

Central Lancashire Online Knowledge (CLoK)

Title	Development of Mucoadhesive Gel Microbicide to Target Mucosal HIV Reservoirs
Type	Article
URL	https://clock.uclan.ac.uk/14572/
DOI	10.17352/2455-3786.000006
Date	2014
Citation	Shegokar, Ranjita and Singh, Kamalinder K (2014) Development of Mucoadhesive Gel Microbicide to Target Mucosal HIV Reservoirs. Journal of HIV for clinical and scientific research, 2 (1). pp. 1-7.
Creators	Shegokar, Ranjita and Singh, Kamalinder K

It is advisable to refer to the publisher's version if you intend to cite from the work. 10.17352/2455-3786.000006

For information about Research at UCLan please go to <http://www.uclan.ac.uk/research/>

All outputs in CLoK are protected by Intellectual Property Rights law, including Copyright law. Copyright, IPR and Moral Rights for the works on this site are retained by the individual authors and/or other copyright owners. Terms and conditions for use of this material are defined in the <http://clock.uclan.ac.uk/policies/>

Ranjita Shegokar and Kamalinder K Singh*

C.U. Shah College of Pharmacy, S N D T Women's University, Santacruz (W), Mumbai-400049, India

Dates: Received: 01 September, 2014; Accepted: 16 December, 2014; Published: 18 December, 2014

*Corresponding author: Kamalinder K Singh C.U. Shah College of Pharmacy, S N D T Women's University, Santacruz (W), Mumbai-400049; E-mail: kksingh35@hotmail.com

www.peertechz.com

Keywords: HIV/AIDS; Microbicide; Stavudine; Gel; Spermicidal activity; Dissolution; Antimicrobial action

Research Article

Development of Mucoadhesive Gel Microbicide to Target Mucosal HIV Reservoirs

Abstract

The wide use of microbicide is mainly depends on its effectiveness, less frequent application, ready availability and most importantly cost. The aim of this work was to develop affordable microbicide mucoadhesive gel formulation of synthetic anti HIV drug, stavudine and to characterise it in terms of its physical properties, mucoadhesiveness and spreadability. The purpose of the present study was also to compare different dissolution media used for in vitro release of vaginal dosage form. The gels were tested for antimicrobial, spermicidal and anti-HIV activity. Gels prepared using Carbopols and Polycarbophil were transparent and homogenous and had excellent mucoadhesion index - and showed fast drug release profile. Gels showed very good antimicrobial action against pathological microorganism.

Introduction

Lots of efforts are being done in the field of new products and combination product for antiretroviral therapy based on novel as well as conventional drug delivery technology. Researchers are trying to combine substances with different mechanism of action to synergies the effectiveness [1-3]. The most effective routes of HIV transmission are sexual transmission from infected individuals. The exact mechanism of such transmission remains unclear. Currently an estimated 47-53 million women worldwide are living with HIV/AIDS [4]. Women's are 4-16 times more likely to contract HIV from infected males than vice versa and young women are especially vulnerable.

The cervix is the main port of entry for HIV in women, blocking the cervix may have a big impact on HIV transmission. The genital viral load might serve as a better estimate for the transmission, being the distinct microenvironment that permits viral replication independent from the systemic circulation. Microbicide would provide protection by directly inactivating HIV or preventing HIV from attaching, entering or replicating in susceptible target cells as well as dissemination from target cells percent in semen or the host cells that line the vaginal or rectal wall [5].

Microbicides are products for vaginal administration that can be used to prevent HIV infection and other sexually transmitted diseases (STDs), it may be gel or cream or other form that could be applied to genital mucosae to prevent or significantly reduce the transmission of HIV and other disease causing organisms [6,7]. Most microbicides under development acts by either killing or inactivating pathogens, by breaking down the surface or envelop of the virus or pathogens, by creating physical barriers, by strengthening the body's normal defenses such as vaginosis natural acidity, by inhibiting viral entry by blocking site or preventing HIV attachment and inhibiting viral replication.

There is urgent need to develop a suitable drug delivery system

to combat costs, side effects, and local tissue drug distribution limitations. In this research, an attempt was made to formulate stavudine, a synthetic thymidine nucleoside analogue mucoadhesive microbicide gel. Three polymers were used at three different concentrations to produce gels. Prepared gels were evaluated mainly for their mucoadhesiveness, drug release profile and antimicrobial action.

Materials and Method

Stavudine was obtained as gift sample from Alkem Laboratories, Mumbai, India. Gelling agents such as Carbopol 934P (C934P), Carbopol 974P (C974P), Hydroxy propyl methylcellulose (HPMC), methylcellulose (MC) and carboxy methyl cellulose (CMC) were obtained as generous gift sample from Signet Chemicals, Mumbai, India. Polycarbophil (PCL) was provided by Noveon, Mumbai, India.

Experimental

Formulation of microbicide gel

Microbicide gels were prepared using various percentages (1 to 3%) of two Carbopol grades 934P and 974P and Polycarbophil. Varying percentages from 1-5% of gelling agent were tried to produce smooth gel of HPMC, MC and CMC. Weighed quantity of gelling agent was soaked overnight in measured quantity of water. Next day the polymer was mixed with drug solution and humectant (glycerin 5%) under slow stirring to get a uniform mix. Gels prepared with both the Carbopol type were neutralized using 10% NaOH solution and pH was maintained between 4 and 5 (Table 1).

Evaluation of microbicide gels

Gels were evaluated for various physiochemical parameters such as appearance, pH, drug assay, spreadability, mucoadhesiveness, *in vitro* drug release and *in vitro* penetration through sheep vaginal mucosa. In addition, antimicrobial, anti-spermicidal and anti-HIV activity was carried out for developed microbicide gel.

Table 1: Formulation optimization of stavudine microbicide gel.

Independent variables	Levels		
	C-934	C-974	PCL
A) Conc. of gelling agent (%w/w)	1	2	3
B) Stirring speed	100	200	300
C) % Drug	1	1	1

Percent drug content

Percent drug content of stavudine gels was determined partitioning drug into aqueous phase. Accurately weighed 100 mg gels were taken in a separating flask containing 100 ml of water or pH 7.2 buffer flask was shaken vigorously for 10 min., followed by filtration (0.45 μ m). The percent drug extracted in aqueous phase was determined by UV spectrophotometry (Jasco Instrument) at wavelength of 265 nm.

Spreadability index

The *in vitro* spreadability check was performed on drug containing gels which can act as an indicator how is the spread on the skin or mucosa surface when applied. The spreadability apparatus equipped with a pan, rectangular glass side and a wooden block was used for this study. The gel was placed in the middle of wooden block and glass slide was placed on it and allowed to stand for 5 min under weight of 300g to allow uniform gel film formation and to expel entrapped air. Later, the time was noted to move glass slide from the wooden box by adding weight of 100 g in pan till it completely separates from box surface. The spreadability index was calculated by time required in seconds for the plate to detach divided by total weight of the gel placed on the wooden surface in grams.

Mucoadhesiveness

Fresh sheep vaginal mucosa was obtained from a local slaughterhouse. The underlying fat and loose tissues very removed to obtain uniform mucosal lining. The mucosal membrane was washed with distilled water twice followed by saline solution. The bioadhesive strength of the gel was determined in triplicate. The mucosa was cut and tied to the Teflon cylinder and placed in 50 ml beaker filled with simulated vaginal fluid. A gel was applied on the surface of mucosa. Another hanging Teflon cylinder from balance was placed on mucosal surface containing gel and allowed in contact for 30 sec. The weights were added in another pan of balance slowly. The mass, in grams, required to detach the Teflon cylinder from the mucosal surface was noted. The mucoadhesion index was calculated using following equation.

$$\text{Force of adhesion (N)} = \frac{\text{bioadhesive strength}}{1000} \times 9.81$$

In vitro drug release

Literature reports use of various media for dissolution of vaginal dosage form. We studied the effect of dissolution media on *in vitro* drug release from mucoadhesive vaginal gel of an anti HIV drug.

Simple USP paddle apparatus was used for carrying out dissolution studies in 200ml of dissolution media at $37 \pm 0.5^\circ\text{C}$ and at 50 rpm. Total nine different media were investigated reported in literature e.g. distilled water pH7 (DW), citrate buffer B.P pH4.8 (CBBP), normal saline pH4.8 (SLN), citrate buffer pH 4.8 (CB) [8], acetate buffer pH6 (AB), acetate buffer with dioxane pH4.8 (ABWD) [9], phosphate citrate buffer pH4.8 (PCB)[10], modified citrate buffer pH5.5(MCB) [11] and simulated vaginal fluid pH4.2 (SVF) [12].

About 100 mg of gels was placed on glass slide and inserted in medium. Five milliliter of aliquot was withdrawn at each time intervals 30 min, 1-8 and 24 h from each dissolution vessel and filtered through 0.45 μ m membrane. The dissolution for respective blank gel was also performed. The drug content in aliquots was analyzed using spectrophotometer at wavelength of 265 nm.

In vitro drug penetration studies

In vitro drug penetration studies on selected batches were carried out using Franz diffusion cell at 50 rpm. The best medium selected from release study i.e. a simulated vaginal fluid (pH 4.8) was used as dissolution media (20 ml) maintained at $37 \pm 0.2^\circ\text{C}$. The weighed quantity gels (containing 100 mg drug) were placed on sheep vaginal mucosa as barrier. One milliliter of aliquots were withdrawn at time intervals 30 min, 1-8 and 24 h and filtered through 0.45 μ m. The drug content of samples was determined spectrophotometrically at 265 nm against blank gels.

Spermicidal activity

Spermicidal activity was determined on freshly obtained semen samples using modified Sander Cramer technique [13]. The spermicidal activity on pure drug at concentrations ranging from 0.25% to 10% and formulated gel was performed. This test indicates the minimum concentration of spermicidal agent required to kill 100% of the sperms within 20 secs. The various test concentrations were mixed with 20 μ L sperm suspension and observed under microscope at 400x magnification, the time was recorded at which the sperm loses its motility.

Anti-microbial activity

Developed gel formulations were checked against various gram +ve (e.g. Staphylococcus aureus (ATCC 25923), Bacillus subtilis (ATCC 6633), Streptococcus faecalis (ATCC 29212)) and gram – ve microorganisms (e.g. Pseudomonas aeruginosa (ATCC 27853), Klebsiella pneumoniae (ATCC 13883), Escherichia coli (ATCC 25922), Salmonella typhi (ATCC 6539)), Antifungal activity was checked against Candida albicans (ATCC 10231). The antimicrobial activity was tested by using agar diffusion method under aseptic conditions. Primarily inhibitory concentrations were determined using ditch plate method. A solution of stavudine in phosphate buffer saline and cloramphenicol (100 μ g) and clotrimazole (25 μ g) in DMSO was used in this study. Antimicrobial assay was performed using a nutrient agar and Sabourad dextrose agar as dehydrated media by applying cup plate technique. Initially, culture solution was prepared to get concentration of 1×10^6 microorganisms/ml which

was matched with McFarlands Standard No. 1 (0.1ml of 1% aqueous barium chloride solution added to 9.9ml of 1% H₂SO₄). On the other hand, 30ml of sterile agar bed containing one milliliter of culture suspension (1x10⁶ microorganisms/ml) was prepared under sterile conditions. In the solidified agar, cups of 13 mm diameter were bored using sterile cork borers. A test and standard solutions were added to the cups on the agar plates aseptically and incubated at 37 ± 2°C for 24 hours. The plates culturing *Candida albicans* were incubated at lower temperature of 25 ± 2°C. After 24h, the zones of inhibition were measured in millimeter.

Anti-HIV activity

The p24 antigen assay detects the soluble viral p24 protein content and is more sensitive than the reverse transcriptase assay. A H9-based *in vitro* method has been developed for the evaluation of anti-HIV activity of developed gel. The H9 cells were procured from National Centre for Cell Sciences, Pune, India. The virus was allowed to adsorb onto the cells followed by centrifugation and resuspension into fresh growth medium ensuring complete removal of soluble virus non-associated p24 antigen. These infected H9 cells were incubated in presence of test sample. The cell free supernatant was collected by centrifugation, and analyzed for p24 antigen. This detection was performed using a commercially available ELISA kit (Zeptometrix, California, USA) and percent cell inhibition was calculated.

Results and Discussion

Formulation of microbicide gel

The transparent gels of Carbopol 934, 974 and Polycarbophil, blank and stavudine loaded can be prepared easily and has pH in range of 4.6 - 4.8 without any sign of precipitation. All gels showed very high mucoadhesive strength when tested on vaginal mucosa, increase in polymer percentage showed increased degree of mucoadhesiveness. The viscosity of prepared gels was in between 1.5 to 2.5 cps depending upon the polymer concentration. This mucoadhesive property can serve as an important parameter in sustaining drug delivery, and simultaneously providing a good physical barrier for HIV migration from semen to vaginal tissue until reasonable residence time. All three gels were homogeneous, odorless and readily water washable. As stavudine is freely water soluble, it was easier to prepare varied concentrations of drug in gel (Table 2). The percent drug content was found to be 97-98%, suggested that drug was distributed evenly at all speed of stirring. The low and moderate stirring speed resulted in formation of homogeneous gels with minimum air entrapment, whereas high speed of 500 rpm showed significant degree of air entrapment. Gels prepared with HPMC, MC and CMC even at 5% concentration were less mucoadhesive and showed poor spreadability. Literature reports incorporation of synthetic antiretroviral drug like saquinavir [14], theaflavin [15] tenofovir [16,17] and dapivirine [18-20], IQP-0528, a pyrimidinedione analog [21], retrocyclin 1 analog RC-101 [22], octylglycerol [23] and for HI-443 (N'-[2-(2-thiophene) ethyl]-N'-[2-(5-bromopyridyl)] thiourea [24] as microbicide candidate. The spreadability of the gels was found to be in the range of 5-26 seconds.

Table 2: Evaluation parameters of gels prepared with Carbopol 934P (C934P), Carbopol 974P (C974P) and Polycarbophil (PCL). Batch C934P-1% showed ideal characteristic for use as microbicide gel.

Batch	Appearance	pH	Mucoadhesive index	Spreadability
C934P-1 [0.75%]	Transparent	4.2	1.30	6.26
C934P-1 [1%]	Transparent	4.7	9.67	21.48
C934P-1 [1.5 %]	Transparent	4.3	12.33	28.81
C974P-1 [0.75%]	Transparent	4.6	5.21	5.34
C974P-1 [1%]	Transparent	4.3	10.65	24.01
C974P-1 [1.5 %]	Transparent	4.5	13.15	26.42
PCL-1 [0.75%]	Transparent	4.8	9.63	6.91
PCL-1 [1%]	Transparent	4.6	12.89	24.56
PCL-1 [1.5 %]	Transparent	4.6	15.01	25.78
MBHPMC	Transparent	4.3	3.15	6.12
MBMC	Transparent	4.3	1.87	3.83
MBCMC	Transparent	4.4	2.32	5.11

In vitro drug release

The objective was to evaluate the effect of type of media and its composition on drug release. Many reports are published using single media for release depending on type of vaginal dosage forms, but none of the report compares different media for one formulation containing single drug. In this study, the type of dissolution media showed a marked effect on release of stavudine all gel formulations. In the citrate buffer and simulated vaginal fluid comparative faster drug release to other media with t50% of 9.25 min and 90% of 30 min in case of simulated vaginal fluid and 1h in case of citrate buffer B.P. The initial drug release was slow in acetate buffer containing dioxane (t50% = 12.78 min) with comparable t90% (29.75 min) (Figure 1). When dioxane was not added to the acetate buffer slow stavudine release was obtained (t50% of 23.44 min.), although within one hour 90% of drug was released. The release of stavudine was very slow in modified citrate buffer with t50% of 43.32 min and t90% of the 2.94h (Table 3). In distilled water as dissolution media, the drug release was found to be the slowest (t50% of >1h, t90% > 3h). The order of release of drug in dissolution media was arranged from fastest to slowest in following sequence SVF > CBBP > AB > CB= PCB=SL> ABWD > MCB> DW. The type of dissolution media, pH of media and composition played critical role in determining drug release from gels. Simulated vaginal fluid showed fastest drug release had an acidic pH of 4.2 while the water which showed slowest drug release had pH 7. The other media had pH of 4.8. The presence of dioxane in acetate buffer resulted in faster drug release as compared to ABWD which did not have dioxane. Similarly with increasing quantities of sodium citrate in modified citrate buffer, citrate buffer and citrate buffer B.P., respectively the drug release was found to increase. Thus, selection of dissolution media is an important consideration for carrying out *in vitro* drug release studies from vaginal formulation.

In vitro drug penetration studies

Permeation studies through vaginal sheep mucosa showed all most similar release behavior from all gel formulations. About 28%

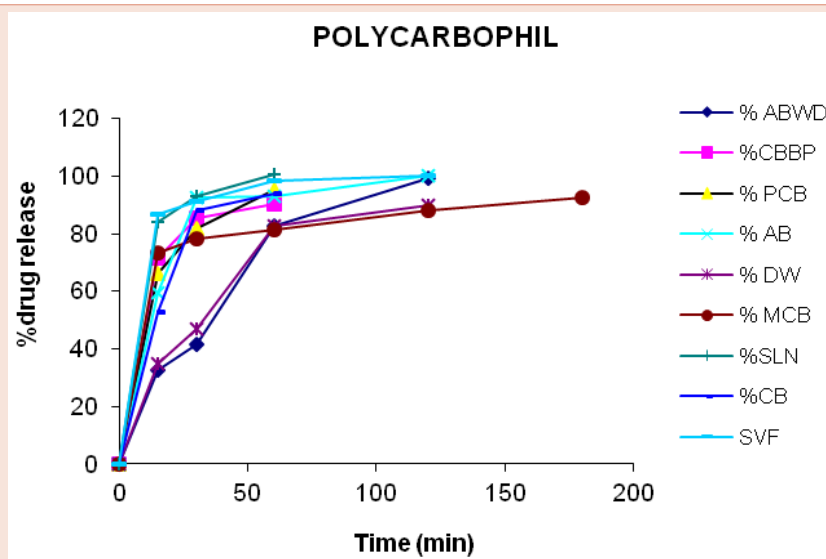


Figure 1: *In vitro* release profile of stavudine at various time points from microbicide gel prepared using Carbopol 934P, Carbopol 974P and Polycarbophil (1%) in acetate buffer without dioxane (ABWD), Citrate buffer/ British pharmacopoeia (CBBP), phosphate citrate buffer (PCB), acetate buffer (AB), distilled water (DW), modified citrate buffer (MCB), saline (SLN), citrate buffer (CB) and simulated vaginal fluid (SVF).

Table 3: Drug content, release pattern in terms of t50% and t90% when performed in simulated vaginal fluid as dissolution medium and *in vitro* penetration of selected stavudine gel batches from sheep vaginal mucosa.

Batch	% Assay	<i>In vitro</i> drug release [%]		<i>In vitro</i> penetration [%]	
		t50%	t90%	12 h	24 h
C934P-1 [0.75%]	99.37	7.50	24.26	-	-
C934P-2 [1%]	98.43	9.24	29.96	28.12	60.01
C934P-3 [1.5 %]	97.20	12.22	35.86	-	-
C974P-1 [0.75%]	98.45	8.34	28.56	-	-
C974P-1 [1%]	97.87	10.07	32.15	30.88	65.03
C974P-1 [1.5 %]	99.01	17.45	41.04	-	-
PCL-1 [0.75%]	98.34	7.56	27.89	-	-
PCL-1 [1%]	97.99	13.98	36.25	35.71	73.45
PCL-1 [1.5 %]	99.45	16.77	45.23	-	-
MBHP	96.68	11.52	56.12	42.37	80.12
MBMC	98.32	11.51	54.42	69.12	70.11
MBNAC	101.01	21.39	57.05	46.87	74.12

drug was release at end of 12 hours from C934P and C974P while it took 2 more hours for Polycarbophil gel to reach that level. At 24^h, almost 60-65% drug was penetrated from both Carbopol gels and 73% for Polycarbophil gel (Table 3). This penetrated drug concentration can kill virus which escaped by physical barrier formed by gelling polymer. It is a sufficient concentration to kill mucosal viral reservoirs.

Spermicidal activity

Sander Cramer test indicated spermicidal activity within one minute. All the gels were compared with the control (0.9% saline). The sperms when mixed with the control maintained its motility for more than 4h. A drug dose dependant decrease in the immobilization

of sperm was observed. The respective blank (placebo) gels did not show any effect on the sperm motility till 60 min. The drug solution and gels formulated with Carbopol 934, 980 and Polycarbophil containing 1% drug were also evaluated for their *in vitro* spermicidal activity. At the concentration of 6%, almost 100% of the sperms were immobilized within 20 seconds (Table 4).

Anti-microbial activity

The stavudine drug solution showed a good antibacterial activity on cup plate method, a concentration dependent increase was found in the zone of inhibition, with 50 mg/ml having the maximum inhibition. The results are depicted in Table 5 and Figure 2. The gels at 1% drug concentration showed good activity against the gram-

Table 4: *In vitro* spermicidal activity of stavudine solution and microbicide gel when mixed with semen sample at ratio of 20 μ l: 20 μ l and 50 μ l: 20 μ l at different time points.

Time [min] / % drug sol	0	1	2	3	4	5	6	7	8	9	10
	% spermicidal activity 20 μ l : 20 μ l										
0.25	20	15	10	10	10	5	5	3	3	3	3
0.5	2	1	1	1	1	1	1	1	1	-	-
1	1	1	1	1	1	1	1	1	1	-	-
2	1	1	1	1	1	1	Nil	-	-	-	-
4	1	1	1	Nil	-	-	-	-	-	-	-
6	1	1	Nil	-	-	-	-	-	-	-	-
8	1	1	Nil	-	-	-	-	-	-	-	-
10	1	1	Nil	-	-	-	-	-	-	-	-
	50 μ l : 20 μ l										
4%	0.5	Nil	-	-	-	-	-	-	-	-	-
6%	Nil	Nil	-	-	-	-	-	-	-	-	-
8%	Nil	Nil	-	-	-	-	-	-	-	-	-
C934P-1% gel	0.5	Nil	-	-	-	-	-	-	-	-	-

Table 5: Antimicrobial activity of stavudine microbicide gel in various gram positive, gram negative organism and fungi compared to standard cloramphenicol when tested by cup plate method.

Sr. No	Microorganisms	Zone of inhibition [mm] [50 mg/ml]			Standard (100 µg/cup) Chloramphenicol
		1	2	Avg	
	Gram + Ve				
1.	Staphylococcus aureus [ATCC 25923]	19.80	21.12	20.46	20.12
2.	Bacillus subtilis [ATCC 6633]	16.38	17.98	17.18	18.73
3.	Streptococcus faecalis [ATCC 29212]	20.12	20.98	20.55	21.71
	Gram -Ve				
4.	Escherichia coli [ATCC 25922]	19.78	21.32	20.55	20.30
5.	Klebsiella pneumonia [ATCC 13883]	16.10	16.80	16.45	19.50
6	Pseudomonas aeruginosa [ATCC 27853]	18.60	19.36	18.98	20.20
7	Salmonella typhii [ATCC 25923]	17.30	18.05	17.67	19.20
	Fungi				
8	Candida albicans [ATCC 6539]	18.30	19.01	18.65	22.00

negative organisms, *Escherichia coli* and *Pseudomonas aeruginosa* and slightly less activity against *Klebsiella pneumonia* and *Salmonella typhi* with the zones of inhibition of 20.55, 18.98, 16.45 and 17.67 mm, respectively. These gels also showed moderate activity with the zones of inhibition of 17.18 and 20.55mm against gram positive organisms *Bacillus subtilis* and *Streptococcus faecalis* at 175mg. The zones of inhibition of gel (18.65 mm) were comparable to clotrimazol (22.00

mm) when tested against fungi *Candida albicans*.

Anti-HIV activity

The Carbopol934P gel (batch C934P-1%) showed promising anti-HIV activity when tested by p24 antigen method. About 80 to 90% anti-HIV activity was observed for gels containing 1% drug. The MTT assay showed more than 85% of cell viability. An increase in drug

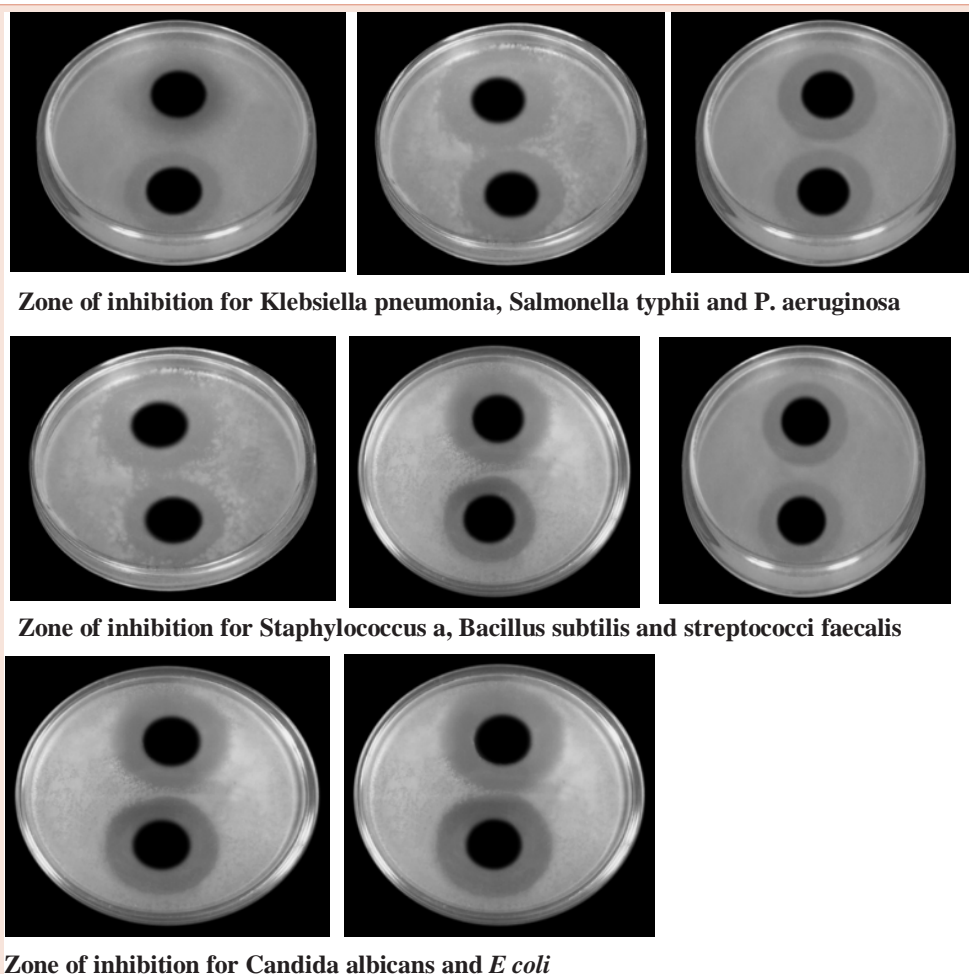


Figure 2: Photomicrographs agar plates showing the zone of inhibition of stavudine microbicide gel against various microorganisms and fungi.

concentration showed further increase in activity. The placebo gel also showed anti-HIV activity to lesser extent (10-15%).

Conclusion

Antiretroviral drug Tenofovir as gel microbicide significantly inhibited HIV transmission in clinical trial. This study indicates that a synthetic drug "stavudine" which is easily available and affordable can be considered as suitable candidate for development of microbicide formulation. The multifunctional mucoadhesive gels have a very good antimicrobial, anti HIV and spermicidal activity and can be easily prepared using three different polymers. The developed gels need to be tested *in vivo* in suitable animal model to confirm the results obtained *in vitro*.

Acknowledgements

Authors would like to thank All India Council of Technical Education (AICTE) for Financial Support.

References

- Shah LK, Amiji MM (2006) Intracellular delivery of saquinavir in biodegradable polymeric nanoparticles for HIV/AIDS. *Pharm Res* 23: 2638-2645.
- Shegokar R (2010) Development and evaluation of novel drug delivery systems for an anti-HIV Drugs. Ph.D thesis, S.N.D.T. Women's University, Mumbai, India.
- Sosnik A, Chiappetta DA, Carcaboso AM (2009) Drug delivery systems in HIV pharmacotherapy: what has been done and the challenges standing ahead. *J Control Release* 138: 2-15.
- Global HIV and AIDS estimates, <http://www.avert.org/worldstats.htm> accessed on 30.08.2012.
- Mahalingam A (2010) Design of a semisolid vaginal microbicide gel by relating composition to properties and performance. *Pharma Res* 27: 2478-2491.
- Rohan LC, Sassi AB (2009) Vaginal drug delivery systems for HIV prevention. *AAPS J* 11: 78-87.
- Nunn A (2009) Microbicides Development Programme: design of a phase III trial to measure the efficacy of the vaginal microbicide PRO 2000/5 for HIV prevention. *Trials* 10: 99.
- Bachhav YG, Patravale VB (2009) Microemulsion-based vaginal gel of clotrimazole: formulation, *in vitro* evaluation, and stability studies. *AAPS PharmSciTech* 10: 476-481.
- Sharma G (2006) Once daily bioadhesive vaginal clotrimazole tablets: design and evaluation. *Acta pharmaceutica* 56: 337-345.

10. Yellanki SK (2010) Development of Metronidazole Intravaginal gel for the treatment of bacterial vaginosis: Effect of Mucoadhesive Natural Polymers on the Release of Metronidazole. *International Journal of PharmTech Research* 2: 1746-1750.
11. Bilensoy E (2007) Thermosensitive mucoadhesive gel formulation loaded with 5-Fu: cyclodextrin complex for HPV-induced cervical cancer. *J Incl Phenom Macrocycl Chem* 57:363-370.
12. Liu Y (2009) Effect of carrageenan on poloxamer-based in situ gel for vaginal use: Improved in vitro and in vivo sustained-release properties. *Eur J Pharm Sci* 37: 306-312.
13. Dhuru S, Tatke PA, Singh KK (2008) Nanoneem®: A nanotechnology based herbal vaginal contraceptive. *International Seminar on Medicinal Plants and Herbal products Tirupati, India: MPIO-01*.
14. Vermeire K (2010) Development and In Vitro Evaluation of Gel-formulated Saquinavir as Vaginal Microbicide: Anti-HIV-1 Activity and Pharmaceutical Availability in Biorelevant Media. *23rd ICAR Abstract Issue / Antiviral Research* 86: A1-A78. 106.
15. Yang J (2012) Vaginal gel formulation based on theaflavin derivatives as a microbicide to prevent HIV sexual transmission. *AIDS Research and Human Retroviruses* 28: 1498-1508
16. Sokal DC (2013) Safety of tenofovir gel, a vaginal microbicide, in South African women: results of the CAPRISA 004 Trial. *Antivir ther* 18: 301-310.
17. Kiser PF (2012) Design of tenofovir-UC781 combination microbicide vaginal gels. *J Pharm Sci* 101: 1852-1864.
18. Nel AM (2010) Pharmacokinetics of 2 dapivirine vaginal microbicide gels and their safety vs. Hydroxyethyl cellulose-based universal placebo gel. *J Acquir Immune defic Syndr* 55: 161-169.
19. Woolfson AD (2010) Freeze-dried, mucoadhesive system for vaginal delivery of the HIV microbicide, dapivirine: optimisation by an artificial neural network. *Int J Pharm* 388: 136-143.
20. Nel AM (2009) Safety, tolerability, and systemic absorption of dapivirine vaginal microbicide gel in healthy, HIV-negative women. *AIDS* 23: 1531-1538.
21. Mahalingam A (2011) Vaginal microbicide gel for delivery of IQP-0528, a pyrimidinedione analog with a dual mechanism of action against HIV-1. *Antimicrob Agents Chemother* 55: 1650-1660.
22. Sassi AB (2011) Formulation development of retrocyclin 1 analog RC-101 as an anti-HIV vaginal microbicide product. *Antimicrobial Agents and Chemotherapy* 55: 2282-2289.
23. Wang L (2012) Development of a liposome microbicide formulation for vaginal delivery of octylglycerol for HIV prevention. *Drug Development and Industrial Pharmacy* 38: 995-1007.
24. D'Cruz OJ (2012) A novel vaginal microbicide containing the rationally designed anti-HIV compound HI-443 (N'-[2-(2-thiophene)ethyl]-N'-[2-(5-bromopyridyl)] thiourea]. *Expert Opin Investig Drugs* 21: 265-279.

Copyright: © 2014 Shegokar R, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Citation: Shegokar R, Singh KK (2015) Development of Mucoadhesive Gel Microbicide to Target Mucosal HIV Reservoirs. *J HIV Clin Scientific Res* 2(1): 001-007.