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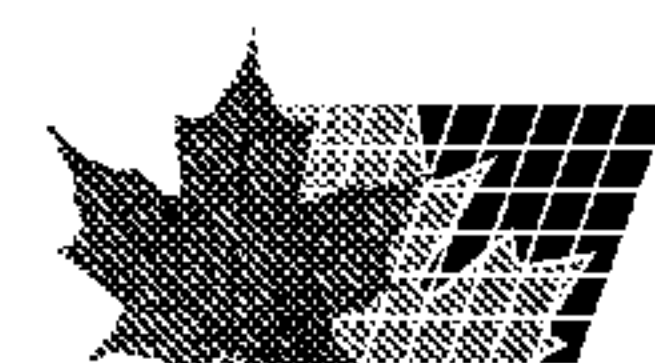
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VASCULAIRES

(54) Title: USE OF HYALURONIDASE FOR PREVENTING AND TREATING CARDIO VASCULAR DISEASES

(57) **Abrégé/Abstract:**

The invention relates to the use of the enzyme hyaluronidase in a dosage of at least 6000 units per day for preventing and/or treating cardiovascular diseases. The hyaluronidase is administrated intravenously over a period of at least four weeks. Hyaluronidase is especially suitable for treating coronary heart disease and for prevention in patients at risk of cardiac infarction.



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(54) Title: USE OF HYALURONIDASE FOR PREVENTING AND TREATING CARDIO VASCULAR DISEASES

(54) Bezeichnung: VERWENDUNG VON HYALURONIDASE ZUR PROPHYLAXE UND BEHANDLUNG VON
HERZ-KREISLAUF-ERKRANKUNGEN(57) Abstract: The invention relates to the use of the enzyme hyaluronidase in a dosage of at least 6000 units per day for prevent-
ing and/or treating cardiovascular diseases. The hyaluronidase is administrated intravenously over a period of at least four weeks.
Hyaluronidase is especially suitable for treating coronary heart disease and for prevention in patients at risk of cardiac infarction.(57) Zusammenfassung: Die Erfindung betrifft die Verwendung des Enzyms Hyaluronidase in einer Dosierung von mindestens
6000 Einheiten pro Tag zur Prophylaxe und/oder Behandlung von Herz-Kreislauf-Erkrankungen, wobei die Hyaluronidase i.v. über
einen Zeitraum von mindestens 4 Wochen verabreicht wird. Insbesondere eignet sich Hyaluronidase für die Behandlung der koro-
naren Herzkrankheit sowie zur Prophylaxe bei herzinfarktgefährdeten Patienten.

WO 01/91785 A3

USE OF HYALURONIDASE FOR PREVENTING AND
TREATING CARDIO VASCULAR DISEASES

The present invention relates to the use of hyaluronidase for prevention and treatment of cardiovascular diseases such as coronary heart disease as well as other arterioscleroses as well as for prevention of restenosis in patients who have already been subjected to a coronary intervention. Coronary interventions include, for example, stent implantation, balloon dilatation, rotablation and bypass operations (arterio-occlusive disease treatments).

- 10 The present invention furthermore relates to prevention and treatment of acute cardiac infarction.

Cardiovascular diseases and the disturbance of well-being and the appearance of physical impairments as its sequelae have in recent years affected an increasing number of people. By now numerous medications have been developed in order to treat heart diseases. These cardiopharmaceuticals include complex compounds like vegetable extracts as well as individual chemical compounds such as those listed in the "2000 Red List". But none of the currently marketed medications has shown really satisfactory results in treating cardiovascular diseases.

- 20 The application NL8102880 describes the use of 20,000 - 80,000 IU of hyaluronidase for treatment of arteriosclerosis, enhanced blood supply and embolism. The hyaluronidase is in these cases applied intra-arterially for some 15-30 minutes at intervals of three weeks.

The object of the present invention is to solve the technical problem of indicating an additional possibility of preventing and treating cardiovascular diseases in humans or animals.

- 30 This problem is solved by the present invention according to patent Claim 1 by using hyaluronidase in a dosage of at least 6,000 units per day for prevention and/or treatment of cardiovascular diseases where the hyaluronidase is

PCT1317-03196/tc

2

administered intra venam over a minimum period of four weeks, preferably in a bolus such as within ten seconds.

Additional advantageous and preferred embodiments of the invention are the subject of subclaims.

According to one embodiment of the invention, the hyaluronidase is used in a dosage of at least 12,000 units per day.

According to a further embodiment of the invention the hyaluronidase is used in a dosage of at least 15,000 units per day. However, it is also possible to work with a dosage of more than 100,000 units per day.

According to still another embodiment of the invention the hyaluronidase is additionally used in combination with carnitin and/or CSE inhibitors and/or nootropics and/or blood supply enhancement agents and/or vitamins and/or cations.

It is obvious that combined use with simultaneous administration such as in the form of a combination medicine or by means of directly successive or temporally staggered administration of the various components can also be made. As a matter of course the combination medicine as well as the various components can be used in connection with pharmaceutically acceptable carriers or dilution agents.

According to yet another embodiment of the invention dosage can be spread out over at least two months and particularly preferable is over at least three months. However, this should only be understood as an example and the concrete dosage duration can naturally be varied in each individual case. Optimum results are obtained by administering a working dose of 12,000 units per day, five to seven times a week over a period of six to eight weeks.

As an example, the cardiovascular disease or disorder may involve a coronary heart disease (CHD), all forms of arteriosclerosis, carotid stenosis, stenosis of the

arteries leading to the brain, cardiac infarction, apoplexy, cardiac hypertrophy or fatty disease of the heart.

Additionally, the use of the enzyme for prevention of restenosis in patients who have already been subjected to a coronary intervention is indicated. Coronary interventions include, for example, stent implantation, balloon dilatation, rotablation and bypass operations (arterio-occlusive disease treatments).

It has moreover been found that according to the invention prevention or treatment of acute cardiac infarction can be carried out.

Here below the invention is described in more detail without limitation.

Hyaluronidase is in itself well-known in the state-of-the-art and belongs to the so-called $\beta(1-4)$ -glycosidases. These enzymes are also designated as hyaluronate glycan hydrolases, EC 3.2.1.35 through 3.2.1.36. Hyaluronidase hydrolyses hyaluronic acid, a linear heteroglycan with alternating glucuronic acid and N-acetylglucosamine residues (acidic glycosaminoglycan (mucopolysaccharide)) and hyaluronate (the ionic form of hyaluronic acid) but also does the same to chondroitin sulphate.

The hyaluronidase can be derived from any source whatsoever and, for instance, may be recovered from bovine protein (bovine type), leeches or bacteria (e.g. in the form of hyaluronate lyase). The hyaluronidase can also be of vegetable origin. The hyaluronidase can be isolated, for instance, from potatoes, tobaccos and peas. Genetic engineering techniques in the art can likewise be used to produce hyaluronidase.

Particularly preferred is any hyaluronidase which splits and thus depolymerises hyaluronic acid, chondroitin-4-sulphate, chondroitin-6-sulphate and mucotin sulphate where the most preferred hyaluronidase is an enzyme available commercially such as, by way of example, the hyaluronidase marketed under the trade name of Hylase® "Dessau" by the firm of Pharma Dessau.

The hyaluronidase can of course not only be used to therapy an already existing disorder or disease of the cardiovascular system but also for preventive treatment, that is prophylactically, of the indications cited above in order to avoid or delay the occurrence of such disorders.

For treatment and/or prevention of the disorders indicated above, a mixture of hyaluronidases of different origins can also be used.

Particularly well suited is any hyaluronidase for prevention in the case of patients at risk of cardiac infarction and/or for treatment of coronary heart disease. Here the enzyme can also be administered directly intra-coronarily in addition to intravenous administration.

Coronary heart disease is also known under the terms of "stenotic coronary sclerosis", "degenerative coronary disease" and "ischaemic heart disease". A pathophysiologic basis is reduction in blood supply caused by sclerosis or occlusion of coronary vessels and thus the supply of energy-providing substrate and oxygen to the heart muscle entailing a discrepancy between supply and demand and clearly noticeable under strain (limited coronary reserves). As syndromes the following are known: angina pectoris, cardiac infarction as well as left ventricular failure. A CHD can also progress for a long time asymptotically while in the exercise ECG under certain conditions signs of a lack of blood supply can be shown.

The success of treatment can be checked, for example, by means of an ECG, an exercise ECG, a long-term ECG, echocardiography, stress echo as well as with myocardial scintigraphy and coronary angiography as well as coronary angioscopy. It has generally been shown that by means of administration according to the present invention blood supply to the coronary vessels was improved, to some extent even attaining complete healing.

The following examples will illustrate the invention without being limiting.

Numerous patients with cardiovascular diseases were treated, some of whom in particular suffered from coronary heart disease. The patients were treated for at least four weeks, preferably for at least two months, and most preferably for at least three months, with a hyaluronidase-containing preparation, preferably with the medicament Hylase® "Dessau". When administering 6,000 units per day of this hyaluronidase preparation, intravenously, by way of example, five times (up to seven times) per week, after several weeks a significant reduction in complaints was observed with the patients.

Some patients were also treated with dosages of Hylase® "Dessau" in the range of 12,000 units per day, five to seven times per week, and with dosages extending up to 15,000 units per day, five to seven times per week. Besides the preferably used intravenous application, other kinds of application such as intramuscular or subcutaneous application, also led to success but where the intravenous application was the most effective method of administration.

When using other hyaluronidases than Hylase® "Dessau" other dosages may be required which the person skilled in the art can easily determine according to the practical circumstances of the case.

According to another aspect of the invention, treatment of cardiovascular diseases with hyaluronidase is done by way of exemplification in combination with carnitin. By using this combination, cardiac hypertrophy and fatty disease of the heart can be counteracted and thus the risk of an infarction can be significantly reduced.

Biochemically, the use according to the invention is directly connected with the degradation of fatty acids. Since the fatty acid synthesis preferably takes place in the cytoplasm while oxidative breakdown of fatty acids takes place in the mitochondria, the fatty acids to be broken down must first migrate from the cytoplasm to the mitochondria. Carnitin (4-trimethylamino-3-hydroxybutyric acid) assists in such migration and can react both with acetyl CoA as well as with acyl CoA with the formation of acetyl-carnitin or acyl-carnitin. The formation of carnitin-activated acetic acid or fatty acid is catalysed by means of acyl-CoA-carnitin-

transferase of which there are two distinct types (for acetyl transfer as well as for acyl transfer). By substituting the acetyl residue for a fatty acid residue a carnityl-fatty acid compound is produced which can pass through the mitochondria membrane. This explains the favourable effect of carnitin on fatty acid oxidation. After entering the mitochondria, the fatty acid is transferred from the acylcarnitin to the CoA and the carnitin is then again available for fatty acid transport.

Additional active ingredients neither possessing any coenzyme function nor demonstrable as essential components for humans but being indispensable cellular components and to some extent performing functions as yet not researched are collectively referred to as vitamin-like active agents. They to some extent influence the permeability of membranes and capillaries.

These include, besides carnitin:

- Meso-inositol (myo-inositol)
- Essential fatty acids (vitamin F)
- Flavanoids (vitamin P)

A further aspect of the invention relates to the use of hyaluronidase in combination with so-called CSE inhibitors (CSE = cholesterol synthesising enzymes) such as pravasin and all other so-called "statins". In this case it is unambiguously proven (e.g. in the WCS study) that pravasin in particular and other statins as well have a cardio-protective effect. Additionally, they act against arteriosclerosis precisely as hyaluronidase does. The purpose of the combination is to achieve optimisation of therapy by means of the two substances' synergies.

Additional substances which may favourably influence the effect of hyaluronidase and which can be used in combinations according to the invention:

- 1) Magnesium and other cations
- 2) Nootropics (e.g. piracetam)
- 3) Agents enhancing blood supply (e.g. pentoxifillin, ginkgo, etc.)

PCT1317-03196/tc

7

- 4) Various vitamins (e.g. vitamin C)
- 5) Substances increasing NO synthesis
- 6) Glycoprotein antagonists such as eptifibatid
- 7) Plasminogen activators such as alteplase, reteplase, etc. in case of acute coronary thrombosis (cardiac infarction)

Use according to the invention is furthermore suitable for the following indications:

- 1) Cerebral sclerosis
- 2) Multiple sclerosis
- 3) Oncology: improvement of resorption of cytostatics in diseased tissue
- 4) Ophthalmology (glaucoma)
- 5) Diabetic micro-angiopathy
- 6) Increase in fertility in men and women

Patent claims

1. Use of hyaluronidase in a dosage of at least 6,000 units per day for prevention and/or treatment of cardiovascular diseases whereby the hyaluronidase is administered intra venam over a period of at least four weeks.
2. Use according to Claim 1 characterised in that the hyaluronidase is used in a dosage of at least 12,000 units per day.
3. Use according to Claim 2 characterised in that the hyaluronidase is used in a dosage of at least 15,000 units per day.
4. Use according to one of the previous claims characterised in that the hyaluronidase is used in combination with carnitin and/or CSE inhibitors and/or nootropics and/or agents enhancing blood supply and/or vitamins and/or cations.
5. Use according to one of the previous claims characterised in that administration is carried out over at least two months.
6. Use according to one of the previous claims characterised in that the disease involves coronary heart disease (CHD), arteriosclerosis, carotid stenosis, stenosis of arteries leading to the brain, cardiac infarction, apoplexy, cardiac hypertrophy or fatty disease of the heart.