereinabove.

Especially preferred R groups are $SO_2C(CH_3)_3$, $SOC(CH_3)_3$, $SO_2C_6H_5$, SOC_6H_5, 2-pyridyl, or 4-pyridyl or COOEt.

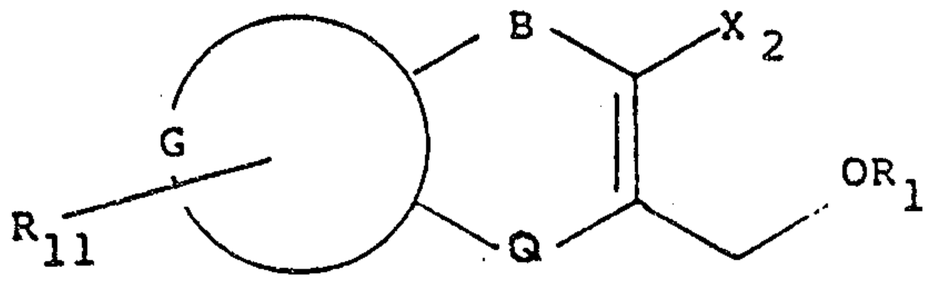
As used herein the term solid support when used singly or in combination refers to polymer resins typically used in peptide synthesis. These polymers are generally employed in the form of beads. The polymer resins preferred for peptide synthesis are polystyrenes, polyacrylamides and the like. The preferred polystyrene resin is a copolymer of styrene-divinylbenzene.

As noted hereinabove, R and X_1 taken together with the carbons to which they are attached may be cyclic containing one, two, or three rings. Said cyclic structure may contain from four to fifteen ring carbon atoms and up to two heteroatoms. These heterocyclic ring atoms may be sulfur, oxygen or nitrogen. For example, when R and X_1 taken together form a ring, the compounds of Formula I may have the formula III or its isomer IIIa



III

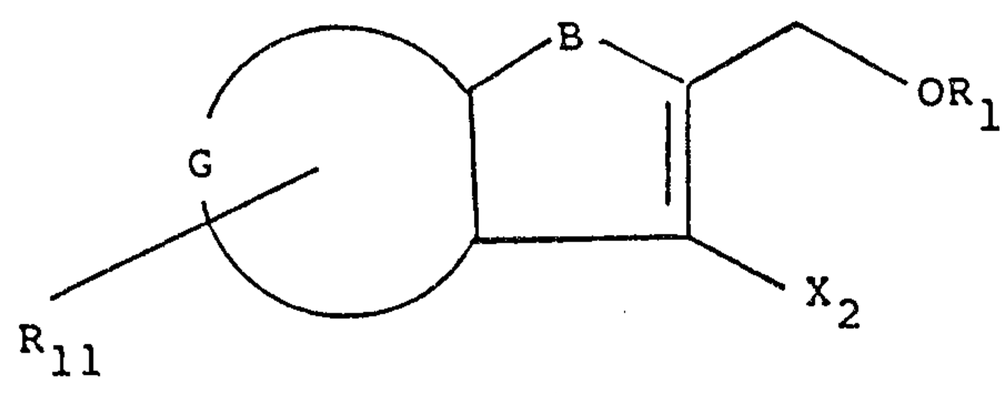
or



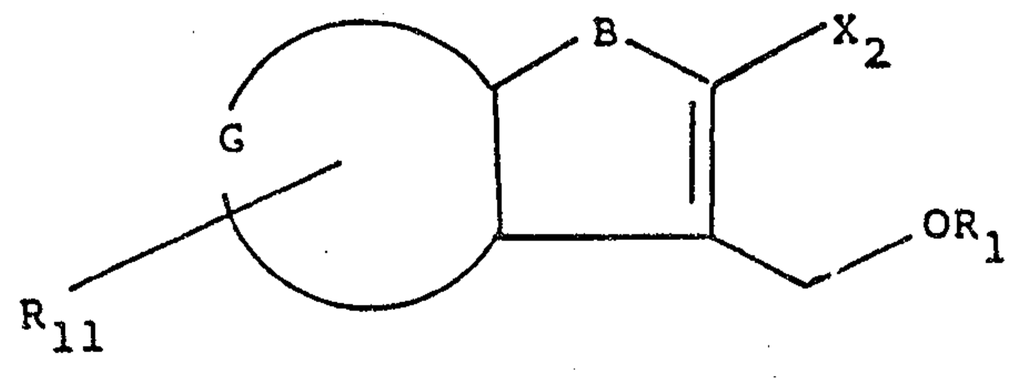
IIIa

wherein B is a chemical bond, CR_8R_9, $\overset{O}{\overset{\|}{C}}$, SO_2, SO, RP(O), or S, R is lower alkyl or OR_{10}, R_{10} is lower alkyl, R_8, R_9, R_{12} and R_{13} are independently hydrogen or lower alkyl, G is a mono or bicyclic fused ring system containing 5 to 10 carbon atoms and may contain up to 2 heteroatoms, G is a chemical bond, $CR_{12}R_{13}$, SO_2, SO, PP(O) or S, R_{11} is hydrogen or lower alkyl and X_2 and R_1 are as defined hereinabove. G may be completely saturated, partially unsaturated or fully aromatic (e.g. a benzo system). Examples of G include cyclopentyl, cyclohexyl, cyclopentenyl, cyclohexenyl, decalinyl, phenyl, naphthyl, pyridyl azaindenyl and the like.

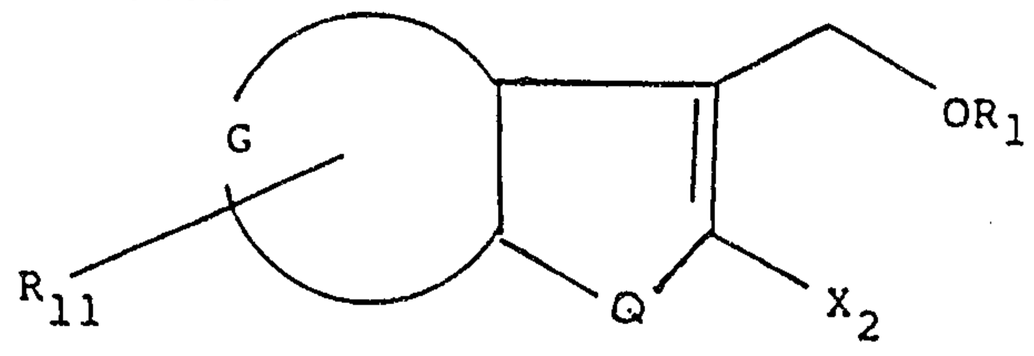
As defined hereinabove, when G is a chemical bond, then III or IIIa has the structure



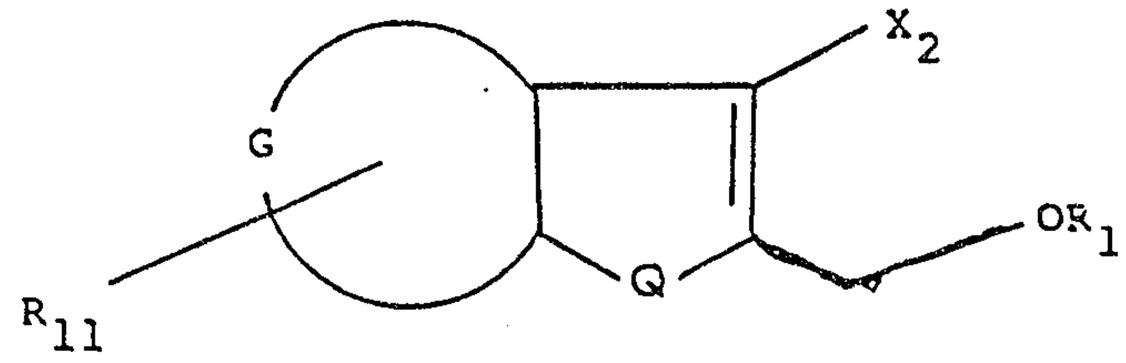
or



Similarly, when B is a chemical bond, then 3 or 3a has the structure

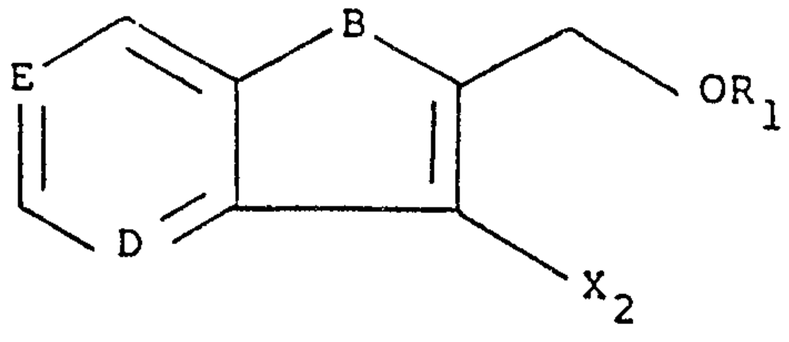


or



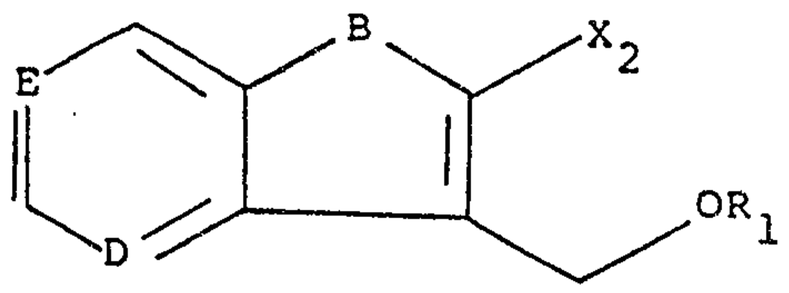
However, in order to be effective, either B or Q is an electron withdrawing group or ring G is itself electron withdrawing. In other words, if ring G is heterocyclic, i.e., contains ring heteroatoms, then B and Q are independently any of the values described hereinabove. However, if ring G is not heterocyclic (e.g., is aromatic or cycloalkyl), then at least one of B or Q is $\overset{O}{\overset{\|}{C}}$, SO_2, SO, RP(O) or S wherein R is as defined hereinabove.

A preferred class of III or IIIa has the formula



IIIb

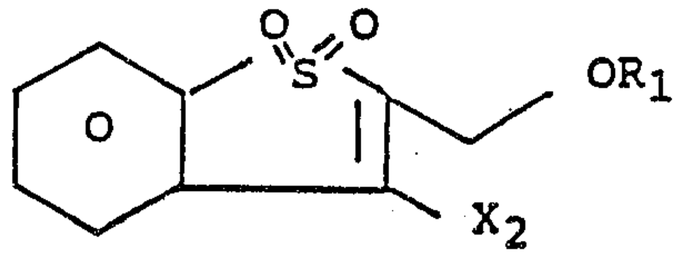
or



IIIc

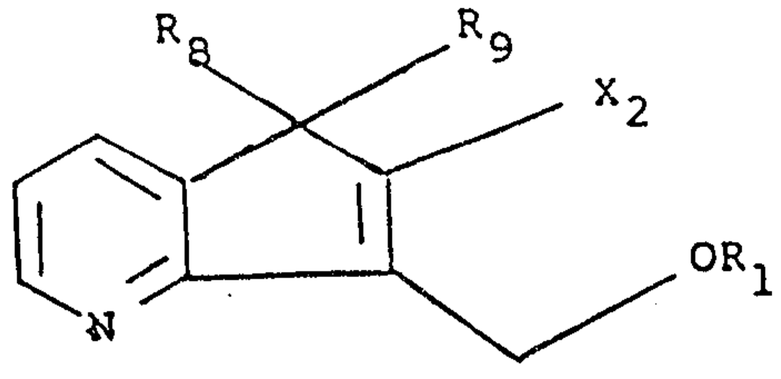
wherein B is CR_8R_9 or SO_2 and E and D are independently CH or N, provided that when B is CR_8R_9, then E or D is N.

A preferred embodiment has the formula IV:



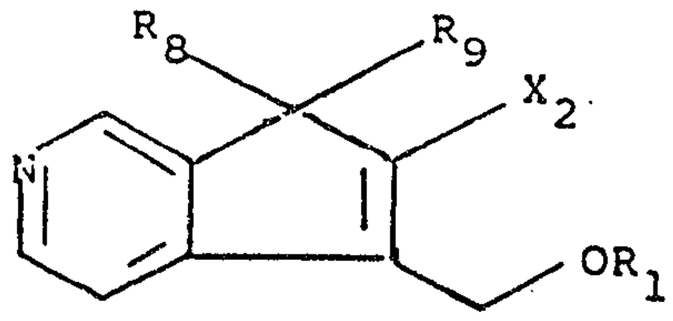
IV

or the formula IVa or IVb:



IVa

or



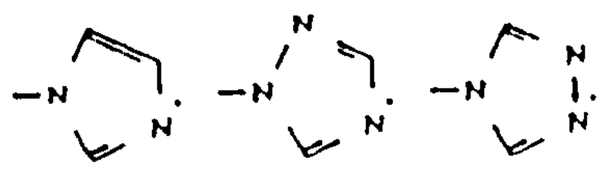
IVb

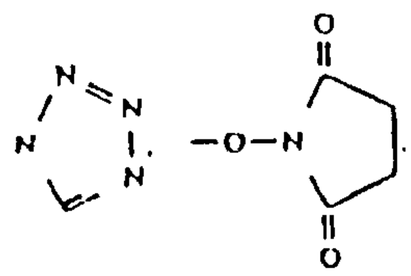
wherein X_2 and R_1 are as defined hereinabove and R_8 and R_9 are independent.

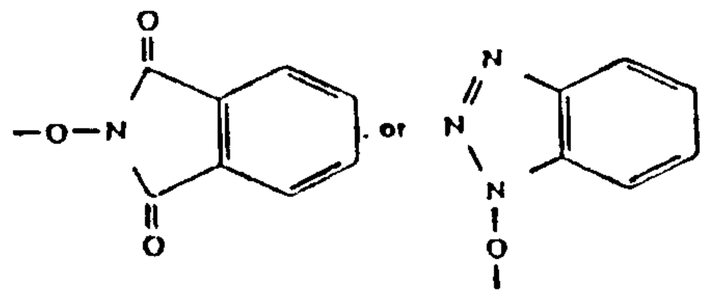
When compounds of Formula I are used to protect a primary or secondary amino group and introduce the 2-propenyloxy carbamate compound of Formula I, Z is a leaving group. As is generally known in the art and for the purposes of the present invention "a leaving group" is defined as a group which is readily broken away from its union with the carbon atoms. It is one which readily joins, for example, with an active hydrogen atom to split out a compound containing the hydrogen atom and the leaving group. Leaving groups are generally electron attracting groups either

because of their electronegativity or because they have an inductive effect. Leaving groups are defined in U.S. Patent No. 4,394,519 to Carpino, et al.

The preferred leaving groups Z are halo, OR_4, SR_4, SAr, N_3, OAr,





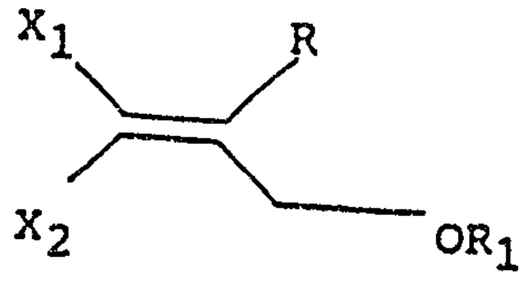


wherein R_4 is lower-alkyl, aryl or aryl lower-alkyl, wherein the alkyl or aryl groups are unsubstituted, or mono- or di-substituted with halides, SO_2R_4, SOR_4, $COOR_4$, COR_4, CHO, CN, CF_3 or NO_2.

The most preferred leaving groups are halo, especially Cl and Br.

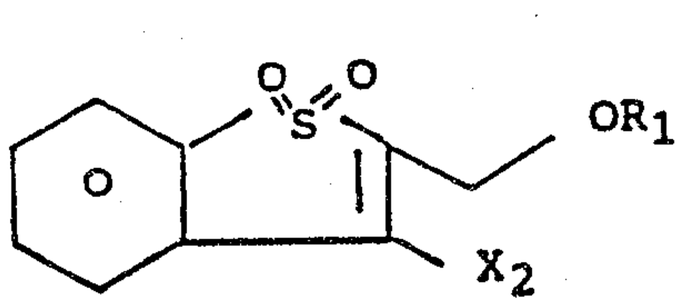
The preferred groups in the vinyl position, X_1 and X_2, are independently H, phenyl, lower alkyl having from 1 to 4 carbon atoms or a solid support. Especially preferred groups for X_1 and X_2 are independently H or phenyl. The X_1, and X_2 positions may also be used as the site of attachment to a polymer resin, on Z, when, Z is an amino acid or peptide residue.

The preferred compounds of Formula I have the formula :

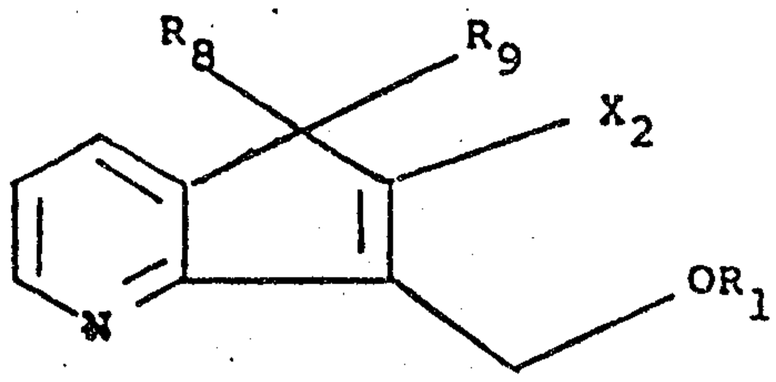


wherein R is SO_2R_2, SOR_2, $COOR_2$, $CONR_2R_3$, aryl or 2- or 4-pyridyl, wherein R_2 and R_3 are as defined hereinabove or R and X_1 taken together with the carbon atoms to which they are attached form a ring, the ring having from 4 to 15 ring carbon atoms and up to a total of 3 rings, often forming a fused benzo-system; R_1 is H or COZ and Z is halo; and X_1 and X_2 are independently H, phenyl or lower alkyl.

The preferred groups in compounds wherein R and X_1 are taken together with the carbon atoms to which they are attached are $SO_2C_6H_4$ or $C_5H_3NCH_2$ thereby describing compounds of the formula IV or IVa or IVb described hereinabove:

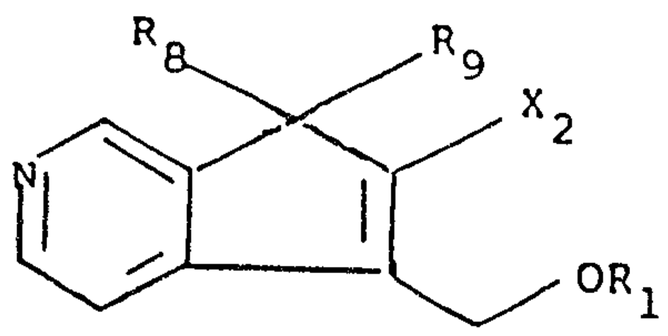


IV



IVa

or



IVb

wherein R_1 and X_2, R_8 and R_9 are as defined hereinabove, and R_8 and R_9 are independently hydrogen or lower alkyl.

The most preferred compounds of Formula I are the 2-propenyloxy chloroformates wherein R is $SO_2C(CH_3)_3$, $SOC(CH_3)_3$, $SO_2C_6H_6$, SOC_6H_6, COOEt, 2-pyridyl or 4- pyridyl; Z is Cl or Br; and X_1 and X_2 are H or phenyl.

The preferred alcohols of Formula II are:

2-(t-butylsulfonyl)-2-propenyl alcohol,
2-carboethoxy-2-propenyl alcohol,
2-(phenylsulfonyl)-2-propenyl alcohol,
(E)-3-phenyl-2-(phenylsulfonyl)-2-propenyl alcohol,
(Z)-3-phenyl-2-(phenylsulfonyl)-2-propenyl alcohol,
3,3-diphenyl-2-(phenylsulfonyl)-2-propenyl alcohol,
Benzothiophenesulfone-2-methanol,
3,3-dimethyl-2-(phenylsulfonyl)-2-propenyl alcohol.
(Z)-3-phenyl-2-(methylsulfonyl)-2-propenyl alcohol

The preferred chloroformates of Formula I are:

2-(t-butylsulfonyl)-2-propenyl chloroformate,
2-carboethoxy-2-propenyl chloroformate,
2-(phenylsulfonyl)-2-propenyl chloroformate,
(E)-3-phenyl-2-(phenylsulfonyl)-2-propenyl chloroformate,
(Z)-3-phenyl-2-(phenylsulfonyl)-2-propenyl chloroformate, or

3,3-diphenyl-2-(phenylsulfonyl)-2-propenyl chloroformate,

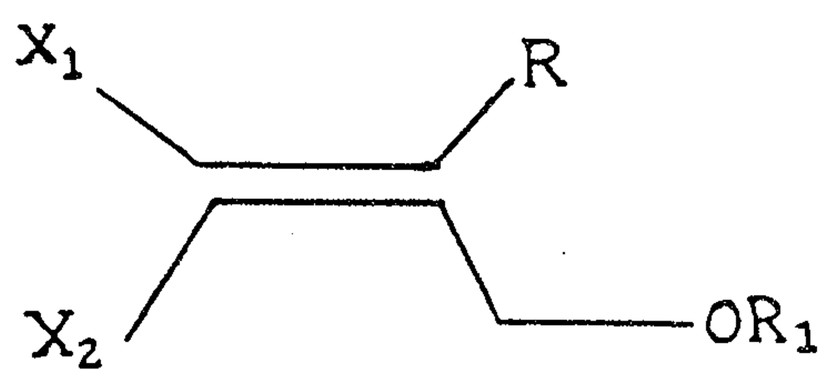
Benzothiophenesulfone-2-methyl chloroformate,

3,3-dimethyl-2-(phenylsulfonyl)-2-propenyl-chloroformate,

(Z)-(3-phenyl)-2-methylsulfonyl-2-propenyl-chloroformate.

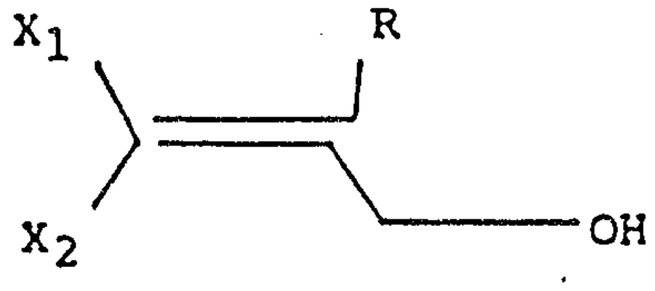
The 2-propenyloxy carbonyl compounds outlined hereinabove can be formed by art-recognized techniques known to one skilled in the art. Exemplary procedures are outlined hereinbelow.

The compounds having the formula Ia:



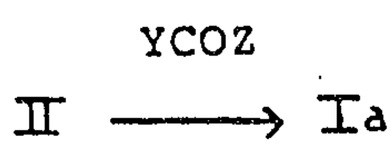
Ia

can be prepared from the corresponding alcohol II:



II

as illustrated in the following equations:



(1)

wherein Z=Y=Cl, Br, Fl, CN;

Z=Cl, Y=SR_4, SAr, OAr, Fl;

(2)

wherein Z=Cl, Br; Y=Fl, N_3, CN.

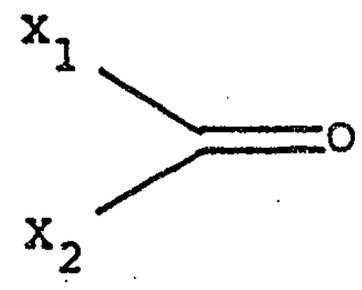
In the above equations, X_1, X_2, R, and R_4 are as defined herein.

Typically, reactions such as indicated by Equation (1) are carried out in an inert organic solvent. As defined herein an inert solvent is a reaction inert solvent, i.e., one which will not react with the reagents or reactants under the given reaction conditions. Suitable solvents are halogenated or non-halogenated hydrocarbons containing up to about eight carbon atoms, e.g., methylene chloride, ethylene dichloride, benzene, isooctane and the like. Reactions are

conducted at temperatures of from about 0°C to about 25°C during a reaction period of from about 1 to about 6 hours. Suitable yields are obtained with equimolar quantities of reactants, although the yield may often be appreciably increased by utilizing an excess of either one of them, for example, up to about a 20% molar excess. Generally speaking, the halogen substituted compounds are prepared under less rigorous reaction conditions than are required for the preparation of those compounds wherein the substituent is of higher molecular weight. The presence of a weak base, may increase the rate of reaction.

Reactions of Equation (2) in which the substituent placed on the carbonyl carbon atom is initially present in an ionic form are carried out in inert polar organic solvents which will enhance ionization, including, for example, acetonitrile, dimethylformamide, dimethylsulfoxide, tetrahydrofuran, dioxane and the like. The reaction is normally carried out at a temperature of from about 0°C to about 25°C during a period of from about 1 to 5 hours. Preferably equimolar quantities of reactants are employed to minimize side reactions but a moderate excess of either reactant would not introduce appreciable difficulties.

Compounds of Formula II can be prepared by reacting an aldehyde or ketone of Formula V:



V

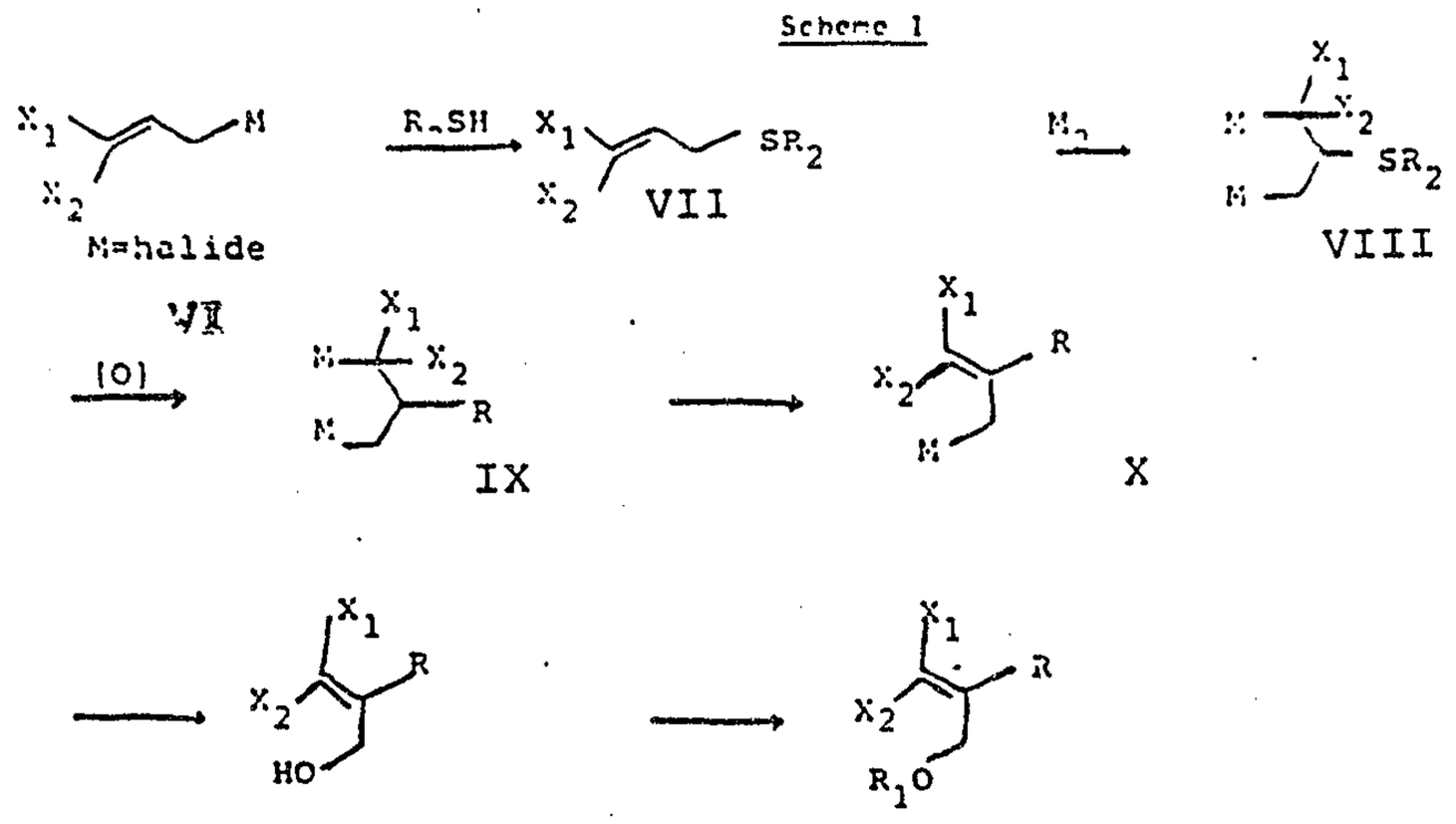
with a Wittig reagent, $\emptyset_3\overset{+}{P}\overset{-}{C}HR$, $(OR_{11})_2\overset{O}{\overset{\|}{P}}CH_2\text{-}R$ or $(R_{11})_3S_iCH_2\text{-}R$ such as,

$(EtO)_2\overset{O}{\overset{\|}{P}}-CH_2-R$ or Me_3Si-CH_2-R

where R is defined hereinabove, and R_{11} is lower alkyl under Wittig reaction conditions; followed by the addition of formaldehyde under Prins reaction conditions, wherein X_1, X_2 and R are as hereinbefore described and R_{11} is lower alkyl.

Compounds of Formula I wherein R contains a sulfur atom can be prepared by an alternative route. The following procedures as depicted in Scheme I is illustrative for the preparation of these compounds:

Scheme I



An allylic halide such as allyl bromide VI , is treated with a thiol, R_2SH, such as t-butylthiol in strong base to form the corresponding thioether VII. Halogenation of the thioether forms the dihalo thioether VIII which in turn is oxidized with an oxidizing agent to form the sulfonyl or sulfinyl compound IX. Various oxidizing agents can be used to effect said reaction, such as MCPBA. Compound IX is then

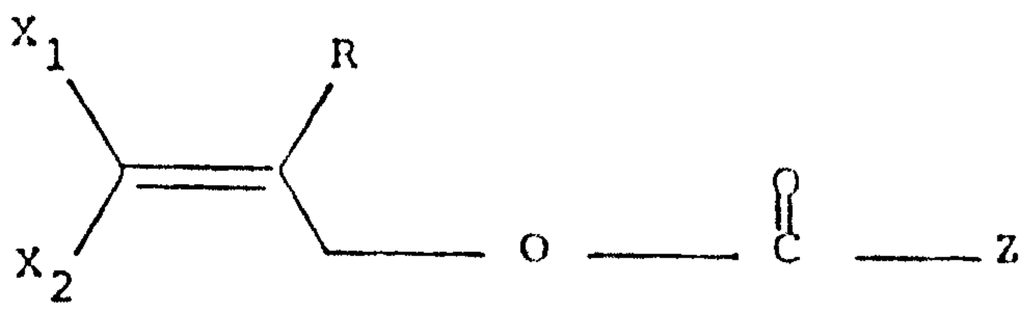
reacted with a strong base, such as lutidine, to form the corresponding unsaturated compound X. Substitution of the halide in compound X with hydroxide forms the compound of Formula II. The compound of Formula I can be readily prepared from II according to the procedure described hereinabove.

Typically, the reactions for synthesis of the compounds described in Scheme 1 hereinabove are carried out in an inert organic solvent. Suitable solvents include alcohols, such as methanol, ethanol, isopropanol, t-butanol and the like, ethers such as diethyl ether, 1,4-dioxane, tetrahydrofuran (THF) and the like, hydrocarbons, such as benzene, hexane, cyclohexane, toluene, *Skelly solvents and the like, and halogenated hydrocarbons such as $CHCl_3$, CCl_4, CH_2Cl_2 and the like.

Temperatures for these reactions range from about -78°C to the reflux temperature of the solvent being employed. Unless indicated to the contrary in the discussion of the various reaction schemes described hereinabve and hereinafter, the preferred temperatures are from about 0°C to about 100°C.

The compounds of Formula I described herein wherein Z is a leaving group protect the amino group in an amino acid during peptide synthesis. Thus, a method for the preparation of a peptide comprises:

a) reacting an amino acid having a free amino group with a compound of the formula:



*Trade-mark

wherein

Z is a leaving group;

X_1 and X_2 are independently H, lower alkyl, aryl, aryl lower-alkyl or a solid support;

R is an electron withdrawing group; or

R and X_1 with the carbon atoms to which they are attached form a ring containing from 4 to 15 ring carbon atoms;

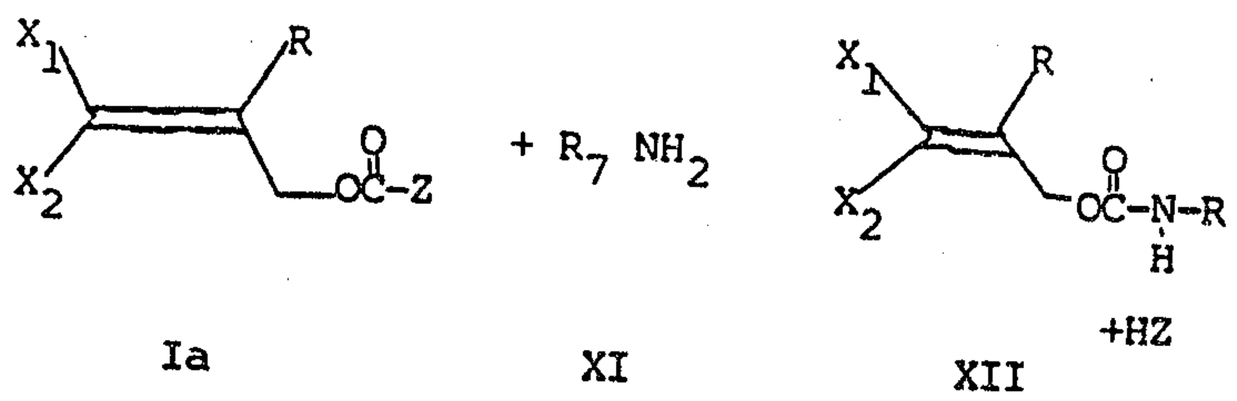
b) reacting the product of (a) with an amino acid-fluoride or peptide having a free amino group; and

c) removing the protecting group.

Thus, the present invention can be used as blocking groups for amino acids during peptide synthesis. The preferred amino acids are alpha-amino acids.

More specifically, the compounds of Formula Ia, wherein Z is a leaving group, can react with a carboxyprotected amino acid as indicated hereinbelow in Scheme II:

Scheme II



Ia XI XII

In the above scheme, X_1, X_2, R, Z are as defined above, and R_7NH_2 (XI) is an alpha-amino acid fluoride. The alpha-amino acids are the alpha-amino acids described herein above and include phenylalanine, glycine, valine, and the like. Often the amino acids are protected by a carboxy protecting group known in the art.

A variety of carboxy protecting groups known in the art may be employed. Examples of many of these possible groups may be found in "Protective Groups in Organic Synthesis" by T.W. Green, John Wiley and Sons, 1981. The preferred carboxy protecting group is the t-butyl ester.

In the above scheme, a compound of Formula Ia is reacted with a carboxy protected amino acid to form XII which is then hydrolyzed in acid to form a compound of Formula I wherein Z is an amino acid adduct. The most preferred protecting group are Bspoc; i.e., 2-(t-butylsulfonyl)-2-propenyloxycarbonyl and Bsmoc, i.e, Benzothiophenesulfone-2-methyloxycarbonyl, and 2-methyl-sulfonyl-3-phenyl-2-propenyloxycarbonyl (Mspoc).

If the leaving group is a halogen, especially chlorine, the reaction may be effected in an inert, polar organic solvent such as dioxane, tetrahydrofuran, dimethylformamide, pyridine or other solvent containing, for example, up to eight carbon atoms. The reaction is run under alkaline conditions, typically dilute aqueous alkali metal base such as sodium or potassium hydroxide or carbonate, at low temperatures, for example, from about 0°C to 25°C during a period of from about 2 to 3 hours. Usually the protected amino acid or peptide will precipitate upon acidification of the mixture, and may be purified by any appropriate method such as recrystallization. Excess blocking reagent may be employed, even up to 0.5 molar excess, but equimolar quantities of reactants generally give better results.

The protected amines can also be prepared by reaction of the 2-propenyl alcohols with isocyanates. This reaction will form a 2-propenyl carbamate directly without requiring the conversion of the alcohols to chloroformate.

Preferred protecting groups of Formula I are 2-t-butylsulfonyl-2-propenyloxycarbonyl (Bspoc), benzothiophene sulfone-2-methyloxycarbonyl (Bsmoc) and 2-methylfulfonyl-3-phenyl-2-propenyloxycarbonyl (Mspoc).

Other amino protecting groups are described in an article entitled "Solid Phase Peptide Synthesis" by G. Barany and R.B. Merrifield in Peptides, Vol. 2, edited by E. Gross and J. Meienhoffer, Academic Presser, New York, New York, pp. 100-118 (1980). These N-amino-protecting groups include such groups as the FMOC, Bspoc, Bsmoc, t-butyloxy-carbonyl (BOC), t-amyloxycarbonyl (Aoc), β-trimethyl-ethyl-oxycarbonyl (TEOC), adamantyloxycarbonyl (Adoc), 1-methyl-cyclobutyloxycarbonyl (Mcb), 2-(p-biphenylyl)propyl-2-oxy-carbonyl (Bpoc), 2-(p-phenylazophenyl)propyl-2-oxycarbonyl (Azoc), 2,2-dimethyl-3,5-dimethyloxybenzyloxycarbonyl (Ddz), 2-phenylpropyl-2-oxycarbonyl (Poc), benzyloxycarbonyl (Cbz), p-toluenesulfonyl aminocarbonyl (Tac), o-nitrophenylsulfenyl (Nps), dithiasuccinoyl (Dts), phthaloyl, piperidinooxy-carbonyl, formyl, trifluoroacetyl and the like.

These protecting groups can be placed into four categories:

1) a base labile N α-amino acid protecting group such as FMOC, and the like.

2) protecting groups removed by acid, such as Boc, TEOC, Aoc, Adoc, Mcb, Bpoc, Azoc, Ddz, Poc, Cbz, 2-furanmethyloxycarbonyl (Foc), p-methoxybenzyloxycarbonyl (Moz), Nps, and the like.

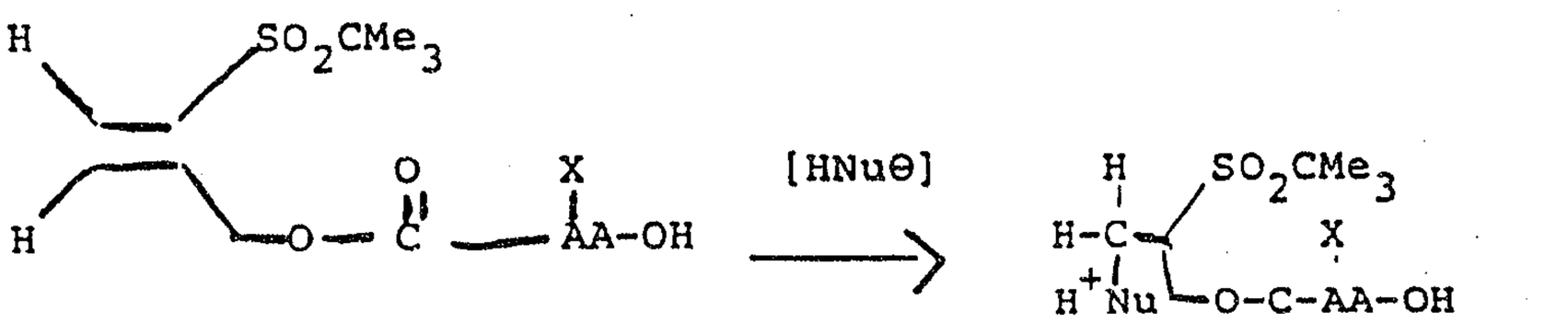
3) protecting groups removed by hydrogenation such as Dts, Cbz.

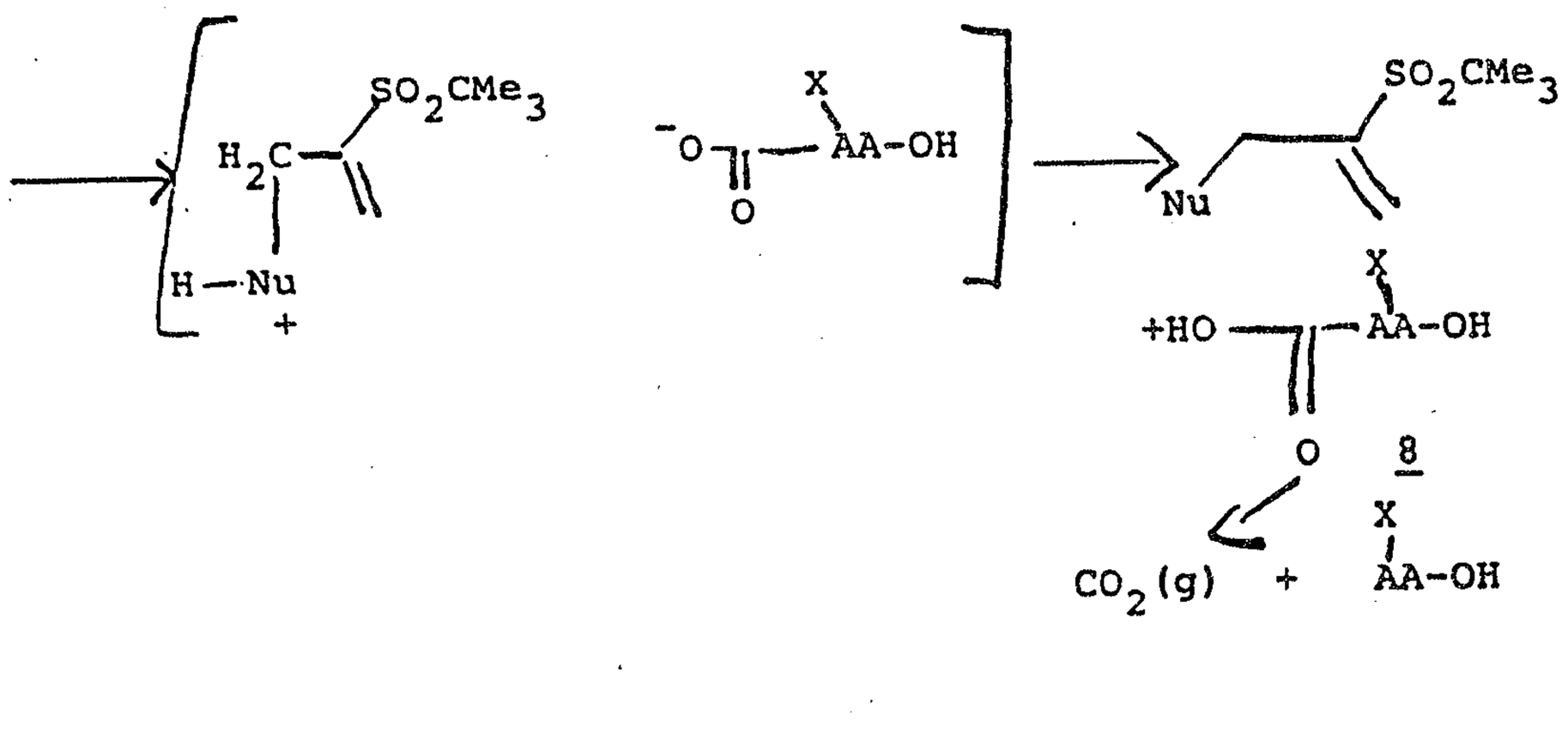
4) protecting groups removed by nucleophiles, such as Bspoc, Bsmoc and Nps and the like.

5) protecting groups derived from carboxylic acids, such as formyl, acetyl, trifluoroacetyl and the like, which are removed by acid, base or nucleophiles.

As defined herein, a nucleophile is an electron-rich atom, i.e., an atom which can donate an electron pair, which tends to attack a carbon nucleus but does not act as a Bronsted Lowry base. For example, a nucleophile, as defined herein, includes those molecules which are used for nucleophilic addition across a double bond and behaves in a manner similar to that described in the schemes herein below.

The general mechanism for cleavage of Bspoc and Bsmoc groups are similar in that the nucleophile is believed to react through a Michael-type addition across a double bond. Although the following schemes are shown for Bspoc, it is also illustrative of Bsmoc:





The nucleophile is believed to attack at the terminal carbon atoms of the propenyl group (Michael acceptor) forming a zwitterion which can eliminate the OCOAA(X)OH anion and H^+ to form an alkene-amine and the amide (8) after protonation. Loss of CO_2 will furnish the free amino acid.

The nucleophiles which will function in concert with this invention must have an active hydrogen atom, i.e., a hydrogen atom attached to the nucleophilic atom.

It is preferred that the nucleophile is a simple amine. It is especially preferred that the simple amine is a primary or secondary amine of the formula HNR_5R_6 wherein R_5 and R_6 are independently hydrogen, lower alkyl or substituted lower alkyl, the lower alkyl being substituted with OH, CH_3, or CH_2CH_3 or R_5 and R_6 taken together form a mono or bicyclic ring containing from 4 to 10 ring carbon atoms and 1 or 2 heteroatoms selected from the group consisting of nitrogen, sulfur or oxygen.

Typical examples of useful amines include ethanolamine, morpholine, piperidine, diethylamine, 2,6-dimethylpiperidine, piperazine, diethylamine, ethylamine and the like.

An organo mercaptan can also be used as a nucleophile, e.g., alkyl mercaptans, cycloalkyl mercaptans, aryl mercaptan or aralkyl mercaptans. The most preferred mercaptan is benzyl mercaptan.

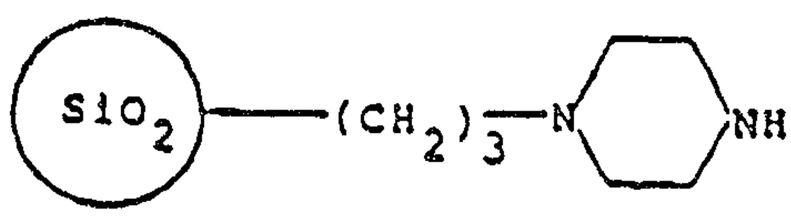
The nucleophile can be added as a free compound or as an insoluble reagent attached to a solid support i.e., polystyrene or silica dioxide. These are represented by the formula:

p-[alk]-NuH

wherein p is an organic polymer as defined hereinabove or a silica gel polymer; alk is a chemical bond, alkyl or aroyl chain having from about one to about ten

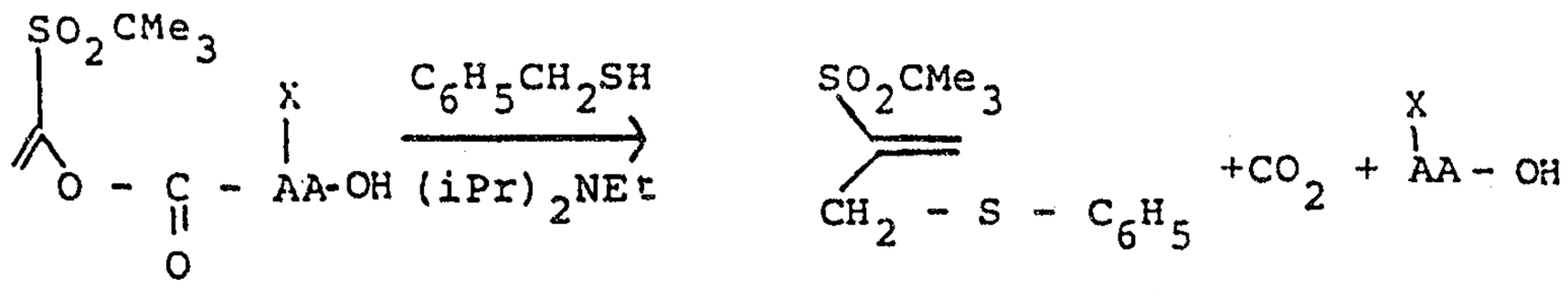
carbon atoms and Nu-H is a nucleophile as defined hereinabove.

A preferred insoluble reagent is the silica based piperazine reagent 9:



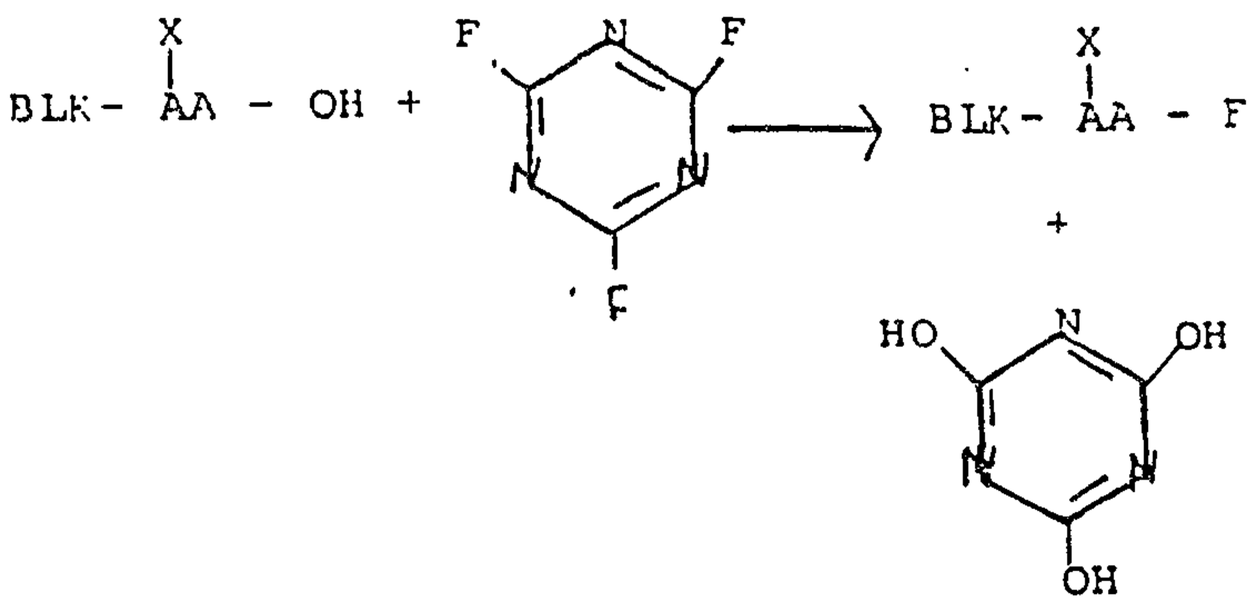
9

Another useful nucleophile is benzylmercaptan as shown in the following scheme.



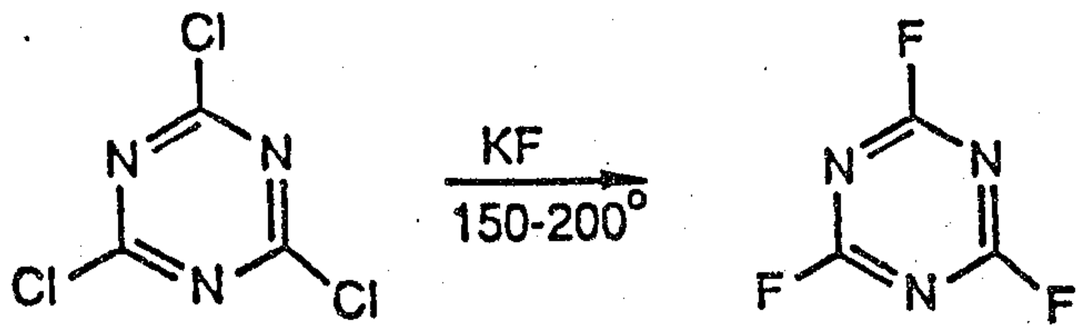
In this scheme the thio-group reacts in a Michael fashion to remove the Bspoc protecting group.

The amino acid fluorides of the present invention can be prepared by art recognized techniques. More specifically, it can be prepared by reacting an N-protected amino acid with the reagent cyannuric fluoride according to the following equation:



wherein BLK is an amino protecting group as defined herein and X is defined herein. It is preferred that BLK is the FMOC CLIMOC, BIMOC, DBD-TMOC, Bspoc, Bsmoc, Mspoc, or related base sensitive group. This reaction can be run at temperatures as low as 0° and up to the refluxing temperature of the solvent, but it is preferred that reaction is run at room temperature. It also can be run in an inert solvent such as pyridine/CH_2Cl_2 and the like.

The cyanuric fluoride can be prepared from the corresponding chloride in the presence of potassium flouride at elevated temperatures ranging from 150° to 250°C, according to the following equation:



Other fluorinating agents well known in the art, such as thionyl fluoride, 2,4,6-trinitrofluorobenzene, N-methyl-2-fluoropyridinium salts, and the like may be used in place of KF to effect the formation of cyanuric fluoride.

The amino acid fluorides of the present invention are useful in peptide bond formation. The scope is quite broad, as the amino acid fluorides of the present invention can be coupled with an amino acid, a dipeptide, tripeptide, or higher peptide, having a free terminal amino group. As used herein, the term first amino acid is meant to include amino acids as well as dipeptides, and the higher peptides.

The synthesis of peptides according to the present invention requires the following steps:

1) protection of the carboxyl group on a first amino acid.

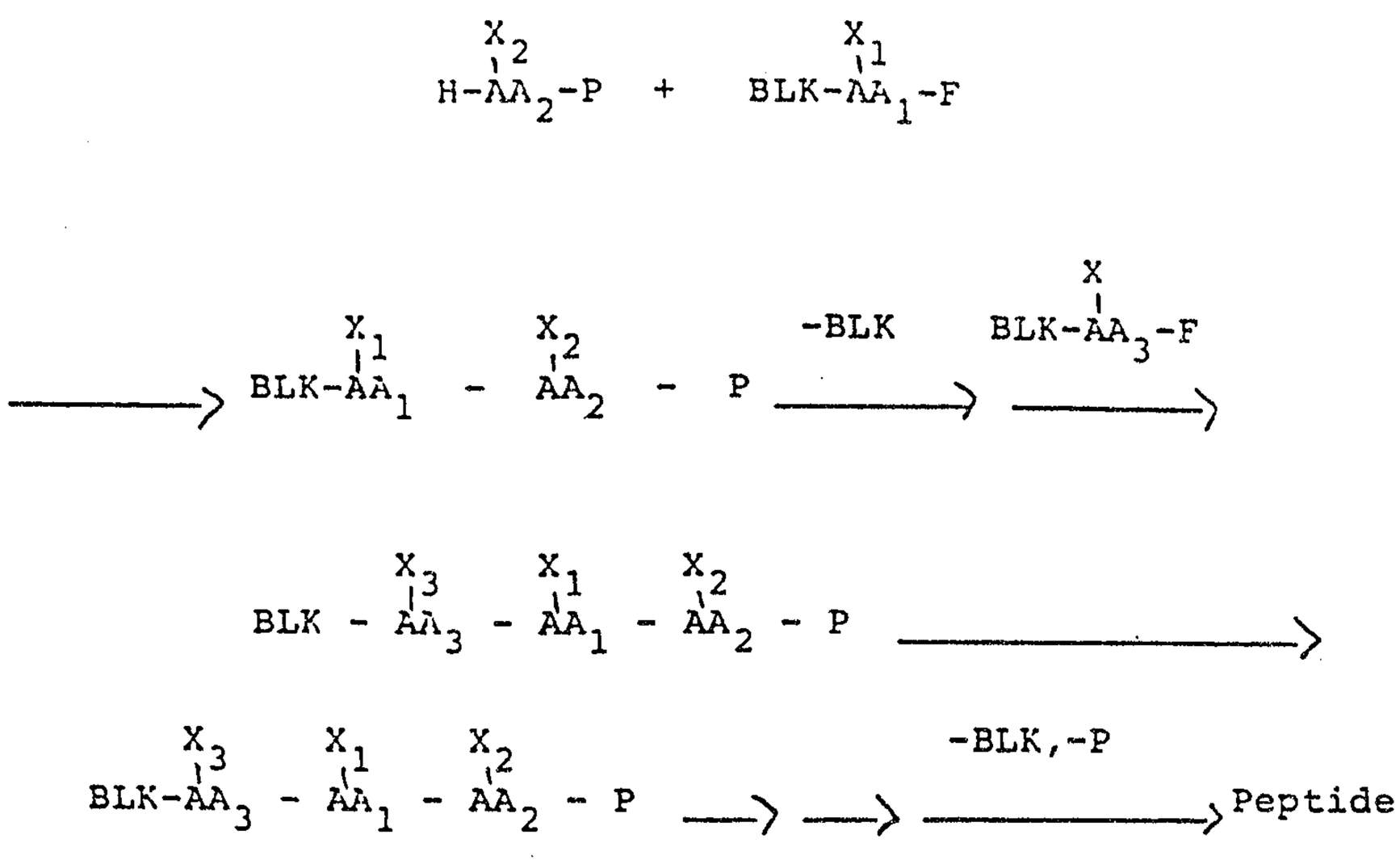
2) formation of the amino acid fluorides of the present invention in accordance with the procedure herein.

3) formation of the peptide bond by coupling the amino acid fluoride with the first amino acid.

4) removal of the protecting groups.

A variety of carboxy protecting groups known in the art may be employed. Examples of many of these possible groups may be found in "Protective Groups in Organic Synthesis", by T.W. Green, John Wiley & Sons, 1981.

The following sequence is illustrative of the coupling of an amino acid fluoride of the present invention with an amino acid having a free amino group:



In the above scheme, BLK is as defined hereinabove, X_1, X_2 and X_3 are independently defined as X hereinabove, and P is a carboxy protecting group, e.g., methyl ester, t-butylester, β-trimethylsilylethyl ester, benzyl ester and the like.

As shown by the above scheme, the Nα-amino protected amino acid fluoride is reacted with a second amino acid in which the carboxy group is protected. A peptide is formed between the first amino acid and the second amino acid. The peptide chain can be increased by removing the alpha amino protecting group by techniques known to one skilled in the art, and then reacting the corresponding di-peptide with another Nα-amino protected amino acid fluoride to form the corresponding tri-peptide. The Nα-amino protecting group of the tri-peptide is removed and the above cycle is repeated until the desired peptide has been obtained.

The coupling of the N-α protected amino acid fluoride with the carboxy protected amino acid by the normal two phase techniques takes place without racemization.

The present invention can readily be utilized in solid phase peptide synthesis. Solid phase peptide synthesis is based on the stepwise assembly of a peptide chain while it is attached at one end to a solid support or solid phase peptide resin. Two methods are generally well known in the art.

One, the Merrifield method, employs a solid support for attachment of the amino acid or peptide residues. This method employs N-protected amino acids as building blocks which are added to an amino acid or peptide residue attached to the solid support at the acyl (acid) end of the molecule. After the peptide bond has been formed, the protected group is removed and the cycle repeated. When a peptide having the desired sequence has been synthesized, it is then removed from the support.

The second method, the inverse Merrifield method, employs reagents attached to solid supports in a series of columns. The amino acid or peptide residue is passed through these columns in a series to form the desired amino acid sequence.

These methods are well known in the art as discussed in U.S. Patent Nos. 4,108,846, 3,839,396, 3,835,175, 4,508,657, 4,623,484, 4,575,541, 4,581,167, 4,394,519 as well as in Advances in Enzymology, 32, 221 (1969) and in PEPTIDES, VOL, 2, edited by Erhard Gross and Johannes Meienhoffer, Academic Press, New York, New York pp. 3-255 (1980).

The amino acid fluorides of the present invention can exist in various stereoisomeric forms, viz., the D or L stereoisomers. Moreover, the amino acid fluorides may be present in mixture of the D and L forms such as in racemic mixtures. All of these forms are comtemplated to be within the scope of the present invention. It is preferred that the stereoisomer of the amino acid fluorides of the present invention exist in the L form.

EXAMPLES

The invention will now be illustrated by examples. The examples are not intended to be limiting of the scope of the present invention. In conjunction with the general and detailed descriptions above, the examples provide further understanding of the present invention.

I PREPARATIVE EXAMPLES: PREPARATION OF CYANURIC FLUORIDE

A 250 mL three necked round bottomed flask equipped with a thermometer, stirring bar, distilling apparatus and powder dropping funnel was charged with oven-dried sodium fluoride (53 g, 1.25 mole) and tetramethylene sulfone (sulfolane, TMS) (145 g, 115 mL, 1.20 mole, d=1.26). To the suspension was added cyanuric chloride (62 g. 0.33 mole) in small portions by use of the powder dropping funnel over a 10-min period. The reaction mixture was gradually heated to 250°C by means of a heating mentle. The product distilled from the flask as formed and was collected in glass traps cooled in a Dry Ice-acetone bath. Collection of the distillate (29 mL) continued until distillation stopped (bp 72-78°C). Redistillation gave 24 mL (86.19 g, 38.4% d=1.6 g/mL) of the fluoride as a clear colorless liquid, bp 72-73°C, lit bp 74°C.

II GENERAL PROCEDURE FOR PREPARATION OF N-(9-FLUORENYL-METHOXYCARBONYL)AMINO ACID FLUORIDES:

A solution (or suspension) of FMOC-amino acid (1 mmole) in dry CH_2Cl_2 (5 mL) was kept under nitrogen and treated with cyanuric fluoride (1.08 g, 8 mmol, 700 uL, d=1.6) and pyridine (81 uL, 1 mmole). A clear solution was obtained which was refluxed (or stirred at room temperature) for 45-120 min. Completion of reaction was checked by TLC. During the reaction a white precipitate separated. The mixture was extracted with ice-water (2 x 15 mL) which caused the precipitate to dissolve. The organic layer was dried over $MgSO_4$. Filtration and solvent removal gave a residue (solid or oil) which was crystallized from CH_2Cl_2/hexane or Et_2O/hexane to give the corresponding FMOC-N-protected amino acid fluoride as a white crystalline solid. The crude and recrystallized acid fluorides were analyzed by HPLC following the same technique described for the corresponding chlorides except that it was necessary to wait for 15-300 min following addition of the fluoride to dry methanol in order to allow time for complete conversion to the methyl ester. For example, a mixture initially analyzing for 93.45% FMOC-Gly-F (as methyl ester) came to complete conversion after 50 min with a measured content of 98.89% FMOC-Gly-OMe and 1.00% FMOC-Gly-OH. In case of FMOC-Val-F, initially analyzing for 82.70% FMOC-Val-F (as methyl ester) complete conversion occurred after 5 hours with a measured content of 98.19% FMOC-Val-OMe and 0.94% FMOC-Val-OH. Fischer esterification of the free acid in the methanolic HF solution did not occur. For example, a mixture initially containing 52.27% of FMOC-Gly-OH showed no significant change after 15 hours (52.19% acid) and the same results were observed in case of FMOC-Val-OH.

EXAMPLE 1

N-(9-Fluorenylmethoxycarbonyl)glycine Fluoride:

The reaction was used in accordance with the procedure described above.

Reaction was completed after 2 hours of reflux, the fluoride being obtained in 80.5% yield as pale yellow needles, mp 140.1°C, (98.9% pure according to HPLC analysis); IR (KBr) 3337 (NH), 1843 (COF), 1680 (OCON) cm^{-1}; ^1H NMR ($CDCl_3$) δ 4.1-4.3 (m, 3, $C\underline{H}CH_2$), 4.5 (d, 2, NCH_2CO), 5.3 (bs, 1, NH), 7.15 - 7.8 (m, 8, aryl).

Anal. Calcd for $C_{17}H_{14}FNO_3$: C, 68.22; H, 4.71; N, 4.67.
Found: C, 68.25, H, 4.63; N, 4.79.

EXAMPLE 2

N-(9-Fluorenylmethoxycarbonyl)alanine Fluoride:

The reaction was run in accordance with the procedure described hereinabove.

Reaction was complete after 2 hours of reflux, the fluoride being obtained in 75.4% yield as a white solid, mp 111-2°C, (98.66% pure according to HPLC analysis); $[\alpha]D^{23}$ + 3.6° (c 0.5, EtOAc); IR (KBr) 3326 (NH), 1845 (COF), 1690 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.6 (d, 3, CH_3), 4.2 (t, 1, $C\underline{H}CH_2$), 4.5 (m, 3, CH_2O, NCHCO), 5.2 (d, 1, NH), 7.2-7.8 (m, 8, aryl).

Anal. Calcd for $C_{18}H_{16}FNO_3$: C, 69.00; H, 5.14; N, 4.47.
Found: C, 69.16; H, 5.30; N, 4.30.

EXAMPLE 3

N-(9-Fluorenylmethoxycarbonyl)valine Fluoride:

The reaction was run in accordance with the procedure described hereinabove.

Reaction was complete after 2 hours of reflux, the fluoride being obtained in a yield of 70.2% as a white solid, mp 113-4°C, (98.62% pure according to HPLC analysis); $[\alpha]D^{24}$ + 10.7° (c 1, CH_2Cl_2); IR (KBr) 3312 (NH), 1843 (COF), 1688 (OCON) cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.0 (d, 6, CH_3), 2.3 (m, 1, $C\underline{H}CH_2$), 4.2 (t, 1, $C\underline{H}CH_3$), 4.5 (m, 3, CH_2O, NCHCO), 5.15 (d, 1, NH), 7.2-7.8 (m, 8, aryl).

Anal. Calcd for $C_{20}H_{20}FNO_3$: C, 70.36; H, 5.90; N, 4.10.
Found: C, 70.27; H, 5.92; N, 4.19.

EXAMPLE 4

N-(9-Fluorenylmethoxycarbonyl)leucine Fluoride:

The reaction was run in accordance with the procedure described above.

Reaction was complete after 1 hour of reflux, the fluoride being obtained in a yield of 75.2% as a white solid, mp 95.5-6.5°C, (98.58% pure according to HPLC analysis); $[\alpha]D^{23}$ - 5.8° (c 1, EtOAc); IR (KBr) 3336 (NH), 1938 (COF), 1699 (OCON) cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.00 (d, 6, CH_3), 1.6-1.8 (m, 3, CH_2CH), 4.2 (t, 1, $C\underline{H}CH_2O$), 4.5 (m, 3, CH_2O, NCHCO), 5.1 (d, 1, NH), 7.2-7.8 (m, 8, aryl).

Anal. Calcd for $C_{21}H_{22}FNO_3$: C, 70.96; H, 6.23; H, 3.94.
Found: C, 70.70; H, 6.48; N, 4.15.

EXAMPLE 5

N-(9-Fluorenylmethoxycarbonyl)isoleucine Fluoride:

The reaction was run in accordance with the procedure described above.

Reaction was complete after 1.5 hours of reflux, the fluoride being obtained in a yield of 73.3% as a white solid, mp 115-6°C, (97.13%) pure according to HPLC analysis); $[\alpha]D^{23}$ + 15.6° (c 0.5, EtOAc); IR (KBr) 3304 (NH), 1840 (COF), 1996 (OCON) cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.00 (m, 6, $CH(\underline{Me})CH_2\underline{Me}$), 1.1-1.6 (m, 2, $CHC\underline{H}_2CH_3$), 2.00 (m, 1, CH), 4.2 (t, 1, $C\underline{H}CH_2O$), 4.5 (m, 3, CH_2O, NCHCO), 5.2 (d, 1, NH), 7.2-7.8 (m, 8, aryl).

Analy. Calcd for $C_{21}H_{22}FNO_3$: C, 70.96; H, 6.23; N, 3.94.
Found: C, 68.30; H, 6.06; N, 3.87.

EXAMPLE 6

N-(9-Fluorenylmethoxycarbonyl)proline Fluoride:

The reaction was run in accordance with the procedure described above.

Reaction was complete after 12 hours of stirring at room temperature, the fluoride being obtained in a yield of 78.2% as a white solid mp, 88-9°C; $[\alpha]D^{30}$ - 28.6° (c 0.5, EtOAc).
Anal. Calcd for $C_{20}H_{18}FNO_3$: C, 70.78; H, 5.34; N, 4.12.
Found: C, 70.86; H, 5.43, N, 4.21.

EXAMPLE 7

N-(9-Fluorenylmethoxycarbonyl)phenylalanine Flouride:

The reaction was run in accordance with the procedure described above.

Reaction was complete after 1 hour of reflux, the fluoride being obtained in a yield of 63.9% as white crystals, mp 118-20°C, (99.3% pure according to HPLC analysis); $[\alpha]D^{24}$ + 35.5° (c 1, CH_2Cl_2); IR (KBr) 3318 (NH),

1843 (COF), 1700 (OCON) cm^{-1}; ^1H NMR ($CDCl_3$) δ 3.2 (d, 2, $\underline{CH_2}C_4H_5$), 4.2 (t, 1, $\underline{CH}CH_2O$), 4.45 (m, 2, CH_2O), 4.85 (m, 1, NCHCO), 5.1 (d, 1, NH), 7.1-7.8 (m, 13 aryl).

Anal. Calcd for $C_{24}H_{20}FNO_3$: C, 74.03; H, 5.14; N, 3.59; F, 4.88. Found: C, 74.03; H, 5.13; N, 3.69; F, 4.68.

EXAMPLE 8

N-(9-Fluorenylmethoxycarbonyl)tryptophan Fluoride:

The reaction was run in accordance with the procedure described above.

Reaction was complete after 1 hour of stirring at room temperature, the fluoride being obtained in a yield of 70.7% as a white solid, mp 125-8°C (dec.) (98.2% pure according to HPLC analysis); $[\alpha]D^{24}$ - 5.2° (c 1, EtOAc); IR (KRr) 3390 and 3360 (NH), 1845 (COF), 1697 (OCON) cm^{-1}, ^1H NMR ($CDCl_3$) δ 3.4 (d, 2, $C\underline{H_2}CHCO$), 4.2 (t, 1, $C\underline{H}CH_2O$), 4.4 (d, 2, $CHC\underline{H_2}O$), 4.9 (m, 1, NCHCO), 5.3 (d, 1, NH), 7.0-8.2 (m, 14, NH + aryl).

Anal. Calcd for $C_{33}H_{31}FNO_3$: C, 72.88; H, 4.94; N, 6.53. Found: C, 72.83, H, 5.01; N, 6.43.

EXAMPLE 9

N-(9-Fluorenylmethoxycarbonyl)-O-(t-Butyl)serine Fluoride

The reaction was run in accordance with the procedure described above.

Reaction was completed after 1 hour of stirring at room temperature, the fluoride being obtained in a yield of 72.7% as white crystals, mp 89-91°C, (98.26% pure according to HPLC analysis); $[\alpha]D^{26}$+ 28.8° (, 5, EtOAc); IR (KBr) 3444 (NH), 1868 (COF), 1733 (OCON) cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.2 (s, 9,

CMe_3), 3.6 (q, 1, CHHOCMe), 3.9 (q, 1, CHHOCMe_3), 4.22 (t, 1, CHCH_2OCO), 4.45 (m, 2, CH_2OCO), 4.7 (m, 1, NCHCO), 5.65 (d, 1, NH), 7.25-7.8 (m, 8, aryl).

Anal. Calcd for $C_{22}H_{24}FNO_4$: C, 68.55; H, 6.27; N, 3.63.
Found: C, 68.49; N, 6.32; N, 3.67.

EXAMPLE 10

N-(9-Fluorenylmethoxycarbonyl)-O-(t-Butyl)threonine Fluoride:

The reaction was run in accordance with the procedure described hereinabove:

Reaction was complete after 1 1/2 hours of stirring at room temperature, the fluoride being obtained in a yield of 72.6% as white crystals, mp 53.5°C, (98.03% pure according to HPLC analysis); $[\alpha]D^{27}$ + 12.3° (c 0.4 EtOAc); IR (KBr) 3320 (NH), 1857 (COF), 1726 (OCON) cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.15 (s, 9, CMe_3), 1.3 (d, 3, CH_3), 4.3-4.5 (m, 5, CHCH_2OCO, NCH(CHO-)CO), 5.6 (d, 1, NH), 7.2-7.8 (m, 8, aryl).
Anal. Calcd for $C_{23}H_{22}FNO_4$: C, 69.15; H, 6.56; N, 3.50.
Found: C, 69.11; H, 6.83; H, 4.00.

EXAMPLE 11

N -(9-Fluorenylmethoxycarbonyl)-Nϵ-(t-Butyloxycarbonyl)-lysine Fluoride:

The reaction was run in accordance with the procedure described above.

Reaction was completed after 1 hour of stirring at room temperature, the fluoride being obtained in a yield of 65.9% as white crystals, mp 128-30°C (99.5% pure according to HPLC

analysis); $[\alpha]D^{23}$ - 2.2° (c 0.5, CH_2Cl_2); IR (KBr) 2254 (NH), 1854 and 1836 (COF), 1693 (OCON) cm^{-1}; ^1H ($CDCl_3$) 1.4 (s, 9, CMe_3), 1.5-2 (m, 6, CH_2-CH_2-CH_2), 3.15 (m, 1, $\underline{CH_2}NH$), 4.2 (t, 1, $\underline{CH}CH_2O$), 4.4-4.6 (m, 4, $\underline{CH_2}O$, N$\underline{CH}$CO, $CH_2\underline{NH}$), 5.7 (d, 1, NH), 7.2-7.8 (m, 8, aryl).

Anal. Calcd For $C_{28}H_{31}FN_2O_5$: C, 66.36; H, 6.64; H, 5.95.
Found: C, 66.16; H, 6.50; N, 5.92.

EXAMPLE 12

N-(9-Fluorenylmethoxycarbonyl)aspartic Acid Fluoride-β-(t-Butyl) Ester:

The reaction was run in accordance with the procedure described above.

Reaction was completed after 30 minutes of stirring at room temperature, the fluoride being obtained in a yield of 67.8% as white crystals, mp 74-5°C, (97.97% pure according to HPLC analysis); $[\alpha]D^{23}$ + 4.00° (c 0.5, EtOAc); IR (KBr) 3320 (NH), 1856 (COF), 1725 (COO), 1695 (OCON) cm^{-1}, ^1H NMR ($CDCl_3$) δ 1.45 (s, 9, CMe_3), 2.9 (dq, 2, CH_2COO), 4.3 (t, 1, $\underline{CH}CH_2OCO$), 4.5 (m, 2, $CH\underline{CH_2}O$), 4.85 (m, 1, HCHCO), 5.85 (d, 1, NH), 7.2-7.8 (m, 8, aryl).

Anal. Calcd for $C_{23}H_{24}FNO_5$: C, 66.81; H, 5.85; N, 3.38.
Found: C, 67.03; H, 5.95; N, 3.70.

The results of the above synthesis are summarized in the Table hereinbelow:

Table 1

Synthesis of FMOC-Amino Acid Fluorides

Compound	Yield %	mp(°C)	$[\alpha]_D^{t°C}$
FMOC-Gly-F	80.5	140-1	
FMOC-Ala-F	75.4	111-2	+3.6° (c 0.5, EtOAc, 23)
FMOC-Val-F	70.2	113-4	+10.7° (c 1, CH_2Cl_2, 24)
FMOC-Leu-F	75.2	95-6	-5.8° (c 1, EtOAc, 23)
FMOC-Ile-F	73.3	115-6	+15.6° (c 0.5, EtOAc, 23)
FMOC-Phe-F	63.9	118-20	+35.5° (c 1, CH_2Cl_2, 24)
FMOC-Trp-F	70.7	125-8	-5.2° (c 1, EtOAc, 24)
FMOC-Ser(tBu)-F	72.7	89-91	+28.8° (c 0.5, EtOAc, 26)
FMOC-Thr(tBu)-F	72.6	53-5	+12.3° (c 0.4, EtOAc, 27)
FMOC-Lys(BOC)-F	65.9	128-30	-2.2° (c .5, CH_2Cl_2, 24)
FMOC-Asp(OtBu)-F	67.8	74-5	+4.0° (c 0.5, EtOAc, 23)

These results indicate that cyanuric fluoride is suitable not only for the preparation of simple FMOC-amino acid fluorides but also for those containing t-BOC, t-Bu or CBZ-groups on the side chain. The amino acid fluorides described hereinabove were obtained in crystalline formed.

The use of amino acid fluorides of the present invention being used as peptide coupling agents is illustrated hereinbelow.

EXAMPLE 13

t-Butyl Allyl Sulfide. To a solution of 350 mL of anhydrous ethanol maintained under nitrogen was slowly added 22.99 g (1 mol) of sodium spheres. The sodium dissolved within 90 minutes, and to the resulting sodium ethoxide solution was added 90.19 g (1 mol) of t-butyl mercaptan with mechanical stirring. Allyl bromide (120.98 g; 1 mol) was then added dropwise to the mechanically stirred sodium t-butyl thiolate solution. After the addition was complete, the mixture was refluxed for 10 minutes, the solution allowed to cool, the precipitated sodium bromide filtered, and the ethanol removed by distillation at atmospheric pressure. The residue was diluted with 200 mL of water, and the layers separated. The aqueous layer was extracted with five 40-mL portions of ether. The combined organic layers were extracted with 150 mL of water, the organic layer dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give a yellow liquid. Distillation through a 0.8 x 15-cm fractionating column gave 56.16 g (45%) of the sulfide as a colorless liquid, bp 139-141°C; IR (neat), cm^{-1} 3090, 2960 1635, 1455, 1360, 1160, 985, 915; ^1H NMR ($CDCl_3$) 1.35 (s, 9H, t-butyl), 3.20 (d, 2H, CH_2), 4.90-6.25 (m 3H, vinyl).

EXAMPLE 14

1,3-Dibromo-2-(t-butylsulfonyl) Propane. To a stirred solution of 17.37 g (0.13 mol) of t-butyl allyl sulfide in 133 mL of CCl_4 at -24°C (CCl_4/dry ice) was added dropwise a solution of 21.33 g (0.13 mol) of Br_2 in 67 mL of CCl_4. A yellow solid precipitated during the addition. The mixture was warmed to room temperature and stirred for 10 minutes following complete solution of the yellow solid. The resulting solution was poured into a mixture of 55.50 g (0.27 mol) of 85% m-chloroperbenzoic acid in 490 mL of CH_2Cl_2 kept at -24°C, and the mixture stirred for 30 minutes at this temperature. The cooling bath was then removed and the mixture stirred at room temperature overnight. The precipitated m-chlorobenzoic acid was filtered and the filtrate washed with three 200-mL portions of saturated $NaHCO_3$, followed by 200 mL of water. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was recrystallized from 20% EtOAc/Skelly B to give 32.02 g (75%) of the dibromide, mp 139-140°C; IR (KBr), cm^{-1} 2980, 1470, 1365, 1285, 1105, 860, 840, 685; ^1H NMR ($CDCl_3$) 1.45 (S, 9H, t-butyl), 3.80-4.00 (m, 5H, CH and CH_2).

EXAMPLE 15

2-(Phenylsulfinyl)-2-propenyl N-p-Chlorophenyl Carbamate. A solution of 0.57 g (3.1 mmol) of 2-(phenylsulfinyl)-2-propenyl alcohol and 0.48 g (3.1 mmol) of p-chlorophenyl isocyanate in 10 mL of benzene was refluxed overnight. The solvent was removed in vacuo from a water bath at 45°C to give a brown oil which was recrystallized from CCl_4/Skelly B to give 0.72 g (69%) of the urethane, mp 106-107.5°C; IR (KBr) cm^{-1} 3240, 1730, 1600, 1545, 1490, 1310, 1220, 1030, 745, 680; ^1H NMR ($CDCl_3$) δ 4.65 (s, 1H, allylic), 4.70 (s, 1H, allylic), 6.00 (s, 1H, vinyl), 6.30 (s, 1H, vinyl), 7.10-7.85 (m, 10H, phenyl and NH).

Anal. Calcd for $C_{16}H_{14}ClNO_3S$: C, 57.23; H, 4.20; N, 4.17. Found: C, 56.95; H, 4.14; N, 4.10.

EXAMPLE 16

t-Butyl 2-(t-Butylsulfonyl)-2-propenyloxycarbonyl-L-phenylalaninate. To a stirred solution of 0.185 g (0.768 mmol) of 2-(t-butylsulfonyl)-2-propenyl chloroformate in 2 mL of benzene at 0°C was added dropwise a solution of 0.34 g (1.53 mmol) of t-butyl L-phenylalaninate in 5 mL of benzene. Upon addition, a white precipitate began to separate. Upon completion of the addition, the slurry was stirred at room temperature for 30 minutes, poured into 15 mL of ether and washed with three 15-mL portions of 5% HCl, and 15 Ml of water. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The resulting oil was recrystallized from 20% ether/pentane to give 0.23 g (70%) of the colorless ester, mp 67-69°C; IR (KBr) cm^{-1} 3410, 2980, 1715, 1510, 1365, 1290, 1155, 1095, 1060, 940, 750, 700, 625; ^1H NMR ($CDCl_3$) δ 1.45 (s, 9H, t-butyl), 1.50 (s, 9H, t-butyl), 3.12 (m, 2H, benzyl), 4.56 (q, 1H, CH), 4.90 (s, 2H, CH_2O), 5.38 (d, 1H, NH), 6.14 (s, 1H, vinyl), 6.29 (s, 1H, vinyl), 7.15-7.40 (m, 5H, Phenyl); $[\alpha]_D^{28}$ +25.4° (c=1, CH_2Cl_2), also, $[\alpha]_{546}^{28}$ +30.9° (c=1, CH_2Cl_2).

Anal. Calcd for $C_{21}H_{31}NO_6S$: C, 59.27; H, 7.34; N, 3.29. Found: C, 59.31; H, 7.45; N, 3.23.

EXAMPLE 17

2-(t-Butylsulfonyl)-2-propenyloxycarbonyl-L-phenylalanine. A solution of 4.57 g (19.0 mmol) of 2-(t-butylsulfonyl)-2-propenyl chloroformate and 5.64 g (18.6 mmol) of t-butyl L-phenylalaninate hydrophosphite in 90 mL of CH_2Cl_2 was stirred in the presence of 165 mL of 5% $NaHCO_3$ at room temperature for 2 hours. The aqueous phase was separated, and the organic phase washed with three 75-mL portions of 5% HCl. The organic phase was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The resulting oil was dissolved in 36 mL of 50% CH_2Cl_2/triflouroacetic acid, and the solution stirred at room temperature for

(91%) of the colorless acid, mp 88.0-89.5°C; IR (KBr) cm^{-1} 3270, 1760 1690, 1520, 1290, 1200, 1100, 1060, 960, 755, 700, 630; ^1H NMR ($CDCl_3$) 1.40 (s, 9H, t-butyl), 3.20 (m, 2H, benzyl), 4.75 (q, 1H, CH), 4.90 (s, 2H, CH_2O), 5.35 (d, 1H, NH), 6.15 (s, 1H, vinyl), 6.32 (s, 1H, vinyl), 7.15-7.40 (m, 5H, phenyl); $[\alpha]_D^{24}$ -31.0° (c=0.5, DMF), also $[\alpha]_{546}^{24}$ -37.5° (c=0.5 DMF).

Anal. Calcd for $C_{17}H_{23}NO_6S$: C, 55.27; H, 6.27; N, 3.79. Found: C, 55.02; H, 6.47; N, 3.71.

EXAMPLE 18

2-(t-Butylsulfonyl)-2-propenyloxycarbonyl-L-phenylalanyl fluoride. The compound of Example 17 is reacted with cyanuric fluoride as described herein to prepare the above product.

EXAMPLE 19

2-(t-Butylsulfonyl)-2-propenyloxycarbonyl Glycine.

A mixture of 3.34 g (13.9 mmol) of 2-(t-butylsulfonyl)-2-propenyl chloroformate and 2.95 g (13.8 mmol) of t-butyl glycinate hydrophosphite in 85 mL of CH_2Cl_2 was stirred in the presence of 100 mL of 5% $NaHCO_3$ at room temperature for 4 hours. The aqueous phase was separated, and the organic phase washed with three 33-mL portions of 5% HCl. The organic phase was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The resulting oil was dissolved in 20 mL of 50% CH_2Cl_2/triflouroacetic acid, and the solution stirred at room temperature for 2 hours. Excess triflouroacetic acid and solvent were removed in vacuo from a water bath at 45°C. The resulting oil was recrystallized from 30% EtOAc/Skelly B to give 3.25 g (84%) of the acid, mp 105.0-105.5°C; IR (KBr) cm^{-1} 3355, 1765, 1695, 1560, 1290, 1190, 1100, 1050, 770; ^1H NMR ($CDCl_3$) δ 1.41 (s, 9H, t-butyl), 4.05 (d, 2H, CH_2), 4.94 (s, 2H, CH_2O), 5.44 (t, 1H, NH), 6.26 (s, 1H, vinyl), 6.33 (s, 1H, vinyl).

Anal. Calcd for $C_{10}H_{17}NO_6S$: C, 43.00; H, 6.13; N, 5.01. Found: C, 43.00; H, 6.03; N, 4.97.

EXAMPLE 20

2-t-butylsulfonyl-2-propenyloxycarbonyl glycine fluoride. The product of Example 19 is reacted with cyanuric fluoride as described herein to prepare the above product.

EXAMPLE 21

2-(t-Butylsulfonyl)-2-propenyloxycarbonyl-D-valine. A solution of 1.01 g (4.15 mmol) of 2-(t-butylsulfonyl)-2-propenyl chloroformate and 0.87 g (4.15 mmol) of t-butyl D-valinate hydrochloride in 22 mL of CH_2Cl_2 was stirred in the presence of 45 mL of saturated $NaHCO_3$ for 90 minutes. The aqueous phase was separated, and the organic phase was washed with two 20-mL portions of 5% HCl. The organic phase was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The resulting oil was dissolved in 10 mL of 50% CH_2Cl_2/trifluoroacetic acid, and the solution stirred at room temperature for 90 minutes. Excess triflouroacetic acid and solvent were removed in vacuo from a water bath at 45°C. The resulting oil was recrystallized from ether/Skelly F to give 1.01 g (75%) of the colorless acid, mp 113-114°C; IR (KBr) cm^{-1} 3380, 3160, 1750, 1700, 1540, 1400, 1295, 1100, 1025, 760, 665; ^1H NMR ($CDCl_3$) δ 0.9 (d, 3H, CH_3), 1.05 (d, 3H, CH_3), 1.4 (s, 9H, t-butyl), 2.2 (m, 1H, CH), 4.3 (d of d, 1H, CHN), 4.95 (s, 2H, CH_2O), 5.5 (d, 1H, NH), 6.25 (s, 1H, vinyl), 6.35 (s, 1H, vinyl), 11.6 (s, 1H, CO_2H); $[\alpha]_D^{25}$ +3.2° (c=1, $CHCl_3$), also $[\alpha]_{546}^{25}$+3.7° (c=1, $CHCl_3$).

Anal. Calcd for $C_{13}H_{23}NO_6S$: C, 48.58; H, 7.21; N, 4.36. Found: C, 48.70; H, 6.99; N, 4.29.

EXAMPLE 22

2-t-butylsulfonyl-2-propenyloxycarbonyl fluoride. The product of Example 21 is reacted with cyanuric fluoride as described herein to prepare the above product.

EXAMPLE 23

2-(t-Butylsulfonyl)-2-propenyl Bromide. A mixture of 16.78 g (0.052 mol) of 1,3-dibromo-2-(t-butylsulfonyl) propane and 14 mL (0.12 mol) of 2,6-lutidine in 55 mL of CH_2Cl_2 was refluxed for 75 minutes. The solution was allowed to cool to room temperature and extracted with three 80-mL portions of 5% HCl followed by 80 mL of water. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give 11.46 g (91%) of the allyl bromide as a white solid, mp 40.5-42.0°C, which was used without further purification; IR (KBr) 2980 cm^{-1}, 1480, 1395, 1230, 1100, 970, 730, 640; ^1H NMR ($CDCl_3$) δ 1.40 (s, 9H, t-butyl), 4.30 (s, 2H CH_2Br), 6.50 (s, 2H, vinyl).

EXAMPLE 24

2-(t-Butylsulfonyl-2-propenyl Alcohol. A mixture of 8.55 g (35.3 mmol) of 2-(t-butylsulfonyl)-2-propenyl bromide and 5.31 g (78.1 mmol) of sodium formate in 150 mL of methanol was refluxed overnight. The solution was allowed to cool and concentrated to 50 mL with the aid of a water aspirator, resulting in the precipitation of excess sodium formate. The residue was diluted with 150 mL of water and extracted with five 50-mL portions of CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was recrystallized from 15% EtOAc/Skelly F to give 4.30 g (68%) of the alcohol as a colorless solid, mp 53.5-54.5°C; IR (KBr), cm^{-1} 3470, 3120, 3000, 1450, 1370, 1270, 1095, 1050, 960, 900, 800, 750, 630; ^1H NMR ($CDCl_3$) δ 1.39 (s, 9H, t-butyl), 2.57 (t, 1H, OH), 4.56 (d, 2H CH_2O), 6.30 (s, 1H, vinyl), 6.31 (s, 1H, vinyl).

Anal. Calcd for $C_7H_{14}O_3S$: C, 47.17; H, 7.92; S. 17.99. Found: C, 47.07; H, 7.95; S, 17.70.

EXAMPLE 25

2-(t-Butylsulfonyl)-2-propenyl Chloroformate. To a solution of 6.67 g (37.4 mmol) of 2-(t-butylsulfonyl)-2-propenyl alcohol in 27 mL of dry THF at 0°C was added in one portion 27 mL of phosgene. The solution was stirred for 1 hour at 0°C and allowed to stand at room temperature overnight. Excess phosgene and solvent were removed under reduced pressure. The crude product was recrystallized from 25% ether/Skelly B to give 8.23 g (91%) of the chloroformate as a colorless solid, mp 56.5-57.7; IR (KBr) cm^{-1} 2980, 1755, 1430, 1380, 1290, 1140, 1100, 965, 915, 810, 750, 680, 630; ^1H NMR ($CDCl_3$) δ 1.41 (s, 9H, t-butyl), 5.11 (s, 2H, CH_2O), 6.37 (s, 1H, vinyl), 6.47 (s, 1H, vinyl).

Anal. Calcd for $C_8H_{13}ClO_4S$: C, 39.92; H, 5.44; S, 3.32. Found: C, 40.10; H, 5.40; S, 13.07.

EXAMPLE 26

2-(t-Butylsulfonyl)-2-propenyl N-p-Chlorophenyl Carbamate. To a solution of 0.24 g (1.0 mmol) of 2-(t-butylsulfonyl)-2-propenyl chloroformate in 3 mL of benzene at 0°C was added dropwise 0.26 g (2.0 mmol) of p-chloroaniline 3 mL of benzene. A white precipitate separated almost immediately. After the addition was complete the mixture was stirred at 0°C for 10 minutes and for 2 hours at room temperature. The mixture was then diluted with 15 mL of benzene and extracted with two 15-mL portions of 5% HCl, followed by 15 mL of water. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was recrystallized from 20% EtOAc/Skelly B to give 0.26 g (79%) of the urethane, mp 124-125°C. The same compound was obtained in 85% yield by treatment of 2-(t-butylsulfonyl)-2-propenyl alcohol with p-chlorophenyl isocyanate in refluxing benzene; IR (KBr) cm^{-1} 3340, 3120, 2980, 1730, 1600, 1490, 1430, 1280, 1210, 1070, 975, 915, 830, 750, 620; ^1H NMR

($CDCl_3$) δ 1.4 (s, 9H, t-butyl), 4.0 (t, J=1 Hz, 2H, CH_2O), 6.3 (s, 1H, vinyl), 6.35 (s, 1H, vinyl), 7.15-7.5 (m, 5H, phenyl and NH).

Anal. Calcd. for $C_{14}H_{18}ClNO_4S$: C, 50.68; H, 5.47; N, 4.22. Found: C, 50.46; H, 5.47; N, 4.28.

EXAMPLE 27

2-Phenylthio-2-propenyl Alcohol. To a stirred solution of 36.0 g (0.64 mol) of propargyl alcohol and 0.12 g (1.8 mmol) of powdered potassium hydroxide, was added dropwise at 125°C over 30 minutes, 60.0 g (0.54 mol) of thiophenol. The mixture was stirred for an additional 90 minutes, and allowed to cool to room temperature. The brown solution was diluted with 200 mL of ether, and extracted with three 100-mL portions of 2 N sodium hydroxide, and two 100-mL portions of water. The organic phase was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give 90.4 g of a brown liquid, which, according to NMR analysis, contained 34% of the desired isomer. The mixture was fractionally distilled through a 0.8 x 20-cm column packed with glass helices to give 29.5 g of a colorless liquid, which by NMR analysis contained 67% of the desired isomer, bp 109-116°C/1.2 Torr. This liquid was chromatographed on silica gel (100-200 mesh, 50 g/gram of compound) with 20% EtOAc/Skelly B as eluant, to give 16.4 g (18%) of the sulfide as a colorless liquid; IR (neat) cm^{-1} 3360, 1610, 1580, 1470, 1430, 1040, 740, 690; ^1H NMR ($CDCl_3$) δ 3.70 (t, 1H, OH), 4.10 (d, 2H, CH_2), 5.2 (t, J=1 Hz, 1H, vinyl), 5.55 (t, J=1 Hz, 1H, vinyl), 7.15-7.55 (m, 5H, phenyl).

EXAMPLE 28

2-(Phenylsulfonyl)-2-propenyl Alcohol. A mixture of 5.03 g (30.3 mmol) of 2-(phenylthio)-2-propenyl alcohol and 12.74 g (62.7 mmol) of 85% m-chloroperbenzoic acid in 250 mL of CH_2Cl_2 was stirred overnight at room temperature. The mixture was extracted with three 100-mL portions of saturated $NaHCO_3$, followed by 100 mL of water. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The residue was chromatographed on silica gel (100-200 mesh, 50 g/gram of compound) with 40% EtOAc/Skelly B as eluant, to give 4.30 g (72%) of the sulfone as a colorless oil; IR (neat) cm^{-1} 3500, 1580, 1440, 1300, 1170, 1130, 1050, 950, 900, 750, 690; ^1H NMR ($CDCl_3$) δ 3.70 (bs, 1H, OH), 4.20 (bs, 2H, CH_2) 6.05 (bs, 1H, vinyl), 6.35 (bs, 1H, vinyl) 7.30-7.95 (m, 5H, phenyl).

Anal. Calcd for $C_9H_{10}O_3S$: C, 54.53; H, 5.08; S. 16.17. Found: C, 54.57; H, 5.13; S. 16.02.

EXAMPLE 29

2-(Phenylsulfonyl)-2-propenyl N-p-Chlorophenyl Carbamate. A solution of 3.38 g (17.1 mmol) of 2-(phenylsulfonyl)-2-propenyl alcohol and 2.62 g (17.1 mmol) of p-chlorophenyl isocyanate in 15 mL of benzene was refluxed overnight. The solvent was removed in vacuo from a water bath at 45°C to give a solid which was recrystallized twice from CCl_4 to give 4.25 g (71%) of the urethane, mp 104-106°C; IR (KBr) cm^{-1} 3320, 1730, 1600, 1530, 1300, 1215, 1065, 960, 820, 750, 680; ^1H NMR ($CDCl_3$) δ 4.85 (bs, 2H, CH_2), 6.16 (bs, 1H, vinyl), 6.50 (bs, 1H, vinyl), 7.05-8.00 (m, 10H, phenyl and NH).

Anal. Calcd for $C_{16}H_{14}ClNO_4S$: C, 54.63; H, 4.01; N, 3.98. Found: C, 54.54; H, 3.99; N, 4.05.

EXAMPLE 30

2-(Methylsulfonyl)ethyl N-p-Chlorophenyl Carbamate. A solution of 2.06 g (16.6 mmol) of 2-(methylsulfonyl)ethyl alcohol and 2.55 g (16.6 mmol) of p-chlorophenyl isocyanate in 15 mL of benzene was refluxed for 30 minutes. The solvent was removed in vacuo from a water bath at 45°C to give a solid which was recrystallized from $CHCl_3$ to give 2.98 g (65%) of the urethane, mp 147-147.5°C; IR (KBr) 3360 cm^{-1}, 1720, 1600, 1490, 1310, 1230, 1130, 1070, 830, 760; ^1H NMR (DMSO-d_6-$CDCl_3$) δ 3.0 (s, 3H, CH_3), 3.40 (t, 2H, CH_2SO_2), 4.60 (t, 2H, CH_2O), 7.15-7.55 (m, 4H, phenyl), 9.20 (bs, 1H, NH).

Anal. Calcd for $C_{10}H_{12}ClNO_4S$: C, 43.25; H, 4.36; N, 5.04. Found: C, 43.16; H, 4.14; N, 4.99.

EXAMPLE 31

2-Carboethoxy-2-propenyl Alcohol. To a stirred solution of 12.07 g (53.8 mmol) of triethyl phosphonacetate and 21 mL of 35-40% aqueous formaldehyde at room temperature was added dropwise a solution of 13.2 g (95.5 mmol) of K_2CO_3 in 50 mL of water. The temperature of the reaction mixture rose to 48°C over the course of the addition. After the addition was complete, the mixture was stirred for 1 hour, quenched with 22 mL of saturated NH_4Cl solution, and extracted with three 25-mL portions of ether. The organic layer was dried over Na_2SO_4, filtered, and the solvent removed in vacuo from a water bath at 45°C to give a colorless liquid which was distilled to give 4.57 g (65%) of the colorless alcohol, bp 60-65°C/0.9 Torr; IR (neat) cm^{-1} 3450, 1710, 1635, 1450, 1400, 1300, 1260, 1160, 1050, 945, 820; ^1H NMR ($CDCl_3$) δ 1.30 (t, 3H, CH_3), 3.5-4.5 (m, 5H, CH_2OH and CO_2CH_2), 5.90 (bs, 1H, vinyl), 6.25 (bs, 1H, vinyl).

EXAMPLE 32

2-Carboethoxy-2-propenyl N-p-Chlorophenyl Carbamate. A solution of 2.12 g (16.3 mmol) of 2-(carboethoxy)-2-propenyl alcohol and 2.50 g (16.3 mmol) of p-chlorophenyl isocyanate in 15 mL of benzene was refluxed overnight. The solvent was removed in vacuo from a water bath at 45°C to give a solid which was recrystallized from CCl_4 to give 2.99 g (65%) of the urethane, mp 93.0-93.5°C; IR (KBr) cm^{-1} 3360, 1720, 1600, 1525, 1390, 1310, 1220, 1170, 1060; ^1H NMR (DMSO-d_6/$CDCl_3$) δ 1.25 (t, 3H, CH_3), 4.25 (q, 2H, CO_2CH_2), 4.90 (s, 2H, CH_2O), 5.90 (bs, 1H, vinyl), 6.35 (bs, 1H, vinyl), 7.15-7.50 (m, 4H, phenyl), 8.0 (bs, 1H, NH).

Anal. Calcd. for $C_{13}H_{14}ClNO_4$: C, 55.04; H, 4.97; N, 4.94. Found: C, 54.89; H, 4.69; N, 4.89.

EXAMPLE 33

2-(Phenylsulfinyl)-2-propenyl Alcohol. To a stirred solution of 0.91 g (5.5 mmol) of 2-(phenylthio)-2-propenyl alcohol in 60 mL of CH_2Cl_2 at -78°C was added 1.11 g (5.5 mmol) of 85% m-chloroperbenzoic acid. The reaction mixture was stirred for 1 hour at -78°C. The cooling bath was removed and the mixture allowed to warm to room temperature. The mixture was filtered, and then extracted with three 50-mL portions of saturated $NaHCO_3$. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The residue was flash chromatographed on silica gel (230-400 mesh, 4 x 12-cm packed column) with 80% EtOAc/Skelly B as eluant, to give 0.57 g (57%) of the sulfoxide as a colorless oil; IR (neat) cm^{-1} 3360, 1440, 1030, 990, 930, 750, 690; ^1H NMR ($CDCl_3$) δ 4.00 (s, 1H, allylic), 4.15 (s, 1H, allylic), 4.40 (bs, 1H, OH), 5.90 (t, J=1 Hz, 1H, vinyl), 6.05 (s, 1H, vinyl), 7.35-7.80 (m, 5H, phenyl).

EXAMPLE 34

trans-Phenyl β-Styryl Sulfide. To a stirred solution of 14.0 g (0.137 mol) of freshly distilled phenylacetylene at 0°C was added dropwise 15.1 g (0.137 mol) of thiophenol. The solution was stirred at room temperature overnight. The reaction mixture was distilled to give 22.2 g (76%) of a colorless liquid, bp 177-181°C/5 Torr. NMR analysis showed a trans/cis-ratio of 80/20. ^{1}H NMR ($CDCl_3$) δ 6.53 (d, J=1, Hz, 1H, cis-vinyl), 6.81 (d, J=2 Hz 1H, trans-vinyl).

EXAMPLE 35

trans-Phenyl β-Styryl Sulfone. To a slightly frozen mixture of 8.17 g (38.5 mmol) of an 80/20 trans-cis-mixture of phenyl β-styryl sulfide in 160 mL of acetic acid was added dropwise 16 mL of 30% hydrogen peroxide. The solution was refluxed for 1 hour, poured on crushed ice to give an oil which soon solidified, and was filtered. The crude solid was dissolved in 100 mL of CH_2Cl_2, dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give 6.64 g (71%) of a solid which contained approximately 15% of the cis-isomer as determined by ^{1}H-NMR analysis. Recrystallization from 30% EtOAc/Skelly B gave 3.2 g (34%) of the pure colorless sulfone, mp 72.5-73.5°C; ^{1}H NMR ($CDCl_3$) 6.8 (d, J=15 Hz; 1H, vinyl), 7.2-8.1 (m, 11H, vinyl and phenyl).

EXAMPLE 36

(E)-3-Phenyl-2-(phenylsulfonyl)-2-propenyl Alcohol.

To a stirred solution of 2.0 g (8.2 mmol) of trans-phenyl β-styryl sulfone in 50 mL of dry THF at -78°C was added dropwise under a nitrogen atmosphere 6.0 mL (8.4 mmol) of 1.4 M n-BuLi within 5 minutes. The reddish-purple solution was stirred at -78°C for 30 minutes. Gaseous formaldehyde, generated by heating paraformaldehyde at 170°C, was passed through a 5-mm tube into the reaction mixture with the aid of a slow stream of nitrogen at -78°C for 45 minutes. The mixture was allowed to come to room temperature over a period of 30 minutes while continuing to pass formaldehyde through the reaction mixture until a pale yellow solution was obtained. The reaction was quenched with 75 mL of 5% HCl and extracted with 50 mL of ether. The organic layer was washed with two 50-mL portions of water, dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give 2.1 g (93%) of the crude alcohol as a brown oil. The oil was flash chromatographed in two 1.05 g batches on silica gel (230-400 mesh, 4 x 19-cm column) with 50% ether/Skelly B as eluant to give 0.99 g (44%) of a light yellow solid, mp 85-87°C. Recrystallization from 40% ether/Skelly B gave the pure colorless alcohol, mp 88-89°C; IR (KBr) cm^{-1} 3470, 1625, 1445, 1285, 1145, 1020, 770, 755, 735, 700, 680; ^1H NMR ($CDCl_3$) δ 2.63 (t, 1H, OH), 4.37 (d, 2H, CH_2O), 7.27-8.13 (m, 11H, phenyl and vinyl).

Anal. Calcd for $C_{15}H_{14}O_3S$: C, 65.67; H, 5.14; S, 11.69. Found: C, 65.60; H, 5.12; S, 11.31.

EXAMPLE 37

(E)-3-Phenyl-2-(phenylsulfonyl)-2-propenyl N-p-Chlorophenyl Carbamate. A solution of 0.56 (2.04 mmol) of (E)-3-phenyl-2-(phenylsulfonyl)-2-propenyl alcohol and 0.32 g (2.08 mmol) of p-chlorophenyl isocyanate in 10 mL of benzene was refluxed for 120 hours. The solvent was removed in vacuo from a water bath at 45°C. The crude solid was recrystallized from 20% EtOAc/Skelly B to give 0.41 g (47%) of the colorless urethane, mp 138.5-139.5°C; IR (KBr) cm^{-1} 3320, 1695, 1590, 1520, 1305, 1230, 1150, 1045; ^1H NMR ($CDCl_3$) δ 5.07 (s, 2H, CH_2O), 6.52 (bs, 1H, NH), 7.25-8.08 (m, 14H, phenyl), 8.15 (s, 1H, vinyl).

Anal. Calcd for $C_{22}H_{18}ClNO_4S$: C, 61.75; H, 4.24; N, 3.27. Found: C, 61.54; H, 4.27; N, 3.11.

EXAMPLE 38

Thiophenyl Trimethylsilyl Methane. To a stirred solution of 143 mL of 1.4 M n-BuLi (0.20 mol) in 55 mL of dry THF, was added dropwise at room temperature under a nitrogen atmosphere 24.8 g (0.20 mol) of thioanisole. Upon completion of the addition, the yellow mixture began to reflux spontaneously. The mixture was stirred at room temperature for 3 hours. Upon addition of 21.84 g (0.20 mol) of chlorotrimethylsilane, the mixture again began to reflux spontaneously. After stirring at room temperature overnight, the mixture was quenched with 100 mL of 5% of HCl. The aqueous layer was separated, and the organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was distilled to give 19.22 g (49%) of the colorless sulfide, bp 84-86°C/0.3 Torr; ^1H NMR ($CDCL_3$) δ 0.17 (s, 9H, $Si(CH_3)_3$), 2.13 (s, 2H, CH_2S), 7.0-7.42 (m, 5H, phenyl).

EXAMPLE 39

Benzenesulfonyl Trimethylsilyl Methane. To a stirred mixture of 14.1 g (69.4 mmol) of 85% m-chloroperbenzoic acid in 300 mL of CH_2Cl_2 at 0°C was added dropwise 6.82 g (34.7 mmol) of thiophenyl trimethylsilyl methane. The mixture was stirred at 0°C for 3 hours and then at room temperature overnight. The mixture was extracted with three 75-mL portions of saturated $NaHCO_3$. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was *Kugelrohred at 115°C/0.1 Torr to give 6.83 g (86%) of the clear sulfone; ^1H NMR ($CDCl_3$) δ 0.27 (s, 9H, $Si(CH_3)_3$), 2.8 (s, 2H, CH_2SO_2), 7.13-8.05 (m, 5H, phenyl).

EXAMPLE 40

2,2-Diphenyl-1-(phenylsulfonyl) Ethene. To a stirred solution of 2.0 g (8.76 mmol) of benzenesulfonyl trimethylsilyl methane in 20 mL of dry THF at 0°C, was added dropwise under a nitrogen atmosphere 6.25 mL of 1.4 M n-BuLi (8.75 mmol). The reddish-orange solution was stirred at 0°C for 30 minutes. To the solution was added 1.60 g (8.78 mmol) of benzophenone. The solution was stirred at 0°C for 2 hours, then at room temperature overnight. The reaction mixture was diluted with 50 mL of 5% HCl, and extracted with three 25-mL portions of water, dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was recrystallized from 15% EtOAc/Skelly B to give 1.25 g (44%) of the slightly yellow sulfone, mp 111.0-112.5°C; ^1H NMR ($CDCl_3$) δ 7.02 (s, 1H, vinyl), 7.12-7.72 (m, 15H, phenyl).

*Trade-mark

EXAMPLE 41

3,3-Diphenyl-2-(phenylsulfonyl)-2-propenyl Alcohol.

To a stirred solution of 1.25 g (3.90 mmol) of 2,2-diphenyl-1-(phenylsulfonyl) ethene in 20 mL of dry THF at -78°C, was added dropwise under a nitrogen atmosphere 2.8 mL of 1.4 M n-BuLi (3.90 mmol). The dark black solution was stirred at -78°C for 30 minutes. Gaseous fomaldehyde, generated by heating paraformaldehyde at 170°C, was passed through a 5-mm tube into the reaction mixture with the aid of a slow stream of nitrogen at -78°C for 30 minutes. The mixture was allowed to come to room temperature over a period of 30 minutes while continuing to pass formaldehyde through the reaction mixture until a pale yellow color was observed. The mixture was stirred overnight at room temperature. The mixture was quenched with 50 mL of 5% HCl, and extracted with three 25-mL portions of CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The resulting crude oil was flash chromatographed on silica gel (230-400 mesh, 5 x 16-cm column) with 30% ether/Skelly B as eluant to give 0.47 g (34%) of the colorless alcohol. In a melting point capillary the compound does not melt but decomposes above 290°C with blackening; IR (KBr) cm^{-1} 3480, 3040, 1590, 1480, 1440, 1370, 1280, 1130, 1020, 960, 790, 730, 700, 680, 610; ^1H NMR ($CDCl_3$) δ 3.47 (t, 1H, OH), 4.61 (d, 2H, CH_2O), 6.75-7.58 (m, 15H, phenyl).

Anal. Calcd for $C_{12}H_{18}O_3S$: C, 71.98; H, 5.18; S, 9.15. Found: C. 71.97; H, 5.22; S, 9.02.

EXAMPLE 42

3,3-Diphenyl-2-(phenylsulfonyl)-2-propenyl N-p-Chlorophenyl Carbamate. A solution of 0.35 g (1.0 mmol) of 3,3-diphenyl-2-(phenylsulfonyl)-2-propenyl alcohol and 0.15 g (1.0 mmol) of p-chlorophenyl isocyanate in 2 mL of benzene was refluxed for 18 hours. The solvent was removed in vacuo from a water bath at 45°C. Recrystallization from 95% EtOAc/EtOH gave 0.38 g (78%) of the colorless urethane. In a melting point capillary the compound does not melt but decomposes above 250°C; IR (KBr) cm^{-1} 3310, 1730, 1590, 1530, 1490, 1300, 1205, 1140, 1050, 825, 700, 680; ^1H NMR ($CDCl_3$) δ 5.13 (s, 2H, CH_2O), 6.80 (bs, 1H, NH), 6.91-7.58 (m, 19H, phenyl).

Anal. Calcd for $C_{28}H_{22}ClNO_4S$: C, 66.73; H, 4.40; N, 2.78. Found: C, 66.51; H, 4.44; N, 2.54.

EXAMPLE 43

cis-Phenyl β-Styryl Sulfide. To a solution of 150 mL of anhydrous ethanol maintained under nitrogen was added 3.45 g (0.15 mol) of sodium. The sodium dissolved within 25 minutes, and to the resulting sodium ethoxide solution was added 16.5 g (0.15 mol) of thiophenol. The solution was brought to reflux, and 15.0 g (0.15 mol) of phenylacetylene was added dropwise. The solution was refluxed overnight, cooled, and poured over crushed ice. The precipitate was filtered, dissolved in 100 mL of CH_2Cl_2, dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was recrystallized from Skelly F to give 22.61 g (71%) of the sulfide, mp 43.0-44.5°C; ^1H NMR ($CDCl_3$) δ 6.51 (d, 2H, vinyl), 7.13-7.67 (m, 10H, phenyl).

EXAMPLE 44

(Z)-3-Phenyl-2-(thiophenyl)-2-propenyl Alcohol. To a solution of 1.0 g (4.7 mmol) of cis-phenyl β-styryl sulfide in 20mL of dry THF at -78°C was added dropwise under a nitrogen atmosphere 5.0 mL (7.0 mmol) of 1.4 M n-BuLi within 5 minutes. The light yellow solution was stirred at -78°C for 30 minutes. Gaseous formaldehyde, generated by heating paraformaldehyde at 170°C, was passed through 5-mm tube into the reaction mixture with the aid of a slow steam of nitrogen at -78°C for 3 hours. The mixture was allowed to come to room temperature over a period of 30 minutes and stirred overnight. The mixture was quenched with 30 mL of 5% HCl and extracted with three 25-mL portions of CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give a brown oil. The oil was flash chromatographed on silica gel (230-400 mesh, 4 x 18-cm packed column) with 20% EtOAc/Skelly B as eluant to give 0.50 g (45%) of the alcohol, mp 64.0-65.0°C; IR (KBr) cm^{-1} 3240, 3140, 1580, 1470, 1090, 1070, 1010, 740, 695; ^1H NMR ($CDCl_3$) δ 1.82 (broad, 1H, OH), 4.17 (d, 2H, CH_2O), 7.15-7.75 (m, 11H, phenyl and vinyl).

Anal. Calcd for $C_{15}H_{14}OS$: C, 74.35; H, 5.82; S, 13.23. Found: C, 74.35; H, 5.90; S, 13.18.

EXAMPLE 45

(Z)-3-Phenyl-2-(phenylsulfonyl)-2-propenyl Alcohol. A mixture of 0.47 g (1.94 mmol) of (Z)-3-phenyl-2-(thiophenyl)-2-propenyl alcohol and 0.79 g (3.89 mmol) of 85% m-chloroperbenzoic acid in 10 mL of CH_2Cl_2 was stirred at room temperature overnight. The mixture was diluted with 40 mL of CH_2Cl_2 and extracted with three 50-mL portions of saturated $NaHCO_3$. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to

give a clear oil. The oil was recrystallized from 20% EtOAc/Skelly B to give 0.27 g (51%) of the sulfone, mp 67.5-69.0°C; IR (KBr) cm^{-1} 3280, 3180, 1445, 1300, 1150, 1080, 1025, 990, 750, 730, 650, 610; ^1H NMR ($CDCl_3$) 2.85 (s, 1H, OH), 4.67 (s, 2H, CH_2O), 7.21-7.83 (m, 11H, phenyl and vinyl).

Anal. Calcd for $C_{15}H_{14}O_3S$: C, 65.67; H, .5.14; S, 11.69. Found: C, 65.45; H, 4.97; S, 11.82.

EXAMPLE 46

(Z)-3-Phenyl-2-(phenylsulfonyl)-2-propenyl N-p-Chlorophenyl Carbamate. A solution of 0.22 g (0.80 mmol) of (Z)-3-phenyl-2-(phenylsulfonyl)-2-propenyl alcohol and 0.12 g (0.78 mmol) of p-chlorophenyl isocyanate in 4 mL of benzene was refluxed overnight. The solvent was removed in vacuo from a water bath at 45°C. The crude product was recrystallized from 15% EtOAc/Skelly B to give 0.23 g (70%) of the urethane, mp 124.0-125.0°C; IR (KBr) cm^{-1} 3320, 1740, 1590, 1525, 1300, 1210, 1120, 1050, 830, 740, 690; ^1H NMR ($CDCl_3$) δ 5.23 (s, 2H, CH_2O), 7.05 (bs, 1H, NH), 7.22-7.83 (m, 15H, phenyl and vinyl).

Anal. Calcd for $C_{22}H_{18}ClNO_4S$: C, 61.75; H, 4.24; N, 3.27. Found: C, 61.70; H, 4.09; N, 3.21.

EXAMPLE 47

2-(Phenylsulfonyl)-2-propenyl chloroformate. Using the procedure from Example 5, 2-(phenylsulfonyl)-2-propenyl alcohol can be readily converted to 2-(phenylsulfonyl)-2-propenyl chloroformate.

EXAMPLE 48

2-Carbethoxy-2-propenyl chloroformate. Using the procedure from Example 5, 2-carbethoxy-2-propenyl alcohol can be readily converted to 2-carbethoxy-2-propenyl chloroformate.

EXAMPLE 49

2-(Phenylsulfonyl)-2-propenyl chloroformate. Using the procedure from Example 5,2-(phenylsulfonyl)-2-propenyl alcohol can be readily converted to 2-(phenylsulfonyl)-2-propenyl chloroformate.

EXAMPLE 50

(E)-3-Phenyl-2-(phenylsulfonyl)-2-propenyl chloroformate. Using the procedure from Example 5, (E)-3-phenyl-2-(phenylsulfonyl)-2-propenyl alcohol can be readily converted to (E)-3-phenyl-2-(phenylsulfonyl)-2-propenyl chloroformate.

EXAMPLE 51

(Z)-3-Phenyl-2-(phenylsulfonyl)-2-propenyl chloroformate. Using the procedure from Example 5, (Z)-3-phenyl-2-(phenylsulfonyl)-2-propenyl alcohol can be readily converted to (Z)-3-phenyl-2-(phenylsulfonyl)-2-propenyl chloroformate.

EXAMPLE 52

3,3-Diphenyl-2-(phenylsulfonyl)-2-propenyl chloroformate. Using the procedure from Example 5, 3,3-diphenyl-2-(phenylsulfonyl)-2-propenyl alcohol can be readily converted to 3,3-diphenyl-2-(phenylsulfonyl)-2-propenyl chloroformate.

EXAMPLE 53

Benzothiophenesulfone-2-methanol

Oxidation of 0.5 g of benzothiophene-2-methanol [F.F. Blicke and D.G. Sheets, J. Am. Chem. Soc., 71, 2856 (1949)] with 2 eqs. of m-chloroperbenzoic acid in 20 mL methylene dichloride gave in 51% yield the sulfone alcohol, mp 112-113°C.

Anal. Calcd for $C_9H_8O_3S$: C, 55.10; H, 4.08. Found: C, 54:81; H, 4.10.

The corresponding urethane derived from p-chlorophenyl isocyanate had mp 154-56°C.

Anal. Calcd for $C_{16}H_{12}ClNO_4S$: C, 54.94; H, 3.43; N, 4.01; Cl, 10.16. Found: C, 54.87; H, 3.48; N, 3.94; Cl, 10.35.

EXAMPLE 54

Similarly, by using the procedures described herein and the appropriate starting materials, the following compounds are prepared:

3,3-dimethyl-2-(phenylsulfonyl)-2-propenyl alcohol

3,3-dimethyl-2-(phenylsulfonyl)-2-propenyl chloroformate

Benzothiophenesulfone-2-methyl chloroformate.

EXAMPLE 55

(Z)-3-phenyl-2-thiomethyl-2-propenyl alcohol

Using the procedure of Example 44 and substituting cis-methyl-B-stryl sulfide for cis-phenyl B-stryl sulfide, the above compound can be prepared.

EXAMPLE 56

Z-3-phenyl-2-(methysulfonyl)-2-propenyl alcohol

Using the procedure of Example 45 and substituting the Z-3-phenyl-2-thiomethyl-2-propenyl alcohol for (Z)-3-phenyl-2-thiophenyl-2-propenyl alcohol, the above compound can be prepared.

EXAMPLE 57

Z-3-phenyl-2-(methylsulfonyl)-2-propenyl chloroformate. The product of Example 56 is reacted with phosgene in accordance with the procedure described in Example 25 to prepare the above product.

EXAMPLE 58

The products of Examples 25,26,28, and 29, 30,37, 42, 46, 47, 48, 49, 50, 51, 52, 54 and 58 can react with a carboxy protected amino acid, such as L-Phe-OH, in accordance with the procedure described hereinabove in Example 17 to prepare the corresponding protected amino acid. These amino acids are in turn hydrolyzed with acid, e.g., triflouoracetic acid, and then reacted with cyanuric fluoride as described herein to prepare the corresponding protected amino acid fluoride. For example, when the product of Example 57 is reacted with a carboxy protected amino acid, such as L-Phe-OH in accordance with the procedure described in Example 17, the product obtained is Mspoc-L-Phe. This product is then reacted with cyanuric fluoride in accordance with the procedure described herein to provide Mspoc-L-Phe-F.

EXAMPLE 59

USE OF FMOC-AA-F IN PEPTIDE SYNTHESIS

I General Method for Executing Rapid FMOC/4-AMP or FMOC/TAEA Peptide Synthesis. Five millimeters of a 0.1 M solution of the C-terminal starting amino acid ester, 5 mL of $CHCl_3$ containing 0.75 mmol of the appropriate FMOC-$\overset{X}{\underset{1}{AA}}$-Y, (wherein Y is halo, i.e., chloro or fluoro) and 5 mL of 5% Na_2CO_3 solution were combined and the two phase mixture stirred vigorously for 10 minutes. The organic phase was separated and treated with 5 mL of 4-AMP or TAEA. After 30 minutes 40 mL of $CHCl_3$ was added and the organic phase washed with five 25-mL portions of 10% phosphate buffer of pH 5.5. Alternatively two 25-mL portions of saturated NaCl solution was used to remove excess 4-AMP or TAEA prior to extractions with the phosphate buffer. The organic phase was concentrated in vacuo to a volume of 5 mL on a rotary evaporator and the resulting $CHCl_3$ solution used analogously for treatment with the next FMOC amino acid halide.

II Preparation of FMOC-Val-Asp(CMe_3)-Val-Leu-Leu-Ser$(CMe)_3$-Tyr(CMe_3)-$OCMe_3$. The general procedure described above was followed beginning with 330 mg (1 mmol) of H-Tyr-(CMe_3)-$OCMe_3$ HCl, 462 mg (1.2 mmol) of FMOC-Ser-(CMe_3)-F, 10 mL of methylene dichloride and 10 mL of 5% sodium carbonate solution. The mixture was stirred for 15 minutes, the organic layer separated and treated with 7.5 mL of TAEA for 30 minutes. The solution was washed with three 10-mL portions of satd sodium chloride solution and three 15-mL portions of phosphate buffer (pH 5.5). The organic layer was then treated in the same way in sequence with the remaining FMOC amino acid halides (FMOC-Leu-Cl twice, FMOC-Val-Cl,

FMOC-Asp(CMe_3)-F and FMOC-Val-Cl a second time). Evaporation of the organic layer was followed by column chromatorgraphy to give the protected heptapeptide, mp 249-251°C, in 50-60% yield MSFAB: 1255 (M+1); calculated 1253.8 (M).

EXAMPLE 60

Coupling of FMOC-Phe-F with H-Leu-OMe. Five millimeters of a 0.1 M solution of H-Leu-OMe in $CHCl_3$ was treated with 5 mL of $CHCl_3$ containing 0.6 mmol of FMOC-Phe-F (mp 118-120°) and 5 mL of 5% Na_2CO_3 solution. The two-phase mixture was stirred vigorously for 10 minutes, the layers separated and the organic layer dried over $MgSO_4$ and evaporated with a rotary evaporator. Without purification the crude residue (90%) was examined by HPLC analysis which showed no evidence for the DL-diastereomer (0.1%). The two diasteromers are readily separated with a mobile phase consisting of 1% isopropyl alcohol in hexane (retention times: LL, 13.26 minutes; DL-17.13 minutes at flow rate 1.5 mL/minutes) using a Waters Radial Pak normal silica gel column 10 u, Z-module fitting.

EXAMPLE 61

CBZ-alanine Fluoride

CBZ-Ala (1 mmole) in dry CH_2Cl_2 is kept under nitrogen and treated with cyannuric fluoride (8 mmol) and pyridine (1 mmole) to form the corresponding CBZ-Ala-F.

EXAMPLE 62

N-formyl-O-(t-butyl) serine Fluoride

N-formyl serine (1 mmole) in dry CH_2Cl_2 under nitrogen is treated with cyannuric fluoride (8 mmol) and pyridine (1 mmol) to form the above-identified product.

EXAMPLE 63

2-(t-butylsulfonyl)-2-propenyloxycarbonyl-l-phenylalanine Fluoride

A. t-Butyl Allyl Sulfide. To a solution of 350 mL of anhydrous ethanol maintained under nitrogen was slowly added 22.99 g (1 mol) of sodium spheres. The sodium dissolved within 90 minutes, and to the resulting sodium ethoxide solution was added 90.19 g (1 mol) of t-butyl mercaptan with mechanical stirring. Allyl bromide (120.98 g; 1 mol) was then added dropwise to the mechanically stirred sodium t-butyl thiolate solution. After the addition was complete, the mixture was refluxed for 10 minutes, the solution allowed to cool, the precipitated sodium bromide filtered, and the ethanol removed by distillation at atmospheric pressure. The residue was diluted with 200 mL of water, and the layers separated. The aqueous layer was extracted with five 40-mL portions of ether. The combined organic layers were extracted with 150 mL of water, the organic layer dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give a yellow liquid. Distillation through a 0.8 x 15-cm fractionating column gave 56.16 g (45%) of the sulfide as a colorless liquid, bp 139-141°C.

B. 1,3-Dibromo-2-(t-butylsulfonyl) Propane. To a stirred solution of 17.37 g (0.13 mol) of t-butyl allyl sulfide in 133 mL of CCl_4 at -24°C (CCl_4/dry ice) was added dropwise a solution of 21.33 g (0.13 mol) of Br_2 in 67 mL of CCl_4. A yellow solid precipitated during the addition. The mixture was warmed to room temperature and stirred for 10 minutes following complete solution of the yellow solid. The resulting solution was poured into a mixture of 55.50 g (0.27 mol) of 85% m-chloroperbenzoic acid in 490 mL of CH_2Cl_2 kept at -24°C, and the mixture stirred for 30 minutes at this temperature. The cooling bath was then removed and the mixture stirred at room temperature overnight. The precipitated m-chlorobenzoic acid was filtered and the filtrate washed with three 200-mL portions of saturated $NaHCO_3$, followed by 200 mL of water. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was recrystallized from 20% EtOAc/Skelly B to give 32.02 g (75%) of the dibromide, mp 139-140°C.

C. 2-(t-Butylsulfonyl)-2-propenyl Bromide. A mixture of 16.78 g (0.052 mol) of 1,3-dibromo-2-(t-butylsulfonyl) propane and 14 mL (0.12 mol) of 2,6-lutidine in 55 mL of CH_2Cl_2 was refluxed for 75 minutes. The solution was allowed to cool to room temperature and extracted with three 80-mL portions of 5% HCl followed by 80 mL of water. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C to give 11.46 g (91%) of the allyl bromide as a white solid, mp 40.5-42.0°C, which was used without further purification.

D. 2-(t-Butylsulfonyl-2-propenyl Alcohol. A mixture of 8.55 g (35.3 mmol) of 2- (t-butylsulfonyl)-2-propenyl bromide and 5.31 g (78.1 mmol) of sodium formate in 150 ml of methanol was refluxed overnight. The solution was allowed to

cool and concentrated to 50 mL with the aid of a water aspirator, resulting in the precipitation of excess sodium formate. The residue was diluted with 150 mL of water and extracted with five 50-mL portions of CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The crude product was recrystallized from 15% EtOAc/Skelly F to give 4.30 g (68%) of the alcohol as a colorless solid, mp 53.5-54.5°C.

E. 2-(t-Butylsulfonyl)-2-propenyl Chloroformate. To a solution of 6.67 g (37.4 mmol) of 2-(t-butylsulfonyl)-2-propenyl alcohol in 27 mL of dry THF at 0°C was added in one portion 27 mL of phosgene. The solution was stirred for 1 hour at 0°C and allowed to stand at room temperature overnight. Excess phosgene and solvent were removed under reduced pressure. The crude product was recrystallized from 25% ether/Skelly B to give 8.23 g (91%) of the chloroformate as a colorless solid, mp 56.5-57.7.

F. 2-(t-Butylsulfonyl)-2-propenyloxycarbonyl-L-phenylalanine. A solution of 4.57 g (19.0 mmol) of 2- (t-butylsulfonyl)-2-propenyl chloroformate and 5.64 g (18.6 mmol) of t-butyl L-phenylalaninate hydrophosphite in 90 mL of CH_2Cl_2 was stirred in the presence of 165 mL of 5% $NaHCO_3$ at room temperature for 2 hours. The aqueous phase was separated, and the organic phase washed with three 75-mL portions of 5% HCl. The organic phase was dried over $MgSO_4$, filtered, and the solvent removed in vacuo from a water bath at 45°C. The resulting oil was dissolved in 36 mL of 50% CH_2Cl_2/trifluoroacetic acid, and the solution stirred at room temperature for two hours. Excess trifluoracetic acid and solvent were removed in vacuo from a water bath at 150°C. The resulting oil was crystallized from ether/Skelly F to give approximately 6.25g (91%) of the colorless acid, mp 88.0-89.5°C.

G. 2-(t-butylsulfonyl)-2propenyloxycarbonyl-L-phenyl-alanine fluoride. The product formed hereinabove in Example 17F (1 mmol) is placed in dry CH_2Cl_2 under nitrogen and treated with cyannuric fluoride (8 mmol) and pyridine (1 mmol) to form the above-identified product.

EXAMPLE 64

BOC-Phe-F.

BOC-Phe (1 mmole) in dry CH_2Cl_2 under nitrogen is treated with cyannuric fluoride (8 mmol) and pyridine (1 mmole) to form the above-identified product.

EXAMPLE 65

Coupling of Bspoc-Phe-F with H-Leu-OMe

Five millimeter of 0.1 M solution of H-Leu-OMe in $CHCl_3$ was treated with 5 ml of $CHCl_3$ containing 0.6 mmol of Bspoc-phe-F and 5 ml of 5% Na_2CO_3 solution. The two phase mixture is stirred vigorously for 10 minutes, the layers separated and the organic layer dried over $MgSO_4$ and evaporated to afford the coupled depeptide.

Bspoc-Phe-Leu-OMe

EXAMPLE 66

BIMOC-Phe-F

A. 1-Bromo-2-bromomethylnaphthalene. To a solution of 103 g of 1-bromo-2-methylnaphthalene (bp 98-120°/0.5 mm, prepared in 90% yield by the method of Adams and Binder in JACS, 63, 2771(1941) and 82 g or N-bromosuccinimide in 1030 mL of CCl_4 was added 0.54 g of dibenzoyl peroxide. The reaction mixture was refluxed with stirring for 3 h. After another 0.54g of dibenzoyl peroxide was added, the mixture was refluxed for 3 hours. The solution was allowed to stand

at room temperature overnight, and the resulting suspension was brought to the boiling point and filtered while hot. Concentration of the filtrate gave 139 g (99.4%) of the bromide as a light yellow solid, mp 104-107°C, which was recrystallized from CCl_4 to give 128 g (91.6%) of pure 1-bromo-2-bromomethylnaphthalene as colorless crystals, mp 106.5-107.5°C.

B. Diethyl 2-(1-Bromo-2-naphthylmethyl)malonate. To a solution of NaOEt prepared from 9.66 g of sodium and 210 mL of dry EtOH was added 63.76 mL of diethyl malonate and the reaction mixture refluxed for 2 h. To the resulting yellow solution was added in small portions 126 g of 1-bromo-2-bromomethylnaphthalene and the reaction mixture refluxed for 16 h. Distillation of ethanol from an oil bath (100-110°C) through a simple Claisen head gave a yellow suspension, to which was added 350 mL of CH_2Cl_2 and 350 mL of H_2O. The aqueous layer was extracted twice with 120-mL portions of CH_2Cl_2, and the combined organic layer was washed three times with 100-mL portion of H_2O, dried ($MgSO_4$) and evaporated to give 152 g (95.6%) of the ester as a yellow solid, mp 60-65°C. Recrystallization from acetic acid gave 137 g (86.2%) of the pure diester as a colorless solid, mp 77-79°C.

C. β-(1-Bromo-2-naphthyl)propionic Acid. To a solution of 160 g of the product formed in B hereinabove in 239 mL of methanol was added 538 mL of 5N NaOH solution. The mixture was refluxed for 135 min, and cooled by means of an ice bath to 0°C. To the reaction mixture was added 320 mL of ice water, and the resulting precipitate was collected by filtration and washed several times with small portions of ice water. To the ice-cold, stirred filtrate was added dropwise 5N HCl solution until the solution was weakly acidic. The precipitate was filtered and washed several

times with small portions of water. Drying gave 86 g (63.1%) of crude diacid as a light yellow solid, mp 154-157°C. Recrystallization from water gave 80 g (58.7%) of pure diacid as colorless crystals, mp 157-159°C. A suspension of 47.5 g of the crude malonic acid in 968 mL of 6N HCl solution was refluxed for 16 h. The mixture was cooled by means of an ice bath to 0°C, treated with 650 mL of CH_2Cl_2, and stirred for 15 min. The aqueous layer was extracted twice with 100-mL portions of water, dried ($MgSO_4$), and evaporated to give 30.3 g (74%) of yellow solid, mp 116-119°C, which was recrystallized from alcohol to give 28 g (68%) of the acid as colorless crystalls, mp 123-124°C.

D. β-(1-Bromo-2-naphthyl)propionyl chloride. To a solution of 40.16 g of the product formed in C hereinabove in 802 mL of CH_2Cl_2 was added 19.3 mL of thionyl chloride. The mixture was refluxed for 4 h, cooled to room temperature, and the solvent evaporated from a water bath (40-50°C) with a rotary evaporator (10 mm) to give a red-brown residue. In order to remove traces of thionyl chloride, small portions of CH_2Cl_2 were added and the solution reevaporated three times. Eventually the crude acid chloride was obtained as a red-brown oil. The crude product was used immediately for the next step without further purification.

E. 4-Bromobenz[f]indan-1-one. To an ice cold, stirred solution of the above crude β-(1-bromo-2-naphthyl)propionyl chloride in 802 mL of dry CH_2Cl_2 was added 26.11 g of anhydrous $AlCl_3$ carefully. The reaction mixture was refluxed for 2 h, cooled to room temperature and treated carefully while stirring with 900 mL of ice-water followed by 75 mL of conc. HCl. The brown precipitate was filtered and washed five times with small portions of CH_2Cl_2. The aqueous layer was separated and extracted twice with 100-mL portions of CH_2Cl_2. The combined CH_2Cl_2 extracts were washed three times with small portions of H_2O, dried ($MgSO_4$) and evaporated from a water bath (50-60°C) with a rotary evaporator (7 mm) to

give 30.1 g (80.1%) of the crude ketone as a yellow solid, mp 146-149°C, which could be recrystallized from acetic acid to give 28.5 g (76%) of the pure ketone as colorless crystals, mp 149-151°C.

F. Benz[f]indan-1-ol. To an ice-cold solution of 22 g of the product formed in E hereinabove in 150 mL of dry THF was added carefully in small portions 16.1 g of $LiAlH_4$. Subsequently, another 270 mL of dry THF was added to the suspension and the mixture was refluxed for 8 days. The reaction mixture was cooled to 0°C by means of an ice-bath and treated dropwise with 50 mL of ice-water followed by 1080 mL of 10% H_2SO_4 solution. The mixture was extracted three times with 100-mL portions of ether and the combined ether extracts washed three times with 150-mL portions of H_2O, dried ($MgSO_4$), and evaporated from a water bath (60-70°C) with a rotary evaporator (10 mm) to give 9.2 g (59.3%) of the crude alcohol as a light yellow solid, mp 135-139°C. Recrystallization from benzene (45 mL) gave 8.6 g (55.4%) of pure alcohol as colorless crystals, mp 139-141°C.

G. Benz[f]indene. A solution of 8.6 g of benz[f]indan-1-ol in 250 mL of 10% H_2SO_4 was refluxed for 24 h. After cooling to room temperature the reaction mixture was extracted with three 150-mL portions of a mixture of benzene and hexane (1:2). The extracts were washed three times with 100-mL portions of water, dried ($MgSO_4$), and evaporated to give 7.6 g (98%) of a colorless solid, mp 160-163°C. Recrystallization from 340 mL of 95% ethanol gave 6.6 g (85%) of the hydrocarbon as colorless crystals: mp 163-164°C.

H. Benz[f]indene-1-methanol. A 1.0 M solution of n-butyllithium (44 mL, 44 mmoles) was added dropwise under a nitrogen atmosphere to a stirred solution of 5 g (30.12 mmoles) of benz[f]indene in 140 mL of anhydrous ether and 20

mL of anhydrous THF cooled by means of a Dry Ice-acetone bath to -70°C. The temperature of the reaction mixture was not allowed to exceed -50°C. Benzindenyl lithium soon started to precipitate as small red crystals. After completion of the addition (about 2 hours), the reaction mixture was stirred at -70°C for another 45 minutes before introduction of formaldehyde. Paraformaldehyde, 13.5 g. dried overnight in vacuum over phosphorus pentoxide, was stirred and heated in a dry flask placed in an oil bath at 175-195°C. The formaldehyde gas was led through a 7-mm glass tube into the benz[f]indenyl lithium solution (held below -50°C) by a stream of dry nitrogen. The temperature was not allowed to exceed -50°C. After completion of the addition, 280 mL of 10% HCl solution was slowly poured into the stirred reaction mixture. The mixture was stirred for 15 minutes at room temperature. After the ether layer was separated, the aqueous solution was extracted twice with 50-mL portions of ether and the combined ether solution was washed with small portions of water until neutral. The ether solution was dried ($MgSO_4$) and evaporated to give a light brown oil (6 g). Storage in the freezer gave a soft yellow solid which was purified by chromatography (100 g of silica gel, 1:1 ethyl acetate/hexane) to give 3.5 g (59%) of the alcohol as a yellow solid. The NMR showed the solid to be a mixture of benz[f]indene-1-methanol (95%) and benz[f]indene-3-methanol. Several recrystallizations from ligroin (bp 88-89°C) gave 3.0 g (50.6%) of pure, colorless benz[f]indene-1-methanol: mp 115-116°C.

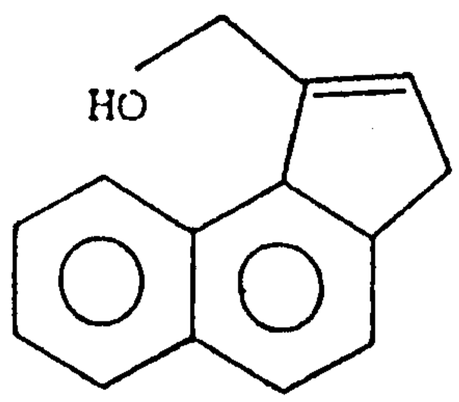
M. The BIMOC-Phe (1 mmole) formed in L hereinabove in dry CH_2Cl is kept under nitrogen and treated with Cyannuric fluoride (8 mmol) and pyridine (1 mmol) to form the corresponding BIMOC-Phe-F.

EXAMPLE 67

Benz [e] indene-3-methyloxycarbonyl-phenylalanine acid fluoride

A. Benz [e]-indene-3-methanol

1-Keto-4,5-benzindane-3-carboxylic acid, [which was prepared in accordance with the procedure described by T.N. Poltabiraman and W.B. Lawson in J. Biol. Chem, 247, 302a (1972)] is reduced by sodium borohydride to form the corresponding 3-carboxylic acid -1-ol. Dehydration with sulfuric acid give the corresponding unsaturated acid. The unsaturated acid was then reduced by $LiAlH_4$ to form the 3-methanol derivative:



B. N-[Benz[e] indene-3-methyloxycarbonyl] phenylalanine

This product is prepared from the product of A hereinabove by following the procedure described in Ex. 20 I-L hereinabove.

C. N-[Benz[e] indene 1-methoxycarbonyl] phenylalanine acid fluoride

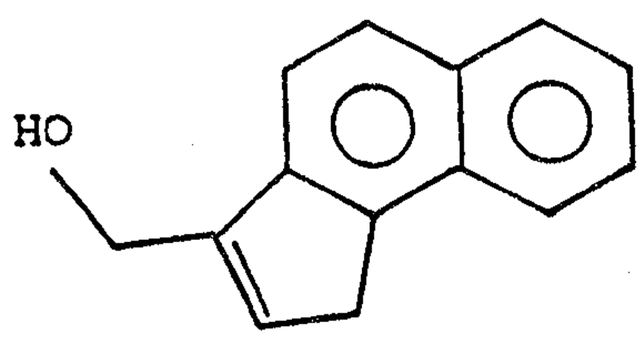
The product B formed hereinabove (1 mmol) in dry Ch_2Cl_2 is kept under nitrogen and treated with cyannuric fluoride (8 mmol) and pyridine (1 mmol) to form the above product.

EXAMPLE 68

Benz [e] indene-1-methyloxy carbonylphenylalanine acid fluoride

A. Benz [e]-indene-1-methanol

3-Keto-4,5-benzindan-1-carboxylic acid, which was prepared in accordance with the procedure described by T.N. Paltabiraman and W.B. Lawson, in J. Biol. Chem. 242, 3029(1941) is reduced to the corresponding 1-carboxylic acid-3-ol- by $NaBH_4$. Dehydration with H_2SO_4 gave the unsaturated acid which was then reduced to benz (e)-indene-1-methanol by means of $LiAlH_4$.



B. N-(Benz[e]indene-1-methyloxycarbonyl) phenylalanine

This product is prepared from the product of A hereinabove by following the procedure described in Ex. 20 I-L hereinabove

C. N-[Benz(e) indene-1-methoxyoxy carbonyl] phenylalanine acid fluoride.

The product B formed hereinabove (1 mmol) in dry CH_2Cl_2 is kept under nitrogen and treated with cyannuric fluoride (8 mmol) and pyridine (1 mmol) to form the above product.

EXAMPLE 69

N-[Benz(e) indene 1-methoxy carbonyl]phenylalanine acid fluoride and N[Benz(e) indene 3-methoxy carbonyl] phenylalanine acid fluoride

2(β)-naphthylpropronic acid is treated with thionyl chloride and cyclized in the presence of aluminum chloride to

form the corresponding ketone. Reduction of the ketone with sodium borohydride, followed by dehydration with sulfuric acid gives the benz[e] indene. Formylation with ethyl formate and sodium hydride followed by treatment with sodium borohydride gave a mixture of the Benz[e] indene-3-methanol and Benz[e]-indene-1-methanol. The two alcohols are separated by chromatography.

Following the procedures in Examples 22 B-C and 23 B-C, the above-identified products are prepared.

It is to be noted from the preparations hereinabove that Benz[e]indene 1-methoxycarbonyl and the benz[e] indene-3-methoxy carbonyl can be used to protect the N α -amino group of an amino acid.

These groups are formed from the corresponding alcohol, as described in Ex. 21-23. The alcohols are then used as starting materials to form the corresponding chloroformates and azidoformates, as described in Ex. 20. The azidoformate can then be reacted with an amino acid in accordance with the procedure described in Ex. 20-L to form the corresponding N α -protected amino acid. This then is treated with cyannuric fluoride to form the corresponding amino acid fluoride.

The above preferred embodiments and examples are given to illustrate the scope and spirit of the present invention. These embodiments and examples will make apparent, to those skilled in the art, other embodiments and examples. These other embodiments and examples are within the contemplation of the present invention. Therefore, the present invention should be limited only by the appended claims.

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

1. A compound of the formula:

or the acid fluoride salts thereof wherein

BLK is an N-amino protecting group;

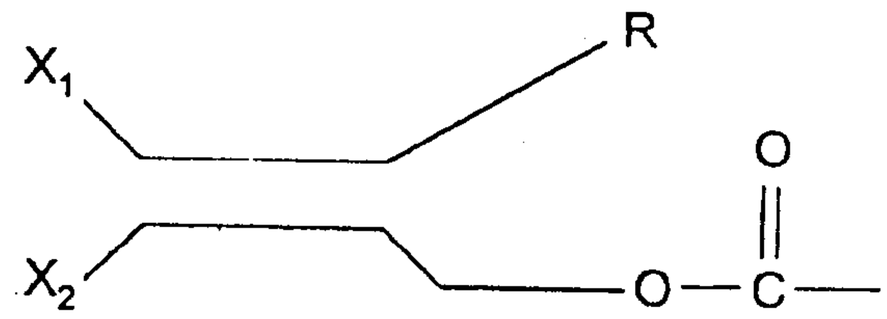
AA is an α-amino acid residue;

X is - or a protecting group on the side chain of the amino acid, wherein - signifies the side chain of the amino acid has no protecting group thereon.

2. The D or L stereoisomer of the compound of Claim 1 or mixtures thereof.

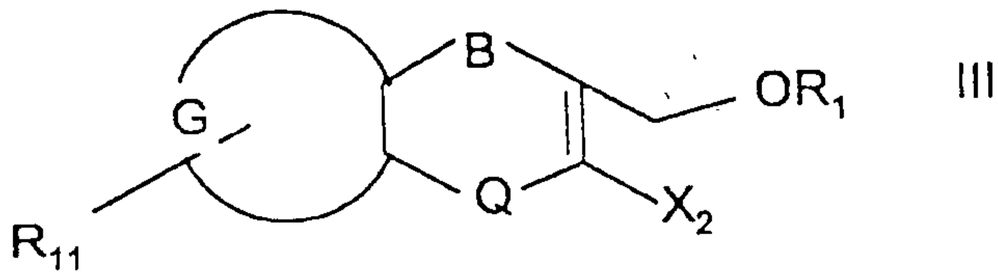
3. The compound according to claim 1 or 2, wherein BLK is a base labile N-α-amino acid protecting group, an acid labile N-α-amino acid protecting group, a nucleophile labile N-α-amino acid protecting group, or a N-α-amino acid protecting group removable by hydrogenation.

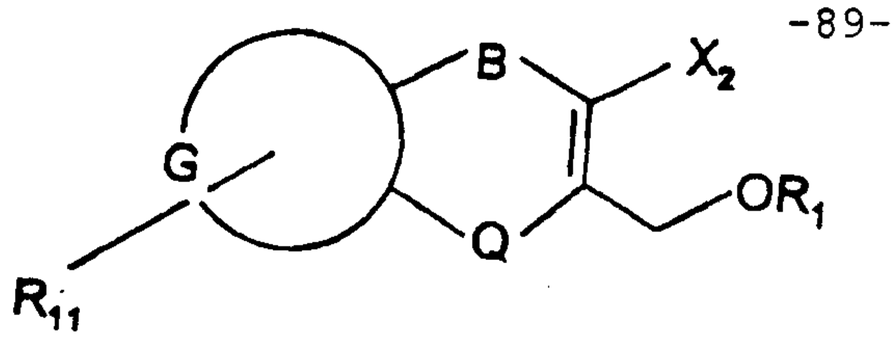
4. The compound according to any one of the claims 1-3, wherein BLK has the formula:



wherein R is an electron withdrawing group,

X_1 and X_2 are independently H, lower alkyl, aryl, aryl lower alkyl, or a solid support or R and X_1 may be taken together with the carbon to which they are attached to form a ring so that BLK has the formula





IIIa

wherein B is a chemical bond, CR_8R_9,

O
‖
C,

SO_2, SO, ŔP(O), or S, Ŕ is lower alkyl or OR_{10}, R_{10} is lower alkyl, R_8, R_9, R_{12}, and R_{13} are independently hydrogen or lower alkyl, G is a mono or bicyclic fused ring system containing 5 to 10 carbon atoms and optionally up to 2 heteroatoms, wherein the heteroatoms are O, S or N, Q is a chemical bond,
CR_{12}, R_{13}, SO_2, SO, ŔP(O) or S and R_{11} is hydrogen or lower alkyl; R_1 is COZ, and Z is a leaving group.

5. The compound according to claim 4, wherein R is SO_2R_2, SOR_2, $COOR_2$, COR_2, CHO, $CONR_2$ R_3, CN, CF_3, NO_2, aryl, 2-pyridyl or 4-pyridyl, wherein R_2 and R_3 are independently lower alkyl, aryl, aryl lower alkyl, or polystyrene and the alkyl or aryl groups are unsubstituted or mono or disubstituted with halide, lower alkyl-, aryl- or aryl lower alkylsulfonyl, lower alkyl-, aryl- or aryl lower alkylsulfinyl, lower alkyl-, aryl- or aryl lower alkylcarboxy, lower alkyl-, aryl- or aryl lower alkylcarbonyl, CHO, CN, CF_3, or NO_2.

6. The compound according to claim 4 wherein R is SO_2R_2, SOR_2, $COOR_2$, COR_2 $CONR_2R_3$, aryl, 2-pyridyl, or 4-pyridyl, wherein R_2 and R_3 are independently lower-alkyl having from 1 to 6 carbon atoms, aryl, aryl lower-alkyl, hetero-aryl or a solid support and the alkyl, aryl or hetero-aryl groups are unsubstituted, or mono- or di-substituted with halides, lower alkyl-, aryl-, hetero-aryl- or aryl lower alkylsulfonyl, lower alkyl-, aryl-, hetero-aryl- or aryl lower alkylsulfinyl, lower alkyl-, aryl-, hetero-aryl- or aryl lower alkylcarboxy, lower alkyl-, aryl-, hetero-aryl- or aryl lower alkylcarbonyl, CHO, CN, CF_3 or NO_2.

7. The compound according to any one of claims 4-

6, wherein R is $SO_2C(CH_3)_3$, $SOC(CH_3)_3$, $SO_2C_6H_5$, SO_2 CH_3, COO Et, 2-pyridyl, or 4-pyridyl or R and X_1 when taken together with the carbon to which they are attached are $SO_2C_6H_4$ or $C_5H_5NCH_2$.

8. The compound according to any one of claims 1-7, wherein AA is glycine, alanine, leucine, isoleucine, proline, hydroxyproline, phenylalanine, tyrosine, methionine, norleucine, serine, threonine, cystine, cysteine, aspartic acid, glutamic acid, asparagine, glutamine, lysine, arginine, hydroxylysine, ornithine, histidine or tryptophan and X is -.

9. The compound according to any one of claims 1-7, wherein

X							
\							
AA is							
DMB	TMB	DMB	TMB				
\	\	\	\				
Gln,	Gln,	Asn,	Asn,				
Mtr	Pmc	FMOC					
\	\	\					
Arg,	Arg,	His,					
BOC	Trt	Bom	t-Bu	Ada	Bz	TMSE	
\	\	\	\	\	\	\	
His,	His,	His,	Ser,	Ser,	Ser,	Ser,	
Cp	Ch	t-Bu	Ada	Bz	TMSE		
\	\	\	\	\	\		
Ser,	Ser,	Thr,	Thr,	Thr,	Thr,		
Cp	Ch	t-Bu	Ada	TMSE			
\	\	\	\	\			
Thr,	Thr,	Tyr,	Tyr,	Tyr,			
Bz	Cp	Ch	t-Bu	Trt			
\	\	\	\	\			
Tyr,	Tyr,	Tyr,	Cys,	Cys,			

Acm S-t-Bu
\ \
Cys, Cys,

Bz t-Bu Cp Ch Ada TMSE
\ \ \ \ \ \
Asp, Asp, Asp, Asp, Asp, Asp,

Bz t-Bu Cp Ch Ada TMSE
\ \ \ \ \ \
Glu, Glu, Glu, Glu, Glu, Glu,

Cbz Adoc CHO TFA
\ \ \ \
Lys, Lys, Lys, or Lys.

10. The compound according to any one of claims 1-7, wherein the side chain of the AA is hydroxy lower alkyl, carboxy lower alkyl, mercapto lower alkyl, hydroxy aryl or hydroxy benzyl and X is t-butyl.

11. The compound according to any one of claims 1-3 and 8-10, wherein BLK is Cbz, FMOC, BOC, CLIMOC, BIMOC, Dbd-TMOC, Aoc, Adoc, $ArSO_2$, Bpoc, Ddz, HCO, TFA, Teoc, benz [e]indene-1-methoxycarbonyl, Benz-[e]-indene-3-methoxy-carbonyl, Bspoc, Bsmoc, or Mspoc.

12. The compound according to claim 1 which is

t-Bu
\
BLK-Ser-F

t-Bu
\
BLK-Thr-F,

t-Bu
\
BLK-Tyr-F,

t-Bu
\
BLK-Asp-F,

BOC
\
BLK-Trp-F

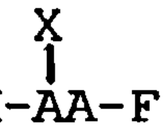
BLK-Lys-F-,

$C(C_6H_5)_3$
\
BLK-Cys-F, BLK-Gly-F, BLK-Ala-F, BLK-Val-F,

BLK-Leu-F, BLK-Ile-F or BLK-Phe-F wherein

BLK is Bspoc, FMOC, Bsmoc, or Mspoc.

13. A process for forming a peptide bond between a first amino acid having a free amino group and a second amino acid or between a peptide having a free amino group and a second amino acid which comprises reacting the first amino acid or peptide with an amino acid fluoride of the formula



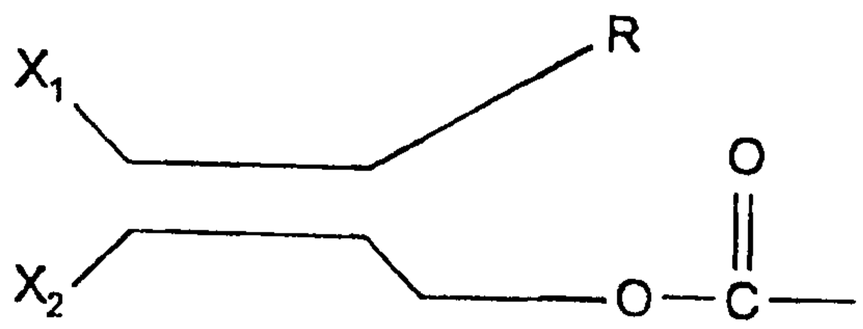
or the acid fluoride salts thereof wherein,

BLK is an N-amino protecting group,

AA is an α-amino acid residue having a side chain and X is - or a protecting group on the side chain of the amino acid wherein - signifies that the side chain of the amino acid has no protecting group thereon.

14. The process according to claim 13, wherein BLK is a base labile N-α amino acid protecting group, an acid labile N-α amino acid protecting group, a nucleophile labile N-α amino acid protecting group or N-α amino acid protecting group removable by hydrogenation.

15. The process according to claim 13 or 14, wherein BLK has the formula:



wherein R is an electron withdrawing group,

X_1 and X_2 are independently H, lower alkyl, aryl, aryl lower alkyl or a solid support or R and X_1 taken together with the carbon to which they are attached form a ring containing from 4-15 carbon atoms and optionally containing

up to 2 heteroatoms, wherein the heteroatoms are O, S or N.

16. The process according to claim 15, wherein R is SO_2R_2, SOR_2, $COOR_2$, COR_2, CHO, $CONR_2R_3$, CN, CF_3, NO_2, aryl, 2-pyridyl or 4-pyridyl, wherein R_2 and R_3 are independently lower alkyl, aryl, aryl lower alkyl, or polystyrene and the alkyl or aryl groups are unsubstituted or mono or disubstituted with halide, lower alkyl-, aryl-, or aryl lower alkylsulfonyl, lower alkyl-, aryl- or aryl lower alkylsulfinyl, lower alkyl-, aryl- or aryl lower alkylcarboxy, lower alkyl-, aryl- or aryl lower alkylcarbonyl, CHO, CN, CF_3 or NO_2.

17. The process according to claim 16, wherein R is SO_2R_2, SOR_2, $COOR_2$, COR_2, $CONR_2R_3$, aryl, 2-pyridyl, or 4-pyridyl wherein R_2 and R_3 are as defined in claim 16.

18. The process according to any one of claims 15-17, wherein R is $SO_2C(CH_3)_3$, $SOC(CH_3)_3$, $SO_2C_6H_5$, SO_2 CH_3, COO Et, 2-pyridyl, or 4-pyridyl or R and X_1 when taken together with the carbon to which they are attached are $SO_2C_6H_4$ or $C_5H_5NCH_2$.

19. The process according to any one of claims 13-17, wherein AA is glycine, alanine, leucine, isoleucine, proline, hydroxyproline, phenylalanine, tyrosine, methionine, norleucine, serine, threonine, cysteine, cystine, aspartic acid, glutamic acid, asparagine, glutamine, lysine, arginine, hydroxylysine, ornithine, histidine or tryptophan.

20. The process according to any one of claims 13-19, wherein

X
\
AA is

DMB TMB DMB TMB
\ \ \ \
Gln, Gln, Asn, Asn,

Mtr Pmc FMOC
\ \ \
Arg, Arg, His,

BOC Trt Bom t-Bu Ada Bz TMSE
\ \ \ \ \ \ \
His, His, His, Ser, Ser, Ser, Ser,

Cp Ch t-Bu Ada Bz TMSE
\ \ \ \ \ \
Ser, Ser, Thr, Thr, Thr, Thr,

Cp Ch t-Bu Ada TMSE
\ \ \ \ \
Thr, Thr, Tyr, Tyr, Tyr,

Bz Cp Ch t-Bu Trt
\ \ \ \ \
Tyr, Tyr, Tyr, Cys, Cys,

Acm S-t-Bu
\ \
Cys, Cys,

Bz t-Bu Cp Ch Ada TMSE
\ \ \ \ \ \
Asp, Asp, Asp, Asp, Asp, Asp,

Bz t-Bu Cp Ch Ada TMSE
\ \ \ \ \ \
Glu, Glu, Glu, Glu, Glu, Glu,

Cbz Adoc CHO TFA
\ \ \ \
Lys, Lys, Lys, or Lys.

21. The process according to any one of claims 13-18, wherein the side chain of AA is hydroxyl lower alkyl, carboxyl lower alkyl, mercapto lower alkyl, hydroxyaryl or hydroxy carbonyl and X is t-butyl.

22. The process according to any one of claim 13-21, wherein the D or L stereoisomer or mixture of the amino acid fluoride is used.

23. The process according to claim 13, wherein BLK is Cbz, FMOC, BOC, CLIMOC, BIMOC, Dbd-TMOC, Aoc, Adoc, $ArSO_2$, Bpoc, Ddz, HCO, TFA, Teoc, benz[e]indene-1-methoxycarbonyl or benz[e]indene-3-methoxycarbonyl, Mspoc, Bspoc or Bsmoc.

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