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Design and evaluation of aceclofenac gel containing different fixed oils as permeation enhancers

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Abstract

The objective of present work is to study the permeation effect of fixed oils taking poorly water soluble aceclofenac as model drug. Aceclofenac is a non-steroidal anti-inflammatory drug (NSAID) analogue of diclofenac, it is used for the relief of pain and inflammation in rheumatoid arthritis, osteoarthritis and ankylosing spondylitis. Oils as permeation enhancers are substances that interact with the major constituents of skin barrier, stratum corneum, to promote penetration of drugs into skin. In this study three types of fixed oils (olive oil, coconut oil and sesame oil) were used in two different concentrations (1% and 5%). Xanthan gum in two different concentrations was used as gelling agents. The gels were evaluated for several physico-chemical parameters. All the formulations have shown good physicochemical properties. Ex-vivo diffusion studies showed 90% drug release in 8 hrs. Anti-inflammatory studies on rats and skin irritations studies were performed. The F1 formulation was stable at room temperature for one month.

Keywords: Fixed oils, aceclofenac, permeation enhancers, ex-vivo, olive oil, coconut oil, sesame oil

Introduction

Oils as permeation enhancers are substances that interact with the major constituents of skin barrier, stratum corneum, to promote penetration of drugs into skin. It is a neutral, non-polar chemical substance which is thick at room temperature. Oils are lipophilic ("fat loving" or miscible with oils) and hydrophobic ("water fearing" or water immiscible) in nature. Sloppy and flammable nature is present in oils having high content of hydrogen and carbon. Oils could be from vegetable, animal or petro chemical sources and are volatile or non-volatile. Oil is qualified as virgin or refined rendering to the way they have been extracted. Virgin oils are extracted by pressing whereas refined oils are extracted out by solvents. Refined oil is the "standardized" oil obtained by modifying its fatty acids. But for the case of "virgin" oils, and therefore it is pure, without any foreign substances, and cannot have been submitted to any other treatments.

Natural oils play a promising role as permeation enhancers in TDDS. Penetration enhancers may be chemical, physical or formulation-based enhancer [1]. A penetration enhancer is a substance that is used to promote the delivery of an active pharmaceutical ingredient across the stratum corneum [2]. The most employed approach in penetration enhancement is the utility of chemical penetration enhancers; they interfere with well-arranged lipid layer of stratum corneum [3]. Penetration enhancers produce their action either by interrupting highly sequenced lipid structure of stratum corneum or by interacting with intercellular proteins. Another mode of action is improvement in the partition co-efficient of the drug which acts as co-enhancer into the stratum corneum [4].

Aceclofenac is a non-steroidal anti-inflammatory drug (NSAID) analogue of diclofenac. It is used for the relief of pain and inflammation in rheumatoid arthritis, osteoarthritis and ankylosing spondylitis [5, 6]. A unique feature of aceclofenac's pharmacology is that it stimulates glycosaminoglycans (GAG) synthesis, which in turns enhances skin permeation of NSAIDS [7]. Coconut oil, olive oil and sesame oil are used as penetration enhancers that increase the skin permeability by certain alteration in the lipid layer of stratum corneum and by perturbation of intercellular lipids and protein domain integrity [8]. Fixed olive oil expressed from the ripe fruit of *Olea europaea* is also known as oleum olive. Sesame oil obtained by expression from the seeds of *sesamum indicum* (pedaliaceae) is also known as Teel oil, gingelly oil, benne oil.

Coconut oil also known as edible oil rich in short and medium chain fatty acid along with 92% saturated fatty acid and 45- 56 % lauric acid^[9]. The objective of present work is to study the permeation effect of fixed oils taking poorly water soluble aceclofenac as model drug which used in the treatment of osteoarthritis, and ankylosing spondylitis.

Materials and Methods

Aceclofenac (Gift sample, Suven Life Sciences limited), carbopol-934, Xanthan gum, Sodium CMC, Methanol, Propylene glycol (S.D. Fine Chemicals Ltd), Triethanolamine (Molychem. Ltd.), Olive oil (Oleomonterreal SL), Marketed olive oil (Deoleo S.A.), Marketed coconut oil (ITC Ltd, Marico), Marketed sesame oil (V.V.V and Sons Edible oil Ltd), Coconut oil, Sesame oil (Cold compressed).

Preparation of gels

Different transdermal gels were formulated by dispersion method. Xanthan gum & Carbopol 934 P, were weighed and soaked for 2-3 hrs in a beaker containing distilled water. After 2-3hrs the swollen polymers were stirred for 400-600 rpm. Then 5-6ml of propylene glycol was added slowly with stirring. Accurately weighed aceclofenac was dissolved in 3ml of ethanol and the ethanolic solution of drug was added slowly in the previously prepared polymer gel under stirring (400-600 rpm). Penetration enhancers were then added directly and was stirred. The final quantity was made up to 10gm with distilled water.

Drug excipient compatibility studies

The spectrum analysis of aceclofenac and polymer which employed in the preparation of gels was studied by Fourier Transform Infra-Red (FTIR) Spectroscopy. FTIR spectra were recorded by preparing potassium bromide (KBr) disks using a Shimadzu Corporation (Kyoto, Japan) facility (model - 8400S). Potassium bromide (KBr) disks were prepared by mixing few mg of sample with potassium bromide and then by compacting it using hydrostatic press under vacuum of 6-8 tons pressure. The resultant disc was mounted in a suitable holder in IR spectrophotometer and the IR spectrum was recorded from 4000 cm⁻¹ to 200 cm⁻¹. The resultant spectrum was compared for any spectral changes. Then they were observed for the incidence of characteristic peaks for the respective functional group in the compound. FTIR study was carried out to check compatibility of drug and excipients^[10].

Solubility studies

Solubility studies were done using orbital shaker bath. Saturated solutions of aceclofenac were prepared by adding drug, distilled water and appropriate quantity of fixed oil as permeation enhancers. Saturated solutions were kept in an orbital shaker, centrifuged for 15 min at 3000 rpm. Aliquots were filtered through Whatman No. 41 filter paper. The filtrates were diluted in distilled water and assayed spectrophotometrically at 273.4 nm. The experiment was repeated three times in the same medium and a calibration curve was determined from the mean value^[10, 11].

Determination of pH

Digital pH meter was used to determine the pH of the prepared gel. One gram of gel was dispersed in 100 ml of distilled water and stored for two hours at constant temperature. The measurement was done in triplicate and average values were calculated.

Drug content

Aceclofenac content in gel was measured by dissolving 100 mg of gel (equivalent to 5mg of drug) in 10 ml solvent (methanol) by Sonication. The solution was passed through the Whatmann filter paper no.42 and filtered. Absorbance was measured after suitable dilution at 274 nm in UV/VIS spectrophotometer. The experiment was done in triplicate and average values were calculated^[12].

Spreadability

Spreadability of the formulated gel was determined by measuring the spreading diameter of 1g of gel between 20x20 cm glass plates after 1 min. The mass of the upper plate was standardized at 150 g.

The spreadability was calculated by means of the formula

$$S = \frac{ml}{t}$$

Where,

S = spreadability,

m = weight tied to the upper glass slide,

l = length of the glass slide,

t = time taken in seconds

Extrudability

Pfizer hardness tester was used to determine the extrudability of the gel. A 15 gm of gel was filled in aluminium tube. The plunger was attuned to hold the tube properly. Pressure of 1kg/cm² was applied for 30 sec. The quantity of gel extruded was weighed. The procedure was repeated at three equidistance places of tube. Test was carried in triplicate^[13].

Homogeneity

It was determined by visual inspection for the presence of any aggregates.

Ex vivo permeation studies using rat skin

Ex vivo drug permeation studies were performed using rat skin in Franz diffusion cell. The cell was locally fabricated, and the volume of receptor compartment was 25 ml. The thawed rat skin was mounted onto diffusion cell such that the dermis side was in constant contact with receptor containing pH 6.4 phosphate buffer saline. 250 mg of gel was applied to the stratum corneum facing the donor compartment and the hydrodynamics in the receptor compartment were maintained by stirring on magnetic stirrer at 800 rpm at 37°C. 1ml sample was withdrawn at predetermined time intervals for 12 hours and drug content was analysed by UV-VIS double beam spectrophotometer at 274 nm.

Ex-vivo permeation rate studies such as % drug release, steady state transdermal flux (SSTF), permeability coefficient, lag time and enhancement ratio for percutaneous absorption of aceclofenac were calculated^[14].

Skin irritation studies

Skin irritation studies were performed on rabbits after it was approved by the Institutional animal ethical committee (IAEC) in G. Pulla Reddy College of Pharmacy Registration number 320/CPCSEA and student ID number: GPRCP/IAEC/20/16/2/PCE/ACE-8. The experiment was carried out for 3 days and the application sites were graded according to a visual scoring scale.

Anti-inflammatory of rats

Anti-inflammatory studies were performed on rats. The paw thickness was measured at 0, 1, 2, 3 and 4 hr after carrageenan administration using Vernier calliper. The anti-inflammatory activity was calculated as percentage inhibition of oedema in the animals treated with extract under test in comparison to the carrageenan control group.

The percentage (%) inhibition of edema is calculated using the formula

$$\% \text{ Inhibition} = (T_o - T_t) / T_o \times 100$$

Where *T_t* is the thickness of paw of rats given test extract at corresponding time and *T_o* is the paw thickness of rats of control group at the same time. Results were summarised in

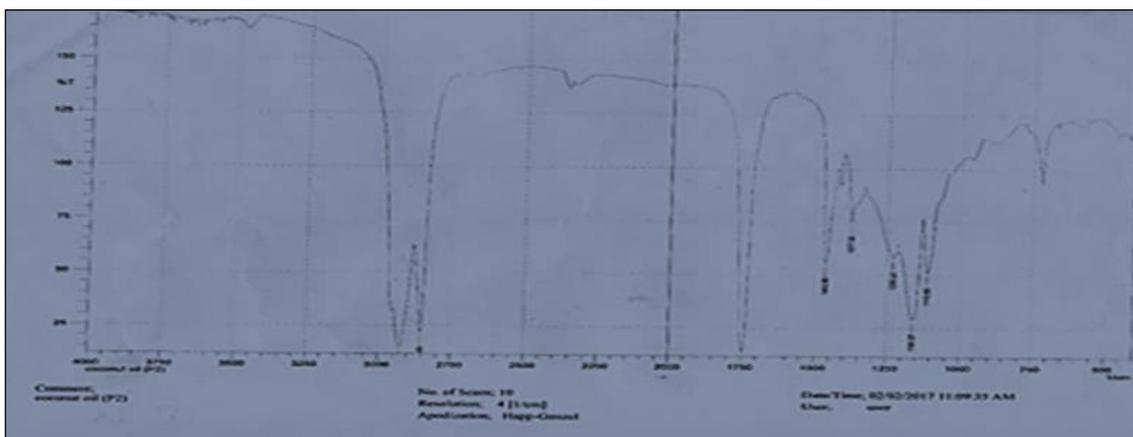


Fig 1: FT-IR of the optimized formulation

Solubility studies

The solubility studies of aceclofenac indicated that the drug solubility increases with increase in the concentration of ethanol. Aceclofenac has higher solubility in unsaturated fatty acids than in triglycerides. Olive oil and sesame oil contains more percentage of unsaturated fatty (Oleic acid and linoleic acid) and this could be the reason for the high solubility of aceclofenac in olive oil than in other oils which do not contain unsaturated fatty acid [11].

pH of prepared transdermal gels

The pH was found to be in range from 5.2 to 6.93 thus indicating suitability for skin application along with good extrudability and spreadability.

Spreadability

The value of spreadability varies from 10.1–12.9 g.cm/s indicating that the gels easily spreadable by small amount of shear. All gel preparations indicated a good spreadability. The

table 4.

Stability study

Stability studies were carried out by keeping optimized formulations in glass containers for one month at room temperature.

Result and Discussion

Drug excipient compatibility studies

Aceclofenac compatibility with excipients was studied by FTIR. From FTIR spectrums of the identified in the pure drug were relatively same when compared with the blend, indicating no drug polymer interaction i.e. the pure drug was not altered functionally and compatible with polymers.

results were abridged in table 1.

Homogeneity

It was evaluated by visual observation. All formulated transdermal gel showed good homogeneity without lumps. The physical appearances of all gel formulations were opaque in nature. The results were abridged in table 1 [15].

Drug content

The content of drug per 100mg of gel ranged from 95.16% to 99.03% which indicates that uniform dispersal of drug in the formulations.

Extrudability

The extrusion of the gel from the tube is important during its application and for patient acceptance. The extrudability of all the formulations was found to be good and compatible. The results were abridged in table 1.

Table 1: Physico-chemical evaluation of prepared transdermal gel

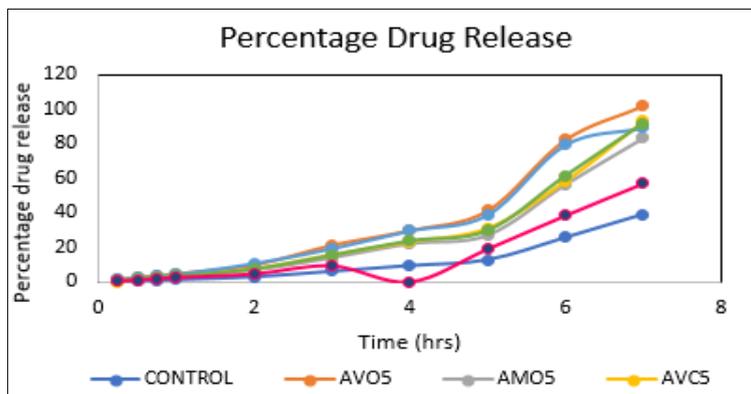
Formulation code	pH	Drug content (%)	Homogeneity	Spreadability (g.cm/sec)	Extrudability
FX	7.09±0.03	98.31±0.25	+++	10.3 ±0.04	+++
FC	7.06±0.08	97.7±0.08	+++	10.5±0.54	+++
F1	6.17 ±0.26	99.03±0.71	+++	10.7±0.87	+++
F2	5.35 ±0.01	98.75±0.58	+++	12.9±0.55	+++
F3	6.87±0.06	97.07±0.21	++	11.4 ±0.19	+++
F4	5.51 ±0.01	97.72±0.08	+++	12.3±0.02	++
F5	6.92±0.08	96.63±0.94	+++	10.2±0.09	+++
F6	5.60 ±0.03	95.16±1.03	+++	12.0±0.05	++

Ex-vivo permeation studies

The ex-vivo permeation studies on rat abdominal skin, showed varied results because the aceclofenac transdermal gel formulations containing fixed oils act as permeation enhancers showed effect on the surface of lipophilic skin by interacting with skin or by rupturing skin integrity, which does not show on dialysis membrane hence ex-vivo studies were directly performed.

Drug release from gels containing olive oil was faster than those containing the same percentage of sesame oil and coconut oil. Formulation F1 containing virgin olive oil as permeation

enhancer has shown maximum permeation i.e., more than 90% of aceclofenac was permeated from the gel formulations, this could be due to the presence of high concentration of poly unsaturated fatty acids compared to other formulations.¹⁶ Virgin olive oil contain 84% of oleic acid (mono-unsaturated fatty acid), which could have enhanced the permeation by non-polar pathway.^{17,18} It was observed that the rate of drug release increased with increase in concentration of oil as permeation enhancer. Figure 1 represents the percentage drug release of the formulations.



Note: AVO5(F1)- Aceclofenace virgin olive oil 5%, AMO5(F2)-Aceclofenace marketed olive oil 5%, AVC5(F3)- Aceclofenace virgin coconut oil 5%, AMC5(F4)-Aceclofenace marketed coconut oil 5%, AVS5(F5)- Aceclofenace virgin Sesame oil 5%, AMS5(F6)- Aceclofenace marketed sesame oil 5%.

Fig 2: Percentage Drug Release

Permeability parameters of optimized gel formulations

The ex-vivo permeability parameters (lag time, Q₈, flux, permeability coefficient, and enhancement ratio) were

calculated for all optimised formulation and the results were tabulated in table 2.

Table 2: Permeability parameters of control and optimized gel formulations

Formulation	Lag time (hr)	Q ₈ (µg/cm ²)	Flux (µg/cm ² /hr)	Permeability coefficient (cm/ hr)	Enhancement ratio
F1	0.27±0.006	231.71	31.21	4.26	7.6
F2	0.99±0.0085	211.71	28.69	3.83	5.2
F3	1.09±0.007	225.62	30.0	4.16	6.6
F4	0.69±0.003	170.06	20.76	3.55	6.5
F5	0.72±0.004	221.39	26.59	4.01	6.3
F6	0.34±0.07	168.45	19.2	2.76	5.0

Note: AVO5(F1)- Aceclofenace virgin olive oil 5%, AMO5(F2)- Aceclofenace marketed olive oil 5%, AVC5(F3)- Aceclofenace virgin coconut oil 5%, AMC5(F4)- Aceclofenace marketed coconut oil 5%, AVS5(F5)- Aceclofenace virgin Sesame oil 5%, AMS5(F6)-Aceclofenace marketed sesame oil 5%.

Model dependent kinetics of optimized aceclofenac gel formulations

Model dependent kinetics was done for all optimized gel formulations in order to determine the release kinetics, release mechanism drug transport mechanism; the results were tabulated in table 3. It was found that F2, F4 & F6 formulations follow zero order drug release kinetics and F1, F3 and F5 formulations follow first order kinetics; the drug release

mechanism for F4 was found to be follow Higuchi drug release mechanism while F1, F2, F3, F5, and F6 was found to be follow Korsmeyer-peppas drug release mechanism.

From the values of release component “n” it can be concluded that all formulations have Anomalous diffusional release mechanism.

The optimized formulations from ex-vivo studies were further evaluated for skin irritation studies and stability studies.

Table 3: Ex-vivo drug release kinetics of optimized formulations

Form. code	r ²				N	Drug transport mechanism
	Zero	First	Higuchi	Peppas		
F1	0.985	0.990	0.963	0.991	0.881	Anomalous transport
F2	0.961	0.935	0.845	0.967	0.913	Anomalous transport
F3	0.980	0.991	0.967	0.989	0.956	Anomalous transport
F4	0.977	0.989	0.966	0.937	1.069	Case-II transport
F5	0.985	0.994	0.845	0.971	0.697	Anomalous transport
F6	0.984	0.972	0.9332	0.990	0.944	Anomalous transport

Skin irritation studies

Skin irritation studies were conducted on depilated rabbit. Skin reaction at the site of application was assessed and scored according to Draize method. The formulation F1 & placebo gel showed irritation potential of “0”, thus proving to be non-irritant. The “0” value in an irritancy test indicates that the applied formulations are generally non-irritant to the human skin. No obvious erythema and edema was observed after 72 hours of the application of the formulations.

Anti-inflammatory studies



Fig 3: Skin Irritation Studies

Anti-inflammatory studies of optimized formulation F6 were performed and compared with marketed preparation HIFENAC GEL®.

The results obtained from the table no 4; have shown a favourable decreasing of the hind paw volume of the rat after using optimized formulation F1 when compared with the paw volume of the control and marketed product (HIFENAC GEL®). The optimized formulation contained permeation enhancers (virgin olive oil) which is enhancing the permeation of the gel formulation through stratum corneum. Hence, increasing the flux, permeability coefficient and finally increasing the therapeutic effectiveness of the gel formulation in minimum time was achieved.

Table 4: Anti-inflammatory studies on rats

Groups	Change in paw Thickness (mm) Mean ± SD (% inhibition)			
	1 hr	2hr	3 hr	4 hr
Control	0.82±0.008	0.82±0.01	0.83±0.009	0.83±0.014
Group I	1.33±0.03	2.35±0.03	3.64± 0.07	3.41±0.02
Group II	0.55±0.04 (55.9%)	0.77±0.02 (66.24%)	1.14±0.03 (68.82%)	0.92±0.03 (72.13%)
Group III	1.00±0.005 (25%)	1.53±0.01 (35.02%)	1.83±0.02 (50.09%)	1.42±0.014 (56.34%)

Note: Values are expressed as mean ± SD, n=3,
Group I- Inflammation with carrageenan 1% (w/v),
Group II- Inflammation treated with F1.
Group III- Inflammation treated with marketed gel. P<0.05 v/s carrageenan control,
Note: P< 0.05 is significant; P< 0.001 is highly significant



Fig 4: Photographs of Rat Paw after the treatment

Stability studies

The stability studies were performed on the F1 formulation. The stability studies were conducted for one month at room temperature and it was found to be stable, with insignificant change in the appearance, drug content, viscosity and pH.

Conclusion

Aceclofenac gels were prepared using virgin and marketed fixed oils as permeation enhancers and various concentrations of the same were optimized. The results were more promising that virgin olive oil containing maximum concentration of oleic acid (84%) have shown better permeation compared to marketed oils and thereby enhancing penetration of drug through stratum corneum.

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None

Conflict of interest

The authors declare that they have no conflict of interest to disclose.

References

- Rao S, Barot T, Rajesh K, Jha L. Formulation, optimization and evaluation of microemulsion based gel of butenafine hydrochloride for topical delivery by using simplex lattice mixture design. *J Pharm Investig.* 2016; 46, 1-12.
- Sloan K, Wasdo S. The role of prodrugs in penetration enhancement. Taylor and Francis, NY, USA. 2006; 51-64.
- Walker RB, Smith EW. The role of percutaneous penetration enhancers. *Adv Drug Deliv Rev.* 1996; 18:295-301.
- Barry BW. Dermatological formulations: percutaneous absorption. 1st ed., Info health care, 1983, 18-23.
- Dooley, Mukta, Spencer, Caroline M. Dunn, Christopher J. Aceclofenac: A Reappraisal of its Use in the Management of Pain and Rheumatic Disease, *Drugs* 2001; 61(9):1351-1378.
- Ina Morton, Judith Hall. The Royal society of medicines. Bloomsbury Publishing Plc38 Soho square London 2002 W1D3HB.
- Chika J, Charles O. Transdermal delivery of Gabapentin: Effect of cosolvent and microemulsion on permeation through rat skin. *Pharmacology & pharmacy.* 2014; 5:471-478.
- Hussain A, Khan GM, Shah SU, Shah KU, Rahim N and Wahab A. Development of a novel ketoprofen transdermal patch: Effect of almond oil as penetration enhancers on in-vitro and ex-vivo penetration of ketoprofen through rabbit skin. *Pak J Pharm Sci.* 2012; 25:227-232.
- Gopala KAG, Gaurav RASB, Prasanth KPK, Preeti C. Coconut oil: chemistry, production and its application - a review. *Ind Cocunut J.* 2010; 8:15-27.
- Amit Kumar Nayak. Comparative Evaluation of in Vitro Diclofenac Sodium Permeability Across Excised Mouse Skin from Different Common Pharmaceutical Vehicles, *International Journal of pharmtech Research.* 2010; 2(1): 920-930.
- Amit C, Upendra N, Neha G, Sharma VK, Khosa RL. Enhancement of solubilization and bioavailability of poorly soluble drugs by physical and chemical modifications: A recent review. *J Adv Pharm educres.* 2012; 2(1):32-67.

12. Tas C, Ozkan Y, Savaser S, Baykara T. In vitro release studies of chlorpheniramine maleate from gels prepared by different cellulose derivatives. *IL Farmaco*. 2003; 58:605-11.
13. Jeevan Sen. Formulation and Evaluation of Muco-adhesive In-situ Gel for Site-specific Delivery of Clotrimazole. *Research & Reviews: Journal of Pharmaceutics and Nanotechnology*, 2014.
14. Japan Patel. Formulation and Evaluation of Topical Aceclofenac Gel Using Different Gelling Agent. *Journal of Drug Delivery & Therapeutics*. 2013; 3(6):51-53.
15. Patel J, Patel B, Banwait H, Parmar K, Patel M. Formulation and evaluation of topical aceclofenac gel using different gelling agent. *Int J Drug Dev Res*. 2011; 3:156-64.
16. Hussain A, Khan GM, Jan SU, Shah S, Shah K, Akhlaq M, Rahim N, Nawaz A, Wahab A. Effect of olive oil on transdermal penetration of flurbiprofen from topical gel as enhancer. *Pak Journal of pharmasciences*. 2012; 25(2):365-369.
17. Suchika Sharma, Geeta Aggarwal, Sanjudhawan. Design and evaluation of Olanzapine transdermal patches containing vegetable oils as permeation enhancers. *Scholars Research Library. Der Pharmacia Lettre*. 2010; 2(6):84-98.
18. Muhammad Zubair Malik. Solubility and Permeability Studies of Aceclofenac in Different Oils, *Tropical Journal of Pharmaceutical Research*. 2014; 13(3):327-330.