Bulletin of High Institute of Public Health
Vol. 37 No. 4 [2007]

Nutritional Biochemistry of Imbalanced Dietary Systems in Rats

Abear H. El-Dakak*, Mona Hanafi*, Ghada M. Yossife*, Ali I.S. Ahmed*

Abstract: Food is of great role in human health. The best goal in this concept is variety in the diet and moderation. This hypothesis has been observed on bases of nutritional biochemistry. A single food, i.e., Phaseolus bean, was used at rates of 30, 50, and 100% replacements of the basal diet for aged rats. It was clear that the higher rate of morbidity and mortality due to this sort of malnutrition, or semistarvation, has been associated with some biological changes. Although beans are rich in containing proteins, the partial replacement of this plant food origin at 30% has shown to be a good help in keeping an optimal body weight (BW) of those aged animals, but any more degree of replacement has seen to be vital in affecting the animal health, hence 50% or over might resulted in animal death. It seemed that 25% may be useful in maintaining BW, meanwhile, a further rate of this plant origin replacement up to a specific level should be an effective way in treating obesity if used in a proper dietary system. In more details, organ morphologies and histopathological examination of some of which as spleen, brain, and liver are greatly affected by imbalanced diets. In another word, the metabolic differences rolled up by this sort of feeding negatively affected liver and brain expectancy or longevity. A number of tissues up normality, such as liver tissues dilatation and congestion of hepatoportal vessels, hepatocytes karyomegaly nuclei, congestion of central vein, and granular degeneration of hepatocytes with both most higher rates of replacement groups. Moreover, there were focal gliosis, pyknosis of neurons, brain edema, and hemorrhage in cerebrum and cerebellum in brain of the same groups. It is obvious that all these organ dysfunctions are frequently occurred as a response to these and similar kind of malnutrition, other than energy one, that has been existed for a long enough time. This weak homeostatic system has been shown as less control on the metabolic constants such as blood glucose (G), triglycerides (TG) and total cholesterol (TC). All those are most likely correlated with the organs subclinical defects mentioned before. It seems that this biological control that actually supported in the presence of balanced diet can be a biological up regulatory mechanisms, and the death eventually occurs due to a protein catabolic pathway due to a protein, the quality but not only quantity, malnutrition. It is an emergence metabolic system (EMS) that runs for longer time caused by a strengthen need for some micronutrients as well as high rate of nitrogenous metabolite accumulation. This protein turnover in such high rates beyond the biological capacity of both intracellular and extracellular compartments negatively affect the whole biological system including cells, tissues, organs, and eventually the blood chemistry as well. Refeeding with a proper system of animal high quality protein and some micronutrients might be vital.

INTRODUCTION

Food is of greatest role on human health. Food shouldn't be judged in isolation, but in relation to the total diet and the individual's needs (Guthre, 1993[1]). In selecting food moderation is a virtue. Moderate of calories and balanced

*Special Food and Nutrition, Meat and Fisheries, Food Technology Research Institute, Agric. Res. Ccenter, Giza. Egypt

nutrients are neckties to provide essential requirements. A 10 to 15% RDA calories from food of limited nutritional value is reasonable, more can lead to nutritional problems (Guthre, 1993)[1]. Although the food production has markedly increased according to the world total production, several areas in this earth are suffering from food shortage. Understanding the biology of hunger might be one great step to rescue those people wherever they are. Biologically, however, with starvation progress and lean body (LB) mass decrease, both BMR and protein oxidation decrease. It has been very early to state that one of the most consistent autopsy findings of animal and human who died of pure starvation is the virtual absence of depot fat, both subcutancously and inernally (Keys *et al*., 1950)[2]. In subjects ingesting hypocaloric diets, the effect of initial adiposity on protein turn over (PTO) and composition of N loss in relation to other confounding variables, e.g., composition of dieting, exercise, etc. is not fully understood**.** However, brain, gonads and skeleton appeared to be preferentially preserved (Elia, 1991, Forbes and Drenick 1979)[3,4].

The biology of starvation stress has been explored at level of enzymes, hormones and genetics most recently **(**Harbison, *et al*., 2005[5]; Mackay, 2005[6]; Lee *et al*., 1999[7], Lee *et al*., 2000[8]; Schadt *et al*., 2003[9]; Teichert *et al*., 1989[10]; Zinke *et al*., 2002[11]; Nelson *et al*., 1995[12]; and Seglen and Bohley 1992)[13]. These studies involved man, animals, insects, and microorganisms. Moreover, relation between diet and hormonal balance was observed (Ahmed *et al*., 2003[14]). In this concern, Svanberg *et al*., (2000)[15] evaluated the effect of insulin like growth factor-I (rhIGF-I) in complex with binding protein 3 (IGFBP-3) compared to the effect of free IGF-I on muscle protein biosynthesis in undernourished animals. The female Sprague–Dawley rats (200 g)

were initially semistarved in order to carry out this investigation. The rhIGF-I/rhIGFBP-3 (SomatoKine) was a significant stimulator of muscle protein synthesis in chronically semistarved animals whereas IGF-I alone failed to increase protein synthesis during the same experimental conditions. This stimulation was because of increased initiation of translation, likely induced by more physiologic concentrations/kinetics of plasma IGF-I and amino acids following rhIGF-I/rhIGFBP-3 treatment, compared to IGF-I in its free form (Svanberg *et al*., 2000)[15].

Likewise, the effects of dietary restriction on the kinetics of absorption in vivo of glucose, galactose and alpha-methyl glucoside were assessed by electrical and chemical methods in the rat jejunum. Fasting for 3 days greatly decreased the apparent Kms obtained from electrical or chemical data for all the sugars but had no effect on those for L-valine or L-methionine. Semistarvation caused a less pronounced reduction of the apparent Kms for the sugars. The dietary-induced change in apparent Km for glucose was also observed in the fasted hamster. One interpretation of these changes is that the affinity of the carriers for sugars increases during dietary restriction, the greater the level of restriction the greater the increase . A standard diet obtained from two commercial sources was found to differ greatly in its effect on the electrogenic transfer system for alpha-methyl glucoside but had no effect on those for galactose and glucose (Debnam and Levin, 1975[16].

From a metabolic point of view, previous work in humans and rats has revealed a link between perinatal growth retardation and glucose intolerance in adulthood. Both maternal semistarvation and severe diabetes are accompanied by

perinatal growth retardation in rats. This study compared the effect of these conditions on tissue glucose uptake in their female offspring. Glucose uptake was measured as glucose metabolic index (GMI), using 2-deoxy- [1-^{3}H]-glucose, in the postabsorptive state and during euglycemic hyperinsulinemia (Holemans**,** *et al*., *1997)*[17]*.* The GMI was measured in insulin-sensitive tissues, e.g., skeletal muscles, diaphragm and white adipose tissue and in two noninsulin-sensitive tissues such as duodenum and brain of adult offspring of normal dames. Dames rendered diabetic with streptozotocin on d 11 of pregnancy, and dames fed half-normal rations from d 11 of pregnancy. Whole-body insulin resistance, measured by decreased glucose infusion rate during hyperinsulinemia was milder in offspring of semistarved rats (O-SR) than in offspring of diabetic rats (O-DR) (Holemans**,** *et al, 1997*[17]). The basal GMI did not differ among the three groups in any tissue except tibialis anterior; during hyperinsulinemia, GMI was significantly greater in the insulin-sensitive tissues of all three groups. GMI of skeletal muscles and adipose tissue during hyperinsulinemia did not differ between control rats and O-SR; in contrast, the GMI was 25-50% lower in skeletal muscles of O-DR during hyperinsulinemia than in those of control rats or O-SR. Thus, maternal semistarvation and diabetes have dissimilar effects on peripheral insulin sensitivity of the adult female offspring. Because both conditions are associated with perinatal growth retardation and fetal hypoinsulinemia, other mechanisms must be identified to explain impaired glucose uptake by skeletal muscles in the offspring of diabetic rats *(Holemans**,** et al.,* 1997)[17].

The other mechanisms must be identified to explain impaired glucose, or any other nutrient, uptake by skeletal muscle upon semistarvation or some sort of diabetes is what so called the

emergence metabolic system of a protein catabolic status (EMS). To put it more clear, some biological factors other than loosing weight or factor other than the shift in base metabolic ratio (BMR) may posses the main reason to death. The other metabolites that deviated with sever reduction in energy were the elevation of some blood parameters (Ahmed *et al*., 2006)[18].

More accurate investigation is conducted to biologically explore this metabolic conjugation. It is clear that this emergence catabolic status of proteins ECS saves the animals, but its existence for longer time may posses a reverse effect. In another word, this ECS is a sort of hormonal oxidative imbalance, which abuses the biological system at the long run[18]. Here, Attkin's diet, as a reverse condition to starvation, may be used as a suitable answer for such scientific confusion.

Here, we are observing the metabolic changes in conjugation with consuming imbalanced diet for longer run in order to figure out the real metabolic complication that lead to death. One main cause of designing this sort of study is to help the UN experts in proper treating such types of human disasters

MATERIAL AND METHODS

Animals and Rationales: Male Albino adult rats sorgue-Dawley western strains aged 3 months have been selected of average weight 310+/-10g. These 24 (4x6) animals were obtained from the biological unit of FTRI, ARC, Cairo. All rats fed on basal diet consisted of 10% protein, 10% cottonseed oil, 5% cellulose, 4% salt mixture, 1% vitamin mixture and 70% corn starch of almost 410 calories/100g diet (Lane-Peter and Pearson, 1971)[19]. After an adaptation period of 7 days, they were divided into four groups. The control (C): a 6 animals that continued fed on complete

basal diet as isocaloric balanced diet, another three groups of same number and weight each started to be fed with 30, 50, and 100% of the basal a single beans replacement diet.

Analytical methods: Estimating body weight gain (BWG) according to Chapman *et al*., (1959)[20] has been carried out as one of the main factor for the biological evaluation. The blood picture in rat's specimens, i.e., blood glucose (G) and triglycerides (TG) were examined as described by Dacie and Lowis (1984)[21]. Total cholesterol in serum was determined enzymatically by kit based on the technique of Richmond (1973)[22]. The alanine amino transferase (ALT) and aspartate amino transferase (AST) were measured colorimetrically as described earlier (Richman and Frankely, 1957)[23].

***Histopathological examination of liver and brain*:** The organs were collected and post-mortal examination was done as soon as possible. Fixation was done in 10% of natural formalin, dehydrated, cleared, and ended paraffin then sectioned at (4-6 mm), and stained with Harris hematoxylin, and casein for histopathological examination (Frankel and Reitman, 1963)[24]. Data have been expressed as main of six animal measurements.

RESULTS AND DISCUSSION

Food has a greatest effect on human health. One of the scientific ways to explore the ranges of this fact is the starvation and semistarvation studies. In the current investigation on rats, the trial was carried out on male animals of almost the same weight and age. The most important fact that may extracted from Table (1) is that the partial replacement of plant food of single origin, i.e., 30% bean, considerably helps in keeping an optimal BW of those aged animals, but any more degree of replacement vitally affected the animal. It seemed, for instance, that 25% may be useful in mentaining BW,meanwhile, a further rate of

replacement up to a proper level might become an effective way for treating obesity. It is, therefore, conform the delicate balance between macro and micronutreints.

Table (1) The body weight response to shift in balanced diet in rats of the same age and 310±10g IBW.

IBW(298g)	1ST wk	2nd wk	3rd wk	4th wk	5th wk	6th wk	FBW/IBW
C	318/100	331/100	375/100	380/100	384/100	390/100	1.31
30%	327/104.6	310/93.7	305/81.3	295/77.1	290/75.5	277/71.0	0.93
50%	308/96.9	278/84.0	268/71.5	239/62.9	195/50.8	-	0.63
100%	310/97.5	299/90.3	291/77.6	266/70.0	192/50.0	-	0.62

IBW and FBW are initial and final body weight, respectively.

However, since there were no remarkable changes in the isocaloric condition of the dietary system, the partial elimination of most minor nutrients had greatly affected the overall health situation. The Table showed the gradual effect among all animals throughout the entire time of the experiment. The FBW/IBW shows also that both 50 and 100% had the same final consequences. It is clear that more than 30% substitution of that single food source, or most probably any other single food even whose nutrition value is higher, is not nutritionally accepted. However, it has been stated that food shouldn't be judged in isolation, but in relation to the total diet and the individual's needs (Guthre, 1993)[(1)]. That means, in selecting food, moderation is a virtue and moderate calories and balance diet are essential to provide the required nutrients. More clearly, a 10 to 15% RDA calories from food of limited nutritional value is reasonable, more can lead to nutritional

problems (Guthre, 1993)[1]. In this respect, diets containing more CHO, however, have been reported to be associated with greater hunger (Davies *et al*., 1984)[25].

More variations are also recorded in case of organs morphology. Table (2) demonstrated that spleen and liver are greatly affected by imbalanced dietary system, followed in this respect with lung. However, the changes did not make any particular trend, but must be connected to a physiological disorder, which is diet dependent in particular.

Table (2) Effect of feeding on some imbalanced diet on organ weight in rats.

Group	Heart		Kidney		Spleen		Liver		Brain		Lung	
C	0.786	100	1.224	100	0.408	100	4.130	100	1.506	100	1.750	100
30%	0.750	95.42	1.590	129.9	1.070	262.2	8.510	206.0	1.460	96.9	1.650	94.3
50%	0.775	98.60	1.250	102.1	0.420	102.9	4.130	100.0	1.640	108.9	2.280	130.3
100%	0.720	91.60	1.480	120.9	0.564	138.2	4.460	107.9	1.650	109.6	1.550	88.6

This, as a matter of fact, was proved in some blood analysis. As seen in Table (3,) semistarvation caused a strong elevation in both blood glucose and total triglycerides, which is a kind of metabolic disorder.

Table (3) Effect of feeding rats with imbalanced diet on some blood parameters.

Group	G		TG		TC	
C	71.8	100	44.7	100	55.3	100
30%	70.4	98.1	41.9	93.7	61.2	110.7
50%	111.5	155.3	67.6	151.2	74.9	135.4
100%	151.7	211.3	98.8	221.0	69.8	126.2

Actually, the total cholesterol shows similar trend, but with a little variation. These metabolic changes must be correlated with the damage of body tissues and cells out of

hunger or by using the stored protein more frequently as an emergent metabolic system (EMS). Here, protein catabolism is important, not only as an alternative source of fuel, but also as a source for amino acids. Teichert *et al*., (1989)[10] mentioned that Lysosomal (vacuolar) proteinases of yeast are essential catalysts for protein degradation, differentiation, and cell survival. In fact, one purpose of EMS is to secure a steadily proper secretion of these enzymes (Gottesman and Maurizi, 2001)[26].

Likewise, and according to the tissues histopathology, the metabolic differences rolled up by the imbalanced diet are connected with organ, i.e., liver and brain's expectancy. Tables 4 and 5 recorded number of upnormalities such as dilatation, and congestion of hepatoportal vessels, hepatocytes karyomegaly nuclei, congestion of central vein and granular degeneration of hepatocytes have been disposed to liver of both groups 3 and 4. There is no doubt that the degree of imbalancing in diet in long run negatively affects the biological system in those organs.

It is such a weak homeostatic control of metabolic system in liver, as seen in Table (4) and brain as in Table (5) that affect the blood constants such as G, TG, and TC. This biological control supported by balanced diet is an upregulatory biological mechanism. *Escherichia coli* bacterium is anything but idle when its environment shifts from nutrient-rich to nutrient-poor. The new findings show that a string of phosphate residues in *E. coli* kicks the Lon protease into action. This enzyme then chops up ribosomal proteins to release free amino acids that are used to manufacture biosynthetic enzymes, which help the bacterium to adjust to its nutrient-poor-environment (Gottesman and Maurizi, 2001)[26].

Table (4) Effect of feeding rats on some imbalances diet on liver histopathology.

Group	C	30%	50%	100%
Dilatation and congestion of hepatoportal vessels	_	_	3+	_
Kupffer cells activation	_	_	_	_
Hepatocytes necrosis	_	_	_	_
Hepatocytes active nuclei	_	_	_	_
Epithelial vascular lining bile duct	_	_	_	_
Portal infiltration	_	_	_	_
Lucocytic cells	_	_	_	_
Hepatocytes cytomegaly nuclei	_	_	_	_
Hepatocytes sinusoids	_		_	_
Hepatocytes karyomegaly nuclei	_	+	_	_
Congestion of central vein	_	+	_	_
Granular degeneration of hepatocytes	_	_	_	+
Vacuolar degeneration of hepatocytes	_	_	_	_
Ranking number	1	2	3	2

On the other side, there were focal gliosis, pyknosis of neurons, brain edema, and hemorrhage in cerebrum and cerebellum in brain. These organ disease is frequently occurred as a response to a sort of malnutrition that has been predisposed in groups 3 and 4. Similar ranking number to that of liver is recorded for brain as seen in Table (5).

Table (5) Effect of feeding rats on some imbalance diet on brain histopathology.

Group	C	30%	50%	100%
Focal gliosis	_	_	+	+
Pyknosis of neurons	_	_	+	+
Neurons central chromatolysis	_	_	_	_
Neuronal degeneration and neuronophagia	_	_	_	_
Fibrosis of meningese	_	_	_	_
Brain edema	_	_	_	+
Multiple small focal gliosis	-	_	_	_
Degeneration of neuron	_	_	_	_
Hemorrhage in cerebrum and cerebellum	_	_	_	+
Ranking number	1	1	3	4

It can be concluded, however, that the balanced diet is necessary to sustain important functions of liver, which role up the nutritional homeostatic and metabolic systems that may be manifested by a proper hormonal diverse secretions. In some other words, the EMS connects starvation, as mentioned above, involves special hormonal imbalance. For example and in connection to our matter, Nelson *et al*., (1995)[12] suggested a neuroendocrinal involvement in aging. They recorded evidence from studies of reproductive aging and caloric restriction to establish their hypotheses. However, the growth hormones and insulin would play important role in correcting this sort of metabolic deviation. Growth hormone (GH) acutely stimulates forearm muscle protein synthesis in normal humans. The short-term effects of GH on skeletal muscle protein synthesis and degradation in normal humans are unknown (Fryburg *et al*., 1991)[27], but according to the present data, an animal protein formula is the answer. In this concept, the EMS is mainly a sort of catabolism reaction directed by hormones such as epinephrine. This may

explore the raising up of TC. The elevation of TC to some extant has been found to associate organ enlargement and hormonal imbalance (Ahmed *et al*., 2005)[28]. Although dietary therapy of muscle degradation with aging or due to EMS is not simple with the absence of a proper hormonal deviation, starving and semistarving data may help in aging studies. The increase in muscle strength and size, however, was not influenced by the predominant source of protein consumed by older men with adequate total protein intake (Haub, *et al*., 2002)[29]. Meat consumption may enhance protein synthesis and muscle hypertrophy by providing creatinine as one of the dietary solution. But, to control the action of epinephrine, insulin alone might not help as a main anabolic direction to cure. The cytoplasmic serine-threonine kinase (S6K1) is critical for transitional regulation of genes that encode essential components of the protein synthesis apparatus. The protein and insulin administration regulate S6K1 activity in skeletal muscles (Bigot *et al*., 2003)[30]. Fernandez *et al*., (1995)[31], for instance, studied the effect of epinephrine administration on the metabolism of red and white muscle and found it inducing plasma metabolites with overall significant glycogen depletion.

In conclusion, this study may prove another biological parameters for death other than loosing BW. This is true in our data. Death, however, is not connected only with losing fat free bodies as stated before (Elia, 1991)[4]. According to the data revealed here, another biological consequences are more accurate. A metabolic systemic deviation must be carried out due to unusual type of hard work that temporarily takes place or carried out under this type of stress. According to these findings, a protein catabolic pathway (EMS) that runs for longer time is the real reason for death occurring in groups 3 and

4. These proteins turn over in such high rates beyond the biological capacity of both intracellular and extracellular compartments negatively affect the whole biological system including cells, tissues, organs, and blood chemistry as well. Again, It is clear that this emergence catabolic status of proteins save the animals in short run, but its existence for longer time may posses a reverse effect. This EMS again is a sort of hormonal oxidative imbalance, which abuses the biological system when it existed for long time. Considering that, a risk to special biological factor other than loosing weight is to be followed. Metabolically, the hypothalamic noradrenaline (NE) turnover, as estimated by the concentration of the major metabolite MHPG, was significantly decreased in semistarved sedentary rats compared to controls. Hyperactivity resulted in marked elevation of NE turnover at all time points examined. Semistarvation-induced decreases of dopamine (DA) turnover as estimated by the concentrations of its major metabolite DOPAC, could also be compensated by hyperactivity. The circadian pattern of NE turnover parallels the pattern of running activity. MHPG levels at times of high activity were even higher than in controls fed ad libitum ($p<0.01$) (Broocks, *et al*., 1989)[32]. The availability of the precursor tyrosine, as indicated by the ratio of plasma tyrosine to the large neutral amino acids, was significantly decreased in semistarvation ($p<0.0001$); hyperactivity caused a further decrease ($p<0.001$), indicating that tyrosine availability is not, under these conditions, a limiting factor for noradrenaline turnover. The combined influence of semistarvation and hyperactivity on central catecholamine turnover in the rat is discussed as an animal model for the effects of malnutrition and heavy exercise often observed in

anorexia nervosa (Broocks, *et al*., 1989)[32].

Actually, this hormonal oxidative imbalance induced by severe long existed hungers can not be corrected without a sort of an urgent dietary therapy. For example, Rodriguez *et al*., (2002)[33] found that olive oil induced an up regulating effect on uncoupling protein gene mRNA that was probably not mediated by systemic metabolic changes, but rather related to local effect on interscapular brown adipose tissue and skeletal muscle. Moreover**,** de Jonge, *et al*., (2002)[34] studied the genetic expression effect of food component. Overexpression of arganise I in enterocytes of transgenic mice has been found to elicit a selective argenine deficiency and affects skin, muscles, and lymphoid development.

The death due to long time of starvation, therefore, may involves immune dysfunction deposited in accordance to the hormonal imbalance and the fallen down of organ expectancy. Liver, in particular, is a real scientific adjunct to that issue due to its role in protein synthesis. Undernutrition compromises barrier function, allowing easier access by pathogens, and compromises immune function, decreasing the ability of the host to eliminate pathogens once they enter the body. Along with undernutrition, infection is becoming the primary cause of morbidity and mortality in the developing world. Complex interactions are existed between these two threatening problems (Calder and Jackson, 2000)[35].

Further investigation should be designed to accurately help and secure hunger people. More accurate investigation is conducted to biologically explore this metabolic conjugation, in which, more metabolic pathways are observed, besides, until that, new dense diet of proteins and antioxidants from animal and plant origin are being tailored.

REFERENCES

1. Guthre HA. Junk food. Mosby book, 2nd ed 1993, page 17. Perspectives in nutrition, a book, 2nd ed. USA: Wordlow and Insel; 1993.
2. Keys A, Brozek J, Henschel, A, Taylor HL. The biology of human starvation. Vol 1, PP 81- 535. Minnesota Press; 1950.
3. Forbes, JB, Drenick, EJ. Loss of body nitrogen on fasting. Am J Clin Nutr. 1979; 37: 1570-4.
4. Elia M. Effect of starvation and very low calorie diets on protein energy interrelationships in lean and obese subjects. Proceeding of IDECG, Oct. 1991.
5. Harbison ST, Chang S, Kamdar KP, Trudy FC. Quantitative genomics of starvation stress resistance in *Drosophila*. Gen Biology. 2005. 6,4.
6. Mackay K. Efficacy of whole-genome transcriptional profiling for identifying genes affecting starvation stress resistance. Genome. 2005; 11, 4
7. Lee CK, Klopp RG, Weindruch R, Prolla TA. Gene expression profile of aging and its retardation by caloric restriction. Science. 1999; 285:1390-3.
8. Lee CK, Weindruch R, Prolla TA. Gene-expression profile of the aging brain in mice. Nat Genet. 2000; 25:294-7.
9. Schadt EE, Monks SA, Drake TA, Lusis AJ, Che N, Colinayo V, *et al*. Genetics of gene expression surveyed in maize, mouse and man. Nature. 2003; 422:297-302.
10. Teichert U, Mechler B, Muller H, Wolf DH. Lysosomal (vacuolar) proteinases of yeast are essential catalysts for protein degradation, differentiation, and cell survival. J Biol Chem. 1989; 264:16037-45.
11. Zinke I, Schutz CS, Katzenberger JD, Bauer M, Pankratz MJ. Nutrient control of gene expression in Drosophila: microarray analysis of starvation and sugar-dependent response. EMBO J. 2002; 21:6162-73.
12. Nelson JF, Karelus K, Bergman MD, Felicio LS. Neuroendocrine involvement in aging: Evidence from studies of reproductive aging and caloric restriction. Neurobiol Aging. 1995; 16:837-43.
13. Seglen PO, Bohley P. Autophagy and other vacuolar protein mechanism. Experientia. 1992 48:158-72.
14. Ahmed AIS, Abo Zeid FKS, El-Bagoury AA. "Hormonal Oxidative Balance of Modified Diets and Uncontrollable Cholesterol". Egy J of Biomedical Sciences. 2003;13, 106-15.
15. Svanberg E, Ohlsson L, Lundholm K. RhIGF-I/IGFBP-3 complex, but not free rhIGF-I, supports muscle protein biosynthesis in rats during semistarvation. European Journal of Clinical Investigation. 2000; 30(5): 438-46.
16. Debnam ES, Levin RJ. Effects of fasting and semistarvation on the kinetics of active and passive sugar absorption across the small intestine in vivo. J Physiol. 1975; 252(3): 681–700.
17. Holemans K, Van Bree R, Verhaeghe J, Meurrens K, André Van F. Assche F. Maternal Semistarvation and Streptozotocin-Diabetes in Rats Have Different Effects on the In Vivo Glucose Uptake by Peripheral Tissues in Their Female Adult Offspring. The Journal of Nutrition. 1997; 127(7): 1371-6
18. Ahmed AIS, Sayed-Ahmed EF, Bayomey AM, El-Deeb F. Some biological aspects in consequence of starvation in rats. J Agric Sci Mansoura Univ. 2006; 31(12): 7765-74.
19. Lane–Peter W, Pearson AEG. Dietary requirement in the laboratory animal, principles and practice. London: Acad Press; 1971.

20. Chapman DG, Castilla R, Campbell JA. Evaluation of protein in foods. Can J Biochem Phys. 1959;37:679-86.
21. Dacie JV, Lewis SM. Practical hematology. UK: Churchill Livingston; 1984.
22. Richmond W. Enzymatic calorimetric method for determination of cholesterol. Clin Chem. 1973;19: 1350.
23. 0Richman A, Frankely S. Calorimetric determination of AST and ALT. Am J Clin Path. 1957; 28: 56.
24. Frankel S, Reithman S. Clinical Determination of AST and ALT. Am J Clin Path. 1963;28:102.
25. Davies HJA, Baird KL, Howard AN. Metabolic response of low and very low calorie diets. AM J Clin Nutr. 1984;49, 745-51.
26. Gottesman S, Maurizi MR. Cell Biology: Enhanced: Surviving Starvation. Science. 2001; 293(5530): 614 –5.
27. Fryburg DA, Gelfand RA, Barret EG. Growth hormone (GH) acutely stimulates forearm muscle protein synthesis in normal humans. Am J Physiol Endocrinol Metab. 1991; 260, E499-E504.
28. Ahmed AIS, Habbib GM. Hammad AA, El-Deeb F. Effect of natural plant protein digest on hyperlipemia and hyperglycemia in some animal models. Egy J Biomed Sci. 2005;17: 85-94.
29. Haub MD, Wells AM, Tarnopolsky MA, Camppbell WW. Effect of protei source on body composition and muscle size in older men. Am J Clin Nutr. 2002; 76, 511-7.
30. Bigot K, Taouis M, Tesseraud S. Refeeding and insulin regulate S6K1activity in skeletal muscles. J Nutr. 2003;133, 369-73.
31. Fernandez X, Levasseur P, Ecolan P, Wittmann W. Effect of epinephrine administration. J Sci Food Agric. 1995;68: 231-9.
32. Broocks AJ, Liu I, Pirke KM. Semistarvation-induced hyperactivity compensates for decreased norepinephrine and dopamine turnover in the mediobasal hypothalamus of the rat. J Neural Transmition. 1989;79(1-2):113-24.
33. Rodrigues VM, Portillo MP, Pico C, Macarulla MT, Paloa A. Olive oil feeding up-regulates uncoupling protein genes in rat brown adipose tissue and skeletal muscle Am J Clin Nutr. 2002; 75:213-20.
34. de Jonge WJO, Hallemeesch MM, Kwilkkers KL, Ruijter JM. Overexpression of arganise I in enterocytes of transgenic mice elicits a selective arganise deficiency and affects skin, muscles, and lymphoid development. Am J Clin Nut. 2002; 75:128-40.
35. Calder, PC, Jackson, AA. Undernutrition, infection and immune function. Nutr Res Rev. 2000;13, 3-29.