

DOI: <https://doi.org/10.56936/18290825-2025.19v.4-70>**INFLUENCE OF DIETARY IODINE ON SEMEN QUALITY PARAMETERS IN GENERAL MALE POPULATION: A PILOT STUDY****DUNDOVIĆ M.^{1,3§}, FERENAC KIŠ M.^{2,3§}, MARCIJUŠ L.¹, KLAPEC T.⁴, BANJARI I.^{5*}**¹ Department of Human and Medically Assisted Reproduction, University Hospital Centre Osijek, Osijek, Croatia² University Hospital Centre Osijek, Clinical Institute for Transfusion Medicine, Osijek, Croatia³ Josip Juraj Strossmayer University of Osijek, Faculty of Medicine, Osijek, Croatia⁴ Department of Applied Chemistry and Ecology, Faculty of Food Technology, Josip Juraj Strossmayer University of Osijek, D Osijek, Croatia⁵ Department of Food and Nutrition Research, Faculty of Food Technology, Josip Juraj Strossmayer University of Osijek, Osijek, Croatia*Received 4.03.2024; Accepted for printing 21.10.2025***ABSTRACT**

Iodine is an important trace element in the human body and plays a key role in the overall metabolism and growth. Global iodization programmes resulted in remarkable improvement of iodine status and nowadays almost 90% of global population use iodized salt, suggesting adequate iodine status. However, this iodization programmes could have led to excessive iodine consumption in some population groups, but remains sparsely researched. Reproduction is altered with iodine deficiency and hypothyroidism causes anovulation, infertility, gestational hypertension, increased first trimester abortions, stillbirths and is linked to male infertility.

The aim of this research was to assess the relationship between dietary iodine consumption, iodine status and parameters of semen quality in men. The study was conducted on general male population from Eastern Croatia. Participants completed study-specific questionnaire, along with dietary iodine consumption assessment and provided samples of semen and morning urine. Iodine content was determined with a standardized method.

Only 33.3% of men had normal semen analysis while others had some type of altered semen analysis which is linked to infertility. After comparing men with normal and altered semen analysis, those with normal findings had lower iodine concentration in urine and seminal fluid. However, men with altered semen analysis consumed less iodine from diet, but the difference was not statistically significant. Also, men preferring saltier foods had higher total sperm count.

These are the first results for iodine in urine and seminal fluid of men undergoing semen analysis within potential infertility treatment from Croatia. Despite small sample, the results provide interesting insights into relationship between iodine and semen quality. Also, dietary iodine assessment needs to be thoroughly reassessed to enable more accurate calculations.

KEYWORDS: iodine; diet; semen analysis; sperm count; fertility**CITE THIS ARTICLE AS:**

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INTRODUCTION

Iodine is a trace element important for the synthesis of thyroid hormones, but its importance extends far beyond [Barbaro D et al., 2019]. Reproduction is altered with iodine deficiency that causes anovulation, infertility, gestational hypertension, increased first trimester abortions, and stillbirths [Dunn JT, Delange F, 2001].

Current data shows that between 2003 and 2017 a remarkable improvement was achieved in iodine status, primarily due to salt iodization programmes. Now, 89% of global population use iodized salt [Barbaro D et al., 2019]. More than 90% of dietary iodine is excreted in the urine so its content is considered a valuable biomarker for iodine status. Due to severe extent of endemic goitre, Croatia has introduced mandatory salt fortification with iodine in 1996. The legislation scope is salt used in households and in processed foods, both salt produced domestically or imported. Iodine content should be 19 mg/kg of salt [GFDE, 2022]. Infertility is a disease characterized by the failure to establish a clinical pregnancy after 12 months of regular and unprotected sexual intercourse and factors affecting both genders' fertility includes lifestyle related factors [Vander Borgh M, Wyns C, 2018]. Human sperm count is declining in the past 50 years and an overall 32.5 % decrease in sperm concentration has been noted [Sengupta P et al., 2018]. Subfertility and infertility are a major problem in the modern world and almost 50% of all infertility issues are caused by male factor [Kumar N, Singh A, 2015]. Causes of male infertility include genetics, anatomical problems, hormonal imbalance, previous illnesses like mumps, poor general health in a man, food and lifestyle choices and social factors [Agarwal A et al., 2015; Leaver R, 2016]. Since the introduction of iodine food fortification, some people may be consuming excess iodine, which has been linked to poor semen quality [Sakamoto K et al., 2004; Chakraborty A, 2021]. The objective of this study was to determine the relationship between semen quality and iodine consumption in general male population from Eastern Croatia.

MATERIALS AND METHODS

Study design and population: This was a cross-sectional observational study encompassing men, of at least 18 years of age, who came for a semen

analysis. All study subjects came to the main tertiary health institution in Eastern Croatia for semen analysis, due to various health reasons, including infertility and assisted reproductive technology procedures.

We obtained institutional review board approval from the Ethical Committee of the University Hospital Centre Osijek (decision from October 6, 2021, number of approval R1/13151/2021). Informed consent was obtained from all participants. All methods were carried out in accordance with relevant guidelines and regulations.

Questionnaires: Participants filled in a study-specific questionnaire which included questions about general information (age, education, employment, health issues, etc.), and lifestyle (physical activity, smoking, alcohol consumption). Based on the self-reported body weight and height, Body Mass Index (BMI) was calculated [WHO, 2022].

Dietary iodine was assessed by using a semi-quantitative Food Frequency Questionnaire (sFFQ) containing a list of the main dietary sources of iodine. In Croatia, mandatory iodine fortification of salt for household purposes is in effect since 1950s [Jukić T et al., 2008], and all salt used in households must be fortified with iodine. Therefore, semi-quantitative Food Frequency Questionnaire included assessment of salt consumption, both table (which is fortified) and sea salt (naturally containing iodine). The consumption was assessed on the level of last 30 days, and consumption frequency offered was: 2 or more times a day, once a day, 3 to 5 times a week, 2 to 3 times a week, once a week, 2 to 3 times a month, once a month, and rarely. Medium size portions were provided, and men were asked to assess whether they consumed less or more than the predefined medium size portion. The questionnaire was then analysed by using NutriPro software which uses national food composition database.

Semen analysis: Semen analysis was conducted by an experienced clinical embryologist using standard operative procedure for the examination. Equipment used to conduct the analysis includes Nikon Eclipse e200 microscope and Makler counting chamber. The results of semen analysis are recorded on standard form and include following parameters: total sperm count in sample, sperm concentration per millilitre of seminal fluid, total

sperm motility (%), progressive sperm motility (%), non-progressive sperm motility (%) and immotile sperm (%). Semen quality was assessed according to the standardized methods [WHO, 2009].

Iodine determination

Iodine in urine and semen samples was determined according to WHO method [WHO, 2007]. In short, samples were digested with ammonium persulfate. Iodine in samples acts as a catalyst in the reduction of ceric ammonium sulphate (yellow) to the cerium form (colourless) and is detected by the rate of colour disappearance (Sandell-Kolthoff reaction).

Reagents needed for the procedure include ammonium persulfate, As_2O_3 , NaCl, H_2SO_4 , $Ce(NH_4)_4(SO_4)_4 \times 2H_2O$, deionized water and KIO_3 . All reagents were of p.a. grade.

Procedure: 250 μ l of each urine (or seminal fluid) sample is pipetted into a test tube. Each iodine standard was pipetted into a test tube, H_2O was added to make a final volume of 250 μ l, to each tube 1 ml 1.0 M ammonium persulfate was added. All tubes were heated for 60 minutes at 100 °C, and then cooled to room temperature. Then, 2.5ml of arsenic acid solution was added and mixed on vortex, left to stand for 15 minutes, followed by quickly mixing with 300 μ l of ceric ammonium sulphate solution, and left 30 minutes at room temperature. Absorbance was measured at 420 nm.

Statistical analysis: Normality of data distribution was tested by the non-parametric Kolmogorov-Smirnov test for the comparison of medians and arithmetic mean, and histograms plotting. For the comparison of categorical data within and between groups Fishers exact test was used, and Pearson's test of correlations was used to calculate correlations between numerical data. T-test for independent samples was used to test differences between groups according to change in pain scores. Software tool Statistica 14.0 was used for the statistical analysis.

RESULTS

The study included 51 men. Nine were excluded due to incomplete data so the final number taken for the analysis was 42. The average age was 34.4 ± 4.8 years and BMI 29.6 ± 5.6 kg/m² (Table 1). Iodine concentration in urine was 122.1 ± 43.2 μ g/L and in seminal fluid 10.6 ± 6.9 μ g/L. Other parameters related to semen analysis are given in Table 1.

TABLE 1

Age, Body Mass Index and parameters of semen analysis and iodine concentrations (N=42)

| Variable | Mean \pm SD | Min. | Max. |
|---|------------------|------|-------|
| Age (years) | 34.4 ± 4.8 | 25 | 43 |
| BMI (kg/m ²) | 29.6 ± 5.6 | 23.1 | 53.7 |
| Urine iodine (μ g/L) | 122.1 ± 43.2 | 0,04 | 208.5 |
| Semen fluid iodine (μ g/L) | 10.6 ± 6.9 | -2.1 | 24.5 |
| Total sperm count in sample (10 ⁶) | 31.9 ± 31.4 | 0 | 118 |
| Sperm concentration in sample (10 ⁶ /ml) | 12.2 ± 11.7 | 0 | 59 |
| Sperm motility (PR+NP) (%) | 50.8 ± 25.5 | 0 | 91 |
| Sperm motility (PR) (%) | 43.5 ± 24.2 | 0 | 82 |
| Sperm motility (NP) (%) | 7.4 ± 6.4 | 0 | 37 |
| Sperm motility (IM) (%) | 46.7 ± 25.4 | 0 | 100 |

NOTES: SD – standard deviation; Min – minimum value; Max – maximum value, BMI = Body Mass Index; PR = progressive motility; NP = non-progressive motility; IM = immotile sperm

Only 33.3% of men had a normal semen sample. Oligoasthenozoospermia was found in 21.4%, oligozoospermia in 35.7% and asthenozoospermia, azoospermia and necrozoospermia in 9.5% of men. Normozoospermia is a preferable result since it signifies high chance of pregnancy in couples that are actively trying to conceive. Other diagnoses indicate a reduced chance of pregnancy depending on severity. Azoospermia is the most severe condition since it implies no spermatozoa was found in sample and patients are usually referred to further examination and some form of sperm extraction methods are needed [WHO, 2009].

None of the men had thyroid disease or any other condition which could affect iodine metabolism. Six had other health conditions (hypertension, dyslipidaemia, diabetes), but due to a small number could not be taken in the analysis.

We then divided men into two groups, one encompassing only men with a normal semen analysis (Group 1) and those with all other findings (Group 2). These two groups did not differ in age or BMI. Group 1 had lower iodine concentration in both urine (104.6 ± 42.8 μ g/L) and seminal fluid (9.5 ± 7.6 μ g/L) in comparison to men from Group 2 (130.8 ± 41.5 μ g/L (p=0.063) urine, 11.2 ± 6.7 μ g/L (p=0.468) seminal fluid), though not reaching statistical significance.

After comparing dietary iodine consumption between the two groups of men, we found that Group

2 had lower total iodine consumption in comparison to Group 1 (Table 2). In other words, men with altered semen analysis consumed less iodine from all observed foods, except for milk where consumption was almost double comparing to the Group 1 (69.10 ± 48.28 vs 39.81 ± 37.00 , $p=0.053$).

However, saltwater fish consumption correlates significantly with higher urine iodine concentrations ($r=0.305$, Table 4). Higher preference towards salty foods correlate with higher total sperm count ($r=0.394$), while higher consumption of iodine from freshwater fish ($r=-0.310$) and milk ($r=-0.452$) correlate with lower total sperm count (Table 3). In addition, we found that higher BMI correlates with lower urine iodine concentration ($r=-0.422$, Table 3).

Despite the results did not reach statistical significance, trend towards lower dietary iodine consumption and higher iodine concentrations in both urine and semen are visible. Probable reason for not reaching statistical significance is the small sample size, which is supported by the large standard deviations. Also, potential overestimation and underestimation of food consumption is another probable reason for this unexpected finding. Reassessment of the method used for the dietary iodine consumption calculation should be considered.

DISCUSSION

Since the iodine food fortification, mainly through salt iodization, some researchers have hypothesized that the introduction of dietary iodine supplementation has been a factor in the decline in sperm counts in the United States and some European countries [Sakamoto K et al., 2004]. This is supported by the fact that sperm counts in Japan have not changed, probably due to high traditional consumption of seaweed and other seafood [Sakamoto K et al., 2004]. Having in mind mandatory salt fortification with iodine, Croatians are not expected to have low dietary iodine consumption. Therefore, we aimed to analyse iodine levels in urine and semen, in relation to dietary consumption of iodine, and compare them with semen parameters. Population selected were men who came

TABLE 2
Dietary iodine consumption from observed foods/ food groups among men with normal (Group 1) and men with altered (Group 2) semen analysis

| Variable | Iodine consumption /day (μg) | | |
|-----------------------|---|---------------------|---------|
| | Group 1 (n=14) | Group 2 (n=28) | p-value |
| | Mean \pm SD | Mean \pm SD | |
| Salt | 367.33 ± 239.41 | 295.14 ± 186.66 | 0.289 |
| Meat products (total) | 86.10 ± 97.51 | 47.82 ± 56.36 | 0.114 |
| Dried processed meats | 79.60 ± 94.31 | 44.33 ± 56.04 | 0.136 |
| Saltwater fish | 47.26 ± 58.76 | 40.24 ± 38.98 | 0.646 |
| Freshwater fish | 2.71 ± 4.42 | 1.85 ± 3.55 | 0.497 |
| Eggs | 22.75 ± 29.11 | 20.61 ± 18.11 | 0.771 |
| Milk | 39.81 ± 37.00 | 69.10 ± 48.28 | 0.053 |
| Cheese (total) | 8.59 ± 7.46 | 8.01 ± 6.39 | 0.795 |
| Yoghurt | 100.26 ± 90.33 | 72.36 ± 75.98 | 0.299 |
| Total | 953.75 ± 468.15 | 791.61 ± 419.93 | 0.263 |

TABLE 3
Correlation coefficients between urine and seminal fluid iodine concentrations, and total sperm count among men (N=42)

| Variables | Seminal fluid | Urinary | Total sperm count |
|---|---------------|---------|-------------------|
| Age | -0.270 | 0.001 | 0.161 |
| BMI | -0.140 | -0.422* | 0.208 |
| Preference of salty foods (scale 1 to 10) | 0.050 | 0.081 | 0.394* |
| Total dietary iodine | -0.048 | 0.087 | 0.114 |
| Salt | -0.005 | -0.038 | 0.283 |
| Meat products (total) | 0.037 | -0.042 | 0.133 |
| Dried processed meats | 0.014 | -0.005 | 0.106 |
| Saltwater fish | 0.113 | 0.305* | -0.265 |
| Freshwater fish | 0.118 | 0.111 | -0.310* |
| Eggs | -0.134 | -0.099 | -0.182 |
| Milk | -0.025 | 0.101 | -0.452* |
| Cheese (total) | -0.169 | 0.023 | 0.021 |
| Yoghurt | -0.033 | 0.222 | 0.085 |

NOTES: BMI – Body Mass Index, *statistically significant at $p<0.05$

for a semen analysis due to various health reasons including infertility.

Semen analysis is a valuable tool for epidemiologists and enables insight into regional and temporal shifts of sperm counts and causes of infertility [Ellekilde Bonde J, 2010]. Men who participated in our study had significantly altered semen analysis; only 33.3% had a normal result. Recent meta-anal-

ysis showed a decline in sperm count over time on a global scale [Levine H et al., 2022]. Sperm count and time to pregnancy have nonlinear relationship, meaning that higher sperm count does not necessarily imply a higher probability of conception, while the probability of conception drops rapidly as sperm count drops [Levine H et al., 2023]. Declining fertility rates are a global problem, despite having some high-fertility cluster areas. Assisted reproductive technology use increased significantly over time but varies across countries [Passet-Wittig J, Greil AL, 2021]. In Croatia in 2022, the share of assisted reproductive technology births was 5.3%, and this share increases continuously. It is estimated that about half of infertile women use Assisted reproductive technology, with differences between countries [Passet-Wittig J, Greil AL, 2021].

The amount of urinary iodine found in men from Eastern Croatia is slightly lower than the one found in Norwegian men. Based on the 24h urinary samples, median urinary iodine in Norwegian men was 174 µg/day [Madar A et al., 2020], while our study subjects had 122.1 µg/day. Urinary iodine was even lower in a group of Somali men living in Norway [Madar A et al., 2018]. Our results show that men with any type of pathological semen analysis have higher iodine concentrations in urine and semen. This is in line with Wang N et al. [2021] who found that men with deficient or excessive urine iodine concentrations have lower semen quality. Additionally, their female partners will have longer time to pregnancy in comparison to couples where a man has normal urine iodine concentration [Wang N et al., 2021]. Lower semen quality and longer time to pregnancy was also reported for deficient urine iodine concentrations [Wang N et al., 2021]. Partal-Lorente A et al. [2017] also found that men with higher urine and semen iodine have more morphological alterations in spermatozoa, lower motile sperm count and longer time to pregnancy (> 3 years).

Total dietary iodine was significantly higher in comparison to data from Norway where median dietary iodine, also calculated from a food frequency questionnaire was 318 µg/day [Madar A et al., 2020]. Somali men living in Norway had even lower consumption of dietary iodine [Madar A et al., 2018]. Dietary diversity is the possible explanation to these large differences in dietary iodine

consumption.

In our study, men with altered semen analysis consumed less iodine from all observed foods, except for milk where consumption was almost double the amount men with normal semen analysis consumed. Research by Sun Y et al. [2020] found that men with deficient or excess urine iodine levels had suboptimal dietary iodine consumption which was negatively associated with semen concentration and semen count. Longer time to pregnancy was observed in iodine deficiency and excess group than in those in the optimal group [Sun Y et al., 2020]. In Norway, dietary iodine relies mostly on the consumption of milk and milk products, fish and fish products, and eggs [Madar A et al., 2020]. On the other hand, for adult men in the US, milk and dairy represent the biggest contributor to daily iodine consumption [Lee K et al., 2016]. Dairy, along with eggs, bread and iodine-containing supplements correlate significantly with higher urinary iodine concentrations among adult men in the USA [Lee KW et al., 2016]. Our results show that higher saltwater fish consumption correlates significantly with higher urinary iodine, which is linked to impaired semen analysis. Yet, higher seafood consumption can also point to a shorter time to pregnancy [Gaskins A et al., 2018].

Finally, most researchers comment dietary iodine consumption in relation to iodine deficiency [Haldimann M et al., 2015; Esche J et al., 2020], while excess dietary iodine is commented in relation to impaired thyroid function [Farebrother J et al., 2019]. Our preliminary result however indicate that excess dietary iodine can have impact on other body systems and should be more attentively researched.

CONCLUSION

Our findings support negative relation between low dietary iodine consumption and semen quality parameters. Men with any type of pathological semen analysis have higher iodine concentrations in both urine and semen. These characteristics have been largely supported by the literature and are linked to a longer time to pregnancy. Given the fact that only a third of men had normal semen analysis, and having in mind declining fertility rates, not just in Croatia but in the majority of countries, more interest from researchers is highly needed.

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