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Review Article

Analysis of iodine from human urine and other food samples

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1. Introduction:

1.1. Significance of iodine for human nutrition:

Small quantities of micronutrients are needed to sustain cell growth and function to complete the life cycle through reproduction but in underdeveloped nations like Ethiopia, pregnant women are susceptible to a variety of micronutrient deficits (Afata et al., 2023). Iodine is essential for the synthesis of thyroid hormone and thus is required for normal physical, neurological, and intellectual growth of infants and children, and normal metabolism and function in adults. On a body weight basis, infancy and early childhood have of highest iodine requirements. Pregnant and lactating women also have increased requirements to meet their heightened physiologic needs. It is critical that women who are likely to conceive, pregnant or lactating, have iodine reserves sufficient for their health and also sufficient to provide the fetus and infant with the necessary iodine supply (Boyages, 1993, Abel et al., 2017).

1.2. Dietary sources of iodine:

The native iodine content of most foods and beverages is low. In general, commonly consumed foods provide 3 to 80 µg per serving (Haldimann et al., 2005) foods of marine origin have higher iodine content because marine plants and animals concentrate iodine from seawater. In many countries, the use of iodized salt in households for cooking and at the table provides additional iodine. Boiling, baking, and canning foods containing iodized salt cause only small losses of less than 10% of iodine content (Chavasit et al., 2002)

1.3. Iodine deficiency:

Iodine functions solely as a component of the thyroid hormones (Zimmermann et al., 2008, Zimmermann, 2016). Poor iodine content of soil and water due to environmental iodine deficiency is the main determinant of iodine deficiency disorders in Ethiopia(Kibatu et al., 2014). In 2011, an estimated 12 million school-age children were living with inadequate iodine, and 66 million people in Ethiopia were at risk of iodine deficiency(Andersson et al., 2012).

Several studies have been documented on iodine nutrition status in Ethiopia where deficiency and thyrotoxicity have been observed with a high prevalence of goiter in communities(Aweke et al., 2014) including cretinism and impaired growth. Nevertheless, reproductive outcomes are affected with an increased risk of stillbirths, abortions, and congenital abnormalities. Maternal urinary iodine has also been positively associated with birth weight, length, and head circumference in male offspring in a recent study of a Bangladeshi population of pregnant women (Rydbeck et al., 2014) as well as the well-recognized impact on offspring cognitive impairment as

described below. Unlike most essential dietary nutrients, iodine status is not linked so much to socioeconomic development but more to geography(Rohner et al., 2014). Its critical significance during pregnancy is, rather than on maternal health directly, due to the devastating impact on the fetus of deficiency.

In poor nations like Ethiopia, iodine deficiency is a serious public health issue(UNICEF, 2011). A report published by the Ministry of Health of the Federal Democratic Republic of Ethiopia states that around 50,000 prenatal fatalities occurred, and 685,000 kids were born with IDD, which causes them to have some sort of learning disability(UNICEF, 2011). Even though iodized salt has been required in Ethiopia since 2011 and the country's supply of iodized salt has increased, iodine deficiency and the repercussions it causes remain a problem, according to a 2016 systematic review. This analysis claims that the national goitre rate for women was found to be higher than 35.8%, with rates as high as 60% in four of the nation's regional states(Keno et al., 2017).

The Ethiopian diet is mainly composed of cereals (teff, maize, sorghum, and millet), tubers and root crops (ensete, potatoes, and sweet potatoes), pulses, and oil seeds. According to the nutrition country profile assessment conducted in Ethiopia by the Food and Agriculture Organization of the United Nations/FAO in 2008, the dietary energy supply in Ethiopia was not sufficient to meet population energy requirements and about half of the population was undernourished. The food supply in Ethiopia is quantitatively insufficient and also lacks diversity. As a result, the Ethiopian people are exposed to under nutrition and micro-nutrient deficiencies (Sheehy et al., 2019). Thus, to mitigate the problem of iodine deficiency and its related consequences, adequate knowledge regarding dietary habits and information on the status of iodine nutrition in a population are of paramount importance so that analysis of iodine samples was important.

2. Analysis of Iodine samples from human urine:

Before you analyse your sample prepare the following chemical solutions.

2.1. Ammonium persulfate:

Ammonium sulfate (22.82g) will dissolve in 100 mL of water.

2.2. Arsenious acid:

Arsenic trioxide (1.25g) and sodium chloride (6.251g) will dissolve with heat in 5N sulfuric acid (50ml) and water up to the calibration line of a 250ml volumetric flask.

2.3. Ceric ammonium sulfate:

Ceric ammonium sulfate (1.2g) will dissolve in 3.5N sulfuric acid (50ml).

2.4. Stock iodine standard (1mg/ml):

About 168.5 mg KIO_3 is dissolved in distilled water to make a final volume of 100 ml. This is stored in an amber-colour bottle (This solution is stable for months).

2.5 Dilute iodine standard (1µg/ml):

Take 100 µl of Stock Iodine Standard and make a volume of 100 ml with distilled water.

2.6. Working iodine standard:

Make the following serial dilutions from diluted Iodine Standard (1µg/ml) into volumetric flasks (10 ml) with double distilled water (diluents). These dilutions are made freshly.

µg/dl Dilution factors
5 µg : 0.5 ml of 1 µg/ml standard + 9.5 ml diluents
10µg : 1.0 ml of 1 µg/ml standard + 9.0 ml diluents
15µg : 1.5 ml of 1 µg/ml standard + 8.5 ml diluents
20µg : 2.0 ml of 1 µg/ml standard + 8.0 ml diluents

3.Final procedure:

According to Sullivan *et al.* (2000) before analysis, the urine samples were put out of the refrigerator a day and defrosted to reach room temperature. Then the urine samples were mixed until homogenization of the suspended sediment. Standards were prepared by pipetting 0, 10, 20, 40, 60, 100, and 250µl of standard solution B in duplicate into 12 test tubes containing 250, 240, 230, 210, 190, and 150µl of H2O respectively, to give a volume of 250µl in each tube. The standard curve was obtained with iodine concentrations- 0, 20, 40, 80, 120, 200 and 500µg/L.

A urine sample of 250µl was pipetted into a 13x100mm test tube and 1 ml of the solution of ammonium persulphate was added to each test tube and mixed slowly. All the test tubes were placed in a thermostatic block and heated for 1h at a temperature 100ºC. Then after digesting the sample, all test tubes were cooled to room temperature. 2.5 ml of arsenic acid solution was added to each 38 test tube, mixed with a “Vortex” mixer, and stayed for about 15 minutes. 300 µl of ceric ammonium sulfate were added to each test tube at 30-second intervals between successive tubes, (which is observed with a stopwatch). Upon adding to the solution, it was followed by mixing with the “Vortex” mixer. The sample was allowed to sit at room temperature and after 30 minutes ceric ammonium sulphate solution was added to the first test-tube.

Then after absorbance (at 420 nm) each sample was read using a single-beam spectrophotometer in 30-second intervals. All successive tubes were read at the same intervals as the ceric ammonium sulfate. Iodine concentration in urine was determined in the base of the value of absorbance plotted from the standard solution. The standard curve was constructed by plotting the log of the absorbance at 405- 420 nm on the X-axis versus the standard iodine concentration in µg/l on the Y-axis with a scattered plot by using Excel on a desktop computer (May et al., 1997). Finally, iodine concentration in µg/l of each specimen was calculated by using the equation of the linear trend line of this chart.

4. Data analysis:

The EPI-INFO version 7 software will be used to enter the data, and the SPSS statistical tool will be used to do the analysis and apply descriptive statistics. The iodine status of the respondents as well as their socioeconomic and socio-demographic characteristics will be described in the descriptive section using percentages, frequency distributions, means, and standard deviations. To determine the combined impact of many variables on iodine deficiencies, multivariate analysis, and Pearson's correlation coefficient will be utilized.

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