Comparison of Carbopol 934 and 941 as Thickener on Diffusion Rate of Ketoconazole Microemulsion

Rahmah Elfiyani[1]*, Yudi Srifiana[1], Nauval Audia Andamdewi[1]

[1] Faculty of Pharmacy and Science, University of Muhammadiyah Prof. DR. Hamka, Jakarta, Indonesia 13460
Email *: rahmahelfiyani@uhamka.ac.id

ABSTRACT

Aim: This study aimed to compare the diffusion rate of ketoconazole in microemulsion containing carbopol 934 or carbopol 941 as a thickener.

Methods: Ketoconazole microemulsion was made into 7 formulas with various concentrations in each type of thickener, i.e 0.1%, 0.2%, 0.3%, and one formula without thickener. Each formula were evaluated includes organoleptic, pH measurement, viscosity, particle size, potential zeta, surface tension measurement, and diffusion test.

Results: The physical properties of ketoconazole microemulsion showed liquid to gel form, yellow in color, specific odor, pH was in the range of 7.00-7.65, viscosity ranged from 2196-26037 cps, particle size was 13.6-42.2nm, zeta potential was -9.12--18.44 mV, and surface tension 30.7-64 dyne / cm. The diffusion kinetics obtained followed by the higuchi equation for the use of the two thickener, while the ketoconazole microemulsion without thickener (F1) followed the Korsmeyer-peppas equation with a diffusion rate of 9.444 %/minute. The diffusion rate of ketoconazole microemulsion with carbopol 934 (F2, F3, F4) were 4.354, 4.638, and 4.847 %/minute, respectively. Meanwhile, the diffusion rate using carbopol 941 (F5, F6, F7) were 4.695, 4.742, 4.751 %/minute, respectively.

Conclusion: It can be concluded that the diffusion rate (sig < 0,05,) of ketoconazole in microemulsion containing carbopol 941 was faster than diffusion rate of microemulsion containing carbopol 934 at the same concentration in the microemulsion system.

Keywords: microemulsion, thickener, carbopol 934, carbopol 941, diffusion rate

INTRODUCTION

Previous studies have shown that tween 80 can form ketoconazole microemulsion with Virgin Coconut Oil (VCO) at a concentration of 55% to produce the most stable microemulsion with a diffusion yield of 61.07% at 60 minutes [1]. However, the use of tween 80 above 50% can cause skin irritation [2]. The use of 35% tween 80 can produce ketoconazole microemulsion but does not show stability during 8 weeks of storage due to the low viscosity of the microemulsion [1]. Low viscosity of microemulsions can also prevent clinical applications due to unpleasant use and make preparations less attractive [3]. Addition of thickener can increase the viscosity of microemulsions. Increased viscosity can slow particle movement resulting in slower particle deposition or phase separation and

microemulsions becoming more stable. However, increasing viscosity can reduce the diffusion coefficient so that it affects the rate of diffusion of the active substance [4].
The thickener that can be used is carbopol which is used in most liquid and semisolid preparations, stable at low temperatures and antimicrobial [5]. There are various types of carbopol including carbopol 934 and carbopol 941. Carbopol 934 is acrylic acid which has crosslinking with sucrose allyl ether, whereas carbopol 941 is acrylic acid which has a crosslinked with pentaerithritol allyl ether [6].
This study aims to compare the rate of diffusion of ketoconazole in microemulsion with virgin coconut oil using carbopol 934 and carbopol 941 as thickener.

MATERIALS AND METHODS

Materials

The materials used were Ketoconazole (PT. Kalbe Farma), VCO (CV. Herba Bagoes), Tween 80 (PT. KAO), Sorbitol, Nipagin, Nipasol (PT. Kimia Farma), Carbopol 934 (Shrec Chemical), Carbopol 941 (Zhongtang), triethanolamine, and Aqua destillata.

Ketoconazol Microemulsion

Table 1: Ketoconazol Microemulsion [1]

Formula	(%)
Ketoconazol	0.4
VCO	5
Tween 80	35
Sorbitol	10
Nipagin	0.18
Nipasol	0.02
Aqua Destillata qs to	100

Nipagin, nipasol, and tween 80 are dissolved in aqua fervida ($80^0C \pm 2^0C$), then mixed until homogeneous, then the temperature is reduced to $40^0C \pm 2^0C$ (water phase). Ketoconazole is dissolved in VCO, then stirred using a magnetic stirrer at 500 rpm and a temperature of $40^0C \pm 2^0C$ for 15 minutes (oil phase). The oil phase is added to the water phase gradually while stirring using a magnetic stirrer at 500 rpm and a temperature of $40^0C \pm 2^0C$ for 15 minutes. Sorbitol is added drop by drop while stirring using a magnetic stirrer at 500 rpm and $40^0C \pm 2^0C$. Stirring was continued for 15 minutes [1].
The ratio between microemulsion and thickener used was 90:10 [7]. Ketoconazole microemulsion with the addition of thickener (Table 2) is made by dispersing carbopol in distilled water, then allowed to stand for 24 hours, 50% TEA solution is added dropwise until the pH dispersion of carbopol reaches pH 5.5, then the carbopol dispersion is slowly added to the ketoconazole microemulsion, while stirred using a magnetic stirrer at 300 rpm for 15 minutes at room temperature [7]. After 24 hours, preparations were evaluated including organoleptic, pH, viscosity, particle size, zeta potential, drug content and diffusion test.

Tabel 2: Formulas of Ketoconazol Microemulsion with Thickener

	Formula						
	F1	F2	F3	F4	F5	F6	F7
Ketoconazol Microemulsion	100ml	90ml	90ml	90ml	90ml	90ml	90ml
Carbopol 934 (%)	-	0.1	0.2	0.3	-	-	-
Carbopol 941 (%)	-	-	-	-	0.1	0.2	0.3
TEA 50% solution	-	0.37ml	0.89ml	1.35ml	0.57ml	1.34ml	1.67ml
Aqua Destillata qs to	-	100ml	100ml	100ml	100ml	100ml	100ml

Evaluation of Ketoconazol Microemulsion

Viscosity measurement is done using Brookfield viscometer (LV type) using spindle No. 63 with 30 rpm for F1, F2, F3, F5, F6, F7, and Spindle No. 64 with 20 rpm for F4. Microemulsion is inserted into a beaker glass until it reaches a volume of 250 ml, then attach the spindle to the specified limit. Surface tension measurements were carried out by the Du Nouy ring method. The force was determined by measuring the turning angle of the circular wire used to pull out the platinum-iridium ring from the effect of interfacial forces.

The particle size and zeta potential were measured using Nano Particle Size Analyzer (Beckman). The sample is inserted into the flow cell by using a syringe, then the flow cell is inserted into the device. The instrument will measure the sample for 9 minutes, after that it gives the results of particle size, potential zeta, and PD index samples.

Determination of the sample content was carried out using sample that an equivalent active ingredient of 4.14 mg and then added methanol to 10 ml then centrifuged at a 4000 rpm for 45 minutes, then 0.5 ml of supernatant put into a 10 ml volumetric flask, then add methanol to the boundary mark. Measure absorbance using a UV-Vis spectrophotometer (Shimadzu) at a wavelength of 243 nm [8].

Diffusion test is carried out using a modified flow-through method from Franz diffusion cells, at 37 ± 1^{o}C with receptor fluid 330 ml solution of 30% methanol phosphate buffer pH 7.4, and using 1 gram sample that placed on a membrane. The test was carried out for 8 hours, the samples were taken 10.0 ml and each take was carried out by replacing the new receptor fluid at the same volume. Sampling was carried out at minutes 5, 10, 15, 30, 45, 60, 80, 100, 120, 150, 180, 210, 240, 300, 360, 420, 480. The absorbance of sample was measured at a wavelength of 225.8 nm using UV-Vis Spectrophotometer [1,8].

RESULTS AND DISCUSSION

Results of Organoleptic Test

The results of organoleptic observation can be seen in Table 3. The form of ketoconazole microemulsion preparations becomes thicker due increases of the carbopol concentration.

Table 3: Organoleptic of Ketoconazol Microemulsion

Formula	Appearance	Color
F1	Solution	Clear Yellow
F2	Viscous solution	Clear Yellow
F3	Viscous solution	Cloudy yellow
F4	Gel	Cloudy yellow

F5	Viscous solution	Clear Yellow
F6	Gel	Clear Yellow
F7	Gel	Cloudy yellow

Results of pH Value

The results of pH measurements indicate that pH is in the range of 7.00-7.65 which is close to the normal skin pH range of 5 - 6.5 [9]. All formulas can meet the criteria of good and safe topical preparations.

Measurement of the pH of the microemulsion of ketoconazole before adding carbopol is 7.5-7.8. The thickness of carbopol is affected by pH. Base addition (TEA) will break the carboxyl group in carbopol and increase the negative charge which causes the carboxyl group to be ionized so that an electrostatic repulsion occurs. The repulsion force between ionized groups causes hydrogen bonds in the carboxyl group to stretch so that an increase in viscosity [10,11].

Results of Viscosity

Viscosity measurement results can be seen in Figure 1. The viscosity value of F4 using carbopol 934 is higher than the viscosity of F7 using carbopol 941 with the same concentration.

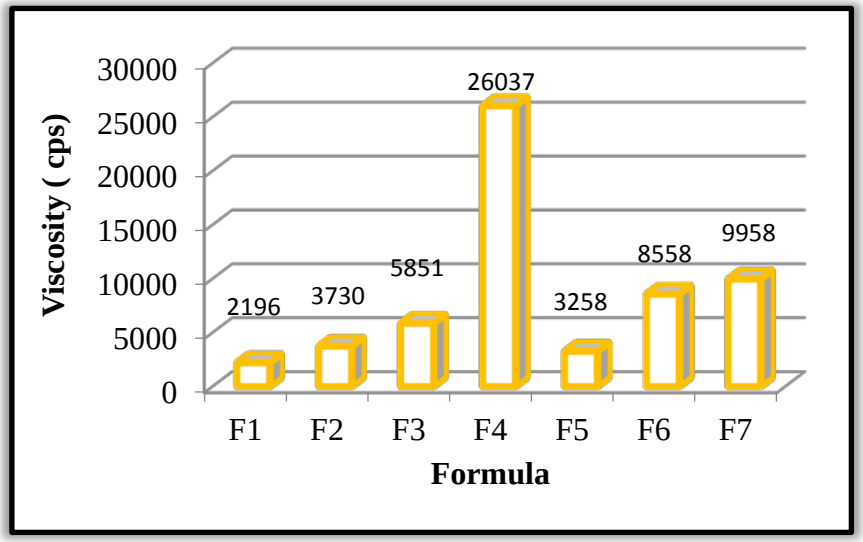

Figure 1: Results of Viscosity

The carbopol 941 dispersion is more acidic than the carbopol 934 dispersion. This is due to the crosslinking of carbopol 941 that is the pentaerithritol allyl ether is more acidic than the sucrose allyl ether which is the crosslinking of carbopol 941 (USP Convention 2007) [6]. These structural differences that affect the ability to stretch the polymer chain to trap water thus affecting the viscosity value.

Increased viscosity is proportional to the increase in the concentration of polymer used [12]. High viscosity will provide stability in the preparation because it will make the disperse medium become more rigid (stiff) and minimize the movement of dispersed phase droplets so that the possibility of coalescence can be prevented [13]. Increased viscosity will also increase retention time at the application site [14].

Results of Particle Size, Polydispersity Index (PdI), and Zetta Potential

Table 4: Particle size, Zetta Potential and PdI of Ketoconazol Microemulsion

Formula	Zetta Potential *	Particle size (nm)*	PdI*
F1	-9.12 ± 0.31	13.6 ± 1.0	0.241 ± 0.06
F2	-9.97 ± 2.47	18.4 ± 1.2	0.571 ± 0.00
F3	-6.17 ± 0.91	24.7 ± 0.1	0.571 ± 0.00
F4	-9.53 ± 0.98	33.6 ± 1.7	0.571 ± 0.00
F5	-9.69 ± 2.20	28.8 ± 0.1	0.571 ± 0.00
F6	-13.01 ± 1.15	37.6 ± 0.2	0.571 ± 0.00
F7	-18.44 ± 5.25	42.2 ± 0.6	0.571 ± 0.00

*n = 3

Polydispersity index value (PdI) provides information about droplet size uniformity of an emulsion [15]. From the measurement results obtained PdI F1 is 0.271 and PdI F2-F7 is 0.571. The addition of thickener can change the microstructure of the microemulsion which results in larger dispersion particles [16]. The enlarged particle size causes the viscosity and specific gravity to increase, but the particle size of the seven formulas is still included in the particle size requirement for microemulsion that is 10-100 nm [17].

The potential zeta indicates the charge of the particle in a specific medium. The stability of nanodispersion during storage can be predicted from potential zeta values. The potential zeta value of F1 is -9.12 while F2-F7 is in the range of -9.97 to -18.44. The addition of thickener can increase the stability of the microemulsion because microemulsion globules are trapped in the thickener base. When the globules are trapped in the thickener base, it will be difficult for the globule to join other globules because the outer phase of the microemulsion is a thick medium. This measure is the basis for the stability of an emulsion to the rate of creaming [4].

Results of Surface Tension

Surface tension was measured by Tensiometer Du Nuoy. The surface tension of the preparation is between 30.7-64 dyne / cm. The lowest surface tension is found in F1 which is a microemulsion without the addition of thickener. Linear surface tension value with viscosity measurement. This is because a larger force is needed to make the lamella torn on a thicker preparation.

Results of Ketoconazole Content

Determination of ketoconazole content in the preparation was carried out using a UV-Vis spectrophotometer at a wavelength of 243.00 nm in the medium of methanol. From the calibration curve, the regression equation y = 0.0322 x + 1.4762.10-3 with a correlation coefficient of 0.9999. The results of determining the ketoconazole content obtained were 88,0453-91,3459%.

Results of Diffusion Rate

The diffusion test used medium 30% methanol phosphate buffer pH 7.4. The ketoconazole calibration curve in 30% methanol phosphate buffer pH 7.4 at a wavelength of 225.80 nm produces a regression equation = 0.0384 x + 2.4367.10-4 with a correlation coefficient of 0.9999. The amount of diffuse ketoconazole can be seen in Figure 2 and the release kinetics can be seen in Table 5.

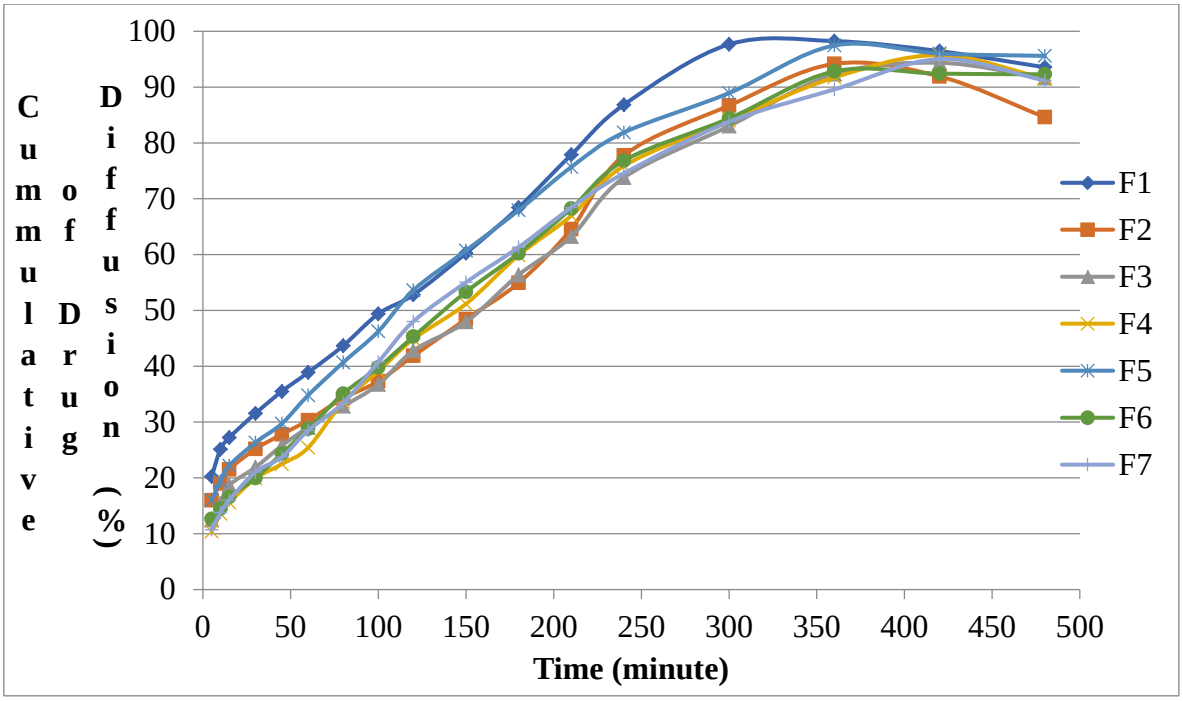

Figure 2: Diffusion profile of Ketoconazol from Microemulsion

From the diffusion profile it is seen that at percent diffused ketoconazole microemulsion with the addition of thickener is lower than the diffused percent microemulsion without thickener. In addition, the peak time at each increase in the concentration of carbopol becomes slower. This happens because the addition of thickener to the microemulsion can inhibit drug release [18].

Table 5: Diffusion Kinetics of Ketoconazole Microemulsion

Formula	Orde 0		Orde 1		Higuchi		Korsmeyer-peppas		
	r	k	r	k	r	k	r	k	n
F1	0.883 ± 0.036	0.177 ± 0.002	0.806 ± 0.056	0.003 ± 0.000	0.945 ± 0.021	4.465 ± 0.074	0.947 ± 0.043	9.474 ± 1.438	0.380 ± 0.031
F2	0.903 ± 0.040	0.176 ± 0.003	0.845 ± 0.041	0.003 ± 0.000	0.928 ± 0.027	4.354 ± 0.135	0.924 ± 0.027	6.529 ± 1.075	
F3	0.948 ± 0.010	0.187 ± 0.006	0.839 ± 0.020	0.004 ± 0.001	0.968 ± 0.014	4.638 ± 0.143	0.963 ± 0.009	4.757 ± 1.060	
F4	0.933 ± 0.023	0.195 ± 0.003	0.801 ± 0.022	0.004 ± 0.000	0.973 ± 0.005	4.847 ± 0.016	0.972 ± 0.004	3.613 ± 0.427	
F5	0.910 ± 0.023	0.187 ± 0.003	0.795 ± 0.028	0.003 ± 0.000	0.974 ± 0.002	4.695 ± 0.039	0.971 ± 0.008	6.611 ± 1.045	
F6	0.924 ± 0.036	0.190 ± 0.005	0.807 ± 0.059	0.004 ± 0.001	0.968 ± 0.004	4.742 ± 0.188	0.963 ± 0.010	4.348 ± 1.052	
F7	0.930 ± 0.016	0.190 ± 0.003	0.792 ± 0.020	0.004 ± 0.000	0.981 ± 0.002	4.751 ± 0.033	0.978 ± 0.003	4.032 ± 0.666	

*n = 3

The microemulsion without the addition of thickener (F1) follows the Korsmeyer-Peppas kinetics. In the Korsmeyer-Peppas equation the release mechanism depends on the value of

'n'. In Table 5 can be seen the value of ‘n 'of the preparation is 0.38. A value of n less than 0.45 indicates the mechanism of release of ketoconazole is the Fickian diffusion mechanism [19]. Drug release following Fickian diffusion means following Fick I law [20]. Whereas for microemulsions with the addition of thickener, F2-F6 follows Higuchi's kinetics. Preparations with release kinetics follow Higuchi's kinetics, their release can occur because the amount of drug in the carrier is greater than the number of drugs in the diffusion medium so that ketoconazole can diffuse from high concentrated areas to low concentrated areas after the diffusion medium penetrates the membrane and dissolves the drug. In addition, small drug particles can facilitate drug particles to penetrate the membrane [21].

The value of the diffusion rate constant (k) on the microemulsion with the addition of thickener has decreased compared to the microemulsion without thickener. This is according to the Stokes-Einstein law which states that viscosity is inversely proportional to diffusion. The thicker the preparation, the harder it will be to release the drug from the carrier. So that the diffusion rate constant is lower [4]. But the diffusion rate constant (k) increases as the concentration of carbopol increases. This can happen because carbomer exhibits very high adhesive bond strength in contact with tissues, enhancing the penetration of drugs [22].

The results of statistical tests on diffusion rate constants using Non Parametric analysis produced sig values < 0.05, then H0 was rejected, meaning that the average diffusion rate constant value of ketoconazole microemulsion without thickener was different from the diffusion rate constant of ketoconazole microemulsion with the addition of thickener. While parametric analysis is performed on the use of thickener types between carbomer 934 and carbomer 941 did not show a significant difference (sig value > 0.05) on the diffusion rate constant. Statistical analysis also showed that there is an interaction between the type of thickener and the concentration of thickener used to produce a significant difference in the diffusion rate constant (sig value < 0.05). based on tukey HSD, it can be seen that significant differences in the diffusion rate occur when using a thickener with a concentration of 0.1 and 0.3% (sig value < 0.05)

CONCLUSION

It can be concluded that the use of carbopol 941 results in faster diffusion rate of ketoconazole compared to carbopol 934 in microemulsion systems.

ACKNOWLEDGEMENTS

Thanks to all those who have helped make this research can run properly until the end.

REFERENCES

[1] Rahmah, E., Fith, K.N., Yunita, A., *Farmasains*. 2011, 1, 3:125 – 129.

[2] Bergh, M., *Allergenic Oxidation Products,* Scandinavian University Press, 1999.

[3] Gohel, M.C., Nagori, S,A., *Indian J.Pharm.Educ.Res.* 2010, 44 *(2)*: 2, 190 – 196.

[4] Sinko, P.J., *Martin’s Physical Pharmacy and Pharmaceutical Sciences: Physical Chemical and Biopharmaceutical Principles in the Pharmaceutical Sciences*. Lippincott Williams & Wilkins, Baltimore 2011.

[5] Ben, E.S., Muslim, S., Chazraj, C., Tomi, Y., *Optimasi Nanoemulsi Minyak Kelapa Sawit (Palm Oil) Menggunakan Sukrosa Monoester*, Prosiding Seminar Nasional Perkembangan Terkini Sains Farmasi dan Klinik III, 2013.

[6] United States Pharmacopeia Convention, *United States Pharmacopoeia National Formulary (USP) 30/NF 25*, Twinbrook Parkway: United States Pharmacopeial Convention, 2007.

[7] Patel, V.A., Makwana, S.B., Patel, B.A., Parmar, SJ., *Formulation and Characterization of Micro-emulsion based gel of Curcumin for the Management of Plaque Psoriasis*, Sardar Patel University, Gujarat 2014.
[8] Ravi, S., Vishnu, T., Chandra, P.M., Chandra, K.S., Dharampal, S., Sandhya, J., *American Journal of PharmTech Research.* 2015, 5 (5), 445 – 462.
[9] Heather, A.E., Adam, C.W., *Transdermal and Topical Drug Delivery : Principles and Practice*, A John Wiley & sons, Inc. New Jersey 2012.
[10] Tristiana, E., Noorma, R., Wing, H., Dien, R.J., *Majalah Farmasi Airlangga.* 2005, 5: 2.
[11] Barry, B.W., *Dematological Formulation*. Marcel Dekker Inc. New York 1983.
[12] Dhanekula, S., Prathia, S., Sadanandam, M., *Journal of Natural Pharmaceuticals.* 2013, *4*: 2.
[13] Laverius, M.F., *Optimasi Tween 80 dan Span 80 Sebagai Emulsifying Agent serta carbopol Sebagai Gelling Agent dalam Sediaan Emulgel Photoprotector Ekstrak Teh Hijau (Camellia sinensis L.):Aplikasi Desain Faktorial*, Skripsi Fakultas Farmasi Universitas Sanata Dharma, Yogyakarta 2011.
[14] Garg, A., Aggarwal, D., Garg, S., Sigla, A.K., 2002, Spreading of Semisolid Formulation, Pharmaceutical technology. www.pharmtech.com. Diakses tanggal 20 November 2016
[15] Agustinasari, I., Endang, Y.P., Niken, H., Sri, Y., *Jurnal Pascapanen.* 2014, 11 (1): 8.
[16] Zhu, W., Guo, C., Gao, Y., Cao, F., Zhai, G., *International Journal of Pharmaceutic.* 2009, *378*: 152-158.
[17] Agoes, G., *Sediaan Farmasi Likuida-Semisolid (SFI-7),* Penerbit ITB, Bandung 2012.
[18] Sah, A.K., Sunil, K.J., Ravi, S.P., *Asian Journal of Pharmaceutical and Clinical Research.* 2011, 4: 143.
[19] Shoaib, H.M., Merchant, H.A., Tazeen, J., Yousuf, R.I., *AAPS PharmSciTech.* 2006, 7 (3): 78.
[20] Kalam, M.A., Humayun, M., Parvez, N., Yadaz, S., Garg, A., Amin, S., Sultana, Y., Ali, A., *Journal Pharmaceutical Siciences*, 2007, 1 : 30-35.
[21] Dash, S.M., Padala, N., Nath, L., Prasanta C., *Acta Poloniae Pharmaceutica-drug Release.* 2010, *67(3)*:217-223.
[22] Alejandro, S,. Jose, dN., Bruno, S., *Progress in Polymer Science*. 2014, 39:2030–2075.