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Biochemical effect of low metabolic calories diets on experimentally induced obesity on rats

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A B S T R A C T

This study aimed to evaluate the effect of weight loss diets in experimental obesity. Forty –five male albino rats, 6-8 weeks old, were randomly assigned into four groups: Group 1 (Control normal group) : Comprised 10 rats, provided only with regular diet and plenty of fresh. Group 2 (Control obese group with 75 calories diet) : Included 10 rats ,was received a high fat diet for 4 weeks period then each rat was fed daily with 75 calories diet for 8weeks .Group 3 (Obese group with 60 calories diet): Included 10 rats ,was received a high fat diet for 4 weeks period then each rat was fed daily with 80% calories of the Control obese diet for 8weeks (60 calories diet) .Group 4 (Obese group with 45 calories diet) : Included 10 rats ,was received a high fat diet for 4 weeks period then each rat was fed daily with 60% calories of the Control obese diet for 8weeks (45 calories diet).Blood samples were collected after 8 and 12weeks of supplementation for biochemical analysis. As compared to the control normal group, control obese group exhibited a significant decrease in serum high density lipoprotein-cholesterol (HDL-c), transferrin and total iron bending capacity(TIBC), along with increase in aspartate aminotransferase (AST) activity, total protein, albumin, urea, creatinine, total cholesterol ,plasma triacylglycerol, very low density lipoprotein-cholesterol (VLDL-c), low density lipoprotein-cholesterol (LDL-c),sodium ,calcium , iron concentration and Plasma ferritin concentration. Meanwhile , the obese group provided with 60 calories diet exhibited a significant decrease in AST activity, total protein, albumin , urea, creatinine, total cholesterol , triacylglycerol ,VLDL-c, LDL-c, sodium ,calcium , iron concentration and ferritin along with increase in HDL-c, transferrin and TIBC when compared with obese group. However, the obese group provided with 45 calories diet, exhibited a significant decrease in total protein concentration, albumin, T. cholesterol, triacylglycerol, VLDL-c, LDL-c, sodium, calcium, iron concentration and ferritin along with increase in AST activity, urea, creatinine, HDL, transferrin and TIBC, when compared with obese group. In conclusion, Low calorie diet reduces calorie intake without incurring malnutrition or a reduction in essential nutrients compared with very low-calorie diet.

Key words: BMI, Iron profiles, Kidney function test, Lipid metabolism, Obesity

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1. INTRODUCTION

Obesity is defined as an excess of body fat (increased fat cell size and number) relative to lean body mass Clinically, obesity is defined on the basis of Body Mass Index (BMI). Any person with BMI between 25 and 29.9 kg/m2 is termed overweight and any individual with BMI 30 kg/m2 or more is classified as obese, severe or class III obesity is defined as BMI 40 kg/m2 or more with or without significant comorbidity. This term is also used for individuals with a BMI between 30 and 39.9 kg/m2 who have significant comorbidities. (Li *et al.*, 2005).

Obesity is a major risk factor for a number of diseases, it has direct effects on glucose and lipid metabolism and on blood pressure, leading via these to type 2 diabetes, coronary heart disease and cerebrovascular disease, these comorbidities are particularly associated with excess intra-abdominal fat, other important diseases which obesity increases the risks of include obstructive sleep apnea, osteoarthritis of weight-bearing joints and a number of types of cancer. In addition to these medical comorbidities obesity has many adverse social and psychological costs (Nightingale *et al.*, 2010).

A combination of excessive food energy intake and a lack of physical activity are thought to explain most cases of obesity. A limited number of cases are due primarily to genetics, medical reasons, or psychiatric illness (Bleich *et al*., 2008). The common methods for assessment of obesity are BMI, waist circumference (WC), waist-to-hip ratio (WHR), and skin fold thickness (Han et al.,2006).A healthy diet is that helps to maintain and improve overall health. It provides the body with essential nutrition: fluid, macronutrients, micronutrients, and adequate calories. (Mitchell and Haroun, 2012). The standard diets which promote weight loss can be categorized as: low-fat, low-carbohydrate, low-calorie, very low calorie and more recently flexible dieting (Strychar, 2006).

The aim of this study is to evaluate the biochemical effect of weight loss diets in experimental obesity.

2. MATERIALS AND METHODS

2.1.Experimental animals: Forty (40) male albino rats, 6-8 weeks old, with average body weight 150-180 g were used in this study. Rats were housed in separated metal cages and fed on constant ration and water was supplied *ad-libitum*. The animals were left 14 days for acclimatization before the beginning of the experiment.

2.2.Materials:

Wheat, Crushed beans, crushed sorghum and Mash 21% protein.

2.3.Experimental design:

Rats were randomly assigned into four groups, placed in individual cages and classified as follow:

Group 1 (Control normal group): Comprised 10 rats, provided only with regular diet and plenty of fresh, clean drinking water *ad-libitum.*

Carbohydrates	58 %	Protein	20.5 %
Lipid	3.4 %	Cellulose	3.1 %
Vitamins	2.3%	Minerals	2.7 %
Moisture	10%		
Total	100%		

Group 2 (Control obese group with 75 calories diet): Included 10 rats ,was received a high fat diet for 4 weeks period then each rat was fed daily with 75 calories diet for 8weeks (Yamaguchi *et al.*, 2012) ,(25g provender 21% protein or 20.5 g crushed sorghum or 21.5 g crushed beans or 22 g wheat).

Group 3 (Obese group with 60 calories diet): Included 10 rats ,was received a high fat diet for 4 weeks period then each rat was fed daily with 80% calories of the Control obese diet for 8weeks (60 calories diet) (Chen,2015), (20g provender 21% protein or 16.5 g

crushed sorghum or 17 g crushed beans or 17.5 g wheat).

Group 4 (Obese group with 45 calories diet): Included 10 rats ,was received a high fat diet for 4 weeks period then each rat was fed daily with 60% calories of the Control obese diet for 8weeks (45 calories diet), (Chen, 2015), (15 g provender 21% protein or 12g crushed sorghum or 13 g crushed beans or 13 g wheat).

Meals were calculated for groups According to food composition table for Egypt, Second edition, 2006.

Food composition table	
Crushed sorghum	366 kcal /100g
Crushed beans	346 kcal /100g
Wheat	344 kcal /100g
Provender 21% protein	3000 kcal /1000g

2.4.Sampling:

Blood samples were collected after 8 and 12 weeks of supplementation for biochemical analysis in dry, clean, screw capped serum separating tubes then serum were separated by centrifugation at 2500 RPM for 15 minutes and kept in deep freeze at - 20 °C until used.

2.5.Biochemical analysis:

Serum AST activity was determined according to the method described by Reitman and Frankel, 1957 ,Total protein (Kaplan *et al.*, 2000),albumin (Doumas et al., 1971), urea (Patton and Crouch, 1977), creatinine (Jaffe, 1986), cholesterol (Allain *et al.*, 1974), triacylglycerol (Fossati and Prencipe, 1982),VLDL-c, LDL-c (Wieland and Seidel,1983), HDL-c (Burnstein et al., 1995) and sodium (Maruna, 1958), calcium (Moorehead and Briggs, 1974), iron (Ceritti and Ceriotti, 1980), ferritin (Lipnisam et *al*., 1981), transferrin and TIBC (Fischl and Cohen, 1962).

2.6.Statistical analysis:

Was carried out using ANOVA with two factors under significance level of 0.05 for the whole results using SPSS (Version 22).

3. RESULTS

Data presented in table (1) revealed that supplementation of 75 calories diet to obese group of male rats daily for 12 weeks influenced significant increase in serum AST, total protein, albumin and urea when compared with normal control group. Meanwhile, the obese group provided with 60 calories diet exhibited a significant decrease in AST, total protein, and albumin and urea. Also, the obese group provided with 45 calories diet, exhibited a significant decrease in serum total protein, albumin, with increase in serum urea level as compared with control obese group.

Data presented in table (2) showed supplementation of 75 calories diet to obese group of male rats daily for 12weeksinfluenced significant increase in total cholesterol, triacylglycerol, VLDL-c with marked in HDL-c compared with normal control group. Meanwhile, the obese group provided with 60 and 45 calories diet exhibited a significant decrease in total cholesterol, triacylglycerol, VLDL-c with increase in HDL-c level compared with control obese group.

Data presented in table (3) revealed that supplementation of 75 calories diet to obese group of male rats daily for 12 weeks influenced significant increase in sodium, calcium, iron and ferritin compared with normal control group. Meanwhile, the obese group provided with 60 and 45 calories diet exhibited a significant decrease in serum sodium, calcium, iron and ferritin compared with control obese group.

Supplementation of 75 calories diet to obese group of male rats daily for 12weeks showed decrease in serum transferrin and TIBC compared with normal control group. Meanwhile, the obese group provided with 60 and 45 calories diet exhibited a significant increase in transferrin and TIBC compared with control obese group (Table 4).

4. DISCUSION

Obesity is a condition in which a person has excess body fat. More than just a number on

a scale or the size of someone's body, obesity can increase a person's risk of diseases and health problems. Serum AST activity result showed a significant increase in 75 calories diet to obese group, after 8 and 12weeks of supplementation, when compared with control normal group while exhibited a significant improvement in 60 calories diet to obese group when compared with obese group.

Table (1) The effect of low metabolic calories diets serum AST, total protein, albumin and urea after 8 and 12 weeks of supplementation.

		AST (U/L)		total protein (g/dl)		Albumin (g/dl)		Urea (mg/dl)	
Duration of treatment/week		8week	12week	8week	12week	8week	12week	8week	12week
Animal groups	Control normal	74.63 ± 3.12^{d}	79.84 ± 3.45^{c}	6.58 ± 0.22^{b}	6.77 ± 0.26^{b}	2.97 ± 0.12^{c}	3.08 ± 0.10^{c}	24.82 ± 1.32^{c}	26.61 ± 1.39^{c}
	Obese +75calories	90.54 ± 3.03^{b}	99.95 ± 3.24^{b}	7.32 ± 0.13^{a}	7.90 ± 0.22^{a}	3.49 ± 0.11^{a}	3.89 ± 0.10^{a}	29.76 ± 1.06^{b}	35.22 ± 1.35^{b}
	Obese +60 calories	81.19 ± 2.99^{c}	75.16 ± 2.63^{c}	6.95 ± 0.16^{b}	6.61 ± 0.18^{b}	3.35 ± 0.12^{b}	3.10 ± 0.10^{b}	25.39 ± 1.04^{c}	23.33 ± 1.29^{c}
	Obese +45 calories	143.67 ± 4.58^{a}	116.17 ± 6.22^{a}	5.52 ± 0.19^{c}	5.33 ± 0.18^{c}	2.99 ± 0.08^{c}	2.89 ± 0.06^{c}	58.09 ± 2.94^{a}	60.83 ± 3.19^{a}

* Data are presented as (Mean ± S.E), S.E = Standard error. Mean values with different superscript letters in the same column are significantly different at (P<0.05).

Table (2) The effect of low metabolic calories diets on serum total cholesterol, triacylglycerol, VLDL-c and HDL-c after 8 and 12 weeks of supplementation.

		Total. Cholesterol (mg/dl)		Triacyl-Glycerol (mg/dl)		VLDL(mg/dl)		HDL(mg/dl)	
Duration of treatment/week		8week	12week	8week	12week	8week	12week	8week	12week
Animal groups	Control normal	71.62 ± 2.92^{b}	75.09 ± 2.36^{b}	66.43 ± 3.12^{b}	71.84 ± 3.45^{b}	14.55 ± 0.42^{b}	16.33 ± 0.64^{b}	25.91 ± 1.75^{a}	25.04 ± 1.51^{b}
	Obese +75calories	126.97 ± 3.28^{a}	142.36 ± 3.79^{a}	176.53 ± 2.73^{a}	185.65 ± 2.69^{a}	22.37 ± 3.03^{a}	26.01 ± 0.38^{a}	13.72 ± 1.18^{d}	9.55 ± 1.43^{d}
	Obese +60 calories	56.67 ± 2.73^{c}	52.62 ± 2.14^{c}	62.45 ± 1.97^{c}	54.54 ± 2.92^{c}	12.83 ± 0.28^{c}	11.05 ± 0.53^{c}	21.86 ± 0.91^{c}	24.92 ± 1.02^{c}
	Obese +45 calories	47.40 ± 1.76^{d}	41.38 ± 1.86^{d}	50.77 ± 2.37^{d}	42.91 ± 2.02^{d}	10.30 ± 0.32^{c}	9.17 ± 0.43^{c}	23.38 ± 1.17^{b}	28.94 ± 1.35^{a}

Data are presented as (Mean ± S.E), S.E = Standard error. Mean values with different superscript letters in the same column are significantly different at (P<0.05).

Table (3) The effect of low metabolic calories diets on sodium, calcium ,iron and ferritin after 8 and 12 weeks of supplementation

		Sodium (mg/dl)		Calcium (mg/dl)		Iron (mg/dl)		Ferritin (ng/ml)	
Duration of treatment/week		8week	12week	8week	12week	8week	12week	8week	12week
Animal groups	Control normal	117.94 ± 2.84^{b}	123.68 ± 2.03^{b}	8.21 ± 0.58^{a}	8.95 ± 0.53^{a}	0.73 ± 0.07^{c}	0.85 ± 0.08^{c}	17.62 ± 0.61^{c}	18.99 ± 0.51^{c}
	Obese +75calories	154.46 ± 2.32^{a}	168.45 ± 2.16^{a}	8.97 ± 0.62^{a}	9.63 ± 0.67^{a}	2.17 ± 0.21^{a}	2.41 ± 0.45^{a}	45.71 ± 1.52^{a}	50.79 ± 1.77^{a}
	Obese +60 calories	125.27 ± 2.66^{b}	119.52 ± 2.38^{b}	8.15 ± 0.47^{b}	7.49 ± 0.41^{b}	1.82 ± 0.14^{b}	1.75 ± 0.13^{b}	23.42 ± 0.51^{b}	21.87 ± 0.48^{b}
	Obese +45 calories	79.84 ± 2.50^{c}	73.76 ± 2.34^{c}	8.04 ± 0.26^{b}	7.23 ± 0.27^{b}	1.61 ± 0.17^{b}	1.31 ± 0.18^{b}	12.52 ± 0.62^{d}	11.31 ± 0.39^{d}

Data are presented as (Mean ± S.E), S.E = Standard error. Mean values with different superscript letters in the same column are significantly different at (P<0.05).

Table (4) The effect of low metabolic calories diets on serum transferrin and TIBC after 8 and 12 weeks of supplementation

		Transferrin (mg/dl)		TIBC (mcg/dl)	
Duration of treatment/week		8week	12week	8week	12week
Animal groups	Control normal	256.12 ± 3.65[a]	249.12 ± 3.65[a]	453.96 ± 5.02[b]	462.32 ± 3.25[b]
	Obese +75calories	219.08 ± 2.58[d]	207.42 ± 2.91[d]	411.35 ± 4.21[d]	426.12 ± 4.82[d]
	Obese +60 calories	237.62 ± 2.57[c]	245.55 ± 3.62[c]	434.25 ± 6.61[c]	445.60 ± 5.81[c]
	Obese +45 calories	242.08 ± 6.22[b]	247.20 ± 3.45[b]	516.49 ± 9.61[a]	492.68 ± 8.57[a]

Data are presented as (Mean ± S.E), S.E = Standard error. Mean values with different superscript letters in the same column are significantly different at ($P<0.05$).

Moreover, additional improvement in serum AST in the obese rats administrated 45 calories diet daily for 12 weeks. Similar results were given by Nassar (2014).

Serum total protein results exhibited a significant increase in 75 calories diet group after 8 and 12weeks of supplementation, when compared with control normal group while showed a significant improvement in 60 calories diet group and showed exhibited a significant improvement in the obese rats group that administrated 45 calories diet daily for 12 weeks .Elevated serum protein level may be indicator for chronic inflammation or metabolic disorders. In accordance, Lozano et al. (2016). This result is also confirmed with Daniel et al.(2014).

Serum albumin result showed exhibited a significant increase in 75 calories diet group after 8 and 12 weeks of supplementation, when compared with control normal group while exhibited a significant improvement in 60 calories diet group. Moreover, additional improvement in serum albumin concentration in the obese rats administrated 45 calories diet daily. This result is also confirmed with Sitar et al. (2013) ,and in accordance, Lozano et al. (2016)

Serum urea result showed exhibited a significant increase in 75 calories diet group , after 8and 12 weeks of supplementation, when compared with control normal group while Supplementation of 60 calories diet to obese group of male rats daily, exhibited a significant improvement in serum urea concentration after 8 and 12 weeks of supplementation, when compared with obese group. Moreover, additional improvement in serum urea concentration in the obese rats administrated 45 calories diet daily. This result is confirmed with Khan et al. (2017) .

Serum creatinine concentration result showed exhibited a significant increase in 75 calories diet group after 8and 12 weeks of supplementation, when compared with control normal group while exhibited a significant improvement in 60 calories diet group when compared with obese group. Moreover, additional improvement in serum creatinine concentration in the obese rats administrated 45 calories diet daily. This result is conformed with Nassar (2014) .

Serum total cholesterol, triacylglycerol, LDL-c, VLDL-c concentration, result showed that supplementation of 75 calories diet to obese group of male rats daily, exhibited a significant increase in after 8and12 weeks of supplementation, when compared with control normal group while Supplementation of 60 calories diet to obese group of male rats daily, exhibited a significant improvement in serum total cholesterol, triacylglycerol, LDL-c, VLDL-c concentration after 8 and 12 weeks of supplementation, when compared with obese group. Moreover, additional improvement in serum total cholesterol, triacylglycerol, LDL-c, VLDL-c concentration in the obese rats administrated 45 calories diet. This result is in the same line with Mooradian et al. (2008).

Serum HDL-c result showed exhibited a significant increase in 75 calories diet group, after 8 and 12 weeks of supplementation, when compared with control normal group while Supplementation of 60 calories diet to obese group of male rats daily, exhibited a

significant improvement in serum HDL-c after 8 and 12 weeks of supplementation, when compared with obese group. Moreover, additional improvement in serum HDL-c in the obese rats administrated 45 calories diet daily. This result is confirmed with Chen et al.(2015).

Serum sodium result showed that supplementation of 75 calories diet to obese group of male rats daily, exhibited a significant increase in serum sodium concentration, after 8 and 12 weeks of supplementation, when compared with control normal group while supplementation of 60 calories diet to obese group of male rats daily, exhibited a significant improvement in serum sodium concentration after 8 and 12 weeks of supplementation, when compared with obese group. Moreover, additional improvement in serum sodium concentration in the obese rats administrated 45 calories diet. In accordance with our result, Vigil et al., 2015 reported a prolonged hypernatremia triggered by hyperglycemic hyperosmolar state.

Serum calcium result showed that supplementation of 60 calories diet or 45 calories diet to obese group of male rats daily, exhibited a significant reduction in serum calcium concentration after 8and 12 weeks of supplementation, when compared with obese group or with normal group. This result is accordance with Choksi et al. (2018).

Serum iron result showed that supplementation of 75 calories diet to obese group of male rats daily, exhibited a significant increase in serum iron conc, after 8and 12 weeks of supplementation, when compared with control normal group while Supplementation of 60 calories diet or 45 calories diet to obese group of male rats daily, exhibited a significant improvement in serum Iron concentration after 8and 12 weeks of supplementation, when compared with obese group. This result agreed with Yamano et al. (2015), who reported that iron excess presence causes oxidative stress.

Serum ferritin result showed a significant increase in 75 calories diet group, after 8 and 12 weeks of supplementation, when compared with control normal group while supplementation of 60 calories diet to obese group of male rats daily, exhibited a significant improvement in serum ferritin concentration after 8 and 12 weeks of supplementation, when compared with obese group. Moreover, additional improvement in serum ferritin. This result is confirmed with KO et al. (2015).

Serum transferrin concentration result showed a significant decrease in 75 calories diet group after 8and 12 weeks of supplementation, when compared with control normal group while supplementation of 60 calories diet to obese group of male rats daily, exhibited a significant improvement in serum transferrin concentration after 8 and 12 weeks of supplementation, when compared with obese group. Moreover, additional improvement in serum transferrin concentration in the obese rats administrated 45 calories diet daily .It is well known that there were negative correlation between ferritin and transferrin as demonstrated with Majoni et al.(2017).

Serum TIBC result exhibited a significant decrease in 75 calories diet group, after 8 and 12 weeks of supplementation, when compared with control normal group while supplementation of 60 calories diet to obese group of male rats daily, exhibited a significant improvement in serum TIBC concentration after 8 and 12 weeks of supplementation, when compared with obese group. Moreover, additional improvement in serum TIBC concentration in the obese rats administrated 45 calories diet daily. This result in the same line with Yamanishi et al. (2012).

6. CONCULOSIONS

Low calorie diet reduces calorie intake without incurring malnutrition or a reduction in essential nutrients, but very low-calorie diet may not be nutritionally complete and provide far fewer calories than most people need to maintain a healthy weight. It is not an easy diet to follow. Our recommendations to those who seeking for weight loss is the low-calorie balanced diet and consult with a certified clinical dietitian to avoid very low-

calorie diets which have serious health hazards.

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