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(54) **Medicament-substituted dextran or levan conjugates, process for their preparation and pharmaceutical compositions containing them.**

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Medicament-substituted dextrin or levan conjugates, process for their preparation and pharmaceutical compositions containing them

The present invention relates to conjugates which are formed between certain medicaments, in particular penicillins, and substituted levans and dextrans said conjugates being useful in providing immunological tolerance in mammals to the administration of the medicaments. The invention also relates to processes for preparing and compositions containing such conjugates.

Many patients suffer from allergic reactions when prescribed particular drugs by their doctor, there being a wide range of drugs which can cause such reactions. In the case of particularly sensitive patients this may prevent certain drugs being prescribed thereby severely restricting the choice of drugs open to the doctor in his fight to combat disease.

The penicillins are a class of drugs which have become increasingly important in the last three decades for the treatment of bacterial infections. Unfortunately however, the penicillins can cause severe allergic reactions and anaphylaxis on administration to animals and humans.

C. H. Schneider and A. L. de Weck, "Immunochemistry" 4 (5), 331—43 (1967) have reported on the reaction of benzylpenicillin with carbohydrates including a delayed type hypersensitivity in guinea pigs. The conjugates described gave an immunogenic reaction and no mention is made that they may create tolerance.

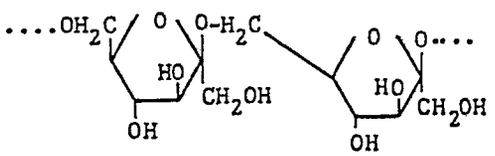
The same authors have reported in Eur. I Immunol., 1 (2), 98—106 (1971) on the low immunogenicity of benzyldextran and nitrobenzyl-dextran conjugates and their usefulness in blocking hapten-specific allergies, however, also this paper does not give any evidence that the conjugates described would create tolerance.

It has now been found that conjugates formed by linking certain medicaments or derivatives thereof to dextran or levan give good tolerance and are poorly immunogenic, that is to say they do not stimulate the production of appreciable amounts of antibodies, after the conjugates have been administered. Furthermore, it has been found that administration of these conjugates after the induction of an allergic reaction to the medicament from which the conjugate is derived effectively counteracts the observed allergic response.

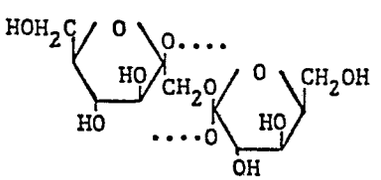
By the term 'medicament' is meant a pharmacologically active substance useful in the field of medicine. The medicaments in the conjugates of this invention are those which cause an allergic reaction on administration to humans and animals and which contain at least one carboxy group or are modified to contain such a group. Commonly only one part of the medicament molecule or a metabolite of the medicament will cause the allergic reaction. For example, it is known that penicilloic acid and penicillamine are two metabolites of benzylpenicillin which contribute greatly to the allergic reaction observed when benzylpenicillin is administered. Thus metabolites of medicaments which cause an allergic reaction on administration, provided they contain at least that part of the molecule which is responsible for the allergic reaction and at least one carboxy group, are also suitable for inclusion in the conjugates of the present invention as derivatives of medicaments.

Suitable for inclusion in the conjugates of the present invention are levans and dextrans, levans being preferred.

Levans are natural polymers of 2,6- and 2,1- linked *β-D*-fructofuranose present in some bacteria and in grasses. Thus the repeating unit of levan has the formulae (I) and (II):



(I)



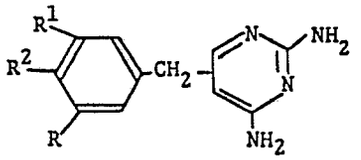
(II)

It can be seen that each furanose ring in levan has three free hydroxy groups to which substituents may be attached.

Dextran is a term applied to polysaccharides produced by bacteria growing on a sucrose substrate, containing a backbone of D-glucose units linked predominantly α-D (1—6).

According to the present invention there is provided a medicament-substituted dextran or levan conjugate having a molecular weight of greater than 20,000, comprising a dextran or levan in which at least 20 hydroxy groups per 1000 fructosyl residues are substituted by —CH_2—CO—NH—X—NHY, wherein X is a C_{1-8} alkylene group optionally substituted by hydroxy groups or a C_{2-8} alkyleneamino

group, wherein Y is a medicament or derivative thereof which causes an allergic reaction on administration to humans and animals which contains at least one carboxy group or is modified to contain such a group, the carboxy group in Y and the amino group —$CH_2CONHXNH_2$ in the dextran or levan forming an amide linkage between the two. Particularly suitable medicaments for inclusion within the conjugates of this invention include penicillins, cephalosphorins, sulphonamides, benzylpyrimidines, extracts of pollens and derivatives thereof, which contain carboxy groups. Suitable derivatives of penicillins include penicilloic acid and penicillamine. Suitable benzylpyrimidines include those of the formula (III):

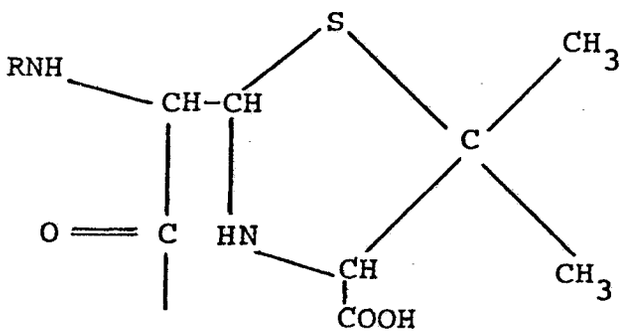


(III)

wherein one of R, R^1, R^2 is a carboxy-group and the others, which may be the same or different are hydrogen, hydroxy, C_{1-4} alkyl or C_{1-4} alkoxy.

By the term X is meant a C_{1-8} alkylene group optionally substituted by hydroxy groups or a C_2—C_8 alkyleneamino group. Conveniently X is a C_{2-5} alkylene group and preferably a propylene group.

In a preferred aspect the present invention provides a penicilloyl-substituted conjugate having a molecular weight of greater than 20,000 comprising a levan in which a plurality of the hydroxy groups are substituted by one or more groups —CH_2—CO—NH—X—NHY wherein X is as hereinbefore defined and Y is a group of the formula (IV):



(IV)

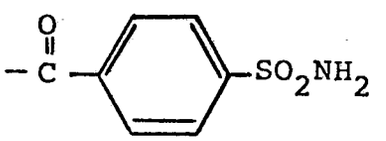
or a pharmaceutically acceptable salt or ester thereof wherein R is a hydrogen atom or an acyl side-chain conventionally linked to the amino group attached to the 6-position in naturally occurring or semi-synthetic penicillins.

Preferably R is a phenylacetyl group.

Pharmaceutically acceptable salts include sodium, potassium, calcium, magnesium, aluminium, ammonium and substituted ammonium salts. The sodium and potassium salts are particularly suitable.

Esters suitable for the purposes of this invention include those notionally derived from an alcohol ROH wherein R is an alkyl, alkenyl, alkynyl, aryl or aralkyl group which may be substituted if desired. Preferably the esters are *in vivo* hydrolysable esters such as a pivaloyl-oxymethyl or phthalimido esters.

In a further preferred aspect of the present invention provides a sulphonamide-substituted levan conjugate having a molecular weight of greater than 20,000 comprising a dextran or levan in which a plurality of the hydroxy groups are substituted by one or more groups —CH_2—CO—NH—X—NHY wherein X is as hereinbefore defined and Y is a group



or a pharmaceutically acceptable salt thereof.

It has been found that although a degree of tolerance is induced when the conjugate has a molecular weight as low as 20,000 higher molecular weight conjugates are preferred, i.e. those with a molecular weight of at least 10^5 and preferably at least 10^6. It has also been found that the degree of induced tolerance is surprisingly increased when the number of Y groups, i.e. the medicament component, attached to the hydroxy groups of the levan or dextran is increased. Whilst it has been

found that tolerance may be induced with a levan or dextran substituted by only 20 Y groups per 1,000 fructosyl residues the levan or dextran is most suitably substituted by at least 60 and preferably by at least 90 Y groups per 1,000 fructosyl residues.

The conjugates of the present invention do not elicit passive cutaneous anaphylaxis, particularly when they are highly substituted, and are therefore preferable to the other known tolerogens. Furthermore the conjugates suppress the immune response to the hapten and are therefore free of potentially troublesome side effects.

The present invention also provides a process for preparing medicament-substituted dextran or levan conjugates which process comprises the reaction of a medicament, or derivative thereof, containing at least one carboxy group with a dextran or levan in which at least 20 hydroxy groups per 1,000 fructosyl residues are substitued by

$$—CH_2—CO—NH—X—NH_2,$$

wherein X is as hereinbefore defined.

In the case of penicillins the reaction will normally be carried out in an aqueous solvent, conveniently water, and at alkaline pH, for example at a pH of greater than 9. This may conveniently be achieved by buffering the solution with a conventional alkaline buffer such as carbonate buffer. Carrying out the reaction at alkaline pH converts the penicillin to a penicilloyl moiety which reacts with the substituted levan to give a penicilloyl-substituted hapten conjugate.

The reaction will be carried out at a non-extreme temperature, for example between 0° and 100°C, and may conveniently be carried out at room temperature.

In the case of other medicaments or derivatives thereof hereinbefore defined the reaction may conveniently be carried out in the presence of a conventional condensation promoting agent such as a carbodiimide. This reaction is normally carried out at acid pH, suitably between 4.5 and 6.5, and at a non-extreme temperature, i.e. between −10°C and 100°C, and conveniently at room temperature, in a suitable solvent, for example water or a mixture of water with a water miscible organic solvent.

Alternatively the carboxylic acid group of the medicament or derivative thereof may be converted into an N-acylating derivative by methods well known to those skilled in the art. Suitable N-acylating derivatives include acid halides and anhydrides. The N-acylating derivative is then reacted with the substituted levan under conditions well known to those skilled in preparing amides.

In a further aspect this invention provides a pharmaceutical composition for the purpose of medical or veterinary treatment which comprises a medicament substituted dextran or levan conjugate hereinbefore defined in combination with a pharmaceutically acceptable carrier for instance in a sealed or sterile state, or in a dosage form.

The compositions of the invention include those in a form adapted for oral, topical or parenteral use and may be used for the treatment of immune hypersensitivity in mammals including humans.

Suitable forms of the compositions of this invention include tablets, capsules, creams, syrups, suspensions, solutions and sterile forms suitable for injection or infusion. Such compositions may contain conventional pharmaceutically acceptable materials such as diluents, binders, colours, flavours, preservatives, disintegrants in accordance with conventional pharmaceutical practice.

Injectable compositions containing the medicament substituted dextran or levan conjugates, especially injectable compositions suitable for intravenous administration, are particularly preferred compositions of this invention. Such compositions will be made up in a sterile form in an aqueous solvent vehicle, such as aqueous polyethylene glycol, or saline. Saline, which may be buffered to physiological pH as required, is a particularly suitable solvent vehicle.

The compositions of the present invention are prepared by conventional formulation techniques well known to those skilled in the art, for example the injectable compositions are prepared by adding the medicament-substituted conjugates to the aqueous solvent vehicle and sterilising the resultant solution.

In a yet further aspect the present invention provides a method of treatment of immune hypersensitivity to a medicament in mammals, including humans, which comprises the administration of an effective dose of the appropriate medicament substituted hapten conjugate hereinbefore defined.

The invention also provides a method of treatment of bacterial infections in mammals, including humans, which comprises the administration of an effective dose of a medicament substituted hapten conjugate hereinbefore defined, wherein the medicament in the conjugate is an antibacterially active medicament.

Suitably betwen 0.01 mg/kg and 100 mg/kg and conveniently 1 mg/kg of the medicament substituted hapten conjugate will be administered daily.

The conjugates of the present invention may be administered to induce immunological tolerance to a medicament so that the patient may then receive a course of treatment with that particular medicament or they be administered when a patient is suffering from an allergic reaction to a medicament to induce tolerance to that medicament. In the latter case the degree of substitution of the hapten needs to be higher than it is in the former. The dosage level of the conjugates of the invention will also normally be higher in this case.

Immunogenic properties of the Conjugates of the Invention

The capacity of pen-HSA (ovalbumin and human serum albumin coupled to the potassium salt of benzylpenicillin), Pen-DAP(1)—CM—LE (a penicilloyl-diaminopropylcarboxymethyl levan conjugate containing 22 penicillin groups/1000 fructosyl residues), Pen-DAP(2)—CM—LE (a penicilloyl-diaminopropylcarboxymethyl levan conjugate containing 95 penicillin groups/1000 fructosyl residues), and Pen-DAP—CM—LE (XM50) (a penicilloyl-diaminopropylcarboxymethyl levan conjugate of low molecular weight (M.W. = 3400) to react with goat anti-Pen serum (containing 6.5 mg precipitating antibody per ml) was measured by direct precipitation and by inhibition of precipitation between the same anti-Pen goat serum and Pen-HSA. The results are shown in Table 1.

TABLE 1

Immunoprecipitation of antibody expressed as % of antibody precipitated by Pen-HSA

Pen — DAP(1)—CM—LE	53%
Pen — DAP(2)—CM—LE	78%
Pen — DAP—CM—LE(XM50)	~0%

The relevance of these results to allergy in mice was investigated by inhibition of PCA (passive cutaneous anaphylaxis) reactions. This reaction was measured incubating a constant dilution of antiserum (calculated to give 10 mm PCA spot) with different amounts of inhibitor for 1 hour at room temperature. Fifty (50) μl samples from each mixture were injected into the skin of Wistor rats that were challenged 24—48 hours later with 2.5 mg Pen-HSA and 5 mg of Evans blue (i.v.). The capacity of the compounds to inhibit PCA was indicated by a decrease in the size of the blue spot in the skin. All three compounds listed in table 1 inhibited specific anti-Pen antibodies. Using amounts of 80, 15 and 2 $\times$ 10^4 ng/site respectively (in the order listed in Table 1) 50% inhibition was achieved.

The capacity of the same compounds to elicit a direct PCA was measured. Several dilutions (50 μl) of anti-Pen serum were injected in the skin of rats and one day later they were injected in the same spot with 1 to 50 μg or antigen right after i.v. injection of Evans blue. The results are shown in Table 2.

TABLE 2

Direct PCA reaction with mouse anti-Pen and penicilloyl substituted levans

Antigen (μg/site)	Antiserum Dilution			No antiserum
	1/10	1/20	1/40	
Pen–HSA (50)	13	9	8	5
(1)	15	10	8	5
Pen–DAP(1)–CM–LE (50)	11	7	6	6
(1)	11	9	6	5
Pen–DAP(2)–CM–LE (50)	5	5	5	5
(1)	5	5	5	5
Pen–DAP–CM–LE(XM50) (50)	5	5	5	5
(1)	5	5	5	4

Tolerance Induction Properties of the Conjugates of the Invention

The tolerance capacity of Pen-DAP—CM-levans was examined first by injecting them into mice 2 and 3 weeks before the immunisation scheme. CBAT6T6 and DBA/2 inbred strains of mice, bred by the

Immunobiology Department of the Wellcome Foundation Limited, were immunised as shown in Figure 1. The immunogen (penicilloyl oralalbumin-Pen—OV) was mixed with aluminium sulphate containing 0.02% phenol red as indicated and precipitated with sodium hydroxide just prior to injection. Immediately after precipitation, *B. pertussis* vaccine was added and the volume made up with saline to give 0.2 ml/mouse. Polysaccharides were injected i.v. (0.2 ml/mouse) diluted in phosphate-buffered saline. DBA/2 and CBAT6T6 mice respond to high and low doses of immugen respectively.

To stimulate the product of IgE antibodies 300 larvae of *Nippostrongylus brasiliensis* were injected subcutaneously in the back of the neck (0.2 ml/mouse).

The modified Jerre PFC (plaque forming cell) assay (Immunology *23*, 843, 1972) was used to determine direct (IgM) splenic PFC specific for Penicilloyl and levan determinants. Estimation of IgE titres was carried out by titrating each serum independently. The animals were bled only once and serial dilutions of the sera (50 μl samples) injected i.v. in the skin of Wistar rats for PCA.

Heterologous cell transfer (HCT) was measured by the method of Kind & Sobrinho (J. Immunol. *111*, 638, 1973). Duplicate 50 and 100 μl samples of washed spleen suspensions in 199 medium (Burroughs Wellcome) were injected into the skin of Ag—B5 HO rats. Twenty-four hours later the animals were challenged in the same way as for PCA reactions and the diameter of the blue spots measured. The number of spleen cells secreting IgE antibodies specific for Pen or OV was assumed to be proportional to the area of the blue spots.

The results in Table 3 indicate that 1 mg doses of Pen-(1)CM—LE and Pen-DAP(2)CM—LE suppressed severely the IgE anti-Pen titre (PCA) measured 2 and 3 weeks after boost with Pen-OV. IgM and IgG PEC/spleen were very low in the immune controls. Both polysaccharide derivatives slightly increased the former and decreased the latter.

To demonstrate that the suppression reflected a diminished number of cells synthesising IgE antibody, rather than peripheral neutralization, the immune response was measured in the spleen of the same animals (Table 3, HCT). The size of the skin reaction in this heterologous transfer is a reflection of the number of cells secreting IgE antibodies, and it is clear from the data presented that tolerance induction resulted in a substantial decrease of spleen cells forming anti-Pen antibodies of the IgE class. A similar experiment was performed using CBA mice, but this time the tolerogen was given after priming, to avoid the problems resulting from the fact that CBA mice had to be immunized several times with low doses of antigen (see Figure 1) to achieve immunity. At the time of tolerogen injection, anti-Pen PCA titres were 1/10. Results of such experiments are presented in Table 4. It is clear that suppression of the immune response also took place in this strain even when tolerogen was given after priming. As with DBA/2 mice the low PCA titre corresponded with smaller numbers of anti-Pen IgE cells in the spleen.

Whether the induction of tolerance could be achieved in mice already showing a high reaginic response was tested according to the following experimental scheme: DBA/2 mice primed, infected and boosted as described in Figure 1 were tested for anti-Pen IgE. On day 33 they were all allergic ($\bar{X}$ = 1/42). On day 47, PCA titres were low (3 positives out of 5, PCA titre 1/23). Subsequently (day 55) mice were divided into 3 groups, the first treated with Pen-DAP(1)—CM—LE, the second with Pen-DAP(2)—CM—LE and the third left as control. Two weeks later they were all boosted in the same way. PCA, HCT, IgM and IgG PFC were measured 14 days after the boost (see Table 5). The results clearly indicate that the highly substituted Pen-DAP(2)—CM—LE provoked a very marked tolerance but the more weakly substituted Pen-DAP(1)—CM—LE only gave a partial, non-significant, suppression. As in the previous experiments, very low spleen IgM and IgG PFC numbers were found. The lack of responsiveness persisted after HCT of the spleen cells.

Preparation of Penicilloyl-diaminopropyl-carboxymethyl-levan

(1) Preparation of carboxymethyl-levan

Purified levan (3 g) from *Corynebacterium levaniformis* was dissolved in water (150 ml) and mixed with 10 N sodium hydroxide (12 ml) and monochloroacetic acid (3 g), stirred for 1 hour at room temperature and at 60°C for 3 hours, neutralised (pH 7) with 5 N hydrochloric acid and dialysed at 4°C for 3 days (checking the conductivity of the water). The material was frozen, dried and analysed for sugar and carboxylic groups (156—COONa/1000 fructosyl residues). The yield was 3.1 g.

(2) Preparation of diaminopropyl-carboxymethyl levan

A 2% solution of carboxymethyl-levan (50 ml) in water was mixed with 1-ethyl-3-(3-dimethyl-aminopropyl)-carbodiimide (0.4 g) in 20% molar excess to the carboxylic groups. The pH was adjusted to 5—6 and a 50 times molar excess of preneutralised diaminopropane (6.4 g) was added at once. The mixture was stirred at room temperature for 24 hours and for the first 3 hours the pH was maintained between 5 and 6. This material was then dialysed at 4°C against 0.01 N sodium hydroxide first and water later, analysed for free amino groups by ninhydrin and the purity checked by gel filtration (Sephadex G—75). The yield was 1.8 g 11% of the carboxy groups had coupled with diaminopropane.

Repetition of the example using different relative amounts of the carbodiimide and diamino-propane alters the degree of substitution of the diamino-carboxymethyl-levan.

(3) Preparation of penicilloyl-diaminopropylcarboxymethyl-levan

Diamino-carboxymethyl-levan (1 g) obtained from (2) was dissolved in a 10% sodium carbonate

solution (50 ml) and potassium benzylpenicillin (4 g) added and dissolved. The solution was kept for two days at room temperature and dialysed at 4°C, frozen and dried (1 g). A full substitution of the amino group was obtained.

Preparation of Penicilloyl-triethylenetetramino-carboxymethyl-levan

The above conjugate was prepared by exactly the same method as penicilloyl-diaminopropyl-carboxymethyl-levan, triethylenetetramine being substituted for diaminopropane.

Preparation of Penicilloyl-triethylenetetramino-carboxymethyl-dextran

The above conjugate was prepared in a similar manner to the levan analogue by substituting dextran (molecular weight about 2×10^6) for levan.

Penicilloyl-triethylenetetramino-carboxymethyl-levan and penicilloyl-triethylenetetramino-carboxymethyl dextran were tested in mice infected with *Nippostrongylus brasiliensis* as described previously for Pen-DAP—CM—LE and found to induce tolerance.

Preparation of Penicilloyl-diaminopropyl-carboxymethyl-dextran

This was prepared as described above for the levan equivalent but substituting dextran (mol. wt. about 2×10^6) for levan.

Preparation of 4-Sulphonamidobenzoyl-diaminopropyl-carboxymethyl-levan

To a solution of diamonopropyl-carboxymethyl-levan (1 g, prepared as described above) and 4-sulphanamidobenzoic acid (1 g) was added 1 ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (10 g) and the pH then adjusted to 8—9 with 1*N* NaOH. The solution was kept at ambient temperature for 2 days, the pH being maintained at the aforementioned value. The reaction mixture was then dialysed against water for 4 days at 4°C and freeze-dried to give product, (SABA—DAP—CM—LE).

Mice were injected with 4-sulphonamidobenzoic acid coupled to chicken gamma globulin (SABA—CGG). The allergic response to (SABA—CGG) in the mice showed cross-reaction with sulphadiazine and sulphaguanidine as measured by inhibition of PCA reactions. A number of the mice were then tolerized with SABA—DAP—CM—LE and sulphamethoxazole, sulphaguanidine and SABA-ovalbumin injected into separate groups of tolerized and untolerized mice. The tolerized mice survived whilst the untolerized mice died. The EIgE antibody titres of the tolerized mice were very low compared to those of the untolerized mice.

Different degrees of substitution can be obtained by changing the proportion of components and the degree of substitution of the diaminopropyl-carboxymethyl-levan used.

Fig. 1

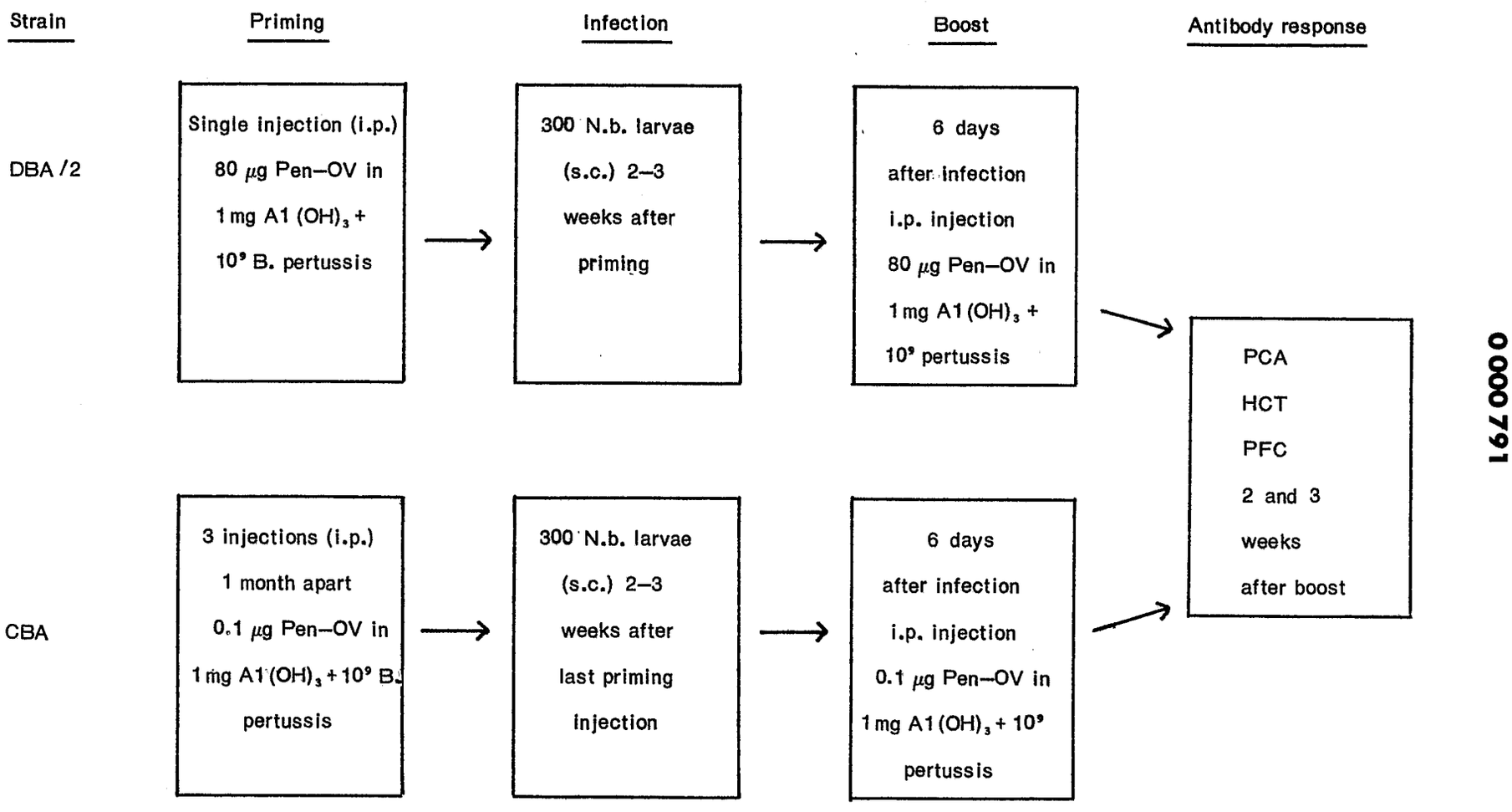


TABLE 3

The effect of high mol. weight Pen–DAP–CM–levans administered prior to induction of allergy to penicilloyl determinants in DBA/2 mice *

	Tolerogen (day 0)	Anti-Pen Immune Response†							
		2 weeks after boost				3 weeks after boost			
		PCA	HCT§	IgM§	IgG§	PCA	HCT	IgM	IgG
Exp. 1	1 mg Pen–DAP(1)–CM–NLE (i.v.)	<1/5	n.d.‡	990(1.34)	307(1.63)	<1/5	n.d.	299(1.49)	179(1.51)
	NIL	1/82(1.48)	n.d.	187(1.53)	680(1.36)	1/35(1.34)	n.d.	112(1.12)	257(1.57)
Exp. 2	1 mg Pen–DAP(2)–CM–NLE (i.v.)	1/7 (1.70)	<1	375(1.30)	105(1.05)	<1/5	<1	<100	<100
	NIL	1/84(1.17)	20.9(2.0)	255(1.38)	336(1.68)	1/79(1.42)	6.0(2.5)	<100	200

* Immunization according to Fig. 1, for Exp. 1 priming, infection and boosting on days 15, 31 and 38 respectively. For Exp. 2, priming, infection and boosting took place on days 23, 44 and 50, respectively.

† Results as geometric means of 5 animals; standard error in parentheses.

§ Blue area in mm.

§ Direct (IgM) and indirect (IgG) PFC/spleen.

‡ Not done.

TABLE 4

Tolerogenicity of high mol. weight Pen–DAP–CM–Levans on IgE response of CBA mice *

Tolerogen	Anti-Pen immune response†		
	2 weeks after boost	3 weeks after boost	
	PCA†	PCA	HCT§ (mm^2)
Pen–DAP(1)–CM–NLE	<1/10	<1/5	<1
Pen–DAP(2)–CM–NLE	<1/1L	<1/5	<1
Nil	1/63(1.73)	1/23(2.2)	12.6(1.59)

Immunization according to Fig. 1

* Priming: days 0, 28 and 59.

Tolerogen: day 78

Infection (N.b.): day 79

Boost: day 85

† Geometric average of 5 mice. Standard error in parenthesis.

§ Area of the blue spot (mm^2).

TABLE 5

Induction of tolerance in DBA/2 mice already allergic to Penicilloyl groups *

Tolerogen (day 55)	No. of animals per group	Immune response (day 83)†			
		PCA	HCT (mm^2)	PFC/spleen IgM	PFC/spleen IgG
Pen–DAP(1)–CM–NLE	7	1/59(1.31)	14.7(1.15)	880(1.33)	1870(1.28)
Pen–DAP(2)–CM–NLE	6	1/18(1.73)	5.9(1.37)	940(1.41)	2400(1.26)
NIL	5	1/149(1.68)	24.9(1.19)	630(1.88)	3630(1.34)

Immunization according to Fig. 1

* Priming: day 0

Infection (N.b.): day 14

Boost: day 20

Test for PCA: days 33 and 47

Tolerogen: day 55

Boost: day 69

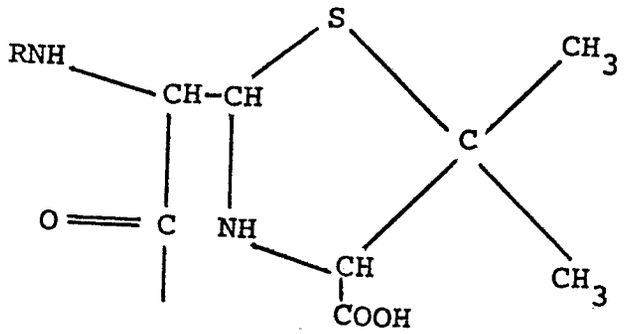
Immune response: day 85.

† Results expressed as geometric means (standard errors in parenthesis).

Claims

1. Medicament-substituted dextran or levan conjugates having a molecular weight of greater than 20,000, comprising a dextran or levan in which at least 20 hydroxy groups per 1000 fructosyl residues are substituted by —CH_2—CO—NH—X—NHY, wherein X is a C_{1-8} alkylene group optionally substituted by hydroxy groups or a C_{2-8} alkyleneamino group, and Y is a medicament or derivative thereof which causes an allergic reaction on administration to humans and animals which contains at least one carboxy group or is modified to contain such a group, the carboxy group in Y and the amino groups —$CH_2CONHXNH_2$ in the dextran or levan forming an amide linkage between the two.

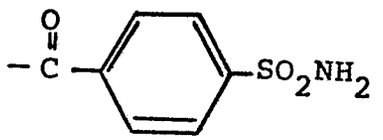
2. Medicament-substituted dextran or levan conjugates according to claim 1 wherein Y is a group of the formula (IV):



or a pharmaceutically acceptable salt or ester thereof, wherein R is a hydrogen atom or an acyl side-chain conventionally linked to the amino group attached to the 6-position in naturally occurring or semi-synthetic penicillins.

3. Medicament-substituted dextran or levan conjugates according to claim 2 wherein R is a phenylacetyl group.

4. Medicament-substituted dextran or levan conjugates according to claim 1 wherein Y is a group:



or a pharmaceutically acceptable salt thereof.

5. Medicament-substituted dextran or levan conjugates according to any one of claims 1—4 wherein X is a propylene group.

6. Medicament-substituted dextran or levan conjugates according to any one of claims 1 to 5 wherein the molecular weight is at least 10^6.

7. Medicament-substituted levan conjugates according to any one of claims 1 to 6 wherein the levan is substituted by at least 90 Y groups per 1,000 fructosyl residues.

8. A pharmaceutical composition which comprises a medicament-substituted dextran or levan conjugate according to any of the preceding claims in the form of an injectable composition for intravenous administration.

9. A process for preparing medicament-substituted dextran or levan conjugates, according to claims 1 to 7, which comprises the reaction of a medicament or derivative thereof, containing at least one carboxy group with a dextran or levan in which at least 20 hydroxy groups per 1,000 fructosyl residues are substituted by

—CH_2—CO—NH—X—NH_2,

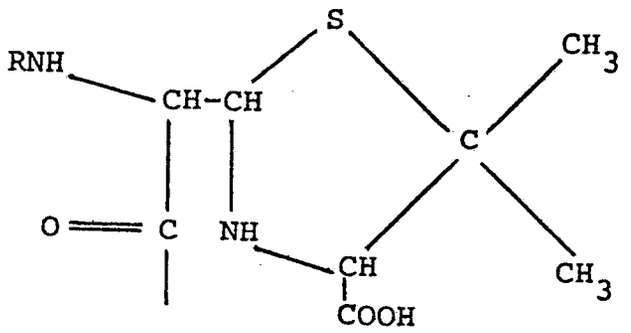
wherein X is as defined in claim 1.

Revendications

1. Conjugués de dextran ou de levan substitués par un médicament, ayant un poids moléculaire supérieur à 20 000, caractérisés par le fait qu'ils sont constitués par un dextran ou un levan dans lequel au moins 20 groupes hydroxy pour 1000 résidus fructosyle sont substitués par un groupement —CH_2—CO—NH—X—NHY, dans lequel X est un groupe alcoylidène de 1 à 8 atomes de carbone éventuellement substitué par des groupes hydroxy ou un groupe alcolyidèneamido de 2 à 8 atomes de carbone, et Y est un médicament ou un dérivé d'un médicament donnant lieu à une réaction allergique

lorsqu'il est administré à des sujets humains et à des animaux qui contient au moins un groupe carboxy ou est modifié de façon à contenir un tel groupe, le groupe carboxy de Y et le groupe amino $—CH_2CONHXNH_2$ du dextran ou du levan formant une liaison amide entre eux.

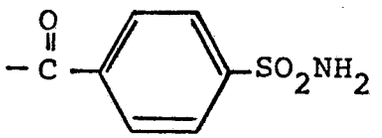
2. Conjugués de dextran ou de levan substitué par un médicament selon la revendication 1, caractérisés par le fait que Y est un groupe de formule (IV):



ou un sel ou ester pharmaceutiquement acceptable de celui-ci, R étant un atome d'hydrogène ou une chaîne latérale acyle liée de façon classique au groupe amino fixé en position 6 dans les pénicillines naturelles ou semi-synthétiques.

3. Conjugués de dextran ou de levan substitués par un médicament selon la revendication 2, caractérisés par le fait que R est un groupe phénylacétyle.

4. Conjugués de dextran ou de levan substitués par un médicament selon la revendication 1, caractérisés par le fait que Y est un groupe



ou un sel pharmaceutiquement acceptable de celui-ci.

5. Conjugués de dextran ou de levan substitués par un médicament selon l'une quelconque des revendications 1 à 4, caractérisés par le fait que X est un groupe propylène.

6. Conjugués de dextran ou de levan substitués par un médicament selon l'une quelconque des revendications 1 à 5, caractérisés par le fait qu'ils présentent un poids moléculaire d'au moins 10^6.

7. Conjugués de levan substitués par un médicament selon l'une quelconque des revendications 1 à 6, caractérisés par le fait que le levan est substitué par au moins 90 groupes Y pour 1000 résidus fructosyle.

8. Composition pharmaceutique, caractérisée par le fait qu'elle contient un conjugué de dextran ou de levan substitué par un médicament selon l'une quelconque des revendications précédentes et se présente sous la forme d'une composition injectable pour une administration intraveineuse.

9. Procédé de préparation de conjugués de dextran ou de levan substitués par un médicament selon les revendications 1 à 7, caractérisé par le fait qu'il consiste à faire réagir un médicament ou un dérivé du médicament contenant au moins un groupe carboxy avec un dextran ou un levan dans lequel au moins 20 groupes hydroxy pour 1000 résidus fructosyle sont substitués par un groupement

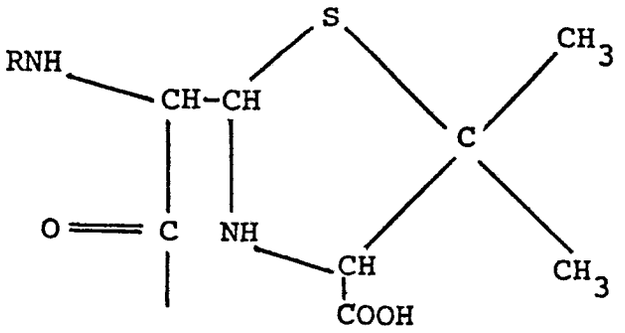
$$—CH_2—CO—NH—X—NH_2,$$

dans lequel X est tel que défini à la revendication 1.

Patentansprüche

1. Medikament-substituierte Dextran- oder Leven-Konjugate mit einem Molekulargewicht von mehr als 20 000, die ein Dextran oder Levan enthalten, in dem mindestens 20 Hydroxylgruppen pro 1 000 Fructosyl-Reste durch $—CH_2—CO—NH—X—NHY$ substituiert sind, wobei X eine gegebenenfalls durch Hydroxylgruppen oder eine C_{2-8}-Alkylenaminogruppe substituierte C_{1-8}-Alkylengruppe ist, und Y ein Medikament oder dessen Derivat ist, das bei der Verabreichung an Menschen und Tiere eine allergische Reaktion bewirkt, das wenigstens eine Carboxylgruppe enthält oder das so modifiziert ist, daß es eine derartige Gruppe enthält, und wobei die Carboxylgruppe in Y und die Aminogruppe $—CH_2CONHXNH_2$ im Dextran oder Levan untereinander eine Amidbindung ausbilden.

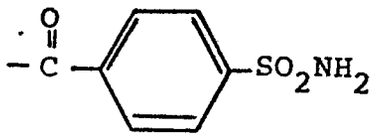
2. Medikament-substituierte Dextran- oder Levan-Konjugate nach Anspruch 1, in denen Y eine Gruppe der Formel (IV)



oder ein pharmazeutisch unbedenkliches Salz oder ein Ester davon ist, worin R ein Wasserstoffatom oder eine Acyl-Seitenkette ist, die in üblicher Weise an die Aminogruppe gebunden ist, die sich in der 6-Position von natürlich auftretenden oder halbsynthetischen Penicillinene befindet.

3. Medikament-substituierte Dextran- oder Levan-Konjugate nach Anspruch 2, in denen R eine Phenylacetylgruppe ist.

4. Medikament-substituierte Dextran- oder Levan-Konjugate nach Anspruch 1, in denen Y eine Gruppe



oder ein pharmazeutisch unbedenkliches Salz davon ist.

5. Medikament-substituierte Dextran- oder Levan-Konjugate nach einem der Ansprüche 1 bis 4, in denen X eine Propylen-Gruppe ist.

6. Medikament-substituierte Dextran- oder Levan-Konjugate nach einem der Ansprüche 1 bis 5, deren Molekulargewicht mindestens 10^6 beträgt.

7. Medikament-substituierte Dextran- oder Levan-Konjugate nach einem der Ansprüche 1 bis 6, in denen das Levan durch mindestens 90 Y-Gruppen pro 1 000 Fructosyl-Reste substituiert ist.

8. Pharmazeutische Zusammensetzung, die ein medikament-substituiertes Dextran- oder Levan-Konjugat nach einem der vorangehenden Ansprüche in Form einer injizierbaren Zusammensetsung für die intravenöse Verabriechung enthält.

9. Verfahren zur Herstellung von medikament-substituierten Dextran- oder Levan-Konjugaten nach den Ansprüchen 1 bis 7, das die Reaktion eines Medikaments oder seines Derivats, das wenigstens eine Carboxylgruppe enthält, mit einem Dextran oder Levan umfaßt, in dem mindestens 20 Hydroxylgruppen pro 1000 Fructosyl-Reste durch

$$—CH_2—CO—NH—X—NH_2$$

worin X wie in Anspruch 1 definiert ist, substituiert sind.