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ANALGESIC ACTIVITY OF ALCOHOLIC EXTRACT OF AZAIRACHTA INDICA (NEEM) ROOT BARK

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Summary

The present study was done to evaluate analgesic activity of alcoholic extract of *Azadirachta Indica* (Neem) root bark(NRBE). 70% alcoholic extract of neem root bark was given in the dose of 200,400 and 800 mg/kg in Wistar Albino rats and mice of either sex in Tail flick and acetic acid writhing method respectively. NRBE was compared with standard drugs like Buprenorphine (0.05 mg/kg S.C.) and Aspirin (100 mg/kg).Our study shows that NRBE is having analgesic activity by both central and peripheral mechanisms.

Keywords:-Azadirachta Indica, root, analgesic, writhing, tail flick.

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Introduction

Neem plant was tested for different activities. Isolation of active gradient from neem was started by Siddiqui in 1942. Then more than 135 compounds were isolated from different parts of neem(1). These extracts were having different activities e.g. aqueous extract of neem leaf have antiviral (2), antiulcer (3), and hepatoprotective activity (4). Neem seed oil has spermicidal activity (5). Chloroform extract of stem bark has anti-inflammatory activity(6). Acetone leaf extract has central nervous system depressant action(7). Fresh juice of young stem bark of neem is having analgesic activity(8). Analgesic activity of neem root bark was not evaluated. So we evaluated the activity of neem root bark in our study.

Material and Methods

Plant material: Roots were collected from neem tree in Navodaya Medical College campus. The roots were shade dried in department of pharmacology. Bark was removed from roots. Shade dried roots were powdered using mixer and grinder.

Preparation of extract: Alcoholic extract of root bark powder was obtained by continuous extraction in percolator using 70% ethyl alcohol. Fresh solution was prepared by dissolving extracts in distilled water before each experimental procedure.

Animals:- Albino rats of either sex (200-250 grams) and Albino mice of either sex (30-50 grams) were used in this study. These animals were procured from National Institute of Nutrition, Hyderabad. Animals were kept under 12 hours light and 12 hours dark cycle.Animals were fed on standard food and water ad libitum. The study was conducted according to guidelines of Institutional animal ethical committee.

Chemicals, drugs and Instrument: Buprenorphine (Buprinor 2 ml injection Astra Zeneca) and Tablet Aspirin 75 mg in each tablet (Ecosprin-USV) were purchased from pharmacy shop. Acetic acid was purchased from s.d. fine-chem Ltd. Boisar. For tail flick method we used Analgesiometer from Inco (Ambala)

Analgesic activity: Analgesic activity of NRBE was evaluated by using tail flick and acetic acid writhing method.

Tail flick method: Albino rats of either sex were fasted for 12 hours. Basal reaction time to radiant heat was taken by placing tip (last 1-2 cms) of the tail on nicrome wire of analgesiometer. Tail withdrawal from the heat is taken as end point. Normal reaction time was 3-4 seconds. Animals failing to withdraw their tail within 3-4 seconds were discarded. Cut off time was taken as 10 seconds to avoid excessive damage to the tail. Animals were divided into 5 groups (each containing 6 rats). Group I (Control) received 2 ml of distilled water orally. Group II (Standard) received Buprenorphine (0.05 mg/kg S.C.) (9) 30 minutes before exposure to radiant heat. Group III, IV, V received NRBE in the dose of 200, 400, 800 mg/kg orally respectively 60 minutes before exposure to radian heat. Reaction time was taken at 15, 30, 60 and 90 minutes. (10)

Acetic acid writhing method: Mice of either sex were fasted for 12 hours. Acetic Acid (0.6%) 0.1 ml was injected intraperitoneally in all mice. Response was observed as abdominal contraction and relaxation with hind limb extension for 20 minutes. Mice failed to produce writhing were discarded. Then animals were divided into 5 groups similar to tail flick method. Group I (Control) received 2 ml of distilled water orally. Group II (Standard) received Aspirin (100 mg/kg orally) (11) 60 minutes before acetic acid injection. Group III, IV, V received NRBE in the dose of 200, 400, 800 mg/kg orally respectively 60 minutes before acetic acid injection. Mice were observed for 20 minutes to count number of writhings in each group (12).

Statistical Analysis: Analysis was done using SPSS software. Analysis of Variance (ANOVA) followed by Dunnet test was used for analysis.

Results

In tail flick method, Buprenorphine showed increase in reaction time at 15 minutes and maximum reaction time at 90 minutes. With neem root extract 200 mg/kg and 400 mg/kg there was no significant analgesic effect. But with dose 800 mg/kg showed maximum increase in reaction time at 60 minutes which was statistically significant (P < 0.05)



Table 1: Analgesic effect of Azadirachta Indica (Neem) root bark extract in albino rats (Tail flick method)

Groups(Drug) (n=6)	Dose	B.D.A.	A.D.A.			
			15 min	30 min	60 min	90 min
I(Control)	2 ml	3.15±0.1				
		3	3.13±0.17	3.23±0.17	3.31±0.25	3.28±0.27
II(Buprenorphine)	0.05 mg/kg	3.51±0.3	5.80±0.22	7.05±0.75	8.05±0.51*	8.66±0.37*
		8	**	**	*	*
III(NRBE)	200 mg/kg	3.15±0.1				
		7	3.13±0.17	3.23±0.17	3.31±0.17	3.28±0.19

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IV(NRBE)	400 mg/kg	3.16±0.1					
		6	4.30±0.40	5.08±0.51	6.28±0.21	6.70±0.32	
V(NRBE)	800 mg/kg	3.28±0.2	4.61±0.26	5.81±0.54			
		3	*	*	6.76±0.56*	7.41±0.39*	
* P < 0.05, ** P < 0.01, *** P < 0.001 Mean±SEM							

In acetic acid writhing method, Aspirin (100 mg/kg) showed significant activity (p < 0.01) in comparison to control. Neem root extract in the dose of 400 mg/kg and 800 mg/kg showed significant (p < 0.05, p < 0.01) analgesic activity in comparison to control.



Table 2: Analgesic effect of Azadirachta Indica (Neem) root bark extract in albino rats (Acetic acid writhing)

2000	Writhing response		
	Number of writhings	% of inhibition	
2ml orally	43± 0.71	00	
100 mg /kg orally	18 ±0.65	58.13***	
200 mg / kg orally	42 ±1.1	2.32	
400 mg / kg orally	41 ±0.33	4.65^{*}	
800 mg / kg orally	35 ±0.71	18.60**	
	2ml orally 100 mg /kg orally 200 mg / kg orally 400 mg / kg orally 800 mg / kg orally	Number of writhings $2ml$ orally 43 ± 0.71 100 mg/kg orally 18 ± 0.65 200 mg/kg orally 42 ± 1.1 400 mg/kg orally 41 ± 0.33 800 mg/kg orally 35 ± 0.71	

* P < 0.05, ** P < 0.01, *** P < 0.001 Mean±SEM

Discussion

Neem leaf and neem seed oil are having analgesic action (13). Effect of neem leaf was seen in both tail flick method and acetic acid writhing suggesting that it has central as well as peripheral mechanism of action (14). Combination of morphine and neem leaf extract produce greater analgesia with lesser side effect(15). In our study we got analgesic activity of neem root bark extract in both peripheral and central mechanism animal models. In

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comparison to standard drugs it was having very weak analgesic action. Neem root bark contains terpanoids like nimbin and nimbidin.(16). Pillai et al (1980) showed that nimbidin has analgesic activity.(17). The reason for weak analgesic activity may be quantity of nimbidin which is present in root bark. Other chemical constituents may also be responsible for analgesic activity. These chemicals can be taken for further studies on analgesic activity.

Conclusion

Neem root bark extract have weak analgesic activity with central and peripheral mechanism. Further studies are required to find out which active ingradients are responsible for analgesic action.

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