



Effect of different level of *Zingiber officinale* and *Salvia officinalis* on semen diluents for Awassi ram

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Abstract

This work was performed to evaluate the effects of hot water extracts of two medicinal plants *Zingiber officinale* and *Salvia officinalis* on Awassi ram sperm activity. Three different concentrations of the two plants were used (0.001, 0.005, and 0.1 mg) and the activity of the sperm were evaluated at the time: 0, 24, 48, 72 hours of experiment. Results showed significant decreased in sperm activity associated with the 0.01mg of *Salvia officinalis* in compare with the other concentrations of the same plant and the second plant. The effect of time also revealed progressive decrease in sperm activity with the time in both plants especially (significantly) in the 48 and 72 hours of experiment. In conclusion, the extract of the two plants decreased sperm activity in proportion with the progression of time and concentration.

Key words: *Zingiber officinale*, *Salvia officinalis*, Water extracts, Awassi ram, Sperm activity.

Introduction

About 15% of couples do not achieve pregnancy within 1 year and seek medical treatment for infertility. Eventually 5% remain unwillingly childless. Infertility affects both men and women. In 50% of involuntarily childless couples a male infertility associated factor is found together with abnormal semen parameters. A fertile partner may compensate for the fertility problem of the men and thus infertility usually becomes manifest if both partners have reduced fertility (Dohle *et al.*, 2010). Many medicinal plants were used for the enhancement of male fertility (Sakr and Badawy, 2011; Kakel and Ahmed, 2008; Santos-Filho *et al.*, 2007; Mbongue *et al.*, 2005).

Medicinal plants are increasingly recognized worldwide as an alternative source of efficacious and inexpensive medications to synthetic chemotherapeutic compound, and high proportion of the world's population rely on plants for their primary Health care (Putheti and Okigbo, 2008; Omogbadegun *et al.*, 2011). The most important active substances in these plants are alkaloids, tannins, terpenoids, glycosides, phenolics, saponins, flavonoids, quinines, lectins and polype-

ptides, and many others (Cowan, 1999; Okigbo and Igwe, 2007).

Ginger *Zingiber officinale* L. belong to the family Zingiberaceae is widely used as a digestive aid for mild stomach upset and is commonly recommended by health care professionals to help prevent or treat nausea and vomiting associated with motion sickness, pregnancy (Chittumma, 2007; Nanthakomon and Pongroj paw, 2006). Also ginger has effective antioxidant and anticancer activity (Cai *et al.*, 2003) and known to significantly increase sperm percentage, viability, motility and serum total testosterone (Khaki *et al.*, 2009). The important active components of the ginger are thought to be volatile oils and pungent phenol compounds such as gingerols, shogaols, zingerone, and gingerols, zingiberene, turmerone, methyl chavicol, and γ -terpinene (Norajit *et al.*, 2007; Sekiwa *et al.*, 2000). *Salvia officinalis* L. (Lamiaceae) is a perennial woody sub-shrub native to the Mediterranean area, used in the food-processing industry but also in the area of human health. It is well known for its fungistatic, virustatic and tannin-based antimicrobial properties. Anti-inflammatory activities were reported to be caus-

ed by some constituents of plants such as triterpenes, oleanolic and ursolic acids, or the diterpene carnosol (Baricevic *et al.* 2001). In Wistar rats there are several phytoconstituents like triterpenoids, tannins, saponins, alkaloids and flavonoids that known to promote wound healing process due to their antioxidant and antimicrobial activities (Estakhr and Javdan, 2011), also *Salvia* species have been used to improve male reproductive functions in traditional medicine (Estakhr and Javdan, 2011). Another species was found to affect liver function, administration caused significant reduction in alkaline phosphatase (ALP) and total bilirubin comparing to control group, while aspartate aminotransferase (AST), alanine aminotransferase (ALT) and total protein were non-significantly changed indicating that *Salvia* administration support the function of liver (Tohamy *et al.*, 2012).

Materials and Methods

Hot water Extract Preparation: The Powder of *Zingiber officinale* and *Salvia officinalis* were mixed with hot water after cooling water to temperature less 50 C. The mixing was done by using a vibrator for 15 minutes. Then the mixture was left for 24 hours. The filtration process was done by using four layers of gauze. Then the leaky was put in a dishes which is heated to 40 C inside an oven for drying the extract. The dry raw solid material for extract then waited and the required concentrations were prepared.

Semen treatments: Semen was collected from one of the rams Awassi 3 years old and weight 70 kg then diluted the semen by Tris- egg yolk diluent (1:10) .then distributed randomly into three groups first one control seconded group contain *Zingiber officinale* extract and the third group contain *Salvia officinalis* extract on three concentration (0.001, 0.005, and 0.1 mg) each.

Refrigerated the samples gradually to 5 Celsius and placed in glass flask contain water in 37 Celsius then keeping the samples in refrigerator. The activity of the sperm were evaluated at the time: 0, 24, 48, 72 hours of experiment. Depending on individual motility of the sperm (0-100) (Walton, 1933).

Statistical analysis: was conducted by using general learner model for SAS (SAS, 2004) and the comparative between means was conducted by using Duncan's Multiple Range Test (Duncan, 1955).

Results and Discussion

The effects of the different concentrations of both plants were shown in the table 1, which revealed significant decrease in sperm activity with the increase of concentrations. The effect of time on sperm activity also showed significant decrease in sperm activity with the progression of time as in the table 2, while table 3 represent the comparison between the effects of the two plants on sperm activity.

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Our results showed undesired decrease in sperm activity associated with the treatment by *Zingiber officinale* and *Salvia officinalis* extracts and these are not the only plants that showed similar effects on male fertility (Priya *et al.*, 2012; Revathi *et al.*, 2010), also these results are contrary to others that showed significant enhancement in male fertility (Sakr and Badawy , 2011; Santos-Filho *et al.*, 2007; Mbongue *et al.*, 2005). These effects may be related to the direct effects on spermatogenesis like the results of Kakei and Ahmed, 2008 , that revealed a significant decrease in the weight of testes, body of epididymis, percentage of live/dead sperms and epididymal sperm count, with a significant increase in the percentage of morphologically abnormal sperms. Another study explained the antifertility of some plants may related to immunoinfertility and the formation of antisperm antibodies (ASA), which can be a major factor of male fertility (Gupta and Sharma, 2006).

Table (1): The effects of different concentrations of *Zingiber officinale* and *Salvia officinalis* on sperm activity.

Plant	Concentration	Sperm activity
<i>Zingiber officinale</i>	0.0	75.0 A
<i>Salvia officinalis</i>	0.0	75.0 A
<i>Zingiber officinale</i>	0.001	75.0 A
<i>Salvia officinalis</i>	0.001	70.0 A
<i>Zingiber officinale</i>	0.005	58.75 A
<i>Salvia officinalis</i>	0.005	58.75 A
<i>Zingiber officinale</i>	0.01	18.75 B
<i>Salvia officinalis</i>	0.01	75.0 A

Means on columns with different superscripts are significantly different ($p > 0.05$).

Table (2): The effects of the time of treatment on sperm activity.

Plant	time	Sperm activity
<i>Zingiber officinale</i>	0.0	81.250 AB
<i>Salvia officinalis</i>	0.0	88.750 A
<i>Zingiber officinale</i>	24	67.500 BC
<i>Salvia officinalis</i>	24	81.250 AB
<i>Zingiber officinale</i>	48	42.5 DE
<i>Salvia officinalis</i>	48	56.25 DC
<i>Zingiber officinale</i>	72	36.250 E
<i>Salvia officinalis</i>	72	36.250 E

Means on columns with different superscripts are significantly different ($p > 0.05$).

Table (3): Comparison between the effects of *Zingiber officinale* and *Salvia officinalis* on sperm activity.

Plant	Concentration	Time			
		0.0	24	48	72
<i>Zingiber officinale</i>	0.0	95 A	85 C	70 F	50 I
	0.001	90 B	80 D	65 G	45 J
	0.005	80 D	75 E	50 I	30 L
	0.01	90 B	85 C	40 K	20 M
<i>Salvia officinalis</i>	0.0	95 A	85 C	70 F	50 I
	0.001	90 B	85 C	65 G	60 H
	0.005	90 B	85 C	30 L	30 L
	0.01	50 I	15 N	5 O	5 O

Means on columns with different superscripts are significantly different ($p > 0.05$).

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