

# The Effect of Age on Male Infertility in Gabon

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# Abstract

Background: Male age significantly affects semen parameters. However, there is no evidence on the impact of aging on semen quality in men residing in Libreville, Gabon. **Objective**: To determine the effects of age on semen quality of patients visiting the Reproductive Laboratory at the Academic Hospital Mère Enfant located in Libreville, Gabon. Methods: This descriptive and prospective study was performed between the 1st of October 2021 and 30st of September 2022. This study involved patients who had semen analysis as part of fertility check-up. Semen analysis was performed following the World Health Organization, 2010 guidelines. Semen parameters such as volume, leucocytes count, sperm concentration, sperm motility, norml morphology and vitality were used in the current study. Descriptive statistics were performed. The Chi-square test and the Fisher test were used as statistical tools for the analysis of data. The ROC curve was used to illustrate the data. Results: A total of 148 patients were included in the study. Male infertility prevalence was 77.03%. The average age was 41 years with minimum and maximum of 24 and 61 years respectively. Secondary infertility was predominant (66.9%). Patients displaying asthenozoospermia (22.3%) followed by those displaying oligoasthenozoospermia (OAT) isolated (1.4%) or not (11.6%) were the most frequent semen parameter abnormalities. Men aged more than 45 years were 4.4 times likely to have abnormal semen parameters. Odd = 4.4 (IC 95% = [3.3 - 14.7]). Staphylococcus haemolyticus infection was more prevalent. Conclusion: Male age was found to significantly influence semen quality.

# **Keywords**

Gabon, Infertility, Semen Analysis

## **1. Introduction**

According to the World Health Organization, infertility affects 1 out of 6 people [1]. Infertility can lead to distress, stigmatization and financial difficulties, which can affect the mental and psychology of the affected people. The geography of infertility shows different zones of low fertility rates in some regions compared to others. Infertility is a major public health issue in most countries [1]. In Africa, infertility affects approximately 25% to 40% of the sub-Saharan African population [2]. Infertility is considered a social taboo. It can lead to divorce [3]. Infertility is considered as a fatality or a curse sent to the couple [3]. The blame often lands on the female partner when a couple cannot conceive, although several studies have highlighted male factor infertility as being solely contributing factor of the couple infertility [4] [5]. Male infertility is a simportant as female infertility (31.7% vs 14.7%), in cases where one of the partner is sub-fertile [4].

Semen analysis is the cornerstone for the investigation of male infertility. Male partner is considered sub-fertile when one of the semen parameters (sperm concentration, motility and/or morphology) is below the threshold value in two analysed semen samples evaluated within 4 weeks [6]. Semen quality can be influenced by several factors, including aging. Many studies have found that advanced male age significantly influences sexual functions, sperm parameters, reproductive hormones, and consequently male fertility [7] [8] [9] [10]. Advanced age may result in a decrease in sperm progressive motility [7] [11], morphology, and vitality [12]. Semen volume, sperm concentration, total sperm count, total motile sperm count, and sperm morphology were found to decrease as age increases [7]. Furthermore, several systematic reviews and meta-analysis have highlighted a decrease in semen quality in men aged between 20 and 40 years [13] [14] [15]. This could be due to lifestyle and environment factors such as tobacco, alcohol consumption, medications, stress, obesity, endocrine disrupting chemicals, etc. [16].

The literature on the impact of agin on male infertility residing in Gabon is almost non-existent. Therefore, the main aim of this study is to evaluate the effects of age on semen parameters.

#### 2. Methodology

This prospective, descriptive analysis was performed from the 1st October 2021 to the 30 Septembre 2022 at the Reproductive Laboratory of the Academic Hospital Mère Enfant (CHUME-FJE) located in Libreville, Gabon.

Patients consulting for semen analysis as part of fertility investigation were asked for their willingness to be part of the study. They were adequately informed of the aims, methods, potential risks of the study and other relevant aspects of the study. Once the patient agreed to be part of the research study, a written informed consent for research purposes was obtained from each participant.

A two page questionnaire was designed in order to collect socio-demographic data such as patient age, marital status, factors associated with infertility and

workplace exposure. Semen analysis was collected by masturbation with an abstinence period between 3 and 5 days.

Semen was analysed according to the World Health Organization (2010) guidelines [7]. Sperm concentration was performed using the Kovacs cells, sperm vitality was done using the eosin and nigrosin stain, sperm morphology was done by preparing frosted semen slides and stained using the RAL 555.

Data was entered in an excel spreadsheet then analysed using the Epi Info 7.2.5.0 software. Results are indicated in sample sizes, percentages, mean and standard deviation. For all statistical tests, P-value of <0.05 was considered statistically significant. Multi-varied analysis and logistic regression to determine the odd ratio were done. The Chi-square test and Fisher's Exact Test were used to determine whether or not there is a significant association between two categorical variables.

- <u>The ROC curve</u>: To differentiate patients presenting with abnormalities, and those who do not display any abnormality, those falling under the ROC curve. near 1.
- <u>The Youden's index</u>: To identify the minimum value used to identify the two groups.

In our case, the abnormality is any decrease in semen parameters.

# 3. Results

During this study, a total of 148 patients had their semen samples evaluated. Amongst them, 114 patients (77.03%) had decreased semen quality and 34 patients had normal semen analysis results (22.94%). The socio-demographic data restult is highlighted in **Table 1**. The majority of patients were between the gae interval 31 - 40 years, with most of the patients being married (84.5%). Secondary infertility was more prevalent (66.9%) compared to primary infertility (33.1%). Coital incontinence (10.8%) and Alcohol consumption (47.3%) were also observed in patients.

## • Socio-demographic data

**Table 2** summarizes the type of infertility, semen volume, semen analysis results and swim-up method according to age group. Secondary infertility was more prevalent in the age group 41 - 50 years while the prevalence of primary infertility was higher in the age group 31 - 40 years. The prevalence of hypospermia was higher in the age group 41 - 50 years. The same age group (41 - 50) had a higher incidence of astheno-necrozoospermia, azoospermia, asthenozoospermia, teratozoospermia, and leukocytospermia isolated.

Although abnormal semen results were observed at all age groups, it was more pronounced in the following age categories: 31 - 40; 41 - 50 and 51 - 60 years. There was no significant correlation between age and semen analysis results (**Figure 1**).

The ROC curve is designed in order to evaluate if minimum age can be a prognostic factor for abnormal semen parameters. The area under the curve AUC = 0.6, IC 95% = [0.5 - 0.7] was significantly different from the 0.5 (P = 0.029).

Characteristics	Ν	(%)
Age (Years)		
≤30	9	6.1
[31; 40]	65	44.0
[41; 50]	51	34.5
[51; 60]	22	15.0
>60	1	0.4
Marital Status		
married	125	84.5
Cohabitation	23	15.5
Infertility		
Primary	49	33.1
Secondary	99	66.9
Workplace exposure		
Heat	30	21.4
Stress	17	12.1
Other	101	66.5
Factors associated with infertility		
Herniorraphy	9	6.1
Varicocele	2	1.4
Posterior valve of urethra	1	0.7
Cryptorchidism	1	0.7
Testicular torsion	1	0.7
Mumps	3	2.0
Coital incontinence	16	10.8
Diabetes	5	3.4
Tobacco	0	0.0
Tobacco + alcohol	16	10.8
Alcohol	70	47.3
Herpes	2	1.4
Hepatitis B	6	4.0
None	16	10.8

Table 1. Socio demographic data distribution of participants.

The maximal Youden index (0.247), obtained for a sensitivity of 37.6% and a specificity of 87.1% was observe at a minimal age of 44.5 years.

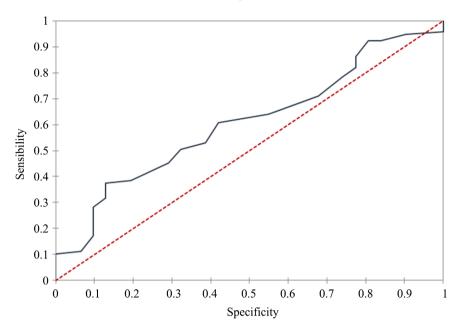
**Table 3** summarizes the distribution of the study population according to age group. The majority of patients were younger than 45 years old (67.6%).

Parameters		Age Group				
	≤30	[31; 40]	[41; 50]	[51; 60]	>60	Р
Infertility Type						
Secondary	7	35	42	14	1	
Primary	2	30	9	8	0	0.0000
Semen Volume (ml)						
<1.5	2	4	9	7	0	
2 - 6	7	58	41	12	1	0.525
>6	0	3	1	3	0	
Semen analysis results						
Astheno-necrozoospermia	2	11	11	9	0	
OAT + Necrozoospermia	2	7	2	2	0	
Necrozoospermia	0	7	5	1	0	
Azoospermia	0	3	4	3	0	
Oligo-astheno-necrozoospermia	0	5	0	2	0	
Asthenozoospermia	0	0	3	3	0	
Teratozoospermia	1	0	4	0	0	
Oligo-asthenozoospermia	0	2	1	1	0	
Necrozoospermia + Leukospermia	0	1	2	0	1	
Asthéno-térato-nécrozoospermie	0	2	1	0	0	
Leukocytospermia isolated	1	2	3	0	0	
Asthénozoospermie	0	2	0	0	0	
Astheno-necrozoospermia + Leukocytospermia	0	2	0	0	0	
OAT	0	2	0	0	0	
OAT + Leukocytospermia	0	1	0	1	0	
OAT + teratozoospermia	0	1	0	0	0	
OAT + Cryptozoospermia	0	0	1	0	0	
Normospermia	3	18	13	3	0	
Results	-				~	
ICSI	1	10	6	5	0	
IUI or IVF	3	23	27	11	1	
Not done	5	32	16	6	0	

Table 2. Type of infertility, semen volume, semen analysis results and swim-up method according to age group.

OAT = Oligoasthenoteratozoospermia; Swim-up test; N = sample size; % = percentage. A correlation between age and type of infertility was found (P = 0.000)

ROC curve/age/AUC = 0.6



**Figure 1.** Determination of the minimum age for the prognostic of abnormal semen parameters.

Table 3. Distribution of the study population according to age group.

Age (years)	N (148)	%
<45	100	67.6
≥45	48	32.4

The effect of age on semen analysis results was highlighted in **Table 4**. More patients aged more than 45 years old had abnormal semen volume (65.6)%. The difference in terms of normal vs abnormal semen volume between the two groups was significant (P = 0.0288). No significant differences were observed in terms of sperm motility and morphology.

The Influence of age on infertility type and semen analysis results was summarized in **Table 5**. Patents aged less than 45 years had 3.4 times more risk of having secondary infertility. Patients aged less than 45 years had 3.3 times more risk of having hypospermia.

The multi varied analysis of risque factors (**Figure 2**) showed that only age is associated with semen quality (P = 0.017). Men aged more than 45 years had 4.4 time risk to have abnormal semen parameters Odd = 4.4 (IC 95% = [1.3 - 14.7]).

# 4. Discussion

## 4.1. Rate of Male Infertility

This study showed that 77.03% of patients had abnormal semen parameters while 22.97% did not. This result is more than previous reported results (57.6%) by Ndoua *et al.* [17] in Cameroon; however, this result is less than previously

 Table 4. Influence of age on semen analysis results.

<45 years	≥45 years	OR [IC 95%]	Р
11 (34.4%)	21 (65.6%)	2.1 [1.1 - 3.9]	0.0288
31 (52.5%)	28 (47.5%)	1.3 [0.9 - 1.9]	0.2161
14 (38.9%)	22 (61.1%)	1,4 [0.8 - 2.5]	0,3939
58 (65.2%)	31 (34.8%)	1.1 [0.9 - 1.5]	0.4689
71 (64%)	40 (36%)	1.2 [1.0 - 1.4]	0.0974
11 (52.4%)	6 (47.6%)	1.1 [0.4 - 2.9]	1.0000
	11 (34.4%) 31 (52.5%) 14 (38.9%) 58 (65.2%) 71 (64%)	11 (34.4%)       21 (65.6%)         31 (52.5%)       28 (47.5%)         14 (38.9%)       22 (61.1%)         58 (65.2%)       31 (34.8%)         71 (64%)       40 (36%)	11 (34.4%)       21 (65.6%) <b>2.1 [1.1 - 3.9]</b> 31 (52.5%)       28 (47.5%)       1.3 [0.9 - 1.9]         14 (38.9%)       22 (61.1%)       1,4 [0.8 - 2.5]         58 (65.2%)       31 (34.8%)       1.1 [0.9 - 1.5]         71 (64%)       40 (36%)       1.2 [1.0 - 1.4]

Table 5. Influence of age on infertility type and semen analysis results.

	Age		Р
Type of infertility	<45	≥45	
Primary	41	8	
Secondary	59	40	0.003 (OR: 3.4)
Semen Volume (ml)			
<1.5	11	14	0.009 (OR: 3.331
2 - 6	86	30	
>6	3	4	
Semen analysis results			
Astheno-necrozoospermia	22	11	
OAT + Necrozoospermia	9	4	
Necrozoospermia	8	4	
Azoospermia	3	7	
Oligo-astheno-necrozoospermia	6	1	
Asthenospermia	1	5	
Teratozoospermia	1	4	
Oligo-asthenozoospermia	2	2	0.077
Necrozoospermia + Leukocytospermie	1	4	
Astheno-terato-necrozoospermia	2	1	
Leukocytospermia isolated	1	5	
Asthenozoospermia	2	0	
Astheno-necrozoospermia + Leukocytospermia	2	0	

Continued			
OAT	2	0	
OAT + Leukocytospermia	1	1	
OAT + teratozoospermia	1	0	
OAT + Cryptozoospermia	1	0	
Hypospermia	0	1	
Normospermia	23	14	
TMS results			
ICSI	12	10	1.000
IUI ou IVF	29	26	

Significant correlation coefficient.

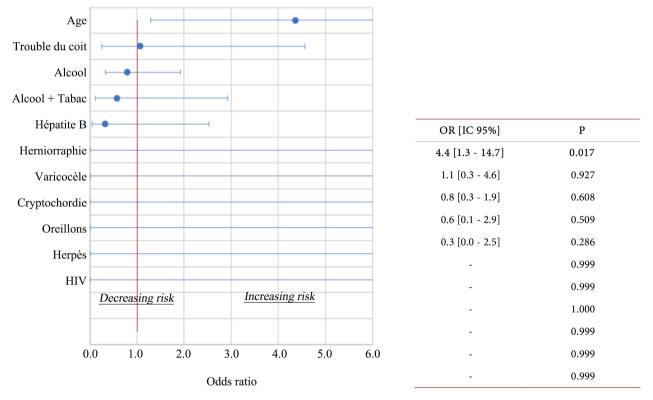


Figure 2. Influence of socio environmental risks factors on semen quality.

reported results (84.3%) by Niang *et al.* in Senegal [18]. The current study did not evaluate female factors infertility, consequently the results are only based on male evaluation.

# 4.2. Socio-Demographic Data

The average age of patients was  $41.53 \pm 7.08$  years with minimum and maximum values of 24 and 61 years. The age group 31 - 40 years was the most represented (44.0%). This age group is almost similar to the reported age interval during

which gabonese men decide to be married 35 - 44 years [19] [20]. Furthermore, the same age group is associated with a desire to have children. The average ages, 42 years, 39 years, 35 years and 31 years were found in Cameroon by Noa Ndoua *et al.* en 2018 au Cameroun [17], Niang L *et al.* in Dakar in 2009 [18], Mohammed Frikh *et al.* in 2021 in Maroc [21] and Matumo *et al.* in Congo in 2020 [22].

Our study population predominantly consisted of businessmen (33.51%), administrators (20.21%), and teachers/lecturers (12.78%). These sectors of activity can represent the population as a whole.

Secondary infertility was mostly predominant (66.9%) compared to 33.1% observed for primary infertility. These results are different from previous ones which indicated primary infertility incidences higher than secondary infertility [21] [23] [24]. The secondary infertility rate found in the current study can be explained by the low fertility rates in young age and adulthood in the Gabonese society with a higher fertility rate observed between the age 25 and 30 years [19]. Furthermore, some economical factors such as cost of living does not favor the population to have children.

#### 4.3. Factors Associated with Male Infertility

In the current study, the main risk factors of male infertility were: Alcohol consumption (47.3%) or associated with smoking (10.8%), coital and erectile dysfunctions (10.8%), Herniorrhaphy (6.1%) and Hepatitis B (4.0%).

Smoking and alcohol consumption are considered as the most harmful risk factors for male infertility. These risk factors can lead to hormonal imbalance through nicotine which induces Leydig cell apoptosis and inhibits biosynthesis of androgens, which are hormones that regulate male reproductive functions. This leads to a decrease in semen quality [23] [24]. Smoking and alcohol consumption were also associated with an increase in seminal ROS with a subsequent rise in sperm DNA fragmentation index [25] [26].

#### 4.4. Influence of Male Age on Semen Parameters

A receiver operator characteristic (ROC) curve was designed for this study (**Figure 1**) in order to evaluate the threshold age for the diagnostic value of semen analysis. The area under the curve AUC = 0.6, IC 95% = [0.5 - 0.7] was significantly different from 0.5 (P = 0.029).

The maximal Youden indice (0.247), obtained for a sensitivity of 37.6% and a specificity of 87.1% was observed at a threshold age of 44.5 years (which was adjusted to 45 years) which helped us to divide our study population in two age categories. (<45 et  $\geq$ 45), with 67.6% (n = 100) for patients less than 45 years and 32.4% (n = 48) for patients aged 45 years and above (**Table 3**).

We have observed a significant decrease in semen volume in men aged more than 45 years (65.6%) compared to those aged less than 45 years (34.4%) with an Odd ratio of 2.1 (IC 95% = [1.1 - 3.9]; P = 0.0288). Furthermore, men aged more

than 45 years had a decrease in smen concentration (47.9%) compared to those aged less than 45 years (37%) although the result was not significant (P = 0.2161). The differences in remaining semen parameters were not significant (**Table 4**). These results were previously found by Frikh *et al.* [27].

## 4.5. Influence of Age on Male Infertility Type

In patients aged more than 45 years, there was a significant association with the type of male infertility (P = 0.003). Additionally, patients aged more than 45 years had 3.4 times more risk to display secondary infertility (OR = 3.4) (**Table 5**). In our study, secondary infertility wasp redominant 66.9% (n = 99), while Benksim *et al.* 2018, in Morocco made different observations [28].

#### 4.6. Age and Semen Parameters

There were differences in semen quality associated with age groups. The most significant changes were observed in the age group 31 to 49 years which represented 72.3% (n = 47) of the total population. Astheno-necrozoospermia was the most present semen abnormality 17% (n = 11), followed by oligo astheno teratozoospermia 17% (n = 11) whether associated with necrozoospermia in 10.8% (n = 7) of cases, or with leukospermia 1.5% (n = 1) of cases.

Results obtained from patients aged less than 45 years showed that astheno-necrozoospermia (66.7%) and oligo astheno teratozoospermie (16.5%) were the most frequent semen abnormalities. The most predominant semen abnormalities observed in patients aged more than 45 years were astheno-necrozoospermie (33.3%) followed by azoospermia (10.5%) (Table 2 et Table 5). Several studies have found that astheno-necrozoospermia and oligo astheno teratozoospermia (OAT) were the most common semen abnormalities. Furthermore, asthenozoospermie was the most frequent abnormality [29] [30].

# 4.7. Influence of Risk Factors on Semen Quality and Male Infertility

The multivariate analysis of risk factors showed that age alone is significantly associated with semen quality (P = 0.017). Patients aged more than 45 years were 4.4 times more likely to have abnormal semen quality. Odd ratio = 4.4 (IC 95% = [1.3 - 14.7]) **Figure 2**. These results are similar to those published by Frickh *et al.* [21]. Several studies have concluded that advanced age could be associated with a decrease insemen volume, sperm motility, with no effect on sperm concentration [31] [32]. Toukam *et al.* highlighted tobacco, exposure to radiation, urogenital chlamydia, syphyllisis, varicoceles, and orchids as main risk of abnormal semen quality [33].

### **5.** Conclusion

Semen quality seems to be impacted by aging in male patients residing in Gabon with an evident threshold.

# **Limitations of the Study**

The limited sample size for the study and the differences in sample size between the evaluated groups were amongst the limitations of the current study.

## **Conflicts of Interest**

The authors declare no conflicts of interest regarding the publication of this paper.

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