**وه‌زاره‌تی خوێندنی باڵا و تۆێژینه‌وه‌ی زانستی**

**Ministry of Higher Education &**

**Scientific Research**

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| **پرۆپۆزەلى توێژینه‌وه‌ بۆ به‌ده‌ستهێنانی بروانامه‌ی دکتۆرا PhD Research Proposal** | | |
| **1. Title of PhD research proposal**  **Role of gene polymorphisms of some cytokines and androgen receptor among infertile men in Erbil province** | | |
| **زانیاری گشتی 2. General information** | | |
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| **3. Summary (Abstract) of PhD research proposal**  Infertility may be defined as the infertility of a couple to conceive after a period of one year of intercourse without use of contraceptive. It is a worldwide problem that characterized by multifactorial etiology. Genetic, physiology, infectious, environmental and immunological factors reported as for as predisposal or even causative agents of this condition. The current study has the aim of evaluation of some cytokines level and gene polymorphism among categories of infertile and fertile males. One of the well-documented cytokines suspected as a hazard to male fertility is tumor necrosis factor-a (TNFa). Genetic factors such as single-nucleotide polymorphisms (SNPs) in the TNF gene cluster impact TNFa levels. In summary, the results several studies indicate that gene poly morphism of some cytokines such as TNF-α, IL-17 and the IL-1 is a molecular marker for male Infertility.  According to physiological parameters, Determination of enzymatic antioxidant such as catalase, glutathione peroxidase,superoxide dismutase , vitamin C and E , Routine Seminal fluid analysis was done for each sample including volume, sperm count, activity and morphology. Then biochemical tests for sperm and seminal plasma were assayed in some types of infertile and fertile men | | |
| **4. Introduction**  The World Health Organization has determined Infertility a major problem affecting approximately married couples; it is defined as the inability to conceive after 1 year of unprotected sexual intercourse. Roughly, 30% of cases involve males, 35% involve female factor and the remaining represent unexplained or idiopathic infertility.  Male infertility's not an entity but reflects a variety of different pathological conditions, and the cause cannot be determined in most men investigated for infertility, the etiology of male infertility is obviously a multifactorial one, could be nutritional, physiological, pathological, accidental or even immunological.  A high percentage of infertile men fail to impregnate their female counterpart because of lack of sperm (azoospermia) or low sperm count (oligozoospermia); infertility may also be due to abnormal sperm morphology (tetratozoospermia) and insufficient sperm motility (asthenozoospermia). It has been estimated that infertility affects 13% to 15% of couples worldwide, with  both male and female factors being present in many of these cases and in roughly half of these cases the defect can be traced to the males. Cytokines important intra-cellular communicators are involved in numerous physiological and pathological processes, which include mediation of inflammatory responses, reproductive physiology and regulation of gonadal steroid production and release. Human sperm contains a wide spectrum of cytokines such as tumor necrosis factor alpha (TNF-á), interleukin 18 (IL-18), IL-6, IL-17 and IL-1. For example: IL-1 is a regulatory cytokine that plays an important role in the maintenance of the immune environment of the testis, regulation of junction dynamics and cell differentiation during spermatogenesis. Members of the IL-1 family are pleiotropic cytokines that are involved in inflammation, immunoregulation and other homeostatic functions in the body. IL1a, IL-1b, and the IL-1 receptor antagonistic molecule (IL-1 Ra) are expressed in the testis under normal homeostasis and they further increase upon infection/inflammation. There is clear evidence indicating the effects of cytokines on spermatozoal functions. Various cytokines are considered as prime candidates for mediating infertility progression. During genital infection cytokines and various soluble receptors of immunoregulatory cytokines are expressed distinctly in seminal plasma. These factors also may be involved in the regulation of sperm cell functions and thus may affect male fertility. Measuring the level of cytokines, both in seminal plasma and serum, does not only expand the diagnostic options, but also, through the growing knowledge of immune processes, can give rise to new therapeutic methods of improving the quality of semen and increasing the chance to reproduce.  Androgens are critical steroid hormones that determine the expression of the male phenotype. Their actions are mediated by a single androgen receptor (AR) which, upon ligand binding, translocates to the nucleus to regulate the expression of androgen-responsive genes. AR mutations that do not lead to complete abrogation of its activity can cause a wide spectrum of milder androgen insensitivity syndromes, from ambiguous genitalia in newborn infants to `idiopathic' male infertility. Mcissense amino-acid substitutions in the ligand-binding domain of the AR result in infccertility through a novel mechanism that involves defective protein-protein interactions between receptor domains and coactivator proteins. Independent of missense mutations, studies indicate that increases in length of a trinucleotide repeat (CAG) tract, encoding a polyglutamine stretch in the transactivation domain of the AR, are associated with increased risk of defective spermatogenesis and under masculinization. | | |
| **5. Research objectives**   1. To determine the association between some cytokines such as (IL-2, IL-10, TNF-α and IFN-γ) and their gene polymorphisms at some positions such as (IL-2-330, IL-2+166, IL10-1082, IL-10-819, IL10-592, TNF-308, TNF-238 and IFNγ+874) with the sperm count, motility and morphology. 2. To investigate the association between AR gene polymorphism with male infertility especially azoospermia patients. | | |
| **6. Methodology and data collection**  Patients married for a minimum of one year, having unprotected intercourse will be considered for the present study. Seminal analyses of (number of participants) infertile males will carry out after three/four days of sexual abstinence to ascertain their infertility status. The patients will categorize in sub-groups as per WHO 1999 criteria. Asthenozoospermic infertile men, Oligozoospermia infertile men and non-obstructive azoospermic infertile men. The control group consists of healthy fertile males who have at least one child and no history of chronic illness.  1-The seminal fluid analyses tests will be done to confirm the diagnosis of infertility, appearance, volume, PH, viscosity and liquefaction of semen, sperm agglutination, morphology, motility and sperm count.    2-Serum or seminal plasma level of cytokines under study will be measured by ELISA.  3-DNA will be extracted from blood or sperm, ampliﬁcation of the different cytokines gene polymorphisms in different positions were accomplished by using of polymerase chain reaction (PCR) method.  4- DNA will be extracted from blood or sperm, ampliﬁcation of the AR gene polymorphisms are accomplished by using of polymerase chain reaction (PCR) method. | | |
| **7. Scope and limit to the research**  Little attention has been paid to the role of cytokines gene polymorphisms and estrogen receptor (ER) gene mutations in male infertility; in this study we will cover these parts. | | |
| **8. Duration and timeline**  **Phase1**: 12 months for sample collection  **Phase2**: 12 months for tests and experiment  **Phase3**: 12 months for data analysis, writing up, and publications | | |
| **9. Conclusions**  We expected significantly association of cytokines gene polymorphisms with abnormal seminograms. Also positive correlation of non-obstructive azoospermia with AR gene polymorphism. | | |
| 10. References سەرچاوەکان  Dousset, B., and F. Hussen et, 1997. Les cytokines dans le sperme humain: une nouvelle voire d'approche de la fertilité masculiné, la Press Med, 26:24-29.  World Health Organization. WHO Manual for the Standardized Investigation and Diagnosis of the Infertile Couple. 2000, Cambridge University Press: Cambridge.  WHO. 2010. Laboratory Manual for the examination of human semen and semen cervical mucus nteraction. WHO. Cambridge Unversity Press, 3rd Ed.  Celinska A, Fracki S, Sangidorj D, Barcz E (2006) : Role of inflammatory cytokines in male infertility Ginekol Pol May;77(5):404-11  Hilaii, H.A.A. and AL-Hasani, H.J. (2012). Role of HLA–DQ Genotyping with Some Immunological Markers in Susceptibility for Infertile Males in Najaf Province. Int. J. Curr. Microbiol. App. Sci., 5(3): 277-287.  Said, M.T. 2008. Emotional stress and male infertility. Ind. J. Med. Research, 128: 228–230.  Huleihel M, Lunenfeld E, Levy A. et al. (1996) :Distinct expression levels of cytokines and soluble cytokine receptors in seminal plasma of fertile and infertile men. Ferti. Steril;66:135–39.  Brackett, N.L., Lynne, C.M., Aballa, T.C., Ferrell, S.M. 2000. Sperm motility from the vas deferens of spinal cord injured men is higher than from the ejaculate. J. Urol., 164: 712–715.  W.H.O. 2010.WHO laboratory manual for the Examination and processing of human semen. Switzerland : World Health Organization .  Yong, E., Loy, C. & Sim, K. J. H. R. U. 2003. Androgen receptor gene and male infertility. 9, 1-7. | | |
| **11. General notes:** هەر زانیارییەکی گشتی دیکە کە سەرپەرشتیار بە گرنگی بزانێت | | |
| **12.**  **په‌سه‌ندكردنی پرۆپۆزەل له‌ لایه‌ن لیژنه‌ی زانستی به‌ش**  ژماره‌ی كۆنووسی كۆبوونه‌وه‌:  رێكه‌وتی كۆبوونه‌وه‌:  بریار: په‌سه‌ند كرا په‌سه‌ند نه‌كرا    ناوی سیانی و واژووی لیژنه‌ی زانستی به‌ش  واژوو:  ناوى سه‌رۆكی لیژنەى‌ زانستی به‌ش مۆری به‌ش  واژوو:  ناوى سه‌رۆكی به‌ش: | | |
| **13.**  **په‌سه‌ندكردنی پرۆپۆزەل له‌ لایه‌ن ئه‌نجومه‌نی كۆلێژ/فاکەڵتى**  ژماره‌ی كۆنوسی كۆبوونه‌وه‌:  رێكه‌وتی كۆبوونه‌وه‌:  بریار: په‌سه‌ند كرا په‌سه‌ند نه‌كرا  واژوو:  ناو راگری كۆلێژ: مۆری كۆلێژ | | |

**تێبینی:** تكایه‌ فۆرمه‌كه‌ ته‌نها به‌ یه‌ك زمان (زمانی توێژینه‌وه‌) پڕ بكرێته‌وه‌.