

ORIGINAL CONTRIBUTION

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Anti-inflammatory, analgesic, antipyretic and antidiabetic activities of *Abutilon hirtum* (Lam.) Sweet

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Abstract

Background: Many plants of genus *Abutilon* are traditionally used for treatment of inflammation, bronchitis, piles, gonorrhoea, diabetes and fever. *Abutilon hirtum* is traditionally used to ease the pain of kidney gravel, to treat diarrhoea, cough and toothache, to cure bladder inflammations, wounds and ulcers and as an antipyretic, demulcent, diuretic and mouth wash. The aim of the study is to evaluate the anti-inflammatory, analgesic, antipyretic and antidiabetic effects of the total ethanolic extract and different fractions of *Abutilon hirtum* (Lam.) Sweet leaves.

Methods: Air dried powder of *A. hirtum* leaves were extracted using 95% ethanol and fractionated successively with petroleum ether, chloroform and finally with ethyl acetate. The extracts were concentrated to afford petroleum ether, chloroform, ethyl acetate and aqueous fractions and were investigated for their anti-inflammatory, analgesic, antipyretic and antidiabetic activities using carrageenan-induced paw edema, hot plate, yeast -induced pyrexia and streptozotocin-induced hyperglycemia methods, respectively.

Results: The total ethanolic extract and the chloroform fraction exhibited the highest anti-inflammatory activity with a percentage of inhibition 50.8% which is close to that of indomethacin (52.4%). The aqueous extract exhibited the maximum analgesic activity (216.6%) with a rapid onset and a longer duration followed by petroleum ether and chloroform fractions and total extract (189.8, 186.9 and 183.0%, respectively), which is almost similar to that of acetylsalicylic acid (186.4%).

The total ethanolic extract showed higher activity compared to the used standard acetylsalicylic acid with a rapid onset (30 min) and a longer duration exhibiting the maximum activity. The crude polysaccharides fraction showed a significant lowering in blood glucose level (81.08%). The total extract and petroleum ether, chloroform, ethyl acetate and aqueous fractions exhibited a significant anti-diabetic activity after 5 h (47.49, 47.79, 50.04, 49.80 and 46.36%, respectively) compared with that of metformin (55.45%).

Conclusion: *Abutilon hirtum* extract and fractions exhibited anti-inflammatory analgesic, antipyretic and antipyretic activities which may be attributed by the presence of active phytoconstituents.

Keywords: Malvaceae, *Abutilon hirtum*, Anti-inflammatory, Analgesic, Antipyretic, Antidiabetic

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Background

Abutilon is a large genus belonging to family Malvaceae comprises about 150 annual or perennial herbs, shrubs or even small trees native to the tropical and subtropical countries of America, Africa, Asia and Australia [1, 2]. Traditionally, many *Abutilon* species are used in treatment of inflammation, piles, gonorrhoea, bronchitis, diarrhea, cleaning wounds and ulcers [3]. In Malaysia, *Abutilon hirtum* is used as a poultice to ease the pain of kidney gravel and often mixed with glutinous rice and applied to ulcers. In Thailand, the roots are used against cough and toothache and as an antipyretic. The leaves or flowers are applied to abscesses. In Kenya the fruits are eaten raw, while the leaves are browsed by goats and camels. Water extract of the bark is given to ease childbirth in Kenya and Uganda. In India, traditionally the leaves are used as demulcent, diuretic and to treat diarrhoea. The decoction of the leaves is used as mouth wash and to cure bladder inflammations, wounds and ulcers, since alkaloids are reported from the roots of the plant [2, 4, 5].

A. hirtum possess hepatoprotective, antioxidant and cytotoxic activities [4, 6, 7], in addition to *A. indicum* and *A. mauritianum* are reported to exhibited anti-inflammatory, analgesic, antipyretic activities [8–12]. Previous phytochemical investigation of *A. hirtum* led to isolation of flavonoids and phenolic acids [13]. For this reasons the current study evaluate some pharmacological activities of total ethanolic extract and different fractions of *A. hirtum* (Lam.) Sweet leaves including anti-inflammatory, analgesic, antipyretic and antidiabetic effects.

Methods

Plant material

The leaves of *A. hirtum* were collected in November 2012 from El-Zohria botanical garden, Cairo, Egypt and identified by Prof. Dr. Mahmoud Abdelhady Hassan Professor of Horticulture, Faculty of Agriculture, Minia University. A voucher sample (Mn-ph-Cog-016) was kept in the Herbarium of Pharmacognosy Department, Faculty of Pharmacy, Minia University, Minia, Egypt.

Preparation of the extract and fractions

The air dried powdered leaves (5 Kg) of *A. hirtum* were extracted with 95% ethanol and concentrated under reduced pressure. The concentrated ethanolic extract (550 g) was suspended in the least amount of distilled water, transferred to a separating funnel and partitioned successively with petroleum ether, chloroform and finally with ethyl acetate. The fractions were concentrated under reduced pressure to afford petroleum ether (150 g), chloroform (8 g) and ethyl acetate fractions (18 g). The

remaining mother liquor was concentrated to give the aqueous fraction (280 g).

Preparation of the crude polysaccharides

The concentrated aqueous fraction (280 g) was dissolved in the least amount of distilled water, transferred to a conical flask and polysaccharides allowed to settle by drop wise addition to 1 l of methanol, followed by vigorous shaking, then filtrated using glass Büchner funnel and vacuum pump. The residue (crude polysaccharides) was collected and dried using vacuum drying oven and then kept for further investigation.

Animals

The animals used in this study include female and male albino rats weighing 200 ± 50 g and mice weighing 30 ± 5 g, obtained from animal house of Faculty of Medicine, Assiut University. They were housed under standardized environmental conditions, and fed with standard diet and water. The study was conducted following approval by the Institutional Animal Ethical Committee of Faculty of Pharmacy, Minia University, Minia, Egypt.

Acute toxicity

The acute toxicity of the total ethanolic extract of *Abutilon hirtum* leaves was determined by measuring the lethal dose for 50% of the laboratory animals (LD_{50}) [14]. Different dose levels (1, 2, 2.5, 3 up to 3.5 g/ kg, p.o) of the total ethanolic extract (suspended in 0.5% CMC) were orally administrated to different groups of mice (30 ± 5 g, each containing six mice). The control group received an equivalent dose of the vehicle (0.5% CMC). Both the test and control groups were observed for 24 h under normal environmental conditions, with free access to food and water.

Anti-inflammatory activity

The total ethanolic extract and different fractions of *A. hirtum* leaves were evaluated for their anti-inflammatory activity using the carrageenan-induced paw edema method [15]. Female albino rats (200 ± 50 g) were randomly divided into seven groups (six animals per group). The specified dose of extract, fractions, and standard drug were suspended in 0.5% CMC solution. The control group administered the vehicle (0.5% CMC solution), while the standard group was given indomethacin orally at a dose level of 8 mg/kg. The total ethanolic extract and different fractions were administrated orally at a dose level of 300 mg/kg through 2 h after carrageenan injection (0.1 ml, 1% w/v in normal saline, s.c.) into the sub-plantar tissue of the right hind paw. The paw thickness (mm) was measured using a vernier caliper at 0, 0.5, 1, 2, 3, 4 and 5 h after administration of the tested extract, fractions and standard drug. The percentage

Table 1 Results of the anti-inflammatory activity of the total ethanolic extract and different fractions of *A. hirtum* leave using carrageenan – induced paw edema method

Group	Thickness of the paw (mm)						
	0 h	30 min	1 h	2 h	3 h	4 h	5 h
Control (–ve)	7.37 ± 0.12	7.37 ± 0.12	7.5 ± 0.0	7.62 ± 0.12	7.62 ± 0.12	7.87 ± 0.23	7.87 ± 0.23
Total extract (300 mg/kg)	7.25 ± 0.14	6.87 ± 0.12	6.25 ± 0.14***	5.62 ± 0.23***	4.62 ± 0.23***	4.0 ± 0.20***	3.87 ± 0.12***
Petroleum ether fraction (300 mg/kg)	7.25 ± 0.14	7.12 ± 0.12	6.75 ± 0.14*	6.37 ± 0.23**	5.75 ± 0.32***	5.25 ± 0.25***	4.87 ± 0.12***
Chloroform fraction (300 mg/kg)	7.25 ± 0.14	6.62 ± 0.23*	6.37 ± 0.23***	5.62 ± 0.23***	4.87 ± 0.31***	4.0 ± 0.2***	3.87 ± 0.12***
Ethyl acetate fraction (300 mg/kg)	7.25 ± 0.14	7.0 ± 0.0	6.75 ± 0.14*	6.5 ± 0.2**	5.87 ± 0.12***	5.37 ± 0.37***	5.37 ± 0.37***
Aqueous fraction (300 mg/kg)	7.12 ± 0.12	7.12 ± 0.12	6.75 ± 0.14*	6.5 ± 0.2**	6.0 ± 0.20***	4.87 ± 0.12***	4.75 ± 0.14***
Indomethacin (8 mg/kg)	7.25 ± 0.14	6.5 ± 0.2**	6.0 ± 0.20***	5.62 ± 0.23***	5.37 ± 0.23***	4.12 ± 0.12***	3.75 ± 0.14***

The results are presented as the mean ± S.E.M Standard error of the mean. Differences with respect to the control group were calculated using the student's T-test (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$)

of inhibition of the rat paw edema was calculated as follows [16]:

$$\% \text{Inhibition} = \frac{(ET_c - ET_t)}{ET_c} \times 100$$

ET_c is Paw edema thickness of control group and ET_t is Paw edema thickness of treated group.

Analgesic activity

The total ethanolic extract and different fractions of *A. hirtum* leaves were evaluated for their analgesic activity using hot plate method [17]. Mice (30 ± 5 g) were grouped into seven groups (six animals each). The control group administered the vehicle (0.5% CMC solution) and the standard group administered 100 mg/kg, p.o. of acetylsalicylic acid. The tested extract and different fractions were suspended in 0.5% CMC solution and were administrated orally at a dose level of 300 mg/kg. The animals were placed on a hot plate and the temperature of the metal surface was maintained at 54 °C. The time (s) of the response produced by the animal as tail withdrawn, licking paws or jumping due to radiant heat is noted and recorded at 0, 0.5, 1, 2, 3, 4 and 5 h after the administration of the tested extract,

fractions and the standard drug. The percentage of thermal pain stimulus protection was calculated according to the following formula [18]:

$$\begin{aligned} \% \text{Protection against thermal stimulus} \\ = \frac{(T_t - T_c)}{T_c} \times 100 \end{aligned}$$

Where T_t is the reaction time of treated group and T_c is the reaction time of control group.

Antipyretic activity

The total ethanolic extract and different fractions of *A. hirtum* leaves were tested for their antipyretic activity using yeast-induced pyrexia method [19, 20]. The test was performed on female albino rats (200 ± 50 g) by subcutaneous injection (in the back, below the nape of the neck) of 20% aqueous suspension of yeast in a dose of 10 ml/kg to induce pyrexia. The pyretic animals were grouped into seven groups (six animals each). The control group orally administered the vehicle (0.5% CMC solution), while the reference group was given acetylsalicylic acid at a dose level of 100 mg/kg, p.o. The tested extract and different fractions were suspended in 0.5% CMC solution and were administrated orally at a

Table 2 Results representing the percentage of edema inhibition of the total ethanolic extract and different fractions of *A. hirtum* leaves

Group	% Edema Inhibition						
	0 h	30 min	1 h	2 h	3 h	4 h	5 h
Total extract (300 mg/kg)	1.62	6.78	16.66	26.24	24.54	33.29	50.82
Petroleum ether fraction (300 mg/kg)	1.62	3.39	10.0	16.40	36.08	49.17	38.11
Chloroform fraction (300 mg/kg)	1.62	10.17	15.06	26.24	22.96	31.76	50.82
Ethyl acetate fraction (300 mg/kg)	1.62	5.02	10.0	14.69	21.25	38.11	31.76
Aqueous fraction (300 mg/kg)	3.39	3.39	10.0	14.69	29.52	47.64	39.64
Acetylsalicylic acid (100 mg/kg)	1.62	11.80	20.0	26.24	39.37	49.17	52.35

Table 3 Results of the analgesic activity of the total ethanolic extract and different fractions of *A. hirtum* leaves in mice using the hot plate method

Group	Time (sec)						
	0 h	30 min	1 h	2 h	3 h	4 h	5 h
Control (–ve)	58 ± 2.51	60.67 ± 0.88	62 ± 2.30	61.33 ± 1.85	59.33 ± 2.02	59.5 ± 1.55	59 ± 1.47
Total extract (300 mg/kg)	60.25 ± 1.88	57.5 ± 1.32	94.25 ± 4.42***	94 ± 5***	152.7 ± 3.28***	159.8 ± 4.19***	167 ± 3.34***
Petroleum ether fraction (300 mg/kg)	59 ± 2.48	60.75 ± 1.93	72.25 ± 3.98	97 ± 1.68***	138.3 ± 7.51***	167.3 ± 2.72***	171 ± 1***
Chloroform fraction (300 mg/kg)	64.33 ± 1.76	68.67 ± 3.84	75.5 ± 5.31	94.33 ± 4.66***	112 ± 4.20***	156.7 ± 7.83***	169.3 ± 5.72***
Ethyl acetate fraction (300 mg/kg)	52.25 ± 3.19	55 ± 0.57	57.25 ± 1.75	57.75 ± 3.49	63.5 ± 2.53	59.25 ± 1.93	58.75 ± 2.95
Aqueous fraction (300 mg/kg)	62.5 ± 2.32	75.33 ± 2.84**	86 ± 4.91**	94.75 ± 2.72***	108 ± 6.24***	155.3 ± 3.70***	186.8 ± 5.51***
Acetylsalicylic acid (100 mg/kg)	56.25 ± 1.70	67.5 ± 3.61	71.5 ± 3.66	79 ± 2.48**	93.67 ± 4.37***	131.7 ± 5.20***	169 ± 7.37***

The results are presented as the mean ± S.E.M Standard error of the mean. Differences with respect to the control group were calculated using the student's T-test (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$)

dose level of 300 mg/kg through 2 h after yeast injection. The rectal temperature of each animal was recorded by inserting a thermometer 2 cm into the rectum at 0, 0.5, 1, 2, 3, 4 and 5 h after administration of the tested extract, fractions and the reference drug.

Anti-diabetic activity

The antidiabetic activity of the total ethanolic extract and different fractions of *A. hirtum* leaves was evaluated using streptozotocin-induced hyperglycemia method [21, 22]. The test was performed on adult male albino rats (200 ± 50 g) by intraperitoneal injection of streptozotocin (80 mg/kg). Blood glucose level was measured after 3 days up to one week for assessment of hyperglycemia. Rats with blood glucose level above (200 mg/dl) were considered to be diabetic and were used in this study. The diabetic rats were divided into eight groups (six rats each). The control group was administered the vehicle (0.5% CMC solution), while the standard group was given of 150 mg/kg p.o metformin. The tested extract and different fractions were suspended in 0.5% CMC solution and were orally administrated at a dose level of 300 mg/kg. Blood glucose levels were measured at intervals of 0 (fasting), 0.5, 1, 2, 3, 4 and 5 h by collecting blood samples from the tail vein (caudal

vein). The percentage of change in blood glucose level was calculated by the following formula [23, 24]:

$$\% \text{lowering blood glucose level} = \frac{(W_c - W_t)}{W_c}$$

Where W_t is the blood glucose concentrations of treated group and W_c is the blood glucose concentrations of control group.

Statistical analysis

Results of all biological studies were expressed as means ± S.E.M. One-way analysis of variance (ANOVA) followed by Dunnett's test was used to determine significance when compared to the control group. p values less than 0.05, 0.01, and 0.001 were considered significant (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$). Graph Pad Prism 5 was used for statistical calculations (Graph pad Software, San Diego California, USA).

Results

Acute Toxicity

There was no mortality or signs of toxicity up to 3 g/kg of the total ethanolic extract of *A. hirtum* leaves.

Table 4 Results representing the percentage of protection against thermal stimulus of the total ethanolic extract and different fractions of *A. hirtum* leaves

Group	% Protection						
	0 h	30 min	1 h	2 h	3 h	4 h	5 h
Total extract (300 mg/kg)	3.87	–5.22	52.01	53.26	157.37	168.57	183.05
Petroleum ether fraction (300 mg/kg)	1.72	0.13	16.53	58.16	133.10	181.17	189.83
Chloroform fraction (300 mg/kg)	10.91	13.18	21.77	53.80	88.77	163.36	186.94
Ethyl acetate fraction (300 mg/kg)	–9.91	–9.34	–7.66	–5.83	7.02	–0.42	–0.42
Aqueous fraction (300 mg/kg)	7.75	24.16	38.70	54.49	82.03	161.00	216.61
Acetylsalicylic acid (100 mg/kg)	–3.10	11.25	15.32	28.81	57.87	121.34	186.44

Table 5 Results of the antipyretic activity of the total ethanolic extract and different fractions of *A. hirtum* leaves on yeast-induced pyrexia method

Group	Rectal temperature (°C)						
	0 h	30 min	1 h	2 h	3 h	4 h	5 h
Control (-ve)	38.73 ± 0.08	38.85 ± 0.02	38.85 ± 0.05	38.88 ± 0.02	39.55 ± 0.13	39.9 ± 0.04	39.98 ± 0.08***
Total extract (300 mg/kg)	38.63 ± 0.10	38.2 ± 0.07***	37.3 ± 0.09***	36.35 ± 0.06 ***	36.23 ± 0.04***	36.13 ± 0.04***	36.05 ± 0.02***
Petroleum ether fraction (300 mg/kg)	38.68 ± 0.08	38.23 ± 0.12***	38.15 ± 0.02***	37.33 ± 0.10***	37.4 ± 0.17***	37.13 ± 0.07***	37.1 ± 0.04***
Chloroform fraction (300 mg/kg)	38.7 ± 0.09	38.73 ± 0.04	38.63 ± 0.04	38.5 ± 0.04***	38.43 ± 0.04***	38.48 ± 0.04***	38.5 ± 0.04***
Ethyl acetate fraction (300 mg/kg)	38.75 ± 0.09	38.5 ± 0.04*	38.2 ± 0.07***	37.63 ± 0.06***	37.55 ± 0.06***	37.2 ± 0.04***	37.05 ± 0.02***
Aqueous fraction (300 mg/kg)	38.73 ± 0.06	38.43 ± 0.08**	38 ± 0.13***	37.48 ± 0.02***	37.43 ± 0.04***	37.25 ± 0.11***	37.13 ± 0.07***
Acetylsalicylic acid (100 mg/kg)	38.78 ± 0.07	38.48 ± 0.06**	37.7 ± 0.16***	37.43 ± 0.04***	37.2 ± 0.04***	37.2 ± 0.07***	37.15 ± 0.02***

The results are presented as the mean ± S.E.M Standard error of the mean. Differences with respect to the control group were calculated using the student's T-test (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$)

Anti-inflammatory activity

The tested fractions and total extract exhibited a significant decrease in the paw edema induced by carrageenan especially after 2 h from the beginning of the experiment (Table 1 and 2 and Additional file 1). After 2 h, both the total ethanolic extract and the chloroform fraction exhibited the same percentage of inhibition (26.2%) as that of the indomethacin, whereas the following hours showed results quite higher than the standard. After 5 h both the total ethanolic extract and the chloroform fraction exhibited the highest anti-inflammatory activity with a percentage of inhibition 50.8% which is close to that of indomethacin (52.4%). The petroleum ether and the aqueous fractions showed a mild inhibition of the inflammation that was increased by time to be 38.1 and 39.6% after 5 h, respectively. On the other hand, the least activity was shown by the ethyl acetate fraction through the 5 h of the experiment (31.8%).

Analgesic activity

The total ethanolic extract and different fractions of *A. hirtum* leaves (Table 3 and 4 and Additional file 2)

demonstrated that the total ethanolic extract and all fractions exhibited potent analgesic activity except the ethyl acetate fraction. One hour from the beginning of the experiment both the total ethanolic extract and aqueous fraction showed the highest percentage of protection against thermal stimulus (52.0 and 38.7%, respectively) in comparison with acetylsalicylic acid (15.3%). Furthermore, after 2 h the petroleum ether and chloroform fractions exhibited effects higher than acetylsalicylic acid. The aqueous extract exhibited the maximum analgesic activity (216.6%) with a rapid onset and a longer duration followed by petroleum ether and chloroform fractions and total extract (189.8, 186.9 and 183.0%, respectively), which is almost similar to that of acetylsalicylic acid (186.4%). On the other hand, the ethyl acetate fraction showed no analgesic activity throughout the experiment time.

Antipyretic activity

The total ethanolic extract and different fractions of *A. hirtum* leaves revealed that the total ethanolic extract and most of fractions exhibited a significant ($P < 0.001$) antipyretic activity up to 5 h (Table 5 and

Table 6 Results of anti-diabetic activity of the total ethanolic extract and different fractions of *A. hirtum* leaves on streptozotocin-induced hyperglycemia

Group	Blood glucose level (mg/dl)						
	0 h (fasting)	30 min	1 h	2 h	3 h	4 h	5 h
Control (-ve)	203.3 ± 2.32	201.8 ± 1.79	201.3 ± 1.49	202.8 ± 1.93	207.3 ± 1.93	208.5 ± 1.70***	208.8 ± 2.78***
Total extract (300 mg/kg)	202.3 ± 1.79	201.8 ± 1.25	184 ± 2.64	150 ± 2.08***	145.5 ± 2.75***	123.3 ± 3.32***	109 ± 4.54***
Petroleum ether fraction (300 mg/kg)	206.3 ± 1.70	199 ± 5.05	183.5 ± 4.34	158.5 ± 4.69***	142.5 ± 3.5***	129.3 ± 4.90***	109 ± 5.35***
Chloroform fraction (300 mg/kg)	206.8 ± 2.32	195.8 ± 3.63	174.8 ± 2.72**	149 ± 4.69***	141.5 ± 3.40***	124.5 ± 4.57***	104.3 ± 3.03***
Ethyl acetate fraction (300 mg/kg)	205 ± 2.85	202.8 ± 2.62	161.3 ± 5.48***	150.5 ± 5.12***	142 ± 3.87***	113.5 ± 3.86***	104.8 ± 2.01***
Aqueous fraction (300 mg/kg)	205.3 ± 3.56	204.3 ± 3.63	181.3 ± 5.51*	160 ± 4.93***	153.5 ± 2.63***	138.5 ± 3.5***	112 ± 5.5***
Crude polysaccharides (300 mg/kg)	207.3 ± 3.77	199.8 ± 1.25	172.7 ± 6.83**	96.3 ± 6.36***	87.25 ± 6.30***	72.25 ± 5.28***	39.5 ± 2.5***
Metformin (150 mg/kg)	204 ± 2.64	199 ± 1.35	166.5 ± 6.70***	137.8 ± 3.19***	126.5 ± 2.21***	113 ± 4.54***	93 ± 4.08***

The results are presented as the mean ± S.E.M Differences with respect to the control group were calculated using the student's T- test (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$)

Table 7 Results of the effect of the total ethanolic extract and different fractions of *A. hirtum* leaves on the percentage of lowering blood glucose level on diabetic rats

Group	Dose mg/kg	Percentage of lowering blood glucose level (%)						
		0 h	30 min	1 h	2 h	3 h	4 h	5 h
Total extract	300	0.34	0.0	0.0	26.03	29.81	40.86	47.79
Petroleum ether fraction	300	-1.62	1.38	0.27	21.84	31.25	37.98	47.79
Chloroform fraction	300	-1.87	2.97	5.0	26.52	31.74	40.28	50.04
Ethyl acetate fraction	300	-0.98	-0.49	12.33	25.78	31.50	45.56	49.80
Aqueous fraction	300	-1.13	-1.23	1.46	21.10	25.95	33.57	46.36
Crude polysaccharides	300	-2.11	0.99	6.14	52.51	57.91	65.34	81.08
Metformin	150	-0.49	1.38	9.51	32.05	38.97	45.80	55.45

Additional file 3). The total ethanolic extract showed higher activity compared to the used standard acetylsalicylic acid with a rapid onset (30 min) and a longer duration exhibiting the maximum activity. After 4 h from the beginning of the experiment the petroleum ether, ethyl acetate and aqueous fractions showed the same effect as that of acetylsalicylic acid that lasted up to 5 h. The least activity was shown by the chloroform fraction where no significant effect was observed throughout the 5 h of the experiment.

Anti-diabetic activity

The total ethanolic extract and all fractions showed a significant ($***P < 0.001$) decrease in blood glucose level (Table 6 and 7 and Additional file 4) especially after 2 h from oral administration and their effects were maintained for 5 h. The crude polysaccharides fraction showed a significant lowering in blood glucose level (52.51%) after 2 h which increased by time after 3 h (57.91%), 4 h (65.34%) and 5 h (81.08%). The blood glucose levels of the mice were reduced significantly when administered total extract and petroleum ether, chloroform, ethyl acetate and aqueous after 5 h, where the percent of lowering blood glucose levels were 47.49, 47.79, 50.04, 49.80 and 46.36%, respectively, compared with that of metformin (55.45%).

Discussion

LD₅₀ of the total ethanolic extract is 3 g/Kg indicating its wide margin of safety. Hence, the chosen experimental dose of the total extract and different fractions is 300 mg/kg which is one-tenth of the lethal dose (3 g/kg) [25]. The total ethanolic extract and the chloroform fraction exhibited the highest anti-inflammatory activity compared to indomethacin and the potent antipyretic activity of the total ethanolic extract may be attributed to their content of sterols and flavonoids which were reported to have anti-inflammatory and antipyretic activities [12, 26, 27]. Also, many mechanisms were proved for the anti-inflammatory

activity of flavonoids as inhibition of cyclooxygenase and 5-lipoxygenase pathways, inhibition of eicosanoid biosynthesis, besides its ability to inhibit neutrophil degranulation [28]. Furthermore, the significant of the total ethanolic extract and aqueous fraction may be attributed to their high content of flavonoids and flavonoidal glycosides, in addition to sterols which were reported to have analgesic activity [27]. The total ethanolic extract and different fractions exhibited a potent anti-diabetic effect and the crude polysaccharides was found to have the highest antidiabetic activity from the first hour till the third hour followed by a severe hypoglycemia after 4 h. This hypoglycemic effect may be attributed to a decrease in blood glucose level or inhibition of glucose absorption [29], an increase in the insulin secretion level, reduced insulin metabolism, through remediating destruction of pancreatic islets and damage of pancreatic β - cells [30, 31].

Conclusion

The present study revealed that total ethanolic extract and different fractions of *A. hirtum* leaves exhibited anti-inflammatory, analgesic, antipyretic and antidiabetic activities which are in accordance with folk medicine of many *Abutilon* plants, in addition to the wide margin of safety of the total ethanolic extract which may assist to develop new drugs from natural source.

Additional files

Additional file 1: Diagram representing the effects of the total ethanolic extract and different fractions of *A. hirtum* leaves on paw thickness using carrageenan – induced paw edema. (DOCX 177 kb)

Additional file 2: Diagram representing the analgesic activity of the total ethanolic extract and different fractions of *A. hirtum* leaves. (DOCX 157 kb)

Additional file 3: Diagram representing the effects of the total ethanolic extract and different fractions of *A. hirtum* leaves on yeast-induced pyrexia. (DOCX 172 kb)

Additional file 4: Diagram representing the effects of the total ethanolic extract and different fractions of *A. hirtum* leaves on streptozotocin-induced hyperglycemia in rats. (DOCX 173 kb)

Abbreviations

A. hirtum: *Abutilon hirtum*; *A. indicum*: *Abutilon indicum*; *A. mauritianum*: *Abutilon mauritianum*; CMC: Carboxy methyl cellulose; LD₅₀: Lethal dose 50; p.o.: Taken orally; s.c.: Subcutaneous

Authors' contributions

AAG performed all of the experiments in the laboratory and data collection, analysis, graphical representation and interpretation. MNS did critical statistical analysis and wrote the manuscript draft. Critical revision of the article was done by SYD. Conception, experiment design, overall monitoring and final approval of the article was done by MSK. All authors read and approved the final manuscript.

Competing interests

The authors declare that they have no competing interests.

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