Original Research Article

An Experimental Study to find out the Additive Analgesic Effect of Hydro-Alcoholic Extract of Ginger with Paracetamol and Ibuprofen in Management of Pain in Mice

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Abstract

Present study was carried out on adult Swiss albino mice of both sex, consisting of six groups with six mice in each group to find out additive analgesic effect of Hydro-alcoholic extract of Ginger with Paracetamol and also to find out additive analgesic effect of Hydro-alcoholic extract of Ginger with Ibuprofen by hot plate and tail flick methods. These six groups were, Groups-A paracetamol, group-B was Ibuprofen, group-C was distilled water, group-D (Hydro-alcoholic extract of Ginger, group-E (Paracetamol + Hydro-alcoholic extract of Ginger), and group-F (Ibuprofen + Hydro-alcoholic extract of Ginger). We find that Paracetamol, ibuprofen has analgesic effect of their own on mice but we did not find any significant analgesic effect of Ginger on mice in singly or additive analgesic effect in combination with ibuprofen or paracetamol. So, we may conclude that Ginger has no additive analgesic effect and no analgesic effect on single use also.
Keywords: Ginger, analgesic, additive, paracetamol, ibuprofen, mice.

INTRODUCTION

Pain as an unpleasant sensory or emotional experience associated with tissue damage. I.A.S.P. committee for Taxonomy defined pain as "an unpleasant sensory or emotional experience associated with actual or potential tissue damage, or describe in terms of such damage [1]". Pain is the most common reason for individuals to seek medical care [2].

Pain motivates the individual to withdraw from damaging situations, to protect a damaged body parts while it heals, and to avoid similar experiences in the future. . Sometimes pain may arise in the absence of any detectable stimulus, damage or disease [3].

Analgesics are drugs that selectively relieve the pain by acting in the CNS or on peripheral pain pathway, without significantly altering consciousness [4]. Analgesic is divided into two groups, Opioids & non-opioids NSAIDs (non-steroidal anti-inflammatory drugs).

Inflammation means pain, redness, swelling and heat that usually developed in response to injury.

The management of acute and chronic pain is important for the patient's wellbeing. Though opioids analgesic like morphine, pethidine is good painkiller but they have abuse potential and lots of side effects may occur such as respiratory depression, tolerance and dependence etc [5]. Non-steroidal anti-inflammatory drugs(NSAIDS) are also good analgesic but also have many adverse effect like hypersensitivity reaction, hepatic damage (Reye's syndrome), acid peptic disorder, gastric ulcer, neuropathy, hemolytic anemia etc [6,7]. The choice of analgesics for acute pain depends on the efficacy, side effects, complications, pharmacokinetics and its cost-effectiveness etc [8].

Ginger (Zingiber *Officinale*) is a flowering plant. Its rhizome or ginger root is called ginger [9, 10]. It is widely used as a spice all over the world and also use as a folk medicine [11]. It is presumed that Ginger has some medicinal property such as analgesic, anti-inflammatory, hypoglycemic, Anti-oxidant, Anti-emetic, Anti-tumorigenic effect [12]. Few research works was published on concomitant use of ginger with NSAIDs (Paracetamol and Ibuprofen) in management of pain. Study also showed that ginger (Zingiber

officinale) rhizomes Hydro-alcoholic extract possesses analgesic, anti-inflammatory and hypoglycaemic properties [13].

In some diseased condition of kidney (such as renal failure), heart (cardiac failure, myocardial infarction, hypertension), liver (hepatic failure), GI tract disorders (gastric Ulcer) dose reduction of NSAID is required and this reduce doses also decrease the analgesic effect of NSAID [14-16]. If we find that Ginger may have additive analgesic effect with these two NSAID (paracetamol or Ibuprofen), we may use this two commonly used NSAID effectively and safely for longer periods in reduced doses with low risk of adverse effects.

In this study we have used two types of NSAIDs (paracetamol and Ibuprofen). Paracetamol is a centrally acting analgesic and Ibuprofen is a peripherally acting analgesic [17, 18]. By using two types of NSAIDS we want to find out whether additive analgesic effect of Ginger is due to central and/or peripheral effect.

OBJECTIVES
Primary objectives
To find outt additive analgesic effect of Ginger in concomitant use with Paracetamol or Ibuprofen in management of pain in mice

Secondary objectives
To find out the analgesic effect of Ginger extract if present acting either centrally or peripherally.

MATERIAL AND METHODS
Type of study: Animal experimental study.
Study Duration: From April 2017 to September 2018.
Place of study- Department of Pharmacology, R.G.Kar Medical College, Kolkata, India.
Sample Size: 36 adult Swiss albino mice
Study Tool: 36 albino mice weighing about 25gm, hot plate apparatus, and tail flick apparatus.

Ethical permission
Necessary permission was taken from the Institutional Animal Ethics
Committee and study was performed following the guidelines of the Committee for the Purpose of Control and Supervision on Experiments on Animals (CPCSEA), Animal Welfare division, Ministry of Environment, Govt. of India. The care and use of the laboratory animals were strictly followed throughout the experimental procedure.

Procedure: - In this study we used adult Swiss albino mice weight 25 ± 2 gm [19]. Mice were divided into Six groups and in each group there are six mice. The groups are as follows:-
Group- A= 6 mice (For Paracetamol)
Group- B = 6 mice (For Ibuprofen)
Group- C = 6 mice (For control, treat with Distilled water)
Group- D = 6 mice (For Hydro-alcoholic extract of Ginger)
Group- E = 6 mice (For Paracetamol+ Hydro-alcoholic extract of Ginger)
Group- F = 6 mice (For Ibuprofen+ Hydro-alcoholic extract of Ginger)

Calculation of dose of drugs:-Dose of drug for mice was calculated by dose conversion from human dose to mouse dose. From human dose to mouse dose conversion factor is 12.3 [20].

Calculation of dose of mouse (weight=25 gm):-Paracetamol dose for human= 15 mg / Kg. per oral. So, Paracetamol dose for mouse = (15 x 12.3) = 184.5 mg/ Kg. Now, 184.5 mg/Kg rounded off to 185 mg/Kg. So, Paracetamol dose for mouse (weight =25 gm) = (185 x 25) / 1000 = 4.62 mg. in this way we calculated the dose of ibuprofen was 3 mg for a mouse weighing 25 gm. and 20 mg Hydro-Alcoholic extract of Ginger(800mg/kg).

Two types of tests were done, (Hot plate and Tail flick) to study of analgesic effect of drugs. Test reading were taken at intervals of 20 minutes, 60 minutes and 90 minutes as per standard protocol. Then data were compared by statistical analysis to obtain the result. Firstly, data of Paracetamol , Ibuprofen and Ginger treated groups were compared with control (distilled water) group to show there analgesic effects individually, then data of paracetamol treated group were compared with data obtained from paracetamol + Ginger treated group and data of Ibuprofen treated group were compared with data obtained from Ibuprofen + Ginger treated group to show additive analgesic effects.

RESULTS
Statistical data N, mean, std. deviation etc. of Hot plate and tail flick method test are given in the table 1 to 10.

Table-01: Group Statistics of Hot plate test

Group = A & C	N	Mean	Std. Deviation	t	P value
@20_min Group- A	6	5.1833	0.50365	10.922	0.000**
Group-C	6	2.4000	0.36878		
@60_min Group-A	6	7.0167	0.51153	19.550	0.000**
Group-C	6	2.3333	0.28752		
@90_min Group-A	6	9.0667	0.79415	19.675	0.000**
Group-C	6	2.3833	0.24833		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table: - 01 shows that, Group-A (Paracetamol) shows strongly significant analgesic effect (P value < 0.01) on mice in all the times (20, 60 and 90 min) when we compare with Group-C (control).

Table-02: Group Statistics of Tail flick test

Group = A & C	N	Mean	Std. Deviation	t	P value
@20_min Group-A	6	5.3000	0.65422	9.393	0.000**
Group-C	6	2.6833	0.19408		
@60_min Group-A	6	7.5333	0.68313	15.828	0.000**
Group-C	6	2.7833	0.27142		
@90_min Group-A	6	9.4667	0.70899	22.168	0.000**
Group-C	6	2.8000	0.20000		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table: - 02 shows that, Group-A (Paracetamol) shows strongly significant analgesic effect (P value < 0.01) on mice in all the times (20, 60 and 90 min) when we compare with Group-C (control).

Table-03: Group Statistics of hot plate test

Group = B & C	N	Mean	Std. Deviation	t	P value
@20_min Group-B	6	6.0333	0.72847	10.900	0.000**
Group-C	6	2.4000	0.36878		
@60_min Group-B	6	8.3167	0.48751	25.895	0.000**
Group-C	6	2.3333	0.28752		
@90_min Group-B	6	10.1500	0.87579	20.899	0.000**
Group-C	6	2.3833	0.24833		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table: - 03 shows that, Group-B (Ibuprofen) shows strongly significant analgesic effect (P value < 0.01) on mice in all the times (20, 60 and 90 min) when we compare with Group- C (control).

Table-04: Group Statistics of Tail flick test

Group = B & C	N	Mean	Std. Deviation	t	P value
@20_min Group-B	6	6.6500	0.39875	21.910	0.000**
Group-C	6	2.6833	0.19408		
@60_min Group-B	6	9.0167	0.62423	22.431	0.000**
Group-C	6	2.7833	0.27142		
@90_min Group-B	6	10.8667	0.85245	22.567	0.000**
Group-C	6	2.8000	0.20000		

*= Significant (P value < 0.05) **= strongly Significant (P value < 0.01)

In this Table: - 04 shows that, Group-B (Ibuprofen) shows strongly significant analgesic effect (P value < 0.01) on mice in all the times (20, 60 and 90 min) when we compare with Group-C (control).

Table-05: Group Statistics of hot plate test

Group = D & C	N	Mean	Std. Deviation	t	P value
@20_min Group-D	6	2.4500	0.32711	0.248	0.809
Group-C	6	2.4000	0.36878		
@60_min Group-D	6	2.3167	0.19408	-0.118	0.909
Group-C	6	2.3333	0.28752		
@90_min Group-D	6	2.3667	0.38816	-0.089	0.931
Group-C	6	2.3833	0.24833		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table: - 05 shows that, Group-D (Ginger) has no significant analgesic effect (P value >0.05) on mice in all the times (20, 60 and 90 min) when we compare with Group C (control).

Table-06: Group Statistics of Tail flick test

Group = D & C	N	Mean	Std. Deviation	t	P value
@20_min Group- D	6	2.7000	0.14142	0.170	0.868
Group- C	6	2.6833	0.19408		
@60_min Group- D	6	2.7667	0.34448	-0.093	0.928
Group- C	6	2.7833	0.27142		
@90_min Group- D	6	2.8167	0.24833	0.128	0.901
Group- C	6	2.8000	0.20000		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table: - 06 shows that, Group-D (Ginger) has no significant analgesic effect (P value >0.05) on mice in all the times (20, 60 and 90 min) when we compare with Group C (control).

7) Statistical data (N, mean, std. deviation, etc) of Hot plate test of Group-A (Paracetamol) and Group-E (Paracetamol + Ginger) are given in the table :-07.

Table-07: Group Statistics of hot plate test

Group = A & E	N	Mean	Std. Deviation	t	P value
@20_min Group-A	6	5.1833	0.50365	-0.349	0.734
Group-E	6	5.3000	0.64498		
@60_min Group-A	6	7.0167	0.51153	0.084	0.934
Group-E	6	6.9833	0.82077		
@90_min Group-A	6	9.0667	0.79415	-0.245	0.812
Group-E	6	9.1833	0.85654		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table:- 07 shows that, Group- E (Paracetamol + Ginger) shows no additive analgesic effect (P value > 0.05) on mice in all the times (20, 60 and 90 min) when we compare with Group- A (Paracetamol).

Table-08: Group Statistics of Tail flick test

Group = A & E	N	Mean	Std. Deviation	t	P value
@20_min Group-A	6	5.3000	0.65422	0.049	0.962
Group-E	6	5.3167	0.51929		
@60_min	6	7.5333	0.68313	0.043	0.967
Group-A Group-E	6	7.5500	0.67454		
@90_min	6	9.4667	0.70899	0.041	0.968
Group-A Group-E	6	9.4500	0.70922		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table:- 08 shows that, Group- E (Paracetamol + Ginger) shows no additive analgesic effect (P value > 0.05) on mice in all the times (20, 60 and 90 min) when we compare with Group- A (Paracetamol).

Table-09: Group Statistics of hot plate test

Group = B & F	N	Mean	Std. Deviation	t	P value
@20_min Group-B	6	6.0333	0.72847	-0.215	0.834
Group-F	6	6.1000	0.21909		
@60_min Group-B	6	8.3167	0.48751	-0.144	0.888
Group-F	6	8.3500	0.28810		
@90_min Group-B	6	10.1500	0.87579	-0.095	0.926
Group-F	6	10.2000	0.94234		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In this Table:- 09 shows that , Group- F (Ibuprofen + Ginger) shows no additive analgesic effect (P value > 0.05) on mice in all the times (20, 60 and 90 min) when we compare with Group- B (Ibuprofen).

Table-10: Group Statistics of Tail flick test

Group = B & F	N	Mean	Std. Deviation	t	P value
@20_min Group-B	6	6.6500	0.39875	0.124	0.904
Group-F	6	6.6167	0.52694		
@60_min Group-B	6	9.0167	0.62423	-0.282	0.784
Group-F	6	9.1167	0.60470		
@90_min Group-B	6	10.8667	0.85245	-0.106	0.918
Group-F	6	10.9167	0.77825		

*= Significant (P value < 0.05)   **= strongly Significant (P value < 0.01)

In the Table: - 10 shows that, Group- F (Ibuprofen + Ginger) shows no additive analgesic effect (P value > 0.05) on mice in all the times (20, 60 and 90 min) when we compare with Group- B (Ibuprofen).

DISCUSSIONS

Data obtained from hot plate and tail flick study of group-A (paracetamol) and group-C (distilled water) at time interval of 20, 60 and 90 min are compared between these two groups. We find that, P value is <0.01 in all the time. So, from this hot plate study we can say paracetamol has significant analgesic effect on mice [21].

In literature also, it is mentioned that paracetamol is highly selective for central COX-2 activities in the brain, which may contribute to its ability to treat fever and pain [22, 23]. This activity does not appear to be direct inhibition by blocking an active site, but rather by reducing COX-3, which must be oxidized in order to function [22].

Data obtained from tail flick and hot plate study of group-B (Ibuprofen) and group-C (distilled water) at time interval of 20, 60 and 90 min are compared between these two groups. We find that, P value is <0.01 in all the time. So, from this study we can say Ibuprofen has significant analgesic effect on mice [24]. Ibuprofen is a non-steroidal anti-inflammatory drug (NSAID) drug [25]. It is used for treating pain, fever, and inflammation. So our study also established its pain killing property.

Data obtained from tail flick and hot plate study of group-D (Hydro-alcoholic extract of Ginger) and group-C (distilled water) at time interval of 20, 60 and 90 min are compared between these two groups. We find that, P value is >0.05 in all the time. So, from this hot plate study we can say Ginger has no analgesic effect on mice.

Data obtained from tail flick and hot plate study of group-A (paracetamol) and group-E (Paracetamol + Hydro-alcoholic extract of Ginger) at time interval of 20, 60 and 90 min are compared between these two groups. We find that, P value is >0.05 in all the time. So, from this hot plate study we can say Ginger has no additive analgesic effect on mice and also has no analgesic effect, which we found in the previous discussion [26-28].

Data obtained from hot plate and tail flick study of group-B (Ibuprofen) and group-F (Ibuprofen + Hydro-alcoholic extract of Ginger) at time interval of 20, 60 and 90 min are compared between these two groups. We find that, P value is >0.05 in all the time. So, from this hot plate study we can say Ginger has no additive analgesic effect on mice and also has no analgesic effect, which we found in the previous discussion [26-28]. In a study Black C.D. *et al*. found that a single 2 g dose of ginger does not attenuate eccentric exercise-induced muscle pain, inflammation or dysfunction 45 min after ingestion. However, ginger may attenuate the day-to-day progression of muscle pain [27].

CONCLUSION

Ginger has no analgesic effect on single use and Ginger has no additive analgesic effect with Paracetamol and Ibuprofen.

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