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SHORT COMMUNICATION

Fertility Suppression of the Male Mouse after Administration of Mint Leaf Extract

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The antifertility effect of an aqueous extract of mint (*Mentha arvensis*) leaves was observed on male albino mice after administration of 10, 5 and 2 mg of the extract every alternate day for 20 days. At the tried experimental protocol, a dose related decrease in the reproductive organ weights was recorded. A marked alteration in the phosphatase content in the reproductive organs was also obtained. While the acid phosphatase mean level recorded a marked decrease, the alkaline phosphatase content of the reproductive organs exhibited a statistically significant increase. Mating performance and libido of the males treated with the mint leaf extract at all the three tried doses was not altered but mating with fertile proestrus females proved to be sterile in each case.

Keywords: antifertility effect; mint extract; male; phosphatases.

INTRODUCTION

The development of fertility regulating drugs from medicinal plants is an attractive proposition for a number of reasons. There is a large volume of information that indicates that plants allegedly have an effect on virtually all steps in the reproductive process. Recently Garg and Jacob (1994) have reported that the aqueous leaf extract of mint (*Mentha arvensis*; Fam. Lamiaceae) effectively interrupts pregnancy in the mouse if administered postcoitally. It was therefore considered worthwhile to evaluate to the effect of mint leaves on the reproductive system, fertility and behaviour of male mice.

MATERIALS AND METHODS

Fresh leaves of mint were dried and powdered, and an aqueous extract was made by Soxhlet extraction. The extract was concentrated and dried under vacuum to constant weight. Colony-bred healthy adult Swiss albino mice kept under controlled conditions of temperature and illumination and provided with a standard pellet diet and water *ad libitum* were used.

Males of proven fertility were selected for experimentation and were randomly divided into the following four groups.

Control group: (i) Vehicle (glass distilled water: GDW) treated group.

Experimental groups (dose/mouse/alternate day): (ii) 10 mg, (iii) 5 mg and (iv) 2 mg. The dose was injected intramuscularly in 0.1 mL GDW for 20 days; the controls received the vehicle alone.

At the termination of the experimental schedule five mice from each group were randomly selected and caged individually with a parous proestrus female. Mating behaviour and mating success was observed. The presence of spermatozoa in the vaginal smear or a vaginal plug was taken as an indicator of positive mating and this was considered to be the first day of pregnancy. Laparotomy was done on day 15 *postcoitum* and the number of implantation sites, if any, were recorded.

The remaining mice from each experimental group were autopsied 24 h after the last injection. Their reproductive organs were dissected out, cleared of adherent tissues and weighed to the nearest mg and then processed for quantitative examination of phosphatases (Fiske and Subarrow, 1925). The enzyme activity was expressed in Bodansky units where each unit corresponds to the liberation of 1 mg of inorganic phosphate per 1 g of tissue during 1 h period of incubation ($mg^{Pi}/g/h$).

RESULTS AND DISCUSSION

At all three tried doses and experimental regimen the sexual behaviour and libido of the experimental males remained unimpaired even after 20 days of treatment with the mint leaf extract. Autopsy of females successfully mated with experimental males revealed that none of them became pregnant, whereas in comparison all the females mated with normal males became pregnant and had an average of 10.2 implantation sites.

The aqueous extract of mint leaves caused a statistically significant dose-related progressive decrease in the mean percent weight of the reproductive organ in relation to the control values (Table 1), excepting that the alteration in the epididymal weight at a dose of 2 mg was not statistically significant and at the same dose regimen the testicular weight exhibited a significant increase in relation to the control values. Similarly, the acid and alkaline phosphatase contents of the testis and accessory reproductive organs of mice treated with the mint leaf extract were also appreciably altered (Table 2). In general, while the acid phosphatase content of the genital organs significantly decreased with increasing dosage of the mint leaf extract there was a

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Table 1. Alteration in the genital organ weight of the male mouse after administration of aqueous extract of mint leaf for 20 days. (Mean values expressed in mg/100 g body weigh±SE; five mice in each group)

Dose	Testis	Epididymis Caput	Cauda	Seminal Vesicle	Prostate
Control	696.44±7.73	130.87±6.94	87.02±2.06	519.75±28.29	199.35±8.07
Treatment (Dose/alternate day/mouse)					
10 mg	545.39±42.93[b]	144.84±8.42[b]	66.39±1.79[c]	239.25±12.36[c]	75.86±7.13[b]
5 mg	635.67±29.75[a]	115.92±5.75[c]	80.65±0.95[b]	280.55±4.75[c]	128.59±2.66[c]
2 mg	727.74±24.51[a]	117.84±3.04[b]	83.67±3.14[a]	290.60±9.82[c]	139.78±10.78[c]

Significant in relation to control: [a] $p<0.05$, [b] $p<0.1$, [c] $p<0.001$.

Table 2. Effect on phosphatase content of the reproductive organs of the male mouse after administration of aqueous extract of mint for 20 days. (Mean values expressed in Bodansky unit±SE; five mice in each group)

Dose	Testis	Epididymis Caput	Cauda	Seminal vesicle	Prostate
Acid phosphatase					
Control	3.42±0.04	2.47±0.05	2.15±0.06	1.21±0.05	1.79±0.08
Treatment (Dose/alternate day/mouse)					
10 mg	3.17±0.06[b]	1.16±0.18[c]	1.22±0.19[b]	0.82±0.14[b]	1.40±0.06[c]
5 mg	3.22±0.02[c]	1.78±0.19[b]	1.29±0.26[a]	1.02±0.09[a]	1.59±0.06[b]
2 mg	3.25±0.11[a]	2.06±0.29[a]	1.42±0.15[b]	1.29±0.09[a]	1.62±0.09[b]
Alkaline phosphatase					
Control	1.98±0.06	5.85±0.39	3.15±0.39	3.89±0.03	3.99±0.26
Treatment (Dose/alternate day/mouse)					
10 mg	4.28±0.22[b]	7.70±0.63[a]	3.41±0.74[a]	5.94±0.31[b]	6.45±0.08[c]
5 mg	3.69±0.14[c]	8.39±0.25[b]	5.79±0.19[b]	4.42±0.18[a]	6.12±0.33[c]
2 mg	3.09±0.26[b]	9.05±0.28[c]	7.75±0.17[c]	4.03±0.03[b]	5.92±0.58[b]

Significance in relation to control: [a] $p<0.05$, [b] $p<0.1$, [c] $p<0.001$.

general statistically significant dose-related increase in the alkaline phosphatase content of the reproductive tissues in relation to the control mean value.

Mint leaf extract has been reported to possess inherent oestrogenicity (Bodhankar *et al.*, 1971; Kanjanapothi *et al.*, 1981; Garg and Jacob, 1994), and in the present study on the male mouse, it also essentially simulates other oestrogenic substances in inhibiting nidation in normal female mice (Wu *et al.*, 1973). In intact mammals exogenous oestrogens reduce the level of circulating androgen and cause functional atrophy of the reproductive organs (Jacob *et al.*, 1991, 1992; Terner and McLaughlin, 1973). The inhibition of nidation in females could be directly related to the induced functional sterility of the substance treated males. On the other hand, according to Ericsson and Baker (1966), it has been surmised that treatment of mammalian male with an oestrogenic substance leads to an accumulation of oestrogen in the semen, which following coitus is sufficient to alter the events essential for conception.

The acid and alkaline phosphatases are androgen-dependent parameters and have been associated with active transport of metabolites, the process of cell growth and differentiation, spermatogenesis and steroidogenesis in the mammalian testis (Mann, 1964; Males and Turkington, 1971). Alteration in the content of these enzymes in the reproductive tissues is possibly indicative of impairment of the functional integrity of the reproductive organs, resulting in disturbances in the spermatogenic process. The present study, therefore, indicates that sterile matings were possibly due to the functional impairment of the male reproductive system as evidenced by a marked reduction in organ weights and alteration in the phosphatase levels of the reproductive structures rather than transfer of seminal oestrogen.

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