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STUDY THE RELATIONSHIP BETWEEN GHRELIN LEVEL AND SOME BIOCHEMICAL MARKER IN ANBAR CITY

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ABSTRACT

The results showed that the sex of the patients in the four groups did not reveal any statistically significant differences in the averages of sex (males and females) between the two groups (controlling healthy people). The data showed that there was a significant difference between the averages of the gum disease patients, and the average age of the patients in the four groups included ranged the study ranged from 24.75 years in patients with obesity without the gum disease group to 32.55 years in patients with obesity and the gum disease group, and the data showed that ghrelin parameters varied in their effect on obesity and gum disease, which appeared in their values for the four tested groups studied. People suffering from obesity and gum disease showed the highest significant value in the level of ghrelin, as it was recorded at 189.90 mol/ml compared to 119.30 mol/ml for the control group. The results revealed that all groups of patients had a significant increase in the concentration of T3 compared to the healthy control group, and the patients also Those suffering from obesity without gum disease had a significant increase in T3 levels, which recorded 97.60 ng/dL with a standard deviation of 19.56, compared to 92.40 ng/dL with a standard deviation of 20.17 for the control group. The group of patients suffering from obesity without gum disease showed a low T4 level value, recording 6.96 micrograms/dL, while the control group recorded 7.73 micrograms/dL. The data showed that there was a significant difference in the average TSH level between the studied groups, as the TSH level in patients suffering from obesity and gum disease increased significantly, recording mIU/L with a standard deviation of 1.11 compared to 1.74 mIU/L with a standard deviation of 1.21 for the control group.

Keywords: Obesity, Ghrelin, TSH, gum disease.

INTRODUCTION

Obesity is a huge public health issue that affects people all over the globe. Since 1975, the prevalence of obesity has climbed by a factor of three, and this trend is expected to continue into the foreseeable future (Engin 2017). In spite of the fact that obesity is defined as an imbalance between a person's energy expenditure and the amount of food they consume, there are a number

of variables, both hereditary and non-genetic, such as environmental and behavioral factors, that contribute to obesity. Because of the inclusion of hereditary variables in addition to hormones, there may be a 40–70% inter-individual variation in body mass index (BMI) within a single family. This is linked to the fact that hormones have a role in the regulation of body weight (Ogden *et al.* 2016). The primary activities of ghrelin in the body are the release of growth hormone, the stimulation of hunger and food intake, the manipulation of the production of gastric acid and motility, and the control of the neuroendocrine and exocrine secretions of the pancreas. Ghrelin also has a role in the motility of the gastrointestinal tract. Ghrelin is not only expressed in the stomach, but also in many other tissues (Ward *et al.* 2019). The body mass index, often known as BMI, is a number that is calculated by applying a formula to a person's weight (in kilograms) and height (in centimeters). Because of the inclusion of hereditary variables in addition to hormones, there may be a 40–70% inter-individual variation in body mass index (BMI) within a single family. This is linked to the fact that hormones have a role in the regulation of body weight (Ogden *et al.* 2016).

MATERIALS AND METHODS

Sample colection

Blood samples were collected from the veins of patients or individuals who recurred Ramad Teaching Hospital in Anbar Governorate, Iraq for hematological investigations that includes thyroid function test (T3, T4, and TSH), besides the estimation of amylase, and ghrelin enzymes levels in obesity patients with periodonatal disease.

Estimation of ghrelin in blood serum

We were labelled of the assay tubes in the following order:

1, 2 Total counts (TC).
3, 4 Non specific bindings (NSB).
5, 6 Zero calibrators (B_0).
7-18 Calibrators 1 to 6.
19, 20 Control C1.

Amylase activity assay

This assay is suitable for the colormetric detection of amylase activity in plasma or serum blood samples by using the amylase activity assay kit (Mak009). Amylase activity is measure using a

coupled enzymatic assay which results in a colormetric product at 405 nm. Proportional to the substrate amount, ethylidene –pNP-G7 is elevated by the enzyme amylase.

Estimation of T3, T4 and TSH levels

Total T3 ELISA kit is a solid phase enzyme-linked competitive immunoassay for the measurement of T3 in human serum.

The total T3 ELISA kit is consist of two key components:

1- Solid microwells pre-coated with T3 analog.
2- Liquid conjugates containing anti-T3 antibody.

The essential reagents for a solid phase enzyme immunoassay T4 include immobilized antibody enzyme- antigen conjugates and native antigen. Upon mixing immobilized antibody enzyme-antigen a competation reaction results between the native antigen and the enzyme – antigen conjugate for a limited number of insolubilized binding sites.

The thyroid stimulating hormone (TSH) ELISA is intended for the quantitative measurement of TSH in human serum. It is a solid phase sandwish ELISA method. The samples and anti-TSH – HRP /Biiotin cinjugate are added to the wells coated with streptividin. TSH in the sample forms a sandwish between two specific antibodies to TSH unbound protein and HRP conjugate are washed off. Upon the addition of the substrate, the intensity of color is proportional to the concentration of TSH in the sample.

Components
Microwells coated with streptividin
TSH standard 7 vials 0.5 mL
TSH conjugate reagent
TMB substrate
Stop solution

Statistical Analysis

SPSS (Statistical Package for Social Science – version 23) is use for statistical analysis. The results are present as mean SE (standard error). The significance of the difference is determine using a

two-sample t-test for two independent means and a paired t-test for two dependent means. For the relationship between two quantitative variables, correlation and regression are use, with P 0.05 set as the lowest level of significance.

Results

4.1 Biochemical Parameters of the Studied Groups

The study included some chemical tests and age, and it was noted that the study variables achieved statistical differences when compared to the control group. The study included age for patients suffering from obesity, average weight, and levels of ghrelin, amylase, and insulin.

4.2 The Gender of Patients with Obesity

The results showed the gender of patients of the four groups revealed no significant differences except group three of gender (males and females) between the groups (control healthy individuals, patients without obesity, periodontal disease, patients with obesity and periodontal disease, patients with obesity without periodontal disease which recorded (as a recorded in Table 1.1) for males and females respectively and Figure 4.1. These results were disagreement with the results of Jain and Bhavsar (2021) which found that the clinical parameters of periodontal disease were higher in females than females, while Al-Abdaly *et al.* (2019) indicated that the periodontal cases were significantly higher sensitivity in females as compared with to male's patients with periodontal disease, they suggested that periodontal disease were related to subclinical atherosclerosis in males but not in females.

Table 1 The gender parameter and study groups

Group of study	N	Male	Female
Healthy individuals	50	26	24
persons with non- obesity with periodontal disease	50	28	22
persons with obesity and periodontal disease	50	32	18
persons with obesity without periodontal disease	50	24	26
Total	200	110	90

4.3 Ghrelin Level and Groups Studied

Data showed that ghrelin parameter varied in their effect on obesity and periodontal disease which seemed in their values of the tested four groups studied, anyway persons with obesity and periodontal disease revealed the upper significant value in ghrelin level which recorded 189.90 mol/ ml as compared with 119.30 mol/ mL for control group. While persons with obesity without periodontal disease group recorded lower value of ghrelin level which recorded 92.70 mol/ mL respectively (Figure 1). Ghrelin supplementing increase obesity prevalence due to its appetizing feature (Iwakura *et al.* 2007). The stimulating effects of appetite are thought to be mediated via the accurate nucleus of the hypothalamus and the messenger peptides neuropeptide Y and Agouti-related protein (Wynne *et al.* 2005).

Table 2 Ghrelin level and groups studied

Group of study	Mean (mol/mL)	N	Std. Deviation	Std. error of mean
Healthy individuals	119.3000	50	30.50988	6.82222
Persons with non- obesity with periodontal	193.3500	50	37.02812	8.27974
Persons with obesity and periodontal	189.9000	50	40.89383	9.14414
Persons with obesity without periodontal	92.7000	50	33.32156	7.45093
Total	148.8125	200	56.30322	6.29489

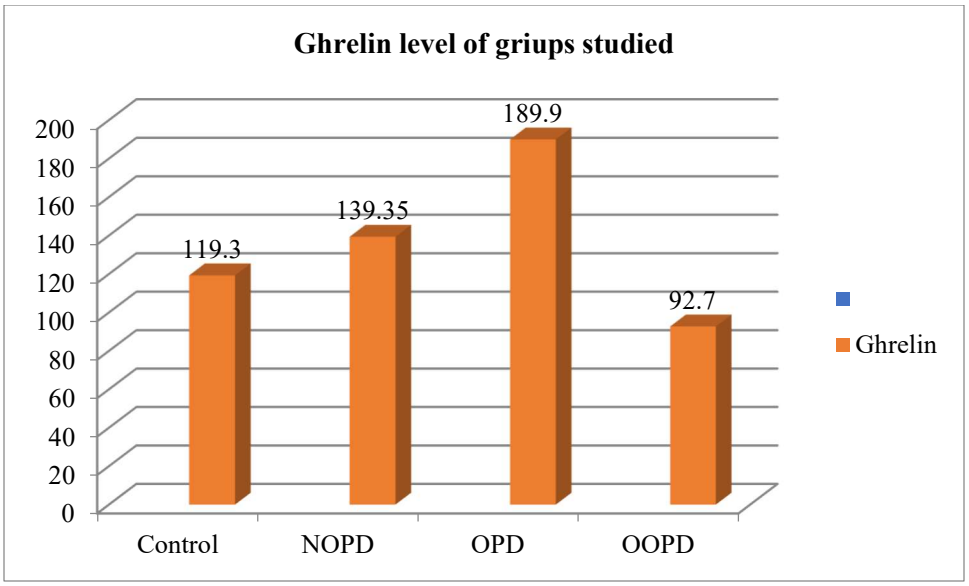

Figure 1 *Ghrelin level of groups studied*

4.4 Homa-IR Level and Groups Studied

The results showed that no significant differences between the mean of Homa-IR levels of the four groups studied while persons with obesity and periodontal group showed upper level of HOMA-IR which recorded 0.96 % as compared with 0.66 % for healthy individuals control group. Singh *et al.* (2013) found that the values of HOMA-IR were increased progressively from normal weight to obese adolescents in both genders, the mean of HOMA-IR values recorded 2.5% had a sensitivity of > 70 % and specify of > 60% for metabolic syndrome, while Qu *et al.* (2011) showed that the best cut-off of HOMA-IR for identifying those with insulin resistance is 3.80 and they found that 30.1% of persons in this Hispanic population with HOMA-IR > 3.80.

Table 3 HOMA- IR level of groups studied

Group of study	Mean	N	Std. Deviation	Std. Error of Mean
Healthy individuals	0.6661	50	0.39292	0.08786
persons with non- obesity with periodontal	0.7631	50	0.42251	0.09448
persons with obesity and periodontal	0.9633	50	0.88264	0.19736

persons with obesity without periodontal	0.9069	50	0.55383	0.12384
Total	0.8249	200	0.59591	0.06662

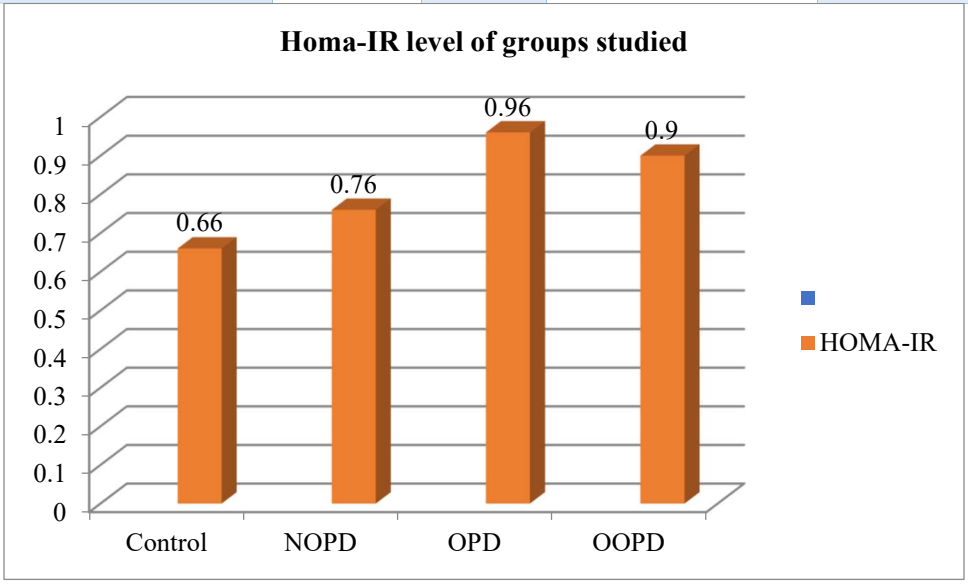

Figure 2 *HOMA-IR level of groups studied*

4.5 Amylase Activity of Groups Studied

The results pointed that no significant differences between amylase levels of the four groups studied, mostly patients' groups deceased the mean of amylase levels as compared with healthy control, patients with obesity and periodontal disease revealed upper decreasing in amylase level which recorded 48.85 U/L as compared with 65.95 U/L for control group (Figure 2). The deceasing in amylase values of patients with periodontal and obesity may be to the infection of the salivary glands or obese in patients may have decreased amylase concentration, the salivary amylase level is more associated with obesity and BMI. These findings were in agreement with the findings of Aldossari *et al.* (2019) which indicated that the amylase hormone activity was significantly lower in overweight individuals of obese groups of males and females and they concluded that the Saudi overweight and obese population seems to be at risk of low amylase activity which correlates with their obesity.

Table 4 Amylase activity of groups studied

Group of study	Mean U/L	N	Std. Deviation	Std. Error of Mean
Healthy individuals	65.9500	50	17.44608	3.90106
persons with non -obesity with periodontal	62.2000	50	13.80541	3.08698
persons with obesity and periodontal	48.8500	50	7.32174	1.63719
persons with obesity without periodontal	60.2500	50	18.24360	4.07939
Total	59.3125	200	15.91162	1.77897

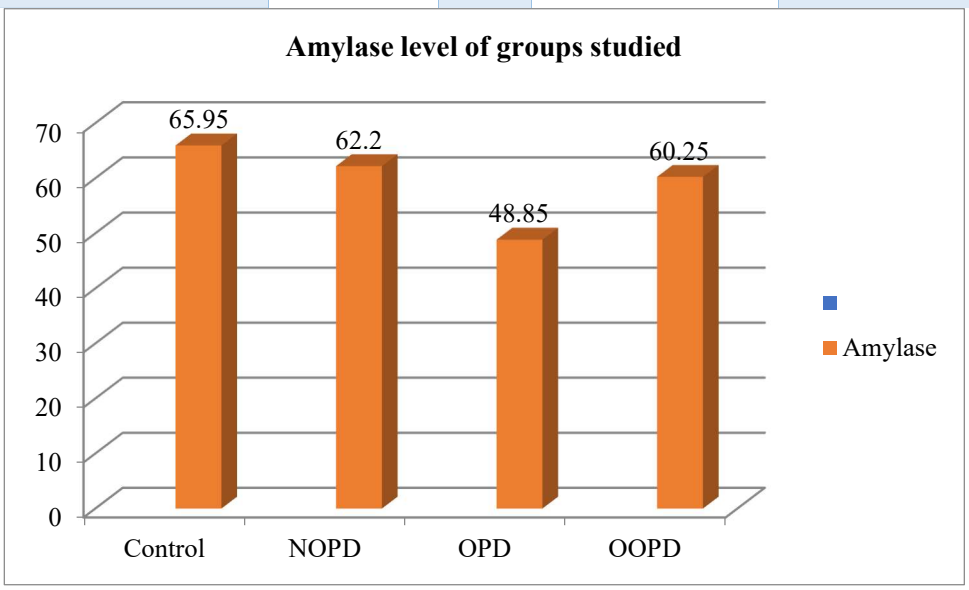

Figure 3 *Amylase level of groups studied*

4.6 T3 Level of Groups Studied

The results revealed all the patients' groups significantly elevated the T3 concentration as compared with healthy control group, patients with obesity without periodontal group significantly superior in increasing the T3 level which recorded 97.60 ng/dL with standard deviation 19.56 as compared with 92.40 ng/dL and standard deviation of 20.17 for control group (Figure 3). Little or moderate increasing inT3 level that is above normal range was described in obesity patients. This increasing is considered more as a result rather than stemming from obesity itself (Reinehr 2010).

Table 5 T3 level of groups studied

Group of study	Mean ng/dL	N	Std. Deviation
Healthy individuals	92.4000	50	20.17920
Persons with non-obesity with periodontal	96.8000	50	18.90586
Persons with obesity and periodontal	95.2500	50	15.77765
Persons with obesity without periodontal	97.6000	50	19.56743
Total	95.5125	200	18.43497

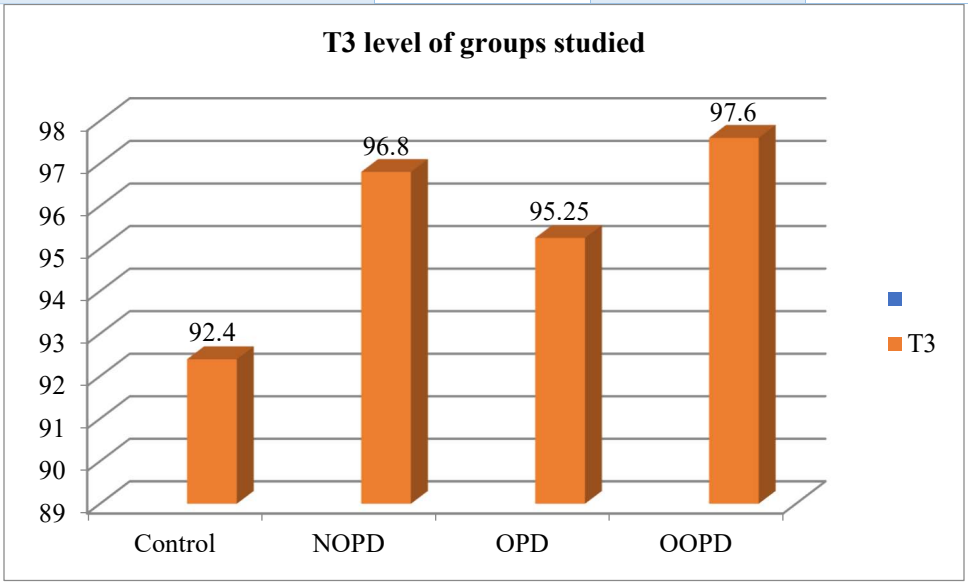

Figure 4 *T3 level of groups studied*

4.7 T4 Level of Groups Studied

The results indicated that there is a significant decreasing in level of T4 of patients groups as compared with healthy control group, patients with obesity without periodontal disease group showed lower value of T4 level which recorded 6.96 mcg/dL while control group recorded 7.73 mcg/dL (Figure 4). These results were in agreement with the results of Sosa-Lopez *et al.* (2021) which indicated that there is a negative correlation between serum T4 and obesity grade. Also, Al-Musa (2017) concluded that there is a negative correlation between T4 and BMI.

Table 6 T4 level of groups studied

Group of study	Mean mcg/dL	N	Std. Deviation
Healthy individuals	7.7377	50	2.16689
Persons with non-obesity with periodontal	7.4639	50	2.09582
Persons with obesity and periodontal	7.5842	50	1.95850
Persons with obesity without periodontal	6.9677	50	1.86101
Total	7.4384	200	2.00636

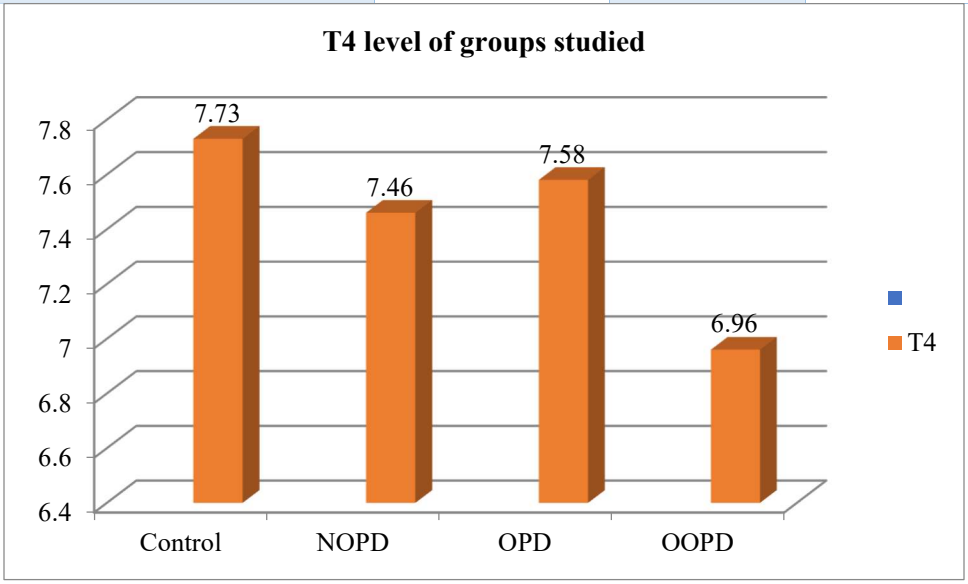

Figure 5 *T4 level of groups studied*

4.7 TSH Level of Groups Studied

Data of Table 4.25 showed there is a significant difference in mean of TSH level between the groups studied, patients with obesity and periodontal disease group significantly elevated the TSH level which recorded mIU/L with standard deviation 1.11 as compared with 1.74 mIU/L with standard deviation 1.21 for control group (Figure 5). Sania *et al.* (2017) found that there was significant uptrend in all parameters of periodontal disease, a significant decreased in TSH and T4 levels in patients with periodontal disease.

Table 7 TSH level of groups studied

Group of study	Mean mIU/L	N	Std. Deviation
Healthy individuals	1.7471	50	1.21626
persons with non-obesity with periodontal	1.5086	50	.94547
persons with obesity and periodontal	2.0665	50	1.11691
persons with obesity without periodontal	1.2270	50	.82015
Total	1.6373	200	1.06258

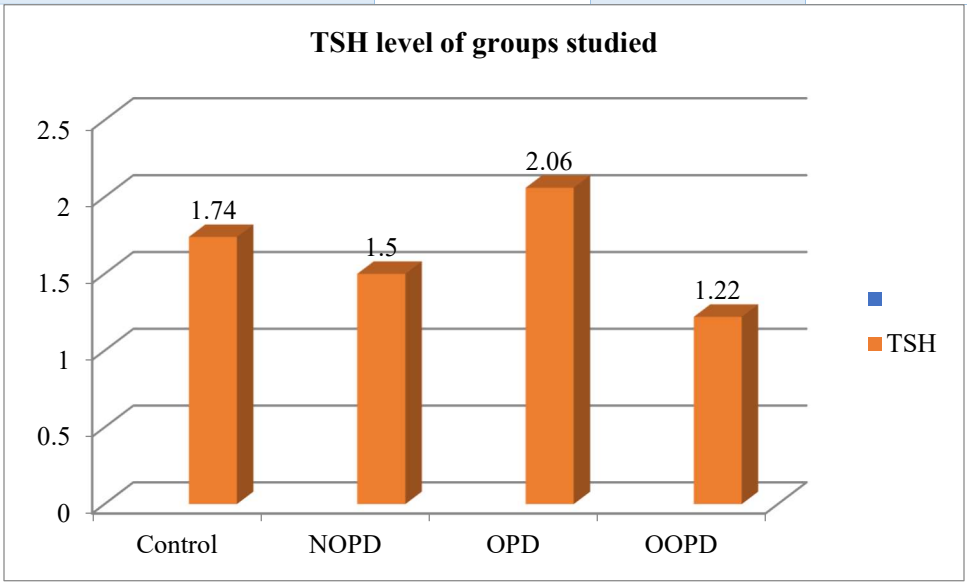

Figure 6 *TSH level of groups studied*

4.8 Conclusions

1. the gender of patients of the four groups revealed no significant differences in mean of gender (males and females) between the groups (control healthy individuals).
2. The results pointed that no significant differences between amylase levels of the four groups studied.
3. The results showed that there is a rise in the level of T3 with a decrease in the level of T4 in all groups of patients compared with the healthy control group, with a variation in the average level of TSH hormone between the studied groups.

4.9 Recommendations

1. The salivary ghrelin could be useful in diagnosis of obesity and salivary ghrelin may be a possible alternative to serum ghrelin for predicting obesity
2. Monitoring of the fast blood glucose level and BMI are positively correlated, and the patients are at a risk of obesity.

Reference

Al-Abdaly, M. A., Al-Qahtany, H. S. and Al-Qahtany, S. S. 2019. The impact of age and gender on severity and types of periodontal diseases among patients from two regions in Saudi Arabia, Open J. Stomatol., 9: 39-50.

Aldossari, N. M., El Gabry, E. E. and Gawish, G. E. 2019. Association between salivary amylase enzyme activity and obesity in Saudi Arabia. Medicine, 98(23): 130-145.

Al-Musa, H. M. 2017. Impact of obesity on serum levels of thyroid hormones among euthyroid Saudi adults. J. Thyroid Res., 10: 1-6.

Engin, A. 2017. The definition and prevalence of obesity and metabolic syndrome. Obesity and Lipotoxicity, 13: 1-17.

Iwakura, H., Akamizu, T., Ariyasu, H., Irako, T., Hosoda, K., Nakao, K. and Kangawa, K. 2007. Effects of ghrelin administration on decreased growth hormone status in obese animals. Amer. J. Physiolo. Endocrinol. Metab., 293(3): 819-829.

Jain, A. and Bhavsar, N. 2021. Role of gender and age in chronic periodontal disease. Int. J. Periodo. and Implantol., 6 (2): 117-125.

Ogden, C. L., Carroll, M. D., Lawman, H. G., Fryar, C. D., Kruszon-Moran, D., Kit, B. K. and Flegal, K. M. 2016. Trends in obesity prevalence among children and adolescents in the United States, 1988-1994 through 2013-2014. Jama, 315(21): 2292-2299.

Qu, H., Li, Q., Rentfro, A., Fisher-Hosh, S. and McCormick, J. 2011. The definition of insuiln resistance using HOMA-IR for americans of Mexican descent using machine learning. Plos One, 6(6): 1-4.

Reinehr, T. 2010. Obesity and thyroid function. Mil. Cell Endocri., 316: 165-171.

Sania, C., Krishan, K., Rajesh, K., Mohan, L., Anupama, B. and Sukanyu, M. 2017. Comparision of serum lipid and thyroid profiles before and after scaling and root planning in periodontitis subjects. Int. Med., 7(91): 1-4.

Singh, Y., Garg, M., Tandon, N. and Marwaha, R. 2013. A study of insulin resistance by HOMA-IR and its cut-off value to identify metabolic syndrome in urban indian adolescents. J. Clin. Res. Pediat. Endocrinol., 5(4): 245-251.

Sosa-Lopez, J., Alarcon-Gonzalez, P., Sanchiz –Hemandez, V., Cruz-Estrada, A., Aguilar–Serralde, C. and Velasco–Medina, A. 2021. Impact of obesity on the thyroid profile, long –term experience at the general hospital of Mexico, Dr. Eduardo Liceaga. Rev. Med. Hosp. Gen. Mex., 84(1): 4-10.

Ward, Z. J., Bleich, S. N., Cradock, A. L., Barrett, J. L., Giles, C. M., Flax, C. and Gortmaker, S. L. 2019. Projected US state-level prevalence of adult obesity and severe obesity. New England Journal of Medicine, 381(25): 2440-2450.

Wynne, K., Sarah, S. McGowan, B. and Bloom, S. 2005. Starling review appetite control. J. Endocrinol., 184: 291- 318.