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Dietary Glycemic Index and Load and Semen Quality: A Cross-Sectional and Prospective Analysis within the FERTINUTS Trial

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Purpose: Infertility is a global health issue and nutrition plays a significant role in fertility outcomes. We aimed to investigate the cross-sectional and prospective associations of glycemic index (GI) and glycemic load (GL) with semen quality parameters in a cohort of healthy young men.

Materials and Methods: The study included 106 men aged 18–35 years from the FERTINUTS trial. Dietary intake was estimated through 3-day dietary records and several semen parameters were assessed. Multivariable linear regression analysis with the Least Absolute Shrinkage and Selection Operator (LASSO) approach was employed.

Results: The cross-sectional analysis revealed positive associations between GI and GL and total sperm count, sperm concentration, and total motility. In the prospective analysis, baseline GI was associated with increases in pH, vitality, immotile sperm or abnormal midpiece and decreases in total sperm count and motility. Conversely, GL was positively associated with changes in vitality and total sperm count.

Conclusions: While these findings suggest that GI may have adverse effects on several sperm quality parameters, the results were not consistently observed in the cross-sectional analysis. However, GL was consistently associated with better sperm quality in both analyses. The impact of carbohydrate quality and quantity on fertility remains uncertain and larger prospective studies are needed.

Keywords: Carbohydrates; Fertility; Glycemic index; Glycemic load; Semen analysis

INTRODUCTION

Infertility is a condition characterized by the inability to achieve pregnancy after at least one year of unprotected sexual intercourse. With more than 186 million individuals affected worldwide, it is evident that

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infertility poses a significant public health challenge [1]. In a recent report published by the World Health Organization (WHO) the overall lifetime prevalence of infertility was estimated at 17.5%, with variations observed based on geographical regions, gender and socioeconomical status [2].

Traditionally, the focus of infertility research is on female factors. However, growing evidence suggests that male-related factors could play an important role. In fact, male-related factors could be the main cause in approximately 25% of infertility cases in couples [3] and contributes to infertillity in up to 50% of cases [4]. Furthermore, it has been estimated that low sperm quality, assessed by several semen parameters including pH, volume, total sperm count, sperm concentration, vitality, motility and morphology as recommended by the WHO [5], affects nearly 90% of couples facing infertility [4].

During the last decades, a decline in sperm quality has been observed and several risk factors have been identified as potential contributors [1,6]. Genetic factors, smoking, stress, pollution and exposure to pesticides have all been implicated in the deterioration of sperm quality [3,6]. Furthermore, emerging research suggests that diet may also have an impact on male fertility. High consumption of red and processed meat, saturated fatty acids, sugar, alcohol, coffee, potatoes and sweets, has been consistently linked to poorer semen quality [3,7,8], while adherence to a Mediterranean diet, characterized by the consumption of vegetables, fruit, fibre, antioxidants, poultry, low-fat dairy products and omega-3 fatty acids, appears to have a beneficial effect on semen quality outcomes [3,7,8].

In this context, glycemic index (GI) and glycemic load (GL) are particularly relevant due to their direct impact on glucose metabolism and subsequent physiological responses. While GI indicates the glycemic response to dietary carbohydrates, GL also takes into account the quantity of carbohydrates consumed. Diets characterized by high GI and GL have been associated with increased risk of cardiometabolic diseases [9,10] and these dietary factors could affect reproductive health through insulin resistance, inflammation, oxidative stress (OS) and changes in hormonal homeostasis. However, the role of GI and GL on semen quality or fertility has never been assessed.

Investigating the associations between GI or GL and semen quality offers a unique opportunity to expand our understanding of the potential impact of the quality and quantity of carbohydrates on semen parameters, uncover new insights and contribute to move forward on potential preventive and therapeutic approaches in the field of male fertility. Therefore, the aim of this study was to examine cross-sectional and prospective associations of GI and GL with semen quality in a cohort of healthy young men.

MATERIALS AND METHODS

1. Study design

The present analysis was performed as an observational prospective cohort study within the FERTINUTS trial. The FERTINUTS study was a parallel 14-week, randomized, controlled clinical trial conducted between December 2015 and February 2017 that aimed to determine the effect of a mixture of 60 g of nuts per day (including 30 g of walnuts, 15 g of almonds and 15 g of hazelnuts) on semen parameters.

The eligible participants were healthy men between 18–35 years old who followed a Western-style diet pattern assessed by a 15-item dietary questionnaire [11]. Exclusion criteria were frequent nut consumption, nut allergy, smoking, taking multivitamins, supplements or chronic medication, having reproductive disorders or severe chronic illness and using illegal drugs. Participants were randomly assigned to either nuts group or control group.

2. Dietary measurements

A 3-day dietary record (including 2 workdays and a weekend day) was administred at baseline and at the final visit by trained dietitians. The average of the 3-day dietary record was used to estimate the total energy intake, macronutrient and micronutrient intakes for each individual by using Spanish food composition tables [12].

GI was estimated through the international tables of GI using glucose as the reference food [13]. Daily GL for each patient was determined summing the GI of each food multiplied by the carbohydrate amount consumed and divided by 100. Additionally, the daily GI was calculated by multiplying the daily GL by 100 and dividing the product by the daily carbohydrate intake.

3. Outcome assessment

In order to evaluate semen quality, fresh semen samples were collected from all participants at the first visit and at the end of the trial after 3 days of sexual abstinence and were analyzed by trained personnel maximum 1 hour after collection. Semen quality was assessed by several semen parameters including pH, volume, total sperm count, sperm concentration, vitality, motility and morphology as recommended by the 2010 WHO report [5].

Semen volume and pH were determined after 30 minutes of liquefaction with Pasteur pipette and pH-indicator strips, respectively. Total sperm count and sperm concentration were estimated with a 100 μm–deep hemocytometer chamber (Neubauer chamber), using bright-field optics at ×400 magnification. Sperm motility included progressive motility, non-progressive motility and immotility and was determined under a light microscope at ×400 magnification. Then, motility was also expressed as a percentage of total motility including progressive and non-progressive motility. Sperm vitality was assessed by evaluating the integrity of the sperm plasma membrane using eosin-nigrosine at ×1,000 magnification. Additionally, sperm morphology was determined on semen smears for staining with Hemacolor (Millipore) at ×1,000 magnification, identifying normal sperm or defects in the head, midpiece, or principal piece. Morphology was also expressed as a percentage of normal forms.

4. Other variables

At baseline and at the end of the study, blood samples were collected under 12-hour fasting conditions and plasma glucose, total cholesterol, high-density lipoprotein, low-density lipoprotein, very low-density lipoprotein, triglycerides, insulin and C-reactive protein were assessed by standard enzymatic automated methods according to the routine laboratory protocols (COBAS; Roche Diagnostics Ltd.). Additionally, some anthropometric and general data such as height, weight (TANITA TBF-300; Tanita), waist circumference, blood pressure (Omron HEM-705CP; Omron) and body mass index (BMI) were recorded. All participants were given at each visit a questionnaire to assess the presence of adverse effects such as headache, diarrhoea, vomiting, dizziness, constipation, allergy, among others during the study. Detailed methodological procedures can be found elsewhere [11].

5. Statistical analyses

Baseline characteristics of study participants are presented as mean and standard deviation or median and interquartile range. The distributions of variables were assessed using the Anderson-Darling test. A natural logarithmic transformation was applied to the seminogram variables to approximate a normal distribution. Missing observations in the covariates were replaced with the median value.

1) Cross-sectional analysis

Multivariable linear regression models were fitted to assess the relationship of GI and GL with semen parameters. All cross-sectional analyses were adjusted for age, alcohol consumption, BMI, total energy intake and dietary intakes of protein, carbohydrates, monounsaturated fatty acids, polyunsaturated fatty acids, saturated fatty acids and fibre intake.

2) Prospective analysis

Changes in semen parameters were calculated by subtracting the final visit measures from the baseline visit. The associations of baseline GI and GL with changes in semen parameters after 14 weeks of follow-up were assessed using multivariable regression models. These models were adjusted for the same potential confounders mentioned above along with the intervention group and the respective ln-transformed seminogram value at baseline. Since GL considers the quantity of carbohydrate consumed, models with GL as exposure were not adjusted for carbohydrate intake.

Considering the number of covariates and the relatively small size, there is a possibility of overfitting the regression models. To address this and account for the dimensionality and collinearity of the data (Supplement Fig. 1), we performed multivariable regression analysis using the Least Absolute Shrinkage and Selection Operator (LASSO) approach ("glmnet" R package) with the argument "lambda.min." LASSO deals with $n \ll p$, known as the large p small n problem, by shrinking many of the coefficients to zero, leaving only the important ones in the large p small n scenario thus minimizing prediction error (mean squared error). Statistical analyses were carried out using R version 4.2.2 (R Foundation for Statistical Computing).

6. Ethics statement

The study protocol was approved by the Institutional

Review Board of the Hospital Universitari Sant Joan de Reus in 2015 (Ref. CEIC: 15-1029/10aclaassN1). The FERTINUTS study was registered at ISRCTN registry as ISRCTN12857940. All participants signed the informed consent, and the study was performed according to the Declaration of Helsinki for Medical Research involving Human Subjects.

RESULTS

1. Study participants

From a total of 244 participants assessed for eligibility, 106 individuals were included in the cross-sectional analysis after excluding those individuals who either did not meet the selection criteria or declined to participate. For the prospective assessment, 8 participants were further excluded because they did not attend the final visit. This resulted in a total of 98 participants being included in the prospective analysis (Fig. 1).

Table 1 shows the anthropometric, biochemical, sperm quality and nutritional data (energy intake, macronutrient intake [expressed as percentage of daily energy intakes], dietary fiber and alcohol intake) of the study population at baseline. The mean GI and GL were 55.55±4.99 and 152.38±46.79, respectively. Moreover, the mean age of the participants was 24.66±4.70 years and the majority of the population was normoweight with a mean BMI of 23.8±3.13 kg/m^2 (Table 1).

2. Cross-sectional associations of GI and GL with semen parameters

Dietary GI was associated with higher total sperm count, sperm concentration and total motility, and lower immotile sperm (Table 2). No significant associations were found for the other semen parameters after adjusting for potential confounders. In addition, GL was associated with higher total sperm count, sperm motility and sperm concentration and lower immotile sperm (Table 2).

3. Prospective associations of GI and GL with changes in semen parameters

The relationship between baseline GI and changes in semen quality is shown in Table 3. GI was positively associated with changes in pH, vitality, immotile sperm or abnormal midpiece and inversely with changes in total sperm count and motility. Baseline GL was positively associated with changes in vitality and total

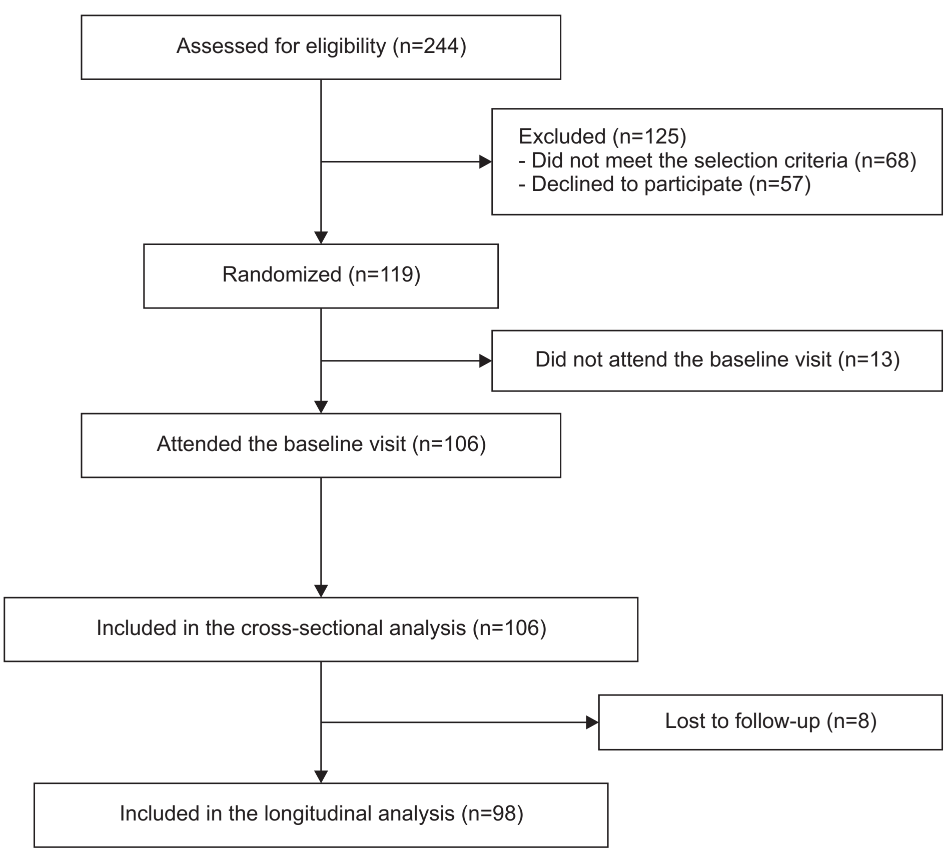

Fig. 1. Flowchart of the study population.

Table 1. Characteristics of the study population at baseline

	Subjects (n=106)
Glycemic index	55.55 (4.99)
Glycemic load	152.38 (46.79)
General characteristics	
Age (y)	24.66 (4.70)
Weight (kg)	75 (10.65)
BMI (kg/m^2)	23.8 (3.13)
Waist circumference (cm)	81.41 (8.07)
Systolic blood pressure (mmHg)	127.73 (11.58)
Diastolic blood pressure (mmHg)	72.37 (8.11)
Biochemical variables	
Fasting plasma glucose (mg/dL)	86 (82.25, 92.75)
Total cholesterol (mg/dL)	168 (150.00, 187.75)
HDL-c (mg/dL)	56 (50.00, 64.75)
LDL-c (mg/dL)	95 (77, 106)
VLDL-c (mg/dL)	13 (11, 18)
Triglycerides (mg/dL)	66 (55.25, 89.50)
Fasting plasma insulin (mcUI/mL)	5.45 (2.90, 7.65)
C-reactive protein (mg/dL)	0.2 (0.2, 0.2)
Semen parameters	
pH	8 (8.0, 8.5)
Volume (mL)	3 (2.00, 4.48)
Total sperm count ($\times10^6$)	72.05 (27.15, 122.50)
Sperm concentration ($\times10^6$)	24.10 (10.85, 42.00)
Vitality (%)	79.3 (72.70, 84.67)
Total motility (progressive and non-progressive motility) (%)	66.45 (48.17, 74.78)
Immotile sperm (%)	33.55 (25.24, 50.92)
Normal form (%)	6.42 (5.21, 7.89)
Abnormal head (%)	53.77 (42.32, 66.29)
Abnormal midpiece (%)	11.34 (8.09, 15.00)
Abnormal principal piece (%)	12.64 (5.01, 29.11)
Combined abnormality (%)	8.2 (6.49, 13.39)
Nutrient intake	
Total energy intake (kcal/d)	2,495.08 (613.61)
Carbohydrate intake (% E)	43.58 (6.50)
Protein intake (% E)	17.15 (3.41)
Fat intake (% E)	36.86 (6.20)
Monounsaturated fatty acids (% FE)	39.55 (6.79)
Polyunsaturated fatty acids (% FE)	12.2 (4.04)
Saturated fatty acids (% FE)	31.51 (6.24)
Dietary fiber intake (g/d)	21.53 (9.42)
Alcohol intake (g/d)	9.51 (15.12)

Values are presented as mean (standard deviation) or median (interquartile range).
BMI: body mass index, E: energy, FE: fat energy, HDL-c: high-density lipoprotein, LDL-c: low-density lipoprotein, VLDL-c: very low-density lipoprotein.

Table 2. Glycemic index and load and their Least Absolute Shrinkage and Selection Operator (LASSO) regression coefficients for semen parameters at baseline

Semen parameter	Glycemic index	Glycemic load
Total sperm count	0.022	0.004
Sperm concentration	0.010	7.764×10^{-4}
Total motility (progressive and non-progressive motility)	0.010	0.001
Immotile sperm	-0.007	-7.183×10^{-4}

The models for glycemic load were adjusted for age, body mass index, total energy intake, intake of protein, alcohol, monounsaturated fatty acids, polyunsaturated fatty acids, saturated fatty acids and fiber. The same potential confounders plus carbohydrates intake were used to adjust models with glycemic index as exposure.

Table 3. Glycemic index at baseline and their Least Absolute Shrinkage and Selection Operator (LASSO) regression coefficients for changes in semen parameters

Changes in semen parameter	Glycemic index
pH	7.24×10^{-4}
Total sperm count	-8.077×10^{5}
Vitality	0.052
Total motility (progressive and non-progressive motility)	-0.150
Immotile sperm	0.110
Abnormal midpiece	0.013

The models were adjusted for age, intervention group, body mass index, total energy intake, intake of protein, alcohol, carbohydrates, monounsaturated fatty acids, polyunsaturated fatty acids, saturated fatty acids, fiber intake and each semen parameter at baseline.

Table 4. Glycemic load at baseline and their Least Absolute Shrinkage and Selection Operator (LASSO) regression coefficients for changes in semen parameters

Changes in semen parameter	Glycemic load
Total sperm count	2.835×10^{5}
Vitality	0.002

The models were adjusted for age, intervention group, body mass index, total energy intake, intake of protein, alcohol, monounsaturated fatty acids, polyunsaturated fatty acids, saturated fatty acids, fiber intake and each semen parameter at baseline.

sperm count after adjusting for potential confounders (Table 4).

DISCUSSION

In this sub-study conducted within the FERTINUTS trial, we found that GI was negatively associated with changes in total sperm count and total motility. We also observed positive associations of GI with changes in pH, vitality, abnormal midpiece and immotility. Given that GI has been related to elevated glucose and insulin levels, as well as increased inflammation and OS, it is plausible to anticipate a relationship between poor carbohydrate quality and a decrease in semen quality [14]. On the other hand, GL was consistently associated with better semen parameters in both cross-sectional and prospective analyses, suggesting a beneficial role of carbohydrate quantity on sperm quality.

Emerging evidence indicates the importance of hyperglycaemia and insulin sensitivity in relation to fertility outcomes. Chronic elevation of blood glucose levels can lead to hyperinsulinemia and insulin resistance. Consequently, reduced glucose uptake by sperm cells could impair sperm motility because of the vital role of glucose as the primary energy source for spermatozoa [15,16].

Furthermore, insulin resistance has been linked to the disruption of the hormonal environment, affecting several fertility-related hormones involved in fertility. Men with diabetes mellitus (DM) exhibit an altered hypothalamus-pituitary-gonadal axis, resulting in reduced levels of follicle-stimulating hormone, luteinizing hormone and testosterone, which in turn affects Leydig cells, Sertoli cells and finally spermatogenesis [17]. A previous meta-analysis of observational studies revealed that DM influenced semen parameters such as volume and sperm motility, although the included studies were highly heterogeneous [18].

According to existing evidence, hyperglycaemia, high glucose variability and insulin resistance are strongly associated with OS [19]. OS has been found to have detrimental effects on sperm cells, through DNA damage, apoptosis and oxidation of plasma membrane lipids [17,20]. In preclinical models, OS has been associated with adverse effects on total sperm count and morphology [17].

Thus, our results show that higher GI is associated with adverse changes in specific semen parameters, supporting the essential role of glucose/insulin metabolism and OS in sperm quality. Previous prospective studies have suggested a potential link between increased consumption of sugar-sweetened beverages and reduced fecundability, although semen parameters were not measured [21]. Also, in a case-control study a higher intake of vegetables and fruit was associated with lower risk of asthenozoospermia, while individuals with a high intake of sweets showed an elevated risk. No significant associations were observed for refined grains or whole grains intake [22]. Considering that high-GI diets contribute to elevated glycemic and insulinemic levels, adopting dietary patterns that reduce chronic glycaemic spikes (i.e., low dietary GI) may be beneficial for enhancing fertility outcomes due to their influence on glucose homeostasis, hormonal balance, inflammation and OS.

Conversely, we found that GL was positively associated with changes in total sperm count and vitality. A plausible explanation for this finding includes the nature of the GL, which is based on the amount of carbohydrate intake and its GI. Interestingly, consuming a high quantity of carbohydrates with a low daily GI can have a similar impact on dietary GL as consuming high-GI products, but with different physiological responses. Specifically, the same amount of carbohydrate from high-GI foods leads to higher glycemia and demand for insulin, resulting in chronic stimulation of pancreatic β-cells that may lead to insulin resistance. Therefore, altering the source of dietary carbohydrates, rather than simply the carbohydrate quantity, could have a specific impact on glucose homeostasis, hormonal environment, metabolic outcomes and fertility [23,24]. In fact, low-GI diets have been suggested to offer greater benefits on metabolic outcomes, indicating the importance of carbohydrate quality in relation to chronic diseases [23,25].

Unexpectedly, both GI and GL were positively associated with total sperm count, sperm concentration or total motility at baseline. Discrepant results have been found in other cross-sectional studies that examined the impact of high-GI foods on semen parameters. For instance, some studies suggested that intake of sugar-sweetened beverages may be associated with lower sperm concentration and total sperm count, indicating potential negative effects on testicular function [26]. However, a recent study found no association between sugar-sweetened beverages intake and semen quality [27] while other cross-sectional studies reported an unexpected positive association between energy drinks or sugar-sweetened beverages and morphologically normal sperm [26,28]. Our results are not in line with these findings, as we did not find a link between carbohydrate quality and sperm cell abnormality. Additionally, other studies have shown that the intake of some carbohydrate-rich food groups, such as cereals and legumes, was positively associated with sperm concentration, while cereals and fruits intake appeared to increase sperm motility. No associations were found for sweets foods and soft drinks [29]. In accordance with these findings, our results suggest that dietary GL, which considers the quantity of carbohydrates consumed, may be associated with higher total sperm count and motility.

In any case, it should be noted that all these studies evaluated the influence of specific carbohydrate-rich food on semen quality, instead of considering the overall dietary carbohydrate quality. Hence, the ability to generalize our findings is limited. One possible biological explanation for our results includes the role of glucose as the main energy substrate for sperm cells. Insufficient of glucose uptake could compromise the formation and functionality of fully competent spermatozoa [16]. Additionally, the absence of insulin can lead to a hormonal dysregulation, as insulin levels influence the hypothalamus-pituitary-gonadal axis and insulin can stimulate the release of gonadotropin-releasing hormone [17]. Therefore, although acute intake of glucose or stimulation of insulin levels may enhance sperm functionality, further research is required to elucidate the acute and chronic effects of carbohydrates intake on fertility.

To the best of our knowledge, this is the first study to examine the associations of GI and GL with semen quality. However, our study presents some limitations that need to be mentioned. First, this is a secondary analysis of the FERTINUTS trial which limits our ability to infer causality. To address potential reverse causality, we conducted both cross-sectional and prospective approaches. Second, although we adjusted the regression models for known potential confounders, we cannot exclude the possibility of residual or unmeasured confounding. Third, the relatively small sample size of our study may have decreased statistical power. Fourth, assigning GI values to each food is subject to variation due to food processing, cooking methods or food origin [13]. Finally, the generalizability of the findings to other male populations cannot be disregarded.

CONCLUSIONS

In summary, our study suggests that higher baseline GI is associated with adverse changes in semen quality among healthy young men, including decreased motility, abnormal morphology and reduced total sperm count. However, these results were not consistently observed in the cross-sectional analysis. On the other hand, higher baseline GL was consistently associated with better sperm quality parameters in both cross-sectional and prospective analyses. The impact of carbohydrate quality and quantity on fertility remains uncertain and larger prospective studies are needed to validate these findings and to gain a deeper understanding of the role of carbohydrate intake in sperm quality and male fertility.

Conflict of Interest

The authors have nothing to disclose.

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Author Contribution

Conceptualization & data curation: MB. Formal analysis: JMF, CP, MB. Funding acquisition: MB. Writing – original draft: JMF, CP, MB. Writing – review & editing: LGT, MR, NNF, HM, MB.

Supplementary Materials

Supplementary materials can be found via https://doi.org/10.5534/wjmh.230328.

Data Sharing Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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