Iodine Fortification of Vegetables Improves Human Iodine Nutrition: In Vivo Evidence for a New Model of Iodine Prophylaxis

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Background: Iodine deficiency is the result of insufficient intake of dietary iodine and as a consequence causes multiple adverse effects. About 2 billion individuals in the world are affected by iodine deficiency. It has been found that the most effective way to control iodine deficiency is through the universal salt iodization. However, salt iodization alone may not be sufficient to assure adequate iodine nutrition. In most industrialized countries, excess consumption of salt has become recognized as a health risk. Therefore, biofortification of vegetables with iodine offers an excellent opportunity to increase iodine intake.

Aim and Methods: The aim of this study was to test the efficiency of a new model of iodine prophylaxis in a group of 50 healthy volunteers through the intake of vegetables (potatoes, cherry tomatoes, carrots, and green salad) fortified with iodine. Each serving of vegetables consisted of 100 g of potatoes, carrots, tomatoes, or salad containing 45 mg of iodine (30% of the Recommended Daily Allowance), and the volunteers consumed a single serving of vegetables, as preferred, each day for 2 weeks. Urinary iodine (UI) excretion was measured before and after intake of vegetables.

Results: The UI concentration measured in volunteers before the intake of vegetables was 98.3 mg/L (basal value), increasing to 117.5 mg/L during the intake of vegetables. Seven days after the discontinuation of vegetable intake, UI was 85 mg/L. UI concentration increment was 19.6% compared with the basal value; therefore, the difference was statistically significant ($P = .035$).

Conclusions: Biofortification of vegetables with iodine provides a mild but significative increase in UI concentration and, together with the habitual use of iodized salt, may contribute to improve the iodine nutritional status of the population without risks of iodine excess. **(*J Clin Endocrinol Metab* 98: E694–E697, 2013)**

Iodine is an essential element for the synthesis of thyroid hormones (1). Iodine deficiency (ID) is the result of insufficient dietary iodine intake and in humans results in multiple adverse effects due to inadequate thyroid hormone production, globally named iodine deficiency disorders (2).

The World Health Organization (WHO) estimates that 2 billion people worldwide and around 45% of continental Europe's population are affected by ID (http://who.int/vmnis/database/iodine/en). The recommended indicator for measuring the prevalence of ID is via urinary iodine

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Abbreviations: FT_3, free T_3; FT_4, free T_4; ID, iodine deficiency; RDA, Recommended Daily Allowance; UI, urinary iodine; USI, universal salt iodization; WHO, World Health Organization.

(UI) concentration in spot urine samples (3–5). ID is defined as a population median UI excretion less than 100 μg/L, with deficiency classified as moderate at 20 to 49 μg/L and severe at less than 20 μg/L (5).

The global strategy used by the WHO to control ID worldwide has been through the introduction of universal salt iodization (USI), consisting of the iodization of all salt for human consumption (food, industry, and domestic) as well as livestock consumption (2). Unfortunately, USI is rarely achieved as food industries are often reluctant to use iodized salt because a certain amount of iodine may be lost during the preparation process, eg, due to the use of high temperatures (2). In addition, in some countries the use of iodized salt for livestock appears to be very difficult. The iodization of domestic salt alone cannot guarantee sufficient intake of iodine; furthermore, inorganic iodine is volatile, and it is difficult to be able to control the loss of iodine during its storage and transport and during cooking, especially with use of high-temperature oils.

Hence, enhancing iodine content in vegetables could prove to be an effective way to control ID, because iodine in food is readily bioavailable (up to 99%) and can be easily assimilated (6). Trace element supplementation through plants is generally known as biofortification and represents a cost-effective way to improve human nutrition (6–8); therefore, the application of iodine to agricultural soils and iodine fortification of crops have been proposed as useful strategies to avoid iodine deficiencies in the human diet (7, 9–11).

In this study 50 volunteers with normal thyroid function were enrolled, and they were given a diet of iodine-fortified vegetables (potatoes, carrots, cherry tomatoes, and green salad) to asses whether intake of these iodine-enriched foods would provide an increase in UI concentration.

Materials and Methods

Vegetables

The vegetables used in this study were kindly donated by Pizzoli SpA 7(Budrio, Italy; http://www.pizzoli.it). Pizzoli supplied potatoes, carrots, tomatoes, and lettuce.

Potatoes of the Cupido variety were grown in open fields in Emilia Romagna (Northern Italy) in accordance with the integrated pest management guidelines (12). Integrated pest management is a broad-based approach that uses a range of practices for economic control of pests. Carrots of the Napoli variety were grown in open fields in Veneto (Northern Italy). Cherry tomatoes of the Piccolo variety were grown in a greenhouse in Sicily (Southern Italy). Baby leaf lettuce of the variety Batavia was grown in a greenhouse in Friuli Venezia Giulia (Northern Italy) in accordance with the hydroponic production system. Iodine-enriched vegetables were obtained by a patented procedure (13) of agronomic biofortification. In brief, iodine is sprayed on the plants through foliar fertilization during the growing season to be absorbed directly; the plants assimilate the iodine, and it is has been found that potatoes, carrots, tomatoes, and lettuce have a functional content for a human diet. The amount of accumulated iodine for 100-g of fresh product corresponded to at least to 30% of the Recommended Daily Allowance (RDA).

A pool of agronomists with experience in the Organoleptic properties of food compared these iodine-enriched vegetables with traditional vegetables and found no differences in color, taste, and smell. Costs for iodine-fortified vegetables are similar to those of foods fortified with other elements and are about 15% to 20% higher than the costs of nonfortified products.

Iodine measurements in vegetables

Iodine levels in vegetables were checked by a certified laboratory of analysis in accordance with the method reported previously (14). Samples of potatoes, carrots, tomatoes, and lettuce were collected, rinsed with water, and air-dried. Each sample was homogenized to a fine pulp, and a small amount was mineralized by alkaline extraction using tetramethylammonium hydroxide at an elevated temperature. Iodine was detected in the extracts by inductively coupled plasma–mass spectrometry.

Study design

Fifty healthy volunteers between the ages of 19 and 50 years were enrolled. The study was approved by the local ethics committee, and informed consent was obtained from all subjects.

To assess the efficiency of this model of iodine prophylaxis, all 50 healthy volunteers consumed vegetables fortified with iodine. Each serving of 100 g of vegetables contained about 45 mg of iodine (30% of RDA for an adult). The volunteers consumed a single serving of one of the vegetables each day for 2 weeks. Salad, tomatoes, and carrots were eaten raw. The daily intake of iodized vegetables represented one third of the regular daily intake. Throughout the study, the volunteers did not use any other product containing iodine such as disinfectant, dietary supplements, beauty cream for weight loss, or drugs, and they did not undergo radiological procedures using iodinated contrast media. All the volunteers were habitual users of iodized salt.

Immediately before the beginning of the 7-day study and at the completion of the study period, thyroid volume, UI excretion, and thyroid hormone levels were measured. UI excretion was measured before, during, and after the discontinuation of vegetable intake.

Measurement of thyroid volume

Thyroid ultrasonographic evaluation was conducted on each volunteer using a Tecnos MP Esaote ultrasonography device. Thyroid volume was calculated by the formula length × width × depth × $\pi/6$ for each lobe, and total volume was obtained by summing the volume of each lobe.

UI excretion measurements

Urine samples were taken from each volunteer early in the morning. The AutoAnalyzer III system (Bran+Luebbe) was used to perform an automated measurement of UI. The principle of the method is based on the catalytic effect of iodide in the oxidation-reduction reaction of Sandell-Kolthoff, after digestion of samples with an ultraviolet lamp (15) as described previously (16). The automated system was sensitive enough to detect UI concentrations of 5.1 μg/L. The within-assay imprecision (co-

Table 1. Average Iodine Content in Iodized and Untreated Vegetables

	Iodized Vegetables, mg Iodine/100 g	Noniodized Vegetables, mg Iodine/100 g
Potatoes	48	3
Carrots	52	2
Tomatoes	45	1
Lettuce	50	1

Iodine was detected by inductively coupled plasma–mass spectrometry.

efficient of variation) was 8%, and the between assay coefficient of variation was 15%.

Laboratory evaluation of thyroid function

Serum free T_4 (FT_4) and free T_3 (FT_3) were measured with a chemiluminescent method (Vitro System; Ortho-Clinical Diagnostics, Rochester, New York). TSH was assessed by an ultrasensitive commercial chemiluminescent method (Immulite 2000; Diagnostic Products, Los Angeles, California). Thyroid peroxidase and thyroglobulin antibodies were measured using a two-step immunoenzymatic assay (AIA-Pack TPOAb and TgAb; Tosoh, Tokyo, Japan). Serum thyroglobulin was measured using an immunometric chemiluminescence assay (Immulite 2000).

Statistical analysis

The Friedman nonparametric test was used to test the difference in iodine concentrations measured before and after vegetable intake. Multiple post hoc comparisons were performed using the Bonferroni correction, when necessary. Statistical significance was assumed for a value of $P < .05$. Data are presented as median and interquartile range or median ± 95% confidence interval or as a percentage of basal concentration.

Results

In Table 1, the average iodine content in iodized and in untreated vegetables is shown. In all cases, the amount of iodine was measured at nearly 50 mg/100 g of vegetable.

UI concentration (expressed as median) measured in volunteers was 98.3 mg/L before the intake of vegetables, 117.5 mg /L during the intake of vegetables, and 85 mg/L 7 days after discontinuation of the intake of vegetables (Table 2). The increment of UI concentration during the intake of vegetables was 19.6% compared with the basal value and thus was statistically significant ($P = .035$) (Table 2). The reduction in the daily UI concentration (−13.5%) after discontinuation of vegetables was not statistically significant compared with the basal value ($P = .683$) (Table 2). Toxic levels of iodine were not reached in any of the subjects.

All 50 volunteers showed normal laboratory thyroid values (FT_3, 3.2 ± 0.9 pg/mL; FT_4, 10.2 ± 1.5 pg/mL; and TSH, 1.9 ± 0.5 mU/mL; absence of thyroid antibodies) and normal thyroid volume before the beginning of the study. No modifications in thyroid hormone levels and TSH were detected during and after discontinuation (FT_3, 3.3 ± 0.7 pg/mL; FT_4, 11.2 ± 1.8 pg/mL; and TSH, 1.8 ± 0.6 mU/mL; absence of thyroid antibodies) of vegetable intake. Thyroid volume remained unchanged in all volunteers during the study.

Discussion

ID has been found to be the most common cause of preventable mental impairment worldwide (2). The mainstay of ID prevention has been providing access to iodized salt (2, 5), an intervention considered1 of the 3 most cost-effective solutions to major world problems by the Copenhagen Consensus in 2008, which included a panel of 8 economists, of whom 5 are Nobel Laureates (http://www.copenhagenconsensus.com/default.aspx?id=953). The global strategy used by the WHO to control ID worldwide has been the introduction of USI, which provides the iodization of all salt for human use (food industry and domestic) as well as for livestock consumption (2, 6, 17). Nevertheless, USI has rarely been achieved, and in industrialized countries, the evidence of the risks for human health caused by excess salt consumption has become compelling. The positive correlation between habitual dietary salt intake and blood pressure has been established through experimental, epidemiological, migration, and intervention studies (18). For these reasons, enhancing io-

Table 2. UI Concentrations Measured Before Intake of Vegetables (Basal), During Intake of Vegetables (Treatment), and 7 Days After Discontinuation of Vegetable Intake (Posttreatment)

		IQR			95% CI			
	Valid N	25th Percentile	Median (μg/L)	75th Percentile	Min	Median (μg/L)	Max	Median (% of Basal)
Basal	50	63.5	98.3	142.0	12.5	98.3	271.0	100.0%
Treatment	48	78.5	117.5	180.0	12.5	117.5	556.0	119.6%[a]
Posttreatment	45	65.0	85.0	134.0	18.8	85.0	294.0	86.5%

Data are medians and interquartile range or median ± 95% confidence interval or percentage of basal concentration. N represents the number of volunteers who provided urine samples useful for the determination of the medians.

[a] Statistical significance among basal, treatment, and posttreatment values was assumed for $P < .05$.

dine content in vegetables can be an important alternative tool to prevent ID.

To assess the effectiveness of this tool to improve human iodine nutrition and to test the bioavailability of iodine in humans after consumption of iodized vegetables, 50 volunteers were given a daily serving of vegetables containing about 45 to 50 mg of iodine (corresponding to 30% of the RDA for an adult) for a period of 2 weeks. All iodized vegetables used in this study were developed by the food industry and are currently available in the Italian market. In particular, lettuce is a good crop to be used in a biofortification with iodine study because, being a leafy vegetable, it is usually consumed raw with no risk of iodine loss. The results of this study showed that the UI concentration increased from 98.3 to 117.5 mg/L during the intake of vegetables and returned to basal levels 7 days after the discontinuation of vegetable intake. The effect of this patented procedure (13) of iodine biofortification of vegetables was a statistically significant increase of the UI level during the intake of vegetables, thus showing that iodine in food is readily bioavailable and assimilated.

In conclusion, iodine-fortified vegetables such as carrots, tomatoes, potatoes, and lettuce may be a successful tool to provide an increase in UI concentration because they are consumed daily in most families. Their consumption, together with the habitual use of iodized salt, may improve the iodine nutritional status of the population without risk of iodine excess.

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