Antiviral agent based on humic acids

Abstract

FIELD: pharmaceuticals.SUBSTANCE: invention relates to the pharmaceutical industry, namely to an antiviral agent. Antiviral agent containing an aqueous solution of a water-soluble fraction of humic acids (WFHA) obtained from oxidized brown coal treated with trypsin enzyme in the ratio of WFHA and trypsin in a buffer solution with subsequent precipitation of WFHA by acidification with the content of WFHA in an aqueous solution in a concentration of from 1.5 to 150 mcg/ml, under certain conditions.EFFECT: above described agent allows you to expand the arsenal of agents with a broad sector of antiviral action against viruses pathogenic for humans.6 cl, 5 tbl, 6 ex

Classifications

A61K31/12 Ketones

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Claims (6)

Hide Dependent translated from Russian

1. An antiviral agent containing an aqueous solution of a water-soluble fraction of humic acids (HFCA) obtained from oxidized brown coal, characterized in that, as HFCA, it contains a water-soluble fraction of humic acids treated with trypsin enzyme in the ratio of HFCA and trypsin (25-35): 1 in 1M Tris buffer solution for at least 5-6 hours, followed by precipitation of HFGC by acidification to a pH of equal to or less than 2.0 units with HFHC content in aqueous solution at a concentration of from 1.5 to 150 μg / ml. 2. The tool according to p. 1, characterized in that it contains an aqueous solution of HFHC in a concentration of from 4.2 to 5.4 μg / ml, having activity against West Nile virus (WNV). 3. The tool according to p. 1, characterized in that it contains an aqueous solution of HFHC in a concentration of from 1.5 to 5.5 μg / ml, having activity against herpes simplex virus type 2 (HSV-2). 4. The tool according to p. 1, characterized in that it contains an aqueous solution of HFHC in a concentration of from 3.0 to 4.0 μg / ml, having activity against the influenza virus A / H1N1 / California / 2009. 5. The tool according to p. 1, characterized in that it contains an aqueous solution of HFHC in a concentration of from 45 to 55 μg / ml, having activity against human immunodeficiency virus (HIV-1). 6. The tool according to p. 1, characterized in that it contains an aqueous solution of HFHC in a concentration of from 100 to 150 μg / ml, having activity against herpes simplex type 1 (HSV-1).

Description

translated from Russian

The invention relates to preparations for the prevention and treatment of viral diseases, in particular caused by influenza viruses, herpes simplex type 1 and type 2, human immunodeficiency (HIV-1) and West Nile virus and can be used in medicine, namely Virology, Pharmacology and Biotechnology.

Humic acids are high molecular weight heteropolymers and are an integral part of objects of natural origin (peat, soil and brown coal). Humic acid molecules are characterized by an irregular structure, heterogeneity of structural elements, and polydispersity. The structural units of humic acids are aromatic condensed systems with side chains and heterocycles. Functional groups are carboxyl, carbonyl, phenolic and alcohol hydroxides, quinoid groups, methoxyls, amino and amido groups, mono-, di-, polysaccharides, peptides, mineral components. The biological activity of humic acids may be due to the presence of various functional groups in these compounds; colloidal properties and component composition. The presence of various functional groups in the HS determines, firstly, the participation of these compounds in chemical oxidation-reduction reactions, and, secondly, the reactivity as a whole, including ion exchange and the formation of chelate compounds. Being colloids, HS show surface-active and electro-surface properties. These properties determine the hydrophilic-hydrophobic and electrostatic interactions of humic compounds. Humic substances, being heteropolymers, can serve as a source of structural fragments of organic macromolecules during biosynthesis in living organisms (Popov A.I., Zelenkov V.N., Teplyakova T.V. Biological activity and biochemistry of humic substances. Part 1. Biochemical aspect (literature review) // Bulletin of the Russian Academy

of Natural Sciences, 2016. - No. 1. - P. 11-18) [1]. It is known that humic substances have a wide range of therapeutic effects: adaptogenic, antitoxic, hepatoprotective, antimicrobial, diuretic and anti-inflammatory effects (Popov A.I., Zelenkov V.N., Teplyakova T.V. Biological activity and biochemistry of humic substances. Part 2 Medical-biological aspect. Literature review) // Bulletin of the Russian Academy of Natural Sciences, 2016. - No. 5. - S. 9-15) [2]. In addition, it was shown that humic acids have antiviral activity against human immunodeficiency viruses (Kornilayeva G.V., Perminova I.V., Gilyazova A.V. et al. Humic substances as promising compounds for creating microbicidal preparations // Russian Immunological Journal 2010. T. 4 (13), No. 3. P.255-260) [3], as well as (Teplyakova T.V., Gashnikova N.M., Balakhnin S.M., Kosogova T.A. Antiretroviral Activity of Extracts from Chaga, Melanin, and Humic Compounds: Modern Mycology in Russia // Materials of the 3rd Congress of Mycologists R Russia, T. 3. M .: National Academy of Mycology, 2012. S. 419-420) [4], herpes simplex and influenza (Ilycheva TN, Balakhnin SM, Gashnikova NM et al. Antiviral Activity of Humic Substances // Third International Conference of CIS IHSS on Humic Innovative Technologies Tenth International Conference daRostim "Humic Substances and Other Biologically Active Compounds in Agriculture" HIT-daRostim-2014 November 19-23, 2014, Lomonosov Moscow State University, Moscow, Russia. P. 42. J Immunol Methods. 1983 Dec 16; 65 (1-2): 55-63) [5].

It is known the use of natural preparations of humic acids isolated from the soil for the treatment of HIV infection (US application No. 20040137085, IPC A61K 35/78, publ. July 15, 2004). The drug is manufactured by the American company Aldrich Chemical Company and is referred to in the application as AVC. Suppression of HIV infection occurs when the leukocytes of an HIV-infected person come in contact with a humic acid preparation. A method of immunostimulating interleukin-2, a method of enhancing the immune response to vaccination using humic acid as an adjuvant for vaccinating a patient with HIV, is also claimed.

However, in this application the United States considers the use of humic acids only for the treatment of HIV infection and there is no data on their use for the treatment of other viral diseases.

Known antiviral agent based on humic acids obtained from medical lignin (brand "Polyphepan", reg. N 80/1211/3 - non-specific enteric sorbent) by oxidation at 150-170 ° C for 0.5-1.5 hours at a pressure of 2.0-2.5 MPa (RF patent No. 2172176, IPC AK61/35/78, publ. 08/20/2001). The drug is effective against herpes simplex virus type 2, type A influenza virus (PR-8), as well as Routh sarcoma virus BH-RSV (RAV-1) serological subgroup A.

However, in the description of the patent for the invention there is no data on the permissible toxicity of an antiviral agent based on humic acids obtained from medical lignin.

The closest analogue (prototype) is an agent against sexual transmission of HIV / AIDS, made in the form of a suppository containing a pharmacologically acceptable base and an active substance (RF patent No. 2531945, IPC A61K 35/02, published on July 27, 2014). As an active substance with anti-HIV activity, a fraction of humic acids isolated from oxidized brown coal is used, and cocoa butter or solid confectionery fat and an emulsifier are used as the basis. The separation of the fraction of humic acids (HAC-1) is carried out by treating brown coal with a 1% solution of sodium hydroxide (NaOH) and heating the mixture for 2 hours in a boiling water bath (at a temperature of 80 ° C). After cooling, the reaction products are centrifuged, the solution is decanted, the insoluble residue is washed once or twice with 100 ml of 1% sodium hydroxide solution and centrifuged again, collecting the main extract and washings in one receiver. To the resulting solution was added a solution of concentrated hydrochloric acid to pH 2 to precipitate humic acids. The resulting precipitate of humic acids is separated by centrifugation and desalted using dialysis. The resulting preparation of humic acids is dried at 80 ° C in an oven.

However, the description of the patent of the Russian Federation No. 2531945 discusses the use of humic acids only for the treatment of HIV infection and there is no data on the use of this drug for the treatment of other viral diseases, in particular caused by influenza viruses, herpes simplex type 1 and type 2, Western Nile. In addition, it is known that the composition of heterogeneous molecules of humic acids (HAs), in particular, contains peptide chains of various compositions and lengths (http://powermatrix-spb.ru/guminovyie_kisloty), which can be released from HAs in the processes of cellular metabolism and have a toxic effect on human cells.

The technical result of the claimed invention is to obtain a preparation of humic acids from oxidized brown coal with a lower content of toxic substances and a wider spectrum of antiviral activity against viruses pathogenic to humans.

The specified technical result is achieved by the fact that in an antiviral agent containing an aqueous solution of a water-soluble fraction of humic acids (HFCA) obtained from oxidized brown coal, according to the invention, as HFCA it contains a water-soluble fraction of humic acids treated with trypsin enzyme in the ratio of HFCA and trypsin (25-35): 1 in 1M Tris buffer solution for at least 5-6 hours, followed by precipitation of HFGC by acidification to a pH value of equal to or less than 2.0 units containing HFHC in an aqueous solution in concentrations from 1.5 to 150 μg / ml.

The tool contains:

- an aqueous solution of HFHC in a concentration of from 4.2 to 5.4 μg / ml, having activity against West Nile virus (WNV);

- an aqueous solution of HFHC in a concentration of from 1.5 to 5.5 μg / ml, having activity against herpes simplex virus type 2 (HSV-2);

- an aqueous solution of HFHC in a concentration of from 3.0 to 4.0 μg / ml, having activity against the influenza virus A / H1N1 / California / 2009;

- an aqueous solution of HFHC in a concentration of from 100 to 150 μg / ml, having activity against herpes simplex virus type 1 (HSV-1);

- an aqueous solution of HFHC in a concentration of from 45 to 55 μg / ml, having activity against human immunodeficiency virus (HIV-1).

Thus, the distinguishing features of the product are, firstly, reduced toxicity (safety) for human cells, due to the purification and enzymatic treatment of humic acids; secondly, a wide range of antiviral effects, shown experimentally. According to the applicant, humic acids have high antiviral activity against five viruses (see examples 2-6) that cause severe human diseases and belong to different families.

Example 1. A method of obtaining an antiviral agent based on humic acids

1.1. Isolation of humic acids. 25 g of brown coal of the Abanskoye field, crushed to a particle size of not more than 0.2 mm, were mixed with 50 ml of 2% ammonia. To the resulting mixture, 40 ml of concentrated hydrogen peroxide was added with continuous stirring. After the end of the gas evolution, the mixture was centrifuged to separate the insoluble part of the coal.

1.2. Purification of humic acids. To precipitate humic acids, the supernatant containing dissolved humic acids was acidified to a pH of less than 2.0 units by the addition of hydrochloric acid and centrifuged again, the supernatant was discarded. Next, the procedure was reprecipitated (successively five times) of humic acids according to the following scheme: the precipitate was resuspended in 10 times the amount of water and the pH was adjusted to a value of at least 10.0 units by adding 20% NaOH; the resulting humic acid solution was then acidified again to a pH of less than 2.0 units and centrifuged.

The precipitate of humic acids was dissolved in a small amount of water and the pH was adjusted to a value of 7-8 units. Then, humic acids were dried at 30 ° C.

1.3. Enzymatic processing. The composition of heterogeneous molecules of humic acids (HA), in particular, contains peptide chains of various compositions and lengths that can be released from the molecules of HA in the processes of cellular metabolism and have a toxic effect on human cells. To reduce toxicity, samples of humic acids were treated with a protease. For this, from 4 to 5.5 g of purified humic acids were dissolved in 100 ml of water, 5 ml of 1M Tris and 150 mg of trypsin were added. The mixture was incubated for at least 5-6 hours at 37 ° C. Humic acids were then precipitated with hydrochloric acid by acidification to a pH of less than 2.0 units. The HA precipitate was dissolved in a small amount of water and the pH was adjusted to 7-8 units. Next, humic acids were dried at 30 ° C.

Example 2. The study of the toxicity and antiviral activity of samples of humic acids against the herpes simplex virus of the second type (HSV-2)

Evaluation of the activity of samples of humic acids (HA) in relation to herpes simplex virus type 2 was carried out in a culture of Vero cells. A strain of MS herpes simplex virus type 2 was used (obtained from the American Type Culture Collection). Prepared dilutions of the preparations were applied to a two-day culture (3 replicates per dilution), except for control wells (virus control and cell control). To assess the toxicity of HA samples, cells were stained with a gentian violet solution. Staining results were recorded using a Zemfira 680 photometer with a BioRed RU Zemf control computer in accordance with the manufacturer's instructions. Received optical density (OD) for each of the wells of the tablet at a wavelength of 570 nm. The average OD value was calculated for each group of three wells with different concentrations of the sample. Based on the obtained values, a graph was constructed of the dependence of OD on the concentration of the sample. 50% toxic concentration (TC_{50}) was determined from the graphs.

To determine the antiviral activity, a virus was added to the wells at a dose of 2×10^{-5} PFU / cell (the volume of the working mixture was 100 μl). The reaction was taken into account on day 4. Assessment of activity was carried out using the method of inhibition of plaque formation. Cells were stained with gentian violet. The percentage of inhibition of plaque formation (% IB) was determined by the formula:% IB = [1 - (number of plaques in the test / number of plaques in the virus control)] × 100. The concentration capable of inhibiting the development of 50% of plaque forming units (PFU) of the number of PFU in control (50% inhibitory concentration - IC_{50}). The average% IB value was calculated for each group of two wells with different concentrations of the sample. Based on the obtained values, a graph of the dependence of% IB on the concentration of the sample was constructed. According to the graphs, $IC_{50\ was}$ determined. Selectivity Index (IS) was defined as the ratio of a 50% toxic dose to a 50% inhibitory dose.

Таблица 1

Сравнение токсичности и активности исходных и модифицированных гуминовых кислот в отношении ВПГ-2

№	Описание образца	TC_{50}, мкг/мл	IC_{50}, мкг/мл	IS
1	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 (без ферментативной обработки).	997,5±15	3,5±2	285
2	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 и 1.3 (после ферментативной обработки).	1867±13	3,5±2	533

Таблица 1

Сравнение токсичности и активности исходных и модифицированных гуминовых кислот в отношении ВПГ-2

№	Описание образца	TC_{50}, мкг/мл	IC_{50}, мкг/мл	IS
1	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 (без ферментативной обработки).	997,5±15	3,5±2	285
2	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 и 1.3 (после ферментативной обработки).	1867±13	3,5±2	533

From the analysis of the data obtained in table 1 it is seen that after enzymatic treatment, the toxicity of the sample of humic acids for human cells decreased by almost 2 times, while maintaining the level of antiviral activity against herpes simplex virus (HSV-2).

Example 3. Testing a sample of humic acids for the presence of antiviral activity against West Nile virus (WNV).

Evaluation of humic acid samples for West Nile fever was performed on a Vero cell culture. In the study, strain Egypt 101 WNV was used (obtained from the State collection of viral strains of the Institute of Virology named after D.I. Ivanovsky of the RAMS, Moscow, Russia). To test the antiviral activity, WNV in a titer of 10^{3} TTCPD / ml was used: at this dose, the virus causes a 100% CPD visible under a microscope on Vero cells.

Таблица 2

Противовирусная активность образца гуминовых кислот в отношении вируса Западного Нила (ВЗН)

№	Описание образца	TC_{50}, мкг/мл	IC_{50}, мкг/мл	IS
1	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 (без ферментативной обработки).	990±30	5,0±0,2	198
2	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 и 1.3 (после ферментативной обработки).	1270±30	4,8±0,6	265

Таблица 2

Противовирусная активность образца гуминовых кислот в отношении вируса Западного Нила (ВЗН)

№	Описание образца	TC_{50}, мкг/мл	IC_{50}, мкг/мл	IS
1	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 (без ферментативной обработки).	990±30	5,0±0,2	198
2	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 и 1.3 (после ферментативной обработки).	1270±30	4,8±0,6	265

Thus, Table 2 shows that humic acids have antiviral activity against West Nile virus belonging to the Flaviridae family.

Example 4. The study of the toxicity and antiviral activity of samples of humic acids against influenza virus A / California / 07/09 (H1N1pdm09) in an MDCK cell culture (dog kidney cells).

MDCK cells were scattered in 96-well plates with a seed concentration of 300,000 cells / ml, 100 µl per well. Dilutions of the studied samples of humic acids were prepared in a support medium. In the work, serial binary dilutions of the samples were used. Prepared dilutions of the preparations were applied to daily culture (2 replicates per dilution), except for control wells (virus control and cell control). To determine antiviral activity, a virus was added to the wells at a dose of 100 $TCID_{50}$ / well. The reaction was taken into account on the 3rd day. Evaluation of the activity of the samples was carried out using a neutral red dye. The test results were recorded using a 680 Zemfira photometer with a BioRad RU Zemf control computer in accordance with the manufacturer's instructions. Indicators of optical density at a wavelength of 490 nm were obtained for each of the wells of the tablet. 50% inhibitory and toxic concentrations of the drugs were calculated using graphs plotted by the average optical density (OD) for each group of two infected and uninfected wells with different drug concentrations. For each dilution, the average OD value was calculated. Based on the obtained values, graphs of the dependence of OD on the concentration of the drug were built. TC_{50} and $IC_{50\ were}$ determined from the graphs. The selectivity index (IS) was calculated as the ratio of TC_{50} to IC_{50} .

Таблица 3

Токсичность и противовирусная активность образца гуминовых кислот в отношении вируса гриппа A/H1N1/California/2009

№	Описание образца	TC_{50}, мкг/мл	IC_{50}, мкг/мл	IS
1	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 (без ферментативной обработки).	997,5±15	3,75±0,5	266
2	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 и 1.3 (после ферментативной обработки).	1200±13	3,5±0,5	343

Таблица 3

Токсичность и противовирусная активность образца гуминовых кислот в отношении вируса гриппа A/H1N1/California/2009

№	Описание образца	TC_{50}, мкг/мл	IC_{50}, мкг/мл	IS
1	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 (без ферментативной обработки).	997,5±15	3,75±0,5	266
2	Гуминовые кислоты, полученные по примеру 1, п.п. 1.1, 1.2 и 1.3 (после ферментативной обработки).	1200±13	3,5±0,5	343

The testing performed (Table 3) confirmed the high antiviral activity of HA against the influenza virus belonging to the Orthomyxoviridae family.

Example 5. The study of the toxicity and antiviral activity of a sample of humic acids against human immunodeficiency virus (HIV-1)

Cells. To work with the human immunodeficiency virus, transplantable human lymphoblastoid cells MT-4 from the cell culture collection of the Institute of Virology named after DI. Ivanovsky "FSBI" NITsEM them. N.F. Gamalei »Ministry of Health of Russia.

Viruses. The HIV-1 899A strain (subtype B) (Germany), HIV-1 IV735 strain (subtype B) (Russia), HIV-1 IV741 strain (subtype AE) (Russia) from the collection of immunodeficiency virus strains were used as a source of human immunodeficiency virus Human Institute of Virology. D.I. Ivanovsky "FSBI" NITsEM them. N.F. Gamalei »Ministry of Health of Russia.

The study of the cytotoxic effect of the drug. Adding to the cells of the studied drug in various concentrations.

Incubation of cells at 37 ° C in an atmosphere with 5% CO_2 and 98% humidity for 5 days. Analysis of results: determination of viability and cell number using dye.

The study of the antiviral effect of the drug. Adding the study drug in various doses simultaneously with infection with the virus at a dose of 0.01 $TCID_{50}$ / cell. Incubation of cell cultures at 37 ° C in an atmosphere with 5% CO_2 and 98% humidity for 5 days. Analysis of results by staining cells with a tetrazolium dye (MTT method) with spectrophotometry and light microscopy: study of the cytopathic effect of the virus (CPE) and virus-induced syncytium formation (syncytium is a conglomerate of several cells with a common cell membrane formed as a result of the fusion of their membranes), determination of the virus antigen in culture fluid infected cells.

The degree of protection of cells from the cyto-destructive effect of the virus was determined by the formula:

$$\% \text{ защиты} = \frac{A - B}{K - B} \times 100, \quad \text{где}$$

$$\% \text{ защиты} = \frac{A - B}{K - B} \times 100, \quad \text{где}$$

A is the number of viable cells in the experimental group;

B - the same in an infected culture (virus control);

The same is in an uninfected culture (cell control).

Determination of the virus antigen in the culture fluid of infected cells was carried out by enzyme-linked immunosorbent assay using a commercial kit GENSCREEN ™ ULTRA HIV Ag-Ab company "BIO-RAD" according to the manufacturer's instructions. The results were taken into account using a US Fax Stat-3200 photometer at a

wavelength of 450/630 nm. The sensitivity of the test system is less than 25 pkg / ml. The results are presented in table 4.

Таблица 4

Противовирусная активность образца водного раствора гуминовых кислот на модели клеток человека, инфицированных ВИЧ-1 (концентрация 50±5 мкг/мл)

Штаммы вируса иммунодефицита человека	Характеристика противовирусной активности	
	Защита клеток от цитодеструктивного действия вируса, %	Снижение уровня вирусного антигена, %
ВИЧ-1 899А (субтип В)	94,8	61,9
ВИЧ-1 ИВ735 (субтип В)	95,1	58,9
ИВ741 (субтип АЕ)	95,9	58,7

Таблица 4

Противовирусная активность образца водного раствора гуминовых кислот на модели клеток человека, инфицированных ВИЧ-1 (концентрация 50±5 мкг/мл)

Штаммы вируса иммунодефицита человека	Характеристика противовирусной активности	
	Защита клеток от цитодеструктивного действия вируса, %	Снижение уровня вирусного антигена, %
ВИЧ-1 899А (субтип В)	94,8	61,9
ВИЧ-1 ИВ735 (субтип В)	95,1	58,9
ИВ741 (субтип АЕ)	95,9	58,7

The data obtained (table 4) showed that a sample of humic acids at a concentration of 50 ± 5 μg / ml had antiviral activity against HIV-1. Marked protection of cells against the cytopathic effect of the virus and a decrease in the level of viral antigen in the culture fluid of HIV-infected cells was noted. Moreover, the drug was effective against strains of HIV-1 of different subtypes isolated in different regions of the world (Western Europe and Russia).

Example 6. The study of the antiviral activity of compounds containing humic acids against herpes simplex virus type 1 (HSV-1)

Materials and methods.

Cells. The green monkey kidney fibroblast cell culture VERO-transplantable cell line sensitive to herpes simplex virus was cultured at 37 ° C and at 5.0% CO_2 .

Virus. The study used herpes simplex virus, type 1, strain L2, obtained from the D.I. Ivanovsky (HSV-1).

The investigated substances. Humic acids.

The experimental design. Test substances were added to a monolayer culture of Vero cells at a concentration of 100, 150, 200 and 300 μg / ml. After 24 hours, cells exposed to the test substances were infected with HSV virus at a dose of 100 $TCID_{50}$. The control was cells not treated with the test substances and infected with HSV-1. The experimental and control cells in 96-well panels were placed in an incubator at 37 ° C and 5.0% CO_2 until 100%) cells were infected in the control. The calculation was carried out microscopically and using the MTT method (using tetrazolium dye).

Таблица 5

Противогерпетическое действие гуминовых кислот в культуре клеток

Концентрация вещества, мкг/мл	Защита от цитопатического действия 100 $ТЦИД_{50}$ ВПГ-1, %	
	Гуминовые кислоты	Контроль ВПГ
100,0	75,0	0,0
150,0	100,0	0,0
200,0	100,0	0,0
300,0	100,0	0,0

Таблица 5

Противогерпетическое действие гуминовых кислот в культуре клеток

Концентрация вещества, мкг/мл	Защита от цитопатического действия 100 $ТЦИД_{50}$ ВПГ-1, %	
	Гуминовые кислоты	Контроль ВПГ
100,0	75,0	0,0
150,0	100,0	0,0
200,0	100,0	0,0
300,0	100,0	0,0

The results of the study.

In control cell cultures (infected with HSV-1 and not treated with the test substances), virus-induced CPE developed after 48 hours. In cell cultures treated with humic acids, protection of cells from the cytopathic effect of HSV-1 was found to be 75-100% at a concentration of 100-150 μg / ml, respectively (table 5).

Thus, examples 1-6 confirm the achievement of the claimed technical result of the claimed invention, namely: obtaining a preparation of humic acids from oxidized brown coal with a lower content of toxic substances and a wider spectrum of antiviral activity against viruses pathogenic to humans.

Patent Citations (3)

Publication number	Priority date	Publication date	Assignee	Title
RU2172176C1 *	2000-06-19	2001-08-20	Общество с ограниченной ответственностью "Нобель"	Method of antiviral agent preparing
US6630179B1 *	1998-09-23	2003-10-07	Enerkom (Proprietary) Limited	Oxihumic acid and its use in the treatment of various conditions
RU2531945C2 *	2013-01-25	2014-10-27	Федеральное государственное бюджетное учреждение "Государственный научный центр "Институт иммунологии" Федерального медико-биологического агентства	Medication against hiv/aids transmission through sexual contact
Family To Family Citations				

* Cited by examiner, † Cited by third party

Non-Patent Citations (4)

Title
"Гумивит" Рег. уд. МЗ РФ N001636.Р.643.06.2000. ТУ 9197-011-46184368-99 [онлайн]. *
"Гумивит" Рег. уд. МЗ РФ N001636.Р.643.06.2000. ТУ 9197-011-46184368-99 [онлайн]. РАШИДА Р.К. БИОЛОГИЧЕСКАЯ АКТИВНОСТЬ ГУМИНОВЫХ ВЕЩЕСТВ, ПОЛУЧАЕМЫХ ИЗ ТОРФА И САПРОПЕЛЯ //Казанский медицинский журнал, 2015 г., том 96, N 1, С. 84-89. *
РАШИДА Р.К. БИОЛОГИЧЕСКАЯ АКТИВНОСТЬ ГУМИНОВЫХ ВЕЩЕСТВ, ПОЛУЧАЕМЫХ ИЗ ТОРФА И САПРОПЕЛЯ //Казанский медицинский журнал, 2015 г., том 96, N 1, С. 84-89. СУХИХ А.С., КУЗНЕЦОВ П.В. ПЕРСПЕКТИВЫ ПРИМЕНЕНИЯ ГУМИНОВЫХ И ГУМИНОПОДОБНЫХ КИСЛОТ В МЕДИЦИНЕ И ФАРМАЦИИ//Медицина Кубани. N9, 2009. С. 10-14. *
СУХИХ А.С., КУЗНЕЦОВ П.В. ПЕРСПЕКТИВЫ ПРИМЕНЕНИЯ ГУМИНОВЫХ И ГУМИНОПОДОБНЫХ КИСЛОТ В МЕДИЦИНЕ И ФАРМАЦИИ//Медицина Кубани. N9, 2009. С. 10-14. *

* Cited by examiner, † Cited by third party

Cited By (2)

Publication number	Priority date	Publication date	Assignee	Title
RU2752872C1 *	2020-11-03	2021-08-11	Федеральное бюджетное учреждение науки "Государственный научный центр вирусологии и биотехнологии "Вектор" Федеральной службы по надзору в сфере защиты прав потребителей и благополучия человека (ФБУН ГНЦ ВБ "Вектор" Роспотребнадзора)	SARS-CoV-2 CORONAVIRUS REPLICATION INHIBITOR BASED ON HUMIC SUBSTANCES
RU2753609C1 *	2020-10-16	2021-08-18	Николай Иванович Милов	Antiviral humic agent
Family To Family Citations				

* Cited by examiner, † Cited by third party, ‡ Family to family citation

Similar Documents

Publication	Publication Date	Title
Jiang et al.	2005	SPL7013 gel as a topical microbicide for prevention of vaginal transmission of SHIV89. 6P in macaques

Lorin et al.	2005	The antimicrobial peptide dermaseptin S4 inhibits HIV-1 infectivity in vitro
Tao et al.	2007	In vitro anti-HIV and-HSV activity and safety of sodium rutin sulfate as a microbicide candidate
Yarchoan et al.	1986	Administration of 3'-azido-3'-deoxythymidine, an inhibitor of HTLV-III/LAV replication, to patients with AIDS or AIDS-related complex
Fernández-Romero et al.	2007	Carrageenan/MIV-150 (PC-815), a combination microbicide
RU2678986C1	2019-02-05	Antiviral agent based on humic acids
WO2005067955A1	2005-07-28	Antimicrobial activity from medicinal mushrooms
Rattanathongkom et al.	2009	Evaluation of chikusetsusaponin IV a isolated from Alternanthera philoxeroides for its potency against viral replication
KR20060017887A	2006-02-27	Agent for inhibiting membrane virus reproduction, method for the production thereof, pharmaceutical composition and method for inhibiting viral infections
Ogura et al.	2010	Evaluation of an edible blue-green alga, Aphanothece sacrum, for its inhibitory effect on replication of herpes simplex virus type 2 and influenza virus type A
Zime-Diawara et al.	2015	The antimalarial action of aqueous and hydro alcoholic extracts of Artemisia annua L. cultivated in Benin: In vitro and in vivo studies
KR20000069296A	2000-11-25	Extracts of Salvia Species Having Antiviral Activity
Stiles et al.	2008	Effects of λ-carrageenan on in vitro replication of feline herpesvirus and on experimentally induced herpetic conjunctivitis in cats
KR900005170B1	1990-07-20	Ant-letrovirul agent
Skopińska-Różewska et al.	2012	The in vivo effect of Rhodiola quadrifida extracts on the antibody production, on the blood leukocytes subpopulations and on the bacterial infection in mice
Barrón et al.	2007	11. Spirulina as an antiviral agent
US20020168416A1	2002-11-14	Indian green mussel (Perna viridis) as a source of anti-HIV activity
RU2531945C2	2014-10-27	Medication against hiv/aids transmission through sexual contact
RU2752872C1	2021-08-11	SARS-CoV-2 CORONAVIRUS REPLICATION INHIBITOR BASED ON HUMIC SUBSTANCES
CN100390200C	2008-05-28	Recombinant targeted fusion protein for treating acquired immunodeficiency syndrome
RU2697887C1	2019-08-21	Agent possessing antiviral action against tick-borne encephalitis viruses and herpes simplex type i
US7056520B2	2006-06-06	Method and composition for the treatment of a viral infection
WO2018142428A1	2018-08-09	Herbal microbicide formulation for preventing hiv
Barrón et al.	2007	Spirulina as an antiviral agent
RU2753609C1	2021-08-18	Antiviral humic agent

Priority And Related Applications

Priority Applications (1)

Application	Priority date	Filing date	Title
RU2018110484A	2018-03-23	2018-03-23	Antiviral agent based on humic acids

Applications Claiming Priority (1)

Application	Filing date	Title
RU2018110484A	2018-03-23	Antiviral agent based on humic acids

Concepts

machine-extracted

Download Filter table

Name	Image	Sections	Count	Query match
humic acid		title,claims,abstract,description	64	0.000
antiviral agent		title,claims,abstract,description	8	0.000
effects		claims,abstract,description	19	0.000
aqueous solution		claims,abstract,description	16	0.000

lignite	claims,abstract,description	9	0.000
Trypsin	claims,abstract,description	7	0.000
Trypsin	claims,abstract,description	7	0.000
pH reduction	claims,abstract,description	4	0.000
trypsin	claims,abstract,description	4	0.000
trypsin	claims,abstract,description	4	0.000
precipitation	claims,abstract,description	3	0.000
Human immunodeficiency virus	claims,description	15	0.000
West Nile virus	claims,description	10	0.000
Human alphaherpesvirus 1	claims,description	9	0.000
solution	claims,description	9	0.000
Herpes Simplex	claims,description	8	0.000
Human alphaherpesvirus 2	claims,description	7	0.000
Influenzavirus A	claims,description	3	0.000
Tris buffer	claims,description	3	0.000
Viruses	abstract,description	24	0.000
anti-viral	abstract,description	20	0.000
substance	abstract,description	17	0.000
drug	abstract,description	13	0.000
chemical substances by application	abstract,description	3	0.000
pathogenic	abstract,description	3	0.000
buffer solution	abstract	1	0.000
pharmaceutical industry	abstract	1	0.000

Show all concepts from the description section