

## Effect of Palm Pit Powder on Nicotine Induced Weight Loss in Albino Mice

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### ABSTRACT

**Background and Objective:** Cigarette smoke contains nicotine that has numerous health hazards ranging from mild to life threatening morbidities. The health benefits of date palm against the toxic substances are well documented in the literature. This study is designed to investigate the effect of nicotine on body weight of mice and its amelioration by date palm pit powder (DPP).

**Methods:** Thirty two adult male albino mice were divided into four groups each having eight animals. Date's pit were first soaked for 48 hours in cold distilled water and then dried by air convection drying method. DPP was administered by oral gavage. Control group 'A' was given saline 1.5 ml per kg for 15 days. The experimental groups were given 0.5 mg/kg nicotine intra-peritoneally for 15 days. For the next 30 days, Group C received 0.9% saline orally and Group D received 500 mg/kg of DPP orally. Solid food intake was measured daily and then calculated as per week intake.

**Results:** The reduction in mean animal weight and food intake of experimental groups on 15<sup>th</sup> day were statistically significant ( $P = 0.001$ ) in comparison with that of the control. The mean animal weight and food intake significantly improved after 30 days in experimental groups D.

**Conclusion:** Nicotine administration to the albino mice significantly reduces their body weight and food intake which are gradually improved by the use of date palm pit powder.

**KEYWORDS:** Nicotine, Body weight, Date palm pit powder, Mice.

### INTRODUCTION

Tobacco use is the foremost avoidable health risk factor in the developed countries. Five million deaths are reported annually because of smoking across the globe. The death toll may rise to one billion during the 21<sup>st</sup> century, mostly affecting the developing countries.<sup>1</sup>

Nicotine is an alkaloid that comprises 1.5 to 4.6% of flue-cured tobacco leaves.<sup>2</sup> According to the protocol set by World Health Organization (WHO), nicotine content must not exceed 0.4mg per gm of cigarette tobacco filler.<sup>3</sup> It affects diverse cellular processes including hormone release, inflection of enzymatic process leading to oxidative stress trauma; diminution of glutathione concentration and suppression of O<sub>2</sub> free radical eliminators.<sup>4,5</sup> It was investigated that nicotinic drugs are anorexogenic primarily through nicotine acetylcholine receptor-dependent activation of hypothalamic pro-opiomelanocortin neurons.<sup>6</sup>

Phoenix *dactylifera* (date palm) is a highly valuable tree farmed in the arid and semi-arid countries of the world.<sup>7</sup> In Arab countries, according to local medicinal practices it is a common belief that taking date fruit alone before any meal can nullify the effects of any noxious substance.<sup>8</sup> It contains 5.0 –

6.3% protein, 7.1 – 10.3% moisture, 65 – 69% neutral detergent fiber, 9.9 – 13.5% fat and 1.0 – 1.8% ash in each of the three date palm pit powder (DPP) acquired from three different sources.<sup>9</sup> Date palm pit exclusively contains selenium related amino acids that play a prophylactic role in cancer and activates various immune mechanisms.<sup>10</sup> The current study is designed to demonstrate the anorexic and cachexic effects of nicotine in albino mice and its amelioration by DPP.

### METHODS

The experimental work was conducted in the department of Anatomy at University of Health Sciences (UHS), Lahore. This study was approved from the Ethical Review Committee with the letter no. UHS/ERC/07/2013. Thirty-two albino mice of age 6-8 weeks of male gender weighing in the range of 30-40 gm were bought from National Institute of Health, Islamabad. Animals were housed at room temperature, and controlled humidity of 50 ± 5% and light & dark cycles of 12 hours. Each of the groups comprised of eight animals allotted by randomization. Group 'A', served as a control and was given 0.9% saline 1.5 ml/kg, intraperitoneally for 15 days. The experimental

groups B, C & D were given 0.5 mg/kg nicotine intra-peritoneally, once daily dissolved in 1.5 ml/kg volume of saline for 15 days.<sup>11</sup> Thereafter, 0.9% saline (1.5 ml/kg) was given orally for next 30 days to the animals of group C and 500 mg/kg DPP orally for next 30 days to the animals of group D. Groups A and B were continued with the same treatment as that of during first 15 days of experiment. Solid food intake was measured daily by weighing the left over feed. For data analysis, weekly food intake was calculated. Liquid nicotine having concentration of 1.01 g/ml was obtained from Merck pharmaceuticals. Nicotine was administered intra-peritoneally in the right lower quadrant of abdomen. Dates (Ajwa) pits were collected by removing its flesh manually; afterwards these were dried and grinded. Fine powder of date palm pit was used daily at a dose of 500 mg/kg.<sup>11</sup>

**STATISTICAL ANALYSIS**

Statistical Package for Social Sciences (SPSS) version 25 was used to enter and analyze the data. Means and standard deviations were computed for the mean body weight. The differences of mean body weight among the groups were determined by applying one-way ANOVA and Post-hoc Tukey’s test. A repeated measure ANOVA was applied for analyzing the differences of food intake within the groups. A *P-value* less than or equaling 0.05 was taken as statistically significant.

**RESULTS**

The mean animal weight in grams on 15<sup>th</sup> day of experiment was 37.38 ± 1.69, 24.88 ± 3.44, 26.13 ± 2.53 and 26.25 ± 1.39 in groups A, B, C and D respectively. These differences were statistically significant when compared with one-way ANOVA (p = 0.001). The mean animal weight on 45<sup>th</sup> day of experiment was 39.29 ± 2.03, 20.76 ± 1.89, 29.35 ± 1.76 and 36.94 ± 2.17 in groups A, B, C and D respectively. These differences were again statistically significant when compared with one-way ANOVA as given in (Table-1). Post-hoc Tukey test on day 15 showed that reduction of weight in groups B, C and D was statistically significant when compared to group A. On day 45, Post-hoc Tukey test revealed that weight was significantly reduced in group B and C when compared to control group A, group D when compared to group B and group D when compared with group C. There were no significant weight differences between groups A and D at day 45 (Table-1).

Average weekly food intake was analyzed by repeated measure ANOVA (RM-ANOVA) within the groups. Groups B and D showed statistically significant difference of food intake during the course of experiment. There was no significant change in weekly food intake in groups A and C as well as in B and C as shown in (Table-2).

**Table -1:** Multiple comparisons of animal weight (gm) on 15<sup>th</sup> and 45<sup>th</sup> day among the groups.

Multiple Comparisons of Animal Weight on 15 <sup>th</sup> Day				Multiple Comparisons of Animal Weight on 45 <sup>th</sup> Day			
(I) Group	(J) Group	Mean Difference (I-J)	P-value	(I) Group	(J) Group	Mean Difference (I-J)	P-value
A	B	12.500*	0.001*	A	B	18.530	0.002*
	C	11.250*	0.001*		C	-9.9400	0.001*
	D	11.125*	0.001*		D	-2.3500	0.102
B	C	-1.250	0.796	B	C	8.5900	0.934
	D	-1.375	0.734		D	16.18	0.001*
C	D	-0.125	1.000	C	D	7.590	0.001*

\*Post-hoc Tukey test

**Table- 2:** Multiple comparisons of Average food intake (gm) in control and experimental groups

Groups	Week 1 n = 8	Week 2 n = 8	Week 3 n = 8	Week 4 n = 8	Week 5 n = 8	Week 6 n = 8	Week 7 n = 8	P-value (RM-ANOVA)
A	18.74±1.42	18.62±1.68	19.25±1.79	19.6±1.87	19.55±2.01	20.33±1.76	20.57±1.89	0.332
B	18.62±1.68	14.24±1.54	14.71±1.76	14.8±1.92	15.56±1.39	15.71±1.98	15.22±1.72	0.001*
C	17.63±1.37	14.78±1.68	16.7±1.45	18.92±1.46	20.99±1.95	21.53±1.81	21.61±2.61	0.160
D	17.51±1.08	13.45±1.69	14.18±0.92	15.74±1.16	18.07±1.11	21.10±0.69	22.88±1.37	0.001*

**DISCUSSION**

All mice of control group remained healthy and alive. However, animals in rest of the groups showed varying degrees of agitation, hypophagia and lethargy. Withdrawal of nicotine in experimental groups C and D enhanced their food intake, reduced their irritability and abnormal behaviors like agitation, hypophagia and lethargy.

The statistical data of present study showed that nicotine usage reduced mice body weight in all experimental groups. This study is in agreement with the previous reports by Mangubat et al.<sup>12</sup> which reveals that antagonizing nicotinic acetylcholine receptors of brain stem by mecamylamine (a nonselective antagonist) would promote food intake. The mean body weight of mice injected by nicotine (Groups B, C

& D) on 15<sup>th</sup> day was significantly lower than that of control group A. Gbadamosi et al.<sup>13</sup> and Stojakovic et al.<sup>14</sup> reported the same finding of weight loss in response to the decreased food intake. DPP significantly ameliorated the weight loss among group D animals. Metwaly et al.<sup>15</sup> supported the current study results by documenting that aqueous extract of date fruit halted the weight reduction in mice infected by *Eimeria papillata*. Reversal of weight loss was also observed in group C animals after withdrawal of nicotine.

This work illustrated significant reduction in food intake among the nicotine injected groups (B, C & D) during initial two weeks. The food intake improved for next 30 days after the cessation of nicotine in groups C and D. The mechanisms of smoking related weight loss are multifaceted and partly understood. It is unanimously agreed that this weight loss is mediated by nicotine present in the cigarettes. Daily caloric intake and the energy consumption establish the body weight. Resting metabolic rate, thermic effects of food and physical exertion are the three energy consumption determining factors and nicotine lift up the resting metabolic rate leading to weight loss.<sup>16</sup>

Martinez de Morentin et al.<sup>17</sup> investigated the anorexigenic capacity of nicotine and revealed for the first time that it inactivates adenosine monophosphate activated protein kinase in hypothalamus which leads to food intake suppression likely via the sympathetic innervations. Another study documented about nicotine restrained appetite via the activation of satiety related melanocortin-4 receptors present in hypothalamus.<sup>18</sup>

Analysis of date pits of United Arab Emirates origin suggested that the DPP can be added in food as a cheap source of starch fibers which will enhance gut efficiency (digestion & absorption) leading to better health.<sup>9</sup> Tareen et al.<sup>19</sup> concluded that the feed containing 3-4% of date palm kernel given to broilers significantly suppressed their food intake and improved water intake and the body weight.

## CONCLUSION

Nicotine administration to the albino mice reduces body weight by suppressing food intake. This weight loss is ameliorated by the use of DPP. The hazardous health effects of cigarette can be attenuated by the simultaneous use of DPP.

## LIMITATIONS OF STUDY

Indirect estimation of food intake instead of direct (actual) measurement is the main limitation of this research work. However, this study may be supplemented with possible future larger scale research in order to strengthen the conclusions drawn about study topic under discussion.

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## AUTHOR'S CONTRIBUTION

**KS:** Conception and design of study.

**AN:** Analysis and interpretation of data.

**AKM:** Acquisition of data.

**AT:** Final approval of the version to be published.

**AJ:** Drafting the article.

**Iz:** Final approval of the version to be published.

## CONFLICT OF INTEREST

None to declare.

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