*Int.J.Curr.Microbiol.App.Sci* (2017) *6*(10): 2147-2153

*International Journal of Current Microbiology and Applied Sciences*
*ISSN: 2319-7706* **Volume 6 Number 10 (2017) pp. 2147-2153**
Journal homepage: http://www.ijcmas.com

**Original Research Article** https://doi.org/10.20546/ijcmas.2017.610.254

# Effect of Andrographolide Co-Administration on Pharmacokinetics of Meloxicam in Rats

**V.L. Gondaliya Vaishali[1], J.H. Patel[1*], R.D. Varia[1], S.K. Bhavsar[1], P.D. Vihol Priti[2], F.D. Modi Falguni[1] and T.H. Solanki Tamanna[1]**

[1]Department of Pharmacology and Toxicology, [2]Department of Veterinary Pathology, Veterinary College, Navsari Agricultural University, Navsari, Gujarat, India
**Corresponding author*

**Keywords**

Andrographolide, Meloxicam, Pharmacokinetic, Interaction.

**Article Info**

***Accepted:***
21 September 2017
***Available Online:***
10 October 2017

**A B S T R A C T**

The pharmacokinetic of meloxicam (5 mg/kg) and effect of co-administration of andrographolide (50 mg/kg) on pharmacokinetics of meloxicam were studied following intramuscular administration in rats. Drug concentration in plasma was determined using High Performance Liquid Chromatography (HPLC). The mean peak plasma drug concentration of 30.48 ± 1.44 µg/mL was achieved at 0.25 h. The pharmacokinetic parameters like mean absorption half-life ($t½_{\alpha}$) (9.71 ± 0.3 h), elimination half-life ($t½_{\beta}$) (12.14 ± 0.006 h), apparent volume of distribution ($Vd_{area}$) (0.23 ± 0.006 L/kg), area under plasma drug concentration-time curve ($AUC_{0-\infty}$) (383.85 ± 8.68 µg.h/mL), area under first moment curve (AUMC) (8813.72 ± 145.23 µg.h$^2$/mL), total body clearance ($Cl_B$) (0.01 ± 0.001 L/h/kg) and mean residence time (MRT) (22.63 ± 0.36 h) were observed. Following co-administration of andrographolide with meloxicam after single intramuscular administration in rats, the mean peak plasma concentration of meloxicam was significantly decreased. The pharmacokinetic parameters like absorption half-life ($t½_{Ka}$) and $AUC_{0-\infty}$were significantly increased whereas elimination half-life, $C_{max}$ and Vd were significantly decreased. This pharmacokinetic interaction might be due to drug-drug interactions.

## Introduction

Meloxicam is a non-steroidal anti-inflammatory drug from the subgroup of enolic acid oxicams. Meloxicam is a novel NSAID that has effectively replaced the old pain killers particularly diclofenac sodium which has high toxicity. Meloxicam has a high intrinsic activity combined with a low ulcerogenic potential (i.e. a high therapeutic index) (Engelhardt *et al.,* 1996).Meloxicam is considered as standard non-steroidal anti-inflammatory drug which metabolized by the cytochrome P450 (CYP) subgroup of isoenzymes, possibly CYP2C9/2C8 and CYP3A4 (Schmid *et al.*, 1995; Chesné *et al.*, 1998). Andrographolide (diterpene lactone) is main chemical constituent of plant *Andrographis paniculata.* It is widely used as home remedies against various aliments like throat infection, pneumonia, tonsillitis, dysentery, gastroenteritis and pyelonephritis (Deng *et al.*, 1982). Moreover, it is reported that andrographolide modulate expression of several cytochromes P450 isoenzymes like CYP2C9, CYP3A4, CYP2C6/11, CYP1A1/2 and CYP3A1/2. (Chesné *et al.*, 1998; Schmid*et al.*, 1995; Pekthong *et al.*, 2008;

Pekthong*et al.,* 2009) and therefore herb preparations containing andrographolide may result to herb–drug interactions in combination therapy.

So effect of using common medicine in combination with modern pharmaceuticals like meloxicam remains to be verified (Ernst, 1998; Patel *et al.,* 2015; Ratndeep *et al.,* 2009). Looking to this background this study was planned to evaluate effect of andrographolide co-administration on pharmacokinetics of meloxicam in rats.

## Materials and Methods

### Experimental animals

The experiment was conducted on male *albino Wistar* rats weighing between 250 to 350 grams. Rats were kept under constant observation for two weeks before the commencement of the experiment and subjected to clinical examination to exclude possibility of any diseases.

The animals were divided into groups and kept in cages. Standard ration and water was provided *ad libitum.* The experimental protocol was approved by Institutional Animal Ethics Committee.

### Drug and chemicals

Pure Andrographolide (98%), meloxicam sodium salt hydrate (>98%) and lambda carrageenan was obtained from Sigma-Aldrich St. Louis, USA. Methanol, acetonitrile and ortho-phosphoric acid (HPLC grade) were purchased from Merck Specialties Private Limited, Mumbai.

### Pharmacokinetic study and data analysis

Animals (n=48) were divided into 12 groups. Each group comprise of four animals. Multiple numbers of rats were used for serial collection of blood at alternating time points. A single intramuscular injection of meloxicam (5 mg/kg) was administered in the left gluteal muscle and for interaction pharmacokinetic meloxicam (5 mg/kg) and andrographolide (50 mg/kg) were administered by intramuscular route left and right gluteal muscle respectively. Blood samples were collected in $K_3$EDTA vials, at different time interval i.e. 0.75, 1, 2, 4, 6, 8, 12, 24, 36, 48 and60 hours from retro orbital plexus. Blood samples were centrifuged at 3000 rpm for 10 minutes at 4°C and plasma was transferred to cryo-vials and, stored at –20°C. Samples were analyzed within 48 h to quantify meloxicam concentration using High Performance Liquid Chromatography.

Meloxicam concentration in plasma samples was determined by reverse-phase High Performance Liquid Chromatography (HPLC) after extraction, using reported assays (Eniko *et al.,* 2014; Gondaliya *et al.,* 2015; Patel *et al.,* 2016) with minor modifications. The high performance liquid chromatography apparatus of Shimadzu comprising binary gradient delivery pump (model LC - 20AP) and UV detector (model CBM-20A) was used for assay. Chromatographic separation was performed by using reverse phase C18 column (ODS; 250 × 4.6 mm ID) at room temperature. The HPLC data integration was performed using software Clarity. Plasma (100 µL) was deproteinized by addition of acetonitrile (100 µL) and vortexed for one minute. This was followed by centrifugation at 8000 rpm for 10 minutes. The clean supernatant was collected and an appropriate aliquot of 20 µl of supernatant was injected into the loop of HPLC system through auto injector. The mobile phase consisted of a mixture of methanol and water (80:20 v/v) adjusted to pH 3.5 with ortho-phosphoric acid. Mobile phase was filtered by 0.2 µ size filter (Axiva N66) and degassed by ultra-

sonication. It was pumped into column by isocratic flow rate of 1.0 mL/min at ambient temperature. The effluent was monitored at 355 nm wavelength. Various pharmacokinetic parameters were calculated from plasma concentration of meloxicam using software PK solution (Version 2.0).

## Standardization and partial validation of assay

Initial stock solution was prepared by dissolving 2 mg pure meloxicam drug in 2 mL free plasma. The assay was sensitive, reproducible and linearity was observed from 0.04 to 200 µg/mL. The mean correlation coefficient ($R^2$) was 0.99. The limits of detection and limits of quantification were determined to be 0.09 and 0.19 µg/mL, respectively. Precision and accuracy of the assay were assessed using samples at concentration 50, 25, 1.56, 0.39 and 0.09 µg/mL. At all concentration studied, the C.V. was less than 8.7 %.

## Statistical analysis

Meloxicam plasma concentration and pharmacokinetic parameters of different treatment groups were compared by using student t-test using Microsoft excel 2007.

## Results and Discussion

The plasma concentration of meloxicam at different time interval following administration of meloxicam alone and concurrent administration of andrographolide is presented as Semilogarithmic plot in Figure 1.

Following intramuscular administration of meloxicam alone, the mean peak plasma drug concentration of 30.48 ± 1.44 µg/mL was achieved at 0.25 h which declined to 14.97 ± 0.43 µg/mL at 4 h. Thereafter, drug concentration decline gradually and detected upto 48 h (2.62 ± 0.11 µg/mL) and beyond then the drug was not detected in plasma. Following co-administration of andrographolide, meloxicam peak plasma concentration was significantly decreased to 23.67 ± 0.48 µg/mL from30.48 ± 1.44 µg/mL. Comparison of pharmacokinetics parameters of meloxicam and co-administration of andrographolide with meloxicam following intramuscular administration are presented in Table 1.

Elimination half-life ($t_{½\beta}$:12.14 ± 0.006 h) of meloxicam (5 mg/kg) following intramuscular administration in rats was in agreement to elimination half-life ($t_{½\beta}$: 13.4 h) reported following intravenous administration in albino rats (Busch *et al.,* (1998). However, shorter elimination half-life were reported in mice following intravenous (6.41h) and oral (4.76 h) administration (Busch *et al.,* 1998), in rabbits following oral administration (8.16 ± 2.19 h and 8.39 ± 1.17 h) of meloxicam (0.3 and 1.5 mg/kg) respectively (Turner *et al.,* 2006) and in flamingo (1.93 ± 0.32 h and 6.05 ± 3.53 h) following intramuscular and oral administration (0.5 mg/kg), respectively (Zordan *et al.,* 2016). Mean apparent volume of distribution ($Vd_{area}$:0.23 ± 0.006 L/kg) of meloxicam following intramuscular administration in rats was found lower in comparison to reported in rabbits (1.46 ± 0.48 L/kg and 4.14 ± 1.03 L/kg) following single oral administration (0.3 and 1.5 mg/kg), respectively (Turner *et al.,* 2006).This finding was supported by higher value of AUC(383.85 ± 8.68 µg.h/mL) of meloxicam in rats,in comparison to findings reported following intravenous administration in male (60.7 µg.h/mL); oral administration in female and male mice (89.5 µg.h/mL and 60.7 µg.h/mL) respectively; intravenous and oral administration in albino rats (70.9 and 83.3 µg.h/mL) (Busch *et al.,* 1998); oral administration in rabbits (5.20 ± 1.29 µg.h/mL) (Turner *et al.,* 2006); intramuscular

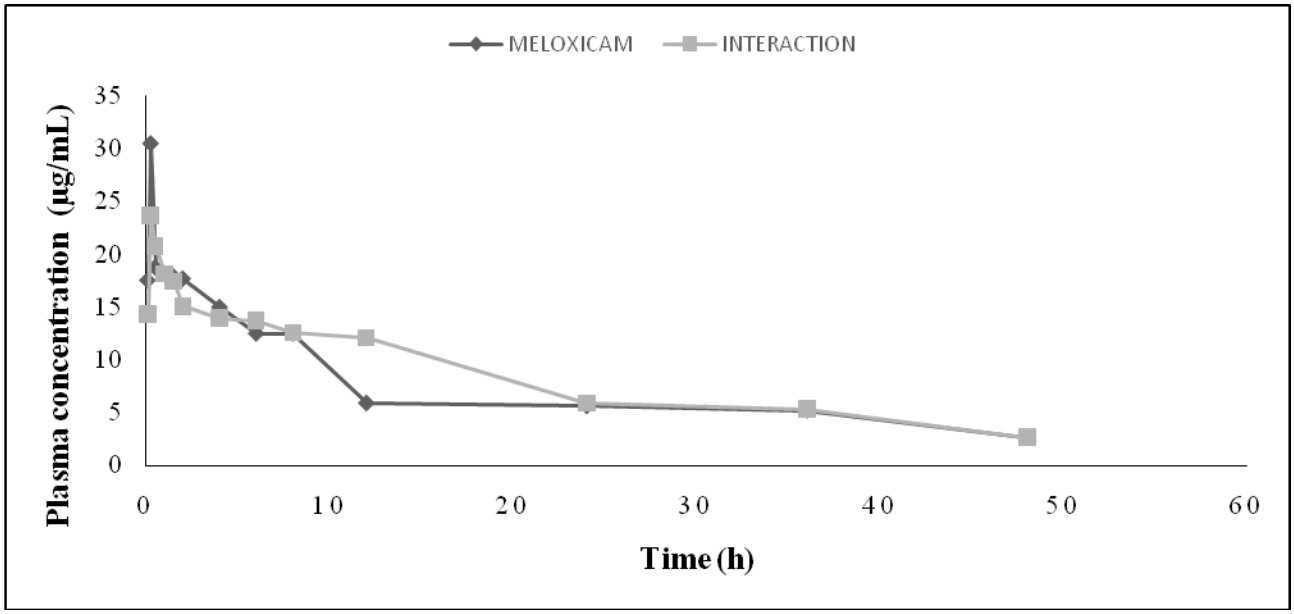
and oral administration in flamingo (17.78 ± 2.79 µg.h/mL and 22.16 ± 7.17 µg.h/mL) respectively (Zordan *et al.*, 2016) and oral, intravenous and subcutaneous administration in dogs (22.9, 21.5 and 24.1 µg.h/mL) respectively (Busch *et al.*, 1998).

**Table.1** Comparison of pharmacokinetics parameters of meloxicam (5 mg/kg) alone and co-administration of andrographolide (50 mg/kg) with meloxicam (5 mg/kg) following intramuscular administration in rats.

| Pharmacokinetic parameters | Unit | Meloxicam | Co-administration of Andrographolide and Meloxicam |
|---|---|---|---|
| $K_a$ | $h^{-1}$ | 0.07± 0.002 | 0.06 ± 0.001* |
| β | $h^{-1}$ | 0.057 ± 0 | 0.058 ± 0.00* |
| $t½_{Ka}$ | H | 9.71 ± 0.3 | 11.8 ± 0.12* |
| $t½_{β}$ | H | 12.14 ± 0.006 | 11.8 ± 0.38* |
| $C_{max}$ | µg/mL | 30.62 ± 1.45 | 23.7 ± 0.48** |
| $T_{max}$ | H | 0.25 ± 0 | 0.25 ± 0.00 |
| $AUC_{(0-\infty)}$ | µg·h/mL | 383.85 ± 8.68 | 435 ± 6.58** |
| AUMC | µg·$h^2$/mL | 8813.72 ± 145.23 | 9451.8 ± 176.19* |
| $Vd_{(area)}$ | L/kg | 0.23 ± 0.006 | 0.19 ± 0.003** |
| $Cl_{(B)}$ | L/h/kg | 0.01 ± 0.001 | 0.01 ± 0.00 |
| MRT | h | 22.63 ± 0.36 | 21.7 ± 0.20 |

* Significant at p<0.05, **Highly significant at p<0.01

**Fig.1** Semilogarithmic plot of comparison of meloxicam (5 mg/kg) and co-administration of andrographolide (50 mg/kg) with meloxicam (5 mg/kg) following intramuscular administration in rats. Each points represents mean ± S.E



In the present study the total body clearance (0.013 ± 0.001L/h/kg) of meloxicam following intramuscular administration (50 mg/kg) in rats was found in accordance with values of total body clearance reported following intravenous administration in male albino rats (0.015 L/h/kg); oral administration male and female albino rats (0.023 L/h/kg and 0.010 L/h/kg) respectively, intravenous administration to dogs (0.010 L/h/kg) (Busch *et al.*, 1998). However, higher value of total body clearance was reported in rabbits (0.12 ± 0.01 L/h/kg and 0.33 ± 0.06 L/h/kg) following single oral administration (0.3 and 1.5 mg/kg), respectively (Turner *et al.*, 2006). Mean residence time (MRT : 22.63 ± 0.36 h) was found in accordance with values reported following oral administration in male (31.8 h) and female (53.4 h) albino rats; intravenous (18 h) administration in male albino rats and oral (40 h), intravenous (34.8 h) and subcutaneous (35 h) administration in dogs (Busch *et al.*, 1998). However, lower MRT values following intravenous (3.02 h) and oral (3.89 h) administration in mice (Busch *et al.*, 1998) were also reported. Low volume of distribution ($Vd_{area}$) of meloxicam following intramuscular administration in present study may be due to strong binding of drug to serum albumin (>99%) in rats (Busch *et al.*, 1998). It is evident that meloxicam gets eliminated slowly following intramuscular administration in rats as elimination half-life and mean residence time values indicates that meloxicam remain longer period of time compare to the intravenous and oral administration in rats.

Following co-administration of andrographolide with meloxicam after single intramuscular administration in rats, absorption half-life ($t½_{Ka}$) and $AUC_{0-\infty}$were significantly increased from 9.71 ± 0.3 h to 11.8 ± 0.12 h and 383.85 ± 8.68 to 435 ± 6.58 µg·h/ml, respectively and elimination half-life, $C_{max}$ and $Vd_{area}$ were significantly decreased from 12.14 ± 0.006 to 11.8 ± 0.38 h, 30.62 ± 1.45 to 23.7 ± 0.48 µg/ml and 0.23 ± 0.006 to 0.19 ± 0.003 L/kg, respectively. In accordance to present study, alteration in pharmacokinetic parameter was observed like clearance of theophylline was significantly increased in *Andrographis paniculata* extract (1g/kg) treated group rats, elimination half-life of theophylline was significantly decreased in andrographolide (154 mg/kg) treated group rats and $AUC_{0-\infty}$of theophylline were significantly decreased in *Andrographis paniculata* extract (2 g/kg) and andrographolide (154 g/kg) treated group rats. While elimination half-life and mean residence time of theophylline were shortened about 14 and 17 % in the andrographolide (77 mg/kg) pretreated group rats (Chao *et al.*, 2010). Similarly *Andrographis paniculata* extract and andrographolide reduced the $AUC_{0–12h}$ of tolbutamide by 37% and 18%, respectively (Chen *et al.*, 2013). However, no significant effect of andrographolide on pharmacokinetics of warfarin (Hovhannisyan *et al.*, 2006) and midazolam (Wongnawa *et al.*, 2012) was observed. Alteration in pharmacokinetic of meloxicam may be due to metabolism of meloxicam caused by major extent through CYP2C9 and, to a much lesser extent, CYP3A4 (Schmid *et al.*, 1995; Chesné *et al.*, 1998) and andrographolide was also found to modulate expression of CYP2C9 and CYP3A4 microsomal enzyme (Pekthong *et al.*, 2008; Pekthong *et al.*, 2009).

## Acknowledgement

The facility and infrastructure provided by Dean, College of Veterinary science and A. H., Navsari to conduct this study is duly acknowledged.

## References

Busch, U., Schmid, J., Heinzel, G., Schmaus, H., Baierl, J., Huber, C. and Roth, W.

1998. Pharmacokinetics of meloxicam in animals and the relevance to humans. *Drug metabolism and disposition*, 26(6): 576-584.

Chao, W. W., Kuo, Y. H. and Lin, B. F. 2010. Anti-inflammatory activity of new compounds from *Andrographis paniculata* by NF-κB transactivation inhibition. *Journal of agricultural and food chemistry*, 58(4):2505-2512.

Chen, H. W., Huang, C. S., Liu, P. F., Li, C. C., Chen, C. T., Liu, C. T. and Lii, C. K. 2013. *Andrographis paniculata* extract and andrographolide modulate the hepatic drug metabolism system and plasma tolbutamide concentrations in rats. *Evidence-Based Complementary and Alternative Medicine*, 2013.

Chesné, C., Guyomard C, Guillouzo A, Schmid J, Ludwig E, Sauter T. 1998. Metabolism of Meloxicam in human liver involves cytochromes P4502C9 and 3A4. *Xenobiotica*.28 (1):1-13.

Deng, W. L., Nie, R. J. and Liu, J. Y. 1982. Comparison of pharmacological effect of four andrographolides. *Yaoxue Tongbao*, 17: 195-198.

Engelhardt, G., Bögel, R., Schnitzler, C. H. R. and Utzmann, R. 1996b. Meloxican: Influence on arachidonic acid metabolism: Part II. *In vivo* findings. *Biochemical pharmacology*, 51(1): 29-38.

Eniko, C., Ibolya, F. and Daniela-Lucia, M. 2014. Development of a HPLC-UV method for determination of meloxicam in human plasma and pharmaceutical dosage forms. *Acta Medica Marisiensis*, 60(4): 142-145.

Ernst, E., 1998. Harmless herbs? A review of the recent literature. *The American journal of medicine*, 104(2):170-178.

Gondaliya, S. R., Bhavsar S. K., Singh R. D., Patel J. H. and Thaker A. M. 2015. Pharmacokinetics and intramuscular bioavailability of Ketoprofen in patanwadi sheep. *Journal of Veterinary Pharmacology and Toxicology*, 14(2): 53-55.

Hovhannisyan, A. S., Abrahamyan, H., Gabrielyan, E. S. and Panossian, A. G. 2006. The effect of Kan Jang extract on the pharmacokinetics and pharmacodynamics of warfarin in rats. *Phytomedicine*, 13(5): 318-323.

Patel, J. H., Vihol Priti D., Patel U. D.,Varia R. D., Bhavsar S. K. and Thaker A. M. 2015. Effect of ketoprofen co-administration and febrile state on pharmacokinetic of Levofloxacin in goats. *Journal of Veterinary Pharmacology and Toxicology*. 14(1): 22-25

Patel, R.L., Bhavsar S.K., Tiwari Shireen, Patel J.H., Varia R.D. and Modi Falguni 2016. Effect of meloxicam co-administration on pharmacokinetic of cefquinome in cow calves. *Journal of Veterinary Pharmacology & Toxicology*. 15(2): 84-86.

Pekthong, D., Blanchard N, Abadie C, Bonet A, Heyd B, Mantion G, Berthelot A, Richert L, Martin H. 2009. Effects of *Andrographis paniculata* extract and Andrographolide on hepatic cytochrome P450 mRNA expression and monooxygenase activities after in vivo administration to rats and in vitro in rat and human hepatocyte cultures. *Chemico-Biological Interactions.* 179(2-3):247-55.

Pekthong, D., Martin, H., Abadie, C., Bonet, A., Heyd, B., Mantion, G. and Richert, L. 2008. Differential inhibition of rat and human hepatic cytochrome P450 by *Andrographis paniculata* extract and andrographolide. *Journal of ethnopharmacology*, 115(3): 432-440.

Ratndeep Singh, Sarita Devi, Patel J. H., Patel U. D., Bhavsar S. K. and Thaker A.M. 2009. Indian herbal bioenhancer: a review. *Pharmacognosy review*.

3(1):80-82.

Schmid, J., Busch, U., Heinzel, G., Bozler, G., Kaschke, S. and Kummer, M. 1995. Pharmacokinetics and metabolic pattern after intravenous infusion and oral administration to healthy subjects. *Drug metabolism and Disposition,* 23(11), 1206-1213.

Turner, P. V., Chen, C. H. and Taylor, M. W. 2006. Pharmacokinetics of meloxicam in rabbits after single and repeat oral dosing. *Comparative medicine,* 56(1): 63-67.

Wongnawa, M., Soontaro, P., Riditid, W., Wongpoowarak, P. and Ruengkittisakul, S. 2012. The effects of *Andrographis paniculata* (Burm. f.) Nees on the pharmacokinetics and pharmacodynamics of midazolam in healthy volunteers. *Songklanakarin Journal of Science and Technology,* 34(5): 533-9.

Zordan, M. A., Papich, M. G., Pich, A. A., Unger, K. M. and Sánchez, C. R. 2016. Population pharmacokinetics of a single dose of meloxicam after oral and intramuscular administration to captive lesser flamingos (*Phoeniconaias minor*). *American Journal of Veterinary Research,* 77(12): 1311-1317.

**How to cite this article:**

Gondaliya Vaishali, V.L., J.H. Patel, R.D. Varia, S.K. Bhavsar, P.D. Vihol Priti, F.D. Modi Falguni and Solanki Tamanna, T.H. 2017. Effect of Andrographolide Co-Administration on Pharmacokinetics of Meloxicam in Rats. *Int.J.Curr.Microbiol.App.Sci.* 6(10): 2147-2153.
doi: https://doi.org/10.20546/ijcmas.2017.610.254