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[54] Title: NEW CONJUGATES OF VINBLASTINE AND ITS DERIVATIVES, A PROCESS FOR THEIR PREPARATION AND PHARMACEUTICAL COMPOSITIONS CONTAINING THE SAME

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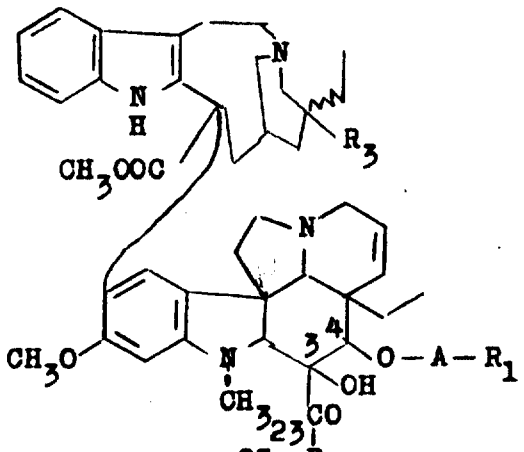
[57]

A B S T R A C T

New conjugates of vinblastine or of derivatives thereof are described. These conjugates correspond to the general formula



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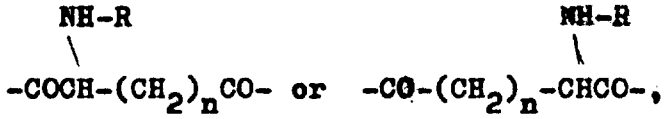
in which A represents an "arm" such as



n varying from 1 to 5 and R representing a hydrogen atom, an acyl group such as acetyl, trifluoroacetyl or carbobenzyloxy; R_1 represents an optionally modified protein radical, R_2 represents a methoxy group, an amino group or an alpha-aminoacide ester which is bonded by a bond of the amide type and in which the ester group contains 1 to 6 carbon atoms, and R_3 represents a hydrogen atom or a hydroxyl group, in each case in the two possible configurations.

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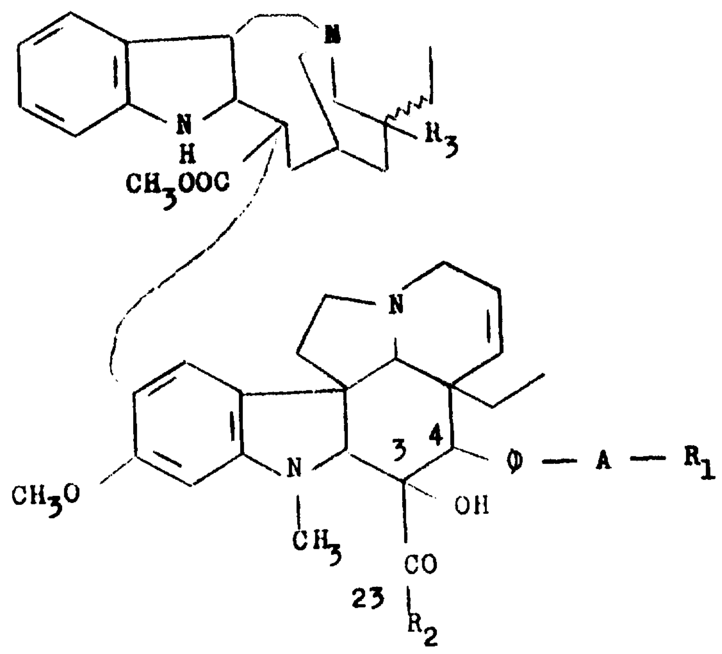


in which A represents an "arm" such as

$$-\,\underset{\displaystyle \overset{|}{\text{NH-R}}}{\text{COCH}}\text{-}(CH_2)_n CO\text{-} \quad \text{or} \quad \text{-}CO\text{-}(CH_2)_n\text{-}\underset{\displaystyle \overset{|}{\text{NH-R}}}{\text{CH}}CO\text{-},$$

n varying from 1 to 5 and R representing a hydrogen atom, an acyl group such as acetyl, trifluoroacetyl or carbobenzyloxy; R_1 represents an optionally modified protein radical, R_2 represents a methoxy group, an amino group or an alpha-aminoacide ester which is bonded by a bond of the amide type and in which the ester group contains 1 to 6 carbon atoms, and R_3 represents a hydrogen atom or a hydroxyl group, in each case in the two possible configurations.

NEW CONJUGATES OF VINBLASTINE AND ITS DERIVATIVES, A PROCESS FOR THEIR PREPARATION AND PHARMACEUTICAL COMPOSITIONS CONTAINING THE SAME

The present invention relates to new conjugates of vinblastine and of some its known derivatives with proteins or fragments of proteins usable as anti-tumour agent. The invention also relates to a process for their preparation and to pharmaceutical compositions containing these conjugates.

Vinblastine and some of its derivatives, in particular vincristine or vindesine, have already been coupled to proteins, for example albumin or various immunoglobulins. Coupling products or compounds called "conjugates" result. The following literature references may be referred to in particular : J.D. Teale, Jacqueline M. Clough and V. Marks, Br.J.Clin.Pharmac. 4, 169-172, 1977; C.H.J. Ford, C.E. Newman, J.R. Johnson, C.S. Woodhouse, T.A. Reeder, G.F. Rowland, R.G. Simmonds, Br.J.Cancer 47, 35-42, 1983; M.J. Embleton, G.F. Rowland, R.G. Simmonds, E. Jacobs, C.H. Marsden, R.W. Baldwin, Br.J. Cancer 47, 43-39, 1983; J.R. Johnson, C.H.J. Ford, C.E. Newman, C.S. Woodhouse, G.F. Rowland, R.G. Simmonds, Br.J. Cancer 44, 472-475, 1981; Eli Lilly Eur. Pat. Applic.,

Publ. No. 56,322, 21.07.82; and R.A. Conrad, G.J. Cullinan, K. Gerzon, G.A. Poore, J. Med. Chem. 22, 391, 1979.

May also be mentioned the European Patent Application No. 84 870057.1 which contains a description of the coupling of bis-indole derivatives to a protein, a fragment of protein or an amine by means of an arm such as $-COCH_2-$, $-CO(CH_2)_n-CO$, a varying from 1 to 5, which may be substituted by an alkyl or amino group.

Coupling of these bis-indole derivatives has been effected not only with the aim of developing new immunological reagents, but in particular with the aim of preparing anti-tumour substances which are more active, more selective and less toxic.

In this last respect, numerous conjugates of proteins with other anti-tumor agents are currently being studied. This applies, in particular, to conjugates of antibodies and a fragment of ricin or conjugates of albumin and methotrexate (French Patent Application 2,437,213, C M Industries; and B.C.F. Chu, S.B. Howell, J. of Pharmacology and Exp. Therapeutics, 219 (2), 389-393, 1981).

Monoclonal antibodies, in particular those of human origin, coupled to known anti-tumour medicaments are more particularly the subject of various studies.

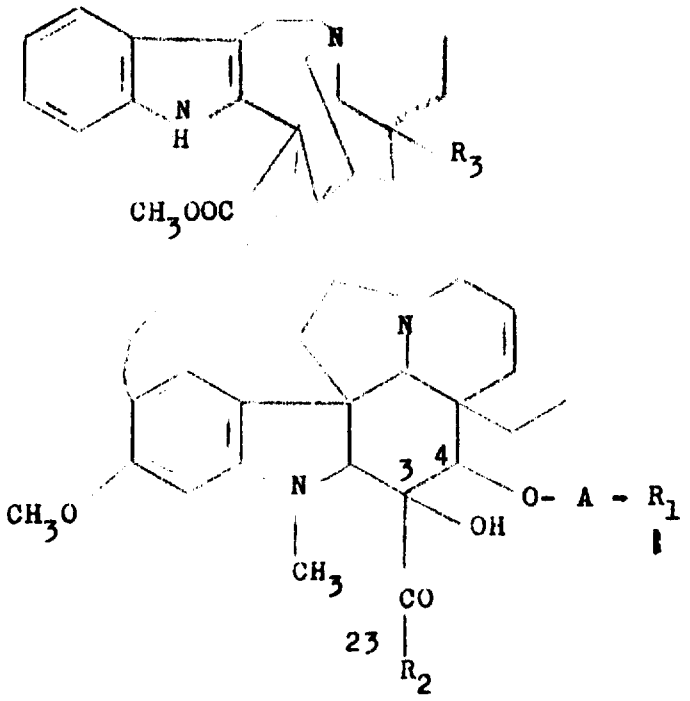
Finally, reference is made to the fact that the use and evaluation of 1:1 complexes of bis-indole alkaloids with tubulin have been described in Belgian Patent 854,053. In some cases, a lower toxicity and a more significant chemotherapeutic activity than from the corresponding free alkaloids may result.

The present invention is particularly directed to new products which consist of conjugates of vinblastine or of derivatives of vinblastine with proteins or fragments of proteins, characterised in that the coupling is effected through an ester group which derives from the hydroxyl group at the carbon 4 of the skeletons of vinblastine.

These conjugates according to the invention are characterised in that the protein or fragment of protein is coupled to the compound of the vinblastine type by means of an arm, by previous condensation of the derivative of vinblastine with a bifunctional organic derivative, optionally activated, according to a preferred embodiment.

The derivative of 4-O-diacetylvinblastine may be, for example, vindesine, 4-O deacetylvinblastine coupled at C-3 with an aminoacid, 4-O-deacetyl-deoxy-4'-vinblastine or 4-O-deacetyl-deoxy-4'-vinblastine coupled at C-3 with an aminoacid.

More particularly, the compounds according to the invention correspond to the following general formula I



in which A represents an "arm" such as

$$-CO-(CH_2)_n-\underset{\displaystyle NH-R}{\underset{|}{CH}}CO- \quad \text{or} \quad -COCH=\underset{\displaystyle NH-R}{\underset{|}{(CH_2)_n}}CO-$$

in which n varies from 1 to 5 and R represents a hydrogen atom, an acyl group such as acetyl, trifluoroacetyl or carbobenzyloxy, R_1 represents an optionally modified protein radical, R_2 represents a methoxy group, an amino group or an alpha-aminoacid ester radical which is bonded by a bond of the amide type and in which the ester group contains 1 to 6 carbon atoms, and R_3 represents a hydrogen atom or a hydroxyl group, in each case in the two possible configurations, as well as their addition salts with an inorganic or organic acid.

The therapeutic activity of the compounds according to the invention is higher than that of the compounds which have been described up to now and their toxicity is substantially lower.

The derivatives according to the present invention are obtained, in a first stage, by condensation of an anhydride, for example aspartic or glutamic anhydride or a higher homologue, optionally substituted on the amine group, onto the hydroxyl at C-4 of 4-O-deacetyl-vinblastine or of 4-O-deacetyldeoxyvinblastine or of one of

the derivatives of the 4-O-deacetyldeoxyvinblastine-3-carboxamide or 4-O-deacetyldeoxyvinblastine-3-carboxamide type.

The hemiaspartate or hemiglutamate or homologue derivatives thus obtained are then condensed with the protein or the fragment of protein in a solvent in which these compounds are soluble. For this purpose, a water/dioxane mixture at a suitable pH maintained by a buffer, for example a phosphate buffer, can be used. The condensation is confirmed by chromatography or electrophoresis. In the later case, radioactive (for example tritiated) vinblastine may be used in order to facilitate the characterisation.

From the chemical point of view, the condensation with the proteins can be explained by the production of covalent bonds resulting from the reaction of the amine groups of the protein lysine with the activated ester group derived from the optionally substituted hemiaspartate or hemiglutamate.

Bovine serum albumin, for example, contains 56 amine residues of lysine. The number of molecules of vinblastine per protein varies as a function of the operating conditions, but is

generally 1 and 34.

The activation can be effected in a conventional manner by treatment with an alkyl chloroformate, preferably ethyl or isobutyl chloroformate, in the presence of an amine base, such as N-methylpiperidine or N-methylmorpholine.

The condensation can be carried out in situ on the reaction mixture containing the activated anhydride. However, in most cases, the activated anhydride can also be isolated.

The conjugate obtained is isolated by means of the conventional methods used in chemistry or, in the case of protein conjugates, in biochemistry. The protein conjugate is accordingly precipitated out of the reaction mixture by addition of acetone and is then centrifuged off, rinsed, lyophilised and purified by gel filtration. If appropriate, the derivative thus obtained may be subjected to a conventional succinylation reaction, which enables the aggregation problems which characterise certain conjugates and non-conjugated proteins to be avoided.

The proteins which can advantageously be used are, in particular, bovine or human serum

albumin, fetuin or immunoglobulins, the latter being obtained, if appropriate, by the monoclonal antibody technique. In the latter case, the use of monoclonal antibodies of human origin which demonstrate a certain specificity towards human tumours has proved to be of particular interest.

The proteins used can also be treated in order to be selectively modified. These modifications enable protein conjugates to be obtained which, during use in therapy, are concentrated preferentially in certain tissues, for example in the liver. It is thus possible prior to the condensation of the derivative of the vinblastine with the protein, to galactosylate the latter. The galactosylation is carried out, for example, by applying the method described by G. Wilson in the Journal of Biochemistry, 253 (7) 2070-2072, 1978.

In vitro experiments carried out with the compounds according to the invention to demonstrate their anti-tumour activity indicate that the formation of the conjugate by a bond at C-4 may be more advantageous than if the bond is effected at C-3.

Alternatively, it is also possible to condense a compound of the $-A-R_1$ type, A and R_1 having the same meaning as indicated above, in a suitable solvent onto the hydroxyl at C-4 of 4-O-deacetylvinblastine or 4-O-deacetyldeoxy-vinblastine or of one of the derivatives of 4-O-deacetylvinblastine-3-carboxamide or 4-O-deacetyldeoxyvinblastine-3-carboxamide. The activation can advantageously be carried out by treatment with an alkyl chloroformate, such as ethyl or isobutyl chloroformate, in the presence of an amine base such as N-methylpiperidine, N-methylmorpholine or triethylamine. The following examples describe some conjugates according to the invention wherein the bis-indole derivatives are coupled at the position C-4 to a protein or a fragment of protein by means of an arm of the substituted hemiaspartate or the substituted hemiglutamate type.

The present invention also relates to industrial and, in particular, pharmaceutical uses of the new bis-indole compounds.

In fact, the compounds according to the invention have particularly useful anti-tumour properties which are capable of being used in human therapy.

In particular, these vinblastine derivatives can be used for the treatment of leukemies, gliomas, lymphosarcomas or other malignant tumours, including so-called "solid" tumours.

In human therapy, they are thus used for the treatment of Hodgkin's disease and for other tumours which may benefit from treatment with vinblastine, vincristine or vindesine.

For use in therapy, the compounds according to the invention, is appropriate in lyophilised form, are preferably administered parenterally, in solution in a pharmaceutically acceptable solvent.

Physiological water or other saline solutions which are buffered, for example with a phosphate, are suitable solvents.

The active substance is in general administered in a dosage which can vary from 50 mg to several grammes. The compounds according to the invention can also be used in combination with other anti-tumour agents.

Example 1

a) α and β isomers of 4-O deacetyl-4-O-L-N-acetyl-hemiaspartate vinblastine

250 mg of N-acetyl-L-aspartic anhydride are added to a solution of 700 mg (0.91 mmol) 4-O-deacetyl-

vinblastine in 20 ml anhydrous dichloromethane.

The solution is stirred during 20 hours at ambient temperature. After distillation under vacuum of the solvent, the obtained residue is purified by chromatography on silica (elution ether/methanol/NH_4OH 25% 60/49.75/0.25).

After trituration in a mixture of dichloromethane and petrol ether, 650 mg 4-O-deacetyl-4-O-L-N-acetylhemiaspartate vinblastine are obtained in the state of a white powder (Yield : 77%).

HPLC analysis of the product indicates the presence of both isomers and in a ratio of 85/15.

* IR spectrum (KBr): 3420, 2960, 2880, 1737, 1660, 1613, 1501, 1460, 1430 cm^{-1}.

* Mass spectrum: DCI (isobutane): 925 (M^{+}), 939 (M^{+} + 14), 857, 769, 693, 635.

b) <u>and isomers of 4-O-deacetyl-4-O-L-N-acetylhemiaspartate methyl ester vinblastine</u>.

A solution of 500 mg (0.540 mmoles) and isomers of 4-O-deacetyl-4-O-L-N-acetylhemiaspartate vinblastine in 10 ml absolute methanol saturated with dry HCl is stirred for 20 hours, at ambient temperature.

After distillation under vacuum of the solvent, the residue is recovered in a mixture of 15 ml distilled water and 15 ml dichloromethane. The mixture is rendered alkaline by NH_4OH addition. The aqueous phase is extracted by three portions of 20 ml dichloromethane. The organic extracts are combined, successively washed with 40 ml water and 40 ml of an aqueous solution saturated with NaCl, dried on magnesium sulfate and evaporated under reduced pressure. The residue is purified by chromatography on silica (elution by CH_2Cl_2:CH_3OH 95:5). Accordingly 410 mg and isomers of 4-O-deacetyl-4-O-L-N-acetylhemiaspartate methyl ester vinblastine are obtained, Yield: 81%.

Fractions resulting from chromatography including individual isomers are analyzed.

- Principal isomer

* Mass spectrum (DCI; isobutane): 939 (M^+), 940 ($M^+ + 1$), 953 ($M^+ + 14$)

* IR spectrum (KBr): 2950, 2880, 1740, 1675, 1615, 1503 cm^{-1}

* NMR spectrum ($CDCl_3$, 360 MHz) δ: 9.65(OH), 8.02 (NH), 7.52 (H-9'), 7.16-7.05 (N-10', H-11', H-12'), 6.61 (H-9), 6.52 (NH), 6.07 (H-12), 5.75 (H-14), 5.52 (H-17), 5.17 (H-15), 4.75 (-CH-NH), 3.95 (H-17A'),

3.80 (-OMe), 3.77 (-OMe), 3.70 (-OMe), 3.60 (-OMe), 2.60 (H-21A', H-21B'), 2.67 (NMe), 2.62 (H-21), 2.00 (MeCO), 0.92-0.75 (2 Me)

* Rf: 0.51 (CH_2Cl_2:CH_3OH 90:10 silica)

- Minor isomer

* Mass spectrum (DCI-isobutane): 939 (M^+), 940 ($M^+ + 1$)

* IR Spectrum (KBr): 1740, 1675, 1615 cm^{-1}

*NMR spectrum ($CDCl_3$, 360 MHz) δ : 9.25 (OH), 8.02 (NH), 7.50 (H-9'), 7.16-7.05 (H-10', H-11', H-12'), 6.71 (H-9), 6.52 (NH), 6.08 (H-12), 5.82 (H-14), 5.47 (H-17), 5.28 (H-15), 4.75 (CH-NHAc), 3.93 (H-17A'), 3.77 (2 x OMe), 3.60 (OMe), 2.70 (N-Me), 2.02 (MeCO-), 0.95-0.77 (2 Me)

* Rf: 0.39 (CH_2Cl_2:CH_3OH 90:10 silica)

Example 2

α and β isomers of 4-O-deacetyl-4-O-L-N-carbobenzyloxyhemiglutamate vinblastine

A solution of 4-O-deacetyl vinblastine (300 mg, 0,39 mmole) and N-carbobenzyloxy-L-glutamic anhydride (144 mg; 0,519 mmole) in dichloromethane (5 ml) is stirred for 20 hours, at ambient temperature.

The solvent is evaporated under vacuum.

The obtained residue is purified by chromatography on silica (elution: ether/methanol/NH_4OH 25% 50/49.5/0.5).

Accordingly, 230 mg of 4-O deacetyl-4-O-L-N-carbobenzyloxyhemiglutamate vinblastine are obtained in the state of an α and β isomers mixture.

A HPLC analysis of the product indicates the presence of α and β isomers in a ratio of 60:40.

IR spectrum (KBr): 3450, 2960, 2880, 1730, 1612, 1593, 1501, 1459, 1432, 1228 cm^{-1}

Mass spectrum (DCI-isobutane): 1032 ($M^+ + 1$), 984, 926, 723 cm^{-1}

Example 3

Coupling of 4-O deacetyl-4-O-L-N-acetylhemiaspartatevinblastine with galactosylated albumin of human origin

a) 80.6 g of 4-O-deacetyl-4-O-L-N-acetylhemiaspartate vinblastine are dissolved in 2ml dioxane. The solution is then plunged in an ice bath. 24.4 µl triethylamine in 0.5 ml dioxane and 22.6 µl isobutyl chloroformate in 0.5 ml dioxane are added. The mixture is stirred for 1 hour.

Further, a solution of 200 mg galactosylated human albumin in 37 ml H_2O is prepared. The pH is adjusted to 8.5 by NaOH 0.1N.

The solution is refrigerated to 4 degrees C. After 1 hour, the activated ester is added to the solution of galactosylated human albumin. The solution is stirred for 14 hours at 4 degrees C, while the pH is maintained at 8.5 by addition of NaOH 1N. Thereafter, the solution is purified by filtration on a Sephadex gel G 25 equilibrated by a solution of NaCl 9/1000, pH - 7.5. The fractions containing the conjugate are combined, concentrated by ultrafiltration and sterilized. The protein content is determined by the Lowry method and the content of alkaloids is estimated by determination of the radioactivity.

The obtained conjugate contains 13 moles of vinblastine per mole of galactosylated human albumin. The HPLC determination of monomers, dimers and polymers of the conjugate composition indicates 82% monomers and 18% dimers.

b) 80,6 mg 4-O-deacetyl-4-O-L-N-acetylhemiaspartate vinblastine are dissolved in 2 ml dioxane. The solution is then plunged in an

ice bath. 24.4 µl triethylamine in 0.5 ml dioxane and 22.6 µl isobutyl chloroformate in 0.5 ml dioxane are added.

Further, a solution of 200 mg galactosylated human albumine in 37 ml phosphate buffer 0.1 M, pH - 8.2, is prepared. The pH is adjusted to 8.5 by NaOH 1N.

The solution is refrigerated to 4 degrees C. After 1 hour, the activated ester is added to the solution of galactosylated human albumin. The solution is stirred for 14 hours at 4 degrees C, while the pH is maintained at 8.5 by an addition of NaOH 1N. The solution is purified by filtration on Sephadex gel G25 equilibrated by a solution of NaCl 9/1000, pH - 7.5. The fractions containing the conjugate are combined, concentrated by ultrafiltration and sterilized. The protein content is measured by the Lowry method and the content of alkaloids is estimated by determination of the radioactivity.

The obtained conjugate contains 9.3 moles vinblastine per mole galactosylated human albumin. The HPLC determination of monomers, dimers and polymers of the conjugate composition indicates 91.5% monomers, 7% dimers and 1.5% polymers.

The sensibility of the conjugate to lysosomial enzymes has been studied by incubation during 48 hours, at 37°C, in the presence of 5 mM cystein, 40 mM acetate buffer and lysosomial enzymes. Aliquot parts are taken and the non decomposed proteins are precipitated by addition of a trichloroacetic acid volume (TCA 40 %). After incubation at 4°C, during one hour, said samples are centrifuged and the radioactivity of the supernatant is estimated by counting of scintillations of an aliquote part of the liquid.

The soluble radioactivity is a measure of the digestion of said conjugate. Practical tests have shown that 75% of the conjugate has been digested after 24 hours. No further evolution has been observed up to 48 hours.

The chemotherapeutic activity of said conjugate has been estimated on P 388 leukemia with female BDF_1 mice : 10^6 tumoral cells are intraperitoneally inoculated at the day 0. The conjugate is intraperitoneally administrated at the day 1. The test results show that the conjugate presents an important activity on this experimental model, because it provides an increase in life time of more than 650 % if it is administered at a rate of 60 mg/kg and the number of surviving mice is 5/5 after 60 days.

EXAMPLE 4

α and β isomers of ethyl N-(4-O-deacetyl-4-O-acetylhemiaspartate-vinblastinoyl-23)-L-tryptophanate.

Following the procedure of example 1, ethyl N-(deacetyl-4-O-vinblastinoyl-23)-L-tryptophanate was converted to ethyl-N-(4-O-deacetyl-4-O-N-acetyl-hemiaspartate-vinblastinoyl-23)-tryptophanate (mixture of isomers α and β).

The obtained residue is purified by chromatography on silica (elution ether/methanol/NH_4OH 25% 50/49,75/0,25).

200 mg of the product are obtained from 314 mg of starting product.

Mass spectrum (DCI, Acetone): 1126 (M^{+1}), 1066, 984, 970, 951, 911.

IR spectrum (KBr): 3400, 2960, 2940, 1740, 1665, 1618 cm^{-1}.

EXAMPLE 5

α and β isomers of ethyl N-(4-O-deacetyl-4-O-L-N-acetylhemi aspartate-vinblastinoyl-23)-L-isoleucinate.

Following the procedure of example 4, ethyl N-(deacetyl-4-O-vinblastinoyl-23)-L-isoleucinate was converted to ethyl-N-(4-O-deacetyl-4-O-L-N-acetyl hemiaspartate-vinblastinoyl-23)-L-isoleucinate.

Mass spectrum (DCI-acetone) : 1051 (M^{+1}), 1036, 1009, 977, 897, 838, 755, 709, 652.

IR spectrum (KBr) : 3410, 2963, 2929, 2880, 1734, 1676, 1612, 1500, 1460, 1430, 1372, 1335, 1293 cm^{-1}

EXAMPLE 6

α and γ isomers of 4-O-deacetyl-4-O-L-N-acetyl-hemiglutamate vinblastine.

Following the procedure of example 20, 4-O-deacetylvinblastine was treated with N-acetyl-L-glutamic anhydride to yield 271 mg of 4-O-deacetyl-4-O-L-N-acetyl-hemiglutamate vinblastine (mixture of isomers α and γ) from 380 mg of 4-O-deacetylvinblastine.

Mass spectrum (DCI-acetone) : 940 (M^{+1}), 871, 707.

IR spectrum (KBr) : 3470, 2960, 2880, 1740, 1665, 1617 cm^{-1}.

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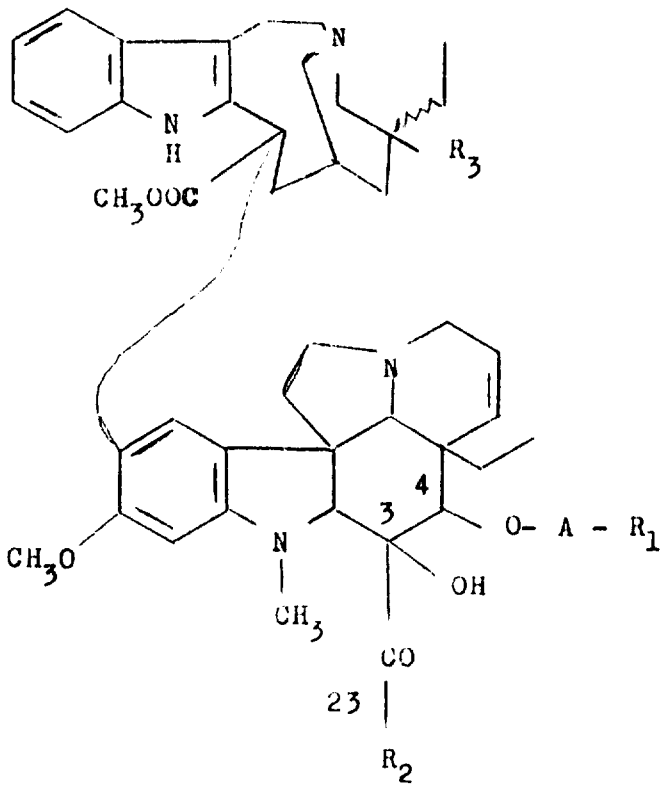
C L A I M S

1. Conjugate of vinblastine of of a derivative thereof corresponding to the general formula :



in which A represents

$$-CO\underset{\underset{NH-R}{|}}{CH}-(CH_2)_n CO- \quad \text{or} \quad -CO-(CH_2)_n \underset{\underset{NH-R}{|}}{CH}CO-$$

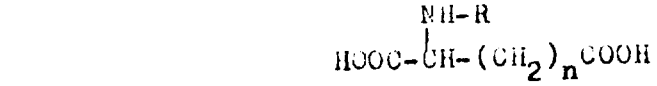
in which n may be 1 or 2 and R represents a hydrogen atom, acetyl or carbobenzyloxy, R_1 represents an optionally modified protein radical T, R_2 represents a methoxy group, and R_3 represents a hydrogen atom or a hydroxyl group, in each case in the two possible configurations.

2. Conjugate according to claim 1 characterized in that the protein radical is one obtainable by monoclonal antibody technique.

3. Conjugate according to claim 1 characterized in that the protein radical consists in monoclonal antibodies which are of human origin.

4. Conjugate according to claim 1 characterized in that the protein radical consists in galactosylated monoclonal antibodies, which are of human origin.

5. Process for the preparation of conjugates according to claim 1 characterised in that an anhydride of an acid of the



type is condensed onto the hydroxyl at C-4 of the vinblastine derivative in dichloromethane

as solvent and in that the derivative thus obtained is then condensed with the protein radical, in a water/dioxane mixture, as solvent.

6. Process for the preparation of conjugates according to claim 1 characterised in that a moiety of the type

$$-CO-\underset{\displaystyle NH-R}{\underset{|}{C}}H-(CH_2)_n-COR_1 \text{ or } -CO-(CH_2)_n-\underset{\displaystyle NH-R}{\underset{|}{C}}HCOR_1,$$

in which R_1 is a optionally modified protein radical T, is condensed onto the hydroxyl at C-4 of the vinblastine derivative and in that an activation of the starting product is effected by treatment with an alkyl chloroformate , in the presence of an amine base.

7. Process according to claim 6 characterised in that the alkyl chloroformate is ethylchloroformate and the amine base in triethylamine.

8. Process according to claim 5, characterised in that a centrifugation, a rinsing, a lyophilisation and/or a purification by gel filtration are furthermore carried out.

9. Process according to claim 8 characterised in that further a conventional succinylation reaction is carried out.

10. Process according to claim 5, characterised in that the protein radical is derived from bovine or human serum albumin, fetuin or immunoglobulins, the latter being obtained, by the monoclonal antibody technique.

11. Process according to claim 6, chacterised in that a centrifugation, a rinsing, a lyophilisation and/or a purification by gel filtration are furthermore carried out.

12. Process according to claim 10, characterised in that further a conventional succinylation reaction is carried out.

13. Process according to claim 6, characterised in that the protein radical is derived from bovine or human serum albumin, fetuin or immunoglobulins.

14. Pharmaceutical composition, characterised in that it comprises the conjugates according to claim 1, in combination with a diluent, a solvent, an excipient or a pharmaceutically acceptable support, the active compound being in lyophilised form or in solution in a buffered solvent, for the treatment of leukemias, lymphosarcomas or other malignant tumours, including so-called "solid" tumours and for the treatment of Hodgkin's disease.