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RESEARCH ARTICLE

COMPARATIVE EVALUATION OF *IN SITU* INTESTINAL ABSORPTION OF ACECLOFENAC FROM SOLID DISPERSIONS, *β*-CYCLODEXTRIN COMPLEXES AND CO-PRECIPITATES IN RATS

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Aceclofenac (AF), a new generation nonsteroidal anti-inflammatory drug with a good tolerability profile in a variety of painful conditions. Aceclofenac is proven to be effective for both acute and chronic inflammatory and degenerative diseases such post traumatic pain, cervical pain, rheumatoid arthritis and osteoarthritis. Aqueous solubility of aceclofenac was enhanced by preparing its solid dispersions and co-precipitates using polyvinyl pyrrolidone (PVP) as water soluble carrier and cyclodextrin complexes with *β*-cyclodextrin. Absorption studies using *in situ* rat gut technique exhibited greater rate of intestinal absorption with co-precipitates of aceclofenac when compared with solid dispersions and *β*-cyclodextrin. The intestinal absorption followed the first order rate kinetics. Statistical correlation of *in vitro* drug dissolution and *in vitro* drug absorption indicates a positive correlation (R^2= 0.931 to 0.964). This increased absorption may be due to the solubilization and improved wetting of AF in PVP rich micro-environment.

Key words: Aceclofenac, Solid dispersion, *β*-Cyclodextrin, *In situ* absorption, Polyvinyl pyrrolidone.

INTRODUCTION

A method is reported for studying gastro-intestinal drug absorption from isolated gut segments of the anesthetized rat *in situ*. The experimental technique is simple and utilizes readily available laboratory equipment. The results are closely reproducible and yield absorption rates which are realistic in terms of the known absorption behavior of drugs in humans and intact animals (Martin and Doluisio, 1977). Solid dispersion is a unique approach in which the drug is dispersed in extremely fine state in an inert water-soluble carrier in solid state (Damian *et al* 2002). Polyvinyl pyrrolidone (PVP) has been used extensively for the enhancement of solubility and dissolution rate of low solubility drugs. PVP-coprecipitate of water-insoluble drugs is formed by dissolving both components in a common solvent and subsequently removing the solvent (Anastasiadou *et al* 1983). Cyclodextrins and their derivatives play an important role in formulation development due to their effect on solubility, dissolution rate, chemical stability and absorption of drugs. Though cyclodextrins have been investigated widely during the last two decades, their commercial application in pharmaceutical formulation was started only in recent years with drugs such as piroxicam and nimesulide (Jun *et al* 2007). Aceclofenac (AF) is a new generational non-steroidal anti-

inflammatory drug showing effective anti-inflammatory and analgesic properties and a good tolerability profile in a variety of painful conditions like ankylosing spondylitis, rheumatoid arthritis and osteoarthritis. Aceclofenac is very slightly soluble in water (Moffat *et al* 1999) and therefore attempt has been made to prepare its solid dispersions and co-precipitates using PVP as water-soluble carrier and cyclodextrin complexes with *β*-cyclodextrin (*β*-CD) with an aim to improve its extent and rate of dissolution and to carry out its absorption studies using *in situ* rat gut technique.

MATERIALS AND METHODS

Aceclofenac (Ipca Laboratories, Mumbai), *β*-cyclodextrin (Cerestar, USA Inc. Hammond Indiana) of commercial purity grade were used. All other chemicals used were of analytical reagent grade.

Preparation of solid dispersions of aceclofenac

Weighed quantities of polyvinyl pyrrolidone (PVP) and aceclofenac (AF) in different ratios such as 1:0.5, 1:1 and 1:2 by weight basis were thoroughly mixed and melted on hot plate with constant stirring to obtain a uniform melt. The melt was shock cooled on an ice cooled stainless steel plate. The solid mass was removed from the stainless steel plate, powdered and kept in a desiccator for two days. The powder was passed through sieve #100 and stored in closed airtight container (Patil and Gaikwad, 2009).

Preparation of co-precipitates of aceclofenac

AF-PVP and PVP co-precipitates were prepared in different ratios such as 1:0.5, 1:1 and 1:2 by weight basis with slow evaporation of ethanolic (95% *v/v*) solutions of drug and carrier in a vacuum oven at 40°C. The resulting solid mass was further dried under vaccum to a constant weight and stored in desiccator (Shah *et al* 1999).

Preparation of molecular inclusion complexes of aceclofenac

Solid inclusion complexes of AF with *β*-CD were prepared in 1:1 and 1:2 molar ratios using kneading method. Accurately weighed quantity of *β*-CD was taken in a glass mortar and water was added slowly followed by mixing to obtain a homogeneous paste. Weighed quantity of AF was added slowly by grinding. The mixture was ground for one hour. During this process, appropriate quantity of water was added to maintain suitable consistency. The obtained solid mass was further dried under vacuum to a constant weight at room temperature (RT) and pulverized followed by sieving through mesh #100. Finally, shieved material was stored in desiccator (Patel *et al* 2007; Dua *et al* 2007a; 2007b).

In situ rat gut technique

The extent of absorption of AF from selected solid dispersions, *β*-CD molecular inclusion complex and co-precipitates which had shown good *in vitro* results were determined in the rat intestine (Dua *et al* 2007a; 2010). The experiments were carried out as per the guidelines of animal ethics committee. Six rats of either sex weighing between 200-250 g were fasted for 2 days prior to experiment. Rats were anaesthetized by administering pentobarbital (60 mg/kg, *i.p.*) and placed on a heated pad to keep normal body temperature. Small intestine of the animals was exposed by a midline abdominal incision. The duodenal and ileal ends of the intestine were cut while keeping the blood supply to intestine intact. Two L-shaped glass cannulae were inserted and secured by ligation with silk suture in the small slits at the duodenal and ileal ends of the small intestine which was returned to the abdominal cavity to maintain its integrity. Four-centimeter segments of Tygon tubing were attached to the exposed ends of both cannulae and a 30 ml hypodermic syringe was fitted with a three way stopcock (**Figure 1**). Perfusion fluid (anhydrous disodium hydrogen phosphate - 40 mM; sodium dihydrogen phosphate - 26 mM and sodium chloride 119 mM) was passed slowly through the duodenal cannula at 37°C and passed out through the ileal cannula until all the intestinal contents were expelled out from the intestine. Air was pumped through the syringe to expel the perfusion fluid from the gut. The drug solution (10 ml) was immediately introduced into the intestine by means of the syringe. An aliquot of 0.1 ml of solution was withdrawn at 0, 15, 30, 45, 60, 75, 90 min from the time of administration of the drug solution. To ensure uniform drug solution concentration throughout the gut segment, aliquots were removed from the two syringes alternatively. Finally, the animal was euthanitized with a cardiac injection of saturated solution of potassium chloride. After making suitable dilutions, absorbance was measured and the amount of drug present in the sample solution was calculated from the regression

equation (Martin-Algarra *et al* 1995; Zakeri-Milani *et al* 2007).

RESULT AND DISCUSSION

The intestinal absorption of AF from these formulations demonstrated the following order: CPs > SDs > β-CD molecular inclusion complex > AF (**Table 1, 2; Figure 2, 3**). The absorption order of AF from these formulations corresponds to its *in vitro* release pattern. A statistically significant difference was observed in the rate of absorption of AF from AF-PVP-CP3 (1:2) and AF-PVP-SD3 (1:2) when compared with AF-βCD2 (1:2 M) and pure drug as well ($P < 0.05$). This increased absorption may be due to the solubilization and improved wetting of AF in PVP rich micro-environment (Kumar *et al* 2008). The correlation coefficient (R^2) values and equations best describing the kinetics of drug absorption are given in **Table 2** and **Figure 3**. The release of AF from all these formulations was found to follow first order release kinetics since value of R^2 for first order was higher in comparison to zero order. The present findings were in agreement with the reports carried out with COX-II inhibitors (Rawat and Jain, 2007).

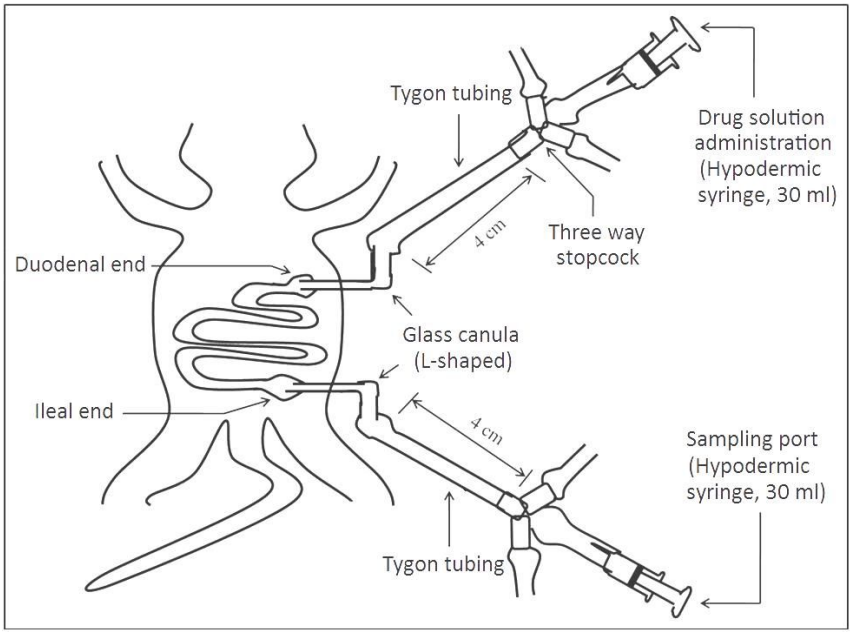

Figure 1. Arrangement for carrying out *in situ* rat gut technique

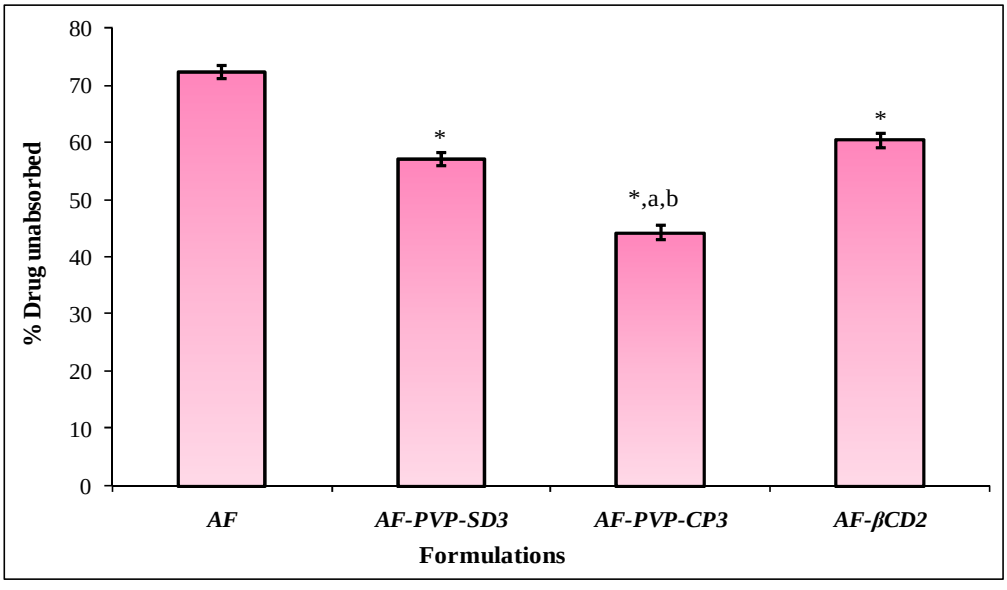

Figure 2. Comparative analysis of intestinal absorption of aceclofenac from selected formulations. Values are mean ± SD. **P*<0.05 *Vs* AF; [a]*P*<0.05 *Vs* AF-PVP-SD3; [b]*P<0.05 Vs* AF-βCD2

Table 1. Comparison of intestinal absorption of selected formulations of aceclofenac using *in situ* rat gut technique

Time (min)	Percent aceclofenac unabsorbed				Log percent aceclofenac unabsorbed			
	AF	AF-PVP-SD3	AF-PVP-CP3	AF-βCD2	AF	AF-PVP-SD3	AF-PVP-CP3	AF-βCD2
0	100 (0.34)	100 (0.23)	100 (0.29)	100 (0.19)	2.000	2.000	2.000	2.000
15	98.43 (0.92)	98.71 (0.73)	98.12 (0.68)	99.01 (0.71)	1.993	1.994	1.992	1.996
30	97.17 (0.91)	84.76* (0.89)	77.76*,a,b (0.95)	84.43* (0.86)	1.988	1.928	1.891	1.926
45	84.64 (0.96)	72.01* (0.92)	68.43*,b (0.88)	74.12* (0.98)	1.928	1.858	1.835	1.870
60	80.51 (0.91)	67.15* (0.89)	58.12*,a,b (0.97)	68.19* (0.87)	1.906	1.827	1.764	1.834
75	79.19 (0.99)	61.54* (0.92)	51.45*,a,b (0.97)	64.18* (1.04)	1.899	1.789	1.711	1.807
90	72.31 (1.19)	57.12* (1.05)	44.24*,a,b (1.34)	60.35* (1.23)	1.859	1.757	1.646	1.781

AF: aceclofenac; βCD: β-cyclodextrin; CP: coprecipitates; PVP: polyvinyl pyrrolidone; SD: solid dispersion. Values in parenthesis indicates the standard deviation (n = 6). *$P<0.05$ *Vs* AF; [a]$P<0.05$ *Vs* AF-PVP-SD3; [b]$P<0.05$ *Vs* AF-βCD2

Table 2. Comparison of orders of intestinal absorption (*in situ* rat gut technique) of selected formulations of aceclofenac.

Formulation	Regression equations	
	Zero order	First order
AF	y = -0.3291t + 102.27 R^2 = 0.9396	y = -0.0017t + 2.0131 R^2 = 0.9408
AF-PVP-SD3	y = -0.5252t + 100.97 R^2 = 0.9574	y = -0.003t + 2.0119 R^2 = 0.9737
AF-PVP-CP3	y = -0.6673t + 101.19 R^2 = 0.9697	y = -0.0042t + 2.0218 R^2 = 0.9852
AF-βCD2	y = -0.4877t + 100.56 R^2 = 0.9498	y = -0.0027t + 2.0085 R^2 = 0.9672

AF: aceclofenac; βCD: β-cyclodextrin; CP: coprecipitates; PVP: polyvinyl pyrrolidone; SD: solid dispersion

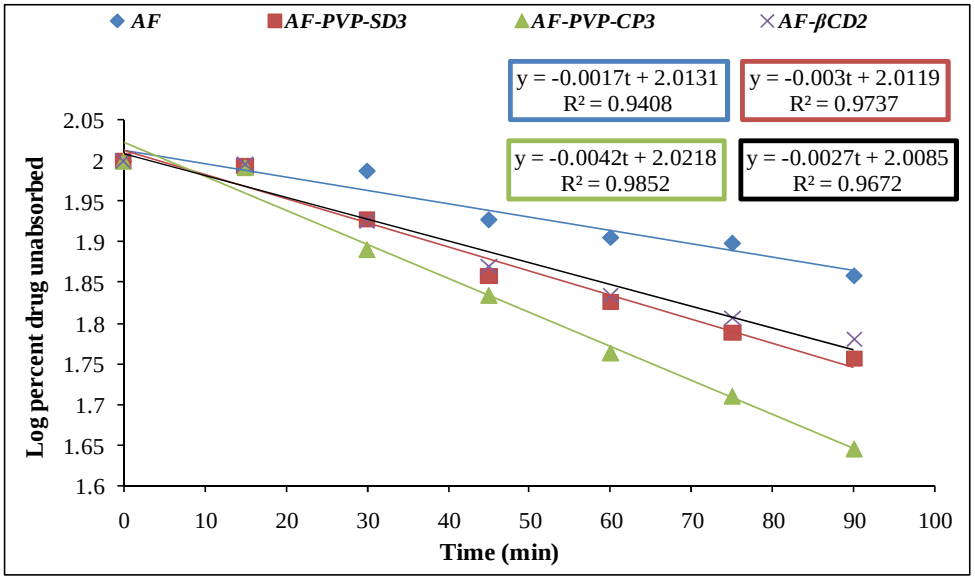

Figure 3. Kinetics plot of selected formulations of aceclofenac

CONCLUSION

The maximum intestinal absorption of the aceclofenac using *in situ* rat gut technique was observed from the co-precipitates with polyvinyl pyrrolidone (PVP) as compared to solid dispersions and *β*-cyclodextrin (*β*-CD) complexes. The co-precipitates of aceclofenac with PVP lends an ample credence for better therapeutic efficacy.

REFERENCES

Anastasiadou C, Henry S, Legendre B, Souleau C, Duchene D. Solid dispersions: comparison of prepared melts and coprecipitates of diazepam and polyoxyethylene glycol 4000. *Drug Dev. Ind. Pharm.* 1983;9(1-2):103-15. [DOI: 10.3109/03639048309048548]

Moffat AC, Osselton MD, Widdop B. Clarke's Analysis of Drugs and Poisons, Pharmaceutical Press, 1999; 3: 17-9.

Damian F, Blaton N, Kinget R, Van den Mooter G. Physical stability of solid dispersions of the antiviral agent UC-781 with PEG 6000, Gelucire® 44/14 and PVP K30. *Int. J. Pharm.* 2002;244(1-2):87-98. [DOI: 10.1016/S0378-5173(02)00316-2]

Dua K, Pabreja K. Ramana MV. Preparation, characterization and *in vitro* evaluation of aceclofenac solid dispersions. *ARS Pharmaceutica* 2010;51(1):57-76.

Dua K, Ramana MV, Sara UVS, Himaja M, Agrawal A, Garg V, Pabreja K. Investigation of enhancement of solubility of norfloxacin *β*-cyclodextrin in presence of acidic solubilizing additives. *Curr. Drug Deliv.* 2007a;4(1):21-5.

Dua K, Sara UVS, Ramana MV. Studies on release of aceclofenac from cyclodextrin systems. *Int. J. Pharm. Excip.* 2007b;4:101-5.

Jun SW, Kim M-S, Kim J-S, Park HJ, Lee S, Woo J-S, Hwang S-J. Preparation and characterization of simvastatin/ hydroxypropyl-*β*-cyclodextrin inclusion using super-critical antisolvent (SAS) process. *Eur. J. Pharm. Biopharm.* 2007;66(3):413-21. [DOI: 10.1016/j.ejpb.2006.11.013]

Kumar N, Jain AK, Singh C, Kumar R. Development, characterization and solubility study of solid dispersion of terbinafine hydrochloride by solvent evaporation method. *Asian J. Pharm.* 2008;2(3):154-58. [DOI: 10.4103/0973-8398.4 3301]

Martin A, Doluisio JT. Industrial bioavailability and pharmacokinetics: Guidelines, regulations, and controls. College of Pharmacy, Drug Dynamics Institute, University of Texas, Austin, TX 78712. 1977;539. [DOI: 10.1002/jps.2600671058]

Martin-Algarra RV, Pascual-Costa RM, Merino M, Casabo VG. Effects of polysorbate 80 on amiodarone intestinal absorption in the rat. *Int. J. Pharm.* 1995;122(1-2):1-8. [DOI: 10.1016/0378-5173(95)00025-E].

Patel HM, Suhagia BN, Shah SA, Rathod IS, Parmar VK. Preparation and characterization of etoricoxib-*β*-cyclodextrin complexes prepared by the kneading method. *Acta Pharm.* 2007;57(3):351-9. [DOI: 10.2478/v10007-007-0028-2]

Patil MP, Gaikwad NJ. Preparation and characterization of gliclazide-polyethylene glycol 4000 solid dispersions. *Acta. Pharm* 2009;59(1):57-65. [DOI: 10.2478/v10007-009 -0001-3]

Rawat S, Jain SK. Enhancement of intestinal absorption of few cox-2 inhibitors through interaction with *β*-cyclodextrin. *Indian J. Pharm. Sci.* 2007;69(4):529-34. [DOI: 10.4103/0250-474X.36939]

Shah JC, Chen JR, Chow D. Metastable polymorph of etoposide with higher dissolution rate. *Drug Dev. Ind. Pharm.* 1999;25(1):63-7. [DOI: 10.1081/DDC-100102142]

Zakeri-Milani P, Valizadeh H, Tajerzadeh H, Azarmi Y, Islambolchilar Z, Barzegar S, Barzegar-Jalali M. Predicting human intestinal permeability using single-pass intestinal perfusion in rat. *J. Pharm. Pharm. Sci.* 2007;10(3):368-379.

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