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p53 Codon 72 arginine/proline polymorphism and cancer in Sudan

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Abstract The aim of this report is to determine frequencies and associations of p53 codon 72 arg/pro polymorphism with different types of cancer in Sudan. p53 codon72 arg/pro polymorphism distribution and allele frequencies in 264 samples of different types of cancers were investigated using PCR. The results were compared to 235 normal controls. The results indicated significant differences in frequency and genotype association between different types of cancers. Breast carcinoma patients most prominently showed excess of homozygous arg genotype as compared to controls with an Odd ratio (OR) of 19.44, 95 %CI: 6.6–78.3, $P < 0.0001$. Less prominently cervical cancer showed genotype effect of 2.4 OR, 95 %CI: 1.12–5.33, $P = 0.015$, while esophageal cancer had an OR of 0.57, 95 %CI: 0.23–1.42, $P = 0.1$. In Burkitt's lymphoma, however, in contrast the homozygous arg accounted for only 6.9 %, (OR 0.18, 95 %CI: 0.02–0.89, $P = 0.018$). We concluded that p53 arg/pro polymorphism has different pattern of frequency in different types of cancer among Sudanese patients, indicating perhaps different etiology and biology of these tumours.

Keywords P53 arg/pro polymorphism-Sudan-cancers

Introduction

The tumor suppressor protein p53 plays a critical role in cell cycle control and apoptosis [1]. Somatic mutations in p53 are found in more than 50 % of human cancers. However, germ line mutations in p53 are rare and only few polymorphisms have been described [2]. One of the most common variants believed to be associated with cancer development is the codon 72 single nucleotide polymorphism (SNP), which results in the substitution of proline for arginine [3]. The association of this polymorphism has long been a subject of reports and controversies with regards to its functions and association with different cancers, including Lung, breast, prostate, esophageal, nasopharyngeal, Burkitt's lymphoma, and cervical cancer (reviewed by Whibley [4]). p53 arg/pro polymorphism allele frequency among normal Sudanese populations was reported previously [5], however, the frequency of the SNP and its association with different cancers has never been investigated.

The aim of this report is to determine frequencies and associations of p53 codon 72 arg/pro polymorphism with some common types of cancers in Sudan, breast and cervical cancer in women, Burkitt's Lymphoma in children, and nasopharyngeal carcinoma (NPC) and Esophageal cancer among males.

Materials and methods

Following the appropriate guidelines and ethical approval, DNA samples from the most common types of cancer in Sudan including, breast, cervical, nasopharyngeal carcinoma, and esophageal cancer were obtained from the Tumour Bank of the Institute of Endemic diseases/University of Khartoum in the form of tissue biopsy. The Burkitt's lymphoma in addition to some breast cancer samples were collected retrospectively from Elhassan Medical specialized Laboratory.

p53 codon72 arg/pro polymorphism distribution and allele frequencies were investigated using PCR as described

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Table 1 Clinical characteristics of the studied patients

Cancer type	Age		Gender		Pathology	Clinical staging IUCC			
	Range (years)	Mean	Female	Male		I	II	III	IV
Breast cancer N:92	28–85	56	89	3	DCIS 2 % NOS 57 % Mixed (DCIS + NOS) 27 % LC 14 %				
Burkit's lymphoma	3–56	16.3	7	22	(100 %) Burkitt's lymphoma	–	–	–	–
Nasopharyngeal carcinoma N:30	10–74	42	8	22	WHO type II (50 %) 15/30 WHO type III (50 %) 15/30	2 (6.7 %)	3 (10 %)	7 (23.3 %)	18 (60 %)
Cervical cancer N:78	21–85	53.6	78	0	SCC 82.1 % (64/78) AC 17.9 % (14/78)	19.2 % (15/78)	51.3 % (40/78)	24.4 % (19/78)	5.1 % (4/78)
Esophageal cancer N:25					SCC 72 % (18/25) AC 28 % (7/25)				

DCIS Ductal carcinoma in situ, *LC* lobular carcinoma, *NOS* invasive ductal carcinoma, *WHO* World Health Organization classification, *SCC* Squamous cell carcinoma. *AC* adenocarcinoma, *AJCC* American Joint Committee on Cancer

Table 2 Frequencies of arg/pro genotypes in different types of cancer

Cancer type genotype	Breast (n:92) (%)	Cervical (n:78) (%)	Burkitt's lymphoma (n:29) (%)	Nasopharynx (n:30) (%)	Esophageal (n:25) (%)	Controls (n:235) (%)
Arg	87	62	32.75	50	51.3	48.75
Pro	13	38	67.25	50	48.7	51.25

Table 3 Allele frequency of p53 polymorphism among different cancer types

Cancer type	Hetero arg/pro			Homo arg/arg		
	OR	95 %CI	*P* value	OR	95 %CI	*P* value
Breast cancer	1.8	0.55–7.75	0.3	19.44	6.6–78.3	>0.0001
Cervical cancer	0.91	0.43–1.96	0.78	2.4	1.12–5.33	0.015
Burkitt's lymphoma	0.57	0.23–1.42	0.17	0.18	0.02–0.89	0.018
Nasopharyngeal carcinoma	0.79	0.29–2.3	0.61	1.1	0.33–3.62	0.86
Esophageal cancer	1.24	0.5–3.43	0.67	1.23	0.39–3.97	0.688

OR Odd ratio, *95 %CI* confident intervals

previously [5] and the results were compared to 235 normal controls. The p53 codon72 genotype distribution and allele frequencies were calculated and statistically analyzed using χ^2 test and logistic regression in the program R. Since proline is believed to be the ancestral allele, the calculation was carried against proline in effect terms.

Results

Two hundred and sixty-four samples were investigated. Available Pathology and clinical staging of the patients are shown in Table 1. The results indicate the presence of considerable differences in frequency and genotype association between different cancers as shown in Tables 2 and 3 respectively. The arg/arg genotype in breast cancer as compared to controls had an Odd ratio (OR) of 19.44, 95 %CI: 6.6–78.3 and $P < 0.0001$. In cervical cancer the homozygous arg genotype was detected in 42.3 % (33/78), while the heterozygous arg/pro was detected in 38.5 % (30/78) and only 19.2 % (15/78) had the pro/pro genotype, with a genotype effect of OR = 2.4, 95 % (CI: 1.12–5.33, $P = 0.015$). NPC had an OR = 1.1, 95 %CI: 0.33–3.62 with P value = 0.86 for the arg/arg. In Burkitt's

lymphoma an opposite pattern seems to exist with a major effect from the pro allele, where the homozygous arg accounted for only 6.9 %, (OR = 0.18, 95 %CI: 0.02–0.89, $P = 0.018$), the arg/pro was 51.7 % and pro/pro 41.4 % (OR = 0.57, 95 %CI: 0.23- 1.42, $P = 0.17$). In esophageal cancer again the heterozygous show the highest frequency with arg/pro 56 %, arg/arg 23.1 %, and pro/pro 20.5 %. But in both esophageal and nasopharyngeal cancers the difference seems to be low and is not supported statistically.

Discussion

The possibility that polymorphic variants in p53 might contribute to cancer susceptibility has been extensively investigated. Studies on codon 72 were reported with its association with the risk of variety of cancers including breast, nasopharyngeal, Burkitt's lymphoma, esophageal and cervical cancer. [4].

Although Buyru et al. [6] and Dosti et al. [7] found positive correlation among Turkish and Iranian patients respectively, our results indicate a spectacular frequency of homozygous p53 arg allele in the breast cancer patients as compared to controls with an OR of 19.4 for the arg/arg genotype. Such association between arg allele and breast cancer may qualify the homozygous form to be considered a susceptibility/resistance marker.

In cervical cancer the result was consistent with a previous study by Michelina et al. [8] on a series of Brazilian women with carcinomas of the cervix. The authors found that 67 % of 148 women were homozygous (arg/arg). However, the published data on the prevalence of p53 arg/pro polymorphism in cervical cancer patients are still controversial. As anticipated, the ethnic background seem to be an important reason for discrepancies in the disease frequency association of this polymorphism as implied by two reports among Italian women [9, 10] In line with the contribution of ethnic stratification is the lack of significant difference between the two allele in the development of NPC in Sudan. This, contradicts previous reports of association of the pro allele with nasopharyngeal carcinoma in Tunisia [11]. The discrepancy may be due to uneven distribution of the allele among different Sudanese Ethnic groups [5]. Nasopharyngeal carcinoma is highly endemic among Nuba tribe [12] who has a high frequency of proline allele [5]. This ethnic stratification may mask the true effect of the allele among studied population.

In esophageal cancer samples, the heterozygotes showed the highest frequency. This result is consistent with that of Piao et al. [13] in Korean population. Jiang et al. [14] who reported that arg decreases the risk of esophageal cancer development.

The association between BL and p53 polymorphism was not previously investigated. The contrasted pattern of the p53 arg/pro polymorphism as compared to other cancers seen in this report warrant further in investigations possible etiologic/biological factors.

It is well-known that cancer occurs as a multistep complex disease involving interaction between the genome and the environment and/or variations and mutations in different loci. From the limited information available in Sudanese cancers, the significance of such interactions are speculated as well as the role of epigenetic modification and viral oncogenesis [15]. The greater combined risk of p53 with Rb LOH reported recently by our group is an example of such possible epistatic effect of several genes/polymorphisms [16].

In conclusion we identified a disparate role of p53 codon 72 in cancers in Sudan and suggested that genotyping and expression profiling of populations at risk could become a useful means of identifying risk, and that the difference in cancer associations with this locus may suggest a possible different etiologies and molecular pathways.

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Conflict of interest This manuscript has been read and approved by all authors. This paper is unique and is not under consideration by any other publication and has not been published elsewhere. The authors and peer reviewers of this paper report no conflicts of interest. The authors confirm that they have permission to reproduce any copyrighted material. Written consent was obtained from the patient or relative for publication of this study.

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