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D-Loop Region Variability in Ovarian Cancer Among Senegalese Patients

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ABSTRACT

Our study aimed to determine the genetic variability of the D-loop region in a Senegalese population with ovarian cancer. This research involved healthy and cancerous tissue samples obtained from Senegalese women diagnosed with ovarian cancer. Extraction of mtDNA from the D-loop region, followed by sequencing, yielded 49 nucleotide sequences. These sequences will be utilized to investigate several parameters of genetic variability: allelic frequencies, nucleotide frequencies, and mutation rates. Additionally, we will explore intra- and inter-individual diversity, assess genetic variability between different tissue types from distinct individuals, and illustrate both intra- and inter-individual haplotypic diversity.

Keywords
D-loop, Ovarian, Cancer, Variability, Women, Senegalese.

Introduction
The human body comprises billions of cells, categorized into various types that form tissues [1]. Cell proliferation, crucial throughout life, varies; some cells persist without renewal, while others undergo constant multiplication. Normal cell proliferation hinges on a delicate balance between activating and inhibitory factors [2].

Mitochondrial DNA (mtDNA), a maternally inherited small genome, spans approximately 16,569 (bp), encompassing a coding region (37 genes) and a non-coding D-loop region (~1,192 bp) [3]. Despite its size, variability in this region underlies numerous hereditary diseases [4].

Cancer ranks among the foremost global causes of morbidity and mortality. Over 60% of new cancer cases arise in Africa, Asia, Central America, and Latin America, with Africa reporting higher cancer-related mortality rates than other regions [5]. Ovarian cancer, is notably prevalent and lethal in Africa, including Senegal with 260 new cases and an estimated mortality of 2.8% [6,7].

mtDNA sequences are extensively utilized for diverse analyses [8]; in our study, they assess the genetic variability of the D-loop region in ovarian cancers among Senegalese women. Genetic diversity, arising from gene variations, mutations, chromosomal recombination, and external influences, underscores the differences between individuals within a species. This study investigates allelic and nucleotide frequencies, intra- and inter-population genetic diversity, and haplotypic diversity to gauge genetic variability.

Amid the array of serious illnesses spotlighted by the WHO, cancers persist as significant public health concerns, given their complexity and impact on medical practices. Hence, exploring genetic variability within the D-loop region remains pivotal.

Methods
Samples
49 tissue samples, including both healthy and cancerous tissues, were collected from Senegalese patients following surgery for ovarian cancer. The tissues were preserved in 96% alcohol and transported to the laboratory, where they were stored at 20°C for subsequent molecular analysis.

Genetic study

DNA extraction and amplification of the Dloop gene with sequencing

Total DNA was extracted from the tissues using the Qiagen protocol, following the recommended steps and kit instructions. The D-loop gene was amplified using specific primers: 5'TGTTAAAAGTGCATACCGCCA3' and 5'AGCCATTTACCGTACATAGCACA3'. This involved PCR amplification with DNA polymerase, dNTPs, and Mg^{2+} ions, repeating the process approximately thirty times to enhance the detection of the target DNA.

Sequencing was performed using a specialized PCR reaction that includes ddNTPs, and modified nucleotides labeled with fluorochromes (ddATP-green, ddTTP-red, ddCTP-blue, and ddGTP-yellow), which terminate DNA strand elongation by lacking a 3' hydroxyl group on the deoxyribose. Sequencing involves randomly fragmenting the genome into pieces of DNA of a few thousand base pairs for easier manipulation. To do this, ddNTPs are incorporated by a polymerase to block the elongation of the complementary DNA during copying, as the nucleotides cannot form phosphodiester bonds with each other due to the absence of a hydroxyl group on the 3' carbon.

Determining the genetic variability of D-loop

Sequences were aligned and cleaned using Bioedit version 8.0.5 [9] with the ClustalW algorithm [10] for the analysis of site homologies, variability indices, and genetic diversity. Various variability parameters, such as the number of polymorphic sites, mutations, average number of nucleotide differences (K), and mutation rate (R), along with analyses of similarity degree, nucleotide diversity (π), and haplotypic diversity (h), were determined for each group (healthy tissue, cancerous tissue) using DnaSp software version 5.10.01 [11].

The Mega software [12] was employed to calculate nucleotide substitution probabilities, providing the percentage of substitution between two bases. In fact, it is possible to calculate the frequency of each nucleotide on the genome, the rules Waston-Crick matching rules: fA = fT and fG = fC. This equidistribution indicates that the G+C rate of the genome is 50%, ensuring a uniform distribution.

DNA methylation, a universal phenomenon, was studied in correlation with microsatellite polymorphism to explore its potential for distinguishing between healthy and diseased tissues. Additionally, nucleotide frequency calculations and the average numbers of synonymous (S) and non-synonymous (N) substitutions per site were investigated using MEGA 6 software [12].

Results and Discussion

Polymorphism and hypermethylation of mtDNA

Firstly, it should be noted that C→T transitions are well-known examples resulting from recombination between genome and epigenome properties, particularly hypermethylation. In this study, analysis of this transition was conducted in 8 healthy and 9 diseased individuals, all at position 544. The repeated occurrence of this transition indicates instability (Figure 1).

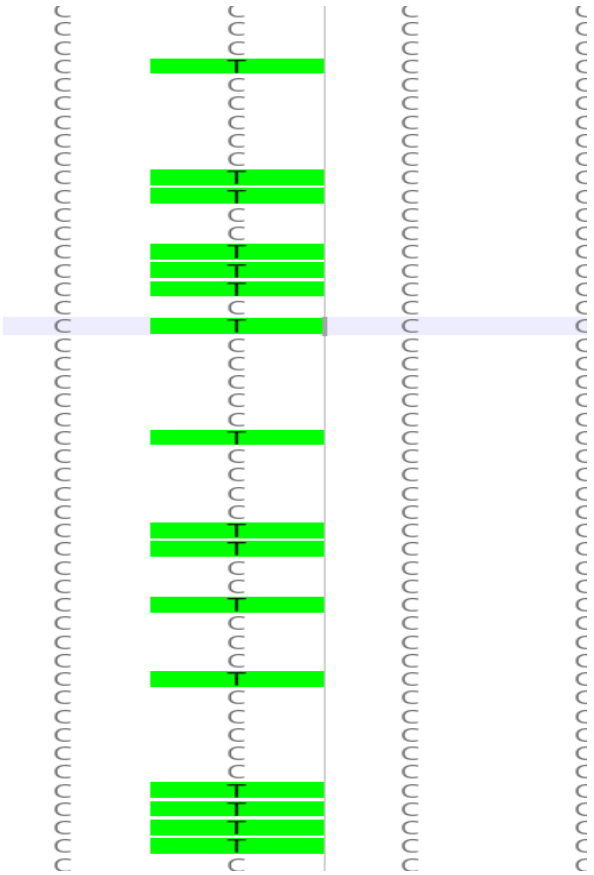

Figure 1: Representation of the C→T transition.

Finally, there is a mitochondrial microsatellite polymorphism located between positions 290 and 310 in the HV2 region, consisting of a cytosine repeat interrupted by a thymine. This supports the hypothesis of hypermethylation (C→T) of mtDNA previously highlighted. This microsatellite polymorphism is found in both healthy and cancerous tissues. The first section of the poly C region has 7Cs, and the second section has 6Cs, a pattern observed in both tissue types (Table 1).

Allele Frequency in Sub-Populations for Each Type of Fluctuation

The table below shows the alternative allele frequencies in four population groups. This represents the number of occurrences of the alternative allele divided by the number of chromosomes in each sub-population. The m.302A>C allele is the most frequent, with an expression rate of 12.63% in the African population, 48.73% in the American population, 15.11% in the European population, and 8.49% in the South Asian population. The most frequent allele in the African population alone is m.183A>G, with a frequency of 2.20%, but it is also present in other populations at lower frequencies. The alleles m.214A>G, m.215A>G, m.227A>G, m.279T>C, and m.292T>C are also present in all sub-populations (Table 2).

Table 1 : Illustration of microsatellite polymorphism.

Healthy tissue			Cancerous tissue
TS1	CCCCCCCTCCCCCC	TC1	CCCCCCCTCCCCCC
TS2	CCCCCCCTCCCCCC	TC2	CCCCCCCTCCCCCC
TS3	CCCCCCCTCCCCCC	TC3	CCCCCCCTCCCCCC
TS4	CCCCCCCTCCCCCC	TC4	CCCCCCCTCCCCCC
TS5	CCCCCCCTCCCCCC	TC5	CCCCCCCTCCCCCC
TS6	CCCCCCCTCCCCCC	TC6	CCCCCCCTCCCCCC
TS7	CCCCCCCTCCCCCC	TC7	CCCCCCCTCCCCCC
TS8	CCCCCCCTCCCCCC	TC8	CCCCCCCTCCCCCC
TS9	CCCCCCCTCCCCCC	TC9	CCCCCCCTCCCCCC
TS10	CCCCCCCTCCCCCC	TC10	CCCCCCCTCCCCCC
TS11	CCCCCCCTCCCCCC	TC11	CCCCCCCTCCCCCC
TS12	CCCCCCCTCCCCCC	TC12	CCCCCCCTCCCCCC
TS13	CCCCCCTTCCCCCC	TC13	CCCCCCCTCCCCCC
TS14	CCCCCCCTCCCCCC	TC14	CCCCCCCTCCCCCC
TS15	CCCCCCCTCCCCCC	TC15	CCCCCCCTCCCCCC
TS16	CCCCCCCTCCCCCC	TC15	CCCCCCCTCCCCCC
TS17	CCCCCCCTCCCCCC	TC17	CCCCCCCTCCCCCC
TS18	CCCCCCCTCCCCCC	TC18	CCCCCCCTCCCCCC
TS19	CCCCCCCTCCCCCC	TC19	CCCCCCCTCCCCCC
TS20	CCCCCCCTCCCCCC	TC20	CCCCCCCTCCCCCC
TS21	CCCCCCCTCCCCCC	TC21	CCCCCCCTCCCCCC
TS22	CCCCCCCTCCCCCC		
TS23	CCCCCCCTCCCCCC		
TS24	CCCCCCCTCCCCCC		
TS25	CCCCCCCTCCCCCC		
TS26	CCCCCCCTCCCCCC		
TS27	CCCCCCCTCCCCCC		
TS28	CCCCCCCTCCCCCC		

Table 2: Alternative frequencies in sub-populations and in Mitomap

HGVS	Alternative frequency in different sub-populations%				Frequency Mitomap%
	Africa	America	Europe	South Asian	
m.109G>A	-	0,0179	-	-	0
m.125T>C	-	3,48	0,0426	0,201	0,33
m.161T>C	-	-	0,00774	-	0
m.168T>C	-	-	0,00774	-	0
m.179T>C	0,0209	0,385	0,00387	-	0,02
m.183A>G	2,20	0,315	0,337	0,134	0,59
m.214A>G	0,147	1,77	0,101	1,14	0,3
m.222C>T	-	0,0175	0,162	0,335	0,06
m.215A>G	0,154	0,773	1,45	0,873	0,88
m.227A>G	0,0418	0,0699	0,403	0,268	0,31
m.251G>A	0,0279	0,0175	0,0658	-	0,04
m.270A>G	-	-	0,00774	0,402	0,01
m.272A>G	0,16	0,0525	0,0271	0,0669	0,09
m.276A>G	-	-	-	-	0
m.279T>C	0,307	0,683	0,36	0,0671	0,56
m.288A>G	-	0,0175	0,0116	-	0
m.292T>C	0,0349	0,122	0,0929	0,067	0,1
m.302A>C	12,63	48,73	15,11	8,49	0,04
m.310_311insTC	0,0209	-	0,882	0,0669	0,19
m.346T>C	0,0209	0,0349	-	-	0
m.16393C>T	-	-	-	-	-
m.16440T>C	-	-	-	-	0
m.16505T>C	-	-	0,00387	0,0669	0,01

Nucleotide Frequencies and Mutation Rates

Table 3 shows 24 substitution patterns for different nucleotides. It should be noted that substitution patterns are used to describe how sequences evolve through nucleotide replacement over time. This table presents the standard nucleotide substitution patterns obtained using the Phase package in the Mega software [12], which can also be used to obtain specific base pair substitutions The transition/transversion rate shows a slight variation in 20 models, where there are differences is at the level of the 4 other models which are those of JC; JC+I; JC+G; JC+G+I with a rate (R) which equals 0.5. This consistency is due to the JC (Jukes-Cantor) model, which is based on the principle of equal base frequencies and induces a uniform transition/transversion rate for all substitutions. This supports the observation that there is little or no diversity in nucleotide composition between the two groups of individuals studied here. The percentage composition of nucleotides in cancer tissues is as follows: %A= 24.8; %G=30.3; %T=27.5 and %C=17.4. These results on nucleotide composition confirm the HKY model, which indicates a higher frequency of guanine nucleotides.

Intra- and inter-individual genetic diversity of D-loop

Amplification of the sequences in the D-loop region produced 559 bp in the control tissues. A total of 52 polymorphic sites were found in healthy tissue, with 23 being informative sites. The total number of mutations (Eta) was 57, and the G+C content was 47.7% across the 559 sites. In the control tissues, there were 27 haplotypes, with a haplotypic diversity (Hd) of 0.99, a nucleotide diversity per site (Pi) of 0.01, and a total number of nucleotide differences (k) of 8.87.

In cancerous tissues, sequence amplification also yielded 559 bp with 27 polymorphic sites, 16 of which were informative. There was a total number of mutations (Eta) of 28, and the G+C content was 47.7%, the same as in the control tissues, indicating that the gene region studied in these two groups was stable. The number of haplotypes (h) was 16, with a haplotypic diversity (Hd) of 0.97, a nucleotide diversity (Pi) of 0.01, and a total number of nucleotide differences (k) of 6.71.

It should also be emphasized that the percentages of transitions and transversions found in both healthy and cancerous tissues show that the mutations found here are not random. Twice as many transitions were noted as transversions, especially in cancerous tissue, with a transition rate of 23%, much higher than the 5% rate of transversions (Table 4).

Genetic Variability Between Two Types of Tissue from Different Individuals

In the table below we have tried to make a comparison between cancerous tissues and supposedly healthy tissues. The choice of tissues to be compared was made randomly, taking care not to compare two types of tissue and tissues from the same individual so as to broaden the field of similarity. The Monte Carlo disparity test used here was repeated up to 1000 times in order to obtain a good estimate of the values. It should be noted that pairs of tissues with a zero nucleotide distance all, without exception, have a similarity rate greater than 99% (Table 5).

Table 3 Maximum likelihood fit for 24 different nucleotide substitution patterns.

Model	R	Freq A	Freq T	Freq C	Freq G
K2+G+I	2,36	0,25	0,25	0,25	0,25
K2+G	2,3	0,25	0,25	0,25	0,25
HKY+G+I	2,34	0,24	0,27	0,17	0,3
T92+G+I	2,35	0,26	0,26	0,23	0,23
HKY+G	2,29	0,24	0,27	0,17	0,3
T92+G	2,28	0,26	0,26	0,23	0,23
TN93+G+I	2,34	0,24	0,27	0,17	0,3
TN93+G	2,31	0,24	0,27	0,17	0,3
JC+G+I	0,5	0,25	0,25	0,25	0,25
JC+G	0,5	0,25	0,25	0,25	0,25
GTR+G+I	2,35	0,24	0,27	0,17	0,3
GTR+G	2,32	0,24	0,27	0,17	0,3
K2+I	2,22	0,25	0,25	0,25	0,25
HKY+I	2,23	0,24	0,27	0,17	0,3
T92+I	2,22	0,26	0,26	0,23	0,23
TN93+I	2,23	0,24	0,27	0,17	0,3
K2	2,21	0,25	0,25	0,25	0,25
JC+I	0,5	0,25	0,25	0,25	0,25
HKY	2,21	0,24	0,27	0,17	0,3
GTR+I	2,24	0,24	0,27	0,17	0,3
T92	2,21	0,26	0,26	0,23	0,23
TN93	2,21	0,24	0,27	0,17	0,3
JC	0,5	0,25	0,25	0,25	0,25
GTR	2,21	0,24	0,27	0,17	0,3

Table 4: D-loop genetic diversity parameters.

	Healthy tissue	Cancerous tissue
Number of sequences	28	21
Number of sites	559	559
Number of sites invariables	507	332
Number of site variables	52	27
Number of gene copies	28	21
Number of loci	57	57
Number of polymorphic loci	52	27
Number average nucleotide difference (K)	8,87	6,71
Nucleotide diversity (Pi)	0,01	0,01
Number of haplotypic (h)	**27**	16
Haplotypic diversity (Hd)	0,99	0,97
Number of sites with segregation (S)	18	28
Tajima test statistics (D)	-1,48	-0,49
FST specificity between populations	0,00232	0,00372
Nucleotide frequencies in healthy tissue		
T(%) C(%)	A(%)	G(%)
27,4 24,9	17,5	30,2
%Transition	35%	
%Transversion	22%	
Nucleotide frequencies in cancer tissues		
T(%) C(%)	A(%)	G(%)
27,5 17,4	24,8	30,3
%Transition	23%	
%Transversion	5%	

Table 5: Similarity search between individuals and different tissues.

Pair of Tissue	Disparity Index	Nucleotide distance	Similarity (%)
TC1-TS5	0	0,01	98,92
TC1-TS37	0,04	0,01	98,92
TC1-TS53	0,04	0,01	98,38
TC1-TS55	0,01	0,03	96,95
TC1-TS61	0,02	0,01	98,38
TC1-TS65	0,01	0	99,10
TC1-TS29	0	0,01	98,74
TC1-TS3	0	0,01	98,74
TC2-TS5	0	0,01	98,92
TC2-TS11	0,04	0	99,28
TC2-TS25	0,03	0	99,28
TC2-TS37	0,04	0,01	98,92
TC2-TS55	0,01	0,03	96,95
TC2-TS61	0,01	0,01	98,38
TC2-TS63	0,04	0	99,28
TC2-TS65	0	0	99,10
TC2-TS29	0	0,01	98,74
TC2-TS3	0	0,01	98,74
TC4-TS29	0,04	0,01	98,92
TC4-TS3	0,02	0,01	99,28
TC9-TC2	0,04	0	99,28
TC10-TS61	0,02	0,01	98,92
TC13-TS45	0,02	0,01	98,92
TC13-TS29	0,04	0	99,28
TC14-TS45	0,02	0,01	98,92
TC16-TS5	0	0,01	98,74
TC16-TS17	0,04	0,02	97,49
TC16-TS55	0,02	0,03	96,77
TC16-TS61	0	0,01	98,21
TC16-TS65	0	0,01	98,92
TC16-TS29	0	0,01	98,56
TC16-TS3	0	0,01	98,56
TC19-TS45	0,04	0	99,10
TC19-TS29	0	0	99,46
TC19-TC16	0,04	0	99,10
TC20-TS29	0	0	99,46
TC33-TS45	0,04	0	99,10
TC34-TS9	0,01	0,01	98,92
TC34-TS45	0	0,01	98,74
TC35-TS29	0,01	0,01	99,46

Haplotypic diversity found within and between individuals

The table below shows the calculation of haplotypic frequency in Excel for each population and enabled us to obtain the average number of different haplotypes (average A = 21.5), the number of haplotypes specific to each group of individuals (average P = 17.5), the effective number of haplotypes (average Ne = 20.18), and the haplotypic richness (average Rh = 17.22), which is estimated by the rarefaction method [13]. This method provides a clear picture of haplotypic diversity both within and between populations (Table 6).

Table 6: Illustration of haplotype diversity.

	A	P	Ne	Rh
Healthy Tissues	27	23	26,13	19,44
Cancerous tissue	16	12	14,22	15
Average	21,5	17,5	20,18	17,22

Regarding the 39 haplotypes found in this study after searching the mtDNA H haplogroup database, we noted 26 that were highlighted, 12 haplotypes that were not listed, and haplotype H22, which has no haplogroup parent. Always mentioning the position of the variant. The two types of tissue have 4 haplotypes in common: haplotype m.3992+5004+9123C>T; haplotype m.146+195+709+13101+16288T>C; haplotype m.3936+14552A>G and haplotype m.55+57+6253T>C (Table 7).

Table 7: List of Dloop haplotypes in ovarian cancer.

Haplogroups	Variant positions	Nb TS	Nb TC	Accession
H1	m.3010G>A	1	0	Ref. GS 1001
H2	m.1438A>C	1	0	Ref. GS 1016
H3	m.6776T>C	1	0	Ref. GS 1027
H4	m.3992+5004+ 9123C>T	2	1	Ref. GS 1032
H5	m.16304T>A	1	0	Ref. GS 1039
H6	m.239 + 16482T>C	1	0	Ref. GS 1042
H7	m.4793A>G	1	0	Ref. GS 1051
H8	m.146+195+709+13101+ 16288T>C	1	2	Ref. GS 1053
H9	m.152 + 13020T>C	1	0	Ref. GS 1054
H10	m.14470AT>C	1	0	Ref. GS 3054
H11	m.8448+13759+16311T>G	1	0	Ref. GS 1055
H12	m.3936+14552A>G	1	1	Ref. GS 1057
H13	m.14872C>T	1	0	Ref. GS 1059
H14	m.7645+10217T>C	1	0	Ref. GS 3068
H15	m.55+57+6253T>C	1	1	Ref. GS 1067
H16	m.10394C>T	1	0	Ref. GS 1005
H17	m.3915G>T	1	0	Ref. GS 1013
H18	m.13708+14364G>A	1	0	Ref. GS 1015
H19	m.6272+14869A>G	1	0	Ref. GS 3076
H20	m.16218C>A	1	0	Ref. GS 3077
H21	m.186+3822+8994G>A	1	0	Ref. GS 1064
H23	m.10211C>T	1	0	Ref. GS 1010
H24	m.16293A>C	1	0	Ref. GS -
H25	m.9620C>A	1	0	Ref. GS 3082
H26	m.11152T>A	1	0	Ref. GS -
H27	m.11719+16093+16316A>G	1	0	Ref. GS -
H28	m.186A+8715+11191T>C	0	2	Ref. GS -
H29	m.93+573.1CC+5582CAdel	0	1	Ref. GS -
H30	m.195+504+8200+8638+8817 + 14241T>C	0	1	Ref. GS 3079
H31	m.7930T+10771A>G	0	1	Ref. GS -
H33	m.11447G>A	0	2	Ref. GS -
H34	m.15519+16291T>C	0	1	Ref. GS -
H35	m.3342T>C	0	1	Ref. GS -
H36	m.152+3525A+16070T>C	0	2	Ref. GS -
H37	m.3531G>A	0	1	Ref. GS -
H38	m.235A>C	0	1	Ref. GS -
H39	m.16299A>C	0	1	Ref. GS -

The evolution of these mtDNA haplotypes through mutations helped to evaluate the historical expansion observed in the two groups of tissues in this study. This expansion was tested by a distribution of mismatches made on all the sequences, and we found a multimodal distribution of mismatches. A first peak was noted with 6 nucleotide differences between two pairs of haplotypes, with an allelic frequency of 0.10289. We also noted two other large peaks, with respective nucleotide differences of 9 and 19, and allelic frequencies of 0.08231 and 0.01029, respectively (Figure 2).

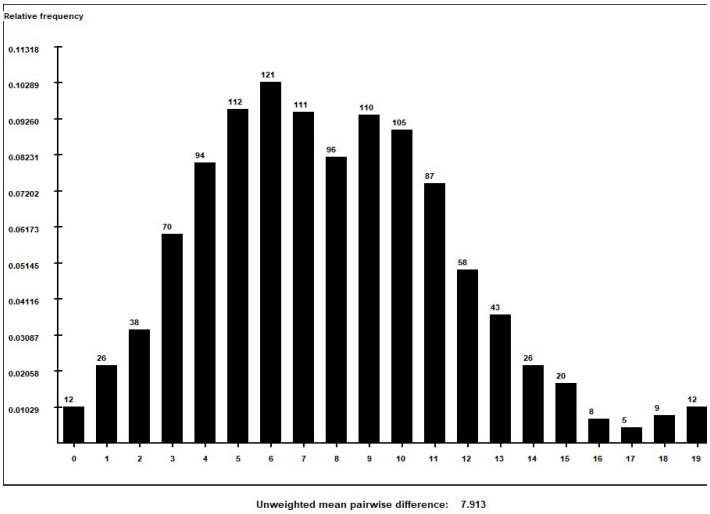

Figure 2: Illustration of mismatches of mtDNA sequences in ovarian cancer.

Discussion

Length changes or microsatellites, which are tandem repeats in the genome inducing instability, are responsible for the formation of new alleles in ovarian tumors through the insertion of a repeat unit found in the two types of tissue studied here. This instability in the D310 region is manifested by poly C insertions, with the first section comprising 7C and the second section 6C, and this distribution is specific to the two types of tissue. This finding is supported by research [14] on intestinal carcinomas, which showed that 10 carcinomas exhibited a mtMSI phenotype. The D310 microsatellite polymorphisms found in this study were accompanied by changes in the methylation level of the D-loop. Recombination between genome and epigenome properties, materialized by hypermethylation of mtDNA for C→T transitions, was found in 8 healthy tissues and 9 cancerous tissues in the ovarian cancers of Senegalese women. Until now, mtDNA

hypermethylation has only been associated with certain diseases such as Alzheimer's in post-mortem brains and Parkinson's disease, as well as studied by Stoccoro et al. [15].

The research we carried out on the altered alleles of the D-loop gene in the gnomAD databases highlighted the altered allele MT-302-A-C, which is the most present in the populations with frequencies of 8.49%, 12.63%, 15.11%, and 48.73% in the Asian, African, European, and American populations, respectively. The alleles MT-183-A-G, MT-214-A-G, MT-215-A-G, MT-227-A-G, MT-279-T-C, and MT-292-T-C were also found in all sub-populations, albeit with low frequencies, showing that the populations share certain alleles due to ancestral heritage. However, the differences in frequencies are the result of diversity, which is considered an important indicator in genetics.

Using the 49 sequences to study genetic diversity and tumor variation in ovarian cancer, differences between the parameters studied were noted. High polymorphism was observed, with haplotypic diversity equal to 0.97 and nucleotide diversity equal to 0.01. The rate of nucleotide diversity is low but consistent in both types of tissue. This indicates that there is no significant difference between the healthy and cancerous tissues studied for ovarian cancers in Senegalese women.

The molecular diversity studied here shows that the fluctuations observed are not random, as the transition rate (23%) is much higher than the transversion rate (5%). The study of sequence evolution using the disparity index, nucleotide distance, and similarity search between pairs of sequences reveals 25 pairs of sequences that are 99% similar, further highlighting the high similarity of the sequences in this study. Analysis of the variability of the mtDNA D-loop region in the ovarian cancers of Senegalese women shows a bimodal distribution within and between the two tissue types. This is due to the fact that there is not a significant difference in all sequences, and the multimodal mismatch distribution curve at the nucleotide level supports our other results. However, given that the tests at the haplotypic level show an expansion between the two groups of tissues studied here, the theory of subdivision of a parent population into two is suggested. The study by Hart DL [16] provides further support, as he suggests that the subdivision of a parent population into two subgroups, taking into account demographic distribution, would reduce the rate of evolution Regarding haplotype analysis, the diversity of genetic structure in the two types of tissue showed an evolution of certain haplotypes. Even though there is an evolution of the haplotypes found in the D-loop region in ovarian cancers, they still belong to the same group or haplogroups. For there to be a change of group, a certain number of mutations are required between the haplotypes, and here in this study, the mutations leading to evolution do not reach 20 mutations in the 5 evolved haplotypes. This can be supported by [17] who report that haplotypes belonging to the same haplogroup differ by a few mutations not exceeding 20. Beyond this number of mutations, the haplotypes will belong to different haplogroups.

Conclusion

This study focuses particularly on the utilization of fundamental molecular parameters and genetic diversity. The results reveal that studying molecular alterations in ovarian cancer is challenging due to the disease's morphological heterogeneity. However, employing new-generation tools and techniques enabled us to identify microsatellite variations and recombinations between the genome and epigenome in this study. These factors are significant contributors to alterations in the D-loop region of mtDNA, accompanied by modifications at the allelic and nucleotide levels, which manifest as high polymorphisms associated with ovarian tumors.

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