Impact of Aging on Some Hormonal Parameters in Human Males

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Abstract

Substantial and important age related change occur in the several hormones such as TSH, T3, T4, cortisol, and testosterone. Physiological aging is an extremely complex, Multiofactorial process, affecting a myriad of genetic, biochemical, and metabolic processes. The present study conducted to evaluate the hormonal disturbances that occurring at advanced age stages. The results of thyroid hormones, showed a significant decrease ($P<0.05$) in the levels of thyroid stimulating hormone (TSH) with significant increase. ($p<0.05$) in the levels of triiodothyronine (T3) and levels of tetraiodothyronine (T4) in aging human males in a comparison with young human males. Concerning the levels of testosterone and cortisol hormone showed a significant decrease ($P<0.05$) of aging human males when compared with young human males. Antioxidants levels (such as reduced glutathione and catalase), also involved in this study and their results reported significant decrease ($P<0.01$) of aging males human when compared with young human males. Data obtained from this study may attributed that at advanced age may be the DNA damage causes the cells to stop dividing or induce apoptosis, often affecting stem cell pool and hence hindering regeneration. Thus, DNA damage is thought to be the common path way causing aging , Hormones and cancer.
Key words: Hormones , Cancer, Aging.

الخلاصة

تحصل العديد من التغيرات الكبيرة والمهمة للعديد من الهرمونات عند التقدم بالعمر خاصة الهرمون المحفز للدرقية (TSH) والهرمون ثلاثي اليود (T3) والهرمون رباعي اليود (T4) والكورتزول والهرمون الذكري التستوستيرون. يعد التقدم بالعمر من اهم التغيرات الفسيولوجية الحاصلة في جسم الكائن الحي وبالاخص الانسان. اذ تحصل العديد من التغيرات في العمليات الايضية والكيموحيوية والوراثية. هدفت الدراسة الحالية الى تقييم مستوى بعض الهرمونات لدى ذكور الانسان المسنين ومقارنتها مع مجموعة من الذكور الشباب الاصحاء. تضمنت هذه الدراسة فحص مستوى الهرمون المحفز للغدة الدرقية (TSH) اذ سجل انخفاضاً معنوياً ($P<0.05$) في الرجال المسنين في حين سجلت قيم كل من هرموني T3 و T4 ارتفاعاً معنوياً ($P<0.05$) في الرجال المسنين عند مقارنتها مع الرجال الشباب. بينت قيم كل من هرمون ألكورتزول وهرمون التستوستيرون انخفاضاً معنوياً ($P<0.05$) في الرجال المسنين عند مقارنتها مع الرجال الشباب. كذلك لوحظ انخفاضاً معنوياً في قيم بعض مضادات الاكسدة (Anti-oxidants) وهي الكلوتاثيون المختزل (GSH) و انزيم الكاتليز (CAT) انخفاضاً معنوياً ($P<0.01$) في الرجال المسنين عند مقارنتها مع الرجال الشباب. ان النتائج الموضحة في اعلاه والمستخلصة من هذه الدراسة تعزى الى ان التقدم في العمر يؤدي الى تحطم الحامض النووي منقوص الاوكسجين (DNA) مؤدياً بدوره الى توقف انقسام وتجدد الخلايا او يؤدي الى الموت الخلوي المبرمج (apoptosis)، ولذا يعد تحطم الحامض DNA العامل الرئيسي في حدوث التقدم في العمر ، الهرمونات وحصول السرطان.

الكلمات المفتاحية:الهرمونات، السرطان، التقدم بالعمر

Introduction

Ageing (British English) or aging (American English) is the accumulation of changes in a person over time (Browen and Atwood, 2004). Numerous aging theories have been proposed to explain mechanisms of aging, no theory has been more lasting in this regard than that known as the "free radical theory of aging" in which damage by free radicals (reactive oxygen species [ROS]) is deemed critical in determination of life span (Beckman and Ames, 1998; Harman, 2003). Multiple changes occur with aging such as decrease in body mass index, osteoporosis, mass of skeletal muscles,and metabolism, and these changes were associated with endocrine disturbances (Mariotti *et al.,* 1995). The hypothalamic-pituitary-adrenal (HPA) axis is and auto-regulating system with many modulatory mechanisms, due to such regulation, the circulating levels of glucocorticoids are highly variable, according both

the spontaneous rhythmic fluctuations and to the responses towards stressful conditions (Mcewen, 1998).

Materials and Methods:-

Subjects of the study

The subjects of the present study included 70 males divided into two groups according to age: first group their age range between 55-66 years and second group their ages ranged between 20-35 years. All subjects were recruited from Babylon governorate and republic health laboratory. All subjects of this study were free from chronic diseases such as diabetes, hypertension, thyroid disorders and others.

Blood collection

Venous blood was collected at time (10 am) in plain tubes (without anticoagulants) and then the tubes put in centrifuge at speed 1000g for 15g minutes. The Buffy coat was removed (Known sample of serum was removed and kept in epindrouph tubes in deep freez (-20)°C to achieve the hormonal analyses. The packed cell were washed with normal saline. Erythrocytes were lysed with hypotonic solution phosphatic buffer (pH 7.5) and the hemolysat Sample was separated by centrifugation at 2500g for 15 minutes.

Determination of hormones:-

The levels of serum thyroid stimulating hormone (TSH), total triiodeothronine (T3), and total thyroxine (T4) were estimated by using enzyme immunoassay (EIA) methods (according to kits supplied by Biocheck, Inc). Testosterone and cortisol hormones were measured by using the instruments of the VIDAS (according to kits supplied by Biomeoiexu).

Measurement of reduced glutathione (GSH):

GSH content of plasma was determined by the method of Ellman's (Ellmans,1959). This method involved plasma 1.0 ml was treated with 0.5ml of Ellman's reagent (19.8mg) of 5.5 dithiobisnitro-benzoic acid (DTNB) in 100ml of 0.1% sodium citrate) and 3.0ml of phosphate buffer (0.2M, pH 8.0). The colour intensity was read at 412nm.

Determination of Catalas (CAT):

Catalase (CAT) was measured colorimetrically at 620nm and expressed as μmol of H_2O_2 consumed min/mg/Hb as described by Shina's method (Shine, 1972). The reaction mixture (1.5ml) contained 1.0 ml of 0.01 mole pH 7.0 phosphate buffer,0.1ml of hemolysate, and 0.4ml of 2mole H_2O_2. The reaction was stopped by addition of 2.0ml of dichromate-acetic acid reagent (5% potassium dichromate and glacial acetic acid were mixed in 1:3 ratio).

Statistical analysis

Data of the present study were statistically analyzed by student's t-test using SPSS version 10.0. The data were expressed as mean ±SD of the number of experiments (Daniel, 1999).

Results

The results which are obtained from this study and illustrated in the following table (1), showed a significant decrease ($P<0.05$) in the levels of thyroid stimulating hormone (TSH), cortisol, and testosterone in aging human males when compared with young human males. Also, this study reported a significant decrease ($P<0.01$) in the levels of reduced glutathione and catalase in aging males when compared with young males. As for, the levels of triiodothyronine (T3) and thyroxine (T4) showed significant increase ($P<0.05$) of aging human males when compared with young human males.

Table (1): Means of age, body mass Indix (BMI), thyroid stimulating hormone (TSH), Triiodothyronine (T3), tetraodothyronine (T4), cortisol hormone, testosterone hormone, reduced glutathione (GSH), and catalase in aging human males.

Parameter	Control group	Test group
Age	28±5	62±6
BMI	25±2	22±4
TSH (MIU/ml)	2.51±1.51	*1.9±1.01
T3 (ng/ml)	0.97±0.41	*1.8±0.3
T4(ug/ml)	10.02±3.12	*12.7±0.71
Cortisol (ng/ml)	115.81±10.52	*97.5±8.5
Testosteron (ng/ml)	10.71±2.51	*7.9±1.71
GSH (mg/dL)	42.78±6.71	**22.95±3.51
CAT (U/mgHb)	70.58±4.51	**55.95±6.42

-Results are means ± SD

-Results with one a strikes (*) are significantly at ($P<0.05$), and results with two a strikes (**) are significantly at ($P<0.01$).

Discussion

The present study aimed to determine the impact of aging on the essential hormones of the body such as (TSH, T3, T4, cortisol, and testosterone). Adequate thyroid function is essential for normal development and retention of cognitive function throughout life. Several changes in thyroid function occur during aging. It had been found that normal aging is associated with changes in thyroid hormone production and metabolism (Mariotti *et al.,* 1995; Davis *et al.,* 2003).

Data obtained from this study were consistent with previous studies (Murialdo *et al.,* 1993; Mariotti *et al.,* 1995). These studies indicated that the secretion of T3 and T4 is reduced in healthy elderly (61-90 years old), but serum concentrations of total and free T4 remain relatively unchanged because T4 degradation is also reduced in the elderly. Also it had been found that circulating total and free T3 concentrations demonstrate a clear, age-dependent decline because of both reduced secretion and reduced Peri pheral conversion of T4. Serum reverse T3 (rT3) seems to increase with age. The decrease in serum T3 levels together with the increase in serum rT3 may indicate a decrease in peripheral hepatic metabolism of iodothyronine during aging. (Mariotti *et al.,* 1993).As for data of TSH during healthy aging are controversial. Some studies showed unchanged TSH concentration while other showed significant increased TSH in both men and women (Murialdo *et al.,* 1993). On the basis of physiological point view, that T3 exerts negative feed back mechanism on the pituitary gland to inhibit the secretion of TSH and a result of decrease T3, this mechanism become low and the secretion of TSH become a high . Results of the present study sowed a significant decrease ($P<0.05$) of the levels of cortisol hormone in aging human males. Cortisol is glucocorticoid and synthesized by adrenal gland. Its primary functions are to increase protein breakdown, inhibit glucose uptake, and increase lipolysis. The level of serum cortisol is affected by many factor such as intensity, duration, and timing of exercise, age, altitude, environmental temperature and psychology (Bernet and Wartofsky, 2000; Minton and Parsons, 1993). The previous studies found significant morphological and functional changes affect with age the (HPA) axis at different with age the experimental animals and in human beings (Mcewen, 1998; Anisman, 1993).In humans, the amount of cortisol present in the blood undergoes diurnal variation, the level peaks in the early morning (approximately 8am) and reaches its lowest level at about mid night (4am), or three to five hours after the onset of sleep (Weerth *et al.,* 2003).

Wilkinson *et al.,* (1997) suggest that the primary defect in allostatic system in aging might be prolonged response to stressful conditions, due to the in ability to sutoff the allostatic response after the end of the stress. However, inspite of the subtle age-related changes of HPA function, the circulating levels of glucocorticoids show a relative constancy or even a trend toward an increase with aging. Indeed, the blood levels of both cortisol and ACTH usually fall within the normal range in physiological aging (Vancauter *et al.,* 1996).A majority of studies have shown that basal circulating levels of cortisol do not vary significantly with aging human. Few studies have reported decreased corisol levels with aging (Drafta *et al.,* 1982; Seem *et al.,* 2000).

The present study reported a significant decrease ($P<0.05$) in the levels of testosterone hormone in aging males in a comparison with young males. Deficiencies in sex steroids, resulting from either menopause or castration, lead to increased bone remodeling and osteoprosis. The mechanisms responsible for this phenomenon are not entirely understood. Sex steroids many suppress the production of cellular cytokines such as interlukin-6 (IL-6) and down regulate the expression for both subunits of its receptors in osteoblasts (Girasole *et al.,* 1995; Lin *et al.,* 1997).Also, it is well recognized as men age, plasma testosterone concentrations decline gradually after age 40 (Kaufman and Vermeulen 2005). Subsequent studies have confirmed progressive loss of testosterone with aging in healthy men. These studies showed that the decline in plasma testosterone concentration is associated with increase sex hormone binding globulin (SHBG) levels (Feldman *et al.,* 2002), a major plasma carrier of testosterone, resulting in even more dramatic decrease in unbound free testosterone (Harman, 2005).More recently, it has been found that low circulating testosterone is associated to age related metabolic abnormalities including body wide reductions in rates of protein synthesis, abdominal obesity, diabetes, prediabetes (insulin resistance), impaired glucose tolerance, and increased risk of cardiovascular diseases (Araujo *et al.,* 2007).Also, decreased testosterone availability in aging has been associated with parallel age related decline in bone density, muscle mass, muscle strength, physical function, and sexual activities (Lin *et al.,* 2005). Several changes for the loss of Lydig cells function has been proposed including: as a) reduction in Lydig cell number; b) a bnormal number of cells, each having defects in one or more enzymatic steps involving testosterone biosynthesis; c) a normal number of cells with reduced responsiveness to tropic hormone (Lutenizing hormone); and d) Leydig cell degeneration and dissolution (Swerdloff and Herber, 1984).The present study pointed out asignificant decrease ($P<0.01$) in the level of reduced glutathione and catalase. Oxygen (O2) is essential to the life of aerobic organisms. However, its metabolits represent a potential threat to all living organisms indeed, O2 is metabolized in animal tissue by successive reductions in superoxide anion ($O_2^{\bullet -}$), hydrogen peroxide (H2O2), and hydroxyl radical (OH). These different metabolites are called reactive oxygen species (ROS). (Nishigori *et al.,* 2004; Zwa *et al.,* 2006).Previous studies had been found that aging is slowed by calori or protein restriction and these studies confirmed that marked effect on aging and cancer is becoming clearer and may in good part be due to reduced oxidative damage. The suggestion that maintenance functions are enhanced in calorie-restricted rats, thus resulting in less oxidative damage, is supported by the findings of more efficient DNA repair,better coupled mitochondrial respiration, and delay in the age dependent decline of some anti-oxidant defenses(Ames *et al.,* 1993).

Also, in vivo and in vitro studies showed that senescence is associated with increased oxidant generation, a decline in robustness of cellular defenses and repair, and an accumulation of the end products of the oxidative damage (Bokov *et al.,* 2004). Cells

of the body are continuously damaged by prolonged oxidative stress that far exceeds the capacity of the body's cells to synthesize antioxidant molecules or to synthesize them from extracellular sources (Komosinska-Vasser *et al.,* 2000).Other studies showed that a decrease in production of sex hormone such as in menopausal women could predispose the women to higher levels of reactive oxygen species (Signorelli *et al.,* 2006). Gredilla *et al.,* (2001) who reported that excess thyroid hormones induces oxidative stress and also resulted in a higher levels of oxidized glutathione (GSSG) to reduced glutathione (GSH).In conclusion, the present study suggests that the aging effects on most body physiological markers, and attribute these changes as a result of DNA damages by dangerous reactive oxygen species.

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