(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
6 May 2010 (06.05.2010)

PCT

(10) International Publication Number
WO 2010/050955 A1

(51) International Patent Classification:
A61K 9/00 (2006.01) *A61K 47/18* (2006.01)
A61K 31/40 (2006.01)

(21) International Application Number:
PCT/US2008/081795

(22) International Filing Date:
30 October 2008 (30.10.2008)

(25) Filing Language: English

(26) Publication Language: English

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(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, NO, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

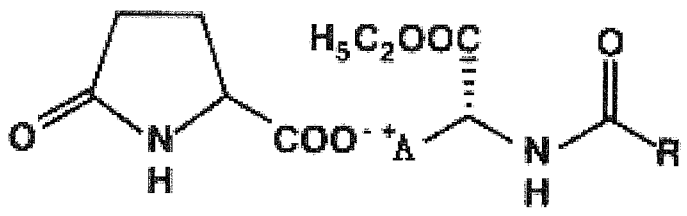
Published:

— *with international search report (Art. 21(3))*

— *with amended claims (Art. 19(1))*

WO 2010/050955 A1

(54) Title: BROAD SPECTRUM MICROBICIDAL AND SPERMICIDAL COMPOSITIONS AND METHODS



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(57) Abstract: The present invention relates to microbicidal and spermicidal compositions, methods and topical pharmaceutical compositions containing N-cocoyl amino acid pyrrolidone carboxylates (N-cocoyl amino acid pyrrolidone salts, CAPS) as active ingredients. The invention is for the prevention and control of sexually transmitted diseases and for the prevention of pregnancy. The microbicidal spectrum of the invention includes various STD pathogens, HIV viruses as well as gram positive and negative bacteria, and yeasts. Spermicidal barriers containing the compositions are also provided. The N-cocoyl amino acid pyrrolidone salts is represented by the formula: Wherein A is an amino acid residue and R is a C.sub.10 -C.sub.14 fatty acid residue.

BROAD SPECTRUM MICROBICIDAL AND SPERMICIDAL COMPOSITIONS AND METHODS

This application is based on, and claims benefit of, U.S. Provisional Application Serial No. 61/001,936, filed on November 7, 2007.

TECHNICAL FIELD

Provided is a composition containing N-cocoyl amino acid pyrrolidone salts for preventing or reducing the transmission of sexually transmitted diseases (STDs). Also provided are compositions and methods for preventing conception and/or reducing the risk of conception. Also provided are methods of inhibiting the activity of pathogens whose mode of transmission is nonsexual. Also provided are compositions effective in the inhibition of bacteria and fungi, which coexist with viruses or viral infections.

BACKGROUND

Sexually transmitted diseases (STDs) are among the most prevalent and communicable diseases, and continue to be a significant public health problem. It is estimated that more than 250 million people worldwide, and close to 3 million people in the United States, are infected annually by gonorrhea. Annual worldwide incidence of syphilis is estimated at 50 million people, with 400,000 in the United States annually needing treatment. The human immunodeficiency virus (HIV), resulting in fatal acquired immunodeficiency syndrome (AIDS), has spread rapidly in both homosexual and heterosexual groups. For many sexually transmitted infections, vaccines do not exist, and therapeutic agents are only partially effective, expensive, and difficult to distribute. In addition, female partners in many relationships do not control pregnancy or STI . One approach to the general control of STDs is the use of topically applied, female controlled microbicides that inactivate the relevant pathogens.

Transmission of HIV is often associated with the co-transmission of other viral and/or microbial pathogens. HIV may not be the sole agent responsible for AIDS (Duesberg, P. H. (1991) Proc. Natl. Acad. Sci. 88:1575-1579; Lemaitre, M., Guetard, D., Henin, Y., Montagnier, L. and Zerial, A. (1990). Res. Virol. 141:5-16). For this reason, antimicrobial agents, such as those described in this invention, with a broad spectrum of activities against viruses, bacteria, and yeasts may be of particular value in the prevention and treatment of Acquired Immune Deficiency Syndrome (AIDS).

It is thought that certain bacteria known to cause STDs may aid in HIV transmission. In persons who have been exposed to HIV, certain bacteria that cause STDs often fail to respond to therapies that are otherwise highly effective. HIV infection may help the spread of a bacterial STD that in turn helps to spread HIV. Sexually transmitted disease (STD) pathogens such as Chlamydeous, syphilis, genital herpes and gonorrhea that cause ulcerations of the genital skin seem to increase the risk of acquiring or transmitting HIV infection sexually.

It remains desirable to develop female controlled microbicides that inactivate STD pathogens.

SUMMARY

Provided is a microbicidal composition comprising 0.003 to 3 weight percent N-cocoyl amino acid pyrrolidone salts and 10 to 99.997 weight percent solvent. The solvent comprises a member selected from the group consisting of water, glycerin, glycerol-gelatin, ethanol, propylene glycol, polyethylene glycol, a water and glycerin mixture, a water and glycerol-gelatin mixture, a water and ethanol mixture, a water and propylene glycol mixture, and a water and polyethylene glycol mixture.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 is the formula for N-cocoyl amino acid pyrrolidone salts in which A is an amino acid residue and R is a C.sub.10 -C.sub.14 fatty acid residue.

DETAILED DESCRIPTION OF THE INVENTION

Provided are a composition and method for the prevention and control of sexually transmitted diseases and for the prevention of pregnancy. The composition and method also relate to treatment of viral infections. More particularly, provided is a method for inhibiting the development of diseases and infections caused by viruses and some sexually transmitted pathogens whose major mode of transmission is sexual. Certain embodiments relate to methods of inhibiting the activity of enveloped viruses and other pathogens whose mode of transmission is nonsexual. Certain embodiments are effective in the inhibition of bacteria and fungi, which coexist with viruses or viral infections. Certain embodiments relate to prevention or treatment for virus related diseases, particularly sexually transmitted diseases related to AIDS, and to diseases related to this and other opportunistic infections of the immune-compromised host.

In certain embodiments, the compositions described may be useful to inactivate viruses, including vaccinia, varicella, herpes zoster, cytomegalovirus, influenza, mumps and measles. In certain embodiments, the compositions prevent and/or treat certain skin diseases, such as, without limitation, ringworm and other fungi infectious skin diseases.

In certain embodiments, the compositions provides a broad spectrum and highly efficient microbicides useful for reducing the risk of transmission of STD-causing organisms to health care providers, laboratory personnel, or other persons who may come in contact with biological samples, specimens, or material.

In certain embodiments, the compositions can also be used as spermicides. In certain embodiments, spermicidal compositions can be used alone, or with other known spermicides, or with or incorporated into contraceptive devices such as condoms, sponges, vaginal inserts, contraceptive films, diaphragms, suppositories, contraceptive patches or sustained release devices. In certain embodiments, the compositions can be incorporated into douches or wipes.

The composition can also be used in or on animals as disinfecting or antiseptic agents.

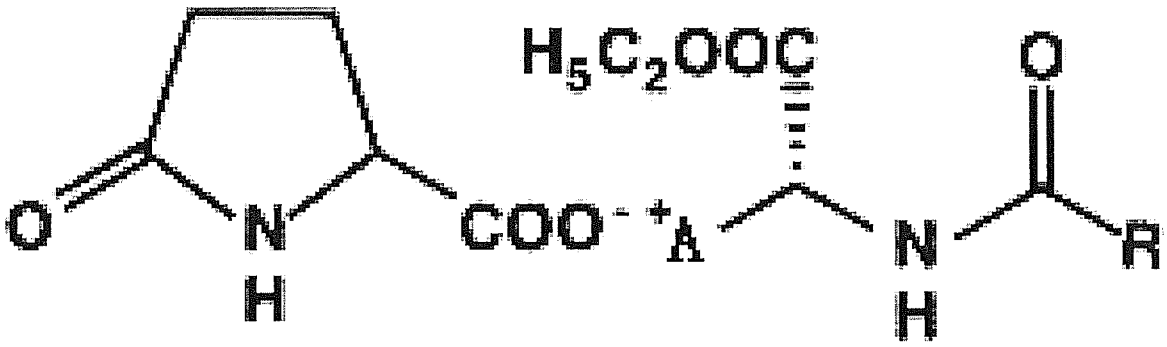
In certain embodiments, without limitation, the compositions can be formulated in the form of a gel, liquid, aerosol, mist, sponge, spray, foam, gel, cream, salve, jelly, suppository and film.

In certain embodiments, without limitation, the compositions can be applied to the external genital organs, vagina, anorectic region and rectum in different dosage forms with appropriate apparatus. In certain embodiments, without limitation, the composition as a gel could be applied to vagina or rectum through an applicator or syringe.

A microbicidal composition may be prepared comprising an amino acid derivative modified with a C.sub.10-14 alkyl group at the N-terminus. In certain embodiments, a N-cocoyl group is included. In certain embodiments, the amino acid residue bears a positive charge and the negative counter ion is an organic molecule. In certain embodiments, the negative counter ion is pyrrolidone salts. There are no halogen elements, such as chloride or bromide, in the structure of the amino acid compound.

A non-limiting example is an amino acid ethyl ester acylated at the N-terminus with a coconut oil fatty acid residue, and having DL-pyrrolidone carboxylic acid added thereto.

Without limitation, N-cocoyl amino acid pyrrolidone salts are represented by the formula:



wherein A is an amino acid residue and R is a C.sub.10 -C.sub.14 fatty acid residue. Without limitation, the amino acid residue could be from that of an arginine, lysine, histidine, homolysine, an unnatural amino acid residue bearing a positive charge, a di-peptide bearing a positive charge group, or a tri-peptide bearing a positive charge group.

In certain embodiments, without limitation, N-cocoyl Arginine pyrrolidone salts may be prepared as follows:

Solution A may be prepared by:

mixing 1.2 equivalent N, N'-Dicyclohexylcarbodiimide (DCC) with 1 equivalent myristic acid ($CH_3(CH_2)_{10}COOH$) in DMF and stir for 45 minute to 2 hours;

adding 1.2 equivalent N-Hydroxylsuccinimide (NHS) dissolved in CH_2Cl_2;

stirring until a dicyclohexane urea precipitate forms; and

filtering the reaction mixture to remove the dicyclohexane urea precipitate.

Solution B may be prepared by:

mixing L-Arginine ethyl ester dihydrochloride and 1 equivalent of NaOH at 0 degree C in DMF or CH_2Cl_2 for 30 minutes;

adding 1.3 equivalent of triethyl amine (Et_3N); and

mixing to form a homogeneous solution.

The amino salt may be prepared by:
mixing together solution A and solution B at room temperature for two hours;
removing most of the solvent by vacuum;
dissolving the product in methanol;
adding one equivalent of NaOH
extracting the product with a mixture of water and ether or a mixture of water and EtOAc;
drying the organic phase under vacuum;
re-precipitating the product by using solvent hexane, or water, or methanol, or a mixture of methanol and water;
re-dissolving the product in methanol;
adding one equivalent of DL-pyrrolidone carboxylic acid; and
removing the methanol under vacuum to obtain the final product.

In certain embodiments, the N-cocoyl amino acid pyrrolidone salts is contained in a proportion of from 0.003 to 3 weight percent.

N-cocoyl amino acid pyrrolidone salts are soluble in water, highly safe with respect to acute toxicity (LD.sub.50), have no substantial irritation to skin or ophthalmic mucosa, have good biodegradable characteristics, and can rapidly be decomposed in wastewaters.

In certain embodiments, microbicide compositions comprise natural plant polysaccharides. In certain embodiments, microbicide compositions comprise a natural plant polysaccharides to enhance antibacterial effects and to protect the epithelium tissues. Without limitation, plant polysaccharides comprise Aloe Vera polysaccharides. Without limitation, plant polysaccharides may comprise powder of freeze dried Aloe Vera gel.

In certain embodiments, microbicide compositions comprise an ampholytic surfactant. Without limitation, ampholytic surfactants comprise amine oxide such as alkyl dimethyl amine oxide.

In certain embodiments, microbicide compositions comprise conventional additive components in conventional amounts. Without limitation, conventional additive components comprise natural polysaccharides, natural plant acid and their salts, fragrance, colorant, flavor, plasticizer, stabilizing agent, emulsifier, moisturizer, and combinations thereof.

In certain embodiments, a liquid microbicide composition is prepared by adding and incorporating the active ingredients in proper amounts into an aqueous medium in an optional order in a conventional method. Without limitation, a liquid microbicide composition may be prepared by mixing the following ingredients by weight percentages in the provided ranges to obtain a homogeneous solution:

N-cocoyl amino acid pyrrolidone salts:	0.003 - 3 weight percent
Water :	10 - 99.995 weight percent

In certain embodiments, a microbicide composition is prepared comprising plant polysaccharides. Without limitation, a microbicide composition comprising plant polysaccharides may be prepared by mixing the following ingredients by weight percentages in the provided ranges to obtain a homogeneous solution:

Plant polysaccharides :	0.001 - 4 weight percent
N-cocoyl amino acid pyrrolidone salts:	0.003 - 3 weight percent
Water :	10 - 99.995 weight percent

EXAMPLE 1

In certain embodiments and without limitation, a microbicide composition is prepared as a gel. Without limitation, a gel microbicide composition may be prepared as described in the steps that follow.

A. Mix the following ingredients by weight percentages in the provided ranges and heat to 60°C -95°C with stirring to obtain a homogeneous solution.

Ampholytic surfactant:	0.0016 - 3 weight percent
Aloe Vera:	0.001 - 4 weight percent
Water	10 - 99.999 weight percent

B. Add Hydroxypropyl ethyl cellulose 0.010 - 5.0 weight percent to the 60° C - 95°C homogeneous solution from step A;

stir for more than 30 minutes;

stop heating and slowly reduce the temperature to 50° C.

C. Add N-cocoyl amino acid pyrrolidone salts 0.003 - 3.0 to the solution from step B;

stir until the temperature of the solution drops to room temperature to obtain a colorless transparent gel.

In certain embodiments, the gel microbicide composition may be stored or applied with a syringe. In certain embodiments, a syringe is filled with 3-5 ml of the gel microbicide composition; the filled syringe is sealed within an aluminum foil bag; and, at use, the user opens the foil bag and inserts the syringe into the vagina or the rectum to apply the material.

Table 1 lists 37 formulations prepared following the above procedures to make liquid or gel preparations. Abbreviations used in Table 1 and hereafter are: Freeze dried Aloe Vera gel powder (AV); Alkyldimethylamine oxide (C1); Water (H_2O); N-cocoyl amino acid pyrrolidone salts (CAPS); Hydroxypropyl methyl cellulose (HP).

TABLE 1. FORMULATIONS OF MICROBICIDES

Units are weight percent

Formula	AV	C1	H_2O	CAPS	HP
1	1.00	1.00	94.50	1.50	2.00
2	0.00	0.00	99.40	0.60	0.00
3	0.25	0.00	99.08	0.67	0.00
4	0.0013	0.002	99.995	0.0034	0.01
5	0.25	0.33	96.75	0.67	2.00
6	0.006	0.008	99.92	0.017	0.05
7	0.013	0.017	99.84	0.034	0.10
8	0.003	0.003	99.9	0.007	0.02
9	0.005	0.007	99.9	0.013	0.04
10	0.025	0.033	99.7	0.067	0.200
11	0.003	0.004	99.96	0.008	0.025
12	1.50	0.50	50.00	1.00	0.50
13	2.00	0.0016	90.0	3.00	5.0
14	0.20	0.25	95.50	0.50	0.30
15	0.00	0.00	10.00	0.003	0.00
16	0.00	0.00	99.995	0.003	0.00
17	0.00	0.00	98.50	1.5	0.00
18	0.00	0.00	97.00	3.0	0.00
19	4.0	0.00	96.00	0.003	0.00
20	1.8	0.00	97.00	1.2	0.00
21	0.001	0.00	97.00	3.0	0.00
22	0.00	3.0	97.00	0.003	0.00
23	0.00	1.7	97.00	1.3	0.00
24	0.00	0.0016	97.00	3.0	0.00
25	0.00	0.00	95.00	0.003	5.0
26	0.00	0.00	96.30	1.4	2.3
27	0.00	0.00	96.99	3.0	0.01
28	0.001	0.00	97.00	0.003	0.00
29	2.0	1.5	94.90	1.6	0.00
30	4.0	0.0016	93.00	3.0	0.00
31	0.00	3.0	96.99	0.0033	0.01
32	0.00	1.3	94.50	1.7	2.5
33	0.00	0.00	92.00	3.0	5.0
34	0.001	0.00	99.995	0.0033	0.01
35	2.2	0.00	93.20	1.8	2.8
36	4.0	0.00	88.00	3.0	5.0
37	0.0	0.0	99.5	0.5	0.0

EXAMPLE 2

In certain embodiments and without limitation, a microbicide composition is prepared as a suppository. In certain embodiments, a microbicide composition is prepared as a suppository comprised of the ingredients in the quantities listed in TABLE 2 by the following method.

Dissolving N-cocoyl amino acid pyrrolidone salts (CAPS), sodium phytate and freeze dried Aloe Vera powder in water;

adding glycerin;

mixing until a homogeneous solution is formed;

adding Glycerogelatin to the homogeneous solution;

mixing at 45°C until homogeneous;

molding the homogeneous solution into a plurality of 4 gram suppositories by injecting the homogeneous solution into suppository molds and cooling the injected homogeneous solution;

ejecting the suppositories for packing.

TABLE 2: MICROBICIDE COMPOSITION OF SUPPOSITORIES

CAPS	4.8 g
Alkyldimethylamine oxide	2.4 g
Aloe Vera powder	1.8 g
Sodium phytate	1.8 g
Glycerogelatin	900.0 g
Glycerin	20.0 g
Water	80.0 g

EXAMPLE 3

In certain embodiments and without limitation, a microbicide composition is prepared as a spray or mist. In certain embodiments, a microbicide composition is

prepared as a spray comprised of the ingredients in the quantities listed in TABLE 3 by the following method.

Dissolving N-cocoyl amino acid pyrrolidone salts (CAPS) in water to form a solution;

slowly adding isopropylene to the solution;

mixing the solution until homogeneous; and

packaging the homogenous solution in container comprising a sprayer.

In certain embodiments, a microbicide composition is prepared as a mist comprised of the ingredients in the quantities listed in TABLE 3 by the following method.

Dissolving N-cocoyl amino acid pyrrolidone salts (CAPS) and Alkyldimethyl amine oxide in water to form a solution;

slowly adding isopropylene to the solution; and

mixing the solution until homogeneous;

adding Dimethyl Ether; and

packaging the homogenous solution in container comprising a sprayer.

TABLE 3: MICROBICIDE COMPOSITIONS OF MIST AND SPRAY

Ingredients	Spray	Mist
CAPS	0.6 kg	0.8 g
Alkyldimethyl amine oxide	0.0 kg	0.4 g
Isopropylene	5.0 kg	5.0 g
Dimethyl Ether	0.0 kg	20.0 g
Water	94.4 kg	80.0 g

EXAMPLE 4

In certain embodiments and without limitation, a microbicide composition is prepared as a film. In certain embodiments, a microbicide composition is prepared as a file comprised of the ingredients in the quantities listed in TABLE 4 by the following method.

Triturating Gelatin with Hydroxyethyl cellulose and glycerin and mixing thoroughly to form a slurry;

dissolving CAPS in warm water to obtain a homogeneous solution;

adding the solution to the slurry to form a mixture;

heating the mixture at 40°C and mixing until the cellulose and the gelatin are completely hydrated;

casting a film of about 2 mm thick by pouring the mixture onto a polyethylene sheet and cooling the mixture; and

cutting the film.

In certain embodiments and without limitation, the film is cut into 3.9X3.9 cm squares.

TABLE 4: MICROBICIDE COMPOSITION OF FILM

CAPS	0.80 kg
Aloe Vera	0.16 kg
Gelatin	6.20 kg
Hydroxyethyl cellulose	0.50 kg
Glycerin	31.00 kg
Water	25.50 kg

TESTING DATA

TEST ON UREAPLASMA UREALYTICUM

The microbicide was tested on Ureaplasma Urealyticum. The details of the test were as follows.

The testing strain was a Ureaplasma Urealyticum clinical strain. The culture medium was a high efficiency Ureaplasma culture medium. The testing sample was the microbicide solution prepared per Example 1, formulation 3.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard). The tests followed the "Standard" with consideration of the characteristics of Ureaplasma Urealyticum. The testing temperature was in the range of 20°C - 25°C. The sample dilutions ratios tested were: 1:100; 1:200; 1:500; and 1:1000. The concentration of Ureaplasma Urealyticum was 10^7. The treatment used a Ureaplasma Urealyticum suspension and sample diluted in a ratio of 1: 9. The reaction times were 1 minute, 3 minutes, and 5 minutes. Diluting methods were used to remove the residue of the sample. Tests were repeated two times.

The results of the testing the samples on Ureaplasma Urealyticum are shown in TABLE 5. In TABLE 5, a "+" indicates that Ureaplasma Urealyticum growth was observed, and a "-" indicates that no Ureaplasma Urealyticum growth was observed.

TABLE 5. THE EFFECTS OF TESTING SAMPLE ON UREAPLASMA UREALYTICUM

Time	Dilution of Sample			Control
(min.)	1:100	1:200	1:500	
1	-	+	+	+
3	-	-	+	+
5	-	-	+	+

The testing results showed that the testing sample diluted for 100 times and reacted with a Ureaplasma Urealyticum suspension for 1 minute killed Ureaplasma Urealyticum.

TEST ON NEISSERIA GONORRHOEAE

The microbicide was tested on Neisseria Gonorrhoeae. The details of the test were as follows.

The testing strain was a Neisseria Gonorrhoeae clinical strain. The culture medium was a 10% blood dish. The testing sample was the microbicide solution prepared per Example 1, formulation 3.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard). The tests followed the "Standard" with consideration of the characteristics of Neisseria gonorrhoeae. The testing temperature was in the range of 25°C - 28°C. The sample dilutions ratios tested were: 1:25; 1:50; and 1:100. The concentration of Neisseria gonorrhoeae was 10^7. The treatment used a Neisseria gonorrhoeae suspension and sample diluted in a ratio of 1:1. The reaction times were 1 minute, 3 minutes, and 5 minutes.

The results of the testing the samples on Neisseria Gonorrhoeae are shown in TABLE 6. In TABLE 6, a "+" indicates that Neisseria Gonorrhoeae growth was observed, and a "-" indicates that no Neisseria Gonorrhoeae growth was observed.

TABLE 6. EFFECTS OF TESTING SAMPLE ON NEISSERIA GONORRHOEAE

React time (min.)	Dilution of Sample			Control
	1:25	1:50	1:100	
1	-	-	-	+
3	-	-	-	+
5	-	-	-	+

The testing results showed that the testing sample diluted for 100 times and reacted with a Neisseria gonorrhoeae suspension for 1 minute killed Neisseria gonorrhoeae.

TEST ON TRICHOMONAS VAGINALIS

The microbicide was tested on Trichomonas vaginalis. The details of the test were as follows.

The testing strain was a clinically isolated Trichomonas vaginalis strain. The culture medium was a liver infusion medium. The testing sample was the microbicide solution prepared per Example 1, formulation 3.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard).

The results of the testing the samples on Trichomonas vaginalis are shown in TABLE 7. In TABLE 7, a "+" indicates that Trichomonas vaginalis was active, and a "-" indicates that Trichomonas vaginalis were not active and were dissolved.

TABLE 7. EFFECTS OF TESTING SAMPLE ON TRICHOMONAS VAGINALIS

time	Dilution of sample			Control
(minutes)	1:50	1:100	1:200	1:50
1	-	-	+	+
3	-	-	+	+
5	-	-	+	+

The testing results showed that the testing sample diluted for 100 times and reacted with Trichomonas vaginalis suspension for 1 minute killed Trichomonas vaginalis.

TEST ON HIV-1, TEST ONE

The microbicide was tested on HIV-1. The details of the test were as follows.

The testing strain was Human Immunodeficiency Virus: HIV-1 IIIB strain. The cell was an MT 4 cell. The testing sample was the microbicide solution prepared per Example 1, formulation 2.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard).

Testing samples were diluted to 1:20; 1:40; 1:80 and 1:160 solutions with distilled water. For each testing sample, 0.9 ml of testing sample was mixed to 0.1 ml HIV suspension solution. The resulting mixtures were each reacted for 1 min.; 3 min.; 5 min. at room temperature. 0.1 ml of each mixture was diluted with culture fluid to 1:1000. The virus concentrations were titrated (96 well plate, each well contains 0.1 ml of 50000/ml MT4 cell). The titrated mixtures were incubated for 7 days at 37°C and 5% CO2. 7) 0.1 ml of fresh culture fluid was added to each titrated mixture at the third day. Observation for cell growth was made everyday. Existence of Virus with CPE was evaluated.

The experimental groups were: Testing group; Drug (sample) removed control group (1000 times diluted Virus + 1000 times diluted sample); Normal cell control group; and Virus control group (Virus + culture fluid).

The results of the testing the samples on HIV-1 are as follows. HIV-1 titration was 10^7 $TCID_{50}$. The "drug removed control group" showed that the dilution could stop the effect of sample to the virus. In the testing samples wherein 1:20 and 1:40 dilutions were reacted with HIV-1 for 1 minute, titrated and cultured with MT4 cell, no cell abnormality were observed. In the testing sample wherein 1:80 dilutions were reacted with HIV-1 for 3 minutes, titrated and cultured with MT4 cell, no cell abnormality were observed. In the testing sample wherein 1:160 dilutions were reacted with HIV-1 for 5 minutes, titrated and cultured with MT4 cell, no cell abnormality were observed.

In testing samples wherein 1:20 (Example 1, formulation 15) and 1:40 solutions were reacted with HIV-1 suspension for 1 minute; wherein 1:80 solution were reacted with HIV-1 suspension for 3 minutes; and wherein 1:160 solution were reacted with HIV-1 suspension for 5 minutes, all the kill rates were 100%.

TEST ON CANDIDA ALBICANS

The microbicide was tested on Candida albicans. The details of the test were as follows.

The testing strain was Candida albicans ATCC 10231, 4th generation. The culture medium was a 10% blood dish. The testing sample was the microbicide solution prepared per example 1, formulation 2.

The testing method used a nutrient broth dilution method at a temperature of 21 +/- 1°C .

The results of the testing the samples on Candida albicans are shown in TABLE 8. In TABLE 8, a "+" indicates that bacterial growth was observed, and a "-" indicates that no bacterial growth was observed.

TABLE 8. THE MBC OF TESTING SAMPLE TO CANDIDA ALBICANS

test	MBC of different dilution of sample									Positive control	Control Negative		Bacterial concentration (CFU/ml)
	1	2	4	8	16	32	64	128	256				
1	-	-	-	-	-	+	+	+	+	+	-	-	2.13×10^6
2	-	-	-	-	-	+	+	+	+				1.89×10^6

To Candida albicans the MBC of testing sample was the 1:16 times dilution at 2 minutes.

TEST ON HIV-1, TEST TWO

The microbicide was tested on HIV-1. The details of the test were as follows.

The testing strain was Human Immunodeficiency Virus: HIV-1 IIIB strain. The cell was an MT 4 cell. The testing sample was the microbicide solution prepared per Example 1, formulation 5.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard).

HIV Virus deactivation Tests, Page: 38. Testing samples were diluted to 1:20; 1:40; and 1:80 solutions with distilled water. For each testing sample, 0.9 ml of testing sample was mixed to 0.1 ml HIV suspension solution. The resulting mixtures were each reacted for 1 minute; 3 minutes; 5 minutes; and 10 minutes at room temperature. 0.1 ml of each mixture was diluted with culture fluid to 1:1000. The virus concentrations were titrated (96 well plate, each well contains 0.1 ml of 50000/ml MT4 cell). The titrated mixtures were incubated for 7 days at 37°C and 5% CO2. 0.1 ml of fresh culture fluid was added to each titrated mixture at the third day. Observation for cell growth was made everyday. Existence of Virus with CPE was evaluated.

The experimental groups were: Testing group; Drug (sample) removed control group (1000 times diluted Virus + 1000 times diluted sample); Normal cell control group; and Virus control group (Virus + culture fluid).

The results of the testing the samples on HIV-1 are as follows. HIV-1 titration was 10^7 $TCID_{50}$. The "drug removed control group" showed that the dilution could stop the effect of sample to the virus. In the testing samples wherein 1:20 and 1:40 dilutions were reacted with HIV-1 for 1 minute, titrated and cultured with MT4 cell, no cell abnormality were observed. In the testing sample wherein 1:80 dilutions were reacted

with HIV-1 for 3 minutes, titrated and cultured with MT4 cell, no cell abnormality were observed.

In testing samples wherein 1:20 (Example 1, formulation 7) and 1:40 (Example 1, formulation 6) solutions were reacted with HIV-1 suspension for 1 minute; and wherein 1:80 solution were reacted with HIV-1 suspension for 3 minutes, the kill rates were 100%.

TEST ON TREPONEMA PALLIDUM

The microbicide was tested on Treponema Pallidum. The details of the test were as follows.

The testing strain was Treponema Pallidum, Nichols strain. The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the "Guidelines for Pre-clinical and Clinical Research of New Drugs" in the 1993 Edition of The Experimental Methods and Procedures for Anti-Treponema Pallidum. The testing temperature was in the range of 20°C - 25°C. The sample dilutions ratios tested were: 1:5; 1:10; 1:20; 1:40; 1:80; and 1:100. 5) Samples were diluted with distilled water. Treponema Pallidum Suspension: each field ≥30 Treponema Pallidum. The reaction times were 1 minute, 3 minutes, 5 minutes, and 10 minutes. No terminating agent was employed in the tests. The deactivating effects for Treponema Pallidum were recorded at different time intervals. The tests were repeated two times.

The results of the testing the samples on Treponema Pallidum are shown in TABLE 9. In TABLE 9, a "+" indicates that Treponema Pallidum was not deactivated, and a "-" indicates that Treponema Pallidum was deactivated.

TABLE 9. THE DEACTIVATING EFFECT OF TESTING SAMPLE TO TREPONEMA PALLIDUM

Treponema Pallidum	React time (min.)	Sample dilution					
		1:5	1:10	1:20	1:40	1:80	1:100
Nichols strain	1	-	-	+	+	+	+
Nichols strain	3	-	-	-	+	+	+
Nichols strain	5	-	-	-	-	+	+
Nichols strain	10	-	-	-	-	-	+

Testing sample diluted at 1:10 (Example 1, formulation 10) deactivated Treponema Pallidum within 1 minute.

Testing sample diluted at 1:20 (Example 1, formulation 7) deactivated Treponema Pallidum within 3 minutes.

Testing sample diluted at 1:40 (Example 1, formulation 6) deactivated Treponema Pallidum within 5 minutes.

Testing sample diluted at 1:80 (Example 1, formulation 11) deactivated Treponema Pallidum within 10 minutes.

TEST ON Chlamydia trachomatis

The microbicide was tested on Chlamydia trachomatis. The details of the test were as follows.

The testing strain was Chlamydia trachomatis, E Bour type,10th generation, provided by CDC of USA. The cells were McCoy cells. The culture medium was RPMI-1640 culture medium. The testing sample was the microbicide gel prepared per example 1, formulation 5. Diluting neutralizing methods were employed, the diluent was 1640 medium with 10% bovine serum.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard). The tests followed the "Standard" with consideration of the characteristics of

Chlamydia trachomatis. The testing temperature was 25°C. The microbicide concentrations tested were: 1:20; 1:50; 1:100; 1:200; and 1:500. The concentration of Chlamydia trachomatis suspension was 10^5. The reaction times were 1 minute, 3 minutes, 5 minutes, and 10 minutes. Diluting methods were used to remove the residue of sample.

The results of the testing the samples on Neisseria Gonorrhoeae are shown in TABLE 10. In TABLE 10, a "+" indicates that there were Chlamydia trachomatis inclusion in the cell, and a "-" indicates that no there were no Chlamydia trachomatis inclusion in the cell. Chlamydia trachomatis growth was observed in the controls.

TABLE 10. THE KILLING EFFECTS OF TESTING SAMPLE ON CHLAMYDIA TRACHOMATIS

strain	time (min.)	Dilution of Testing sample					control
		1:20	1:50	1:100	1:200	1:500	
E Bour strain	1	-	-	+	+	+	+
	3	-	-	-	+	+	+
	5	-	-	-	+	+	+
	10	-	-	-	-	+	+

Testing sample diluted at 1:50 (example 1, formulation 9) and reacted with Chlamydia trachomatis suspension for 1 minute killed Chlamydia trachomatis.

TEST ON UREAPLASMA UREALYTICUM

The microbicide was tested on Ureaplasma Urealyticum. The details of the test were as follows.

The testing strain was Ureaplasma Urealyticum, international standard strain, 6^{th} generation. The culture medium was High efficiency Ureaplasma culture medium. The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard). The tests followed the "Standard" with consideration of the characteristics of Ureaplasma Urealyticum. The testing temperature was 25°C- 28°C. The microbicide concentrations tested were: 1:100; 1:200; 1:500; and 1:1000. The concentration of Ureaplasma Urealyticum was 10^7. The reaction times were 1 minute, 3 minutes, 5 minutes, and 10 minutes. The treatment used a Ureaplasma Urealyticum suspension in a diluted sample at a ratio of 1:9. Diluting methods were used to remove the residue of drug. The test was repeated two times.

The results of the testing the samples on Ureaplasma Urealyticum are shown in TABLE 11. In TABLE 11, a "+" indicates that Ureaplasma Urealyticum growth was observed, and a "-" indicates that no Ureaplasma Urealyticum growth was observed. Ureaplasma Urealyticum growth was observed in the controls.

TABLE 11. THE EFFECTS OF TESTING SAMPLE ON UREAPLASMA UREALYTICUM

time	Dilutions of Testing Sample				control
(min.)	1:100	1:200	1:500	1:1000	
1	-	-	+	+	+
3	-	-	+	+	+
5	-	-	+	+	+
10	-	-	+	+	+

The testing results showed that a testing sample diluted 200 times (Example 1, formulation 4) and reacted with Ureaplasma Urealyticum suspension for 1 minute, killed Ureaplasma Urealyticum.

TEST ON NEISSERIA GONORRHOEAE

The microbicide was tested on Neisseria gonorrhoeae. The details of the test were as follows.

The testing strain was Neisseria gonorrhoeae, Standard strain G, provided by WHO. The culture medium was 10% blood dish. The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard). The tests followed the "Standard" with consideration of the characteristics of Neisseria gonorrhoeae. The testing temperature was 25°C- 28°C. The microbicide concentrations tested were: 1:25; 1:50; 1: 100; 1:250; and 1:500. The concentration of Neisseria gonorrhoeae suspension was 10^7. The treatment times were 1 minute, 3 minutes, 5 minutes, and 10 minutes. The treatment used a Neisseria gonorrhoeae suspension diluted in 1:1 ratio. Diluting methods were used to remove the residue of drug. The test was repeated three times.

The results of the testing the samples on Neisseria gonorrhoeae are shown in TABLE 12. In TABLE 12, a "+" indicates that growth of Neisseria gonorrhoeae was observed, and a "-" indicates that no growth of Neisseria gonorrhoeae was observed.

TABLE 12. THE KILLING EFFECTS OF TESTING SAMPLE TO NEISSERIA GONORRHOEAE

React time	Dilutions of Testing sample				control
(min.)	1:50	1:100	1:200	1:500	
1	-	-	+	+	+
3	-	-	+	+	+
5	-	-	+	+	+
10	-	-	-	+	+

The testing results showed that a testing sample diluted for 100 times (example 1, formulation 8) and reacted with Neisseria gonorrhoeae suspension for 1 minute, killed Neisseria gonorrhoeae.

TEST ON TRICHOMONAS VAGINALIS

The microbicide was tested on Trichomonas vaginalis. The details of the test were as follows.

The testing strain was clinically isolated Trichomonas vaginalis. The culture medium was liver infusion medium. The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard). The tests followed the "Standard" with consideration of the characteristics of Trichomonas vaginalis.

The testing temperature was 20°C- 25°C. The microbicide concentrations tested were: 1:50; 1: 100; 1:200. The concentration of Trichomonas vaginalis suspension was 10^5/ml. The treatment times were 1 minute, 3 minutes, 5 minutes, and 10 minutes. The treatment used a Trichomonas vaginalis suspension diluted in 1:1 ratio. Diluting methods were used to remove the residue of drug. The test was repeated two times.

The results of the testing the samples on Trichomonas vaginalis are shown in TABLE 13. In TABLE 13, a "+" indicates active Trichomonas vaginalis, a "±" indicates most Trichomonas vaginalis were killed, a few active were left, a "-" indicates that Trichomonas vaginalis were not active and were dissolved.

TABLE 13. THE KILLING EFFECTS OF TESTING SAMPLETO TRICHOMONAS VAGINALIS

React time (min.)	Dilutions of Testing sample			control
	1:100	1:200	1:400	
1	-	±	+	+
3	-	±	+	+
5	-	±	+	+

10	-	-	+	+

The testing results showed that a testing sample diluted for 100 times (example 1, formulation 8) and reacted with Trichomonas vaginalis suspension for 1 minute, killed Trichomonas vaginalis.

TEST ON HERPES SIMPLEX VIRUS

The microbicide was tested on Herpes Simplex Virus. The details of the test were as follows.

The testing strain was Herpes Simplex type II strain, standard international strain, provided by the Drug and bio-product office of the Health Department of China. The culture medium 1640 culture medium. The cell was Vero cell strain, introduced by the Cell biology Institute of Shanghai. The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the standards issued by The Health Department of China in the 2002 Edition of Technical Standards for Disinfection (The Standard). The tests followed the "Standard" with consideration of the characteristics of Herpes Simplex.

The virus suspension was 5x10^4/ml. The microbicide concentrations tested were 1:20; 1:40; and 1:50. The reaction times were 1 minute, 3 minutes, 5 minutes, and 10 minutes. The testing temperature was 20°C- 25°C. Diluting methods were used to remove the residue of drug (2% bovine serum culture medium as diluents). The test was repeated two times.

The results of the testing the samples on Herpes Simplex are shown in TABLE 14. In TABLE 14, a "+" indicates virus growth observed, infected cell > 50%; a "±" indicates virus growth observed, infected cell < 10%; a "-" indicates non virus growth.

TABLE 14. THE EFFECTS OF TESTING EXAMPLE ON HERPES SIMPLEX VIRUS

React time (min.)	Dilutions of Testing sample			Control
	1:20	1:40	1:50	
1	-	±	+	+
3	-	±	+	+
5	-	±	+	+
10	-	±	+	+

The testing results showed that in a testing sample diluted for 20 times (Example 1, formulation 7) and reacted with Herpes Simplex Virus, Herpes Simplex Virus was killed with 20 times dilution of the sample in 1 minute.

TEST ON HERPES SIMPLEX VIRUS

The microbicide was tested on Staphylococcus aureus. The details of the test were as follows.

The testing strain was Staphylococcus aureus ATCC6538, 5-6th generations, provided by the reserve center of China microbial reservation committee. The testing medium carrier was sterilized filter paper. The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the《Technical Standard for Disinfection》(2002 Edition) and GB15979-2002《Technical Standard for Testing Disinfection Product》"Procedures for testing bactericide effect".

At 20±1°C temperature, three repeated tests showed that for a testing sample reacted with Staphylococcus aureus for 1 minute, the average bactericide rate was 95.70%.

TABLE 15. THE BACTERICIDE EFFECT OF TESTING SAMPLE TO STAPHYLOCOCCUS AUREUS

Time (min.)	Bactericide rates of three tests			Average bactericide rate (%)	Control (CFU/pc.)
	First test	Second test	Third test		
1	92.13	96.87	98.10	95.70	1.28×10^4
3	99.64	100.00	99.58	99.74	1.25×10^4
5	99.72	100.00	99.95	99.89	1.23×10^4

Testing sample (Example 1, formulation 5) reacted with Staphylococcus aureus for 1 minute; the average bactericide rate was 95.70%.

TEST ON CANDIDA ALBICANS

The microbicide was tested on Candida albicans. The details of the test were as follows.

The testing strain was Candida albicans, ATCC 10231, the 5-6 generation as provided by the "Reserve Center of Chinese Microbial Reservation Committee". The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the《Technical Standard for Disinfection》(2002 Edition) and GB15979-2002《Technical Standard for Testing Disinfection Product》"Procedures for testing bactericide effect".

The bactericide effect of the testing sample to Candida albicans ATCCI0231 at 20±1°C temperature with three repeated tests showed that for testing material reacted with Candida albicans ATCCI0231 for 1 minute, the average bactericide rate was 95.47%.

Table 16. The bactericide effect of Testing Sample to Candida albicans

Time (min.)	Bactericide rates of three tests			Average bactericide rate (%)	Control (CFU/pc.)
	First test	Second test	Third test		
1	97.95	93.81	94.65	95.47	3.07 $\times 10^4$
3	99.66	99.96	99.30	99.53	2.99 $\times 10^4$
5	99.91	99.88	99.75	99.84	2.96 $\times 10^4$

For a testing sample reacted with Candida albicans for 1 minute, the average bactericide rate was 95.47%. The Microbicidal Gel has bactericide effect on Candida albicans.

TEST ON ESCHETICHIA COLI

The microbicide was tested on Eschetichia coli. The details of the test were as follows.

The testing strain was Eschetichia coli 8099, the 5-6 generation as provided by the "Reserve Center of Chinese Microbial Reservation Committee". The testing sample was the microbicide gel prepared per example 1, formulation 5.

The testing methods and standards followed the《Technical Standard for Disinfection》(2002 Edition) and GB15979-2002《Technical Standard for Testing Disinfection Product》"Procedures for testing bactericide effect".

The bactericide effect of the testing sample to Eschetichia coli at 20±1°C temperature with three repeated tests showed that for sample gel reacted with Eschetichia coli for 1 minute, the average bactericide rate was 99.97%.

TABLE 17. THE BACTERICIDE EFFECT OF TESTING SAMPLE TO ESCHETICHIA COLI

Time (min.)	Bactericide rates of three tests			Average bactericide rate (%)	Control (CFU/pc.)	
	First test	Second test	Third test			
1	99.96	100.00	99.95	99.97	1.30	$\times 10^4$
3	100.00	100.00	100.00	100.00	1.22	$\times 10^4$
5	100.00	100.00	100.00	100.00	1.20	$\times 10^4$

For a testing sample reacted with Eschetichia coli for 1 minute, the average bactericide rate was 99.97%.

TEST ON HUMAN SPERM, TEST ONE

The microbicide was tested on human sperm. The details of the test were as follows.

The human sperm was normal sperm obtained from three healthy adults. The testing sample was the microbicide solution prepared per example 1, formulation 37.

The method followed was: mix the diluted samples with equal volume of fresh sperm (0.5 ml sample and 0.5 ml sperm); determine the minimum concentration and time of demobilize all the sperms; use saline as negative control; repeat the tests for three times.

The undiluted solution reacted with sperms for 30 seconds. The survival rate of sperms was zero. Results are shown in Tables 18,19 and 20

Table 18. Testing Results on Sperm Sample 1:

Time		Sample concentration		
		solution	1:5	1:10
30 sec	Sperm count (million/ml)	23	23	23
	Active rate (%)	0	50	60
	Sperm count (million/ml)	23	22	22

	Active rate (%)	0	45	45
2 min	Sperm count (million/ml)	22	22	22
	Active rate (%)	0	42	45
3 min	Sperm count (million/ml)	23	23	23
	Active rate (%)	0	40	40

	Before test	Control / saline
Sperm count (million/ml)	45	23
Active rate (%)	82	72

Table 19. Testing Results on Sperm Sample 2:

Time		Sample concentration		
		solution	1:5	1:10
30 sec	Sperm count (million/ml)	13.5	13.5	13.5
	Active rate (%)	0	50	50
1 min	Sperm count (million/ml)	13.5	13.4	13.4
	Active rate (%)	0	50	50
2 min	Sperm count (million/ml)	13.3	13.4	13.5
	Active rate (%)	0	45	50
3 min	Sperm count (million/ml)	13.2	13.2	13.3
	Active rate (%)	0	40	50

	Before test	Control / saline
Sperm count(million/ml)	25	13.5
Active rate (%)	62	62

Table 20. Testing Results on Sperm Sample 3

Time		Sample concentration		
		solution	1:5	1:10
30 sec	Sperm count (million/ml)	29.7	29.5	29.4
	Active rate (%)	0	60	61
1 min	Sperm count (million/ml)	29.6	29.0	29.0
	Active rate (%)	0	58	59
2 min	Sperm count (million/ml)	29.0	29.0	29.0
	Active rate (%)	0	58	58
3 min	Sperm count (million/ml)	28.5	28.5	28.5
	Active rate (%)	0	58	58

	Before test	Control / saline
Sperm count (million/ml)	58	30
Active rate (%)	75	73

The undiluted solution reacted with equal volume sperm for 30 seconds, 1 minute, 2 minutes, 3 minutes. The sperm survival rate was zero. The spermicidal rate of testing microbicide solution was 100% in 30 seconds (in vitro). For saline reacted with equal volume sperm, the sperm count and active rate was not influenced.

The spermicidal rate of testing sample was 100% in 30 seconds (in vitro).

TEST ON HUMAN SPERM, TEST TWO

The microbicide was tested on human sperm. The details of the test were as follows.

The human sperm was normal sperm obtained from three healthy adults. The testing sample was the microbicide gel prepared per example 1, formulation 5.

The method followed was: dilute the Testing sample to different concentrations (1:10, 1:5, 1:1); mix the diluted samples with equal volume fresh sperm (0.5 ml sample and 0.5 ml sperm); determine the minimum concentration and time of demobilize all the sperms; use saline as negative control; repeat the tests for three times.

The undiluted gel reacted with sperm for 30 seconds, the survive rate of sperm was zero.

The gel was diluted at 1:1, 1:5 and reacted with equal volume sperm for 30 seconds, 1 minute, 2 minutes, and 3 minutes. The sperm survival rate was zero.

The gel diluted at 1:10 and reacted with equal volume sperm for 30 seconds. The sperm survive rate was 10%.

The 1:10 gel dilution reacted with sperm for 1 minute, 2 minutes, and 3 minutes, respectively; the sperm survive rate was zero.

The 1:10 dilution deactivated all the sperm in 60 seconds. The sperm survival rate was zero.

Results are shown in Tables 21, 22, and 23.

TABLE 21. TESTING RESULTS ON SPERM SAMPLE 1

Time		Sample concentration			
		gel	1:1	1:5	1:10
30 sec	Sperm count(million/ml)	12.3	11.4	11.0	10.1
	Active rate (%)	0	0	0	10
1 min	Sperm count(million/ml)	——	10.0	9.8	9.7
	Active rate (%)	——	0	0	0
2 min	Sperm count(million/ml)	——	9.6	9.8	9.5
	Active rate (%)	——	0	0	0
3 min	Sperm count(million/ml)	——	9.5	9.4	9.5
	Active rate (%)	——	0	0	0
		Before test		Control / saline	
Sperm count (million/ml)		20.1		12.5	
Active rate (%)		65		64.5	

Table 22. Testing Results on Sperm Sample 2 :

Time		Sample concentration			
		gel	1:1	1:5	1:10
30 sec	Sperm count(million/ml)	20.0	19.2	19.0	18.9
	Active rate (%)	0	0	0	5
1 min	Sperm count(million/ml)	——	18.4	19.0	18.7
	Active rate (%)	——	0	0	0
2 min	Sperm count(million/ml)	——	18.6	18.6	18.5
	Active rate (%)	——	0	0	0
3 min	Sperm count(million/ml)	——	18.0	18.0	17.9
	Active rate (%)	——	0	0	0
		Before test		Control / saline	
Sperm count(million/ml)		36.3		20.9	

Active rate (%)	70	69

Table 23. Testing Results on Sperm Sample 3 :

Time		Sample concentration			
		gel	1:1	1:5	1:10
30 sec	Sperm count(million/ml)	14.3	14.2	14.2	14.0
	Active rate (%)	0	0	0	15
1 min	Sperm count(million/ml)	——	13.8	13.7	14.0
	Active rate (%)	——	0	0	0
2 min	Sperm count(million/ml)	——	13.6	13.5	13.3
	Active rate (%)	——	0	0	0
3 min	Sperm count(million/ml)	——	13.2	13.1	13.1
	Active rate (%)	——	0	0	0
		Before test		Control / saline	
Sperm count (million/ml)		23.2		15.1	
Active rate (%)		75		75	

The spermicidal rate in testing microbicide gel was 100% in 30 seconds (in vitro), the minimum effective dilution was 1:5.

The spermicidal rate in testing microbicidal gel was 100% in 60 seconds (in vitro), the minimum effective dilution was 1:10.

CLAIMS:

I claim:

1. A composition comprising:
0.003 to 3 weight percent N-cocoyl amino acid pyrrolidone salts; and
10 to 99.997 weight percent solvent, wherein said solvent comprises a member selected from the group consisting of water, glycerin, glycerol-gelatin, ethanol, propylene glycol, polyethylene glycol, a water and glycerin mixture, a water and glycerol-gelatin mixture, a water and ethanol mixture, a water and propylene glycol mixture, and a water and polyethylene glycol mixture; and
wherein said composition is adapted for microbicidal use.

2. The composition of claim 1, further comprising a plant polysaccharide.

3. The composition of claim 1, wherein the amino acid group of the N-cocoyl amino acid pyrrolidone salts, comprises a member selected from the group consisting of arginine, lysine, histidine, homolysine, unnatural amino acid residue bearing a positive charge, di-peptide bearing a positive charge group, and tri-peptide bearing a positive charge group.

4. The composition of claim 1, wherein said water comprises de-ionized water, distilled water, or purified water.

5. The composition of claim 2, wherein said plant polysaccharide comprises Aloe Vera polysaccharides.

6. The composition of claim 1, wherein the microbicidal composition comprises a form selected from the group consisting of a suppository, gel, cream, salve, jelly, spray, liquid, paste, aerosol, lotion, tablet, foam, film, and mist.

7. The composition of claim 6, wherein the microbicidal composition is adapted for application to external genital organs, vagina, anorectic region or rectum.

8. The composition of claim 1, wherein the microbicidal composition is adapted for inhibiting the activity of sexually transmitted disease pathogens, bacteria, viruses, fungi, or protozoa.

9. The composition of claim 1, wherein the microbicidal composition is adapted fordeactivating human sperm.

10. The composition of claim 1, wherein the microbicidal composition is adapted for use in vaginal mucous membrane for preventing and treating sexually transmitted diseases through inhibiting the activity of the sexually transmitted disease pathogens.

11. The composition of claim 1, wherein the microbicidal composition is adapted for use in vaginal mucous membrane for preventing and treating women's diseases caused by birth tract infections through inhibiting the activity of bacteria, viruses, fungi, and protozoan.

12. The composition of claim 1, wherein the microbicidal composition is adapted for use in vagina, exterior sex organs or rectum for preventing AIDS through inhibiting the activity of HIV.

13. The composition of claim 1, wherein the microbicidal composition is adapted for use on skin and mucous to treat skin and mucous diseases.

14. The composition of claim 1, wherein the microbicidal composition is adapted for use as a disinfectant for the disinfection of skin and mucous surfaces.

15. The composition of claim 1, wherein the microbicidal composition is adapted for reducing the transmission of STDs pathogens to health care providers, laboratory personnel, and related persons who may touch biological samples and specimens.

16. The composition of claim 13, wherein the microbicidal composition is adapted for use on animals.

17. The composition of claim 1, wherein the microbicidal composition is adapted for vaginal use to prevent unwanted pregnancy.

AMENDED CLAIMS

received by the International Bureau on 03 December 2009 (03.12.2009)

1. A composition comprising:
   0.03 to 3.0% weight percent N-cocoyl amino acid pyrrolidone salts; and 10 to 99.97% weight percent solvent, wherein said solvent comprises a member selected from the group consisting of water, glycerin, glycerol-gelatin, ethanol, propylene glycol, polyethylene glycol, a water and glycerin mixture, a water and propylene glycol mixture, and a water and polyethylene glycol mixture, and wherein said composition is for use in preventing and treating sexually transmitted diseases.

2. The composition of claim 1, further comprising a plant polysaccharide.

3. The composition of claim 1, wherein the amino acid group of the N-cocoyl amino acid pyrrolidone salts, comprises a member selected from the group consisting of arginine, lysine, histidine, homolysine, unnatural amino acid residue bearing a positive charge group, and tri-peptide bearing a positive charge group.

4. The composition of claim 1, wherein said water comprises de-ionized water, distilled water, or purified water.

5. The composition of claim 2, wherein said plant polysaccharide comprises Aloe Vera polysaccharides.

6. The composition of claim 1, wherein the compositions comprises a form selected from the group consisting of a suppository, gel, cream, salve, jelly, spray, liquid, paste, aerosol, lotion, tablet, foam, film, and mist.

7. The composition of claim 6, wherein the compositions is for use in external genital organs, vagina, anorectic region or rectum.

8. The composition of claim 1, wherein the composition is for use in inhibiting the activity of sexually transmitted diseases pathogens, bacteria, viruses, fungi, and protozoan.

9. The composition of claim 1, wherein the composition is for use in deactivating human sperms.

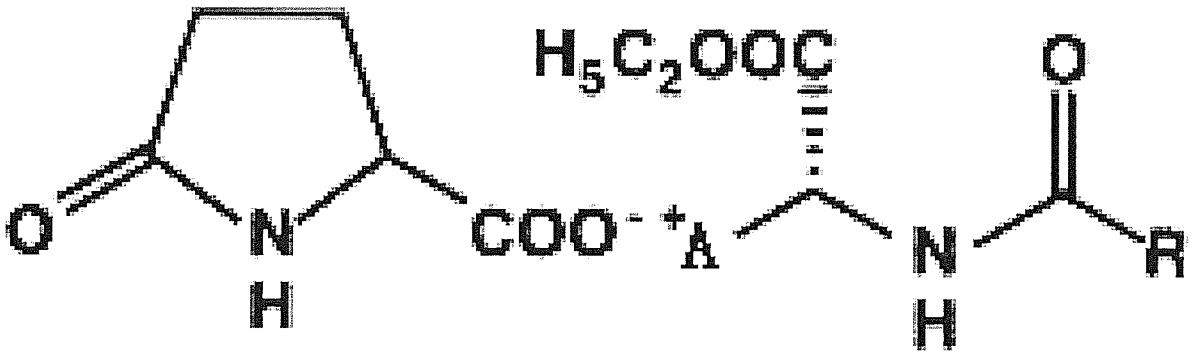
10. The composition of claim 1, wherein the composition is for use in vaginal mucous membrane for preventing and treating sexually transmitted diseases (STDs) through inhibiting the activity of the STDs pathogens.

11. The composition of claim 1, wherein the composition is for use in vaginal mucous membrane for preventing and treating women's diseases caused by birth tract infections through inhibiting the activity of bacteria, viruses, fungi, and protozoan.

12. The composition of claim 1, wherein the composition is for use in vagina, exterior sex organs and or rectum for preventing Aids through inhabiting the activity of HIV.

13. The composition of claim 1, wherein the composition is for use in vagina to prevent unwanted pregnancy.

Figure 1



INTERNATIONAL SEARCH REPORT

International application No
PCT/US2008/081795

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K9/00 A61K31/40 A61K47/18

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal

C. DOCUMENTS CONSIDERED TO BE RELEVANT

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[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

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Date of the actual completion of the international search	Date of mailing of the international search report
23 September 2009	02/10/2009

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31–70) 340–2040, Fax: (+31–70) 340–3016	Giese, Hans-Hermann

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INTERNATIONAL SEARCH REPORT

International application No
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