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Research Article

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Formulation and Evaluation of Dispersed Paroxetine Liposomes in Gel

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ABSTRACT

Paroxetine (PARX) is the most potent serotonin reuptake blocker antidepressant clinically available. This study is aimed to encapsulation of paroxetine in liposomes and formulation and evaluation of dispersed paroxetine in different gel bases. Paroxetine liposomes were prepared by reverse phase evaporation technique using soya lecithin, cholesterol and drug in different weight ratios. The prepared liposomes were characterized for size, shape, entrapment efficiency. The studies demonstrated successful preparation of paroxetine liposomes. The effect of using different weight ratios of soybean lecithin phosphatidylcholine: cholesterol (SLP: CHOL) on entrapment efficiency and on drug release was studied. Liposomes showed entrapment efficiency percent (% EE) of 81.22% ± 3.08 for paroxetine. The optimized paroxetine liposomes formula was F5 (7:7) molar ratio of (SLP: CHOL), which after that was incorporated in different based gels at different concentrations as Pluronic F127 (PF127-G) (20%, 25%and 30%), Carbopol 934 (C934-G) (1%, 1.5% and 2%) and Hydroxypropyl methylcellulose E4M (HPMC-G) (2%, 4% and 6%) and evaluated through in-vitro release, viscosity, pH and drug content.

Keywords: Liposomes, Paroxetine, gel, *In-vitro* drug release study.

INTRODUCTION

Liposomes are closed vesicles consisting of one or more concentric spheres of lipid bilayers (or lamellae) enclosing an equal number of aqueous compartments[1, 2]. Liposomes are hollow structures having dimensions between 0.005 and 100 µm [3-5] (figure a).

Liposomes have been widely used as drug carriers to deliver the entrapped drugs into the skin in treatment of diseases, especially in dermatology. They act as permeation enhancers by virtue of the phospholipids that penetrate into the stratum corneum and subsequently alter the skin lipid bilayers. They are known to act as a depot for sustained release of actives into the skin, and also modulate the rate and extent of systemic drug absorption. Liposomal formulations are known to favour drug deposition in the skin, reduce irritation potential of drugs, and improve drug stability [6].

They are capable to incorporate a variety of hydrophilic and hydrophobic drugs, to enhance the accumulation of drug at the administration site, to provide sustained and/or controlled release of entrapped drug and to reduce side effects and incompatibilities [7].

In search of improved topical products, attempts are being made to design new vehicles or utilize drug carriers to ensure adequate penetration and more importantly, localization of the drug within the skin [8, 9]. To overcome the difficulties of poor skin permeability various vesicular approaches have been proposed including elastic liposomes and ethosomes [10].

However, the major limitation of using liposomes topically is the liquid nature of the preparation. Since topically applied liposomes may leak from the application site. This can be overcome by their incorporation in an adequate

vehicle where original structure of vesicles is preserved and their rheological and /or mucoadhesive properties are adjusted. This can be achieved by adding gelling agents in liposomal dispersions forming liposomal hydrogels [11].

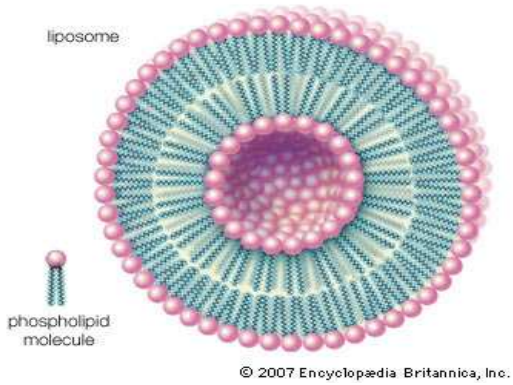

Figure a: A liposome. Cross-section of a unilamellar vesicle with an aqueous interior surrounded by a bilayer membrane

Gels are semi-solid systems comprising small amounts of solid, dispersed in relatively large amounts of liquid, yet possessing more solid-like character. These systems form a three-dimensional, polymeric matrix in which a high degree of physical (or sometimes, chemical) reticulation has been comprised. They are formed of long, disordered chains that are connected at specific points, but the connections must be reversible. The molecular mechanisms of gelation are poorly understood, but researchers are attempting to design and enhance molecules with these properties [12].

Hydrogels are hydrophilic natural or synthetic cross linked polymers that have the ability to swell in an aqueous environment without dissolution; the capacity of these molecular networks to absorb water arises from hydrophilic functional groups attached to the polymeric network, while the inability to dissolve arises from cross linked polymer chain. Water inside the hydrogel allows free diffusion of some solute molecules, while the polymer serves as a matrix to hold water together [13].

The formation of composite hydrogel drug release vehicles may increase the biocompatibility of the particulate vehicle by ‘‘hiding’’ the microparticles within the hydrogel while also preventing microparticle migration away from their targeted site in vivo. Poly(lactic-co-glycolic acid) nanoparticles can be incorporated within a cross-linkable hyaluronan-based hydrogel matrix without compromising the biocompatibility or anti-adhesion properties of the hyaluronic acid carrier [14], facilitating the incorporation of a wider array of anti-adhesion drugs within the matrix.

Hydrogels fabricated in the form of membrane or sheets can suitably be employed in the dermal or transdermal delivery of therapeutic agents [15].

Liposomes can also be entrapped in hydrogels. Liposomes entrapped in carbopol and hydroxyethylcellulose-based hydrogels can control the release of calcein and griseofulvin according to the rigidity of the liposomal membrane [16]. liposomes entrapped in poly(hydroxyethyl methacrylate) hydrogels mimicking contact lenses can control the release of anti-glaucoma drugs for up to 8 days [17].

Thereby, a drug-in-liposomes-in-gel complex formulation is developed. The release of drug molecules from such liposomal gels depends on the stability of the liposomes (membrane integrity and mechanical stability) during their dispersion in the semisolid formulation. This may be determined by the vesicle-membrane rigidity as well as the semisolid system physical properties as viscosity and rheological properties. The membrane integrity of liposome is important when hydrophilic drugs that cannot diffuse across lipid membranes are encapsulated in aqueous compartment of the vesicles. However, when amphiphilic or lipophilic drugs are used, other parameters may also be implicated, as the lipophilicity of the drug (or its ability to diffuse through lipid membranes), its aqueous solubility that will be the driving force moving drug molecules out of liposomes for partitioning in the aqueous dispersion media. Furthermore, it was observed that liposomes vesicles are protected from the disruptive effects of specific excipients when dispersed in hydrogels compared to aqueous media [11].

Because gel base formulations make the drug molecules more easily remove from the system than cream and ointment ones. Over the last decade hydrogels formed from natural, semisynthetic or synthetic polymers have been confirmed as vehicles for different types of pharmaceutical applications. They have good viscosity, satisfactory

bioadhesion, and are without irritating or sensitizing actions. High molecular grades of several commercial polymers derived from cellulose can be used in the formation of viscid, jelly-like aqueous solutions. These include methyl cellulose, sodium carboxymethylcellulose and hydroxypropylmethylcellulose. These are water soluble derivatives of cellulose and have been used as ointment bases so called 'hydrogel bases'. Generally, hydrogel bases can be easily washed out and well adhered to mucous membrane or skin, wet with secreting fluid and thus these are applied to injured skin and also to eyes. Percutaneous penetration, that is, the passage through the skin, involves the dissolution of a drug in a vehicle, diffusion of the solubilized drug from the vehicle to the surface of the skin, and the penetration of the drug through the layers of the skin, mainly the stratum corneum. This penetration may be improved by selecting the appropriate vehicle.

It has been confirmed that liposomes are fairly compatible with viscosity increasing agents such as methylcellulose, as well as polymers derived from acrylic acid (Carbopol resins). Due to the good bioadhesive properties of some Carbopols and the prolonged retention of the formulation at the site of administration, these hydrogels can be good vehicles for incorporation of liposomes destined for vaginal delivery [18].

Paroxetine (PARX) is the most potent serotonin reuptake blocker antidepressant clinically available, but has a lower selectivity for the serotonin reuptake site than either fluvoxamine or sertraline[19]. In addition, it blocks muscarinic acetylcholine receptors to almost the same degree as the Tricyclic Antidepressants (TCAs) imipramine or doxepin, and even more effectively than desipramine or maprotiline. PARX is efficiently absorbed from the gastrointestinal tract, but is readily metabolized during its first pass through the liver. The $t_{1/2}$ is variable, depending on both dose and duration of administration. Half–life up to 21 hours after oral administration of 30 mg of paroxetine/day [20, 21].

In this study PARX will be encapsulated in liposomes and evaluated for the encapsulation efficiency, and in vitro drug release. Then the dispersed PARX liposomal formulations also were incorporated in different based gels at different concentrations as Pluronic F127 (PF127-G) (20%, 25% and 30%), Carbopol 934 (C934-G) (1%, 1.5% and 2%) and Hydroxypropyl methylcellulose E4M (HPMC-G)(2%, 4% and 6%) and evaluated through *in-vitro* release, viscosity, photomicroscopic analysis, pH and drug content.

EXPERIMENTAL SECTION

2.1. Materials:

Paroxetine was obtained from GlaxoSmithKline Beecham (England), L-α-Phosphatidylcholine P3644-25G from soybean and cholesterol, from Sigma Chemical Co (St Louis, MO, USA). Spectra / Pore dialysis membrane, 12,000-14,000 molecular weight Cut off (Spectrum Laboratories Inc., USA, Cellulose Nitrate a Millipore ® Filter pore size (0.45 µm) Sartorius Stadim GmbH 37070 Goettingen Germany. Pluronic F127 (PF127, Sigma Chemical Co., St. Louis, USA). Carbopol 934, B.F., (Goodrich Chemical Company, Ohio, USA). Hydroxypropyl methylcellulose (HPMC)-E4M, (Tama, Tokyo, Japan). Triethanolamine, E. Merck, Germany. All the other chemicals, reagents and solvents used like potassium dihydrogen orthophosphate, disodium hydrogen phosphate, sodium hydroxide pellets, acetone, chloroform, and methanol were of analytical reagent grade.

2.2. Preparation of PARX liposomes:

Large unilamellar and oligolamillar liposomes [22] were prepared using the reverse-phase evaporation [23] technique according to Szoka and Papahadjopoulos [24, 25] as follows:

the liposomal components (lecithin(SLP), either alone or in different molar ratios with cholesterol (CHOL), equivalent to 200 mg as illustrated in table 1 , were weighted into 100 ml round bottomed flask and dissolved in 10 ml of chloroform. The organic solvent system was slowly evaporated under reduced pressure, using rotary evaporator (Heidolph –Laborota 4000, D-91126-Germany), at 40^{o}C and at 60 rpm for 15 minutes, such that a thin film of dry lipid was formed on the inner wall of the rotating flask.

The lipid film was redissolved in 5 ml diethyl ether, and the PARX (10mg) solution in 5 ml of acetone and 10 ml of phosphate buffered saline pH 7.4 was added at this point. The resulting two-phase system was sonicated for 2 minutes in bath-type sonicator. The mixture was then placed on the rotary evaporator and the organic solvents were removed under reduced pressure at 40^{o}C and 60 rpm for 15 minutes. The liposomes were allowed to equilibrate at room temperature, and the liposomal suspension was kept in the refrigerator (4^{o}C) to mature over night [26-28].

Table1.Composition of lipids for preparation of PARX liposome formulations

Composition mg	Formulae				
	F1	F2	F3	F4	F5
SLP	200	180	155.5	127.5	100
CHOL	0	20	44.5	72.5	100
PARX	10	10	10	10	10

2.3. Separation of Entrapped From Unentrapped PARX from its Liposomes:
The PARX trapped in liposomes were separated from unentrapped PARX by cooling centrifugation of a known aliquot (1 ml) of the prepared liposomal suspension at 15000 rpm for twenty minutes at (4^{o}C) in Cooling or refrigerated centrifuge, Haraeus general purpose centrifuge (Megafuge 1.0/1.0R), Kendro laboratory products, Fussex, UK) [29, 30]. The supernatant was separated from the liposomal precipitate. The precipitated liposomes was washed by 1ml of phosphate buffered saline pH 7.4 and recentrifuged for twenty minutes to remove excess unentrapped PARX, then the combined supernatant was diluted to 10 ml by phosphate buffered saline (pH 7.4). The concentration of unentrapped paroxetine was determined spectrophotometrically (Model 6705-Jenway, Multicell changer, Bibby Scientific ltd. U.K.) by measuring the UV absorbance at λ 294.3 nm.

The percentage entrapment efficiency (% EE) of PARX was determined relative to the original drug added, applying the following equation:

$$\% \text{ EE} = [(C_d - C_f)/C_d].100$$

Where %EE is the percentage entrapment efficiency, C_d is concentration detected of total PARX added and C_f is concentration of free PARX.

2. 4. Preparation of PARX liposomal gel
F5 was selected to be incorporated in gels as the best formula according to the release as it showed the highest release and also the lowest deviation (SD) and (CV% or RSD)41.04 ± 1.20 (CV% 2.87)compared to other formulae.

2.4.1. Preparation of PARX liposomal gel by Pluronic F127 (PF127-G):
PF127-G based PARX liposomal gel was formulated in three different ratios (20% w/w, 25% w/w, and 30% w/w) [31] , where the weighed amount of PF127 was sprinkled gradually in distilled water and then stirred with magnetic stirrer at medium speed, and then the dispersion was stored in refrigerator (4°C) for 24 hours for bubbling air to be removed, and increasing orderliness of the cross link for the gel and became transparent. The gels were formed when the solutions are equilibrated at room temperature. Liposomal gel formulations were prepared by mixing the liposomal dispersion with the gels in order to have a final PARX concentration of 0.01% w/w in the gels.

2.4.2. Preparation of PARX liposomal gel by Carbopol 934 (C934-G):
C934-G based PARX liposomal gel was formulated in three different ratios (1% w/w, 1.5% w/w, and 2%w/w), where the weighed amount of C934was sprinkled gradually in distilled water and then stirred with magnetic stirrer at medium speed. Stirring was continued until no lumps were observed and then triethanolamine was added for neutralizing the gel base. The resulting gel was stored in refrigerator (4°C) for at least 24 hours until it was fully swollen and transparent. Liposomal gel formulations were prepared by mixing the liposomal dispersion with the gels in order to have a final PARX concentration of 0.01% w/w in the gels [16].

2.4.3. Preparation of PARX liposomal gel by Hydroxy propylmethylcellulose E-4M (HPMC-G):
HPMC-G based PARX liposomal gel was formulated in three different ratios (2% w/w, 4% w/w, and 6% w/w), where the weighed amount of HPMC-E4M was dissolved gradually in hot distilled water by aid of magnetic stirrer at medium speed. Stirring was continued until the formation of gel base then left overnight for equilibration. Liposomal gel formulations were prepared by mixing the liposomal dispersion with the gels in order to have a final PARX concentration of 0.01% w/w in the gels.

2.4.4. pH measurements:
The pH measurement of the prepared PARX liposomal gel (1g of each gel formula in 9 g of distilled water using magnetic stirrer) were measured by using pH meter, and the measurements were repeated three times for each formula and the average of the readings of three replicated was taken.

2.4.5. Viscosity measurements:
A rotational Brookfield viscometer (Cone and Plate viscometer, with attached computer software, Model III Brookfield, DV-I, USA) was used to measure the viscosities (in cps) of the gels. The prepared PARX liposomal gels

were evaluated, for PF127-G (20%, 25%, and 30%), C934-G (1%, 1.5%, and 2%), and HPMC-G (2%, 4%, and 6%) at 25 ± 1 °C [32].

About 0.5 g of the tested formula was applied to the plate and left until the temperature of the cone reached 25 ± 1°C. The measurements were made over the range of speeding setting from 0.5 to 100 r.p.m. with 10 second between each two successive speeds, and then in a descending order.

The individual rheological data [□ min, □ max, Furrow's constant (N)] for each of the tested gels were calculated.

The rheological data were analyzed by using Farrow' s equation[33] and power law equation[34] to predict the rheological behaviour of each formula.

Farrow's equation: Log D = N Log S - Log □

Where: D: Shear rate (sec $^{-1}$) S: Shear Stress (dyne/cm^2)
N: Farrow's constant □: Viscosity (cp.)

N (Farrow's constant) is the slope of log D against log S plot, which indicates the deviation from Newtonian flow.

When N is less than one, it indicates dilatants flow (shear rate thickening). If N is greater than one, it indicates pseudoplastic flow (shear rate thinning).

Power law equation: □ = □ □^{n}

Where: □: Shear rate (sec $^{-1}$) □: Shear Stress (dyne/cm^2)
n: power constant □: a constant called the consistency Index (apparent viscosity)

n (power constant) is the slope of log Shear Stress (□) against log shear rate (□), which indicates the deviation from Newtonian flow. In case of Newtonian behaviour n=1, whereas in case of (shear thinning) 0<n<1, while in case of dilatants flow (shear thickening) n>1.

2.4.6. Drug content studies:
Drug content of the gels was determined by dissolving an accurately weighed quantity of PARX liposomal gel (about 1 g) in about 10 ml of methanol. These solutions were quantitatively transferred to volumetric flasks and appropriate dilutions were made with the same methanol solution. The resulting solutions were then filtered through 0.45 µm membrane filters [35] before subjecting the solution to spectrophotometric (Model 6705-Jenway, Multicell changer, Bibby Scientific ltd. U.K.) analysis for PARX at 296 nm.

2.4.7. Photomicroscopic analysis:
Samples of PARX liposomal gel preparations were examined microscopically at magnification of 40X and 100X with a binocular Light microscope equipped with camera to study their size (Leica-Queen 550IW- Germany). A sample of PARX liposomal gel is placed on microscope slide and was covered with cover and was examined and photographed for morphological evaluation.

2.4.8. *In-vitro* release studies of PARX **liposomal gelled through artificial membrane:**
The release of PARX from liposomal gels with different compositions was studied using U.S.P. dissolution Tester (Classic Vession6- Vextra-Model BLHMO15K-10 Oriental Motor, Co. ltd.-Japan). Dialysis Spectra / Pore dialysis membrane, 12,000- 14,000 molecular weight Cut off (Spectrum Laboratories Inc., USA) was soaked in phosphate buffer saline (pH 7.4) for 4 hours for moistening of the membrane and removing the preservative [36, 37].

An accurately measured amount of PARX liposomal gels formulations, equivalent to 10 mg of paroxetine was inserted in a glass cylinder having the length of 10 cm and diameter of 2.5 cm. This cylinder was fitted with pre-soaked membrane and was placed in the vesicles of the U.S.P. dissolution Tester, containing 100 ml. of Phosphate buffer saline (pH 7.4) with constant speed (75 rpm) at 32±0.5^{o}C [37]. Sink condition is fulfilled since the saturated solubility was previously determined to be 5.5±0.1mg/mL.

At predetermined time intervals (0.5, 1, 1.5, 2, 4, 8, 12 and 24 hr), 5 ml aliquots of the release medium were withdrawn for analysis and replaced with equal volume of fresh phosphate buffer saline (pH 7.4) solution to maintain a constant volume [38]. The absorbances of the collected samples were measured spectrophotometrically at

λ max 294.3 nm using phosphate buffer saline (pH 7.4) as blank. The results are the mean values of the release experiments.

The release data obtained from this study was analyzed using correlation coefficient (r) and Korsmeyer-Peppas equation.

$$M_t/ M_\infty = K\, t^n$$

The value of K and n were estimated by linear regression of Log (M_t/ M_∞) on Log (t) where Log K is the intercept and n is the slope of the straight line

$$\text{Log } (M_t/ M_\infty) = \text{Log } K + n \text{ Log } t$$

RESULTS AND DISCUSSION

3. 1. Preparation of PARX Liposomes

The reverse-phase evaporation technique yielded the liposomes with a large unilamellar and oligolamillar liposomes. Particle sizes were measured by (Zetasizer ZEN 3600 Nano ZS (Red badge) Malvern Instr., UK) and reported in table (2).This technique was easily to be used in the preparation of liposomes [39].

3.2. Entrapment Efficiency (%EE) of PARX

The entrapment efficiency percent results of PARX of liposomes were shown in table (2) indicated that as the concentration of SLP decreases, drug entrapment efficiency of liposomes decreases which was due to the saturation of lipid bilayer with reference to the drug where low phosphatidylcholine content provides limited entrapment capacity. The encapsulation efficiency of liposomes is governed by the ability of formulation to retain drug molecules in the aqueous core or in the bilayer membrane of the vesicles. CHOL improves the fluidity of the bilayer membrane and improves the stability of bilayer membrane in the presence of biological fluids such as blood/plasma [40, 41].

3.3. Preparation of PARX liposomal gel

3.3.1. Evaluation of the physical properties of the prepared PARX liposomal gels:

All PARX liposomal gels showed gel in appearance except PF127 in concentration of 20% showed highly viscous liquid, White in colour, homogenous and not precipitated.

3.4. pH measurements

All of the PARX liposomal gels exhibited values of pH between five and six. These values are undoubtedly suitable and non irritating to the skin [42, 43].

3.5. Viscosity measurements

All PARX liposomal gels revealed a non-Newtonian shear thinning (pseudoplastic) flow behaviour as illustrated in table (3), where there is a decrease in viscosity by the shear rate. The cause of shear thinning flow may be due to progressive rupture of the internal structure of the formulations (by increasing shear) and its later reconstruction by means of Brownian movement [44].

There was a significant increase in Farrow's constant upon increasing polymer concentration as shown in figure [45]. In addition, a non – thixotropic behavior was revealed. Also, from the power law, the flow index (n) values were <1 revealing a non-Newtonian shear thinning behaviour. There was a significant decrease in flow index (n) upon increasing polymer concentration which was in accordance to Fresno et al [46]. This decrease on the flow index was explained by the formation of full structured three dimensional polymer lattices due to increased polymer concentration as shown in Figure (2). Formulae of PARX liposomal gels with 2% C934-G, and 6% HPMC-G exhibited reading error in torque percent due to high viscosity and therefore this formula was rejected.

By comparing the viscosity values of PARX liposomal gels prepared by the same polymer in different concentration, it was found that, by increasing the concentration of the polymeric material in the liposomal gel, there was an increase in the viscosity values as shown in table (3) and figures (3 and 4). This result was in accordance with Jones et al [47] who explained it by macromolecular entanglement phenomena. Because higher concentrations of polymer increase the entanglement density, the viscoelastic properties increase correspondingly.

3.6. Drug content of PARX liposomal gel

Drug content of prepared PARX liposomal gel containing different concentration of gelling agents were illustrated in table (3) and figure (5). The results showed that the range of drug content uniformity between 93% - 96%.

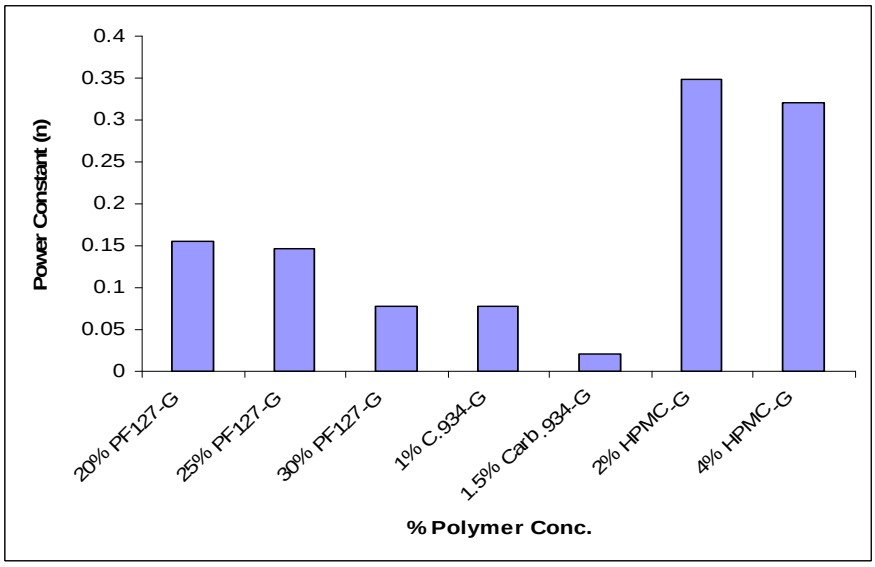

Figure (2): Relation between percent polymer concentration of formulae and Power constant

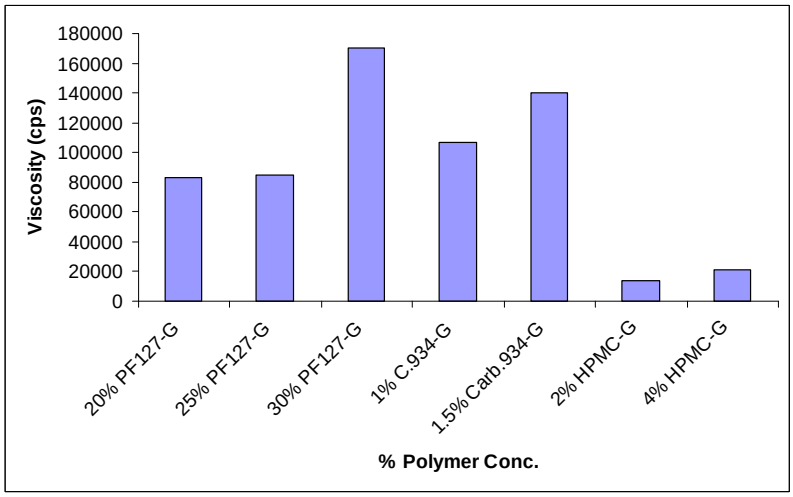

Figure (3): Comparison between viscosities of PARX liposomal gels at minimum shear rate

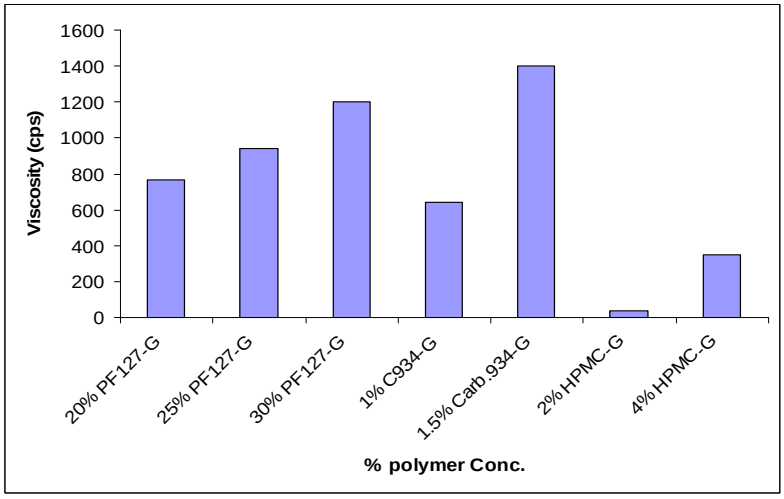

Figure (4): Comparison between viscosities of PARX liposomal gels at maximum shear rate

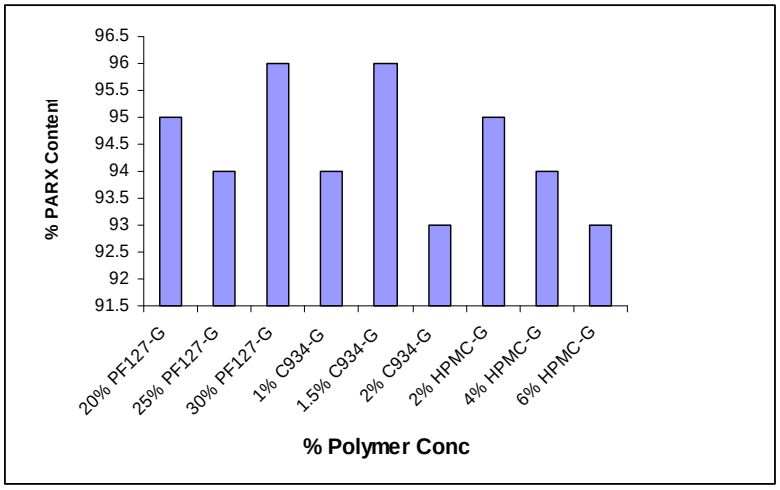

Figure (5): Drug content in PARX liposomal gel containing different concentration of gelling agents

Table (3): Physical parameters of PARX liposomal gels in different concentrations of gelling agents

Polymers		Rheological data of PARX liposomal gels containing different concentration of gelling agents.				Viscosity of PARX liposomal gel at minimum and maximum shear rate.		Drug content in PARX liposomal gel
Gelling agent	% Conc.	Farrow's Constant (N)	Flow index (n)	Consisteny index (n)	Flow Behavior	Min. Shear Rate (□ max)	Max. Shear Rate (n max)	Mean Drug content ± S.D.
PF127-G	20%	6.175	0.156	991.745	Pseudoplastic	83000	765	95% ± 0.1
	25%	6.423	0.146	990.148	Pseudoplastic	85000	940	94% ± 0.15
	30%	11.871	0.078	1356.438	Pseudoplastic	170000	1200	96% ± 0.1
C934-G	1%	11.129	0.077	1160.112	Pseudoplastic	107000	640	94% ± 0.1
	1.5%	48.476	0.0203	1748.236	Pseudoplastic	140000	1400	96% ± 0.1
	2%	----------	---------	----------	----------	----------	----------	93% ± 0.1
HPMC-G	2%	2.784	0.349	33.822	Pseudoplastic	14000	40	95% ± 0.1
	4%	2.811	0.321	172.743	Pseudoplastic	21000	350	94% ± 0.1
	6%	----------	---------	----------	----------	----------	----------	93% ± 0.1

3.7. Photomicroscopic analysis

The photomicrograph of PARX liposomal gels are shown in Figure (6). It showed the presence of homogenous population of unilamellar vesicles with one phospholipid bilayer and oligolamellar vesicles consisting of a few concentric bilayers inside of gels (PF127-G, C934-G and HPMC-G). The liposomes are well-identified spheres that have a large internal aqueous space relative to the sphere diameter.

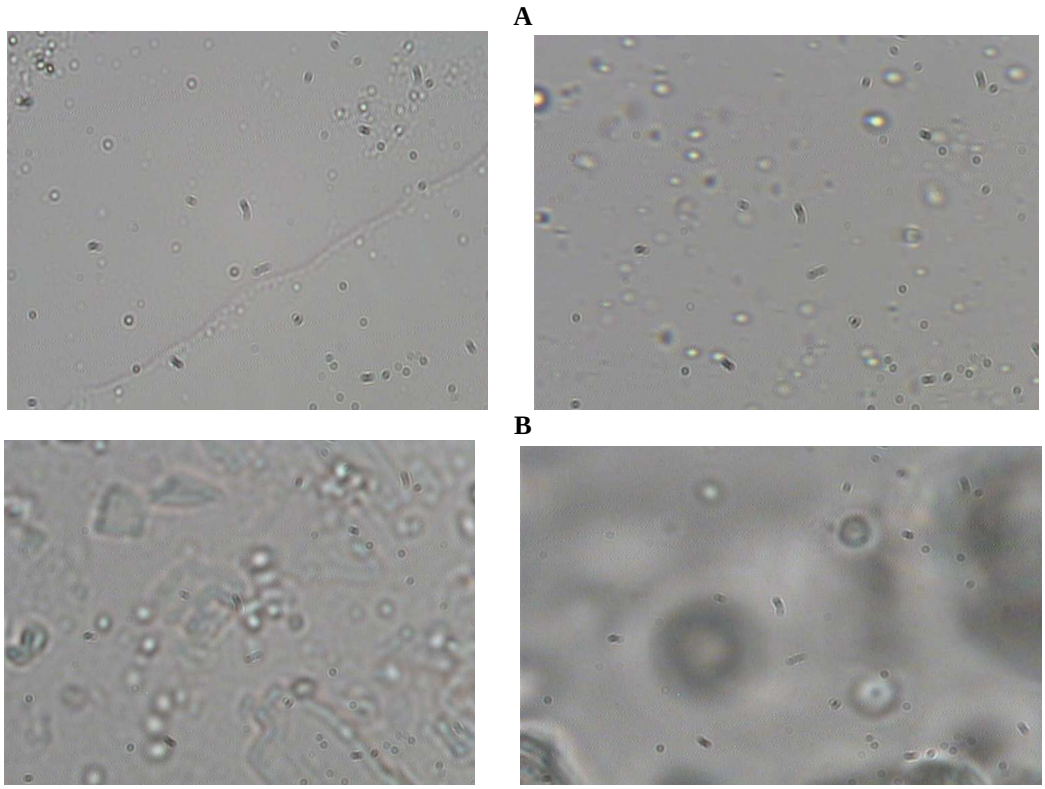

Figure (6): Photomicroscopic of PARX liposomal (A and B) by lance X40 and X100 respectively.

3.8. *In-vitro* release studies of PARX liposomal gelled through artificial membrane

The diffusion of PARX from different liposomal gels through artificial membrane was apparently dependent on polymer concentration as illustrated in Figures (7, 8 and 9)**,** where the increase in the polymer concentration is associated with decrease in rate of release due to increase in viscosity of the prepared gel. These results are in accordance with Jones et al. [47] who stated that the physical reason for slower release rate from viscous gel is most probably due to formation of highly viscous diffusion layers of hydrated polymer chain which entraps the excess of water and reduce migration of drug molecules.

The highest PARX release was obtained from PF127-G (20%, 25% and 30%) and also from HPMC-G (2% and 4%) based liposomal gels as follows: 32.76% (CV% 3.39), 31.66% (CV% 3.75), 27.46% (CV% 1.62), 30.98% (CV% 0.24) and 29.30% (CV% 2.28) respectively compared to 6% HPMC-G and C934-G (1%, 1.5% and 2%) based liposomal gels which show percent release of 18.03% (CV% 2.47), 17.40% (CV% 3.41), 15.57% (CV% 2.38) and 13.99% (CV% 1.59) respectively. This can be due to the low viscosity of these formulations. On the other hand, the lowest PARX release was obtained from Carbopol 934 1%, 1.5% and 2% based liposomal gels due to the high viscosity exhibited by these formulations. These results are in accordance with Attia and Basu [48, 49] who found that PF127-G liposomal gel demonstrated increased release rates for hydrophobic drug indicating easier diffusion of the compound through this type of gel compared to C934-G based liposomal gel.

Statistically, one way ANOVA test was applied - to all formulations of PARX liposomal gels at significant level **(*P* = 0.05),** The results showed there are no significant differences in the release between formulae PF127-G (20%, 25% and 30%), also from HPMC-G (2%, 4% and 6%) and C934-G (1%, 1.5% and 2%) based liposomal gels.

The release parameters of PARX liposomal gel through artificial membrane were graphically compared to the release parameters of PARX liposomal suspension formula F5, which showed that there was statistically significant differences in the extent of release from all formulae PARX liposomal gels when compared to F5 liposomal suspension where **(*P* = 0.044)** as illustrated in Figures (10 and 11). This indicated that the incorporation of liposomal

suspension into gel bases resulted in delayed release due to presence of an additional diffusion barrier to the drug release [50].

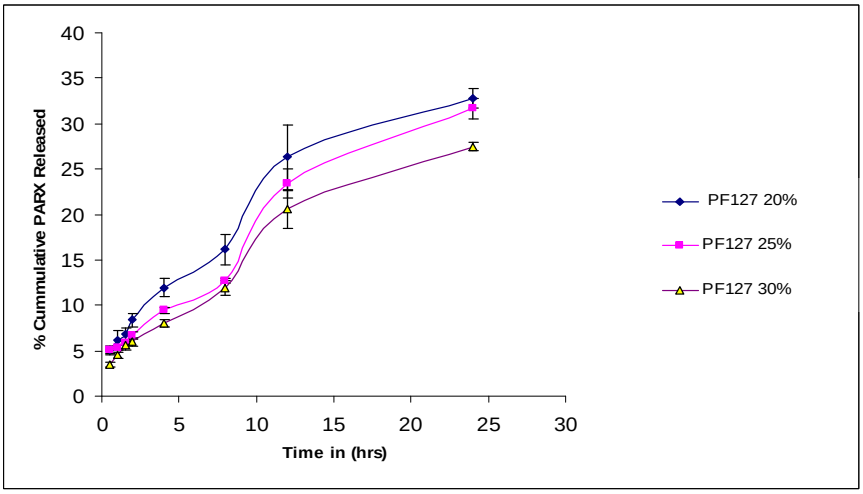

Figure (7): Release studies profile of PARX through artificial membrane from different liposomal PF127-G formulations

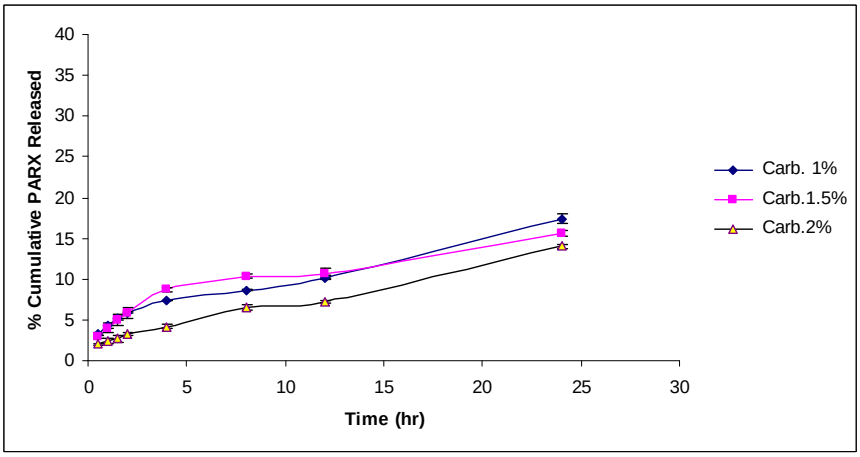

Figure (8): Release studies profile of PARX through artificial membrane from different liposomal C934-G formulations

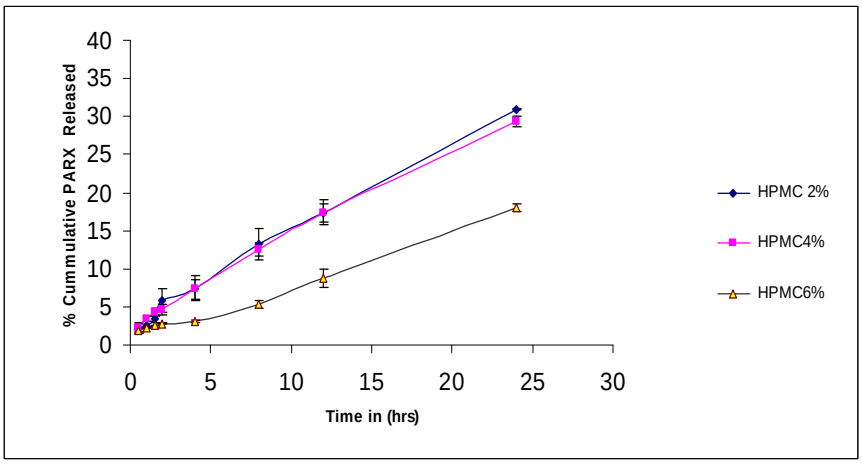

Figure (9): Release studies profile of PARX through artificial membrane from different liposomal HPMC-G formulations

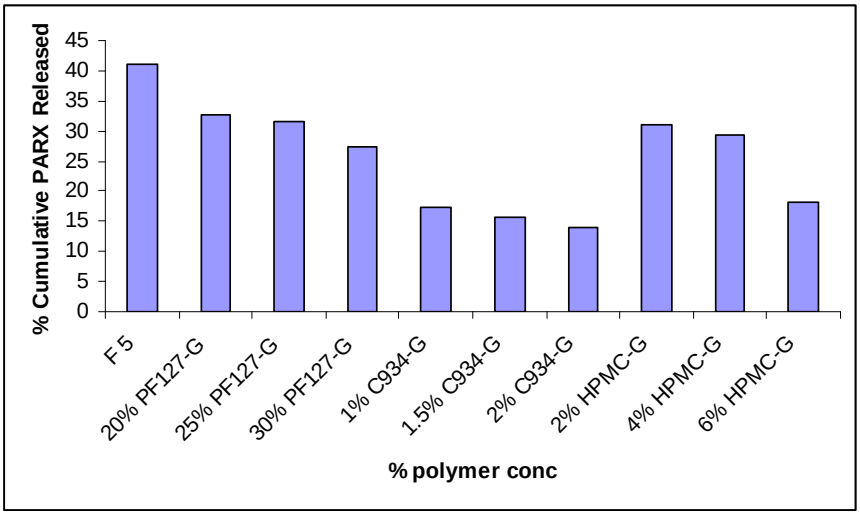

Figure (10): Percent Cumulative PARX released from different concentrations of liposomal gelling agents in comparison with formula 5.

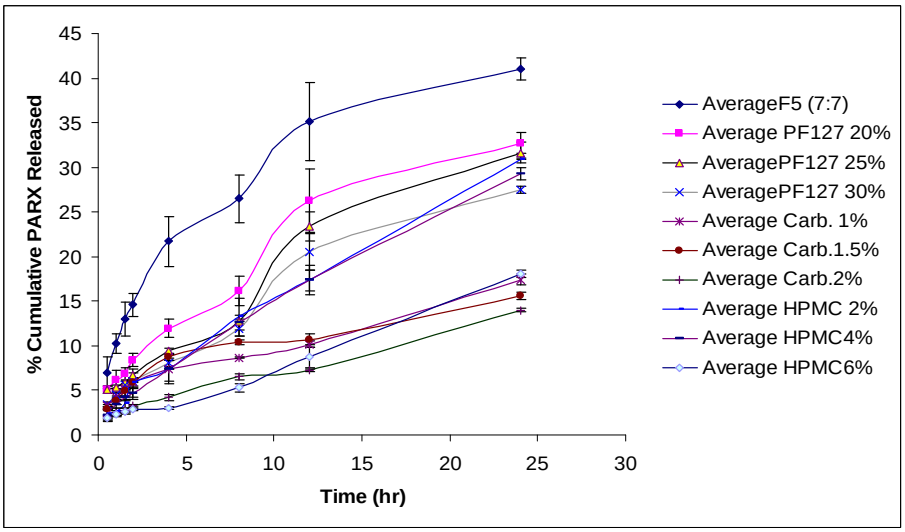

Figure (11): Percent Cumulative PARX released from different concentrations of liposomal gelling agents in comparison with F 5.

3.9. Kinetic analysis of *in-vitro* release of PARX from liposomal gels through synthetic membrane using Korsmeyer – Peppas equation:

Analyzing the results of PARX permeation through artificial membrane according to Korsmeyer-Peppas equation, diffusion exponent (n) for the PARX-liposome containing gels is mostly higher than 0.5 and lower than 1.0 except formulae of paroxetine-liposome containing 1% C934-G, 1.5% C934-G and 2% C934-G in which (n) was less than 0.5 as shown in table (4), so when (n) is more than 0.5 and less than 1.0, indicating that most of the formulae were an anomalous behavior (non-Fickian). Also the release constant was decreased as the concentration of each polymer increased except for formula of PARX-liposome containing 4% HPMC-G.

These results are in accordance with Mourtas et al, who studied the diffusion of griseofulvin from liposomal gel, where it was found that an anomalous behavior with respect to the release profile of drug was observed because other parameters (in addition to diffusion) were also implicated in drug release[16].

Table (4): Kinetic analysis of *in-vitro* release of PARX from liposomal gels synthetic membrane

% Polymer Conc.		**Kinetic analysis of the permeation data of PARX liposomal gel using Korsmeyer – Peppas equation.**			
Gelling agent	% Conc.	Release exponent (n)	Kinetic constant (K)	Regression Coefficient (R^2)	Mechanism
PF127-G	20%	0.516	6.187	0.9753	Anomalous transport
	25%	0.506	5.444	0.9275	Anomalous transport
	30%	0.547	4.441	0.9705	Anomalous transport
C934-G	1%	0.395	4.271	0.9737	Fickian diffusion
	1.5%	0.426	4.136	0.9798	Fickian diffusion
	2%	0.487	2.409	0.9629	Fickian diffusion
HPMC-G	2%	0.7	3.084	0.9795	Anomalous transport
	4%	0.67	3.25	0.9957	Anomalous transport
	6%	0.552	2.123	0.8914	Anomalous transport

CONCLUSION

Visual inspection showed that all PARX liposomal gels were milky white homogenous gels except for the formula consisting of 20% PF127-G which was milky white highly viscous liquid. The measured pH were in the range 5.65 – 6.40. All PARX liposomal gels revealed a non – Newtonian shear thinning pseudoplastic flow behavior with non – thixotropic behavior and increasing polymer concentration led to subsequent increase in liposomal gels viscosity. The diffusion of PARX from liposomal gels was apparently dependent on polymer concentration. Analyzing the release results of paroxetine from liposomal gel formulations through artificial membrane according to Korsmeyer-Peppas equation exhibited an anomalous behavior (non- Fickian Kinetics) corresponding to a coupled diffusion/and other parameter mechanism. Liposomes-based gels demonstrated remarkable advantage in formulating PARX. Further in vivo evaluations of liposomal formulations of PF127-G (25%), HPMC-G (2%) and HPMC-G (4%) will be studied in the future.

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