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***Egyptian Desert Truffles, Androgenic Natural Product,
Possibility of Abusing in Sport.***

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Introduction

Egyptian desert truffles growing naturally in Egypt are used in folklore medicine as aphrodisiac, androgenic, in the treatment of ophthalmic diseases and as nutritive supplement⁽¹⁾. Nothing of the current literature studied the aphrodisiac effect of desert truffles.

The present study was carried out on two species of Egyptian desert truffles (white, *Tirmania nivea* and brown, *Terfezia claveryi*), including study of their nutritive values, steroidal matter (including pheromones) and their effect on the genital system of male mature rats. Their anabolic effect and possibility of abusing in sport will be discussed.

Samples of Egyptian desert truffles (collected from El-Salloum) were purchased from the local market in February 2000. The ascocarps were freed from soil, some samples were kept fresh for the study of steroidal content, others were dried till constant weight at 50⁰C in an oven and powdered for analysis.

Experimental

1) Study of the nutritive value

Study of the nutritive value included amino acids analysis ⁽²⁾, determination of the In-Vitro-Protein-Digestibility (IVPD) ⁽³⁾, fatty acids analysis ^(4,5), determination of soluble and hydrolysable carbohydrates ⁽⁶⁾, determination of mineral elements other than phosphorus ⁽⁷⁾ determination of phosphorus ⁽⁸⁾, determination of vitamin C ⁽⁹⁾, vitamin B₁ and B₂ ⁽¹⁰⁾ and fat soluble vitamins (A, E and D₃) ⁽¹¹⁾. The results are tabulated in tables 1 and 2.

2) Study of the steroidal content

For isolation of fungi steroids from the unsaponifiable matter, preparative TLC was used using hexane: ethyl acetate (9: 1) as developing system. Brassicasterol/Ergosterol ratio was determined by TLC densitometry at 366-400nm by comparison of the corresponding spots areas at each specified wavelength. Pheromone content was studied by TLC screening of the methylene chloride: ethyl acetate extracts as described by Claus et al. (1981)⁽¹²⁾ and the chromatoplates were examined with UV light and sprayed with *p*-anisaldehyde-sulphuric acid for location of spots.

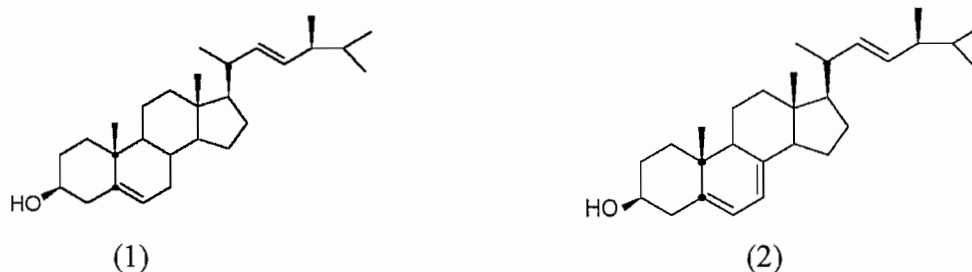
3) Effect on genital system of male mature rats

The alcohol soluble constituents of both species were injected intraperitoneally in doses of 250 and 500 mg/kg body weight of experimental animals (30 animals/group) for six successive days during which the control group was injected with 15% tween 80. Tissue specimens of testes, epididymis, seminal vesicles and prostate glands were taken weekly for six successive weeks for histopathological examination, starting from the last day of injection (3 animals/group/week).

Results and discussion

Amino acids analysis (table 2) revealed the presence of nine essential and eight non-essential amino acids, with glutamic acid as the major and cystine as the minor amino acid in both species. IVPD (table 2) was higher for white truffles than for brown truffles. Fatty acids analysis (table 2), reported for the first time for Egyptian desert truffles, showed that palmitic acid was the major fatty acid in both species while lauric and margaric acids were the minor fatty acids in white and brown truffles respectively. From table 1, the major identified soluble sugar was galacturonic acid in both species, while the minor soluble sugar was arabinose in white truffles and an oligosaccharide in brown truffles. Galactose and trehalose were detected only in brown truffles. On the other hand, glucose was the major sugar in the hydrolysable carbohydrates for both species, followed by glycerol. The mineral composition (table 2), reported for the first time for Egyptian desert truffles, showed that phosphorus and sodium were the major and minor macroelement in both species. Among microelements, copper showed the highest level and Co the lowest in both species. The high percentage of phosphorus may explain the preference of Egyptian desert truffles as aphrodisiac by truffles consumers. Egyptian desert truffles were also found to be a good source of vitamins (table 1), reported for the first time. The high vitamin E content may also play a role in the biological activity.

2 mg of pure brassicasterol (1) and 0.8 mg pure ergosterol (2) were isolated by preparative TLC. The isolated sterols were identified by their physical properties and mass spectral data.



The average Brassicasterol/Ergosterol ratio was found to be 1.8 by TLC densitometry. TLC screening of the methylene chloride: ethyl acetate 1:1 extracts revealed the absence of detectable concentrations of pheromones in both species.

Pharmacological study revealed that morphologically, there was a remarkable increase in the size of the testes of the treated animals at the last day of injection. Histopathologically, the treated rats showed normal structure and marked vasodilatation of blood vessels in genital organs, regardless to the dose, the species of truffles or the week of taking the sample. This vasodilatory effect of truffles in genital organs can explain their aphrodisiac effect, also, no biosynthetic relationship could be found between truffles sterols and testosterone. According to our results, the Egyptian truffles can be considered as a non doping nutritive supplement.

Table 1: Soluble and hydrolysable carbohydrates (as % of dry weight) and vitamins (mg/100g dry weight)

Sugar	White truffle	Brown Truffle	Vitamin	White truffle	Brown Truffle
Oligosaccharide^s	1.519	1.449	Vitamin C	255.827	60.527
Galacturonic acid^s	8.475	8.842	Vitamin B₁	0.310	0.293
Sucrose^s + Maltose^s	8.024	15.054	Vitamin B₂	0.546	0.480
Arabinose^s	0.517	3.065	Vitamin A	3.10x10 ⁻³	1.00x10 ⁻³
Glycerol^s	6.592	8.095	Vitamin E	82.094	932.766
Mannitol^s	1.581	3.001	Vitamin D₃	116.9x10 ⁻³	-
Galactose^s + Trehalose^s	-	6.427			
Glucose^h	34.838	46.676			
Arabinose^h	9.200	12.219			
Glycerol^h	30.227	32.890			

^s soluble carbohydrates

^h hydrolysable carbohydrates

Table 2: Amino acids (as % of dry weight), fatty acids (relative %) and minerals [mg/100g for all elements except P (%) and Se (ppm)]

Amino Acid	White truffle	Brown truffle	Fatty acid	White truffle	Brown truffle	Mineral element	White truffle	Brown truffle
Aspartic	1.57	1.82	Lauric	1.03	2.14	K ^b	1368.700	745.120
Threonine ^a	1.11	1.00	Myristic	3.50	10.06	Na ^b	0.992	1.178
Serine	0.84	0.80	Pentadecylic	20.73	5.46	Mg ^b	44.400	48.690
Glutamic	2.06	3.82	Palmitic	50.17	54.31	Ca ^b	280.614	381.770
Proline	0.86	0.86	Margaric	2.27	0.84	P ^b	2.810	3.530
Glycine	0.83	0.82	Stearic	2.10	1.61	Fe ^c	5.629	7.640
Alanine	0.89	1.20	Oleic	9.19	-	Mn ^c	0.345	0.407
Cystine ^a	0.31	0.21	Linoleic	-	19.25	Cu ^c	255.870	168.490
Valine ^a	0.7	0.78	Linolenic	1.45	-	Zn ^c	4.031	4.452
Methionine	0.11	0.11	Arachidic	3.31	0.97	Co ^c	0.152	0.292
Isoleucine ^a	0.54	0.58	Henecosanoic	2.59	3.55	Se ^c	0.032	0.049
Leucine	0.87	0.98	Behenic	3.66	1.79			
Tyrosine	0.41	0.40						
Phenylalanine) ^a	0.61	0.65						
Histidine ^a	0.32	0.47						
Lysine ^a	0.50	0.69						
Arginine ^a	0.66	0.76						
IVPD	72	69						

^a essential amino acids

^b macroelements

^c microelements

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