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# Measurement of Inhibin-A, Activin-A, Relaxin and Progesterone Hormones in patients with missed miscarriage

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**ABSTRACT**
This study aimed to show the effects of Inhibin–A , Activin-A , Relaxin and Progesterone Hormones in the incidence of missed abortion and normal pregnancy cases in women at first trimester of pregnancy in Thi-Qar Province. This study had been performed in the period between ( November 2023 to September 2024) , included (100) blood random samples of missed abortion and normal pregnancy women, all diagnoses by the Ultrasound scan and other ways as clinical diagnoses and the histery of pregnant , Their range ages between(18-40) years. That samples were obtained from attending women to Bint AL-Huda Hospital and some private medical clinics. The selective samples were divided into two groups , the first group represented 50 blood samples from aborted women, while the second group included 50 blood samples from normal pregnancy women as a control group. The blood samples had taken during the period between (1-3)months of pregnancy.The results revealed a significant decrease in the concentration of Inhibin-A , Activin-A and Progesterone hormones ($P \leq 0.05$) in the group of aborted women when compared with the control group.Also the results had showed a high a significant in Relaxin concentration in abortion group ($P \leq 0.05$) when compared with the control group.
This study confirmed an apparent relationship between the decrease concentrations of inhibin-A , activin-A and Progesterone and incidence of the abortion cases , which emphasizes the important role of their hormones in pregnancy continue

**Keywords:** Progesterone Hormones, Relaxin, abortion, missed miscarriage

## INTRODUCTION

Missed miscarriage: A missed miscarriage, also known as a missed abortion, occurs when a fetus is no longer alive, but the body does not recognize the pregnancy loss or expel the pregnancy tissue. As a result, the placenta may continue to release hormones, so you may continue to experience signs of pregnancy[1][2][3]. A missed miscarriage is often detected during the first-trimester exam, usually between 11 and 14 weeks[4]. After a heartbeat has been detected at the eight-week scan, the chance of a miscarriage drops to only 2%. The chance falls to below 1% after 10 weeks [5].

A missed miscarriage is often known as a silent miscarriage because people often don't exhibit the most common miscarriage symptoms, such as: Vaginal bleeding, Heavy cramping and Expulsion of fetal tissue[6][7] .However, some may notice that their pregnancy symptoms, like breast tenderness, nausea, or fatigue, may disappear.

Some women may also have brownish or red vaginal discharge. Approximately 1-5% of all pregnancies will result in a missed miscarriage[8] .Missed miscarriage was a special type of spontaneous abortion that the embryo or fetus has already died but remained in the uterus for days or weeks and with a closed cervical ostium [9]. Patients might present with or without subtle clinical symptoms such as vaginal bleeding or abdominal pain. Missed abortion, (its occurring in approximately 8–20% of clinically confirmed intrauterine pregnancies) [10][11], was often confirmed

using ultrasonography. The causes of missed abortion are complex and diverse, and recent studies have shown that it is associated with genetic factors, immune factors, endocrine factors, viruses, bacterial, parasitic infections, and so on [11].Missed abortion is undoubtedly a huge physical and psychological setback for women with fertility requirements[12].
Therefore, early identification of women at high risk of missed abortion is pivotal, which might aid in providing possible theoretical basis for implementing clinical measures to prevent missed abortion[13][14].

## SUBJECTS ,MATERIALS AND METHODS

### Design of Study

A total of 100 Missed Miscarriage and Normal pregnancy women with an age range from 18 to 40 years were involved in this study after strict application of the exclusion criteria. Data were collected through direct interviews with the patients . Written consent was obtained from each patient who was participating in this study to fulfill the international research ethical criteria.This study was conducted at Bint AL-Huda Hospital in Thi-Qar, especially, in the Women's Emergency and Biochemistry Laboratory in the College of Science (University of Thi-Qar) in the period between 6/11/2023 to 16/8/2024.The controls and patients were divided into (50) blood samples are collected from normal pregnant as a control group and (50)blood samples from patients with Missed Miscarriage , during the period between (1-3)months of pregnancy.The sample size was determined according to the equation Stephen Thompson [15]. Serum Inhibin-A, Activin-A , Relaxin and Progesterone Hormones were measured using ELISA Kits (Demeditec Diagnostics GmbH, Germany).About (5 mL) of venous blood was collected from pregnant women with missed miscarriage and normal pregnant, were transferred to plain tube and allowed to clot at room temperature to get serum by putting it in empty disposable tubes and centrifuged to separate it at 3000 rotor per minute (rpm) for 10 min, the serum samples were separated and stored at (-20˚C) for later measurement biochemical parameters, unless used immediately. Serum Inhibin-A, Activin-A and Relaxin hormones were estimated by enzyme linked immunoassay method by ELISA Reader, using kit supplied by BT LAB, China.The concentrations of progesterone in the collected sample were determined by using a competitive ELISA technique according to the procedure provided with the kit. The kit has an analytic sensitivity of 0.03 ng/ml. The absorbance of calibrators and specimen was determined by using automated ELISA reader system by using a reference wavelength of 630 nm.

### Statistical Analysis

The statistical analysis was done using spss v 26 the results were expressed as mean ± standard deviation (mean ± SD). It was used t test to compare study groups. P-values (P≤0.05) were considered statistically significant.

## RESULTS AND DISCUSSION :

### Inhibin and Activin

In the table a two tailed t-test samples were performed on the studied groups .
For the inhibin is produced by the placenta. In a miscarriage, the placenta may not develop or function properly, leading to lower levels of Inhibin.[16]. In the table the inhibin shows low concentrations in women have miscarriage (**10.788∓6.027**) compared to women with normal pregnancy (**14.366∓7.228**), this significant difference with **p = 0.008**, indicating that Low levels of activin may be related to intrauterine growth restriction, where the fetus does not grow as expected in the uterus. This condition can lead to various complications, both during pregnancy and after childbirth.[17][18].A study of Yue, C. Y shows that Low levels of inhibin have been marked in the second trimester of pregnancy[19]. Its decleared that Inhibin levels typically rise during normal pregnancy, and lowered abnormally levels related to an increased risk of miscarriage.[18][20]

**Table (1)** Serum level of Inhibin-A for control and patient groups

| | Study Groups | | t. test | P. Value |
|---|---|---|---|---|
| | Patient NO.=50 | Control NO.=50 | | |
| INHIBIN-A | Mean ∓SD | | | |
| | 10.788∓6.027 | 14.366∓7.228 | -2.689 | 0.008 |

For the Activin it shows a low concentration in women miscarriage (**1.310∓0.349**), and a higher concentrations of activin (**2.285∓1.651**) for the control group thus, a tatistical significant difference appeared in this relation, t(53.38) = **--4.086**, p = <0.001. since activin is an important hormone in pregnancy, influencing placental function, fetal development, and the maternal immune response.[21][22] The low concentrations of activin could be an important insights into the health of the pregnancy and the risk of complications leading to may be a conditions like preterm labor or other pregnancy complications.

This is the same as the study of Ahmed, N. H. [23], which indicates that women who suffer a miscarriage often have lower levels of activin, as activin is produced by the placenta, so its levels may reflect placental function and ability. So, In cases where the pregnancy does not progress normally, such as in a miscarriage, the placenta may not function properly, resulting in decreased activin secretion.[24]

Dynamic, as it is up- and downregulated during the process of decidualization [25, 26], and studies in women with nonfunctional ovaries have suggested a fetoplacental origin for activin-A [24]. Serum levels of activin A are higher in pregnant than in nonpregnant women and increase throughout pregnancy until about 28 weeks' gestation . [25, 27] However, in early pregnancy, the expression of activins by the cytotrophoblast is low, which suggests that trophoblast invasion is induced by the maternally derived activins [28].

The source of maternally derived activin A in pregnancy is primarily from newly decidualized cells, and this promotes the decidualization of neighboring cells and thus facilitates the spread of decidualization throughout the endometrium[29] Normal concentrations of serum activin A in pregnancy were reported to rise 69-fold (wide spectrumof values) throughout pregnancy [30].

**Table (2)** Serum level of Activin-A for control and patient groups

| | **Study Groups** | | **t. test** | **P. Value** |
|---|---|---|---|---|
| **ACTIVIN-A** | **Patient NO.=50** | **Control NO.=50** | | |
| | **Mean ∓SD** | | | |
| | 1.310∓0.349 | 2.285∓1.651 | **-4.086** | **0.000** |

**Progesterone**

From the obtained results as shown in table Progesterone has no significance difference among the studied groups where p = 0.218

Progesterone is necessary to maintain the lining of the uterus (endometrium) where the fertilized egg is located. It prevents shedding of the uterine lining, which is crucial for the continuation of pregnancy.[31]

Progesterone also helps prevent uterine contractions that may lead to expulsion of the embryo or fetus.[32]

**Table (3)** Serum level of Progesterone for control and patient groups

| | **Study Groups** | | **t. test** | **P. Value** |
|---|---|---|---|---|
| **PROG** | **Patient NO.=50** | **Control NO.=50** | | |
| | **Mean ∓SD** | | | |
| | 61.977∓17.499 | 71.089∓48.873 | **-1.241** | **0.218** |

**Relaxin**

In the obtained results show that relaxin levels altered during miscarriage (364.872 ± 157.057), compared to normal state of pregnant women (272.473 ± 89.532). this difference is statistically significant with p = 0.002, that leads us to considare relaxin as a potential biomarker in miscarriage in this study[33] .It's well known that Relaxin defined as a hormone primarily associated with preparing the body for childbirth, which plays an important role during pregnancy.and since that most studies focus on its role in the later stages of pregnancy, especially in relation to labor and delivery. Therefore, while there may be changes in relaxin levels during miscarriage.[34-39]

**Table (4)** Serum level of Relaxin for control and patient groups

| | Study Groups | | t. test | P. Value |
|---|---|---|---|---|
| RELAX | Patient NO.=50 | Control NO.=50 | | |
| | Mean ∓SD | | | |
| | 364.872∓157.057 | 272.473∓89.532 | **-3.614** | **0.002** |

**Fig. (1)** Serum level of Inhibin–A , Activin-A , Progesterone and Relaxin Hormones for control and patient groups

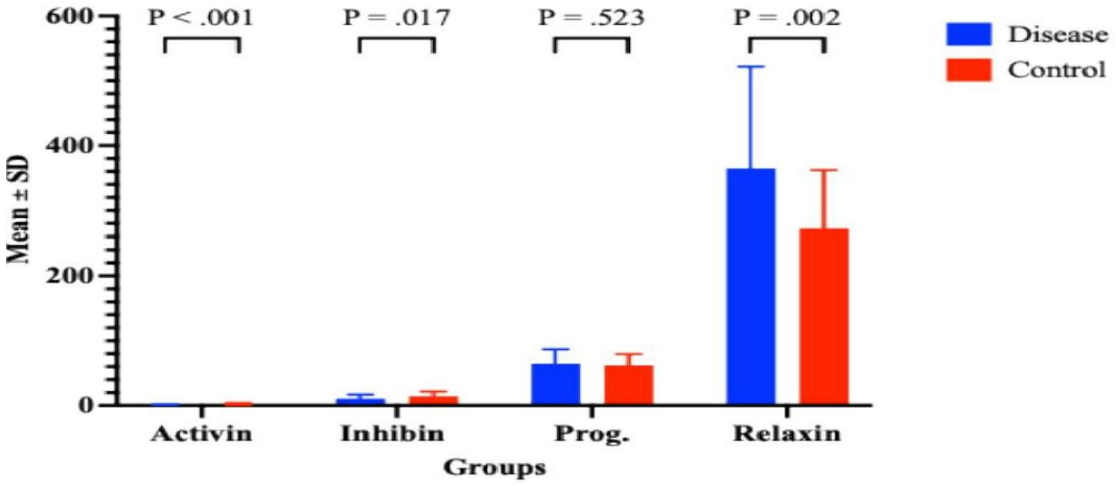


**Table (5)** Serum level of all hormones for control and patient groups

| Parameters | Groups | N | Minimum | Maximum | Mean ± Std. | P – value |
|---|---|---|---|---|---|---|
| Activin | Disease | 50 | 0.7 | 3.29 | 1.31 ± 0.35 | <0.001 |
| | Control | 50 | 0.87 | 9.05 | 2.29 ± 1.65 | |
| Inhibin | Disease | 50 | 0.46 | 29.12 | 10.79 ± 6.03 | 0.017 |
| | Control | 50 | 2.16 | 34.31 | 14.37 ± 7.23 | |
| Prog | Disease | 50 | 33.96 | 132.16 | 61.977 ± 17.49 | 0.218 |
| | Control | 50 | 33.44 | 110.62 | 71.089 ± 48.873 | |
| Relaxin | Disease | 50 | 155.68 | 871.22 | 364.87 ± 157.06 | 0.002 |
| | Control | 50 | 181.44 | 552.82 | 272.47 ± 89.53 | |

**REFERENCES**


1. O'Halloran, T. V. (2017). Zinc sparks induce physiochemical changes in the egg zona pellucida that prevent polyspermy. *Integrative Biology*, *9*(2), 135-144.
2. Hirohashi, N., & Yanagimachi, R. (2018). Sperm acrosome reaction: its site and role in fertilization. *Biology of reproduction*, *99*(1), 127-133.
3. Gupta, S. K. (2021). Human zona pellucida glycoproteins: binding characteristics with human spermatozoa and induction of acrosome reaction. *Frontiers in cell and developmental biology*, *9*, 619868.
4. Rossant, J., & Tam, P. P. (2022). Early human embryonic development: Blastocyst formation to gastrulation. *Developmental cell*, *57*(2), 152-165.
5. Kagawa, H., Javali, A., Khoei, H. H., Sommer, T. M., Sestini, G., Novatchkova, M., ... & Rivron, N. (2022). Human blastoids model blastocyst development and implantation. *Nature*, *601*(7894), 600-605.
6. Al-Salih, R. M. (2023). Role of Folic Acid, Vitamin D, and Progesterone in Pregnancy and Threatened Abortion. *University of Thi-Qar Journal of Science*, *10*(2).Ali, A. T., Mahdi, D. S., & Awad, A. H. (2019).
7. Moseson, H., Herold, S., Filippa, S., Barr-Walker, J., Baum, S. E., & Gerdts, C. (2020). Self-managed abortion: a systematic scoping review. *Best practice & research Clinical obstetrics & gynaecology*, *63*, 87-110.
8. Tan, C. M. J., & Lewandowski, A. J. (2020). The transitional heart: from early embryonic and fetal development to neonatal life. *Fetal diagnosis and therapy*, *47*(5), 373-386.
9. Kirkman, M., Rowe, H., Hardiman, A., Mallett, S., & Rosenthal, D. (2009). Reasons women give for abortion: a review of the literature. *Archives of women's mental health*, *12*, 365-378.
10. Lecanuet, J. P., Fifer, W. P., Krasnegor, N. A., & Smotherman, W. P. (Eds.). (2013). *Fetal development: A psychobiological perspective*. Psychology Press, 5-9
11. Hassun, A. F., & Jarulla, B. A. (2021). Abortion Related of Infectious Agents in Women in Thi-Qar Province. *University of Thi-Qar Journal of Science*, *8*(1), 118-124.Muter, J., Lynch, V. J., McCoy, R. C., & Brosens, J. J. (2023). Human embryo implantation. *Development*, *150*(10), dev201507.
12. Zhai, J., Guo, J., Wan, H., Qi, L., Liu, L., Xiao, Z., ... & Wang, H. (2022). Primate gastrulation and early organogenesis at single-cell resolution. *Nature*, *612*(7941), 732-738.
13. Muttukrishna, S., Fowler, P. A., Groome, N. P., Mitchell, G. G., Robertson, W. R., & Knight, P. G. (1994). Endocrinology: Serum concentrations of dimeric inhibin during the spontaneous human menstrual cycle and after treatment with exogenous gonadotrophin. *Human Reproduction*, *9*(9), 1634-1642.
14. Daponte, A., Deligeoroglou, E., Garas, A., Pournaras, S., Hadjichristodoulou, C., & Messinis, I. E. (2013). Activin A and follistatin as biomarkers for ectopic pregnancy and missed abortion. *Disease markers*, *35*(5), 497-503.
15. MARGIANA, Ria, et al. Functions and therapeutic interventions of non-coding RNAs associated with TLR signaling pathway in atherosclerosis. Cellular Signalling, 2022, 100: 110471.
16. ARIF, Anam, et al. The functions and molecular mechanisms of Tribbles homolog 3 (TRIB3) implicated in the pathophysiology of cancer. International Immunopharmacology, 2023, 114: 109581.
17. LEI, Zimeng, et al. Detection of abemaciclib, an anti-breast cancer agent, using a new electrochemical DNA biosensor. Frontiers in Chemistry, 2022, 10: 980162.
18. LAFTA, Holya A., et al. Tumor-Associated Macrophages (TAMs) in Cancer Resistance; Modulation by Natural Products. Current topics in medicinal chemistry, 2023.
19. HJAZI, Ahmed, et al. The pathological role of CXC chemokine receptor type 4 (CXCR4) in colorectal cancer (CRC) progression; special focus on molecular mechanisms and possible therapeutics. Pathology-Research and Practice, 2023, 154616.
20. HJAZI, Ahmed, et al. Unraveling the Impact of 27-Hydroxycholesterol in Autoimmune Diseases: Exploring Promising Therapeutic Approaches. Pathology-Research and Practice, 2023, 154737.
21. GUPTA, Jitendra, et al. Double-edged sword role of miRNA-633 and miRNA-181 in human cancers. Pathology-Research and Practice, 2023, 154701.
22. SANE, Shahryar, et al. Investigating the effect of pregabalin on postoperative pain in non-emergency craniotomy. Clinical Neurology and Neurosurgery, 2023, 226: 107599.
23. Farhan, Shireen Hamid, et al. "Exosomal non-coding RNA derived from mesenchymal stem cells (MSCs) in autoimmune diseases progression and therapy; an updated review." Cell Biochemistry and Biophysics 82.4 (2024): 3091-3108.

24. Qasim, Maytham T., and Zainab I. Mohammed. "The Association of Helicobacter pylori Infection and Virulence Factors in Gastric Cancer in Thi-Qar, Iraq." Asian Pacific Journal of Cancer Biology 9.4 (2024): 541-545.
25. Zamanian, Mohammad Yasin, et al. "Effects of Resveratrol on Nonmelanoma Skin Cancer (NMSC): A Comprehensive Review." Food Science & Nutrition (2024).
26. Hsu, Chou-Yi, et al. "MicroRNA-enriched exosome as dazzling dancer between cancer and immune cells." Journal of Physiology and Biochemistry (2024): 1-19.
27. Lv, Jing, et al. "A comprehensive immunobiology review of IBD: With a specific glance to Th22 lymphocytes development, biology, function, and role in IBD." International Immunopharmacology 137 (2024): 112486.
28. Ul Hassan Shah, Zameer, et al. "Development of antihyperlipidemic drug loaded β-CD-based microparticulate carrier systems: tuning and optimization." Polymer-Plastics Technology and Materials 63.11 (2024): 1438-1463.
29. Hsu, Chou-Yi, et al. "A comprehensive insight into the contribution of epigenetics in male infertility; focusing on immunological modifications." Journal of Reproductive Immunology (2024): 104274.
30. Patel, Ayyub Ali, et al. "Application of mesenchymal stem cells derived from the umbilical cord or Wharton's jelly and their extracellular vesicles in the treatment of various diseases." Tissue and Cell (2024): 102415.
31. Zamanian, Mohammad Yasin, et al. "Chemopreventive and Anticancer Role of Resveratrol against non-melanoma skin cancer (NMSC): Focusing on cellular and molecular mechanisms and biochemistry." Authorea Preprints (2024).
32. Fakhrioliaei, Azadeh, et al. "Potential Role of Nrf2, HER2, and ALDH in Cancer Stem Cells: A Narrative Review." The Journal of Membrane Biology 257.1 (2024): 3-16.
33. Ramaiah, Pushpamala, et al. "Dietary polyphenols and the risk of metabolic syndrome: a systematic review and meta-analysis." BMC Endocrine Disorders 24.1 (2024): 26.
34. Al-Hawary, Sulieman Ibraheem Shelash, et al. "Tumor-derived lncRNAs; Behind-the-scenes mediators that modulate the immune system and play a role in cancer pathogenesis." Pathology-Research and Practice (2024): 155123.
35. Qasim, M. T., M. N. Fenjan, and H. A. Thijail. "Molecular identification of cystoisospora belli in patients infected with the virus human immunodeficiency." International Journal of Drug Delivery Technology 12.2 (2022): 701-704.
36. Qasim, M. T., et al. "Ovine Pasteurellosis Vaccine: Assessment of the Protective Antibody Titer and Recognition of the Prevailing Serotypes." Archives of Razi Institute 77.3 (2022): 1207.
37. Qasim, Maytham T., and Zainab I. Mohammed. "Investigating Treatment Response and Viral Immunity in Early Rheumatoid Arthritis via Immune Response Profiling." Journal of Rare Cardiovascular Diseases 4.8 (2024): 166-173.
38. Stewart, D. R., Overstreet, J. W., Celniker, A. C., Hess, D. L., Cragun, J. R., Boyers, S. P., & Lasley, B. L. (1993). The relationship between hCG and relaxin secretion in normal pregnancies vs peri-implantation spontaneous abortions 1. *Clinical endocrinology*, *38*(4), 379-385.
39. Sarb, J. (2018). *The role of matrix metalloproteinases and relaxin hormone in trophoblast-endometrium interactions during implantation* (Doctoral dissertation, University of Sheffield).