

Association of the TP53 Codon 72 Polymorphisms with PCOS Female Infertility in Karbala City

Inam Joudah Radhi¹, Abdulmutalb Badr Manhy Alkhaleeli², Hameedah Hadi AbdulWahid³

¹Assist. Lecturer, ²Lecturer, College of Pure Science, Department of Chemistry, University of Kerbala, Iraq,

³Consultant Gynecologist, Kerbala Obstetric and Gynecology Teaching Hospital, Kerbala Health Directorate, Ministry of Health, Iraq

Abstract

To training the association of polycystic ovaries with p53 and 72 polymorphisms in the population of female infertility in central Karbala and to search for possible interaction with the polymorphism. The Iraqi environment was suffered from acts of profanation by due to wars in Iraq, since 1990 and after 2003, An enormous number of damages and deaths were affected by destructive chemicals and radioactive resources. These events resulted in either cancer or infertility. Infertility is one of the medical, social and psychological burdens in Iraqi society. After 12 months or more of failure to a clinical pregnancy lead to a regular unprotected sexual intercourse.

Keywords: Women sterility, PCOS, polymorphism, Codon p53 and 72.

Introduction

Infertility is defined as a disease of the failure to achieve a clinical pregnancy after 12 months or more of regular unprotected sexual intercourse [1].

The disability to become pregnant consequences in an important affecting and profitable tolls, among that artificial nervousness and sadness are more public in sterile couples than sterile couples, also Assisted Reproductive Technologies (ART) is accessible toward care for sterility, then these are pricey. The credit of changeable hazard influences for sterility might guide to economical then helpful interferences expected at preclusion [2]. It is accepted that cancers are the result of the mutation accumulation in the genome. Breast cancer has been a significant by P53 role in the formation and progression, it is also known as the genome guard. The

results of different studies indicate that the p53 gene, one of the tumor suppressor genes, is involved at least in various cancers. P53 has been known “the guardian of the genome”, which aids in DNA stability by preventing genome mutation. (SNPs) in the p53 gene There are some different single nucleotide polymorphisms that the biological special effects of most of these polymorphisms have been planned and their clinical results are well known. According to the molecular epidemiological studies, p53 gene mutations are estimated to be identifiable in breast cancer cases. The codon 72 is one of the main SNPs throughp53 that has been frequently studied in breast cancer. This polymorphism should be 15% worldwide, > 30% in some developing countries, and 17.28% in manufacturing countries. Polymerase chain reaction (RFLP-PCR) will be restricted by several methods such as fragment length and polymorphism. Direct DNA sequencing and amplification-refractory mutation system-PCR (ARMS-PCR) have been used to examine the polymorphism of the p53 gene codon 72. [3].

Corresponding author:

Inam Joudah Radhi

Assist. Lecturer, College of pure Science, Department of Chemistry, University of Kerbala, Iraq, .
anamj@yahoo.com

The 4 exon codon 72 regions are lies by the most significant is rs1042522. This praline-rich region is

primarily responsible for the show of the apoptotic feature of the TP53 gene. It is a functional polymorphism which is found on exon 4 and foundations the replacement of praline with arginine residue at the regulatory portion of the TP53 gene, causing in altered instruction of apoptosis. Different studies have been established by TP53 codon polymorphism in the growth of endometriosis [4]. The p53 is a growing suppressor gene, governing the instruction of cell tumor and the regulation of the cell cycle, which is complicated in cell spreading like the progression of different procedures of cancer and brings apoptosis or hunks the cell cycle in according to DNA damage, which make it possible for cells to be damaged or returned before reinitiating of DNA replication [5]. In contrast, germline variants of the p53 gene are not regularly recognized, and many differences found about their importance in different malignant tumors. The p53 codon 72 gene are derived by two variants labelled from the replacement of a single nucleotide which is resulted in the attendance of proline or arginine in the protein product [6]. The improvement of cancer has been described by revisions the arginine homozygote in codon 72 because it is a danger feature, whereas others exposed a raised danger to persons with the proline homozygous genotype. The malignant transformation of some detectives, abnormalities, chromosomal and aberrations has been related toward p53 by ovarian endometriosis then can be associated with did not notice the different appearance of p53 in endometriosis [7]. The aim of this research has been determined by the frequency of p53 codon and 72 polymorphisms in PCOS in Karbala city women sterility and other symptoms of the disease.

Materials and Methods

The study was showed during the period from (2019) A cross-sectional study was designed; 70 persons, 40 patients and 30 healthies, aged from (19) to (40) years, the diagnosis was based on the clinical history, presentation long-established by UTS and numerous surveys of PCOS biomarker.

DNA extraction:

Two ml of blood sample collected in EDTA tube for

genomic DNA extraction used for molecular analysis. DNA extraction kit was purchased from Gene aid _Gsync DNA Extraction Kit.

P53 genotyping:

For each allele double autonomous polymerase chain reaction (PCR) tests were used. Genomic DNA improved by using PCR primers which notice p53 codon 72 in the proline form (178 bp. (5'GCCAGAGGCTGCTCCCC-3'; 5'CGTGCAAGTCACAGACTT-3') and arginine form (142 bp) (5' T C C C C C T T G C C G T C C C an A - 3' ;5' CTGGTGCAGGGGCCACGC-3') [8].

The PCR was obtained in a 20- μ L final volume PCR (premixTM) kit was used to amplify. It contained 2 μ l of extracted DNA (1 μ l of each primer and of 10 pmol/ μ l primers were mix in total volume of 20 μ l. Then the mixture was added to lyophilized PCR premix formula PCR amplification was carried out (cleaver, USA) thermal cycler using the following protocol: 5 min at 95°C, 30 s at 95°C, 30 s at 58°C for Arg72 and 60°C for Pro72, and 45 Second at 72°C for 35 cycles, with an additional 5 min at 72°C after the last cycle The PCR products were separated on a two percent TBE agarose gel and stained with ethidium bromide. A DNA ladder marker (BIONEER, South Korea) was used to determine the size of DNA fragment.

Result

(PCOS) Poly Cystic Ovarian Syndrome is the most public endocrine complaint of reproductive-age women. The analysis of PCOS is mostly founded on the resulting three components: TSH, FSH, and BMI. Prominently, the diagnostic criteria and complications related to Poly Cystic Ovaries Syndrome PCOS are age-dependent, showed in table-1.

The allelic frequencies in the patient group were Arg 47% and Pro 33%; however, the genotype (P<0.001) and allele (P=0.01) distribution showed significant differences between cases and controls ('P' values < 0.05). There was a significant increase of P/P

genotype frequency in Poly Cystic Ovaries infertility that they were compared with healthies. The allele frequency also followed similar trend indicating that 'P' allele might confer risk of developing PCOS and 'R' allele provides protection against the diseases, showed in table2

Table-1: age and hormones related pcos.

	CASES AVAR± SD NO(40)	CONTROL AVAR± SD NO(30)
Age	27.96±5.5	23.00±5.63
Tsh	2.02±1.01	3.90±1.04
Fsh	7.21±5.77	10.00±2.11
Bmi	26.91±5.16	21.9±4.53

Table-2: Distribution of TP53 Codon 72 Genotypes in PCOS Cases and Controls.

	Cases (n=40) n%	Control (n=35)n%	OR (95%CI)	p value
pro allele	33(41.25)	3(4.29)	1rf	
arg allele	47(58.75)	67(95.71)	15.6(4.5-54.1)	<0.001
p/p	8(20)	0	1rf	
p/r	17(42.5)	3(8.57)	1.41(0.126-15.78)	0.77
r/r	15(37.5)	32(91.43)	17.06(1.95-149.1)	0.01

Discussion

P53 which are participated in the instruction of glucose are: fatty acid, amino acid (glutaminolysis) and influences mitochondrial integrity and oxidative phosphorylation, insulin sensitivity, antioxidant response, autophagy and mammalian Target of Rapamycin [10]. The unstable production and apoptosis of granulosa cells could result to unusual folliculogenesis [9].

Additionally, the use of oocytes with lacking tumor suppressor genes role may be prematurely eliminated, resultant in initial weakening of egg replacement and, as

a result, principal ovarian deficiency [10].

The previous studies say that P/P genotype is related to an enlarged appearance of genes adaptable cell cycle apprehension, which consequently results with the stop of follicle growth [11].

Other Indian study ends with similar conclusion that P/P genotype of the TP53P/R polymorphism was related with developed danger of PCOS, associated to the P/P genotype, in addition to the occurrence of the allele which was expressively higher in Poly Cystic Ovaries Syndrome (PCOS) female than in healthy controls P/P

variant of R/P polymorphism of TP53 is higher in PCOS patients [12]. These studies agree with our results but we still need extra studies and research in this field, because there are no local or even regional studies [13].

Ethical Clearance: Ethics committee refer that there is no plagiarism and there is no mistakes or wrong results or plagiarism in this work.

Conflict of Interest : The authors declare that there is no conflict of interest.

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