Original Article

Effect of Carob Supplement on Spermogram Parameters and Sexual Function of Infertile Men Referred to the Infertility Center, Hamadan, Iran, 2019: A Randomized Controlled Trial

Shamim Pilehvari; M.D.[1], Tayebeh Gharakhani Bahar; B.Sc.[2], Seyedeh Zahra Masoumi; Ph.D.[2], Farideh Kazemi; Ph.D.[2], Shirin Moradkhani; Ph.D.[3,4], Alireza Rastgoo Haghi; M.D.[5], Parvin Maleki; B.Sc.[2]

1 Endometrium and Endometriosis research center, Medicine school, Hamadan University of medical sciences, Hamadan, Iran
2 Department of Midwifery, Mother and Child Care Research Center, School of Nursing and Midwifery, Hamadan University of Medical Sciences, Hamadan, Iran
3 Department of Pharmacognosy, School of Pharmacy, Hamadan University of Medical Sciences, Hamadan, Iran
4 Medicinal Plants and Natural Products Research Center, Hamadan University of Medical Sciences, Hamadan, Iran
5 Department of Pathology, Medicine school, Hamadan University of medical sciences, Hamadan, Iran

Received March 2023; Revised and accepted August 2023

Abstract

Objective: The purpose of this study is comparison of carob with placebo in the treatment of male infertility.
Materials and methods: This study was performed as a clinical trial with two-group pretest-posttest design. Each group consisted of 30 members. The first group received 1.5 grams of carob per day, and the second group received placebo treatments. Treatment lasted for 12 weeks. Semen analysis as well as testosterone, prolactin, (LH), (FSH) and (TSH) were performed before and after drug treatment in two groups. Sexual function was assessed in the groups in two stages before and after the intervention using the standard International Index of Erectile Function. P-value less than 0.05 was considered statistically significant. Statistical analysis of data was performed using SPSS 16.
Results: The participants' mean age was 34.83 ±6.22 in the placebo and 33.67 ±5.82 years in the Carob group. The results showed in the carob group compared to the placebo group, the rate of normal sperm counts increased by 17% and also the normal level of testosterone was 40% higher than the abnormal levels of the placebo group and these differences were statistically significant (P <0.05). And in most areas of sexual function, the mean scores after the intervention were higher than before (P> 0.05).
Conclusion: It is recommended to use carob supplements to improve spermogram parameters and male sex hormones.

Keywords: Carob; Clinical Trial; Male Infertility; Sexual Dysfunction

Introduction

As defined by the WHO, infertility is couples' inability to conceive after one year of unprotected intercourse. Infertility is one of the most common problems in the world, experienced by about 15% of

Correspondence:
Dr. Seyedeh Zahra Masoumi
Email: zahramid2001@gmail.com

Copyright © 2023 Tehran University of Medical Sciences. Published by Tehran University of Medical Sciences.
This work is licensed under a Creative Commons Attribution-Noncommercial 4.0 International license (https://creativecommons.org/licenses/by-nc/4.0/). Noncommercial uses of the work are permitted, provided the original work is properly cited.

couples. About 25% of Iranian couple's experience primary infertility in their marital life. It is due to a male factor and sperm problems in 50% of the cases. Infertility and its associated individual and social problems are an important issue for couples, because the cause of male infertility is pathologically detectable only in 40% of cases (1). Therefore, infertility treatment is more difficult in men than in women, especially in developing countries where treatment is associated with high cost. Some studies have indicated the harmful effects of environmental factors such as toxins, pesticides and radiations on male infertility and suggested that toxins and pesticides can reduce sperm concentration (2) .The major causes of male infertility are congenital or acquired anomaly of the genitourinary system, malignancies, urogenital infections, increased scrotal temperature (such as varicocele), endocrine disorders, genetic abnormalities, and immunological problems. However, infertility is idiopathic in 30-40% of infertile men, but it may have a variety of causes, including environmental pollution, Oxygen free radicals, and genetic and epigenetic abnormalities (3). Male infertility may have other factors, such as vas deferens obstruction, sperm problems (low count, low motility, dysmorphology), and sperm allergy. A male factor is involved in about half of all infertility cases. The presence of the male factor is often based on abnormal sperm parameters (azoospermia to oligozoospermia). Impaired sperm production and function and damaged spermatogenesis process are among the most common causes of male infertility. Trauma or anatomical defects in the genital system and the use of certain drugs to treat diseases can lead to impaired sperm production and consequently male infertility (4). The overproduction of reactive oxygen species (ROS) by leukocytes as well as abnormal sperm in semen and consequent oxidative stress have recently been identified to be among the causes of male infertility . Free radicals are made up of different metabolic pathways in each aerobic cell, but their main source in male seminal fluid is white cells and abnormal and immature sperm. Studies have shown that infertile men are more likely to have lower antioxidant capacity than fertile men. Researchers have sought to prevent reduced motility, increased mortality and DNA damage due to oxidative stress in infertile people using antioxidants (5). Carob or Ceratonia siliqua L is a 7- to 12-meter tall tree belonging to the Legominasae family. Its leaves are a combination of yellow, red or purple flowers with no petals. Its fruit is a curved pod light brown in color, 10 to 30 cm long and containing 12 to 16 hard seeds. Carob is native to the Mediterranean region and is found in southern Syria. It grows rapidly in India, many Mediterranean regions, California, and Fars, Iran (6). Studies on the Ceratonia siliqua seed chemicals have shown that it is high in fiber, polyphenol compounds, arachidonic acid, lignin, fats, proteins, carbohydrates, calcium, potassium and phosphorus. They also contain aspartic acid, glutamic acid, linolenic acid, linoleic acid, vitamin E, beta-sitosterol, silica, iron, and magnesium. These compounds can have different effects on the body of animals. Carob seeds are also known as a potential natural source of antioxidants and their antioxidant activity is associated with phenolic compounds and the presence of substances (7). Sexual desire has an undeniable effect on marital life and its stability. Any factor that disrupts a healthy marital relationship is classified as a sexual disorder (8). The WHO considers sexual health to be the integration and consistency among the mind, feeling, and the body, which leads the social and intellectual aspects of human beings towards the development of their personality and leads to the establishment of communication and love. Therefore, any disorder that leads to inconsistency and consequent dissatisfaction with sexual relationship can lead to sexual dysfunction. There has been little research on the effects of carob seed supplementation on male reproduction, so the present study has tried to explore the effect of carob seed supplementation on changes in sperm parameters, sex hormone levels, and sexual function in infertile men, the results of which may prove helpful in fertility treatment and family planning. This study was completed in accordance with SCARE 2020 guidelines (9).

Materials and methods

Study design: This study was designed as a two-group randomized, double-blind, placebo-controlled clinical trial ((IRCT20120215009014N284) performed on idiopathic infertile men. After obtaining permission from the Vice Chancellor for Research of Hamadan University of Medical Sciences and the code of ethics (IR.UMSHA.REC.1398.155), the present study was done in Fatemieh Infertility Center in Hamadan, from May 10 to September 9, 2019. The sample size was calculated in Stata 13 software with Sampsi module. The sample size was determined 30 for each group

based on the data obtained from Mahdiani *et al*.'s study (10), considering the 25% loss, and based on the following information:

$$M1 = 17, M2 = 21, Sd1 = 4, Sd2 = 1.5, \alpha = 0.05, \text{power} = 0.90$$

Participants: The inclusion criteria were being a man under 40 years of age with primary infertility; abnormality of at least one of the semen parameters (volume, concentration, sperm count, motility and morphology of sperm), lack of infertility-related disorders such as chromosomal abnormalities, testicular failure, varicocele, and cryptorchidism, lack of chronic diseases such as diabetes, kidney disease, infectious diseases, genital infections, thyroid, having a BMI less than 30, lack of the use of narcotic substances, alcohol and any drugs that disrupt spermatogenesis (such as methotrexate, nitrofurantoin, colchicine and chemotherapy), pituitary suppressants (such as testosterone injections, GnRh analogues), anti-androgens (cimetidine, spironolactone), use of drugs that cause ejaculation failure (alpha-blockers, antidepressants, phenothiazines), use of drugs that cause dysfunction (Beta-blockers, thiazide diuretics, metoclopramides), long-term use of drugs such as anabolic steroids, cannabis, heroin and cocaine, no history of testicular and vasodilator surgery, no contact with pesticides, heavy metals and solvents, and non-use of antioxidant supplements in the last three months (11). Infertile men with drug and alcohol use, the use of the creatinine more than twice, strenuous physical activity, infertility during the study, diet for weight loss, and change of location were not included in the study.

Intervention: A semen sample was initially taken from men who had referred for infertility treatment. Samples were collected in case of three-day avoidance of sexual intercourse. Incubation was performed for 30 to 60 minutes to convert the samples from bulk to liquid. To evaluate sperm parameters in accordance with the WHO standards, 200 microliters of fluid sample was examined. Computer semen analysis was used to assess sperm motility. Also, microscopic tests were performed to evaluate and determine parameters such as sperm concentration per milliliter of semen, viability and sperm morphology. The research goals and methods were explained to those who had the inclusion criteria and then written consent was obtained from all volunteers for the research. The data collection form about the general characteristics of the patients was completed. Furthermore, 10 cc of blood was taken from patients at the beginning of the study to measure their sex hormones (LH, FSH, prolactin, and testosterone) and TSH. The packages containing the capsules were coded by individuals other than the researchers as A and B, and the volunteers were randomly assigned to intervention and placebo groups. There were mixed cards with labels A and B based on the number of participants (Figure 1).

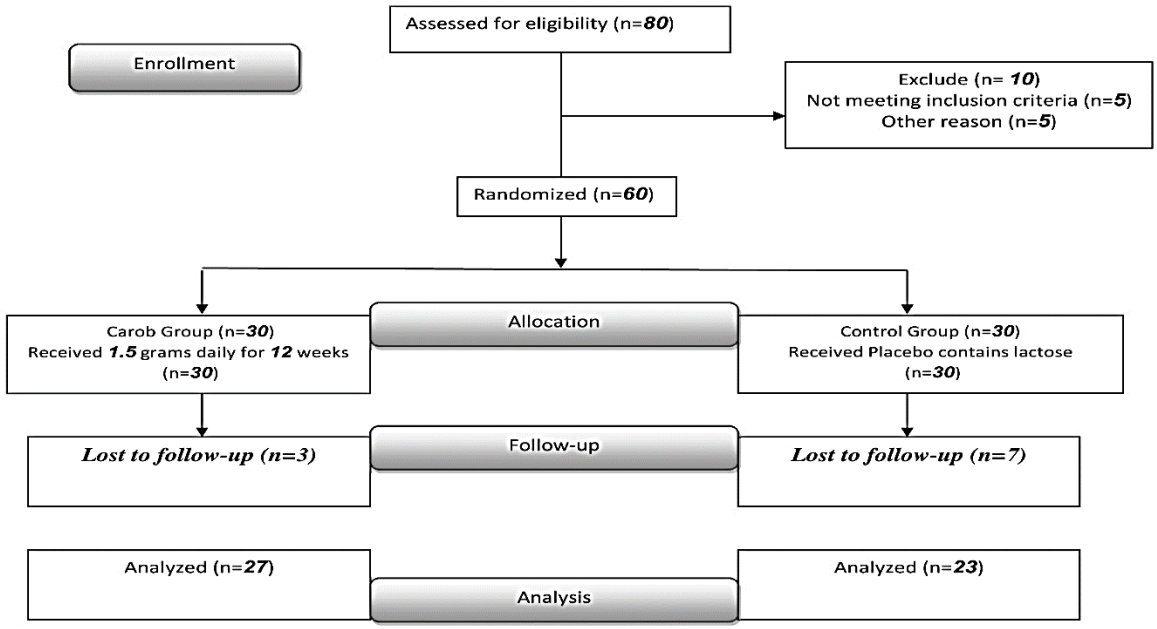

Figure 1: Flowchart of sampling

By taking out the cards, the patients were divided into two groups of carob and placebo. The researcher and the patient were blind to the study groups. The first group received 1.5 grams of carob seed powder per day in three, 500 mg capsules. They received placebo capsules, which were similar in appearance to carob capsules and contained lactose, and both groups received the same routine infertility treatment in 12 weeks. The prescribed dose selection was based on a pilot study which had been conducted by the researchers. This pilot study had been performed on ten idiopathic infertile men as a supplement to carob on sperm parameters. Both carob and placebo drugs are prepared in the form of capsules by the Faculty of Pharmacy of Hamedan University of Medical Sciences. Follow-up of patients was performed by phone once every 15 days in order to control the use of capsules and prevent the loss of samples. Moreover, by counting the remaining capsules, patients who had not used more than 10% of their capsules were excluded at the end of the study. Patients were also advised not to change their diet. Finally, after the intervention, the semen samples were evaluated for spermogram and blood samples were examined for sex hormones. LH and FSH were measured using the ELISA method with the CSB E12654r kit made by the Japanese CUSABIO Company. Serum levels of Testosterone, prolactin and TSH were measured using the ELISA hormone measurement kits made by the German DRG Instruments GmbH Company with hormonal sensitivity of 0.083 ng / ml and the RIA (radioimmunoassay) with hormonal sensitivity of 0.09.

Data measurements: It should be noted that sexual function was examined in the infertile men with the inclusion criteria in groups in two stages before and after the intervention using the IIEF. This index was completed by researchers via interviews. It contains 15 standard questions and is divided into 5 subscales, namely erectile function, orgasmic function, sexual desire, satisfaction with intercourse and overall satisfaction. It is scored from zero to five and the total score is obtained by adding the scores of the questions of each dimension. Higher score indicates the most optimal sexual function. The scores range from 15 to 75, with scores within the 15-25 range indicating low sexual function, scores within the 25-50 range indicating moderate sexual function, and scores higher than 50 indicating high sexual function. In previous studies, the reliability of the questionnaire was confirmed with the Cronbach's alpha of 0.85%.

Statistical analysis: In this study, all the data were shown as mean and frequency for quantitative and qualitative variables, respectively. Initially, the normality of data distribution was evaluated using the Kolmogorov-Smirnov test. The independent sample t-test was used to compare the mean changes of quantitative variables between the two groups. P-value less than 0.05 was considered statistically significant. Statistical analysis of data was performed using SPSS 16.

Results

The participants' mean age was 34.83 ± 6.22 years in the carob group, and 33.67 ± 5.82 years in the placebo group. The majority of participants in all groups had less than a high school diploma and was mostly self-employed. A comparison of the variables presented in Table 1 did not show a statistically significant difference among the groups.

Table 1: Comparison of demographic and contextual characteristics in groups

Variables		Carob group	Placebo group	Statistics	P-value
Age (Year), M (SD)		33.67 (5.82)	34.83 (6.22)	0.97	0.33*
BMI, M(SD)		26.57 (5.01)	27.62 (5.30)	0.94	0.34*
Duration of marriage (Year), M(SD)		8.52 (6.17)	6.37 (4.15)	-0.67	0.50*
Education (N %)	Academic	20 (74.1)	12 (52.2)	2.58	0.10**
	Diploma	7 (25.9)	11 (47.8)		
Occupation (N %)	Employed	3 (11.1)	3 (13)	-	1.0
	Unemployed	24 (88.9)	20 (87)		
Physical activity (N %)	Low	3 (11.1)	2 (8.7)	-	0.42
	Moderate	14 (51.9)	19 (82.6)		
	Excessive	10 (37.0)	2 (8.7)		
Alcohol consumption history (N %)	No	25 (92.6)	21 (95.5)	-	0.67
	Yes	2 (7.4)	1 (4.5)		
History of smoking (N %)	No	21 (77.8)	18 (81.8)	-	0.72
	Yes	6 (22.2)	4 (18.2)		

*Independent t test, **Chi-square test and the rest: Fisher's exact test

Table 2: Comparison of semen analysis results before intervention between groups

Variables		Carob group N (%)	Placebo group N (%)	Statistical test	P-value
Volume	≥1.5	26 (96.3)	18 (78.3)	-	0.08
	<1.5	1 (3.7)	5 (21.7)		
Viscosity	Normal	24 (88.9)	20 (87.0)	-	1.00
	Abnormal	3 (11.1)	3 (13.0)		
Sperm count per cc (million)	≥15	25 (92.6)	16 (69.6)	-	0.06
	<15	2 (7.4)	7 (30.4)		
Progressive motility	≥32%	14 (51.9)	14 (60.9)	0.40	0.52*
	<32%	13 (48.1)	9 (39.1)		
Sperm morphology	≥4%	26 (96.3)	21 (91.3)	-	0.58
	<4%	1 (3.7)	2 (8.7)		
WBC number	Normal	27 (100.0)	22 (95.7)	-	0.46
	Abnormal	0	1 (4.3)		

*Chi-square test and the rest of Fisher's exact test

A comparison of seminal fluid parameters at the pre-intervention stage showed that 96.3% of the carob group members and 78.3% of the placebo group members had normal semen volume. About 93% of the carob group subjects had normal sperm counts, while this rate was 70% in the placebo group. In terms of progressive motility, sperm shape and WBC number, the majority of subjects in the groups were in normal condition. None of these parameters showed a statistically significant difference between the two groups (P>0.05) (Table 2). The results of multivariate analysis showed that by controlling the values of semen at the pre-intervention stage, the normal volume of semen in the carob group was 9% less than that in placebo, and these results were statistically significant (P<0.05). In the carob group, the natural viscosity increased by 15% compared to placebo, which was not statistically significant. In the carob group (P>0.05), the rate of normal sperm counts increased by 17%, which was also statistically significant (P<0.05) (Table 3).

Inter-group and intragroup comparisons of the results revealed that although the mean scores in the carob group after intervention were higher than that in placebo in the subscales of orgasm function, IIEF, satisfaction with sexual intercourse, sexual desire, overall satisfaction, and erectile function, no significant statistical difference was observed in these subscales (P>0.05) (Table 4). A comparison of the level of hormones presented in Table 5 demonstrated that the two groups did not have a statistically significant difference at the pre-intervention stage (P>0.05) (Table 5). The results of multivariate analysis in terms of hormone status showed that by controlling hormone levels at the pre-intervention stage, normal testosterone levels were 40% higher than its abnormal levels in placebo users and this difference was statistically significant (P<0.05).

In the case of prolactin, the normal level of this hormone in the carob group was 37% higher than that in placebo, but this difference was not statistically significant (P>0.05). The changes in the rest of the hormones are negligible, as shown in Table 6.

Table 3: Comparison of semen analysis results after intervention between groups

Variables		Carob group N (%)	Placebo group N (%)	Statistical test	P-value	Risk ratio
Volume	≥1.5	25 (92.6)	21 (91.3)	-	1.00	0.91 (0.86, 0.96)
	<1.5	2 (7.4)	2 (8.7)			
Viscosity	Normal	25 (92.6)	19 (82.6)	-	0.39	1.15 (0.99, 1.33)
	Abnormal	2 (7.4)	4 (17.4)			
Sperm count per cc (million)	≥15	23 (85.2)	18 (78.3)	-	0.71	0.99 (0.85, 1.17)
	<15	4 (14.8)	5 (21.7)			
Progressive motility	≥32%	20 (74.1)	14 (60.9)	0.99	0.31*	1.07 (0.84, 1.37)
	<32%	7 (25.9)	9 (39.1)			
Sperm morphology	≥4%	27 (100.0)	19 (82.6)	-	0.03	1.17 (1.05, 1.30)
	<4%	0 (0)	4 (17.4)			
WBC number	Normal	27 (100.0)	23 (100.0)	-	-	-
	Abnormal	0 (0)	0 (0)			

*Chi-square test and the rest of Fisher's exact test

Table 4: Comparison of hormones levels before the intervention between groups

Hormone		Carob group	Placebo group	Statistical test	P-value
LH	Normal	21 (77.8)	12 (52.2)	3.62	0.06*
	Abnormal	6 (22.2)	11 (47.8)		
FSH	Normal	23 (85.2)	21 (91.3)	-	0.67
	Abnormal	4 (14.8)	2 (8.7)		
Prolactin	Normal	20 (74.1)	12 (52.2)	2.58	0.10*
	Abnormal	7 (25.9)	11 (47.8)		
Testosterone	Normal	23 (85.2)	22 (95.7)	-	0.35
	Abnormal	4 (14.8)	1 (4.3)		
TSH	Normal	22 (81.5)	22 (95.7)	-	0.19
	Abnormal	5 (18.5)	1 (4.3)		

*Chi-square test and the rest of Fisher's exact test

Discussion

Nowadays, herbs are used to treat infertility with male or female factor in many countries of the world, especially in Asian countries. One of the herbal and local medicines widely used in world to treat infertility with male factor and impotence is carob. In our study, the results of comparison between the carob and placebo groups indicated normal viscosity, progressive motility of more than 32%, and normal shape of more than 4% in the carob group after the intervention as compared with the placebo. However, no significant difference was observed only in the progressive motility due to the limited sample size. Since antioxidants play a central role in the defense of cells against free radicals, a decrease in the antioxidant activity of the body's physiological system is likely to be associated with a decrease in the quality of sperm cells (12). Numerous studies have shown that active oxygen species (AOS) can strongly affect various sperm parameters, including sperm morphology and motility. According to studies, sperm quality has decreased over the past decades. Various micronutrients are involved in sperm metabolism. The high content of unsaturated fatty acids in the plasma membrane of sperm is considered to be the reason for the high sensitivity of sperm to ROS (13). Carob has traditionally been used to increase the number of sperm in infertile men. Animal studies have shown carob extract to affect cAMP production and the activity of enzymes involved in steroidogenesis in rat testes. The use of carob has been shown to affect sex hormones, sperm production and spermatogenesis in rats by influencing biochemical factors affecting Leydig cells and testosterone biosynthesis (14). Studies conducted by Alsalman *et al*. in 2013 (15) and Elsheikh MG *et al*. in 2015 (16) have revealed that the use of antioxidants improves sperm quality. A study was conducted by Greco E in 2005, finding that antioxidant administration can be effective in controlling damage to sperm DNA in the short term (17).

Mahdiani *et al*. (2018) found that changes in the mean total antioxidant capacity and plasma malondialdehyde concentration was significant after the intervention in the carob group in comparison with the placebo group.

Table 5: Comparison of hormones levels after the intervention between groups

Hormone		Carob group N (%)	Placebo group N (%)	Statistical test	P-value	Risk Ratio
LH	Normal	25 (92.6)	12 (52.2)	-	0.003	1.18 (0.94, 1.47)
	Abnormal	2 (7.4)	11 (47.8)			
FSH	Normal	21 (77.8)	18 (78.3)	0.001	0.96*	1.04 (0.91, 1.20)
	Abnormal	6 (22.2)	5 (21.7)			
Prolactin	Normal	20 (74.1)	12 (52.2)	2.58	0.10*	1.37 (0.95, 1.99)
	Abnormal	7 (25.9)	11 (47.8)			
Testosterone	Normal	21 (77.8)	14 (60.9)	1.69	0.19*	1.40 (1.10, 1.78)
	Abnormal	6 (22.2)	9 (39.1)			
TSH	Normal	23 (85.2)	21 (91.3)	-	0.67	0.97 (0.86, 1.08)
	Abnormal	4 (14.8)	2 (8.7)			

*Chi-square test and the rest of Fisher's exact test

Table 6: Comparison within and between groups of the total score of sexual function domains before and after the intervention

Domains	Carob group N (%)		Placebo group N (%)		Statistical test*	
	Before intervention	After intervention	Before intervention	After intervention	Before intervention	After intervention
Erectile function	22.81 (6.63)	22.72 (7.89)	22.96 (4.31)	23.80 (3.78)	Z =-0.64 P = 0.52	Z =-0.48 P = 0.62
Statistical test**	Z =-0.74, P = 0.45		Z =-1.25, P = 0.21			
Orgasm function	7.73 (2.11)	7.84 (2.24)	7.85 (1.53)	8.20 (1.29)	Z =-0.28 P = 0.77	Z =-0.15 P = 0.88
Statistical test**	Z =-0.44, P = 0.65		Z =-0.87, P = 0.38			
Sexual desire	7.34 (1.43)	7.61 (1.09)	7.50 (1.47)	7.76 (1.30)	Z =-0.32 P = 0.74	Z =-0.47 P = 0.63
Statistical test**	Z =-0.68, P = 0.49		Z =-1.09, P = 0.27			
Sexual satisfaction	9.45 (3.52)	9.73 (3.84)	10.03 (2.06)	10.12 (1.89)	Z =-0.50 P = 0.61	Z =-0.45 P = 0.64
Statistical test**	Z = -0.28, P =0.77		Z = -0.09, P = 0.92			
Overall satisfaction	8.04 (1.98)	8.57 (1.53)	9.00 (1.26)	8.32 (1.97)	Z =-1.61 P = 0.10	Z =-0.19 P = 0.84
Statistical test**	Z = -0.98, P =0.32		Z = 1.58, P = 0.11			
International Erection Performance Index	54.95 (13.09)	56.52 (14.65)	56.51 (9.56)	58.12 (7.54)	Z =-0.17 P = 0.86	Z =-0.45 P = 0.65
Statistical test**	Z = -0.45, P = 0.64		Z = -1.27, P = 0.20			

*Mann–Whitney U test, **Wilcoxon Test

Researchers have sought to prevent reduced motility, increased mortality and DNA damage due to oxidative stress in infertile people using antioxidants (10). Faramarzi *et al*. (2020) found that an increase in the progressive motility of sperm was observed after the administration of carob in the laboratory environment (18). Sufficient animal and human studies have not been carried out on the effects of the extract of this plant and the mechanisms involved, as well as its effective amounts. The results of the present study revealed that there was no statistically significant increase in many of the sexual function subscales in the carob group after intervention, and the normal levels of prolactin and testosterone were higher in the carob group than in the placebo. Sanagoo *et al*. (2021) found that the use of carob supplement did not affect the sexual function of infertile men. Carob is a rich source of iron, calcium, sodium, potassium, phosphorus and sulfur, as well as vitamins E, C, D, niacin, folic acid and pyridoxine. Its powder contains 11 phenolic compounds with large contents of pyrogallol, catechol and chlorogenic acid and small amounts of other phenolic compounds such as coumarin, cinnamic, ferulic and gallic acid. It also contains 17 fatty acids, mainly the four fatty acids of oleic, linoleic, palmitic and stearic acid (19).

Mokhtari *et al*. (2012) studied the possible effects of carob seeds on levels of FSH, LH, testosterone and dihydrotestosterone, testicular tissue, and fertility improvement in male rats. They found that the use of carob seed extract caused a significant increase in testosterone and dihydrotestosterone concentrations and a decrease in the LH levels in experimental groups, but the FSH concentration did not show significant changes (20). Mahdiani found that changes in testosterone were positive after taking carob supplements, although the changes were not significant. Also, the change in other sex hormones was not significant in that study. The only study conducted on the effect of carob use on the sex hormones of rats revealed that the use of carob seed extract in male mice significantly increased the concentration of testosterone and dihydrotestosterone, but it decreased the LH levels (10) It seems that the increase in testosterone levels by carob is due to its direct effect on Leydig cells and in testosterone biosynthesis. These effects are probably due to the stimulation of PGE2 synthesis. In addition, carob seeds contain gamma linolenic acids and alpha linolenic acid, which can be converted to dihomo-gamma-linolenic acid and PGE2, which ultimately increases the production of cyclic adenosine

monophosphate and stimulates testosterone production (16).

Limitation: One of the limitations of this study is the small number of samples that reduce the generalizability of the results to the whole community.

Conclusion

According to the results of this study, the use of carob supplement is recommended to improve some spermogram parameters such as viscosity and increase the normal levels of testosterone and prolactin hormones.

Conflict of Interests

Authors declare no conflict of interests.

Acknowledgments

The authors wish to thank the Deputy of Research of the Hamadan University of Medical Sciences for supporting the project financially (NO, 9805013364). We would like to express our gratitude to the staff of infertility center at Fatemieh Hospital for helping us with this project.

References

1. Direkvand Moghadam A, Delpisheh A, Sayehmiri K. The trend of infertility in Iran, an original review and meta-analysis. Nursing Practice Today. 2015;1(1):46-52.
2. Okonofua FE, Ntoimo LFC, Omonkhua A, Ayodeji O, Olafusi C, Unuabonah E, Ohenhen V. Causes and Risk Factors for Male Infertility: A Scoping Review of Published Studies. Int J Gen Med. 2022;15:5985-5997
3. Babakhanzadeh E, Nazari M, Ghasemifar S, Khodadadian A. Some of the Factors Involved in Male Infertility: A Prospective Review. Int J Gen Med. 2020;13:29-41.
4. Durairajanayagam D. Lifestyle causes of male infertility. Arab J Urol. 2018;16(1):10-20.
5. Alsalman ARS, Almashhedy LA, Alta'ee AH, Hadwan MH. Effect of Zinc Supplementation on Urate Pathway Enzymes in Spermatozoa and Seminal Plasma of Iraqi Asthenozoospermic Patients: A Randomized Controlled Trial. Int J Fertil Steril. 2020;13(4):315-323.
6. Calislar S, Kaplan Y. Effects of carob (Ceratonia siliqua) pod byproduct on quail performance, egg characteristics, fatty acids, and cholesterol levels. Revista Brasileira de Zootecnia. 2017;46(2):113-7.
7. Mahmoudi S, Mahmoudi N, Benamirouche K, Estévez M, Mustapha MA, Bougoutaia K, Djoudi NEHB. Effect of feeding carob (Ceratonia siliqua L.) pulp powder to broiler chicken on growth performance, intestinal microbiota, carcass traits, and meat quality. Poult Sci. 2022;101(12):102186.
8. u X, Zhang S, Chen L, Zhang XY, Wang Q. High incidence of sexual dysfunction and timed intercourse was found only in infertile males who with known impairment of sperm quality for a long period: evidence from a hospital-based cross-sectional study. Reprod Biol Endocrinol. 2022;20(1):139.
9. Agha RA, Franchi T, Sohrabi C, Mathew G, Kerwan A; SCARE Group. The SCARE 2020 Guideline: Updating Consensus Surgical CAse REport (SCARE) Guidelines. Int J Surg. 2020;84:226-230.
10. Mahdiani E, Khadem Haghighian H, Javadi M, Karami A A, Kavianpour M. Effect of Carob (Ceratonia siliqua L.) oral supplementation on changes of semen parameters, oxidative stress, inflammatory biomarkers and reproductive hormones in infertile men. Scientific Journal of Kurdistan University of Medical Sciences. 2018; 23 (3) :56-66.
11. Ahmadi S, Bashiri R, Ghadiri-Anari A, Nadjarzadeh A. Antioxidant supplements and semen parameters: An evidence based review. Int J Reprod Biomed. 2016;14(12):729-736.
12. Soleimanzadeh A, Malekifard F, Kabirian A. Protective effects of hydro-alcoholic garlic extract on spermatogenic disorders in streptozotocin-induced diabetic C57BL/6 mice. Scientific Journal of Kurdistan University of Medical Sciences. 2017;22(4):8-17.
13. O'Flaherty C. Reactive Oxygen Species and Male Fertility. Antioxidants (Basel). 2020;9(4):287.
14. Vafaei A, Mohammadi S, Fazel A, Soukhtanloo M, Mohammadipour A, Beheshti F. Effects of carob (Ceratonia siliqua) on sperm quality, testicular structure, testosterone level and oxidative stress in busulfan-induced infertile mice. Pharmaceutical Sciences. 2018;24(2):104-11.
15. Alsalman A, Almashhedy LA, Hadwan MH. Zinc supplementation attenuates lipid peroxidation and increases antiperoxidant activity in seminal plasma of Iraqi asthenospermic men. Life Science Journal. 2013;10(4):989-97.
16. ElSheikh MG, Hosny MB, Elshenoufy A, Elghamrawi H, Fayad A, Abdelrahman S. Combination of vitamin E and clomiphene citrate in treating patients with idiopathic oligoasthenozoospermia: A prospective, randomized trial. Andrology. 2015;3(5):864-7.
17. Greco E, Iacobelli M, Rienzi L, Ubaldi F, Ferrero S, Tesarik J. Reduction of the incidence of sperm DNA

fragmentation by oral antioxidant treatment. J Androl. 2005;26(3):349-53.
18. Faramarzi A, Aghaz F, Bakhtiari M, Khazaei M. In vitro application of Ceratonia siliqua improved sperm parameters and chromatin quality after vitrifacation in normozoospermic aged men. Middle East Fertility Society Journal. 2020;24(1):1-5.
19. Sanagoo S, Farshbaf-Khalili A, Asgharian P, Hazhir S, Oskouei BS. Comparison of the effect of *Ceratonia siliqua L*. fruit oral capsule and vitamin E on semen parameters in men with idiopathic infertility: a triple-blind randomized controlled clinical trial. J Complement Integr Med. 2021;18(4):791-796.
20. Sadeghzadeh F, Sadeghzadeh A, Changizi-Ashtiyani S, Bakhshi S, Mashayekhi FJ, Mashayekhi M, Poorcheraghi H, Zarei A, Jafari M. The effect of hydro-alcoholic extract of *Ceratonia Silique L.* on spermatogenesis index in rats treated with cyclophosphamide: An experimental study. Int J Reprod Biomed. 2020;18(4):295-306.

Citation: Pilehvari S, Gharakhani Bahar T, Masoumi SZ, Kazemi F, Moradkhani S, Rastgoo Haghi A, et al. **Effect of Carob Supplement on Spermogram Parameters and Sexual Function of Infertile Men Referred to the Infertility Center, Hamadan, Iran, 2019: A Randomized Controlled Trial.** J Family Reprod Health 2023; 17(3): 142-50.