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Pyruvate attenuate lipid metabolic disorders and insulin resistance in obesity induced in rats

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A B S T R A C T

This study was designed to evaluate the effect of prolonged intake of Pyruvate on insulin resistance, leptin and lipid metabolism in obesity -induced in female rats by feeding high fat diet. Fifty female albino rats were divided into five equal groups of 10 rats each. Group I :(Control negative group): rats fed normal diet. Group II: (Control positive group): rats fed high fat diet (HFD) and administered no drugs. Group III: rats fed HFD and administered pyruvate (270 mg/kg b. wt. /day, orally) for 8 weeks. Group IV: rats received HFD and administered pyruvate (540 mg/kg b. wt. /day, orally). Group V: rats received the control normal diet and administered pyruvate once daily (540 mg/kg b. wt., orally) for 8 weeks. Blood samples were collected at 2, 6, 8 weeks from the onset of pyruvate administration for determination of serum glucose, insulin, insulin resistance, leptin, total cholesterol (TC), triacylglycerol (TAG), phospholipids, Low density lipoprotein-cholesterol (LDL-c), Very low density lipoprotein-cholesterol (VLDL-c) and High density lipoprotein-cholesterol (HDL-c) levels in addition to serum transaminases enzymes (AST, ALT) and creatine kinase-MB (CK-MB) activities. The obtained results revealed that, rats fed HFD exhibited marked hyperglycemia, significant elevation of serum leptin, insulin and insulin resistance, AST, ALT and CK-MB, lipids profile (TC, TAG,LDL-C, VLDL-c) with marked decreased in serum HDL-c concentrations compared to rats fed normal diet. Meanwhile, administration of pyruvate to HFD-fed rats tended to prevent hyperglycemia, improve dyslipidemia and other changes relevant to cardiovascular disease mainly through improving leptin and insulin resistance. These results suggest that, pyruvate is effective in improving the obesity with its associated many important complications such as diabetes mellitus and coronary heart disease.

Keywords: Pyruvate; Obesity; Leptin; Insulin; Lipids profile

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1. INTRODUCTION

Obesity is a severe metabolic disorder, characterized with increases in energy intake and a decrease in energy output concerning body weight and glucose metabolism (Akiyama et al., 1996).Obesity is associated with many important complications such as diabetes mellitus and coronary heart disease.(Abu-Abid et al., 2002). Moreover, obesity is considered the largest public health problem worldwide, especially in industrialized countries (Bravo et al., 2006).

There is growing concern over the increasing numbers of overweight and obese individuals, and management of obesity has become an important component of public health calendar. Several weight-management strategies are now available and a wide variety of slimming aids usually marketed as food supplements are on offer (Joyal, 2004).

The efficacy of these food supplements has not been proven, yet they are sold as over the counter preparations, and on the internet. One such slimming aid is pyruvate (Onakpoya et al., 2012).

Pyruvate, a three-carbon compound generated via glycolysis, has been touted as a natural dietary substance that can enhance the loss of body fat (Mahi, 1998). pyruvate might function as an ideal weight reduction and/or weight maintenance agent, with its ability to enhance body weight and fat loss both with short-term severely restricted hypo-energetic dietary therapy and with minimally restricted, longer-term hypo energetic dietary therapy (Stanko et al., 1994).

Accordingly, the purpose of the present study was to investigate the ameliorative effect of pyruvate against obesity-induced in rats obesity -induced in female rats by feeding high fat diet. Also, to determine whether the pyruvate when administered to obese rats would attenuate serum lipids profile, insulin, insulin resistance, leptin, glucose and some serum markers enzymes.

2. MATERIALS AND METHODS

2.1. Experimental animals:

Fifty white female Albino Wister rats (8-10 weeks old age), weighing 165-225 gm were used in the experimental investigation of this study. Rats were kept at constant environmental and nutritional conditions throughout the period of experiment. Animals were housed in separate metal cages, fresh and clean drinking water was supplied ad-libitum. All rats were acclimatized for minimum period of two weeks prior to the beginning of study.

2.2. Ration and additives:

The animals were fed on constant ration through the course of the experiment in the form of concentrated diet composed of (7-10% fat, 68-70% CHO, 18-20% protein, 1-2% vitamins and minerals; 210 kcal/100 g/day) normal control diet (NCD).

2.3. Pyruvate:

Pyruvate (molecular weight 110.050 the active constituent of the dietary for the treatment with purity 99%(pyruvate 99%,chloride 0.003%, sulphate 0.01%,lead 0.001%,iron 0.001%) was purchased from El-Gomhoria Company for Chemicals, Egypt.

Pyruvate was freshly prepared (dissolved in distilled water) and administered in oral daily dosage 540 mg/kg body weight using stomach tube for group IV, and the same dose were administrated for group V, half dose (270 mg/kg body weight) was given for group III for 8 weeks.

2.4. Induction of obesity:

The experimental induction of obesity in female rats was induced by feeding the rats on the prepared high fat diet (HFD) for one month before the beginning of the experiment. The high fat diet (HFD) was consisted of (30% fat, 50-52% CHO, 18-20% protein, 1-2% vitamins and minerals; 210 kcal/100 gm/day). The diet was prepared and necessary vitamins and minerals were added. For fatty diet the chow, in powder form, was mixed fat until become homogenous in a dough-like consistency. This dough was shaped with a paste injector. Obtained chow blocks were dried and used for feeding (Altunkaynak, 2005). One month after obesity induction, treatment with sodium pyruvate were given and continued for eight weeks

2.5. Design of the experimental work:

The Rats under study were randomly divided into five main equal groups, 10 rats each, placed in individual cages and classified as follows:

Group I: (Normal control diet): Rats received normal control diet (NCD) all over the experimental periods (for 12 weeks).

Group Π :(High fat diet): rats received high fat diet (HFD), served as obesity induced rats group, all over the periods of experiment (for 12 weeks).

Group III: (HFD + pyruvate): rats were fed HFD and administered sodium pyruvate (270 mg/kg b. wt./day/orally) for 8 weeks.

Group IV: (HFD + pyruvate): rats were fed HFD and received sodium pyruvate (450 mg/kg b. wt./day/orally)for 8 weeks.

Group V: (CND + pyruvate): rats were maintained on CND and received sodium pyruvate (540 mg/kg b. wt./day/orally)for 8 weeks.

2.6. Sampling:

Random blood samples were collected from all animal groups (control and experimental groups) three times along the duration of experiment at the 2nd, 6th and 8th weeks from the onset of treatment with sodium pyruvate (one month after obesity induction).

2.6.1. Blood samples:

Blood samples were collected from retro orbital plexus of eyes after overnight fasting in clean dry screw-capped tubes, then allowed to coagulate at room temperature for 30 minutes, and centrifuged at 4000 r.p.m for 10 minutes. The clean, clear-serum was separated by Pasteur pipette and received in dry sterile sample tube, processed directly for glucose, ALT, AST and CK-MP determination, then kept in a deep freeze at -20° C until used for subsequent biochemical analysis.

2.7. Biochemical analysis:

The Serum glucose, TC, TAG, HDL-c, LDL-c, VLDL-c, insulin, insulin resistance, ALT and AST, CK-MP, phospholipids, Leptin, were analyzed according to the methods described by Tietz, (1995), NCEP expert panel, (1988), Stein, (1987), National cholesterol education program re-commendation for measurement of High density Lipoprotein Cholesterol, (1995), Friedewald et al., (1972),Bauer, (1982), Connerty et al., (1961), Breuer, (1996), Friedman and Young, (1997), Wilson and miles, (1977), Matthews et al.(1985), respectively.

2.8. Statistical analysis:

The obtained data were analyzed using the statistical package for social science (SPSS, 13.0 software, 2009), for obtaining mean and standard deviation and error. The data were analyzed using one-way ANOVA to determine the statistical significance of differences among groups. Duncan's test was used for making a multiple comparisons among the groups for testing the inter-grouping homogeneity.

3. RESULTS

3.1. Effect of treatment with Pyruvate on serum Glucose, leptin, insulin and insulin resistance concentrations in normal and obesity-induced in female rats:

The obtained results in table (1) revealed that, a significant increase in serum levels of glucose, insulin, insulin resistance and leptin were observed in obesity induced in rats groups. Treatment with pyruvate to HFD-fed rats significantly decreased serum glucose, insulin, insulin resistance, and leptin concentrations compared to control HFD-fed non treated group.

3.2. Effect of treatment with Pyruvate on serum total cholesterol, triacylglycerols and phospholipids concentrations in normal and obesity-induced in female rats:

The obtained results in table (2) revealed that, a significant increase in serum levels of TC, and TAGs and phospholipids were observed in obesity induced in rats groups. Treatment of obese rats fed HFD with pyruvate

significantly decreased serum TC, TAGs and phospholipids concentrations compared to obese non treated rats group.

3.3. Effect of treatment with Pyruvate on serum HDL-c, LDL-c and VLDL-c concentrations in normal and obesity-induced in female rats:

The present data demonstrated in table (3) revealed that, serum LDL-c and VLDL-c concentration were significantly increased, while serum HDL-c level was significantly decreased in obesity induced in rats groups. Treatment of HFD-fed rats with Pyruvate significantly decreased serum LDL-c and VLDL-c with marked increased in serum HDL-c level compared to HFD-fed non treated control group.

3.4. Effect of treatment with Pyruvate on serum creatine kinase-MB (CK- MB), alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities in normal and obesity-induced in female rats:

The obtained results presented in table (4) showed that, a significant increase in the activity of serum AST, ALT and CK–MB were observed in the obese rats compared to the control group. Treatment of HFD-fed rats with Pyruvate significantly improved all motioned enzymatic changes compared to obese non treated control group.

Administration of pyruvate to rats fed normal control diet did not produce any significant changes in all serum biochemical parameters investigated in comparison to the values in normal control rats.

Table (1): Effect of treatment with Pyruvate on serum Glucose, leptin, insulin and insulin resistance concentrations in normal and obesity-induced in female rats

Experimental groups	Glucose (mg/dl)			leptin (pg/ml)			insulin(µIU/ml)			Insulin resistance		
	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks
Group I: (Control negative NCD)	115.90 ±5.61^{a}	100.75 ±2.5^{d}	98.16 ±1.05^{d}	53.04 ±0.54^{b}	137.58 ±0.64^{a}	87.11 ±1.98^{b}	21.98 ±1.62^{a}	21.26 ±1.13^{c}	20.64 ±0.93^{c}	6.1± 0.50^{b}	5.19± 0.25^{d}	4.96± 0.17^{d}
Group II : (control positive HFD)	122.23 ±5.01^{a}	199.75 ±8.26^{a}	194.83 ±3.85^{a}	61.79± 3.17^{ba}	139.07 ±6.32^{a}	97.46± 15.50^{b}	25.19 ±0.66^{a}	27.75 ±0.25^{a}	25.36 ±0.28^{ab}	7.79± 0.49^{a}	14.2± 0.43^{a}	12.38 ±0.24^{a}
Group III:(HFD+pyruvate270 mg/kg.b.wt)	116.40 ±5.31^{a}	155.55 ±2.86^{b}	157.10 ±3.12^{b}	54.64± 1.08^{b}	133.0± 11.47^{a}	75.04± 5.93^{b}	22.67 ±1.06^{a}	25.36 ±0.57^{b}	23.05 ±0.89^{cb}	6.26± 0.35^{b}	9.59± 0.33^{b}	8.8±0. 48^{b}
Group IV:(HFD+pyruvate540 mg/kg.b.wt)	110.53 ±4.42^{a}	138.07 ±3.18^{c}	134.55 ±5.94^{c}	57.24± 11.27^{b}	62.83± 4.32^{b}	73.46± 3.76^{b}	22.32 ±1.77^{a}	23.58 ±0.06^{b}	21.76 ±2.58^{cb}	6.54± 0.68^{ba}	7.9±0 .16^{c}	6.97± 0.92^{c}
Group V: (NCD+pyruvate540mg/kg.b.wt)	114.05 ±3.99^{a}	98.50± 3.62^{d}	97.03± 1.6^{d}	79.20± 4.45^{a}	56.06± 4.79^{b}	125.80 ±6.04^{a}	15.29 ±3.18^{b}	23.68 ±1.01^{b}	27.63 ±0.53^{a}	5.07± 0.08^{b}	5.61± 0.03^{d}	6.55± 0.12^{c}

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 ($P \leq 0.05$)

Table (2): Effect of treatment with Pyruvate on serum total cholesterol, triacylglycerols and phospholipids concentrations in normal and obesity-induced in female rats

Experimental groups	Total Cholesterol (mg/dl)			Triacylglycerols (mg/dl)			phospholipids (mg/dl)		
	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks
Group I: (Control negative NCD)	103.36 ±0.90^{c}	102.09 ±0.68^{c}	97.75±5 .98^{c}	106.15 ±7.99^{c}	98.90± 0.84^{d}	99.88v 0.43^{c}	83.5±4. 67^{a}	82.7±1 .78^{a}	86.8±5 .74^{a}
Group II : (control positive HFD)	134.04 ±1.15^{a}	135.50 ±1.71^{a}	134.21± 0.84^{a}	151.60 ±2.88^{a}	148.08 ±0.91^{a}	152.75 ±5.85^{a}	94.03± 10.86^{a}	91.23± 2.04^{a}	101.05 ±8.6^{a}
Group III:(HFD+pyruvate 270 mg/kg.b.wt)	122.0± 0.41^{b}	121.63 ±0.63^{b}	121.13± 0.75^{ba}	132.75 ±1.11^{b}	132.59 ±1.51^{b}	136.0± 1.63^{b}	80.29± 3.44^{a}	78.77± 6.43^{a}	78.75± 4.81^{a}
Group IV:(HFD+pyruvate 540 mg/kg.b.wt)	120.38 ±1.38^{b}	118.75 ±0.22^{b}	119.20± 0.58^{b}	128.03 ±0.94^{b}	128.70 ±1.22^{c}	128.43 ±1.24^{b}	88.54± 4.91^{a}	89.73± 5.23^{a}	96.65± 6.77^{a}
Group V: (NCD+pyruvate54 0mg/kg.b.wt)	101.81 ±0.12^{c}	102.12 ±1.03^{c}	94.75±8 .28^{c}	103.90 ±5.51^{c}	98.05± 1.41^{d}	98.43± 1.38^{c}	80.05± 6.01^{a}	82.87± 5.62^{a}	82.02± 6.78^{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): Effect of treatment with Pyruvate on serum HDL-C, LDL-C and VLDL-C concentrations in normal and obesity-induced in female rats

Experimental groups	VLDL-C (mg/dl)			LDL-C (mg/dl)			HDL-C (mg/dl)		
	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks
Group I: (Control negative NCD)	21.23± 1.6^{c}	19.78± 0.17^{d}	19.98± 0.09^{c}	41.96± 1.89^{c}	42.93± 0.62^{c}	37.3±6. 18^{c}	40.18± 0.96^{a}	39.38± 0.58^{ba}	40.47± 1.86^{a}
Group II : (control positive HFD)	30.32± 0.58^{a}	29.62± 0.18^{a}	30.55± 1.17^{a}	70.66± 0.59^{a}	74.86± 1.35^{a}	72.04± 0.96^{a}	33.0±1. 12^{c}	31.03± 0.45^{c}	31.63± 0.63^{c}
Group III:(HFD+pyruvate 270 mg/kg.b.wt)	26.55± 0.22^{b}	26.52± 0.30^{b}	27.2±0. 33^{b}	60.05± 0.76^{b}	58.48± 1.57^{b}	59.24± 1.26^{b}	35.40± 0.78^{bc}	36.63± 1.26^{b}	34.69± 0.65^{cb}
Group IV:(HFD+pyruvate 540 mg/kg.b.wt)	25.61± 0.19^{b}	25.72± 0.24^{c}	25.69± 0.25^{b}	57.08± 1.37^{b}	55.36± 1.72^{b}	56.59± 1.48^{b}	37.70± 1.30^{ba}	37.65± 1.70^{ba}	36.92± 1.01^{b}
Group V: (NCD+pyruvate540 mg/kg.b.wt)	20.78± 1.1^{c}	19.61± 0.28^{d}	19.69± 0.28^{c}	40.49± 1.31^{c}	42.55± 2.02^{c}	34.08± 8.03^{c}	40.55± 0.64^{a}	40.47± 1.07^{a}	41.0±0. 46^{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 (4): Effect of treatment with Pyruvate on serum creatine kinase-MB (CK- MB), alanine amino-transferase (ALT) and aspartate aminotransferase (AST) in normal and obesity-induced in female rats

Experimental groups	AST(U/L)			ALT(U/L)			CK -MB(U/L)		
	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks	2 weeks	6 weeks	8 weeks
Group I: (Control negative NCD)	55.98± 1.22^{c}	55.88± 1.03^{b}	50± 6.72c	18.38± 0.63^{c}	18.25± 0.48^{c}	18.5± 0.96^{d}	140.8± 0.8^{dc}	141.35± 0.91^{c}	141.42± 1.34^{c}
Group Π : (control positive HFD)	85.0± 5.11^{a}	113.0± 19.53^{a}	85.55± 4.97^{a}	36.48± 1.69^{a}	37± 1.48^{a}	36.56± 1.74^{a}	235.3± 13.25^{a}	249.73± 16.74^{a}	251.4± 15.06^{a}
Group III:(HFD+pyruvate270 mg/kg.b.wt)	74.5± 1.67^{b}	75.35± 1.84^{b}	74.63± 1.21^{ab}	28.28± 0.63^{b}	28.38± 1.43^{b}	28.06± 0.42^{b}	165.22± 0.8^{cb}	165.96± 0.41^{b}	139.62± 14.73^{c}
Group IV:(HFD+pyruvate540 mg/kg.b.wt)	71.15± 1.42^{b}	70.38± 0.63^{b}	71.03± 0.93^{b}	24.81± 2.14^{b}	27.53± 1.86^{b}	23.13± 1.48^{c}	119.7± 0.8^{b}	177.14± 0.89^{b}	179.27± 1.93^{b}
Group V: (NCD+pyruvate540 mg/kg.b.wt)	55.3± 0.69^{c}	55.33± 1.13^{b}	54.5± 0.62^{c}	16.75± 2.29^{c}	18.91± 1.1^{c}	19.26± 1.31^{d}	167.7± 12.9^{d}	140.8± 0.96^{c}	140.33± 0.47^{c}

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 ($P \leq 0.05$).

4. DISCUSSION

Obesity is characterized by hormonal changes with a number of metabolic abnormalities which in all may contribute to development of cardiovascular disorders (Vecchione et al., 2002). There is great evidence suggesting obesity as an independent risk factor for a number of health problems, including cardiovascular disease (CVD) (Chandrasekaran et al., 2012). The obtained results in table (1) revealed that, a significant increase in serum levels of glucose, insulin, insulin resistance and leptin were observed in obesity induced in rats groups. Insulin resistance is not only an early and major feature in development of non insulin dependent diabetes mellitus (NIDDM), but also associated with hyperlipidemia, hypertension, obesity, enhanced oxidative stress, endothelial dysfunction and cardiovascular disease, so called insulin-resistance syndrome (syndrome X, metabolic syndrome) (Oudot et al.,2009). Obesity is associated with elevated basal plasma insulin levels and resistance to the metabolic effects of insulin (Ferranti and Mozaffarian, 2008). Independent of obesity, high-fat feeding itself contributes to impaired glucose tolerance and insensitivity to the blood glucose lowering effect of insulin. The fatty acid profile of the diet also, plays a crucial role in insulin resistance (Riccardi et al., 2004). Loss of insulin action causes a shift in balance from oxidation to esterification of free fatty acids (FFAs), resulting in elevated very low-density lipoprotein (VLDL) secretion (Mayes, 1993). In addition, insulin resistance can be considered as additional factor which contributes to increased cardiovascular disease in obesity. Insulin resistance is a state in which higher concentration of insulin is required to maintain normoglycemia (Eckel et al., 2005). The action of insulin is initiated by binding to its receptors and activation of intrinsic protein tyrosine kinase activity of the receptors, resulting in initiation of intracellular signaling cascade that eventually related to glucose and lipid metabolism (Westerbacka et al., 2002). It is well established that increased availability and utilization of free fatty acids (FFAs) play a

critical role in the development of insulin resistance. Excess adipose tissue has been shown to release an increased amount of FFAs which directly affect insulin signaling, diminish glucose uptake in muscles, drive exaggerated triglyceride synthesis and induce gluconeogenesis in the liver leading to elevated levels of glucose and lipids(Mlinar et al., 2007). Leptin is primary involved in the regulation of body weight by centrally inhibiting food intake and stimulating energy expenditure. Leptin enters the circulation and crosses the blood-brain barrier to reach its primary target receptors in the hypothalamus. Binding of leptin to these receptors triggers intracellular pathways in the hypothalamus satiety centers which in turn signal brain for restricting food intake and regulating body weight (Ahima and Osei, 2004). There is provide evidence that, serum leptin was elevated in obese human (Orel et al., 2004) and animals (Scarpace and Zhang, 2008). Moreover, Masoud and Adel, (2006) reported that, serum leptin concentration was increased in relation to increased body fat content. The positive correlation between body fat and serum leptin is probably explained by the increased release of leptin from large fat cells. Thus, leptin can serve as an indicator of fat content and its level may be decreased by reduction of body weight. Additionally, Lin et al., (2000) suggested that, during high fat feeding animals are sensitive to the food lowering effect of leptin. However, despite the reduction in food intake, animals become fat as a result of the increase in food efficiency leading to an increase in plasma leptin levels followed by resistance to its action. Modern investigation suggested that, leptin resistance as contributing factor for incidence of hypertension and cardiovascular complications in obese subjects, which in turn may be linked to impairment of vascular endothelial function (Singh et al., 2010a). On this issue, it was demonstrated that leptin receptors are present on endothelial cells and that increasing doses of hormone are able to exert vasorelaxant response through increasing endothelial production of nitric oxide (NO) (Shiuchi et al., 2001). However, the chronic condition of hyperleptinemia typical of obesity could be accompanied by impaired endothelial vasorelaxation through deficiency of NO production (Tripathy et al., 2003). The obtained results in tables (2 and 3) revealed that, serum TC, TAGs, phospholipids, LDL-c and VLDL-c concentrations were significantly increased while HDL-c level was significantly decreased in obesity induced in rats groups. Increased lipid profile has also suggested being a major risk factor predisposing obese subjects to develop CVD. In different obese states, level of TC is frequently increased possibly through decreased level of HDL-C, together with increased LDL-C concentration. As reported earlier, LDL-C is the major cholesterol carrier in the blood, about 60-80% of cholesterol is carried by LDL-C. Some of cholesterol is used by tissues and other returned to liver (Quinet et al., 2009), but if there is much LDL-C in blood, cholesterol may be deposited. On the other hand, HDL-C picks up cholesterol and takes it back to liver for reprocessing or excretion by a pathway called reverse cholesterol transport (Xie et al., 1999). Consequently, decreased HDL-C is associated with decreased cholesterol removal from extra hepatic tissues and increased risk of developing cardiovascular disorders. Events of cardiovascular disorders may also involve elevations of serum VLDL-c and TAGs with subsequent accumulation of TAGs in the vascular wall and cardiac tissue (Vallance and Chan, 2001). HDL is the most protective because it is rich in surface phospholipids (Goldfarb et al., 2003). Phospholipids are a class of lipids that are a major component of all cell membranes as they can form bilayers. The phospholipids of the plasma lipoproteins are synthesized in the liver and intestinal wall and are incorporated

into the lipoprotein macromolecules before their discharge into the circulation. It seems likely that their function is to stabilize lipoproteins by acting as a link between the protein and the less polar lipids of the protein lipid complex (Campbell et al., 2006). While et al., (1991) reported that, dietary fats can significantly alter the proportions of phospholipids and their fatty acyl constituents in tissue of obese and to a lesser extent lean rat. Based on this, the present findings revealed that, HFD-fed rats showed raised lipids profile characterized by elevation in serum TGs, TC, and Phospholipids concentrations as well as serum VLD-c, LDL-c with decreased serum HDL-c level may indicate development of CVD. Accordingly, the increased insulin level as seen in the present study may indicate a state of insulin resistance which in turn may contribute to incidence of hyperglycemia and raised lipid profile in serum of the obese rats. The results demonstrated in table (4) showed that, a significant increase in the activity of serum AST, ALT and CK–MB were observed in the obese rats. Normally, Nitric oxide (NO) functions to maintain vascular homeostasis, while decreased production of NO is associated with vasoconstriction that accelerates development of atherosclerosis with increased myocardial injury (Dubey et al., 2008). When myocardial cells are injured, many enzymes such as (CK-MB, LDH, ALT, and AST) can be released from the myocardial cells to the extracellular fluid as a result of alterations in plasma membrane integrity and/or permeability (Ramadan et al., 2012). Accordingly, it can said that various events, such as hyperleptinemia, decreased NO level and increased serum ALT, AST, LDH and CK-MB, with reduction in their activities in liver, aorta and cardiac tissue, as seen in the present study may indicate incidence of CVD as consequence of obesity. Treatment of obese rats with pyruvate significantly decreased serum glucose, insulin, insulin resistance, leptin TC, TAGs, phospholipids, LDL-c and VLDL-c concentrations with marked increased in serum HDL-c level. Also, treatment of HFD-fed rats with Pyruvate significantly improved all motioned enzymatic changes compared to obese non treated control group. Leptin is secreted by adipose tissue and has been shown to play an important role in feed intake regulation, energy metabolism and mammalian reproduction (Sun et al. 2006). Furthermore, glucagon is the hormone responsible for controlling lipolysis in fowl (Freeman and Manning 1976). It is well established that the pancreatic hormones, insulin and glucagon regulate intermediary metabolism in birds (Cogburn1991). This study demonstrated that 5 and 10% Cr-Pyr addition enhances the concentrations of serum leptin, insulin and glucagon. Accordingly, glucagon -stimulate catabolic pathway of fat, resulting in reduced accumulation of fat in broilers. Also, Johnstone et al., (1989) reported that, pyruvate/dihydroxyacetone (1:1; P/D) supplemented at 10% of the diet significantly decreased body weight of SCWL pullets. These results are consistent with those reported by Cortez et al. (1991) and Ivy et al. (1994) who recorded that, pyruvate given for 3-5 weeks to rats resulted in decreased body weight. In addition, the abdominal fat, serum and liver TG concentrations were significantly decreased, whereas serum HDL-C concentration was increased in the 5 and 10% groups. Furthermore, (Olson et al. 1991) found that, pyruvate promotes fat loss and regulates cholesterol levels, as it may increase the concentration of plasma HDL-cholesterol. On the other hand, the growth of adipose tissue is a balance between lipogenesis and lipolysis. Stanko and Adibi (1986) found that, the rate of lipid synthesis in the adipose tissue of rats receiving the experimental diet was significantly reduced. The reduction in the rate of lipid synthesis was accompanied with a lower blood level of insulin, which is considered a key hormone in

regulating lipid synthesis. Insulin can influence the production of pyruvate by its modulatory action on glucose metabolism, while pyruvate is an insulin secretagogue (Liu et al., 2002). It is claimed that pyruvate may also enhance the fat loss that accompanies physical training (Stone et al., 1999). Since it is known that patient with hyperlepitinemia are at increased risk for cardiovascular disease through impaired NO production, action of pyruvate such as decreased leptin and increased NO availability as shown in the present study appears to play important role in preventing cardiovascular disease associated with obesity. A result which is further supported by the present finding of normalized activities of ALT, AST and CK-MB in serum, liver of pyruvate administered HFD-rats. Thus, indicating the protective activity of pyruvate against obesity- induced CVD. Furthermore, Shen et al., (2013) reported that, ethyle pyruvate attenuates hepatic ischemia reperfusion (I/R) injury and the histopatholigical changes caused by I/R such as cellular necrosis, neutrophil infiltration and cellular swelling are clearly ameliorated by ethyle pyruvate which are consistent with changes in ALT and AST activities. On the other hand, Cr-Pyr treatment augmented creatine kinase (CK) enzyme activity. Creatine kinase (CK) plays a key role in muscle energy metabolism, keeping the cellular ATP concentration stable during fluctuating rates of ATP turnover, and reversibly transfering a phosphoryl group from ATP to creatine (Wallimann et al. 1992). A possible explanation for this is the administration of Cr-Pyr resulted in higher plasma concentrations of creatine (Jager et al., 2007), which reversibly increased CK phosphorylation activity; with results of that product phosphocreatine was able to donate inorganic phosphate and energy to re-phosphorylate ADP and sustain the proper energetic environment in skeletal muscle (Harris et al. 1992). It is presumed that pyruvate might have maintained the cell integrity and stabilized the myocardial membrane which restricts the leakage of CK-MB from the heart into blood (Ojha et al., 2010).

Conclusion: administration of pyruvate to HFD-fed helped in controlling obesity, tended to prevent hyperglycemia, improve dyslipidemia and other changes relevant to cardiovascular disease mainly through improving leptin and insulin resistance. These results suggest that, pyruvate is effective in improving the obesity and it's associated many important complications such as diabetes mellitus and coronary heart disease. So, we recommended that, administration of diet rich in Pyruvate as a natural dietary product is very important and suitable for weight reduction, attenuate the metabolic disorders of different body tissue and protection of vital organs against obesity complications.

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5. REFERENCES

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البيروفات يحسن اضطرابات أيض الدهون ومقاومة الأنسولين في الفئران المحدث فيها السمنة

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الملخص العربي

السمنة من أكثر الأسباب التي تؤدى الى أمراض القلب. لقد أجريت هذه الدراسة لمعرفة التأثير الواقي والعلاجي للبيروفات الصوديوم على التغيرات الكيميائية الحيوية التي تحدث فى دم وأنسجة الفئران المحدث فيها السمنة تجريبيا. هذا وقد استخدم لأجراء هذه الدراسة 75 من اناث الفئران البيضاء أعمارها تتراوح من 8-10 أسبوع وأوزانها من (165-225جرام) وقد قسمت الى مجموعات متساوية اشتملت كل مجموعة على عدد 15 فأر وتم توزيعها كالاتي: المجموعة الاولى (المجموعة الضابطة) اشتملت على 15 فأر لم تعطى أى ادوية واستخدمت كمجموعة ضابطة للمجموعات الاخرى والمجموعة الثانية (المجموعة المحدث بها السمنة): تكونت من 15 فأر تم زيادة معدل الدهون فى وجباتهم. المجموعة الثالثة (مجموعة ملح بيروفيت الصوديوم) تكونت من 15 فأر تم اعطائها عن طريق الفم جرعة مقدارها 270ميللى جرام لكل كيلو جرام من وزن الجسم. المجموعة الرابعة: اشتملت 15 فأر تم احداث السمنة بها عن طريق زيادة معدل الدهون فى وجباتها ثم اعطائها ملح بيروفيت الصوديوم عن طريق الفم بجرعة مقدارها 540 ميللى جرام لكل كيلو جرام من وزن الجسم. المجموعة الخامسة: اشتملت على 15 فأر تتغذى على وجبات عاديه غير محدث بها سمنه ثم تم اعطائها ملح بيروفات الصوديوم عن طريق الفم بجرعة مقدارها 540ميللى جرام لكل كيلو جرام من وزن الجسم. وقد تم تجميع عينات الدم على فترات بعد 2-6-8 أسابيع من بدء العلاج بملح بيروفات الصوديوم وذلك بعد 4 اسابيع من احداث السمنة فى انابيب نظيفة وجافة ومعقمة. وقد تم فصل مصل الدم واستخدم مباشرة لقياس تركيز سكر الدم والانسولين ومقاومه الانسولين والكوليسترول الكلى والدهون الثلاثية والدهون عالية الكثافة والدهون منخفضة الكثافة والدهون الفوسفورية وانزيمات الكبد والكيرياتين كاينيز ام بى. وقد أسفرت نتائج التحاليل البيو كيميائي عن وجود زيادة فى كلا من سكر الدم والانسولين والكوليسترول الكلى والدهون الثلاثية والدهون عالية الكثافة والدهون الفوسفاتية وانزيمات الكبد والكرياتين كاينيز ام بى. كما اوضحت النتائج ان اعطاء ملح بيروفات الصوديوم يؤدى الى انخفاض فى كلا من سكر الدم والانسولين والكوليسترول الكلى والدهون الثلاثية والدهون عالية الكثافة والدهون الفوسفورية وانزيمات الكبد والكرياتين كاينيز ام بى. وخلصت الدراسة أن ملح بيروفات الصوديوم له تأثير جيد في خفض مستوى سكر الدم وتحسين نسبة الدهون العالية لذلك لديه القدرة من الحد من مضاعفات أمراض القلب التي تنتج من زيادة الدهون. ولذلك ينصح بملح بيروفات الصوديوم لمن يعانون السمنة.

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