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Occupational Determinants of Azoospermia among Patients Attending Ebenezer Clinical Laboratory - Kampala Capital City, Uganda

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Abstract

Background: Azoospermia is one of the social problems affecting families/countries today in the whole world, which has resulted in an involuntary declining birth rate (Sengoku, 2012) [34]. Worldwide, more than 70 million couples suffer from infertility, and it is estimated that azoospermia is found in up to 10 to 20 per cent of the men who present to an infertility clinic (Kumar, 2015) [30]. Uganda is among the countries where male infertility is assumed to be a big challenge, with an estimated 5,000,000 people facing infertility, where 10 to 15% of the couples are unable to have children. Hence the current study aimed at establishing the occupational-related factors associated with azoospermia among patients attending Ebenezer Clinical Laboratory, Kampala, Uganda.

Methods: A retrospective case-control study design was conducted on men who had visited the Microbiology Department for semen analysis from 1st January to 31st December 2015. Cases were azoospermic participants in the ECL database with no sperm cells in the ejaculate whereas controls were normozoospermic participants in the ECL database with normal sperm cells in the ejaculate. Systematic sampling was employed in the selection of respondents using their clinical records. The sample size was 204 (102 cases and 102 controls) clients, determined using a formula from the OpenEpi software package for Kelsey. The sample involved 102 cases and 102 with a ratio of cases to controls being 1:1. The study used a data abstraction semi-structured questionnaire for data collection. Data was analyzed using descriptive statistics to generate frequencies, percentages, means, standard deviations, and ranges. The chi-square test and binary logistic regression analyses were used to determine whether there was a significant association between risk factors and azoospermia at 0.05 statistical significance.

Results: The study found that exposure to phones [AOR = 0.110, (CI 95% = 0.001- 0.171), p=0.001], noise exposure [AOR = 0.041, (CI 95% = 0.009 - 0.189), p=0.000], and having ever had an accident [AOR = 0.041, (CI 95% = 0.002 - 0.923), p=0.000] were statistically significant factors associated with azoospermia among patients ($p < 0.05$) ($p < 0.05$).

Conclusions: Interventions should be focused on improving these factors, such as sensitizing the males on dangers of getting exposed to mobile phones, as well as strengthening policy guidelines on noise pollution and road safety measures.

Keywords: Azoospermia, Normozoospermia, Sperm Count, Occupation Related Factors, Ebenezer Clinical Laboratory, Kampala Capital City Authority

1. Background

Globally one of the social problems affecting families/countries today is the involuntary declining birth rate, although the number of infertile couples is not well documented (Sengoku, 2012) [34]. Worldwide, it is believed that more than 70 million couples suffer from infertility (Agarwal *et al*., 2022) [5]. Approximately 1% of all men in the general population suffer from azoospermia, and azoospermic men constitute approximately 10 to 15% of all infertile men. Male infertility accounts for 40-50% of infertility, affecting approximately 7% of all men. Yet, approximately 10% of infertile men are azoospermic (Drugkar *et al*., 2013). Also, azoospermia is found in up to 10 to 20 percent of the men who present to an infertility clinic (Kumar, 2015) [30]. Thus, this group of patients represents a significant population in the field of male infertility (Ahmetand, 2013).

The highest numbers of male infertility have been recorded in the "African Infertility Belt" of which 43% of the infertility cases are due to men. Among the developing countries, the exact number of male infertility is unknown due to a lack of proper registration and well-performed studies (Alam *et al*., 2018) [7]. According to WHO, one in every four couples in developing countries is affected by infertility. There has been a notable difference in the prevalence according to geographical locations, and environmental, cultural, and socioeconomic influences. E.g., in Nigeria, male infertility is at 11% (Glazer *et al*., 2019) [19]. In some parts of sub-Saharan Africa including the Republic of Sudan and Cameroon, infertility rates could exceed 30% (Sharma, 2017) [44]. In Kenya, Gatonye Gathura (2017) in a national study concluded that azoospermia (no sperm) and Oligospermia (low sperm) were the major causes of infertility among men; responsible for 41 percent of all male infertility.

In Uganda, infertility is a major challenge, with an estimated 5,000,000 people facing infertility, mainly handled by the private centers in urban areas; and about 10-15% of the couples cannot have children due to infertility. According to the MOH-Uganda, 75% of people are affected by infertility, the problem is due to Sexually Transmitted Infections (STIs) which often lead to blockage of sperm ducts among men (Ampurire, 2018). However, the actual percentages showing infertility rates in Uganda are hard to come because people are not open about it and we do not have data, but medical consultants noted there are more Ugandans seeking help for fertility problems (Wanjala, 2013). Social and environmental factors have been cited to be behind the increase in the number of patients with infertility, among men contributing almost half of all cases in advanced nations (Miyamoto *et al*, 2012) [34].

2. Methods

Study design

Th study adopted a retrospective case-control study design descriptive survey design with a cross-sectional design to establish the risk factors associated with azoospermia among men who had visited the Microbiology Department for semen analysis from 1st January to 31st December 2015 (Rothman, 2017). Cases were azoospermic participants in the ECL data base with no sperm cells in the ejaculate whereas controls were normozoospermic participants in the ECL data base with normal sperm cells in the ejaculate. Quantitative research approaches were used, where data from the patient files was analyzed using descriptive and inferential statistics.

Setting

The study was carried out at Ebenezer Limited Clinical Laboratory, Kampala, Uganda. It's found on the First Floor of Sure House Building Plot 1, Bombo Road within the central area of Kampala Capital City Authority (KCCA), Uganda. This laboratory was chosen because it's accredited to ISO 15189 Testing Laboratory No. M0221, by SANAS from South Africa (Ebenezer Ltd Clinical Laboratory, 2015) [16] serves a larger number of people for infertile investigations than any other Laboratory in Kampala and Uganda in general. The laboratory participates in external and internal quality assurance schemes and runs control daily. Over the past two years since 2013, there has been a rapidly increasing number of clients visiting the Ebenezer laboratory, testing positive for azoospermia among those seeking semen analysis tests, whereby, an average of five azoospermic patients were reported per day since 2013 to date. This raised a concern to conduct the current study at Ebenezer laboratory.

Sample

The sample size of was 204 (102 cases and 102 controls) clients, determined using a formula from the OpenEpi software package for Kelsey (Dean *et al*. 2009) which is suitable for determining sample size in an unmatched case-control study design. This sample size was determined with an assumption of a proportion of controls with exposure being 10 % at a 95% confidence interval with 5% marginal error and power of 80% as well as an odds ratio of three detected. The sample involved 102 cases and 102 with a ratio of cases to controls being 1:1.

Sampling

A systematic sampling technique was employed to select 102 cases and 102 controls. According to the records at ECL, a total of 2,880 clients visited the Microbiology Department for semen analysis tests in the period of 1st January to 31st December 2015 (ECL-2015 Annual Report, 2015). Since daily half of the clients were turning out to have azoospermia; this implies that out of the total population of clients, half of them (1,440 clients) were the estimated population of azoospermia clients (cases). Hence from this population, 102 azoospermia clients were picked using a systematic sampling technique, and these were the cases. Whereas, according to the daily trend of normozoospermia (controls), 2 out of 10, were normozoospermia and hence the study population for controls was 576 clients, and the sample size for controls was 102.

Data was captured from a pre-arranged laboratory register book in the microbiology department which is usually filled whenever one comes for semen examination. Systematic sampling was employed to select the study participants. Since the sample size for cases was 102, the sampling interval was 1,440/102 = 14. Therefore, every 14th element in succession was chosen from the sampling frame to be a part of the case group. Using the rotary method, the first case participant between 1 and 14 was selected randomly in the register which was 1. Then, 1, and every 14th element to follow was picked from the target population of 1,440.

The sample size for controls was 102, therefore the sampling interval was 576/102 = 6. Therefore every 6th element in succession was chosen from the sampling frame to be a part of the control group. Using the rotary method, a first-case participant between 1 and 6 was selected randomly in the register which was 4. Then, 4, and every 6th element to follow was picked from the target population of 576. The collection of the data took 2 months to be completed.

For inclusion criteria, all the azoospermic and normozoospermic patients in the data register aged 18 years and above, who visited ECL Microbiology Department for semen analysis from 1st January to 31st December 2015 were considered in the study. Whereas, for exclusion criteria, the following categories of patients were not included in the study; Oligozoospermic (Sperm concentration less than the reference values); Asthenozoospermic (Less than the reference value for motility); Teratozoospermic (Less than the reference value

for morphology); Oligoasthenoteratozoospermic (Signifies disturbance of all three variables); Aspermic (No ejaculate); Incomplete responses in the data register; Men that were below 18 years of age; and non-Ugandans.

3. Data collection

Methods

The study used a questionnaire survey method for data collection. This is because the data to be gathered is quantitative. The questionnaire survey method was used to collect data from the laboratory records.

Instruments

A data abstraction questionnaire captured information from cases and controls on clients who had visited the facility in 2015. The data abstraction semi-structured questionnaire had closed-ended questions developed to address the objectives of the study. The questionnaire was used because it is the most appropriate instrument given the nature of the topic of the study.

4. Data analysis

Data from the respondents was edited to detect errors, cleaned daily, and sorted and questionnaires were given numbers for identification. Filled questionnaires were reviewed at the end of the day for completeness and accuracy. Data was entered into a datasheet and analyzed in the computer using the Statistical Package for Social Scientists (SPSS) version 20.0 while ensuring the accuracy and consistency of data. Different statistical methods were used for instance descriptive statistics and inferential statistics.

Data was analyzed using descriptive statistics to generate frequencies, percentages, means, standard deviations, and ranges. To establish the significant associations between the predictor variables and azoospermia, a chi-square test was used for bivariate analysis. The x^2 test was used to determine whether there was a significant association between risk factors and azoospermia about 0.05 statistical significance. Variables that were significant under chi-square analysis were further subjected to binary logistic regression to obtain crude odds ratios (COR) and adjusted odds ratios (AOR) and their corresponding 95% confidence intervals. To test the hypotheses the researcher used the P-value and a significance level of (α=0.05) and the researcher rejected the null hypothesis (Ho) when the p-value was less than ($\leq$ 0.05) when the findings were statistically significant and then accepted the null hypotheses (Ho) when the p-value is greater than ($\geq$ 0.05).

Ethical considerations

During the retrieval of respondents'' information from their medical documents, the anonymity and privacy of the participants was observed. The participants remained anonymous during the whole process of the study. The participants' information was kept confidential and only used for the research, the local Research Ethics Committee (REC) and Uganda National Council for Science and Technology (UNCST) as entities that may have access to private information that identifies the research participants by name.

5. Results

In reference to the results in Table 1 below, the majority 75 (73.5%) of the cases (azoospermia) were aged 50 and above years, majority 72 (70.6%) of the controls (normozoospermia) were aged below 50 years. The majority 63 (61.8%) of the clients who had azoospermia were primary school level graduates. While the majority 69 (67.6%) of the clients with normozoospermia were primary school graduates. In the same regard, the majority 66 (64.7%) of the men who were found to be having azoospermia were alcoholic religious affiliated; while among the controls, more than half 54 (52.9%) of the respondents were non-alcoholic religious affiliated. Regarding marital status, half 51 (50%) of the cases and more than half 60 (58.8%) of the controls were married.

Table 1: Socio-demographic characteristics of study participants

Variable & Categories	Cases (azoospermia)		Controls (normozoospermia)	
	Frequency (n)	Percent (%)	Frequency (n)	Percent (%)
Age category				
18 - 49	27	26.5	72	70.6
50>	75	73.5	30	29.4
Education level				
Primary and below	63	61.8	69	67.6
Post-primary	39	38.2	33	32.4
Religious Affiliation				
Religious Affiliation _Alcoholic	66	64.7	54	52.9
Religious Affiliation __Non alcoholic	36	35.3	48	47.1
Marital status				
Unmarried	51	50.0	42	41.2
Married	51	50.0	60	58.8

Occupation-Related Factors Associated with Azoospermia among Patients Attending Ebenezer Clinical Laboratory

Chi-square analysis and bivariate logistic regression analyses were conducted to determine the association between the occupation-related factors with azoospermia among patients at Ebenezer clinical laboratory, at a 95% level of significance.

Table 2: Chi-Square Analysis of Occupation-Related Factors Associated with Azoospermia among patients attending Ebenezer Clinical Laboratory

Factors & Categories		Sperm Count		χ^2	df	p-value
		Cases (Azoospermia) N (%)	Controls (Normozoospermia) N (%)			
Pesticides' exposure						
No		21 (20.6)	87 (85.3)	1.214	1	0.270
Yes		81 (79.4)	15 (14.7)			
Heavy metals exposure						
Yes		75 (73.5)	3 (2.9)	**22.510**	**1**	**0.000****
No		27 (26.5)	99 (97.1)			
Radiation exposure						
Yes		78 (76.5)	9 (8.8)	**8.134**	**1**	**0.004****
No		24 (23.5)	93 (91.2)			
Phones exposure						
	Yes	93 (91.2)	21 (20.6)	**5.628**	**1**	**0.018***
	No	9 (8.8)	81 (79.4)			
Laptops' exposure						
	Yes	69 (67.6)	33 (32.4)	0.000	1	1.000
	No	33 (32.4)	69 (67.6			
Stress exposure						
	Yes	57 (55.9)	36 (35.3)	1.659	1	0.198
	No	45 (44.1)	66 (64.7)			
Noise exposure						
	Yes	63 (61.8)	21 (20.6)	**35.700**	**1**	**0.000*****
	No	39 (38.2)	81 (79.4)			
Ever had accident						
	Yes	84 (82.4)	3 (2.9)	**11.944**	**1**	**0.001****
	No	18 (17.6)	99 (97.1)			

From Table 2, findings show that among the males who had azoospermia, the majority 81 (79.4%) of them had ever had exposure to pesticides. Whereas, among the clients who had normozoospermia, the majority 87 (95.3%) were not exposed to pesticides. Among the clients who had azoospermia, the majority 75 (73.5%) of them had exposure to heavy metals. While among the normozoospermia group, the majority 99 (97.1%) were not exposed to heavy metals. Concerning radiation exposure, the majority 78 (76.5%) of the clients who had azoospermia had radiation exposure. Whereas, among the normozoospermia group, the majority 93 (91.2%) of them did not have radiation exposure. Regarding phones exposure, the majority 93 (91.2%) of the azoospermia clients reported having exposure to phones. While, among the normozoospermia clients, the majority 81 (79.4%) were not exposed to phones. Also, it was highlighted that the majority 69 (67.6%) of the clients with azoospermia were exposed to laptops, and the majority 69 (67.6%) of clients who had normozoospermia were not exposed to laptops. Regarding stress exposure, the majority 57 (55.9%) of the clients with azoospermia were exposed to stress. While, among the clients who had normozoospermia, majority 66 (64.7%) of them were not exposed to stress. About noise exposure, majority 63 (61.8%) of men who had azoospermia had noise exposure; whereas, among those who had normozoospermia, majority 81 (79.4%) of them had no exposure to noise. Results also indicated that majority 84 (82.4%) of men found to be having azoospermia reported having ever had an accident; whereas, among those clients with normozoospermia, almost all 99 (97.1%) of clients who had normozoospermia had never had an accident.

Table 3: Multivariate Logistic Regression Analysis of Occupational Factors Associated with Azoospermia among Patients Attending ECL

Factors & Categories	Sperm Count		COR (95%CI)	p	AOR (95%CI)	p
	Cases (Azoospermia) N (%)	Controls (Normozoospermia) N (%)				
Exposure to Radiation						
Yes	78 (76.5)	9 (8.8)	3.179 [1.396 – 7.241]	0.006	1.259 [0.200 - 7.932]	0.806
No	24 (23.5)	93 (91.2)	1		1	
Exposure to Phones						
Yes	93 (91.2)	21 (20.6)	**1**		**1**	
No	9 (8.8)	81 (79.4)	**0.373 [0.162 - 0.0861]**	**0.021**	**0.110 [0.001- 0.171]**	**0.001****
Exposure to Noise						
Yes	63 (61.8)	21 (20.6)	**1**		**1**	
No	39 (38.2)	81 (79.4)	**0.160[0.086 - 0.300]**	**0.000**	**0.041 [0.009 - 0.189]**	**0.000****
Ever had accident						
Yes	84 (82.4)	3 (2.9)	**1**		**1**	
No	18 (17.6)	99 (97.1)	**0.141[0.040 - 0.497]**	**0.002**	**0.041[0.002-0.923]**	**0.000****

Exposure to phones was a statistically significant factor associated with azoospermia among patients attending Ebenezer clinical laboratory. The crude odds ratio [COR = 0.373, (CI 95% = 0.162 - 0.0861), p = 0.021], implied that the odds of developing azoospermia among patients who were not exposed to phones were about 3 times lower compared to those who were exposed to phones. Whereas, on conducting multivariate analysis, the adjusted odds ratio [AOR = 0.110, (CI 95% = 0.001- 0.171), p = 0.001] implied that the odds of developing azoospermia among patients who were not exposed to phones were about 9 times lower compared to those who were exposed to phones.

Results also show that noise exposure was a strong statistically significant factor associated with azoospermia among patients attending Ebenezer clinical laboratory. The crude odds ratio [COR = 0.160, (CI 95% = 0.086 - 0.300), p = 0.000], implied that the odds of developing azoospermia among patients who were not exposed to noise were about 6 times lower compared to those who were exposed to noise. Whereas, on conducting multivariate analysis, the adjusted odds ratio [AOR = 0.041, (CI 95% = 0.009 - 0.189), p = 0.000] implied that the odds of developing azoospermia among patients who were not exposed to noise were about 24 times lower compared to those who were exposed to noise. While, in relation to exposure to accidents, the crude odds ratio [COR = 0.141, (CI 95% = 0.040 - 0.497), p = 0.002], implied that the odds of developing azoospermia among patients who had never had an accident were about 7 times lower compared to those who had ever had an accident. Whereas multivariate analysis results showed that the adjusted odds ratio [AOR = 0.041, (CI 95% = 0.002 - 0.923), p = 0.000] implied that the odds of developing azoospermia among patients who had never had an accident were about 24 times lower compared to those who had ever had an accident.

6. Discussion

Exposure to phones was also a statistically significant factor associated with azoospermia among patients attending Ebenezer clinical laboratory, where exposure to phones increases the chances of one to get azoospermia. This is because phones produce radiation, which affects spermatogenesis, and hence exposure to phones would cause azoospermia. Therefore, sensitization messages need to be dispersed to the population guiding people on how best to use their mobile phones and highlighting the dangers of poor use of mobile phones. The findings are in agreement with studies done by Daniel, 2013, Ariel *et al*. (2015), and Daniel (2013) who proved that human spermatozoa exposed to radiation/electromagnetic force have decreased motility, morphometric abnormalities, and increased oxidative stress, whereas the use of mobile phones may decrease sperm concentration, motility, morphology, and viability.

The abnormalities seemed to be directly related to the duration of mobile phone use. Whereas Tang *et al*. (2022) showed that talking on your cell phone while it is connected to a charging point doubles the chances of a decrease in sperm concentration from 33.3% to 66.7%. in the same lane, findings are consistent with findings of another study done by Meistrich (2013), who statistically proved that prolonged daily use of cell phones has negative effects on semen quality, especially sperm motility, progressive motility, and morphology. However, the findings disagree with Mehmet *et al*. (2015) who proved that there was no significant difference between sperm counts and sperm morphology due to the mobile phone usage period.

Noise exposure also had a statistically significant association with azoospermia, where it was suggested that exposure to noise increases the likelihood of one acquiring azoospermia. This is so because noise exposure results in stress, which affects the mental status of an individual, leading to depression, which eventually affects the production of male hormones that are necessary for spermatogenesis. Hence there is a need to strengthen policy guidelines on noise pollution in society. This is in agreement with a study done by Ali *et al*. (2013) who confirmed that noise and welding in Industry and construction companies is a contributor to male infertility; and also proved that noise stress has a significant effect on the fertility of male rats. Krausz *et al*. (2015) [24] in a study dubbed 'environmental and lifestyle factors associated with sperm DNA damage' confirmed that semen quality in the adult male can be affected by several environmental and lifestyle factors.

Exposure to accidents was a strong statistically significant factor associated with azoospermia in the study, where having accidents was found to increase the likelihood of the individual getting azoospermia. This was attributed to the fact that some accidents like groin or scrotal accidents result in damaging the sperm ducts or even the epididymis that is necessary to produce sperm, and this ends up causing azoospermia. Therefore, road safety guidelines should be strengthened to minimize road accidents in the country. This was consistent with Atia *et al*. (2015) who proved that there was definite evidence of sub-fertility as assessed by abnormal semen analyses and atrophic testes following testicular trauma. The findings are also consistent with Hamada *et al*. (2013), who confirmed that physical genital examination should include an inspection of the external genitalia with emphasis on surgical scars which may indicate possible injuries to the testicular blood supply and/or vas deferens.

7. Limitations

The study faced a recall bias as it was done among patients who visited the facility a year ago. This was overcome by a proper explanation of the purpose of the study to the respondents and the importance of the study which could help formulate policies that would help the communities. Semen analysis studies and responses related to sexual activity resulted in the quality of data being very poor to very good depending on the individual being interviewed since reproductive information was private and couples may not be inclined to be truthful in surveys. Many men were not willing to participate in semen studies.

8. Conclusions

Identifying the occupational-related factors influencing azoospermia helps to appropriately address the burden and consequences of azoospermia, and this would reduce such consequences like stigmatization, isolation, neglect, domestic violence, loss of social status and failure to have children among the affected individuals, as well as improving the mental and social well-being of the affected.

Table 4: Abbreviations

AIDS	Acquired Immune Deficiency Syndrome
AOR	Adjusted Odds Ratio
CI	Confidence Interval
COR	Crude Odds Ratio
CVI	Content Validity Index
DNA	DeoxyRibo Nucleic Acid
ECL	Ebenezer Clinical Laboratory
FSH	Follicle Stimulating Homone
HBM	Health Belief Model
HIV	Human Immune Virus
IRB	Institutional Review Board
KCCA	Kampala Capital City Authority
LH	Luteinizing Hormone
MOH	Ministry of Health
NEMA	National Environment Management Authority
PCR	Polymerase Chain Reaction
SANAS	South African National Accreditation System
SPSS	Statistical Package for Social Scientists
STDs	Sexually Transmitted Diseases
STIs	Sexually Transmitted Infections
TB	Tuberculosis
UBOS	Uganda Bureau of Statistics
UNRA	Uganda National Roads Authority
WHO	World Health Organisatrion

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11. Authors' contributions
SSK was the primary author. NT, SL, DC read, and suggested improvements in the manuscript. All the authors herein read, reviewed and found the final manuscript acceptable.

12. Consent for publication
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