

# The Influence of Vitamin E and Omega-3 Fatty Acids on Reproductive Health Indices Among Male Workers Exposed to Electromagnetic Fields

American Journal of Men's Health  
January-February 1–16  
© The Author(s) 2022  
Article reuse guidelines:  
sagepub.com/journals-permissions  
DOI: 10.1177/15579883221074821  
journals.sagepub.com/home/jmh  


Hamzeh Mohammadi<sup>1</sup>, Farideh Golbabaie<sup>2</sup>,  
Somayeh Farhang Dehghan<sup>2</sup> , Hossein Imani<sup>3</sup>,  
Fahimeh Ramezani Tehrani<sup>4</sup>,  
and Soheila Khodakarim Ardakani<sup>5</sup>

## Abstract

The present study aims to investigate the effects of using the supplementation of vitamin E and Omega 3 fatty acids on reproductive indices among workers in an automobile parts manufacturing plant. The effect of exposure to electromagnetic fields on certain sex hormones and sperm parameters will also be assessed. The participants were deployed into four groups as per the double-blind block randomization method. Semen parameters and sex hormones of the participants were analyzed before and after 3-month consumption of supplements. The level of workers' exposure to low-frequency magnetic and electrical fields was measured through the recommendation of National Institute for Occupational Safety and Health. Univariate analysis of variance indicated that exposure to electric fields had a statistically significant effect on sperm count, morphology, and motility. The simultaneous consumption of vitamin E + Omega 3 had a statistically significant effect on sperm morphology and motility.

## Keywords

occupational exposure, electromagnetic fields, productive health, nutritional supplements

Received October 16, 2021; revised November 24, 2021; accepted January 4, 2022

## Introduction

The knowledge of the effects of hazardous occupational agents on male fertility is limited; this is while workers are increasingly being exposed to hazardous physical and chemical agents in industrial settings which can be detrimental to the male reproductive system. Unlike other diseases such as cancer, problems in the reproductive organs only become apparent shortly before the onset of serious injury. The protection of workers against those occupational exposures that are influential on the reproductive system is of the utmost importance and can also prevent additional adverse health consequences (Abdollahi et al., 2021; Mohammadi et al., 2021).

Technological advancements in the industry and the ever-increasing use of electrical devices emitting electromagnetic fields (EMFs) have caused concern among scientists and the general public regarding the biological

<sup>1</sup>Department of Occupational Health, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran

<sup>2</sup>Environmental and Occupational Hazards Control Research Center, School of Public Health and Safety, Shahid Beheshti University of Medical Sciences, Tehran, Iran

<sup>3</sup>Department of Clinical Nutrition, School of Nutritional Sciences and Dietetics, Tehran University of Medical Sciences, Tehran, Iran

<sup>4</sup>Reproductive Endocrinology Research Center, Research Institute for Endocrine Sciences, Shahid Beheshti University of Medical Sciences, Tehran, Iran

<sup>5</sup>Department of Biostatistics, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran

### Corresponding Author:

Somayeh Farhang Dehghan, Environmental and Occupational Hazards Control Research Center, School of Public Health and Safety, Shahid Beheshti University of Medical Sciences, Daneshjoo Boulevard, Velenjak, Shahid Chamran Highway, Tehran 198353-5511, Iran.

Emails: somayeh.farhang@sbm.ac.ir; somayeh.farhang@gmail.com



effects of exposure to these fields. Studies have reported a potential link between exposure to low-frequency EMFs and hazardous effects on fertility as well as changes in the male reproductive system (Mohammadi et al., 2021; Suri et al., 2020). The male reproductive organs are perhaps the most sensitive organ to electromagnetic radiation (Y. Liu et al., 2015). Exposure to EMFs can affect the polarization of the cell membrane. Inadequate polarization of the cell membrane is the cause of many problems associated with the synthesis and secretion of testosterone. This includes lower testosterone in the bloodstream as well as lower testosterone to estradiol ratio which leads to reduced spermatogenesis and infertility (Kesari & Behari, 2012). Reduced testosterone levels could also be a result of damage to Leydig cells due to EMF exposure, which reduces the reaction of these cells to the Luteinizing hormone (LH) pulse (Sepahmanesh et al., 2014).

Environmental stressors (hazardous agents) can increase the number of free radicals within the body. Of these free radicals, reactive oxygen species (ROS) are most important. ROSs are produced via various metabolic routes such as the aerobic metabolism involved in mitochondrial respiration and thus play an important role in tissue damage caused by metabolic stress (Poprac et al., 2017). Excessive amounts of ROSs are involved in the onset of male infertility. Due to the specific morphology of sperm cells, antioxidant enzymes within the sperm cell are incapable of protecting the plasma membrane as well as around the acrosome and tail. The health and fertility of the sperm cell are highly dependent on the availability of antioxidants (Arbabian et al., 2018). Lower levels of antioxidants within the ejaculate as well as higher levels of antioxidants in infertile men have already been observed in certain studies (Gharagozloo et al., 2016). It is not a surprise that antioxidant supplements have become popular in recent years as they play an extensive role in curing male infertility. These compounds protect the spermatozoa, prevent the maturation of spermatids, and increase the motility of spermatozoa (Gharagozloo et al., 2016).

Sperm cells contain large quantities of polyunsaturated fatty acids (PUFA). Healthy spermatozoa have a higher percentage of PUFAs (n-3 and n-6) compared with that found in blood serum or other cell membranes (Rooke et al., 2001). Evidence suggests that sperm count can be influenced by fatty compounds as well as PUFA levels within the sperm (Safarinejad & Safarinejad, 2012). Due to the lack of enzymes that can synthesize n-3 or n-6, these types of fatty acids are not naturally produced in animals. These fatty acids need to be supplemented via a food diet as they are required for many processes such as growth, reproduction, sight, and brain development (Gurr et al., 2002). Omega 3 ( $\omega$ -3 or n-3) fatty acids have anticoagulant effects and can increase catalase levels in the peroxisome and cytoplasm, thus improving antioxidant properties (Shahi et al., 2017). Nonetheless, a recent study

suggests that high levels of Omega 3 fatty acids can increase the potential for lipid peroxidation although this can be prevented by the simultaneous use of vitamin E (Wander et al., 1996). Vitamin E is an important part of the human diet and is perhaps the most effective antioxidant agent that is soluble in fat (Al-Attar, 2011).

Reproductive indices can be affected by various factors such as heredity, age, background disorders, diet, and the use of pharmaceutical or recreational drugs. Exposure to hazardous physical or chemical agents in the workplace is also important in this regard. The present study aims to investigate the effect of using the supplementation of vitamin E and Omega 3 fatty acids on reproductive indices as preventive administrative measures in workplaces. In addition, the effects of exposure to electromagnetic fields on certain sex hormones and sperm parameters among workers in an automobile parts manufacturing plant (mainly dealing with casting processes) will also be assessed. Literature review on this subject reveals no studies have looked at the role of simultaneous supplementation of vitamin E and Omega 3 fatty acids on the reproductive indices of workers. A relatively low number of field studies were conducted on the effect of EMFs on reproductive indices among workers.

## Method

The present study is a clinical trial that was registered and approved by the Iranian Registry of Clinical Trials with the registration number ID: IRCT20210207050290N1. This was done according to sample size calculations while also considering entry criteria and filling out a written consent form approved by the ethics committee (IR.SBMU.PHNS.REC.1399.157). Due to cultural and religious considerations, semen samples were only taken from married men. The inclusion criteria were age 20 to 50 years (Zitzmann, 2013); having 2 or more years of employment (Klonoff-Cohen, 2005); not using steroids, testosterone, prednisolone, antioxidants (Ball et al., 2001) such as selenium, vitamin C, E or B supplements (Yousef et al., 2003), and body-building supplements (Sinclair, 2000); having no family history of infertility or organic disorders affecting fertility such as diabetes, kidney disorders, angina pectoris, heart disorders, arterial hypertension, pituitary disorders or a chronic pulmonary obstruction disorder; not suffering from testicular infections, orchitis, and varicocele (Sabeti et al., 2016); and no having history of chemotherapy or radiotherapy (Gandini et al., 2006). The background information was obtained from demographic questionnaires and by referring to the participants' medical records (Mohammadi et al., 2021).

According to the formula for the calculation of sample quality, the sample size was determined to be 23 per group, adding to a total of 92 participants (23×4). The participants were deployed into four groups as per the

double-blind block randomization method. The first group was given vitamin E (100 mg) accompanied by a placebo of Omega 3 fatty acids. The second group was given Omega 3 fatty acids (180 mg eicosatetraenoic acid [EPA] and 120 mg docosahexaenoic acid [DHA]) accompanied by a placebo of vitamin E. The third group was given vitamin E along with Omega 3 fatty acids. Finally, the fourth group acted as a placebo group and was given placebos of both vitamin E and Omega 3 fatty acids.

Semen parameters of the participants were analyzed before and after the consumption of supplements. Sex hormone levels within the blood serum were analyzed again after a 3-month supplement consumption period. The workers were asked to complete 3-day self-recall questionnaires in both the initial stage and the final stage of the intervention. The level of physical activity of all participants was thus evaluated per The International Physical Activity Questionnaires (IPAQ).

Of the 92 participants, 80 remained in the study until the end, with others leaving mostly due to changes in employment, complications due to the Covid-19 pandemic, leaving the job, and the unwillingness to continue the project. The placebo sample used for vitamin E and Omega 3 fatty acids were obtained from the Daana Pharma Co. (Tabriz, Iran) and Karen Pharma and Food Supplement Co. (Tehran, Iran), respectively.

The low-frequency magnetic field and electric field were measured based on the guideline provided by National Institute for Occupational Safety and Health (1998) using a calibrated TES 1393 Electromagnetic Field Tester (TES Electronics Co., Taiwan) and a TES 593 Electrosmog meter (TES Electronics Co., Taiwan), respectively.

The Enzyme-Linked Immunosorbent Assay (ELISA) method was used to analyze the serum level of sex hormones including free serum testosterone levels, luteinizing hormone (LH), and follicle-stimulating hormone (FSH). Blood samples taken between 7 and 9 a.m. were prepared and analyzed according to the kit manufacturer's guidelines (Free Testosterone AccuBind ELISA test system, Monobind Inc., USA, and LH/FSH Padtan Gostar Isar Inc., Iran) and using a STAT FAX 2100 EIA analyzer and microplate reader.

Details regarding the particular case industry, methods of EMF monitoring, and sex hormone measurement methods are explained in detail in the authors' previous study (Mohammadi et al., 2021). Supplement consumption and semen analysis will be explained further in the Supporting Information.

### **Nutritional Supplementation**

Throughout the intervention, the participants were told to maintain their regular diet and physical activity and

refrain from changing the dosage of the supplements without informing the researchers. Before and during the intervention, the necessary guidelines regarding dosage, method of consumption, when to stop consumption, potential side effects, cautionary advice, and proper storage methods were provided to the participants. During the intervention, the researchers would contact (via call phone and message) the participants every 2 weeks to ensure the proper execution of protocols by the participants. A run-in period was also implemented prior to the intervention where participants were asked to try and stabilize their life for a 2-week period in terms of their diet, physical activity, and work requirements to prepare for the study. A daily usage chart was also given to participants to remind them to take the supplements. This chart would then be handed back to the researchers after the intervention was complete and would help determine how well the participants had maintained their supplement regiment.

Participants were prescribed one capsule of their respective supplements each day for 3-month period. The placebo group would be given identical capsules containing edible paraffin instead (Supplemental Figure S1) with identical containers being used for both the supplements and the placebos. As per the double-blind method, a third party would be responsible for logging the containers and providing them to the researchers. This means that neither the researchers nor the participants had any knowledge regarding the contents of the containers.

Estimating the amount of nutrients received by the participants was done with the help of a 3-day food intake report made using the Nutritionist software v.4 (Tinuviel Software, Warrington, United Kingdom). The data obtained before and after intervention from the Nutritionist software were entered into Microsoft Excel. As the present study aims to evaluate the effects of supplements on reproductive indices, the food intake of the participants must be monitored via self-report before and after the intervention to detect any significant changes. This is done via the paired sample *t*-test that calculates mean differences for minor and major nutrients both before and after the intervention. The results indicated no significant changes in food intake among the participants ( $p > .05$ ).

The level of physical activity of each participant was evaluated by having them complete the IPAQ before and after the intervention. This questionnaire tracks various day-to-day physical activities that the participant has performed in the past 7 days and includes work or household activities, physical exercise, and leisure-time physical activities. The questionnaire categorizes physical activity into walking, moderate and vigorous, while also providing guidelines on how to calculate the overall energy used for physical activity in the past 7 days. Overall energy intensity of <600 MET/CAL per walking is categorized

as low, while a value of 600 to 3,000 MET/CAL per week is categorized as moderate and a value of above 3000 MET/CAL per week is categorized as vigorous (Moeini et al., 2011).

### Semen Analysis

Semen samples were taken in the sampling room of the fertility clinic at the Sarem Hospital and were further analyzed at their laboratory (Supplemental Figure S2). The participants were required to refrain from ejaculation for at least 3 days before sampling and were instructed to avoid using condoms, soap, or any type of gel during sampling. As per the World Health Organization (WHO), the time between sample collection and transference to the lab was less than 1 hr while the time required to prepare and analyze the samples was less than 3 hr. The semen samples were placed inside a sterile sample container with no contamination. Parameters such as quality of motility, sperm count per 1 ml, and irregular morphology were determined according to the WHO guidelines for human sperm sample laboratory analysis (Cao et al., 2011).

### Statistical Analysis

Statistical data analysis was performed using SPSS v.22 (Chicago, IL, USA). Descriptive statistics such as frequency, percentage, mean, median, percentile, and standard deviation were used to present the results related to the hormone analysis and semen quality parameters both before and after supplement consumption. The Shapiro test was used to determine data distribution normality. The median difference between pre- and post-intervention values of variables was assessed by Wilcoxon Signed Ranks Test. The median of variables between different studied groups was compared using the Kruskal-Wallis test or chi-square test. The effect size of the dietary supplements on the target parameters as well as the predictive model for the effect mechanism was estimated using univariate analysis of variance. A significance level of 0.05 was considered for the present study.

### Results

Table 1 presents descriptive-analytic statistics regarding demographic variables observed among the participants. As per the Shapiro test, the majority of demographic variables followed a non-normal distribution except for body mass index (BMI;  $p < .05$ ). Mean age and employment duration among the participants were 33.57 (5.19) years and 8.30 (5.59) years, respectively. Of the participants, 61% ( $n = 49$ ) were occupied in the grinding section and 64% ( $n = 51$ ) of the participants had a high school

diploma or a higher education. In all, 43% ( $n = 34$ ) of the participants had a normal BMI and 37% ( $n = 30$ ) had a healthy waist-hip ratio (Noble, 2001). Around 44% ( $n = 61$ ) of participants exercised regularly, and 69% ( $n = 55$ ) of participants were nonsmokers (past or present). All of the participants had extreme physical activity according to the IPAQ questionnaires.

Table 2 presents descriptive/analytic statistics regarding exposure to the electrical and magnetic fields for both the exposure groups and the supplement groups. As per the Shapiro test, data regarding exposure followed a non-normal distribution ( $p < .05$ ). Results obtained from comparing supplement groups show no statistically significant difference in the amount of exposure to electrical or magnetic fields whether in terms of overall exposure or in terms of categorized exposure levels ( $p > .05$ ).

Table 3 presents descriptive-analytic statistics regarding sex hormone levels among the participants before and after the intervention. The difference in the level of testosterone before and after the intervention was not statistically significant for any age group. In addition, no statistically significant difference in testosterone levels before and after intervention was observed between the supplement groups. After the intervention, mean testosterone levels had increased in the supplement groups. The lowest testosterone level before intervention was observed among the vitamin E + Omega 3 group while after intervention this was true for the placebo group. Differences in follicle-stimulating hormone (FSH) and LH hormone levels before and after the intervention were not statistically significant in any of the supplement groups (except for FSH in the vitamin E + Omega 3 group). The difference in the level of these hormones before and after the intervention was not statistically significant for any of the various supplement groups.

Table 4 presents descriptive-analytic statistics regarding certain sperm parameters observed among the participants both before and after the intervention. Differences in sperm count and sperm with full motility before and after the intervention were not statistically significant in any of the supplement groups (except for vitamin E + Omega 3,  $p = .016$  and  $p = .003$ , respectively). The difference in sperm count and sperm with full motility between the supplement groups before and after the intervention was not statistically significant either. Overall, mean sperm count and sperm with full motility among supplement groups had increased after the intervention. A significant difference was observed in the percentage of sperm with normal morphology before and after intervention in the Vitamin E + Omega 3 group as well as the vitamin E group. No significant difference was observed in the percentage of sperm with normal morphology between the supplement groups before or after the intervention. Overall, the supplement groups had a higher

**Table 1.** Demographic Characteristics of Participants.

Parameter	Statistical measure (N = 80)				Supplementation group				p value <sup>a</sup>
	Vitamin E (N = 22)		Omega-3 (N = 21)		E+ omega-3 (N = 19)		Placebo (N = 18)		
	M (SD)	Median (IQR)	M (SD)	M (SD)	M (SD)	M (SD)	M (SD)	M (SD)	
Age (years)	33.57 (5.91)	32.50 (11.75)	34.27 (6.27)	31.85 (6.33)	35.52 (5.33)	32.66 (4.99)	32.66 (4.99)	32.66 (4.99)	.144
Employment duration (years)	8.30 (5.59)	6.50 (7.75)	10.31 (5.96)	6.33 (4.79)	10.63 (5.93)	5.66 (3.72)	5.66 (3.72)	5.66 (3.72)	.005
Manufacturing Section									
Aluminum	17 (22)		9 (52.9)	0 (0)	4 (23.5)	4 (23.5)	4 (23.5)	4 (23.5)	.004
Cast iron	14 (17)		3 (21.4)	2 (14.3)	7 (50)	2 (14.3)	2 (14.3)	2 (14.3)	
Grinding	49 (61)		10 (20.4)	19 (38.8)	8 (16.3)	12 (24.5)	12 (24.5)	12 (24.5)	
Education N (%)									
Illiterate	2 (2)		0 (0)	0 (0)	1 (50)	1 (50)	1 (50)	1 (50)	.838
Primary school	7 (9)		2 (28.6)	1 (14.3)	2 (28.6)	2 (28.6)	2 (28.6)	2 (28.6)	
Junior high school	20 (25)		5 (25)	6 (30)	3 (15)	6 (30)	6 (30)	6 (30)	
High school Diploma	44 (55)		13 (29.5)	12 (27.3)	13 (29.5)	6 (13.6)	6 (13.6)	6 (13.6)	
Associate Degree	4 (5)		1 (25)	1 (25)	0 (0)	2 (50)	2 (50)	2 (50)	
Bachelor's degree	3 (4)		1 (33.3)	1 (33.3)	0 (0)	1 (33.3)	1 (33.3)	1 (33.3)	
BMI (kg/m <sup>2</sup> )	25.06 (4.49)	24.79 (6.23)	27.98 (3.48)	23.88 (4.65)	25.93 (4.32)	21.95 (3.13)	21.95 (3.13)	21.95 (3.13)	.001 <sup>b</sup>
N (%)									
Underweight (<18.5)	6 (7)		0 (0)	2 (33.3)	1 (16.7)	3 (50)	3 (50)	3 (50)	.009
Normal (18.5-24.9)	34 (43)		3 (8.8)	10 (29.4)	9 (26.2)	12 (35.3)	12 (35.3)	12 (35.3)	
Overweight (25-29.9)	27 (34)		13 (48.1)	6 (22.2)	5 (18.5)	3 (11.1)	3 (11.1)	3 (11.1)	
Obese (>30)	13 (16)		6 (46.2)	3 (23.1)	4 (30.8)	0 (0)	0 (0)	0 (0)	
WHR	0.93 (0.21)	0.93 (0.12)	0.96 (0.06)	0.89 (0.06)	0.97 (0.43)	0.89(0.05)	0.89(0.05)	0.89(0.05)	.002
N (%)									
Healthy (≤0.9)	30 (37)		3 (10)	10 (33.3)	5 (16.7)	2 (40)	2 (40)	2 (40)	.003
Unhealthy (>0.9)	50 (63)		19 (38)	11 (22)	14 (28)	6 (12)	6 (12)	6 (12)	
Smoker-N (%)									
Yes	25 (31)		7 (28)	8 (32)	6 (24)	4 (16)	4 (16)	4 (16)	.594
No	55 (69)		15 (27)	13 (24)	13 (24)	14 (25)	14 (25)	14 (25)	
Regular exercise N (%)									
Yes	44 (55)		13 (29.5)	15 (34.1)	7 (15.9)	9 (20.5)	9 (20.5)	9 (20.5)	.161
No	36 (45)		9 (25)	6 (16.7)	12 (33.3)	9 (25)	9 (25)	9 (25)	
Physical activity (IPAQ score)									
Walking	—	—	—	—	—	—	—	—	—
Moderate	—	—	—	—	—	—	—	—	—
Vigorous	80 (100)	80 (100)	22 (100)	21 (100)	19 (100)	18 (100)	18 (100)	18 (100)	

Note. IQR = Interquartile Range; BMI = Body Mass Index; WHR = Waist to Hip Ratio; IPAQ = International Physical Activity Questionnaire.

<sup>a</sup>Differences in variables between different manufacturing units (Kruskal-Wallis Test / Chi-squared Test). <sup>b</sup>Mean differences of BMI between different manufacturing units (One-way analysis of variance [ANOVA]).



**Table 2.** Exposure Levels of Participants to Electromagnetic Fields.

Exposure	Categorized values	Supplementation group												p value <sup>a</sup>			
		Total (N = 80)			Vitamin E (N = 22)			Omega-3 (N = 21)			E + omega-3 (N = 19)				Placebo (N = 18)		
		M (SD)	Median (interquartile range)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)		N (%)		
Magnetic field ( $\mu\text{T}$ )	—	1.95 (5.31)	0.57 (0.7)	—	2.25 (7.73)	—	0.79 (0.63)	—	3.48 (6.5)	—	1.31 (2.72)	—	0.384				
	<0.39 (N = 25)	0.22 (0.08)	0.21 (0.16)	8 (32)	0.16 (0.07)	4 (16)	0.3 (0.04)	5 (20)	0.18 (0.03)	8 (32)	0.26 (0.06)	8 (32)	.121				
	0.39–0.86 (N = 28)	0.55 (0.14)	0.57 (0.23)	8 (28.6)	0.53 (0.08)	12 (42.9)	0.55 (0.17)	4 (14.3)	0.6 (0.14)	4 (14.3)	0.53 (0.18)	4 (14.3)					
Electrical Field (V / m)	>0.86 (N = 27)	4.99 (8.44)	1.61 (1.65)	6 (22.2)	7.33 (14.45)	5 (18.5)	1.73 (0.64)	10 (37)	6.28 (8.13)	6 (22.2)	3.22 (4.3)	6 (22.2)	.951				
	—	0.61 (0.72)	0.25 (0.77)	—	0.6 (0.69)	—	0.59 (0.59)	—	0.8 (0.96)	—	0.53 (0.64)	—					
	<0.18 (N = 26)	0.13 (0.04)	0.13 (0.05)	7 (26.9)	0.13 (0.03)	6 (23.1)	0.13 (0.001)	6 (23.1)	0.1 (0.06)	7 (26.9)	0.14 (0.01)	7 (26.9)	.977				
	0.18–0.37 (N = 25)	0.24 (0.04)	0.25 (0.07)	7 (28)	0.25 (0.05)	8 (32)	0.23 (0.03)	6 (24)	0.26 (0.04)	4 (16)	0.23 (0.03)	4 (16)					
	>0.37 (N = 29)	1.36 (0.47)	1.5 (1.03)	8 (27.6)	1.32 (0.71)	7 (24.1)	1.19 (0.61)	7 (24.1)	1.87 (0.79)	7 (24.1)	1.08 (0.76)	7 (24.1)					

<sup>a</sup>Differences in variables between different studied groups (Kruskal-Wallis Test / Chi-square Test).

**Table 3.** Descriptive-Analytical Statistics Regarding Sex Hormone Levels of Participants Before and After Intervention.

Hormone	Categorized values	Supplementation group																							
		Vitamin E (N = 22)				Omega-3 (N = 21)				E+ omega-3 (N = 19)				Placebo (N = 18)											
		Before	After	Before	After	Before	After	Before	After	Before	After	Before	After												
Free Testosterone (pg/ml) for 20-39 years old (N = 57) for		p-value <sup>c</sup>	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	M (SD)	N (%)	p-value <sup>a</sup>	p-value <sup>b</sup>
<9.2		.460	—	5.44 (4.89)	—	6.34 (3.62)	—	6.27 (2.55)	—	7.22 (2.71)	—	5.07 (1.86)	—	6.08 (2.22)	—	6.21 (1.91)	—	6.05 (2.18)	—	6.21 (1.91)	—	6.05 (2.18)	—	.497	.109
Testosterone (pg/ml) for 20-39 years old (N = 57)			13 (86.7)	5.42 (1.59)	11 (73.3)	4.42 (1.59)	15 (88.2)	6.18 (1.36)	14 (82.4)	6.18 (1.36)	9 (100)	5.46 (1.27)	8 (88.9)	5.46 (1.27)	16 (100)	5.54 (1.81)	14 (87.5)	5.54 (1.81)	—	16 (100)	5.54 (1.81)	14 (87.5)	—	—	—
Normal Range (9.2-34.6)			2 (13.3)	110.6 (1.60)	4 (26.7)	11.60 (1.60)	2 (11.8)	12.10 (1.96)	3 (17.6)	12.10 (1.96)	0 (0)	—	1 (11.1)	—	0 (0)	—	2 (12.5)	—	—	0 (0)	—	2 (12.5)	—	—	—
>34.6		.611	—	5.97 (2.39)	—	6.28 (0.77)	—	5.97 (2.39)	—	6.28 (0.77)	—	5.64 (2.49)	—	7.01 (2.44)	—	5.10 (0.98)	—	4.05 (0.14)	—	5.10 (0.98)	—	4.05 (0.14)	—	.122	.911
Free Testosterone (pg/ml) for 40 years and older (N = 23) for			3 (42.9)	3.93 (0.75)	4 (57.1)	5.42 (0.47)	3 (42.9)	3.93 (0.75)	4 (57.1)	5.42 (0.47)	5 (50)	3.56 (0.97)	2 (20)	3.90 (0.47)	2 (100)	5.10 (80.9)	2 (100)	5.40 (0.14)	—	2 (100)	5.10 (80.9)	2 (100)	5.40 (0.14)	—	—
Normal Range (6.1-30.3)			4 (57.1)	8.05 (1.17)	3 (42.9)	6.70 (0.20)	4 (57.1)	6.70 (0.20)	3 (42.9)	6.70 (0.20)	5 (50)	7.27 (1.51)	8 (80)	7.78 (1.86)	0 (0)	—	0 (0)	—	—	0 (0)	—	0 (0)	—	—	—
>30.3		.258	—	2.50 (1.79)	—	2.03 (1.68)	—	2.50 (1.79)	—	2.03 (1.68)	—	5.64 (2.49)	—	1.75 (1.89)	—	3.55 (4.10)	—	3.05 (2.67)	—	3.55 (4.10)	—	3.05 (2.67)	—	.460	.410
FSH (mIU/ml)			5 (22.7)	0.38 (0.52)	9 (40.9)	0.56 (0.29)	6 (28.6)	0.52 (0.38)	9 (40.9)	0.56 (0.29)	4 (21.1)	3.56 (0.97)	8 (42.1)	0.48 (0.22)	4 (22.2)	0.77 (0.25)	4 (22.2)	0.77 (0.25)	—	4 (22.2)	0.77 (0.25)	8 (42.1)	0.48 (0.22)	—	—
<1			17 (77.3)	3.08 (1.60)	13 (59.1)	3.05 (1.48)	14 (66.7)	3.08 (1.60)	13 (59.1)	3.05 (1.48)	15 (78.9)	7.72 (1.51)	11 (57.9)	2.67 (2.04)	13 (72.2)	3.29 (1.95)	18 (100)	3.05 (2.76)	—	13 (72.2)	3.29 (1.95)	18 (100)	3.05 (2.76)	—	—
Normal Range (1-14)			—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
>14		.413	—	1.65 (1.29)	—	1.79 (0.76)	—	1.72 (1)	—	1.56 (0.56)	—	1.62 (0.7)	—	1.50 (0.33)	—	2.15 (1.16)	—	1.54 (0.18)	—	2.15 (1.16)	—	1.54 (0.18)	—	.945	.213
LH (mIU/ml)			5 (22.7)	0.24 (0.15)	2 (9.1)	—	2 (9.5)	0.14 (0.14)	0 (0)	—	2 (9.5)	0.45 (0.21)	0 (0)	—	3 (16.7)	—	—	—	3 (16.7)	—	—	—	—	—	—
<0.7			17 (77.3)	2.07 (1.17)	20 (90.9)	0.71 (0.71)	19 (90.5)	1.90 (0.94)	21 (100)	1.56 (0.56)	19 (90.5)	1.75 (0.59)	19 (100)	1.50 (0.33)	15 (83.3)	2.44 (1.05)	17 (94.4)	1.59 (0.38)	—	15 (83.3)	2.44 (1.05)	17 (94.4)	1.59 (0.38)	—	—
Normal Range (0.7-7.4)			—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
>7.4			—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—

Note. FSH = Follicle-stimulating hormone; LH = Luteinizing hormone.

<sup>a</sup>Median differences in variables between different studied groups before intervention (Kruskal-Wallis Test). <sup>b</sup>Median differences in variables between different studied groups after intervention (Kruskal-Wallis Test). <sup>c</sup>Median difference between pre- and post-intervention values of variables (Wilcoxon Signed Ranks Test).

**Table 4.** Descriptive-Analytical Statistics Regarding Some Sperm Parameters Before and After Intervention.

Sperm parameter	Supplementation group																				
	Vitamin E(N = 22)				Omega-3(N = 21)				E+ omega-3(N = 19)				Placebo(N = 18)								
	Before		After		Before		After		Before		After		Before		After						
Count (10 <sup>6</sup> /ml)	.155	42.86 (30.00)	42.50 (15.56)	46.81 (15.56)	49.00 (30.80)	.148	38.23 (15.22)	38.00 (27.50)	44.23 (12.28)	42.00 (15.00)	.016	41.50 (20.29)	49.00 (36.25)	53.38 (18.31)	63.50 (22.00)	.670	39.61 (19.14)	40.00 (30.00)	38.55 (19.46)	42.00 (37.00)	.799
Normal Morphology (%)	.007	12.86 (6.47)	13.00 (5.25)	16.40 (5.38)	17.00 (8.00)	.080	10 (7.35)	10 (9.50)	12.33 (7.39)	12.00 (13.50)	0.008	10.77 (5.19)	10.50 (6.75)	18.22 (6.70)	22.00 (9.00)	.333	8.94 (5.16)	10.00 (5.25)	9.61 (8.30)	15.00 (15.75)	.114
Full motility (%)	.114	0.40 (1.18)	00 (0.00)	1.00 (1.92)	0 (0.75)	.317	0.38 (1.2)	0 (1.2)	0.61 (1.56)	0 (0)	.003	0 (0)	0 (0)	2.22 (2.12)	3.00 (4.00)	.258	0.05 (0.23)	0 (0)	0.07 (0.82)	0 (0)	.391
Sluggish motility (%)	.162	31.13 (18.91)	33.50 (29.00)	37.09 (15.61)	44.00 (27.00)	.326	19.66 (17.43)	19.00 (26.50)	23.66 (18.98)	23.00 (40.00)	.003	23.94 (19.47)	24.00 (44.00)	39.94 (17.73)	52.00 (30.00)	.325	19.22 (15.19)	16.00 (27.50)	20.00 (19.73)	27.50 (41.25)	.161
Low motility (%)	.770	37.04 (14.62)	35.50 (26.25)	34.81 (12.62)	31.00 (17.75)	.852	37.14 (15.33)	38.00 (9.00)	36.66 (12.72)	40.00 (16.00)	.268	33.72 (11.22)	31.00 (24.00)	30.50 (14.47)	23.50 (22.00)	.153	33.88 (14.84)	42.00 (24.00)	32.55 (16.52)	41.00 (27.75)	.818
Immotile (%)	.155	31.40 (14.12)	27.00 (15.75)	27.09 (6.09)	27.00 (6.75)	.179	42.80 (20.20)	35.00 (22.50)	38.04 (19.53)	30.00 (26.50)	.010	42.33 (13.05)	48.00 (20.00)	27.33 (17.45)	22.00 (10.00)	.174	46.83 (20.66)	40.00 (27.75)	46.16 (16.23)	39.00 (17.00)	.015

Note. According to the laboratory report, the normal limit of sperm count is >15 million / ml. According to the laboratory report, the normal limit of sperm motility is >32. IQR = Interquartile Range.

<sup>a</sup>Median differences in variables between different studied groups before intervention (Kruskal-Wallis Test). <sup>b</sup> Median differences in variables between different studied groups after intervention (Kruskal-Wallis Test). <sup>c</sup>Median difference between pre- and post-intervention values of variables (Wilcoxon Signed Ranks Test).



**Table 5.** Results of Univariate Analysis of Variance.

Variable	Free testosterone		Immotile		Full + sluggish motility		Normal morphology		Count	
	B	p value	B	p value	B	p value	B	p value	B	p value
Supplementation Group	2.795	.749	-7.206	.759	8.034	.764	5.205	.564	5.260	.917
	3.646	.722	-16.175	.521	21.818	.412	13.499	.167	24.572	.352
	5.160	.377	-48.128	.097	72.211	.021	28.998	.011	48.777	.106
Magnetic fieldp	-0.031	.723	0.506	.418	-1.308	.053	-0.423	.083	-0.471	.469
Electrical Field	-0.593	.373	9.541	.053	-11.564	.027	-4.961	.010	-11.116	.032
Age	-0.293	.014	0.585	.317	-1.344	.034	-0.529	.023	-0.895	.147
Employment Duration	-0.156	.595	0.001	.999	-1.863	.022	-0.625	.026	-1.172	.135
BMI	-0.063	.117	0.856	.228	-0.203	.784	-0.011	.966	-0.489	.505
Free testosterone	—	—	0.975	.331	-1.0418	.182	-0.323	.398	-0.949	.363
FSH	0.205	.522	-3.941	.047	8.201	.001	3.196	.001	4.933	.019
LH	0.110	1.59	6.631	.461	5.033	.334	1.537	.415	3.905	.448
Smoking	-1.399	.888	19.865	.484	-94.24	.003	-41.005	.001	-80.056	.011
Physical Activity	-8.390	.159	-3.221	.829	6.780	.666	1.183	.835	-3.368	.828

Note. BMI = Body Mass Index; FSH = Follicle stimulating hormone; LH = Luteinizing hormone.

mean percentage of sperm with normal morphology after the intervention. The difference in the percentage of immotile sperm before and after the intervention was not statistically significant in any of the supplement groups (except for vitamin E + Omega 3 group,  $p = .001$ ). Overall, the mean percentage of immotile sperm was lower after intervention in the supplement groups. The comparison of the percentage of workers with normal sperm parameters before and after the intervention has been presented in Supplemental Figure S3.

Partial correlation tests with controlling for the effect of demographic parameters indicate that before the intervention, exposure to electric fields had no significant correlation with sperm parameters or testosterone levels ( $R = -.144, p = .230$ ). The sperm parameters including the sperm count ( $R = -.002, p = .988$ ), percentage of sperm with normal morphology ( $R = -.078, p = .517$ ), percentage of motile sperm ( $R = -.029, p = .809$ ), and testosterone levels had a weak yet reverse correlation with all exposure groups. The same applies to magnetic fields. After the intervention, the correlation between exposure to electric fields and both percentages of sperm with normal morphology ( $R = -.276, p = .020$ ) and the percentage of sluggish sperm ( $R = .254, p = .033$ ) was statistically significant.

The effects of the target variables (demographics, exposures, and sex hormones) on free testosterone levels and sperm parameters were analyzed and modeled using univariate analysis of variance, with the results presented in Table 5. Among the exposure variables, electric fields had the largest effect on testosterone levels ( $B = -.593$ ), but this was not statistically significant ( $p = .0373$ ). The use of supplements resulted in an increase in testosterone levels in all groups but this increase was not statistically

significant. The largest effect was observed in the vitamin E + Omega 3 supplement group. Among demographic variables, age had a significant effect on testosterone levels ( $B = -.293, p = .014$ ). The largest significant effect observed among the exposure variables was that of electric fields on sperm count ( $B = -11.116; P = .032$ ). Supplement use improved sperm count in all supplement groups, but this was not statistically significant. The largest effect was observed in the vitamin E + Omega 3 group. Among demographic variables, the effect of smoking on sperm count was also statistically significant ( $B = -80.005, p = .011$ ). Electric fields had the largest significant effect on normal sperm morphology among the exposure variables ( $B = -4.961; p = .001$ ). The effect of supplement use on sperm morphology was significant in the vitamin E + Omega 3 group ( $B = 28.995, p = .011$ ). The effect of supplement use on normal morphology was incremental in other supplement groups but was not statistically significant. Smoking also had a significant effect on sperm morphology ( $B = -41.005, p = .001$ ). Electric fields had the largest effect on full and sluggish sperm motility ( $B = -11.564; p = .027$ ). The effect of supplement use on full and sluggish sperm motility was also significant in the vitamin E + Omega 3 group ( $B = 72.211, p = .021$ ). Supplement use had an incremental effect on sluggish and full sperm motility in all other supplement groups but this was not statistically significant. As for demographic parameters, age ( $B = -1.344, p = .034$ ), employment duration ( $B = -1.863, p = .022$ ), and smoking ( $B = -94.24, p = .003$ ) had significant effects on sluggish and full sperm motility. Electric fields had the largest effect on the percentage of immotile sperm among exposure variables ( $B = 9.541; p = .053$ ). Supplement

use had a decremental effect on the percentage of immotile sperm in all supplement groups but this was not statistically significant.

## Discussion

### *The Effects of Nutritional Supplementation on Reproductive Indices*

As was detailed earlier, the participants were divided into 4 groups in a double-blind random clinical trial study. Comparing the initial and final energy intake, macronutrients and nutrients influential on target variables reveal no significant difference in any of the groups. This indicates that the participants had not made any major changes to their diets as asked.

As per the results, the difference in the level of testosterone, before or after the intervention, was not significant in any of the supplement groups. Overall, a higher testosterone level was observed in the supplement groups after the intervention compared with before intervention. The highest testosterone levels after intervention belonged to the vitamin E + Omega group. The mean level of FSH and LH had gone down overall after the intervention.

Animal studies have established the efficacy of vitamin E and Omega 3 fatty acids on sex hormone levels and sperm parameters. Tran et al. (2016) indicate that Omega 3 fatty acid supplements lead to higher plasma testosterone levels compared with the other two supplement groups in male buffalo (Tran et al., 2016). They observed that a diet rich in Omega 3 fatty acids causes increased testosterone secretion, reduces maturation time, and improves the quality of semen in male buffalo (Tran et al., 2016). Putri et al. (2018) studied the effects of Omega 3 fatty acids on testosterone and the quality of spermatids in rats (Putri et al., 2018). Their results indicated no significant difference in serum testosterone levels or sperm vitality among the groups being studied. The sperm motility ( $p = .039$ ) and sperm morphology ( $p = .047$ ) had been significantly affected by Omega 3 fatty acids. They concluded that overall, Omega 3 can improve sperm motility and morphology resulting in improved fertility (Putri et al., 2018). Gholamhasan et al. (2018) evaluated the effects of vitamin E on the levels of certain sex hormones in male mice and reported that the injection of vitamin E caused a significant increase in testosterone levels as well as a reduction in gonadotropins (FSH and LH).

The physiology of fertility is very complex and many factors are influential in infertility. One of the problems with the male reproductive system is the risk of free radicals. Oxidants have a negative effect on the quality and motility of sperm. Antioxidants such as vitamin E can have beneficial effects on sperm parameters. Free

radicals cause changes in the levels of fertility hormones and can lead to sterility or even a still-birth. The antioxidant effects of this vitamin have already been observed, and it is clear that they can be used to improve the containment of free radicals in the testicles and sperm (Kao et al., 2008; Niki, 2014). Evidence has been growing regarding the benefits of dietary Omega 3 fatty acids, but further research is needed in this regard. Essential fatty acids such as DHA and EPA are not created in the human body, and it is necessary that everyone get a suitable amount each day. These fatty acids can increase testosterone in males by producing prostaglandins (Riediger et al., 2009; Von Schacky, 2006). In the study by Jokar et al., it was demonstrated that Omega 3 fatty acids had caused increased testosterone levels in adult male rats by affecting the testicles and the hypothalamic–pituitary–gonadal axis. These effects were not dose-dependent, and the incremental changes in testosterone were clearly observable (Jokar et al., 2016).

According to our findings, before the intervention, around 6% of participants had sperm with normal morphology and 32% had normal sperm motility. After a 3-month supplement intervention, a 22% increase in the number of sperm with normal morphology as well as a 44% increase in sperm motility was observed. The difference in sperm count before and after the intervention was not statistically significant in any of the supplement groups (except for vitamin E + Omega 3 fatty acids group). The difference in the percentage of sperm with normal morphology before and after the intervention was statistically significant in the vitamin E + Omega 3 fatty acids group. The difference in the percentage of sperm with full motility before and after the intervention was not statistically significant in any of the supplement groups (except for vitamin E + Omega 3 group). Overall, mean sperm count, percentage of sperm with normal morphology, and sperm with full motility was higher after intervention in the supplement groups. The difference in the percentage of immotile sperm before and after the intervention was not statistically significant in any of the supplement groups (except for vitamin E + Omega 3 group). Overall, the mean percentage of immotile sperm was lower after the intervention in all supplement groups. Univariate analysis of variance indicated that the effects of supplement use on sperm count were incremental but not statistically significant. The largest effect was observed in the vitamin E + Omega 3 fatty acids group. The effect of supplement use on sperm morphology and sluggish and full motility was statistically significant in the vitamin E + omega 3 fatty acids group. The effect of supplement use on the percentage of immotile sperm was decremental in all supplement groups but this was not statistically significant. The largest effect was observed in the vitamin E + Omega 3 fatty acids group.

Eslamian et al. (2020) evaluated the effects of simultaneous consumption of DHA and vitamin E on spermogram results among men with asthenospermia (Eslamian et al., 2020). They reported that sperm count and sperm concentration had significantly increased in the vitamin E group compared with others, while other semen parameters showed no significant difference among the groups after intervention. They concluded that, overall, the simultaneous consumption of DHA and vitamin E supplements led to increased sperm motility, with no discernable effect on sperm morphology or sperm vitality in men (Eslamian et al., 2020). Hosseini et al. (2019) looked at the effects of Omega 3 fatty acid consumption on male infertility in a systematic meta-analysis review (Hosseini et al., 2019). They observed significant improvements in sperm motility and semen plasma DHA concentration in infertile men who had used Omega 3 fatty acid supplements (Hosseini et al., 2019). Q. Liu et al. (2015) studied the effects of using Omega 3 and Omega 6 fatty acid supplements along with vitamin E on the quality of sperm and antioxidant response in wild boars (Q. Liu et al., 2015). They reported that a 6/6 ratio of Omega 6 to Omega 3 had a significantly higher effect on sperm motility compared with the other two ratios. They concluded that a vitamin E supplement of 400 mg/kg as opposed to 200 mg/kg had improved sperm motility and had increased superoxide dismutase and total antioxidant capacity (parameters within the semen while also reducing malondialdehyde; (Q. Liu et al., 2015). Gulliver et al. (2012) conducted a review study on the role of unsaturated Omega 3 fatty acids in the reproductive system of sheep and cows (Gulliver et al., 2012). They reported strong evidence that indicates the use of Omega 3 supplements with high dosage is accompanied by lower inflammation indices such as Prostaglandin F<sub>2</sub>alpha (PGF<sub>2</sub>α). Eicosanoid inflammatory mediators such as PGF<sub>2</sub>α can significantly affect reproductive outcomes such as the onset of oestrus, fetus vitality, and pregnancy. They conclude that the effects of dietary supplements containing a high dose of unsaturated Omega 3 or Omega 6 fatty acids on male fertility are mostly unknown (Gulliver et al., 2012).

### *The Effects of Exposure to Electromagnetic Fields on Reproductive Indices*

Results show that exposure to electric and magnetic fields was far lower than the threshold limit values (TLV) of occupational exposure for all three exposure groups and in all three production sections. In studies conducted in similar industries such as that of Don et al. (2020), exposure levels were reported as being lower than TLV as well (Don et al., 2020). It must be remembered that when devising TLVs for occupational exposure, only the main effect of that particular exposure is taken into account.

This means that other potential effects on the reproductive system are neglected (American Conference of Governmental Industrial Hygienists, 2021), and thus, it is not enough to simply compare results with TLVs.

Certain studies have suggested a potential link between exposure to low-frequency electromagnetic fields and detrimental effects on the reproductive system (Johansson, 2009). These studies also suggest that these effects are dependent on frequency, wavelength, polarity, intensity, power density, and exposure duration (Gye & Park, 2012). Univariate analysis of variance revealed that the largest effect observed among the exposure variables was that of electric fields on testosterone levels ( $B = -0.593$ ), this was not statistically significant ( $p = .373$ ). It seems that a single-unit increase in the intensity of the field results in a half-unit drop in free testosterone levels. Exposure to electric fields had a statistically significant effect on sperm count ( $p = .032$ ), suggesting an 11-fold reduction in sperm count for every single unit increase in field intensity. As for morphology and motility (sluggish and full), the largest effect observed was that of electric fields, which was statistically significant.

The largest effect among exposure variables was that of electric fields on the percentage of immotile sperm, but this was not statistically significant. As for magnetic fields, negative effects were observed on sperm count, motility (sluggish and full), and morphology ( $p > .05$ ). The effect on motility (full, sluggish) is near significance levels ( $p = .053$ ), suggesting a one-unit decrease in sperm motility for every unit increase in the flux density of the field. Correlation tests reveal that exposure to electromagnetic fields had no statistically significant correlation with testosterone levels or sperm parameters before supplement intervention. After the intervention, the relationship between exposure to electric fields and the percentage of sperm with normal morphology, as well as with the percentage of sperm with sluggish motility, was statistically significant. Still, a weak reverse relationship was observed between exposure to electromagnetic fields and free testosterone levels or sperm parameters (count and motility).

Gamberale et al. (1989) looked at the effects of exposure to low-frequency electromagnetic fields on reproductive hormones among linesmen. The only significant difference observed was that of testosterone levels between control conditions and exposure conditions. Testosterone levels were actually higher under exposure in their study. This does not agree with our findings, as the lowest observed free testosterone levels belonged to the third exposure group (highest level of exposure; Gamberale et al., 1989). In the present study, blood samples were only taken at their highest levels (between 7:00 am and 8:00 am) to reduce the effect of the daily variance in the levels of reproductive hormones (Gamberale et al., 1989). Gamberale et al. (1989) instead took samples 3

times per shift and the variance in testosterone levels throughout the day are observable in their results. Other reasons for the disparity between the results of Gamberale et al. and the present study include the difference in the study design and other influential factors that may have affected hormone levels.

In the follow-up study conducted by Hjollund et al. (1999) on welders, no significant correlation was reported between exposure to low-frequency electromagnetic fields and human reproductive markers including duration of pregnancy, sperm quality, and reproductive hormones (LH, FSH, estradiol, and testosterone). The results of Hjollund et al. (1999) are in agreement with the present study as neither indicated significant correlations between exposure to low-frequency magnetic fields and reproductive hormone levels.

Wang et al. (2016) conducted a study at the Zhejiang power station with the aim of evaluating the effect of exposure to electromagnetic radiation on plasma hormone biomarkers (testosterone, estradiol, melatonin, and heat shock protein) among high-exposure workers (Wang et al., 2016). The results are in agreement with the present study and suggest lower testosterone levels in those exposed to low-frequency EMFs compared with the control. This also suggests the possibility of reduced plasma testosterone levels and a lower ratio of testosterone to estradiol in men with chronic exposure to EMFs (Wang et al., 2016). Suri et al. (2020) observed that exposure to various levels of ELF had no significant effects on serum levels of certain reproductive hormones among workers of a power plant (Suri et al., 2020). As stated by Suri et al., this may have been due to their small sample size, similar exposure levels, and the variance in the target hormones measured among the participants (Suri et al., 2020).

Numerous studies have been conducted on animals for determining the effects of electromagnetic fields on reproductive indices. One study looked at the cellular effects of extremely low frequency (ELF) magnetic fields on spermatogenesis in mice. They demonstrated the effects of an ELF-EMF at 1 mT and 50 Hz on the division and differentiation of the Spermatogonium and reported increased meiosis, higher overall Germ cell division, and higher serum testosterone concentration (Furuya et al., 1998). These results are not in agreement with the present study.

de Bruyn and de Jager (2010) investigated the effects of prolonged exposure to varying levels of ELF-EMFs (0.5–77  $\mu$ T) on reproduction in male mice (BALB/c). Their results indicated no significant difference between the exposure and the control groups in terms of the number of offspring, duration of pregnancy, sperm count, or testosterone levels. In addition, exposure to magnetic fields caused by electrical currents (simulated as those fields produced by high voltage power lines) lead to no

significant change in plasma testosterone concentration in the exposure group compared with the control. This varying magnetic field did have detrimental effects on sperm motility. de Bruyn and de Jager (2010) report a significant reduction in the quality of sperm motility and the number of live sperm for two generations but with no significant effect on any of the other target parameters.

In the ex vivo study by Gholampour et al. (2012), seminiferous tubules atrophied after 135 days along with hyperplasia of the Leydig cells. Gholampour et al. (2012) state that testosterone is essential to maintaining a high-functioning reproductive system. Atrophy in the seminiferous tubules may have been the cause of the observed reduction in testosterone levels in their study. It is also possible that the hyperplasia of the Leydig cells is a compensatory reaction to low testosterone (Gholampour et al., 2012).

Overall, it seems that some studies report a correlation between exposure to ELF-EMFs and reduced sperm quality and lower reproductive hormones, while others observed no such correlation. The differences in these results may be due to a number of issues including the type of study (human/animal/cell culture), type of EMF emitting device, type of exposure (chronic or acute), and exposure duration and field intensity. It is possible that the contradictory reports regarding the toxicity of these fields are influenced by the type of frequency, field intensity, exposure protocol, type and species of animal, or differences in exposure duration (Saito et al., 2006).

The potential effect mechanism of ELF-EMFs on serum testosterone levels remains unclear. A number of studies conducted on mice exposed to ELF-EMFs report increased serum LH along with reduced testosterone levels (Al-Akhras et al., 2006; Mostafa et al., 2007). This indicates that the effect of ELF-EMFs on testosterone levels is not due to its central limiting of pituitary hormone secretions but rather it is environmental (Taherianfard et al., 2013).

As it was stated earlier, the numerous studies conducted on humans and animals regarding the effects of EMFs on reproductive indices have controversial and contradictory reports (Wang et al., 2016). Certain studies on humans report that exposure to EMFs does not cause any changes to male reproductive indices (Hjollund et al., 1998; Møllerløkken et al., 2012). Many animal studies report that exposure to EMFs can cause reductions in reproductive indices such as testosterone (Kesari & Behari, 2012; Sepehrimanesh et al., 2014; Shahin et al., 2014) but can in some cases increase testosterone (Forgács et al., 2004). It is clear that the male reproductive system is sensitive to electromagnetic radiation (Y. Liu et al., 2015). As was mentioned earlier, exposure to EMFs can affect the membrane polarization of the interior Leydig cells in the testicles which are responsible for the secretion of testosterone. The reduction in the amount of testosterone may be due to damaging of the Leydig cells due to EMFs which results in an



inadequate response to the LH pulse (Sepehrimanesh et al., 2014). Despite the numerous studies conducted on the interaction between EMFs and biological systems, various aspects of it remain unclear and the results of the literature reviews remain controversial (Saito et al., 2006).

The particular challenge in EMF exposure evaluation is that it exists everywhere and finding a group with no prior exposure is difficult. This is why very little contrast exists between comparing low levels of exposure with high levels of exposure. Other challenges in this regard include the lack of understanding regarding the biological and biophysical effect mechanisms of EMFs at the level of environmental exposures (Scientific Committee on Emerging Newly Identified Health Risks, 2015). Many disorders caused by EMFs are chronic in nature and researching them requires retrospective studies that itself complicates things even more. Another limitation is the relatively low number of participants who have experienced high levels of exposure to EMFs making it impossible to draw conclusions about the effects of high exposure (Feychting et al., 2005; Mohammadi et al., 2021; Suri et al., 2020).

It should be noted due to the limited number of words in the article, the discussions regarding the effect of demographic parameters on reproductive indices have been presented in Supporting Information.

## Conclusion

The difference in sperm count, sperm motility, and the number of immotile sperm before and after the intervention was not significant in any of the supplement groups (except for the vitamin E + Omega 3 group). The difference in the level of sex hormones before and after the intervention was not statistically significant for any of the various supplement groups. Univariate analysis of variance showed that the exposure to electric fields had a statistically significant effect on sperm count, morphology, and motility. The simultaneous consumption of vitamin E + Omega 3 had a statistically significant effect on sperm morphology and motility. Considering the findings, it is difficult to draw definite conclusions regarding the effects of exposure to electromagnetic fields on reproductive indices as well as the role of supplementation in their improvement. Although an adequate sample size has been obtained and the entry criteria have been designed to remove influential factors, it is not possible to control all confounding factors. This is a problem in human studies, especially field studies. Nonetheless, theories that are tested in field studies are valuable as they better reflect real conditions (Mazlomi et al., 2017). Some of the limiting factors that make it difficult to draw definitive conclusions include:

- Wide-ranging levels of sex hormones observed among participants, especially testosterone.

- Variation in hormone levels throughout the day.
- Inability to repeat monitoring due to budget limitations, strict role of the industry, and cultural/religious considerations.
- Intervening factors such as hazardous chemical agents (heavy metals and polyaromatic hydrocarbons), physical agents (heat stress, noise, and vibration), psychological stressors, and background disorders.
- Limitations and sample drop caused by the Covid-19 pandemic.
- Lack of data on the participants' reproductive indices from the beginning of employment.
- Inability to conduct pathological monitoring of testicular tissue.
- Budget limitations in determining the amount of DNA fragmentation.

## Recommendation

It is highly suggested that a follow-up study with adequate sample size and proper control group be conducted from the beginning of employment in occupations involving exposure to environmental stressors so that a definitive conclusion can be made regarding the effect mechanism of these hazardous occupational agents. Despite the limitations mentioned earlier, the presents study is still highly valuable. Studies that consider the amount of exposure when looking at the effects of physical stressors, such as electromagnetic fields, on reproductive indices in occupational environments are hard to find.

## Authors' Note

Farideh Golbabaee is now affiliated to Department of Occupational Health, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran.

## Acknowledgments

The authors would like to thank the HSE office of the automobile parts manufacturing industry who helped us conduct the biological and environmental assessment, and the authors gratefully acknowledge the Sarem Hospital for its helpful assistance in the semen sampling and analysis. The authors mention a special thanks to Dr. Nriman Moradi and Dr. Maryam Chamari who conducted hormone and nutritional analysis, respectively.

## Author Contributions

HM carried out experiments and analyzed data. FG supervised the research and provided critical revision of the article. SFD contributed to the study design, managed and planned the project, and drafted and provided critical revision of the article. SK supervised data analysis and interpretation. HI supervised nutrition intervention, and FRT contributed substantially to the conception and design of the study. All authors reviewed and provided final approval of the version to publish.

## Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

## Funding

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This study was part of a research project supported by Shahid Beheshti University of Medical Sciences (Grant no. 25821). This work was not funded by or on behalf of any governmental agency.

## Ethics Approval and Consent to Participate

Ethical approval for this study was obtained School of Public Health & Neuroscience Research Center, Shahid Beheshti University of Medical Sciences (IR.SBMU.PHNS. REC.1399.157). All participants filled out the consent form and were participated voluntarily in the study.

## ORCID iD

Somayeh Farhang Dehghan  <https://orcid.org/0000-0002-6607-6396>

## Supplemental Material

Supplemental material for this article is available online.

## References

- Abdollahi, M.-B., Dehghan, S. F., Balochkhaneh, F. A., Moghadam, M. A., & Mohammadi, H. (2021). Comparison of mice' sperm parameters exposed to some hazardous physical agents. *Environmental Analysis Health and Toxicology*, *36*(3), Article e2021013. <https://doi.org/10.5620/eaht.2021013>
- Al-Akhras, M. A., Darmani, H., & Elbetiha, A. (2006). Influence of 50 Hz magnetic field on sex hormones and other fertility parameters of adult male rats. *Bioelectromagnetics*, *27*(2), 127–131.
- Al-Attar, A. M. (2011). Antioxidant effect of vitamin E treatment on some heavy metals-induced renal and testicular injuries in male mice. *Saudi Journal of Biological Sciences*, *18*(1), 63–72.
- American Conference of Governmental Industrial Hygienists. (2021). *TLVs and BEIs, threshold limit values for chemical substances and physical agents and biological exposure indices*. Signature Publications.
- Arbabian, M., Amirzadegan, M., Tavalaee, M., & Nasr-Esfahani, M. H. (2018). Oxidative stress and its effects on male infertility: A review study. *Journal of Rafsanjan University of Medical Sciences*, *17*(3), 253–274.
- Ball, B., Medina, V., Gravance, C., & Baumber, J. (2001). Effect of antioxidants on preservation of motility, viability and acrosomal integrity of equine spermatozoa during storage at 5 C. *Theriogenology*, *56*(4), 577–589.
- Cao, X. W., Lin, K., Li, C. Y., & Yuan, C. (2011). A review of WHO laboratory manual for the examination and processing of human semen (5th edition). *Zhonghua Nan Ke Xue*, *17*, 1059–1063.
- de Bruyn, L., & de Jager, L. (2010). Effect of long-term exposure to a randomly varied 50 Hz power frequency magnetic field on the fertility of the mouse. *Electromagnetic Biology and Medicine*, *29*(1–12), 52–61.
- Don, R. K. M. A., Jung, J.-S., Lee, Y.-J., & Hong, S.-C. (2020). ELF-MF occupational exposure in die-casting and electroplating workers in Korea. *International Journal of Occupational Safety and Ergonomics*, *26*, 624–631.
- Eslamian, G., Amirjannati, N., Noori, N., Sadeghi, M.-R., & Hekmatdoost, A. (2020). Effects of coadministration of DHA and vitamin E on spermatogram, seminal oxidative stress, and sperm phospholipids in asthenozoospermic men: A randomized controlled trial. *The American Journal of Clinical Nutrition*, *112*(3), 707–719.
- Feychting, M., Ahlbom, A., & Kheifets, L. (2005). EMF and health. *Annual Review of Public Health*, *26*, 165–189.
- Forgács, Z., Somosy, Z., Kubinyi, G., Sinay, H., Bakos, J., Thuróczy, G., . . . Lázár, P. (2004). Effects of whole-body 50-Hz magnetic field exposure on mouse Leydig cells. *The Scientific World Journal*, *4*, 83–90.
- Furuya, H., Aikawa, H., Hagino, T., Yoshida, T., & Sakabe, K. (1998). Flow cytometric analysis of the effects of 50hz magnetic fields on mouse spermatogenesis. *Nippon Eiseigaku Zasshi/Japanese Journal of Hygiene*, *53*(2), 420–425.
- Gamberale, F., Olson, B. A., Eneroth, P., Lindh, T., & Wennberg, A. (1989). Acute effects of ELF electromagnetic fields: A field study of linesmen working with 400 kV power lines. *Occupational and Environmental Medicine*, *46*(10), 729–737.
- Gandini, L., Sgrò, P., Lombardo, F., Paoli, D., Culasso, F., Toselli, L., . . . Lenzi, A. (2006). Effect of chemo-or radiotherapy on sperm parameters of testicular cancer patients. *Human Reproduction*, *21*(11), 2882–2889.
- Gharagozloo, P., Gutiérrez-Adán, A., Champroux, A., Noblanc, A., Kocer, A., Calle, A., . . . Moazamian, A. (2016). A novel antioxidant formulation designed to treat male infertility associated with oxidative stress: Promising preclinical evidence from animal models. *Human Reproduction*, *31*(2), 252–262.
- Gholamhasan, V., Abbas, N., & Soude, N. D. (2018). Effect of vitamin e on male rat fertility in simulated microgravity condition. *Pajouhesh Dar Pezeshki*, *42*(1), 28–33. (In Persian)
- Gholampour, F., Owji, S., Javadifar, T., & Bahaoddini, A. (2012). Long term exposure to extremely low frequency electromagnetic field affects sex hormones level and structure of testis in rats. *International Journal of Zoological Research*, *8*(3), 130–136.
- Gulliver, C., Friend, M., King, B., & Clayton, E. (2012). The role of omega-3 polyunsaturated fatty acids in reproduction of sheep and cattle. *Animal Reproduction Science*, *131*(1–2), 9–22.
- Gurr, M. I., Harwood, J. L., & Frayn, K. N. (2002). *Lipid biochemistry* (Vol. 409). Springer.
- Gye, M. C., & Park, C. J. (2012). Effect of electromagnetic field exposure on the reproductive system. *Clinical and Experimental Reproductive Medicine*, *39*(1), 1–9. <https://doi.org/10.5653/cerm.2012.39.1.1>
- Hjollund, N. H. I., Bonde, J. P. E., Jensen, T. K., Ernst, E., Henriksen, T. B., Kolstad, H. A., . . . Olsen, J. (1998).



- Semen quality and sex hormones with reference to metal welding. *Reproductive Toxicology*, *12*(2), 91–95.
- Hjollund, N. H. I., Skotte, J. H., Kolstad, H. A., & Bonde, J. (1999). Extremely low frequency magnetic fields and fertility: A follow up study of couples planning first pregnancies. The Danish first pregnancy planner study team. *Occupational and Environmental Medicine*, *56*(4), 253–255.
- Hosseini, B., Nourmohamadi, M., Hajipour, S., Taghizadeh, M., Asemi, Z., Keshavarz, S. A., & Jafarnejad, S. (2019). The effect of omega-3 fatty acids, EPA, and/or DHA on male infertility: A systematic review and meta-analysis. *Journal of Dietary Supplements*, *16*(2), 245–256.
- Johansson, O. (2009). Disturbance of the immune system by electromagnetic fields—A potentially underlying cause for cellular damage and tissue repair reduction which could lead to disease and impairment. *Pathophysiology*, *16*(2–3), 157–177.
- Jokar, A., Tvakoli Sarvestani, F., Changizi Ashtiyani, S., Zarei, A., Ramezani, M., & Aghababa, H. (2016). Effect of omega-3 fatty acids on testosterone plasma level in Wistar rat. *Alborz University Medical Journal*, *5*(1), 28–33. <https://doi.org/10.18869/acadpub.aums.5.1.28>
- Kao, S.-H., Chao, H.-T., Chen, H.-W., Hwang, T. I., Liao, T.-L., & Wei, Y.-H. (2008). Increase of oxidative stress in human sperm with lower motility. *Fertility and Sterility*, *89*(5), 1183–1190.
- Kesari, K. K., & Behari, J. (2012). Evidence for mobile phone radiation exposure effects on reproductive pattern of male rats: Role of ROS. *Electromagnetic Biology and Medicine*, *31*(3), 213–222.
- Klonoff-Cohen, H. (2005). Female and male lifestyle habits and IVF: What is known and unknown. *Human Reproduction Update*, *11*(2), 180–204.
- Liu, Q., Zhou, Y., Duan, R., Wei, H., Jiang, S., & Peng, J. (2015). Effects of dietary n-6: N-3 fatty acid ratio and vitamin E on semen quality, fatty acid composition and antioxidant status in boars. *Animal Reproduction Science*, *162*, 11–19.
- Liu, Y., Liu, W.-b., Liu, K.-j., Ao, L., Zhong, J. L., Cao, J., & Liu, J.-y. (2015). Effect of 50 Hz extremely low-frequency electromagnetic fields on the DNA methylation and DNA methyltransferases in mouse spermatocyte-derived cell line GC-2. *BioMed Research International*, *2015*, Article 237183. <https://doi.org/10.1155/2015/237183>
- Mazlomi, A., Golbabaee, F., Farhang Dehghan, S., Abbasinia, M., Mahmoud Khani, S., Ansari, M., & Hosseini, M. (2017). The influence of occupational heat exposure on cognitive performance and blood level of stress hormones: A field study report. *International Journal of Occupational Safety and Ergonomics*, *23*(3), 431–439.
- Moeini, B., Jalilian, F., Jalilian, M., & Barati, M. (2011). Predicting factors associated with regular physical activity among college students applying BASNEF model. *Avicenna Journal of Clinical Medicine*, *18*(3), 70–76.
- Mohammadi, H., Dehghan, S. F., Moradi, N., Suri, S., Pirposhteh, E. A., Ardakani, S. K., & Golbabaee, F. (2021). Assessment of sexual hormones in foundry workers exposed to heat stress and electromagnetic fields. *Reproductive Toxicology*, *101*, 115–123. <https://doi.org/10.1016/j.reprotox.2020.12.015>
- Møllerlækken, O. J., Moen, B. E., Baste, V., Magerøy, N., Oftedal, G., Neto, E., . . . Mild, K. H. (2012). No effects of MRI scan on male reproduction hormones. *Reproductive Toxicology*, *34*(1), 133–139.
- Mostafa, R. M., El Hefnawi, A., Moustafa, K., Ali, F., Moustafa, Y., Kamal, S., & El Hefnawi, M. (2007). Effect of 50 Hz, 10 mTesla magnetic field on sex hormones level in male rats. *Journal of Medical Sciences Research*, *1*, 31–36.
- National Institute for Occupational Safety and Health. (1998). *Manual for measuring occupational electric and magnetic field exposures* (No. 98-154). U.S. Department of Health & Human Services.
- Niki, E. (2014). Role of vitamin E as a lipid-soluble peroxy radical scavenger: In vitro and in vivo evidence. *Free Radical Biology and Medicine*, *66*, 3–12.
- Noble, R. E. (2001). Waist-to-hip ratio versus BMI as predictors of cardiac risk in obese adult women. *Western Journal of Medicine*, *174*(4), 240–241.
- Poprac, P., Jomova, K., Simunkova, M., Kollar, V., Rhodes, C. J., & Valko, M. (2017). Targeting free radicals in oxidative stress-related human diseases. *Trends in Pharmacological Sciences*, *38*(7), 592–607.
- Putri, N. D., Lipoeto, N. I., & Reza, M. (2018). Effects of omega 3 on testosterone hormone levels and quality of spermatozoa in obese Rattus norvegicus Wistar Albino Strain. *International Biological and Biomedical Journal*, *4*(3), 156–163.
- Riediger, N. D., Othman, R. A., Suh, M., & Moghadasian, M. H. (2009). A systemic review of the roles of n-3 fatty acids in health and disease. *Journal of the American Dietetic Association*, *109*(4), 668–679.
- Rooke, J., Shao, C., & Speake, B. (2001). Effects of feeding tuna oil on the lipid composition of pig spermatozoa and in vitro characteristics of semen. *Reproduction*, *121*(2), 315–322.
- Sabeti, P., Pourmasumi, S., Rahiminia, T., Akyash, F., & Talebi, A. R. (2016). Etiologies of sperm oxidative stress. *International Journal of Reproductive Biomedicine*, *14*(4), 231–240.
- Safarinejad, M. R., & Safarinejad, S. (2012). The roles of omega-3 and omega-6 fatty acids in idiopathic male infertility. *Asian Journal of Andrology*, *14*(4), 514–515.
- Saito, K., Suzuki, H., & Suzuki, K. (2006). Teratogenic effects of static magnetic field on mouse fetuses. *Reproductive Toxicology*, *22*(1), 118–124.
- Scientific Committee on Emerging Newly Identified Health Risks. (2015). Opinion on potential health effects of exposure to electromagnetic fields. *Bioelectromagnetics*, *36*(6), 480–484.
- Sepehrmanesh, M., Saeb, M., Nazifi, S., Kazemipour, N., Jelodar, G., & Saeb, S. (2014). Impact of 900 MHz electromagnetic field exposure on main male reproductive hormone levels: A Rattus norvegicus model. *International Journal of Biometeorology*, *58*(7), 1657–1663.
- Shahi, M., Nejati, V., & Najafi, G. (2017). Effect of omega-3 on ovarian tissue and blastocyst formation on diabetic rats

- in vitro. *Journal of Mazandaran University of Medical Sciences*, 26(145), 176–185.
- Shahin, S., Mishra, V., Singh, S., & Chaturvedi, C. (2014). 2.45-GHz microwave irradiation adversely affects reproductive function in male mouse, *Mus musculus* by inducing oxidative and nitrosative stress. *Free Radical Research*, 48(5), 511–525.
- Sinclair, S. (2000). Male infertility: Nutritional and environmental considerations. *Alternative Medicine Review: A Journal of Clinical Therapeutic*, 5(1), 28–38.
- Suri, S., Dehghan, S. F., Sahlabadi, A. S., Ardakani, S. K., Moradi, N., Rahmati, M., & Tehrani, F. R. (2020). Relationship between exposure to Extremely Low Frequency (ELF) magnetic field and the level of some reproductive hormones among power plant workers. *Journal of Occupational Health*, 62(1), 1–10. <https://doi.org/10.1002/1348-9585.12173>
- Taherianfard, M., Bahaddini, A., Keshtkar, S., Fazeli, M., & Shomali, T. (2013). Effect of extremely low frequency electromagnetic field and GABAA receptors on serum testosterone level of male rats. *International Journal of Endocrinology and Metabolism*, 11(4), Article e11029.
- Tran, L., Malla, B., Sharma, A., Kumar, S., Tyagi, N., & Tyagi, A. (2016). Effect of omega-3 and omega-6 polyunsaturated fatty acid enriched diet on plasma IGF-1 and testosterone concentration, puberty and semen quality in male buffalo. *Animal Reproduction Science*, 173, 63–72.
- Von Schacky, C. (2006). A review of omega-3 ethyl esters for cardiovascular prevention and treatment of increased blood triglyceride levels. *Vascular Health and Risk Management*, 2(3), 251–262.
- Wander, R. C., Du, S.-H., Ketchum, S. O., & Rowe, K. E. (1996).  $\alpha$ -Tocopherol influences in vivo indices of lipid peroxidation in postmenopausal women given fish oil. *The Journal of Nutrition*, 126(3), 643–652.
- Wang, Z., Fei, Y., Liu, H., Zheng, S., Ding, Z., Jin, W., . . . Chen, G. (2016). Effects of electromagnetic fields exposure on plasma hormonal and inflammatory pathway biomarkers in male workers of a power plant. *International Archives of Occupational and Environmental Health*, 89(1), 33–42.
- Yousef, M., Abdallah, G., & Kamel, K. (2003). Effect of ascorbic acid and vitamin E supplementation on semen quality and biochemical parameters of male rabbits. *Animal Reproduction Science*, 76(1–2), 99–111.
- Zitzmann, M. (2013). Effects of age on male fertility. *Best Practice & Research Clinical Endocrinology & Metabolism*, 27(4), 617–628.