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NOVEL METHOD THYROID HORMONE MEASUREMENT

Ari Tri Wanodyo Handayani
Department of Dental Public Health, Faculty of Dentistry, University of Jember,
aritri@ymail.com

INTRODUCTION

Iodine is an essential mineral of thyroid hormones produced by the thyroid gland. These are essential for life which therefore, makes iodine crucial. Although goiter is the most visible sequelae of iodine deficiency, the major impact of hypothyroidism due to iodine deficiency (1). There are 2 billion people are at risk of goiter due to insufficient intake of iodine. Nearly 266 million school-aged children world-wide have insufficient iodine intake (2). The school children formed representative study population for iodine status estimation as they represent community (3). Decreased thyroid hormones levels (hypothyroidism), by contrast, is associated with decreased metabolic rate. Most of these effects are due to the direct action of thyroid hormones on target tissues and direct actions on genes expression (4).

Thyroid stimulating hormone (TSH), secreted by the anterior pituitary in response to feedback from circulating thyroid hormone. TSH regulates iodide uptake mediated by the sodium/iodide symporter, followed by a series of steps necessary for normal thyroid hormone synthesis and secretion. Thyroid hormone is essential for normal development, growth, neural differentiation, and metabolic regulation (5).

Although the presence of thyroid hormone is crucial, it has not become part of a routine check up on the public service because it is costly. Central public health laboratory does not serve the thyroid hormone tests (6). Besides, blood test to measure thyroid hormone is considered quite painful. Lately, research on oral fluid (saliva) which can be used as the unit of test analysis emerged. saliva can be seen in many cases as a reflection of the physiological function of the body. There have been concerns about the use of saliva for diagnostic purposes due to its low concentration of analytes in comparison to blood (7). However, the examination of the thyroid hormones using saliva have not been used. The present investigation was aimed to analyze the thyroid hormone assays using saliva.

MATERIAL AND METHODS

The research was an experimental laboratory. The used animals were male Wistar rats aged 10-11 weeks as many as 21 rats which were divided into 3 groups. K was control group which received the 6-week standard food and drink. P1 was the first treatment group (iodine deficiency) which received PTU (6mg/kg/BW) using *intragastric intubation* method for 6 weeks. P2 was the second treatment group which received PTU (6mg/kg/BW) using *intragastric intubation* method for 6 weeks followed by *Levothyroxine* (10µg/100mg/BW) administration using the same method for 4 weeks. At the end of the study, saliva and blood of rats was taken and then the rats were sacrificed. The specimen were examined for the levels of T_3, T_4, TSH using ELISA procedure using Rat U-T3 (*Ultrasensitivity Triiodothyronine*) kit, Catalog No: E-EL-R1050 (*Elabscience*); Rat T4 (*Thyroxine*) kit, Catalog No: E-EL-R0981 (*Elabscience*); and Rat TSH (*Thyroid Stimulating Hormone*) kit, Catalog No: E-EL-R0976 (*Elabscience*). The data were collected and statistically analyzed using *ANOVA Mutivariate*.

RESULT

The results showed that levels of T_3 and T_4 and TSH in all groups are similar between serum and saliva.

Table 1. Mean and Standard Deviation of T_3 Hormone Level in Serum and Saliva (ng/mL)

Variable	Groups	Serum	Saliva	p-value
T_3	K	87.551 ± 4.980	89.201 ± 1.726	0.106
	P1	57.120 ± 9.488	58.201 ± 10.839	
	P2	87.713 ± 6.197	84.568 ± 4.193	

Table 2. Mean and Standard Deviation of T_4 Hormone Level in Serum and Saliva (ng/mL)

Variable	Groups	Serum	Saliva	p-value
T_4	K	78.604 ± 4.933	80.460 ± 1.076	0.213
	P1	60.181 ± 4.777	62.818 ± 7.607	
	P2	78.927 ± 6.176	79.081 ± 0.208	

Table 3. Mean and Standard Deviation of TSH Hormone Level in Serum and Saliva (ng/mL)

Variable	Groups	Serum	Saliva	p-value
TSH	K	7.519 ± 4.799	7.506 ± 0.587	0.688
	P1	31.882 ± 4.039	32.587 ± 3.659	
	P2	20.047 ± 2.922	23.343 ± 3.435	

Shown in the Table 1, Table 2 and Table 3 that T_3 hormone level in serum and saliva for all groups there was no significant difference ($p > 0.05$). Not different with T_4 and TSH hormones level in serum and saliva also showed the same level in all groups. These results are also presented in histogram form below.

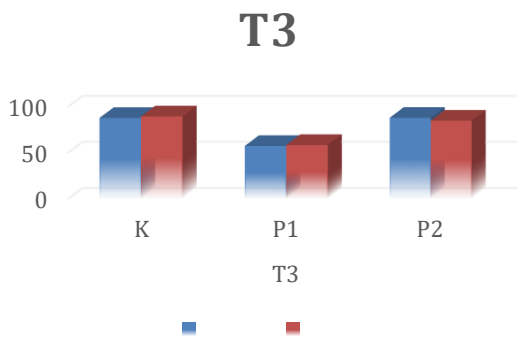

Figure 1. Mean and Standard Deviation of T_3 Hormone Level in Serum and Saliva (ng/mL)

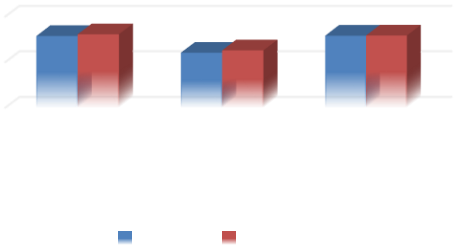

Figure 2. Mean and Standard Deviation of T_4 Hormone Level in Serum and Saliva (ng/mL)

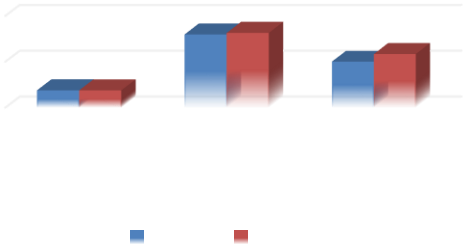

Figure 3. Mean and Standard Deviation of TSH Hormone Level in Serum and Saliva (ng/mL)

DISCUSSION

Based on Riskesdas 2013, IDD prevention program activities are now focused on household salt sample inspection, iodine urine test and examination iodine content of household drinking water. The result is 14.8% and 8.1% use less salt does not contain iodine and iodine. In children aged 6-12 years, the percentage of the risk of shortages and risk of excess iodine in 2013 tended to be more higher than in 2007. About 40.1% of drinking water consumed does not contain of iodine and 52% of drinking water contains little iodine (8). According to the Minister of Health stated that the prevalence of stunting in children under 2 years was 32.9% (9). One of the causes of stunting is iodine deficiency. The data indicate that people are at risk of suffered IDD due to iodine deficiency.

Iodine deficiency problems have not been followed by adequate laboratory facilities to measurement of thyroid hormone levels in the health care. Based on Minister Of Health, The Implementation Of Central Public Health Laboratory mentions that the ability of the laboratory examination at the health center include basic checks such as: a. Hematology: Hemoglobin, hematocrit, erythrocyte count, Count platelet, leucocyte count, Count type of leukocytes, ESR, b. Clinical chemistry: glucose, protein, albumin, total bilirubin, Direct bilirubin, SGOT, SGPT, alkaline phosphatase, uric acid, Urea / BUN, creatinine, triglycerides, total cholesterol, cholesterol HDL and LDL cholesterol. c. Microbiology and Parasitology: BTA, Diplococcus gram negative, Trichomonas vaginalis, Candida albicans, bacterial vaginosis, Malaria, microfilaria and Mushrooms surface. d. Immunology: A pregnancy test, blood group, Widal, VDRL, HBsAg, Anti Hbs, Anti HIV Antigen / antibody dengue. e. Urinalisa: macroscopic (color, clarity, odor, volume), pH, Density, Protein, Glucose, Bilirubin, Urobilinogen, Ketones, Nitrite, leukocytes, erythrocytes and Microscopy (sediment). f. Feces: Macroscopic, Blood faint and microscopy (6).). There is not include of thyroid hormones test.

Over the past forty years, improvements in the sensitivity and specificity of thyroid testing methodologies have dramatically impacted clinical strategies for detecting and treating thyroid disorders. In the 1950s, only one thyroid test was available an indirect estimate of the serum total. Since 1970, technological advances in radioimmunoassay and more recently spectrometry methodologies (10).

As shown in Table 1, Table 2 and Table 3 that T_3, T_4 and TSH hormones in all groups were not significantly different, it means thyroid hormone levels in serum and saliva almost equal. It was clear in Figure 1, Figure 2 and Figure 3. Saliva is critical for preserving and maintaining the health of oral tissues and has been used as a source of non-invasive investigation of metabolism and detection of disease. At present, saliva represents an increasingly useful auxiliary means of diagnosis. Many researchers have made use of sialometry and sialochemistry to diagnose systemic illnesses, monitoring general health, and as an indicator of risk for diseases creating a close relation between oral and systemic health. Health disorder can alter secretion and induces changes in various salivary components, such as immunoglobulins, hormones, lactate, proteins, and electrolytes (11). Recently, the utility and advantages of saliva as a screening tool for diagnostic diseases more potentially than serum. Like serum, saliva also contains hormones, antibodies, growth factors, enzymes, microbes and their products (12).

Interest in rapid and less invasive diagnostic tests has grown exponentially in the past decade, which has led to extensive research on saliva as a biological fluid for clinical diagnosis. Saliva has some advantages compared to blood and urine, two of the most used diagnostic fluids in laboratory setting. Saliva collection is easy and non-invasive requiring relatively simple instructions for collection and it possesses lower protein content, less complexity and varying composition than serum (13).

Salivary glands fulfil a huge range of functions in different species, and even amongst mammals there is great variety in salivary gland morphology and the control of salivation by nerves, reflectin adaptation to diet and environment (14). Saliva provides a “window” into the oral and systemic health of an individual, and like other bodily fluids, saliva can be analyzed and studied to diagnose diseases. With the advent of new, more sensitive technologies to detect smaller concentrations of analytes in saliva relative to blood levels, there have been a number of critical developments in the field that we will describe (15). All these characteristics make saliva an appealing diagnostic candidate for the detection and monitoring of several biomarkers in infants, children, adults and uncooperative patients.(12)

CONCLUSION

This study concluded that thyroid hormone measurement using saliva could as alternative and second opinion cause the rate of the hormones on serum and saliva are similar. Saliva can contribute significantly to disease screening, risk assessment, intervention evaluation, recurrence prediction and other prognostic outcome assessments. Progress in salivary diagnostics will also depend on establishing clinical utility of macromolecules and low molecular weight components. In this regard, salivary variations observed in specific pathological conditions have been compared with blood or other body fluids

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