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ANTI-INFLAMMATORY, ANALGESIC AND ANTIPYRETIC ACTIVITIES OF GINGER (ZINGIBER OFFICINALE)

BY

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ABSTRACT

In the present study, the *ginger extracts* (either alcoholic or watery) *were* evaluated for pharmacological activity in rats and mice. The anti-inflammatory and analgesic activities were evaluated in mice (40 mice) while, the antipyretic activity was evaluated in rats (16 rats). The obtained results revealed that both extracts (300 and 300 mg/kg, p. o.) were found to possess, anti-inflammatory, analgesic and antipyretic activities in a time-dependent manner and the effect was comparable with that produced by the standard drug (indomethacin).

INTRODUCTION

Medicinal plants are of great importance to the health of individuals and communities. The medicinal value of these plants lies in some chemical substances that produce a definite physiological action on the human body **(Edeoga et al, 2005).** The most important of bioactive constituents of plants are alkaloids, tannins, flavonoids, and phenolic compounds **(Hill, 1952).** The use of herbs to treat disease is almost universal among non-industrialized societies. Many of the pharmaceuticals currently available to physicians have a long history of use as herbal remedies. The World Health Organization (WHO) estimates that 80 % of the world's population presently use herbal medicine for some aspect of primary health care. **(Edgar et al., 2002).**

Kar et al. (2004) reported that several plant products are claimed and proved to possess analgesic , anti-inflammatory ,and antipyretic properties such as ginger (Zingiber officinale), Citrullus colocynthis, ,Senna italica Mill (Sana mekki), Curcuma longa, Ricinus communis, Hyoscyamus muticus, Azadirachta indica, and Carissa caranda.

Ginger (Zingiber officinale, Z. officinale) has been used as a medicine since ancient time. In Asian medical practices, dried ginger has been used to treat stomachache, diarrhea and nausea. Recent studies have shown that Z. officinale has anti-inflammatory effects and reduces pain and swelling associated with either rheumatoid or osteoarthritis. It has also antioxidant effect, analgesic and anti-pyretic properties **(Murakami et al., 2002).**

The present study was designed to investigate:

1-The anti-inflammatory and analgesic effects of ginger in mice.

2- The antipyretic effect of ginger in adult albino rats.

MATERIALS AND METHODS

1-Materials

A- Medicinal plant:

A.1.Zingiber officinale L. (ginger)

- **Proper Name:** *Zingiber officinale* Roscoe (Zingiberaceae*)* **(USDA, 2003).**
- **Common Name (English name):** Ginger **(McGuffin *et al*., 2000).**

A.2. Dosage and administration:

Ginger was purchased from a local market and stored in dry atmosphere. The dry extract of ginger dissolved in 0.9% physiological saline solution and given (i.p) to the animals at a dose of 150 mg /kg b. wt. **(Raji Y. et al., 2002).**While the watery extract was administered to the animals at a dose of 150 mg /kg b. wt. **(Zahra et al., 2005).**

B. Drugs and chemicals:

B.1. Indomethacin

Indomethacin (50 mg) was purchased from PHARCO Company (PHARCO, Alexandria, Egypt) and it was injected intra-peritoneal in a dose 10 mg/kg b.wt**. (Amit et al., 2011)**.

B.2. Formalin:

Formalin (1%) was purchased from **El- Gomhoria Co., ARE** and was injected in a dose 0.1 ml for each mice.

B.3. Brewer's yeast (15%):

It was purchased from **Sigma Company** (Sigma, St. Louis, Mo, USA) and it was injected subcutaneous in a dose rate 10 ml/ kg b. wt**. (Mahesh et al., 2009).**

B.4.Ethyle alcohol. 95% (El- Nasr Company)

C. Experimental Animals:

A total of sixthteen (16) adult healthy male rats (average body weight 160-200 g), and forty (40) male mice (average body weight 25-30 g) were used in this study. Animals purchased from animal house in Helwan, and housed in Department of Pharmacology, Faculty of Veterinary medicine, Mansoura University. Animals were left for one week to acclimatize the place. Animals were kept in cages in a controlled environment, maintained under a 12 hours light: dark cycle, 24°C (± 3°C) and 50-70% humidity. Rats and mice were provided with standard diet and water ad-libitum.

METHODS

1. Preparation of medicinal plants extracts:

1. a. Ethanolic extract of ginger:

Ginger alcoholic extract was prepared according to **Raji Y. et al., (2002).**

1. d. Aqueous extract of ginger:

Ginger aqueous extract was prepared according to **Zahra et al., (2005).**

3. Experimental Design for ginger:

The experiment was conducted on 40 male mice for estimation of anti-inflammatory and analgesic activity and 16 rats for determination of antipyretic activity. After one week period of acclimatization in cages condition, mice were divided into 2 main groups (each of 20 mice). The 1st main group was served for anti-inflammatory effects, while the 2nd was served for analgesic effect. Each of the 1st and 2nd main groups were divided into 4 subgroups (each of 5 mice) and were served as the following:

Group I **:** control +ve given 0.2 ml (i.p) physiological saline

Group II : treated with 10 mg/kg b.wt (i.p) Indomethacin.

Group III : treated with 150 mg/kg b.wt (i.p) of aqueous ginger extract.

Group IV **:** treated with 150 mg/kg b.wt (i.p) of alcoholic ginger extract.

In case of pyrexia 16 rats were used and divided into 4 groups (each group consist of 4 rats) as the following:

Group I **:** control feverish given 0.2 ml (i.p) physiological saline

Group II : Feverish treated with 10 mg/kg b.wt (i.p) Indomethacin.

Group III : Feverish treated with 150 mg/kg b.wt (i.p) of aqueous ginger extract.

Group IV : Feverish treated with 150 mg/kg b.wt (i.p) of alcoholic ginger extract.

6- Anti-inflammatory activity of ginger:

The anti-inflammatory activity was evaluated by formalin induced hind paw edema method **(Fhernanda et al., 2008).** The percentage inhibition of inflammation was computed using the formula by **Adedapo et al. (2008)** using the formula:

$$\% \text{ inhibition} = \frac{D_0 - D_t}{D_0} X100$$

Where,

D_0 = the average inflammation (hind paw oedema) of the negative control group at a given time period.

D_t = the average inflammation (hind paw oedema) of the treated group at a given time period.

7- Analgesic activity of ginger:

Analgesic activity of ginger extracts were evaluated using hot plate method. **(Mohamed et al., 2011).**

8- Antipyretic activity of ginger:

The antipyretic activities of extract were evaluated using Brewer's yeast induced pyrexia in Wister rats. **(Mahesh et al., 2009).**

11- Statistical Analysis:

Data were subjected to statistical analysis using statistical software program (SPSS for Windows, version 20, USA, **Levesque, 2007)**. Means and standard error for each variable were estimated. Differences between means of different groups were carried out using one way ANOVA with Duncan multiple comparison tests. Dissimilar superscript letters in the same column show a significance ($P<0.05$) **(Snedecor and Cochran, 1981)**.

RESULTS

3- Anti-inflammatory effect of ginger:

The obtained results in Table (1) recorded the paw volumes and percent inhibition of oedema for the various groups at different time intervals. The paw oedema showed gradual reduction with the time. The reduction caused by the extract was significantly at $P<0.05$ when compared with the control group and indomethacin treated group. The percent inhibition of oedema in group treated with 150 mg/kg of alcoholic extract of ginger were 27.69, 50.67, 60, 67.07 and 75.29% for the 1st, 2nd, 3rd, 4th and 5th hour respectively. While the percent inhibition of oedema in group treated with 150 mg/kg of watery extract of ginger were 20, 44, 53.75, 58.54 and 68.24% respectively. The oedema reduction for the ginger treated groups was more than that observed for the standard indomethacin, anti-inflammatory drug, which were 10.77, 36, 46.25, 51.22 and 61.18% for the 1st, 2nd, 3rd, 4th and5th hour respectively.

4- Analgesic activity of ginger:

It was evident from Table (2) that there was an increase in the animal reaction time to the heat stimulus. Values were found to be significant ($p < 0.05$) from 30 to 210 min after treatment with indomethacin, ethanol and aqueous extracts of ginger, respectively, when compared with control group. The highest reaction time in seconds was observed for ethanol and aqueous extracts of *ginger* at a dose of 150 mg/kg body weight (15.18±0.10 and 14.20±0.09 sec., respectively) when compared with indomethacin treated group (13.34±0.11).

5- Antipyretic effect of ginger:

The results of the effect of either alcoholic or watery ginger extracts at 150 mg/kg b.wt against brewer's yeast induced pyrexia were recorded in Table (3). There was a progressive time dependent reduction in temperature of rats treated with alcoholic extract (34.9 ± 0.07), watery extract (35.35 ± 0.06) and indomethacin (35.8 ± 0.11) at 180 minutes post treatment. The reductions induced by both extracts were significantly decreased ($P<0.05$) lower when compared to that obtained by indomethacin (10mg/kg).

DISCUSSION

Ginger is an essential ingredient in many traditional medicine and has been used since the 4th century BC. Ginger has extensive medicinal history. It is used as spice in food and beverages and in traditional medicine as carminative, antipyretic and in the treatment of pain, rheumatism and bronchitis **(Afzal et al., 2001).** Its extracts have been extensively studied for a broad range of biological activities including antibacterial **(Azu et al., 2007)**, analgesic and anti-inflammatory **(Grzanna et al., 2005)**.

Anti-inflammatory effect of ginger:

The present study revealed some of the pharmacological basis for the ethnomedicinal use of ginger in the treatment of inflammation. The ethanol extract of ginger showed a good anti-inflammatory activity against inflammation, suppressing the rat paw edema both at the early and later phases. Oedema results from the action of inflammatory mediators such as histamine, serotonin and bradykinin at the site of a local inflammatory insult **(Harriot et al., 2004)**. The early phase of edema, beginning from 1 h after the administration of the irritant, is due to the release of histamine and serotonin, while the later phase, occurring from 3 to 5 h after the administration of the irritant is induced by bradykinin, protease, prostaglandin and lysosome **(Wallace, 2002; and Harriot et al., 2004).**

In current study, administration of ginger extracts produced a significant reduction in the paw edema and percent inhibition of edema in rats in comparison with control group and indomethacin treated group. The reduction in edema evinced by ginger extract in this study suggests that it contains active constituents which block the release of histamine and serotonin from mast cells and inhibit the activity of other inflammatory mediators. This result agree with the earlier results of **Suekewa and Yuasa (1986)** who showed that (6)-shogaol isolated from ginger extract inhibited experimentally- induced swelling of the hind paw in rats, due to the ability of (6)-shogaol to inhibit cyclooxygenase enzyme. In addition, **Sharma and Srivastava (1994)** also reported that ginger inhibited paw oedema in an experimentally induced arthritis in the right knee and paw of rats. Moreover, **Srivastava and Mustafa (1992)** stated that 75% of patients suffering from arthritis, osteoarthritis or muscular discomfort experienced relief in pain and swelling to varying degrees after powered ginger treatment for 3 months to 2.5 years. Also, **Shen et al. (2005)** stated that the anti-inflammatory effect of ginger roots on osteoarthritic cow chondrocytes due to its strong inhibition of COX-2 enzyme, pro-inflammatory cytokines and prostaglandins which are all components of the inflammatory

response. Furthermore, **Middleton and Kandaswami (1992) and Read (1995)** reported that the plant flavonoids, apart from their many pharmacological actions, it has the anti-inflammatory activity. Phytochemical results of many studies showed that ginger is abundantly rich in flavonoids. This suggests that the flavonoids in ginger may be one of its main active anti-inflammatory constituents (**Illavarasan et al., 2006**).

Antipyretic effect of ginger:

Spacer and Breder, (1994) stated that pyrexia was a result of secondary impact of infection, tissue damage, inflammation, malignancy or other diseased states. The infected or damaged tissue initiates the enhanced formation of pro-inflammatory mediators (cytokines like interleukin 1β, α, β and TNF-α) which increase the synthesis of PGE2 near pre-optic hypothalamus area thereby triggering the hypothalamus to elevate the body temperature. Most of the anti-pyretic drugs inhibit COX-2 expression thus inhibiting PGE2 biosynthesis and reduce the elevated body temperature.

The results of this study showed that there were a progressive time dependent reduction in the temperature of rats treated with both alcoholic, watery extracts and indomethacin at 180 minutes post treatment. The reductions caused by both extracts were significant at $P<0.05$ when compared to that induced by indomethacin (10mg/kg). The results are supported by the results recorded by **Thomson et al., (2002) and Ficker et al., (2003)** who stated that ginger may be a stronger inhibitor of prostaglandin (PGE_2) synthesis than indomethacin . In addition, **Shen et al., (2003)** reported that the ginger constituents inhibit arachidonic acid metabolism thus prostaglandin synthesis.

Analgesic activity of ginger:

The present study showed that there was an increase in the rat reaction time to the heat stimulus. Values were found to be significant ($p < 0.05$) from 30 at the beginning to 210 min after treatment with indomethacin, alcoholic and aqueous extracts of ginger, respectively, when compared with the control group.

The obtained results are in agreement with that of **Nurtjahja et al., (2005)** who reported that ginger has a strong analgesic action which in many cases act by COX-1 inhibition. In addition, **Sulaiman et al., (2009)** found that ginger produced analgesic activity at the central and peripheral levels based on its inhibitory activity in the hot plate test and abdominal constriction test, respectively. Furthermore, the extract was effective in blocking chemically

and thermally induced analgesic effects, a characteristic of strong analgesics, e.g. opioid agonists. The strong analgesic activity claimed was further supported by the findings stated that the ginger prolonged the latency to discomfort/pain in the hot plate test and inhibited nociception in both phases of the formalin test, as seen with many centrally acting analgesic drugs, e.g. morphine **(Chan et al., 1995; Pini et al., 1997).**

Hosseinzadeh and Younesi (2002) claimed that any plant extract is considered to have centrally mediated analgesic activity if it demonstrates inhibitory activity in the abdominal constriction and hot plate tests. It is generally accepted that the peripherally acting analgesics such as non-steroidal anti-inflammatory drugs exert their antinociceptive activity only in the former test, while centrally acting analgesics such as opioid agonists exhibit their antinociceptive activity in both tests.

Table (1): Anti-inflammatory effect of ginger extracts (either alcoholic or watery) at 150 mg/ kg b.wt on formalin -induced paw edema in mice. (Mean ± SE) (n=5).

Parameters / Groups	Paw volume and %inhibition of edema						
	0 time	30min	1hr	2hr	3hr	4hr	5hr
G1 (Control inflammed given 0.2ml normal saline)	0.22 ± 0.04^{a}	0.50 ± 0.07^{a}	0.65 ± 0.02^{a}	0.75 ± 0.02^{a}	0.80 ± 0.02^{a}	0.82 ± 0.01^{a}	0.85 ± 0.02^{a}
G2 (Inflamed treated with indomethacin at 10mg/kg b.wt)	0.21 ± 0.03^{a}	0.42 ± 0.06^{a}	0.58 ± 0.01^{b} (10.77%)	0.48 ± 0.01^{b} (36.0%)	0.43 ± 0.01^{b} (46.25%)	0.40 ± 0.01^{b} (51.22%)	0.33 ± 0.01^{b} (61.18%)
G3 (Inflammed treated with water extract of ginger at 150mg/kg b.wt)	0.15 ± 0.02^{a}	0.38 ± 0.06^{a}	0.52 ± 0.01^{c} (20.0%)	0.42 ± 0.01^{c} (44.0%)	0.37 ± 0.01^{c} (53.75%)	0.34 ± 0.01^{c} (58.54%)	0.27 ± 0.03^{c} (68.24%)
G4 (Inflammed treated with alcoholic extract of ginger at 150mg/kg b.wt)	0.16 ± 0.03^{a}	0.34 ± 0.05^{a}	0.47 ± 0.02^{d} (27.69%)	0.37 ± 0.02^{d} (50.67%)	0.32 ± 0.02^{d} (60.0%)	0.27 ± 0.01^{d} (67.07%)	0.21 ± 0.01^{d} (75.29%)

Table (2): Analgesic effect of ginger extracts (either alcoholic or watery) at 150 mg/ kg b.wt by hot plate test in mice. (Mean ± SE) (n=5).

Parameters / Groups	Reaction time (seconds)							
	0 time	30min	60min	90min	120min	150min	180min	210min
G1 (Control given 0.2ml normal saline)	8.30 ± 0.25[a]	8.60 ± 0.14[d]	8.48 ± 0.22[d]	9.40 ± 0.24[d]	9.60 ± 0.14[d]	9.76 ± 0.12[d]	9.86 ± 0.09[d]	9.64 ± 0.07[d]
G2 (Indomethacin treated group with10mg/kg b.wt)	8.10 ± 0.29[a]	10.96 ± 0.20[c]	12.04 ± 0.16[c]	12.56 ± 0.16[c]	13.30 ± 0.07[c]	13.60 ± 0.14[c]	13.48 ± 0.15[c]	13.34 ± 0.11[c]
G3 (Ginger water extract treated group with 150mg/kg b.wt)	7.86 ± 0.29[a]	11.68 ± 0.10[b]	13.36 ± 0.16[b]	13.82 ± 0.13[b]	13.90 ± 0.07[b]	14.22 ± 0.09[b]	14.58 ± 0.04[b]	14.20 ± 0.09[b]
G4 (Ginger alcoholic extract treated group with150mg/kg b.wt)	7.66 ± 0.15[a]	12.66 ± 0.11[a]	14.04 ± 0.16[a]	14.62 ± 0.20[a]	15.52 ± 0.07[a]	15.34 ± 0.12[a]	15.86 ± 0.05[a]	15.18 ± 0.10[a]

Table (3): Antipyretic effect of ginger extracts (either alcoholic or watery) at 150 mg/ kg b.wt on feverish rats. (Mean ± SE) (n=4).

Parameter / Groups	Temperature(^{0}C)							
	BBT	0.0hr (after 18hr)	0.5hr	1.0hr	1.5hr	2.0hr	2.5hr	3.0hr
G1 (Control feverish given 0.2ml normal saline)	35.03 ± 0.11[a]	37.05 ± 0.06[a]	37.23 ± 0.09[a]	38.13 ± 0.08[a]	38.35 ± 0.06[a]	38.45 ± 0.06[a]	38.55 ± 0.06[a]	38.65 ± 0.06[a]
G2 (Feverish treated with indomethacin at 10mg/kg b.wt)	35.01 ± 0.08[a]	37.01 ± 0.08[a]	36.90 ± 0.11[b]	36.80 ± 0.11[b]	36.68 ± 0.09[b]	36.58 ± 0.09[b]	36.00 ± 0.11[b]	35.80 ± 0.11[b]
G3 (Feverish treated with water extract of ginger at 150mg/kg b.wt)	34.83 ± 0.09[a]	36.83 ± 0.09[a]	36.55 ± 0.06[c]	36.38 ± 0.11[c]	36.28 ± 0.11[c]	36.20 ± 0.11[c]	35.65 ± 0.06[c]	35.35 ± 0.06[c]
G4 (Feverish treated with alcoholic extract of ginger at 150mg/kg b.wt)	34.78 ± 0.08[a]	36.80 ± 0.12[a]	35.95 ± 0.10[d]	35.88 ± 0.09[d]	35.78 ± 0.09[d]	35.70 ± 0.09[d]	35.08 ± 0.09[d]	34.90 ± 0.07[d]

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الملخص العربى

أنشطة الزنجبيل كمضاد للالتهاب ومسكن للالم وخافض للحرارة

ا.د/ مجدى صلاح عامر ، إيمان صالح العشرى

قسم الأدوية – كلية الطب البيطرى- جامعة المنصورة

استهدفت هذه الدراسة استبيان مدى فعالية الزنجبيل كمضاد للالتهاب ومسكن للالم (أجريت التجربة على عدد ٤٠فأر) ومخفض للحرارة(أستخدم عدد ١٦جرز ، وقد تم تقسيم الجرذان والفئران إلي مجاميع لأجراء التجارب على النحو التالى :

المجموعة الأولى: مجموعة ضابطة حقنت بمحلول فسيولوجى بجرعة (٠,٢ مل / جرذ) فى الغشاء البروتينى .

المجموعة الثانية : مصابة ومعالجة بالإندوميثاسين (١٠ مجم / كجم) فى الغشاء البروتينى

المجموعة الثالثة والرابعة : مصابة ومعالجة بالمستخلص المائي والكحولى للزنجبيل(١٥٠مجم / كجم) لكلا منهما على التوالى فى الغشاء البروتينى .

وقد اسفرت النتائج مايلى:

بالنسبة لتأثير الزنجبيل كمضاد للالتهاب: أظهرت النتائج أن هناك انخفاض تدريجي فى حجم المخلب للجرذان مع الوقت، وكان الانخفاض الناجم عن مستخلص الزنجبيل (الكحولى والمائى) أكثر بشكل ملحوظ مقارنا مع المجموعة الضابطة والمجموعة المعالجة بالإندوميثاسين. وكانت النسبة المئوية لتثبيط الورم لمجموعات الزنجبيل أكثر من تلك التي لوحظت للإندوميثاسين فى الساعة الاولى والثانية والثالثة والرابعة والخامسة على التوالى.

أما بالنسبة لاستخدام الزنجبيل كمسكن للالم: أظهرت النتائج التي تم الحصول عليها أن هناك زيادة في وقت رد فعل الحيوان إلى الحرارة (٣٠-٢١٠) دقيقة بعد العلاج بالإندوميتاسين والزنجبيل ، مقارنة مع المجموعة الضابطة. وقد لوحظ ان الزنجبيل له أعلى وقت رد فعل بالمقارنة مع الإندوميثاسين.

وبالنسبة لتأثير الزنجبيل كخافض للحرارة: أوضحت الدراسة أن هناك انخفاض تدريجي في درجة حرارة الفئران التى عولجت بمستخلصات الزنجبيل والإندوميثاسين (١٨٠) دقيقة بعد العلاج. وكان الانخفاض الناجم عن الزنجبيل أقل بالمقارنة من تلك التي تم الحصول عليه عن طريق الإندوميثاسين.

نستنتج من هذا أن مستخلصات الزنجبيل (الكحولية والمائية) لها تاثير كمسكنات للالم، وكمخفضات للحرارة ومضادات للالتهاب.