Comparative anti-inflammatory effects of azithromycin, clarithromycin and roxithromycin using animal models of inflammation in Wistar rats

Mahesh M.Bharambe[1], Vandana M. Thorat[1*], Somnath M.Matule[2]
[1]Department of Pharmacology, Krishna Institute of Medical Sciences, Karad, India, 415539,
[1*]Departmentof Pharmacology,Krishna Institute of Medical Sciences, Karad, India, 415539,
[2]Department of Pharmacology, Ashwini Rural Medical College& Research Centre,Solapur,India, 413006,

***Corresponding author name: Dr.Vandana M. Thorat**

Abstract

Background: The anti-inflammatory effect of macrolideshas led the way to establish a platform for their exploratory use in asthma as well as other various inflammatory and infective airway conditions. This has been reported in few clinical, in vitro and animal studies.
Aims & Objective: To assess the anti-inflammatory activity of azithromycin, clarithromycin and roxithromycin in acute and sub-acute models of inflammation in experimental Wistar rats and to compare their activities with the control group and standard drug aspirin.
Materials and Methods: This study was implemented after the approval from animal ethics committee. Animals were divided into five groups each for acute and sub-acute model of inflammation. The control group were administered (4ml/kgsuspension) of 1% gum acacia and the test groups were administered azithromycin, clarithromycin, roxithromycin (20mg/kg each) and aspirin(200mg/kg). In acute model (Carrageenan induced rat paw edema), using a mercury plethysmograph,the rat paw volume was measured at regular time intervals and then we estimated the percentage inhibition of edema. In model of sub-acute inflammation (Granuloma induced by foreign body), we implanted two sterile cotton pellets of 10 mg each subcutaneously in bilateral axillae of rats. The treatment was initiated onday 1 of pellet implantation and it was repeated once daily till day 10. The rats were sacrificed on day 11 and then we calculated mean dry weight of cotton pellets covered with granuloma for various groups.
Results:
Acute model: This study clearly shows the anti-inflammatory potential of azithromycin, clarithromycin and roxithromycinwhen compared with the control group. However, the anti-inflammatory activity of azithromycin and aspirin was comparable to each other at 5-hour interval whereasthe anti-inflammatory activity of clarithromycin and aspirin was comparable to each other at4and 5-hour interval. Roxithromycin activity appeared to be inferior when compared with aspirin.
Sub-acute model: Onlyazithromycin showed significant anti-inflammatory activitywhen compared with the control group and its activity was also comparable to aspirin.

*e*ISSN1303-5150

Conclusion: All three selected macrolides possess significant anti-inflammatory activity in comparison to control in acute model of inflammation and only Azithromycin has significant anti-inflammatory activity in sub-acute model of inflammation in comparison to control.

KEY WORDS: Macrolide, aspirin, anti-inflammatory, Carrageenan, foreign body granuloma
DOINumber:10.14704/nq.2022.20.8.NQ44759 **NeuroQuantology2022;20(8):7355-7364**

INTRODUCTION

Inflammation continues to be an area of keen interest in the field of research possibly due to inadequacy of a safer and more effective anti-inflammatory agent. Inflammation is defined as a complex and dynamic condition which involves molecular and cellular responses that are designed to combat the invading pathogens and getting rid of damaged or necrotic tissues[1]. The body's cardinal defenders against foreign intruders are the plasma proteins, leukocytes and tissue phagocytes that are derived from the circulating cells

Acute inflammation is followed by chronic inflammation which is long lastinginvolving prolonged inflammatory response that involves the proliferation of blood vessels as well as the presence of macrophagesand lymphocytes,fibrosis, and tissue destruction.The inflammatory response acts by commencing thehealing process and abolishing deleterious stimuli[2].

Therapy of inflammation is a matter of debate and is also insufficient since long. Multipletreatment modalities are available for the treatment of different inflammatory conditions. The currently used threemajor groups of anti-inflammatory medications involves corticosteroids, NSAIDs & DMARDs. These drugs yielded good results till now but they have got potential toproduce serious adverse effects, sometimes even life-threatening events have resulted in widespread limitation of their use[3].Some dipeptidyl peptidase 4 inhibitors[4], Angiotensin receptor blocker telmisartan[5,6],some adrenergic agonists[7],calciumchannel blockers[8] and sulfonamides[9] have been documented to have anti-inflammatory potential in experimental studies. As these drugs are not completely devoid of adverse effects[3] there is a need to investigate for better and safer anti-inflammatory agents.

Macrolides had long been used in therapeutics in the treatment of infections caused by several Gram-positive and Gram-negativebacteria namely Haemophilus influenzae, Legionella pneumophilia, Chlamydia and Mycobacteria.Macrolide antibiotics have better pharmacokinetic profile such as once daily administration,good tolerability and greater bioavailability[10].Macrolides not only possess antimicrobial activity but also modify many components of the immune response[11].Some studies proposed that several macrolides possess antioxidant property that is responsible for the anti-inflammatory activity of these agents[12].

The anti-inflammatory effect of macrolides has led the way to establish a platform for their experimental use in bronchial asthma as well as other various inflammatory and infective airway conditions[13]. Several studies have been carried out to investigate the anti-inflammatory effect of macrolides in animal models but there has been widespread speculation about their anti-inflammatory effects. However, the mechanisms responsible for these actions are still unclear. There is a paucity of published literature regarding anti-inflammatory activity of macrolides in animal studies hence, in the present study we planned to assess the anti-inflammatory potential of macrolides in models of inflammation in Wistar rats.

MATERIALS& METHODOLOGY

Animals used in experiment

Wistar rats including both the sexes weighing 200-300 grams were taken from our central animal house, Krishna Institute of Medical Sciences, Karad. The animals were accommodated to light - dark cycle for 10 days before experimentation. They had free access to food and water ad libitum under strict hygienic conditions. Institutional Animal

Ethics Committee approval was taken and CPCSEA guidelines were strictly followed throughout the study.

Drugs

Azithromycin,Clarithromycin and Roxithromycin were obtained in pure powder form from Century Pharmaceuticals Ltd, Vadodara. Injection Ketamine (10ml vial of 50mg/ml) (Troikaa Pharmaceuticals Ltd), Aspirin (Reckitt Benckiser India Ltd), carrageenan and gum acacia powder were obtained from Central Pharmacy of KIMS, Karad

a. Azithromycin, Clarithromycin and Roxithromycin: 100 mg pure powder form of each test drug was added in 10 ml of 1% gum acacia. Gum acacia was used as a suspending agent as all the drugs were poorly soluble in sterile water

b. Aspirin: 325 mg of the dispersible tablet of aspirin was dissolved in 5 ml of sterile water.

c. Carrageenan was prepared as suspension (1% in 0.9% normal saline)[14].

Groups

The rats were divided into 5 groups. Each group consisted of 6 rats for acute and sub-acute model of inflammation. Total Wistar rats utilised for the study = 60.

Group	Treatment	Dose	Route
I	Gum Acacia	4ml/kg	Oral
II	Aspirin	200mg/kg[15]	Oral
III	Azithromycin	20mg/kg[16]	Oral
IV	Clarithromycin	20mg/kg[16]	Oral
V	Roxithromycin	20mg/kg[16]	Oral

METHODS:

1. <u>Model of acute inflammation</u>

It was carried out by inducing rat paw edema by carrageenan injection[17]:

Mercuryplethysmograph was used to assess anti-inflammatoryactivity byinducing rat paw edema after carrageenan injection A red ink mark was put on one paw of each animal at the level of lateral malleolus to enable uniform dipping at subsequent readings. One hour before the edema induction, all the drugs were administered by oral gavage feeding needles to the respective groups. Induction of paw edema was done by injecting0.05 ml of 1% suspension of carrageenan intradermally intoone of the hind paws of the rats. By mercury plethysmograph, the volume of pawedema was measured in ml as the displacement of mercury at 0 hour i.e., immediately after the carrageenan injection and the same technique was carried out at 1, 2, 3, 4 and 5 hours. The actual edema volume was assessed by noting the difference between 0 hour and subsequent reading.

The percentage inhibition of edema for control and test groups was calculated by,

$$\frac{EC - ET}{EC} \times 100.$$

(EC:Mean edema in control; ET: mean edema in drug treated group)

2. <u>Sub-acute model of inflammation</u>

It was carried out by foreign Body Induced Granuloma Method[18]:

On Day 1 of experiment, all drugs were given to the respective groups by oral gavage feeding needles and then the rats were anesthetized by ketamine (50mg/kg i.p)[19].Under all aseptic precautions after clipping the hair over axillae, a small incision was given in bilateral axillae of the rats and after that two cotton pellets (sterilized by autoclaving at 120°C) weighing 10 mg eachwere implanted subcutaneously. The wounds weresutured and the rats were then

*e*ISSN1303-5150 www.neuroquantology.com

kept in clean cages individually after recovery from anaesthesia. The treatment was initiated on Day 1 of pellet implantationand was repeated once a day for the next ten days. The rats were sacrificed on eleventh day withketamine anaesthesia overdose and the cotton pellets were removed. After removing the extraneous tissue from the cotton pellets, thepelletswere dried overnight at 60°C in hot air oven and their dry weight was noted. We calculated the net granuloma weight by noting the difference in the weight of cotton pellets before and after the implantation. The mean dry weight of granuloma for study groups was recorded and expressed as mg/100 g of body weight of rat. The percentage inhibition of mean granuloma dry weightfor control and test groups was calculated by,

$$\frac{CW - TW}{CW} \times 100$$

CW: Mean dry weight of granuloma among control group
TW: Mean dry weight of granuloma among drug treated

STATISTICAL ANALYSIS:

The data is exhibited as mean ± SD and the analysis was performed by one way ANOVA which was followed by post hoc Dunnett'stest. Dunnett's multiple comparison test was used to compare aspirin and macrolides. All the statistical methods were carried out through the software Graph pad Instat 3.06 version and $p < 0.05$ was considered as statistically significant.

RESULTS

Acute model of inflammation (Paw edema induced by carrageenan injection):

Mean of the paw edema volumes in 'ml' measured using plethysmographfor control, aspirin, azithromycin, clarithromycin and roxithromycingroups are shown in (Table.1,Graph.1).

Statistically significantinhibition of paw edema volumewas seen with aspirin as well as all the three macrolides in comparison tocontrol group. Further anti-inflammatory activity of azithromycin, clarithromycin and roxithromycin was compared with the anti-inflammatory activity of aspirin. The anti-inflammatory effect of azithromycin and aspirin was comparable to each other ($p > 0.05$) at 5hr whereas the effect of clarithromycin and aspirin was comparable to each other($p > 0.05$) at 4hr and 5hr. Anti-inflammatory effect of roxithromycin appeared to be inferior to aspirin at all time intervals.(Table.1).Percentage inhibition of paw edema of all the test drugs are mentioned in (Table.2,Graph.2)

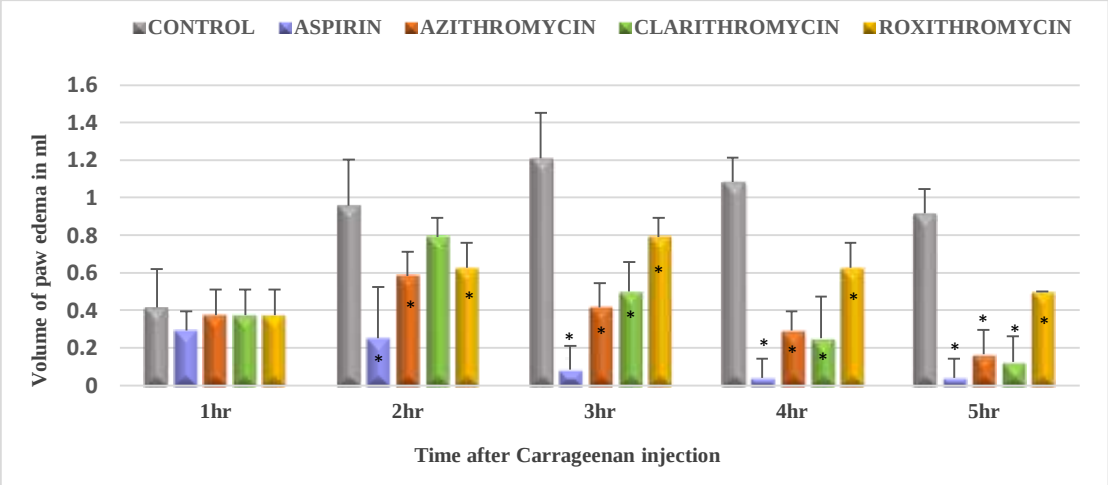
TABLE 1: Effect of 200mg/kg aspirin and 20mg/kg azithromycin, clarithromycin,roxithromycin treatments on paw edema induced by carrageenan

Time after carrageenan injection	(Mean ± SD) Volume of paw edemain ml					ANOVA	
	Control (4ml/kg of 1% gum acacia)	Aspirin (200mg/kg)	Azithromycin (20mg/kg)	Clarithromycin (20mg/kg)	Roxithromycin (20mg/kg)	F value	p value
1 hr	0.416±.204	0.291±0.102	0.375±0.136	0.375±0.136	0.375±0.136	0.5769	0.682
2 hr	0.958±0.245	0.250± 0.273 *	0.583±0.129 *, #	0.791±0.102 #	0.625±0.136*	11.580	< 0.0001
3 hr	1.208±0.245	0.083± 0.129	0.416±0.129	0.5±0.158*,	0.791±0.10	41.653	<0.0001

		*	*, #	#	2*		
4 hr	1.083±0.129	0.041±0.102 *	0.291±0.102 *, #	0.25±0.223*	0.625±0.136*	46.814	<0.0001
5 hr	0.916±0.129	0.041±0.102*	0.166±0.129 *	0.125±0.136 *	0.50±0.00*	62.750	<0.0001

Post hoc analysis: - by Dunnett's Test: *p < 0.05; by Dunnett's multiple comparison test: # p < 0.05

GRAPH 1:Effect of different treatments onpaw edema volume after carrageenan injection

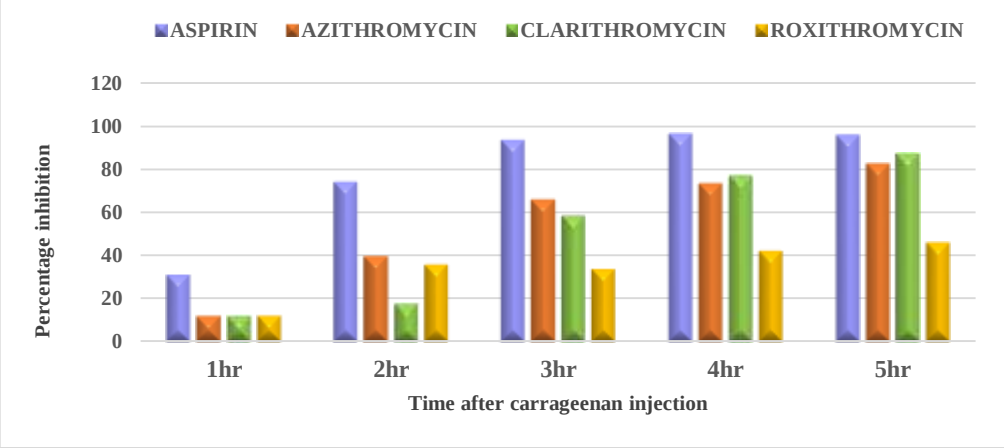


Post hoc analysis by Dunnett's Test: * *p*< 0.05

Groups	1 hour	2 hours	3 hours	4 hours	5 hours
ASPIRIN (200mg/kg)	30.95 %	73.95 %	93.33 %	96.29 %	95.65 %
AZITHROMYCIN (20mg/kg)	11.90 %	39.58 %	65.83 %	73.14 %	82.60 %
CLARITHROMYCIN (20mg/kg)	11.90 %	17.70 %	58.33 %	76.85 %	86.95 %
ROXITHROMYCIN (20mg/kg)	11.90 %	35.41 %	33.33 %	41.66 %	45.65 %

TABLE 2: Percentage inhibition of paw edema in Azithromycin, Clarithromycin and Roxithromycin treated group compared with Aspirin group at different time intervals.

GRAPH 2: Percentage inhibition of paw edema induced by carrageenan at different time interval



Sub-acute inflammation (Granuloma induced by cotton pellet implantation):

The mean of dry weight of ten-day old granuloma which is expressed as (mg/100 g) body weight of rat as well as its percentage inhibitionin control, aspirin, azithromycin, clarithromycin and roxithromycin groups are shown in (Table 3, Graph 3 and Graph 4).

Azithromycin treated group showed statistically significant decrease indry weight of granuloma ($p < 0.05$) in comparison to control. Clarithromycin and roxithromycin treated group did not show any statistically significant difference in meandry weight of granuloma ($p > 0.05$) in comparison to control. Hence both of them further were not compared with aspirin group.

When the mean dry weight of granuloma of azithromycin group was compared with that of aspirin group, no statistically significant difference was noted in the mean granuloma dry weight of azithromycin in comparison toaspirin ($p > 0.05$) group. It indicates that in sub-acute model of inflammation, the anti-inflammatory activity of azithromycin was comparable to aspirin (Table 3).

Table 3: Effect of 200mg/kg aspirin and 20mg/kg azithromycin, clarithromycin,roxithromycin treatments on dry weightof granuloma

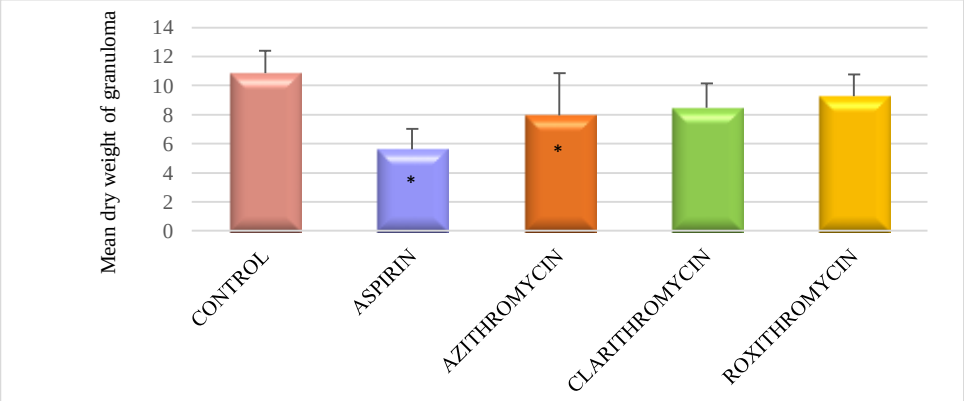
S. No	Treatment	Meandry weight of granuloma (Mean ± SD)	Percentage inhibition
1.	CONTROL (4ml/kg)	10.86 ± 1.555	____
2.	ASPIRIN (200mg/kg)	5.60 ± 1.436 *	48.43%
3.	AZITHROMYCIN (20mg/kg)	7.96 ± 2.884 *	26.70%
4.	CLARITHROMYCIN (20mg/kg)	8.50 ± 1.638	21.73%
5.	ROXITHROMYCIN (20mg/kg)	9.25 ± 1.528	14.8%

ANOVA: F = 6.227, p =0.0013

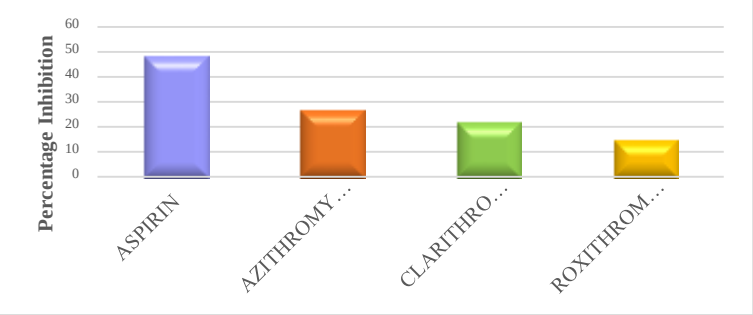
Post hoc analysis by Dunnett's Test: *p< 0.05

Dunnett's multiple comparison Test: - Aspirin Vs Azithromycin p> 0.05

GRAPH 3: Effect of different treatments on mean dry weight of granuloma



GRAPH 4:The percentage inhibition of dry weight of granuloma



DISCUSSION

Inflammation plays a vital role in body's defense mechanism. Amongst the modern perspectives to treat inflammation, macrolidesnot only decreasethe infection but also appears to be advantageous in reducing the inflammation. Evidences specify that macrolides affect the generation of many pro-inflammatory cytokines like IL1, IL 2, IL 6, leukotrienes, TNF-α, prostaglandins, and nitric oxide[20].

ACUTE INFLAMMATION (PAW EDEMA INDUCED BY CARRAGEENAN INJECTION):

Paw edema induced by carrageenan comprises of biphasic response in which the response of first phase is associated with the release of serotonin, histamine and kinins, whereas the second phase response is associated with prostaglandins and the release of slow reacting substances[21].We estimated the mean paw volume in acute inflammatory model. The reduction in mean paw volume after inflammation induction was considered as anti-inflammatory activity. The results of our study indicates that azithromycin, clarithromycin and roxithromycin at dose of 20mg/kg each, have shown significant anti-inflammatory activity in comparison to control group in acute model. The anti-inflammatory effect of azithromycin and aspirin was comparable to each other at 5hr interval whereas the effect was inferior to aspirin at 2hr, 3hr and 4hr intervals. This indicates that azithromycin might have delayed anti-inflammatory effect beyond 5 hours. The anti-inflammatory effect of Clarithromycin and aspirin was comparable to each other at 4hr and 5hr intervals whereas the effect was inferior to aspirin at 2hr and 3hr intervals. This indicates that clarithromycin might have delayed anti-inflammatory effect beyond 4 and 5 hours. However, anti-inflammatory effect of Roxithromycin appearedto be inferior to aspirin at 2hr, 3hr, 4hr and 5hr intervals when compared to aspirin. The inhibition of synthesis of histamine, serotonin prostaglandins, and various inflammatory mediators might be the most plausible

mechanisms of macrolide antibiotics in acute model of inflammation[12,14,22].

There is no significant inhibition of paw edema with any of the study drugs at 1hr interval. This might be either due to inadequate progression of inflammation at 1 hour interval or due to delayed onset of action of study drugs or due to both the mechanisms. According to Winter et al.[17], after injecting the phlogistic agent into the hind paw of rat, paw edema reaches a peak in 3-5hrs and then the same degree of edema is retained for few hours. This might explain that sufficient inflammation was not produced at 1hr after injecting carrageenan and hence the activity of all the three macrolides was comparable to aspirin at 1hr. As the inflammation progressed from 2nd hour, the individual drugs effects were also seen in the form of suppression of the edema till the 5th hour.

SUB-ACUTE INFLAMMATION(GRANULOMA INDUCED BY COTTON PELLET IMPLANTS)

This model of granuloma induced by foreign body implantation comprises ofthree response phases which includes transudative phase, exudative phase and proliferative phase.The proliferative phase response is measured as the increase in granuloma dry weight[23].We evaluated the mean dry weight of granuloma and the reduction of this weight was considered as anti-inflammatory action. In this model, it was found that only azithromycin resulted in significant decrease in dry weight of granuloma in comparison to control and also its anti-inflammatory effect was comparable with aspirin.This significant anti-inflammatory activity of azithromycin might be due to promotion of apoptosis of neutrophils or inhibition of fibroblast proliferationin sub-acute model of inflammation [20,24].

Thus, all the three macrolides - azithromycin, clarithromycin and roxithromycin has shown significant anti-inflammatory activity in comparison to control treated groupin acute model of inflammationand only azithromycin has shown significant anti-inflammatory activity in comparison to control groupin sub-acute model of inflammation.

The observations of our study are in agreement with Gosavi et al.[20], Scaglione et al.[22] and Ianaro et al.[25] propounding that macrolidehave anti-inflammatory activity in different animal models of inflammation. The results of our acute model of inflammation correlates with the findings of the earlier studies however according to them roxithromycin has better anti-inflammatory potential as compared to azithromycin and clarithromycin whereas we found roxithromycin to be inferior in our study. According to Gosavi et al.[20], no significant anti-inflammatory activity of macrolideswas seen in cotton pellet induced granuloma model whereas we found azithromycin to be effective in decreasing the mean granuloma dry weight.In view of results obtained from the present study, we suggest that macrolides have anti-inflammatory activity. Moreover, these drugs have better pharmacokinetic profile including once daily administration,good tolerability and greater bioavailabilitywhich make them a potential new anti-inflammatory agent for inflammatory conditions associated with acute infections. As this is an animal study and animal data cannot be directly hypothesized on humans, well planned human studies are required further to support these findings in order to reveal the impact of macrolides on inflammation as well as in the treatment of COPD, asthma, bronchiectasis, bronchiolitisand osteomyelitis.

CONCLUSION

Based on the results obtained from our study, we conclude that: -

- Azithromycin, Clarithromycin and Roxithromycin in their therapeutic equivalent doses have significant anti-inflammatory activity in comparison to control and modest anti-inflammatory activity in comparison to aspirin in animal model of acute inflammation.
- Only Azithromycin has significant and modest anti-inflammatory activity incomparison to control and aspirin

respectively in sub-acute model of inflammation.

- Clarithromycin appears to be more effective in controlling acute inflammation whereas Azithromycin appears to be effective in both acute and sub-acute inflammation.
- The current study suggests that the use of macrolides may have an added benefit in various inflammatory disorders associated with acute infections as monotherapy as well as along with the conventional medications.
- However, we suggest that macrolides should not be given in chronic inflammatory conditions as long term use in chronic conditions will lead to development of resistance against macrolides but it will be beneficial to treat acute infections developed in chronic inflammatory conditions.
- Though the anti-inflammatory activity of macrolides is secondary to its anti-infective activity, more studies are needed to be done in other different models of acute as well as chronic inflammation. Along with these, pre-clinical evaluation, human studies are also required to be done, in order to strengthen the results and prove their efficacy in the long-term use of macrolides as potential anti-inflammatory agents in routine clinical practices.
- Hence, we suggest that macrolides along with its anti-infective, immunomodulatory and additional anti-inflammatory property might be beneficial for patients with infective conditionsassociated with inflammation like chronic sinusitis, COPD, asthma,bronchiolitis, bronchiectasis, cystic fibrosis and osteomyelitis

ACKNOWLEDGEMENT:
We would like to acknowledge Century Pharmaceuticals Ltd, Vadodara., Gujarat, for providing drugs in pure powder form.We acknowledge the support given by the Central animal house of the institution and the technical staff of Department of Pharmacology during study.

CONFLICT OF INTEREST
No conflict of interest

BIBLIOGRAPHY

1. Chensue SW, Ward PA.Inflammation. In: Damjanov I, Linder J, editors. Anderson's Pathology.10thEdition.St Louis, Missouri: Mosby Year Book; 1990: 387-413
2. Kumar V, Abbas AK, Fausto N, Aster JC. Robbins and Cotran pathologic basis of disease. 9th ed. Philadelphia: Saunders An imprint of Elsevier; 2015.
3. Brunton L, Chabner BA, Knollmann BC. Goodman and Gilman's the pharmacological basis of therapeutics. Twelfth.
4. Kagal UA, Angadi NB, Matule SM. Effect of dipeptidyl peptidase 4 inhibitors on acute and subacute models of inflammation in male Wistar rats: an experimental study. International Journal of Applied and Basic Medical Research. 2017 Jan;7(1):26.
5. Matule SM, Hogade AP, Kangle RP, Torgal SS, Kothari N. Effect of telmisartan on acute model of inflammation in male Wistar rats: an experimental study. Int J Res Med Sci. 2016;4:135e138.
6. Matule, S., Hogade, A., Kangle, R., B., N., &Kolla, R. (2016a). Effect of telmisartan on sub-acute model of inflammation in male Wistar rats - an experimental study. International Journal of Research in Medical Sciences, 1988–1992. https://doi.org/10.18
7. Green KL. The anti-inflammatory effect of catecholamines in peritoneal cavity and hind paw of mouse. Br J Pharmacol 1972;45(2):322-32.
8. Shrivastava VK, Saxena KK, Gupta B. Calcium channel blockers in acute inflammation. Indian J Exp Biol 1988;26(1):70-1.
9. Hiremath SV, Gouripur VV, Patil PA. The anti-inflammatory activity of some sulphonamides in albino rats. Ind J Med Res 1996;103:120-5.
10. Ungureanu V.: Macrolides, lincosamides, streptogramines (MLS):mechanisms of

action and resistance. BacteriolVirusolParazitolEpidemiol., 55, 131, 2010.
11. Altenburg J. et al.: Immunomodulatory effects of macrolide antibiotic-part 1: biological mechanisms. Respiration, 81, 67, 2011
12. Labro MT, El Benna J and Babin-Chevaye C (1989) Comparison of the in vitro effect of several macrolides on the oxidative burst of human neutrophils. J AntimicrobChemother 24:561–572.
13. Khobragade AA, Patel SB, Pophale RR, Vallish BN, Kosale SP. Analgesic and anti-inflammatory activity of roxithromycin and erythromycin, alone and in combination with ibuprofen: an animal study. IOSR J Pharmacy. 2012;1:15-21.
14. Morris CJ. Carrageenin induced paw edema in the rat and mouse. In: Winyard PG, Willoughby DA, editors. Inflammation Protocols. UK: Humana Press; 2003: 115-21.
15. Laurence DR, Bacharach AL. Evaluation of Drug Activities:Pharmacometrics Vol. 2. New York and London: Academic Press Inc; 1964.
16. Gajendra NJ, Madhavi E, Konda VG, Venkata RY, Rajeshwaramma G. Comparative study of anti-inflammatory activity of newer macrolides with etoricoxib. Journal of Evolution of Medical and Dental Sciences. 2014 Mar 10;3(10):2413-20.
17. Winter CA, Risley EA, Nuss GW. Carrageenan induced edema in hindpaw of the rat as an assay for anti-inflammatory drugs. Proc Soc exp Biol Med. 1962;111:544-7.
18. Meier R, Schuler W, Desaulles P. ZurFrage des mechanismus der hemmung desBindegewebswachstumsdurch cortisone. Experientia 1950;6:469-71.
19. Karwacki Z, Kowiański P, Moryś J. General anaesthesia in rats undergoing experiments on central nervous system. Folia morphologica. 2001;60(4):235-42.
20. Gosavi PA, Jaju JB, Ubale VM, Pawar GR, Dharmadhikari SC. Study of evaluation of anti-inflammatory activity of macrolide antibiotics in rats: an experimental study.
21. Brahmbhatt MR, Patel JM, Patel VB, SalujaAK.Analgesic and anti-inflammatory activity of leaves of riveahypocrateriformis. J Pharm Phytother. 2010;1(1):001-3.
22. Scaglione F, Rossoni G. Comparative anti-inflammatory effects of roxithromycin, azithromycin and clarithromycin.JAntimicrobChemother. 1998;41 Suppl B:47-50
23. Swingle KF, Shideman FE. Phases of the inflammatory response to subcutaneous implantation of a cotton pellet and their modification by certain anti-inflammatory agents.JPharmacol Exp Ther. 1972;183(1):226-34.
24. Kanoh S, Rubin BK. Mechanisms of action and clinical application of macrolides as immunomodulatory medications. Clinical microbiology reviews. 2010 Jul;23(3):590-615.
25. Ianaro A, Ialenti A, Maffia P, Sautebin L, Rombolà L, Carnuccio R, Iuvone T, D'Acquisto F, Di Rosa M. Anti-inflammatory activity of macrolide antibiotics. Journal of Pharmacology and Experimental Therapeutics. 2000 Jan 1;292(1):156-63.