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(54) Titre : PROCEDE DE PRODUCTION DE LOVASTATINE
(54) Title: METHOD OF PRODUCTION OF LOVASTATIN

(57) Abrégé/Abstract:
The method finds application in the pharmaceutical industry. Lovastatin is derived by this method from culture broth by filtration at values of pH 9.5-13.0, included in a solid mass it is precipitated from the filtrate obtained, pH 2.5-4.0, in the presence of an inert filler, antioxidant and a non-miscible with water organic solvent. It is extracted and lactonized in the medium of a chlorine-containing organic solvent. The latter is concentrated, and the residue is dissolved in a mixture of solvents having different polarity. After cooling at -10-- 30°C, lovastatin is crystallized, dried and several times recrystallized.

ABSTRACT

The method finds application in the pharmaceutical industry. Lovastatin is derived by this method from culture broth by filtration at values of pH 9.5–13.0, included in a solid mass it is precipitated from the filtrate obtained, pH 2.5–4.0, in the presence of an inert filler, antioxidant and a non-miscible with water organic solvent. It is extracted and lactonized in the medium of a chlorine-containing organic solvent. The latter is concentrated, and the residue is dissolved in a mixture of solvents having different polarity. After cooling at -10–-30°C, lovastatin is crystallized, dried and several times recrystallized.

METHOD OF PRODUCTION OF LOVASTATIN

Field of technics

The invention is related to a method of production of lovastatin finding application in the pharmaceutical industry.

Preceding state of technics

Methods of isolation of lovastatin are known where the filtered culture broth, containing lovastatin, is extracted using ethyl acetate, the extract obtained is lactonized by boiling in toluene, and the isolated product is purified by chromatographing on silica gel (US 4444784, US 4450171).

A method of lovastatin isolation is known, where a sample of culture broth, mycelium or filtrate is extracted by butyl acetate, at pH 3-5, the resulting extract is concentrated through simultaneous lactonization, under vacuum, then through cooling lovastatin in lactone form is directly crystallized from it. (WO 94/29292).

A method of lovastatin isolation and purification is known, where the culture broth is alkalized to pH 8.5-9 and it is filtrated. The separated mycelium is extracted by alkaline water, at continuous agitation, it is filtrated, combined with the filtrate, obtained during culture broth filtration and it is purified through butyl acetate extraction at pH 4 or through ultrafiltration. The obtained filtrate is concentrated at simultaneous lactonization under vacuum. Gaseous ammonia is passed through the concentrate, the resulting precipitate is filtrated, and the filtrate is reconcentrated to 1/10 of its initial volume, then lovastatin in lactone from is crystallized through cooling to -20°C. The obtained crystals are filtrated, dried under vacuum at about 40°C and recrystallized in ethyl acetate, butyl acetate or ethanol. (BG 60460A)

A disadvantage of these methods is the necessity for using large volumes of extracting agents which leads to burdening expenses including also the ones for their reclamation with the view of protecting the environmental elements. The purification by extraction or by ultrafiltration/reverse osmosis demands usage of sophisticated and expensive equipment, the process are multistage, which contributes to the decrease of the yield. As a result of oxidation by-products are formed in the process of isolation of lovastatin and during the storage of the finished product obtained by any of the known methods. This leads to a decrease of the purity and the stability of the product.

Another method of lovastatin isolation and purification is known, where culture broth is diluted with water, treated at continuous agitation for 2 hours at pH 7.5-10 in presence of organic ingredient in quantity not less than 0.1 weight %, then it is filtrated. From this filtrate at pH 1.0-4.5, in presence of salts of alkali-earth or earth metals, lovastatin is directly precipitated, it is filtrated and purified, preferably by recrystallization. (WO 94/10328).

A disadvantage of this method is the possibility of oxidation of the active ingredient during the process, as well as the high residual activity of lovastatin in mother liquor at precipitation, which is carried out in a volume considerably higher than the initial culture broth volume.

The goal of the invention is creation of a method for production of lovastatin of high purity and stability, which is simplified, reliable, and employs small quantities of solvents.

Technical nature of the invention

The goal of the invention is achieved by a method, in which the culture broth containing lovastatin is treated with an alkaline base in the presence of an antioxidant at a pH value 9.5-13.0, filtered and washed with a diluted solution of an alkaline base. The solution obtained is acidified with a mineral acid to pH 2.5-4.0 in the presence of an antioxidant, inert filler and a 0.1-0.3 % non-miscible with water organic solvent. The precipitate produced is filtered, dehydrated, extracted and lactonized by boiling in the medium of a chlorine-containing organic solvent. The latter is distilled to a high concentration of the residue and dissolved in a mixture of acetonitrile-tert.-butylmethylether-butyl chloride in the presence of an antioxidant, and after cooling from –10°C to -30°C to crystallized lovastatin, which is isolated and dried.

The filtered and dried raw lovastatin is dissolved in a nitrile such as acetonitrile or propionitrile, it is decoloured using a mixture of activated carbon and aluminium oxide and after filtration it is precipited with water at room temperature.The filtered product is dissolved in a mixture of acetate of a lows alcohol-alkane C_7-C_9, then it is cooled at a temperature from 0° to -20°C. It is the filtered and the filtered product is dissolved in acetone and the solution is cooled at a temperature from –10°C to -30°C. The crystalline product obtained is filtered and dried at a temperature of 50°C, and at a pressure of 1 kPa.

More particularly, the method for producing the lovastatin comprises the steps of:

a) providing a culture broth containing lovastatin;

b) treating the culture broth with an alkaline base in the presence of an antioxidant at a pH value of 9.5-13.0;

c) filtering and washing the culture broth treated in step b) with a diluted alkaline base to obtain a filtered and washed solution;

d) adding to the filtered and washed solution of step c) an inert filler, an antioxidant and a 0.1-3.0% non miscible with water organic solvent and acidifying the solution to a pH of 2.5-4.0 to obtain a precipitate of lovastatin;

e) filtering and azeotropically dehydrating the precipitate of step d);

f) lactonizing the precipitate filtered and dehydrated in step e) by boiling in a medium of a chlorine-containing organic solvent;

g) distilling the solvent of step f) to a high concentration of the residue;

h) dissolving the solvent of step g) in a mixture of solvents of different polarity;

i) cooling the solvent dissolved in step h) to a range of -10°C to -30°C to crystallize the lovastatin present in the solvent dissolved in step h);

j) isolating the lovastatin crystallized in step i);

k) drying the lovastatin of step j);

l) dissolving the lovastatin of step k) in a nitrile to obtain a mixture;

m) decolouring the mixture of step l) with aluminum oxide and activated carbon;

n) filtering the mixture of step m);

o) precipitating the lovastatin present in the mixture of step n) by adding water;

p) filtering the lovastatin precipitated in step o);

q) crystallizing the lovastatin filtered in step p) in a mixture of acetate of a low alcohol-alcane C_7-C_9 at a temperature from 0°C to -20°C;

r) filtering the lovastatin crystallized in step q); and

s) crystallizing the lovastatin filtered in step r) in acetone at a temperature from -10°C to -30°C.

As antioxidants are used : butylhydroxtoluene, butylhydroxyanisole, nordihydroguaiaretic acid, propyl ester of gallic acid, 2,2-methylenå-bis(6-tert.-butyl-4methyl-phenol), sodium pyrosulphite, sodium bisulphite, d,l-α-tocopherylacetate, hydroquinone, etc.

Silica gel, activated carbon, diatomite, kieselguhr, perlite, sawdust, wood shavings, zeolite are used as fillers for the precipitation of lovastatin.

Esters of aliphatic acids (amyl acetate, ethyl propionate, butyl formiate, butyl acetate, etc.), chlorine-containing carbon hydrides (methylene chloride, chloroform, trichloroethylene, trichloroethane, perchloroethylene, dichloroethane, etc.) are used as non-miscible with water organic solvent.

Methyl acetate, ethyl acetate, propyl acetate or butyl acetate can be used as acetates of low alcohols, as the system of propyl acetate-isooctane is recommended.

The crystallization of lovastatin is performed in a mixture of solvents of different polarity in a suitable ratio, as mixtures of isopropanol-acetone, acetonitrile-butyl chloride, acetonitrile-tert.-butylmethylether-butyl chloride are used.

Advantages of the method according to the invention are production of lovastatin of high purity and yield, stable during storage, by using a simplified technological scheme and small quantities of solvents.

Examples for performance of the invention

To culture broth, containing 792 g of lovastatin (calculated as lactone), are added 16.3 g of butylhydroxytoluene in 165 ml of ethyl alcohol, the mixture is alkalified with a 40 % sodium hydroxide to pH 11.5 and stirred for 3 hours. The mycelium is filtered, washed six-fold - each time with 100 l of a 0.05 % sodium hydroxide. To the solution obtained are added 20 g of butylhydroxytoluene and 10 kg of perlite, and the mixture is acidified with a 20 % nitric acid to pH 2.7. After stirring for 30 min, 18 l of chloroform are added, and the mixture is stirred for another 1 h. The residue is separated from the filter-press. The product obtained is suspended in 100 l of chloroform and boiled in order to be to dehydrated using a metal Florence flask type of vessel until lovastatin acid is lactonized thoroughly. The process is monitored by HPLC. The chloroform solution is distilled to the consistence of oil which is dissolved in a mixture of 0.5 l of acetonitrile, 0.5 l of tert.-

butylmethylether and 3.0 l of butyl chloride. After cooling to -20°C for 48 h the product crystallizes.

After filtration and drying, the raw lovastatin is dissolved in 10 l of propionitrile at 35°C, decoloured using 280 g of activated carbon and 280 g aluminium oxide, and after filtration it is precipitated with 46 l of water at room temperature for 2 h. The filtered product is dissolved at 70°C in 3 l of propyl acetate, and after addition of 4 l of isooctane it is cooled to -20°C for 30 h. The filtered product is recrystallized in 2.5 l of acetone at a final temperature of -30°C, filtered and dried at a temperature of 50 °C and at a pressure of 1 kPa.

601 g of lovastatin of 99.5 % purity are produced (HPLC).

Yield of lovastatin from culture broth - 75.5 %.

Optical rotation - -330° (c=5.0 mg/ml, CH_3CN).

IR and NMR spectra of the product are identical to these of Lovastatin USP Reference Standard.

WHAT IS CLAIMED IS:

1. Method for producing lovastatin comprising the steps of:

a) providing a culture broth containing lovastatin;

b) treating the culture broth with an alkaline base in the presence of an antioxidant at a pH value of 9.5-13.0;

c) filtering and washing the culture broth treated in step b) with a diluted alkaline base to obtain a filtered and washed solution;

d) adding to the filtered and washed solution of step c) an inert filler, an antioxidant and a 0.1-3.0% non miscible with water organic solvent and acidifying the solution to a pH of 2.5-4.0 to obtain a precipitate of lovastatin;

e) filtering and azeotropically dehydrating the precipitate of step d);

f) lactonizing the precipitate filtered and dehydrated in step e) by boiling in a medium of a chlorine-containing organic solvent;

g) distilling the solvent of step f) to a high concentration of the residue;

h) dissolving the solvent of step g) in a mixture of solvents of different polarity;

i) cooling the solvent dissolved in step h) to a range of -10°C to -30°C to crystallize the lovastatin present in the solvent dissolved in step h);

j) isolating the lovastatin crystallized in step i);

k) drying the lovastatin of step j);

l) dissolving the lovastatin of step k) in a nitrile to obtain a mixture;

m) decolouring the mixture of step l) with aluminum oxide and activated carbon;

n) filtering the mixture of step m);

o) precipitating the lovastatin present in the mixture of step n) by adding water;

p) filtering the lovastatin precipitated in step o);

q) crystallizing the lovastatin filtered in step p) in a mixture of acetate of a low alcohol-alcane C_7-C_9 at a temperature from 0°C to -20°C;

r) filtering the lovastatin crystallized in step q); and

s) crystallizing the lovastatin filtered in step r) in acetone at a temperature from -10°C to -30°C.

2. The method of claim 1, wherein the antioxidant is selected from the group consisting of butylhydroxytoluene, butylhydroxyanisole, nordihydroguaiaretic acid, propyl ester of gallic acid, 2,2-methylene-bis-(6-tert.-butyl-4-methyl-phenol), sodium pyrosulphite, sodium bisulphite, d,1-α-tocopheryl acetate and hydroquinone.

3. The method of claim 1, wherein the chlorine-containing organic solvent is selected from the group consisting of methylene chloride, chloroform, trichloroethylene, trichloroethane, perchloroethylene and dichloroethane.

4. The method of claim 1 or 2, wherein the mixture of solvents is selected from the group consisting of iso-propanol-acetone, acetonitrile-butyl chloride, acetonitrile-tert.-butylmethylether-butyl chloride, propyl acetate-isooctane and ethyl acetate-n-heptane.