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Research Article

ANALGESIC ACTIVITY OF TELMISARTAN AND ROSUVASTATIN IN VARIOUS ANIMAL MODELS

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ABSTRACT

Objective: To study the analgesic activity of telmisartan and rosuvastatin in various animal models

Materials and Methods: Analgesic activity of telmisartan and rosuvastatin alone and in combination is assessed in hot plate method in rats; acetic acid induced writhing in mice and tail clip method in rats, in all these tests the comparator is aspirin.

Results: In hot plate method rosuvastatin showed some analgesic activity, while telmisartan is almost nil. Effect of the combination group in causing analgesia is almost similar to that of aspirin producing more than 70% inhibition of writhing. In tail clip method to test the mechanical pain stimulus both aspirin and combination group had 50% inhibition of response to application of tail clip.

Conclusion:These results shows that rosuvastatin has some analgesic activity and it is stronger when combined with telmisartan, but this may not justify its use as standalone analgesics. But it can be a potential adjuvant for coexisting inflammatory disorders and dyslipidemia, hypertension.

Keywords: Group, telmisartan, aspirin, rosuvastatin.

INTRODUCTION

Many drugs like non-steroidal anti-inflammatory drugs and corticosteroids are being used in modern medical practice to suppress pain and inflammation. The drawback of these drugs is they provide symptomatic relief and long-term use of these drugs is associated with serious adverse effects. Hence, the search for a new, safe analgesic drug is on-going.

The elderly people almost have one or more other major disease conditions, and most of the disease is related with pain as a major symptom. Painful arthritis which is a chronic disorder must be managed with continuous analgesic treatment. If at all understanding the pathogenesis of the symptoms and signs to establish a diagnosis and prognosis is difficult, it becomes even more challenging for managing them with drugs with their geriatric changes like age related reduction in hepatic blood flow and hepatocyte mass and primary aging changes in hepatic sinusoidal endothelium. This has an effect on drug transfer and oxygen delivery and causes reduction of hepatic drug clearance. Age related changes in renal clearance is evident, although renal clearance reduction in older people is predominantly disease-related and is poorly estimated by standard methods. The geriatric dosing axiom, "start low and go slow" is based on pharmacokinetic considerations and concern for adverse drug reactions since there is lack of clinical trial data.

If a single drug can be given to a patient for more than one indication with such coexisting comorbid conditions, it can potentially reduce their drug load and minimize the drug toxicity. This may also prove to be a cost effective method of treatment.

Rosuvastatin a lipid lowering drug and Telmisartan an anti-hypertensive drug if found to be having analgesic property, then it would be a great boon to geriatric patients and others suffering with chronic patients who are suffering with number of comorbidities. Hence, this study was planned to evaluate and compare the pain reducing actions of rosuvastatin and telmisartan in different animal models of pain.

MATERIALS AND METHODS

Animals

Adult Wistar albino rats (*Rattusnorvegicus*) weighing between 200 to 240 gram and Swiss Mice (*Musmusculus*) of either sex weighing 25 – 30 gram were purchased from King Institute of Preventive Medicine, Guindy, Chennai and maintained in the Central Animal House, SreeBalaji Medical College and Hospital, Chennai, India for acclimatization. All experiment was performed with Institutional Animal Ethics Committee approval and under CPCSEA guidelines.

Experimental Drugs

Rosuvastatin (from Micro Labs Ltd), Telmisartan (from Micro Labs Ltd) and Aspirin (from ZydusCadila Healthcare Ltd) were obtained by pure powdered form and given in dosage of Rosuvastatin 5 mg/kg, Telmisartan 2 mg/kgand aspirin dose of 100 mg/kg orally by gavage feeding tube. For all the oral drugs carboxymethyl cellulose is used as a solvent and 1% Acetic acid via intra-peritoneal route is used as algesic agent.

In this study, dosages used for evaluation of analgesic activities were in accordance with their respective indication provided in previous published studies .

Hot Plate method – Thermal Method

To assess nociceptive responses to thermal stimuli, hot plate method was used. The experimental drugs were given 30 minutes before the start of experiment. Then animals were placed on the Eddy's Hot plate analgesiometer with temperature setting controlled at 55±0.2 °C. Cut-off time was 15 seconds, with in that if they don't react they are removed from the hotplate without getting injured. Nociceptive response was defined as licking which is considered as a rapid response to painful thermal stimuli and jumping represents an emotional component of escaping. Time duration between placing the animals on hot plate and licking or jumping was considered as reaction time. The reaction time in control and treated animals was recorded at 0(basal time point), 30, 60, 120 minutes after treatment.

Acetic acid induced writhing – Chemical Method

To assess nociceptive responses to chemical stimuli, acetic acid induced writhing was used. The experimental drugs were given 30 minutes by oral gavage before the start of experiment. With the help of 27 gauge ½-inch needle 0.1 ml of 1% acetic acid solution was injected intra-peritoneally. The mice were kept in glass beaker for monitoring and five minutes were allowed to elapse during which the onset of writhes is noted. Then for a period of 10 minutes animals were monitored for number of writhes. For scoring purpose, a writhe is taken to be indicated by stretching of the abdomen with simultaneous stretching of at least one hind limb.

Percentage Inhibition (W %) is calculated as

$= \{(W_c - W_t)/ W_c\} \times 100$

Where,

W_c, No. of writhes in control group

W_t, No. of writhes in test group

Tail Clip Method - Mechanical Method

To assess mechanical stimulus of pain Haffener's tail clip method is used. Tail clip was applied before and after 60 minutes after oral administration of experimental drugs and the attempt to dislodge the clip is noticed. Animals not attempting to dislodge the clip within 10 sec were discarded. This is to ensure the safety of the animal to avoid any injury. Attempt to dislodge the clip is called as a response.

Percentage Inhibition of response (R %) is calculated by

$= \{(R_c - R_t)/ R_t\} \times 100$

Where,

R_c, Time in sec to response in control group

R_t, Time in sec to response in test group

STATISTICAL ANALYSIS

Results are expressed as mean ± Standard Error of Mean (SEM). Data was analysed using IBM SPSS Version 20. Comparison between different groups was done by One-Way Analysis of Variance (ANOVA) followed by a post hoc test Tukey's. P value less than 0.05 was considered statistically significant.

RESULTS

From the table 1, the mean reaction time at 30 minutes for all groups showed no apparent increase except for aspirin this increased significantly. This had a P value <0.001 not only with control but also with other study groups. But at 60 minutes and 120 minutes the mean reaction time of aspirin, rosuvastatin and combination group were statistically significant with P value <0.001. But telmisartan showed no increase in reaction time when compared with its basal reaction time. There is no significant difference of combination group and rosuvastatin in producing analgesia i.e. the contribution of telmisartan in causing analgesia must be negligible to none. Telmisartan effect on mean reaction time was only comparable to the control.

Table 1: Mean reaction time in seconds in Hot Plate method.

Groups	Basal	30 Min	60 Min	120 Min
Group I: Control	4.500±0.224	4.667±0.333	4.667±0.333	4.500±0.428
Group II: Aspirin (standard)	4.833±0.307	8.500±0.563 *	11.167±0.477 *	14.167±0.307 *
Group III: Rosuvastatin	4.833±0.543	5.333±0.211	7.667±0.422 *	9.333±0.211 *
Group IV: Telmisartan	4.667±0.333	5.000±0.258	5.333±0.422	5.833±0.477
Group V: Rosuvastatin + Telmisartan	4.833±0.307	5.333±0.333	7.833±0.477 *	10.333±0.494 *

***P<0.001 when compared with control of the same time point**

From table 2, onset of writhing was early in control and Telmisartan, Writhing occurred only after 5 minutes in Aspirin, Rosuvastatin and Combination groups, with Aspirin having the late mean onset. These three groups were having p <0.001 when compared to control.

The numbers of writhes in ten minutes were significantly less in Aspirin, Rosuvastatin and Combination groups with P value < 0.001 when compared to control, while there is no statistical significant difference from telmisartan to control. Also when compared with aspirin, rosuvastatin and telmisartan showed significant difference with P value <0.05 meaning there is a lack of analgesia produced by these drugs alone compared to aspirin. No such statistical difference existed when the means of combination group is compared with aspirin.

Percentage of analgesia is maximum for Aspirin with 89.57% and lowest for telmisartan with 12.80%. Percentage of analgesia more than 70% is taken as having at least minimal analgesia. Only Aspirin and Combination group has percentage of analgesia more than 70% with 89.57% and 72.04% respectively.

Table 2: Mean onset of writhing and number of writhing in 10minutes with percentage of analgesia developed from control in acetic acid writhing test.

Groups	Onset in seconds	No of Writhes in 10 min	% of analgesics from control
Group I: Control	2.717±0.268	35.167±1.682 ^	Not Applicable
Group II: Aspirin (standard)	6.783±0.101 *	3.667±0.494 *	89.57%
Group III: Rosuvastatin	5.317±0.119 *	13.667±0.803 *^	61.14%
Group IV: Telmisartan	3.017±0.266	30.667±2.951 ^	12.80%
Group V: Rosuvastatin + Telmisartan	6.033±0.067 *	9.833±0.601 *	72.04%

***p<0.001 when compared with control, ^p<0.05 when compared with aspirin**

It is evident from table 3 the mean reaction time of mice was higher for aspirin group. But aspirin group, Rosuvastatin group and rosuvastatin and telmisartan group had a very high significance of p<0.001 when compared to control. Also the telmisartan group has a significance level of p<0.05 when compared to control. Aspirin group and rosuvastatin and telmisartan combination group had the percentage of inhibition of above 50%, while rosuvastatin alone had inhibition of 31% which 10% higher than telmisartan alone.

Table 3: Mean response time in seconds before administration of test drug and after 60 minutes of drug administration. Also % of inhibition of response is given.

Groups	Basal in sec	60 min after treatment in sec	% of inhibition
Group I: Control	1.58±0.268	1.58±0.268	Not Applicable
Group II: Aspirin (standard)	1.21±0.101	3.267±0.494 *	62.96%
Group III: Rosuvastatin	1.60±0.119	2.337±0.803 *	31.53%
Group IV: Telmisartan	1.55±0.266	1.98±2.951 ^	21.71%
Group V: Rosuvastatin + Telmisartan	1.24±0.067	2.687±0.601 *	53.85%

***p<0.001 when compared with control, ^p<0.05 when compared with control**

DISCUSSION

Hot plate method is used to test the pain response in animals. It is used in basic pain research an in testing the effectiveness of analgesics by observing the reaction to pain caused by heat. It was first proposed by Woolfe and MacDonald, although the version most often used today is as modified by Eddy and Leimbach in 1953. This is a behavioural model of nociception effective in estimating the efficacy and potency of centrally acting analgesics. This was evident in this study wherein the pain threshold increased significantly during the period of observations in all treatment groups. But the increase in rosuvastatin and telmisartan alone is comparatively less than when these drugs are combined. Above findings also shows that if any synergistic analgesic action exist in combination group it is clearly not significant. It is also evident that there is no pain reducing effect of telmisartan, which can be only comparable with control.

Acetic acid induced writhing experiment is used to test nociceptive response due to chemicals in animals. This is a largely developed and still used on mice. The technique dates back to the mid 1950's when two groups of people described a syndrome of writhing – lengthwise stretches of the torso accompanied by concave arching of the back produced by phenylquinone or acetic acid, which was sensitive to abolition by a wide range of analgesics including Non-Steroidal anti-inflammatory drugs. In this model, compounds with percentage analgesia of less than 70% are considered to have minimal analgesic activity. Percentage analgesia with both rosuvastatin and telmisartan was less than 70%, and it was more than 80% only in the aspirin treated animals. But the combination group showed activity more than 70%. The writhing response induced by acetic acid is a sensitive procedure to establish peripherally acting analgesics. This response is thought to involve local peritoneal receptors by causing peritonitis like condition. As this method mainly evaluates peripherally acting analgesics, maximum analgesic activity of aspirin was observed in this model. Rosuvastatin alone also exhibited significant analgesic activity though less than aspirin. Selective analgesic action of statins or angiotensin receptor blocker has not been evaluated in commonly used experimental models of analgesia; hence, there is no comparative data. Data regarding angiotensin receptor blocker as analgesic agent is scarce. But from our experiment we can come to a conclusion that telmisartan has no analgesic activity and rosuvastatin having a percentage inhibition closer to 70% helps us in declaring rosuvastatin having minimal analgesic activity. It is also seen that the combination group has a higher analgesic effect when compared to the drugs given alone. Also this difference is statistically significant only with telmisartan and not with rosuvastatin.

Haffner's tail clip method is done to study the mechanical pain stimulation. These experiments provide the least burden to the animals being tested. The reflex mechanism involved in dislodging the tail clip is based on the higher centres. Here more than 50% inhibition of response to mechanical painful stimulation occurred in obvious aspirin group and the combination of rosuvastatin and telmisartan group.

Analgesic action of these drugs is poor when compared to aspirin but some significant analgesic action exists in rosuvastatin and combination group. It is also clear that there is no synergistic action exists between these drugs for relieving pain. There were also no investigation done to identify the underlying mechanism by which Rosuvastatin inhibits nociception; however, it has been reported that bradykinin, (BK), tumour necrosis factor-a (TNF- a), interleukin-1b (IL-1b) and the chemokine CXCL induce approximately the same intensity of hypernociception, and pretreatment with statins reduced each of these hypernociceptive states, to about the same level. Rosuvastatin also has been reported to reduce prostaglandin E2 (PGE2)-induced hypernociception.

There are many superior analgesics available, so use of these drugs solely for that indications cannot be justified. But conditions with coexisting hypertension, dyslipidemia and nociception may benefit from these additional properties new studies are needed to identify the additional properties of these chronically used drugs so that newer indications of these drugs may benefit population by reducing the drug load.

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