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Bahaa Aldin A *

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Research Article

# The Association between changes in C reactive protein and Pregnancy rate in IVF/ICSI.

**Bahaa Aldin A*, Elyan A., Sadoon A. and Fathy H**

Department of ob/gyn Ain Shams University Maternity Hospital

***Corresponding Author:** Bahaa Aldin A, Assistant professor of obs/gyn, Ain Shams University,

**Received Date: March 05, 2021; Accepted Date: March 22, 2021; Published Date: March 26, 2021**

**Citation:** Hashaad A., Raafat T., Kamal A., Ahmed Essam Eldin Mansour (2021) The Association between changes in C reactive protein and pregnancy rate in IVF/ICSI. *J Women Health Care Issues*, 4(2); Doi:**10.31579/2642-9756/047**



## Abstract

**Objectives:** The aim of this study is to evaluate the association between changes in C reactive protein and pregnancy rate in IVF/ICSI.

**Patients and methods:** A prospective cohort study that was conducted in Ain Shams University Maternity Hospital, Infertility Clinic during period time from September 2018 to September 2020. A total of 100 infertile women who were candidate for IVF/ICSI, over a period of 2 years, beginning in September 2018 were recruited to the study after they met the inclusion and exclusion criteria. The participants gave written informed consent and approval for the ethical aspects of the study.

**Results:** This study showed no statistically significant differences according to clinical pregnancy regarding the sociodemographic data as BMI, duration of infertility and type of infertility and regarding Oocyte retrieval and embryo transfer, also no significant differences according to clinical pregnancy regarding CRP at baseline and at Oocyte pickup as well as CRP change at oocyte pickup. But age was significantly lower in cases with clinical pregnancy and CRP at embryo transfer and CRP change at embryo transfer were significantly higher in cases with positive pregnancy.

**Conclusion:** Finally, we concluded that patients whose CRP level decreased on transfer day, had lower chance of pregnancy, whereas patients whose CRP level elevated on embryo transfer day had high chance of pregnancy

**Key words:** reactive protein; pregnancy, ivf/icsi

## Introduction

C-reactive protein (CRP) is a marker in inflammatory reactions. Its level is changed with gender and age. Researches validated that women at the time of parturition have elevated levels of CRP compared to those who are not. (Wood et al., 2000). There is an association between the rise of this 110,000-120,000 KD protein and occurrence of atherothrombosis, pre-term delivery, low weight of the fetus, and preeclampsia. (Garcia et al., 2007). This protein enforces the innate immunity and protection against tissue damage through increase in phagocytosis and removing cells and damaged, dead or dying organisms (Coussons et al., 2007). This protein as a sensitive marker in inflammatory processes rises following hormonal stimulation. (De Maat et al., 2007). CRP does not have diurnal alterations, but administration of exogenous estrogen increases its level. (Störk et al., 2008). In vitro fertilization (IVF) steps occur over about a two-week interval of time, which is called an IVF cycle. The first pregnancy after thE fertilization of a human egg in vitro and the first birth from an in vitro fertilized embryo were reported in 1976 and 1978. (Ormond et al., 2017). Controlled hyper-ovulation of the ovary, and specially puncture of the ovaries in in-vitro fertilization (IVF) may affect the successful rate of IVF/ICSI, implantation, and pregnancy. Also, administration of human chorionic gonadotropin (HCG), regardless of the response rate of the ovaries, causes activation of endothelial cells and neutrophils. (Orvieto et al., 2004).

## PATIENTS AND METHODS

It is a prospective cohort study that was conducted in Ain Shams University Maternity Hospital, Infertility Clinic and assisted reproductive unit during period time from September 2018 to September 2020. A total of 100 infertile women who were candidate for IVF/ICSI, over a period of 2 years, beginning in September 2018, were recruited to the study.

The participants gave written informed consent and approval for the ethical aspects of the study.

The participants included in the study met the following criteria:

### Inclusion Criteria:

1. Infertile women between 20 to 35 years.

2. Women with negative CRP before starting IVF/ICSI protocol.

3. Using standard long GnRH agonist protocol

4. Serum estradiol on the day of HCG injection between 1000: 3000 (pg/mL).

5. BMI: 18-29 kg/m2

**Exclusion Criteria:**

1. Fertile women < 20 and > 35 years

2. Severe male factor

3. Serum estradiol on the day of HCG injection < 1000 or > 3000 (pg/mL).

4. Factors that increase CRP like: PID, hydrosalpinx, endometriosis, infection. Procedure: Women who were selected eligible for the study underwent ovulation stimulation cycle by standard long protocol and written consent was taken from them.

**Controlled ovarian stimulation (COS):**

(COS) is the first step with the purpose of inducing maturation ofmultiple oocytes, and hence maximizing the chance of achieving successful pregnancy (Zhang et al., 2020). GnRH agonist (long) protocol:

Short-acting GnRH agonist long protocol, GnRH agonist [Triptorelin Acetate) (DECAPEPTYL ®) 0.1 mg by subcutaneous injection was administered daily starting from the mid-luteal phase of the menstrual cycle, lasting for 10–14 days until the pituitary down-regulation was confirmed, pituitary gonadotropin production was inhibited, thereby maximizing control of the cycle. Pituitary suppression of LH secretion was important to prevent a surge of endogenous LH prior to full maturation of the ovarian follicles. Then the ovarian stimulation with gonadotropin (Gn) commenced (ESHRE, 2019). Down-regulation was verified by serum estradiol measurement less than 50 pg./ml and LH less than 5 IU/ml. If the GnRH agonist was initiated in the luteal phase of the cycle prior to the IVF cycle, the initial stimulatory effect of the agonist was blocked by the endogenous secretion of progesterone and estradiol (ESHRE, 2019). One regimen uses two weeks of oral contraceptives with the GnRH agonist initiated after one week and used concurrently for one week, then the GnRH agonist is continued alone for another week (Adel, 2020; ESHRE, 2019). When stimulation begins, hMG (or FSH, or both) was administered in a dose of 225 to 300 IU/day subcutaneously to stimulate follicular growth, with the GnRH agonist being continued at a lower dose to prevent a premature surge in luteinizing hormone (LH) secretion. (Huang et al., 2019). The hMG dose was subsequently adjusted according to follicular growth (as determined by transvaginal ultrasonography) and target follicle size is >17mm two follicle or more and serum estradiol concentrations > 600Pg/ml (an indicator of granulosa cell proliferation), then oocyte retrieval by TVS guided after 34-36 hours from HCG injection. After that invitro fertilization of oocyte with prepared sperm and embryo transfer after 2-3 days at 4-8 cell stage or 5-6 days at blastocyst stage, then luteal support by natural progesterone from day of oocyte retrieval until beta HCG done after 2 weeks from oocyte retrieval (Huang et al., 2019).

**Serum sampling of IVF/ISI cases in different stages:**

Serum samples of women who were candidate for IVF/ICSI were drawn in controlled ovarian hyper-stimulation (COH) four times as follows: the day of ovulation stimulation start (Day-S); the day of ovum pick-up (Day OPU); and the day of transfer (Day-Transfer). Also, on ovum pick-up day.

All samples were centrifuged for 10 minutes at 1000 g, and were stored at -20°C until the final assay. The gathered samples were evaluated by comparative enzyme linked immunosorbent assay (ELISA) method for determining the level of CRP.

**Primary outcome:**

Assess CRP changes in different stages of IVF/ICSI cycles; and the association between these changes with pregnancy rate.

## Statistical methods

The collected data were coded, tabulated, and statistically analyzed using IBM SPSS statistics (Statistical Package for Social Sciences) software version 22.0, IBM Corp., Chicago, USA, 2013. Quantitative normally distributed data described as mean±SD (standard deviation) after testing for normality using Shapiro-Wilk test, then compared using independent t-test if normally distributed, while Pearson test was used for correlations. Qualitative data described as number and percentage and compared using Chi square test and Fisher's Exact test for variables with small expected numbers. Logistic regression was used to find out factors affecting stress, depression and anxiety. The level of significance was taken at P value < 0.050 was significant, otherwise was non-significant. Diagnostic characteristics was calculated as follows:

- Sensitivity = (True positive test / Total positive golden) x 100

- Specificity = (True negative test / Total negative golden) x 100

- Diagnostic accuracy = ([True positive test + True negative test] / Total cases) x 100 - Youden's index = sensitivity + specificity – 1

- Predictive positive value = (True positive test / Total positive test) x 100

- Predictive negative value = (True negative test / Total negative test) x 100

- LR+ = (sensitivity/ 1-specificity)

- LR- = (1- sensitivity / specificity)

- LR= LR+/ LR-

- Kappa=Observed agreement–chance agreement / 1–chance agreement

ROC curve was used to evaluate the performance of different tests differentiate between certain groups. McClish test was used to compare two independent AUCs (McClish, 1989). Linear regression model was used to find out independent factors affecting certain conditions. Log rank test was used to compare death rate. The level of significance was taken at P value < 0.050 is significant, otherwise is non-significant.

## RESULTS

Among the studied 100 cases, one case had chemical pregancny, 53 cases had clinical pregnancy. CRP at embryo transfer and CRP change at embryo transfer were significantly higher in cases with positive pregnancy. CRP at embryo transfer and CRP change at embryo transfer had significant moderate diagnostic performance in predicting pregnancy; was higher in CRP change at embryo transfer. CRP at embryo transfer ≥7.0 and CRP change at embryo transfer ≥5.1 had high specificity and PPV, but low sensitivity and NPV. CRP change at embryo transfer ≥5.1 had higher diagnostic characteristics in predicting pregnancy.

Table (1) shows that: Demographic characteristics among the studied cases. Mean±SD of age (years) was 29.4±4.0 with range 20.0–35.0. Mean±SD of BMI (kg/m2 ) was 26.1±3.4with range 4.8–29.0. Mean±SD of Duration of infertility (years) was 5.1±3.0 with range 1.0–18.0. Primary infertility was in 66 (66.0%) of cases, while secondary infertility was in 34 (34.0%) of cases

**Table (1): Demographic characteristics of the studied cases**

| Characteristics | | Mean±SD | Range |
|---|---|---|---|
| Age (years) | | 29.4±4.0 | 20.0–35.0 |
| BMI (kg/m$^2$) | | 26.1±3.4 | 4.8–29.0 |
| Duration of infertility (years) | | 5.1±3.0 | 1.0–18.0 |
| | | N | % |
| Type of infertility | Primary | 66 | 66.0 |
| | Secondary | 34 | 34.0 |

**Total=100**

Table (2) shows that: Hormonal profile of the studied cases. Mean±SD of FSH (IU/mL) was 6.8±2.5 with range 0.1–16.0. Mean±SD of LH (IU/mL) was 5.6±2.9 with range 0.3–17.0. Mean±SD of Estradiol (ng/mL) was 40.1±21.6 with range 10.0–150.0. Mean±SD of Prolcatin (µg/L) was 13.2±8.3 with range 1.0–46.0. Mean±SD of TSH (mIU/L) was 2.0±0.9 with range 0.4–5.2.

**Table (2): Hormonal profile of the studied cases**

| Hormons | Mean±SD | Range |
|---|---|---|
| FSH (IU/mL) | 6.8±2.5 | 0.1–16.0 |
| LH (IU/mL) | 5.6±2.9 | 0.3–17.0 |
| Estradiol (ng/mL) | 40.1±21.6 | 10.0–150.0 |
| Prolcatin (µg/L) | 13.2±8.3 | 1.0–46.0 |
| TSH (mIU/L) | 2.0±0.9 | 0.4–5.2 |

**Total=100**

Table (3) shows that: Oocyte retrival and embryo transfer among the studied cases. Mean±SD of Oocyte retrival was 11.9±7.6 with range 1.0–30.0. Mean±SD of Oocyte transfer was 2.5±0.8 with range 1.0–4.0.

**Table (3): Oocyte retrival and embryo transfer among the studied cases**

| Characteristics | Mean±SD | Range |
|---|---|---|
| Oocyte retrival | 11.9±7.6 | 1.0–30.0 |
| Embryo transfer | 2.5±0.8 | 1.0–4.0 |

**Total=100**

Table (4) shows that: CRP (mg/L) among the studied cases. Mean±SD at baseline was 2.4±0.8 with range 0.1– 4.3. Mean±SD at oocyte pickup was 2.7±0.9 with range 0.8–5.7. Mean±SD at embryo transfer was 6.4±2.5 with range 0.1–11.9. Mean±SD of CRP change at oocyte pickup was 0.3±0.4 with range -0.4–2.1. Mean±SD of CRP change at embryo transfer was 4.0±2.4 with range -1.7–9.4.

**Table (4): CRP (mg/L) among the studied cases**

| Time | Mean±SD | Range |
|---|---|---|
| Baseline | 2.4±0.8 | 0.1–4.3 |
| Oocyte pickup | 2.7±0.9 | 0.8–5.7 |
| Embryo transfer | 6.4±2.5 | 0.1–11.9 |
| Change at oocyte pickup | 0.3±0.4 | -0.4–2.1 |
| Change at embryo transfer | 4.0±2.4 | -1.7–9.4 |

Total=100. Change= time – baseline, negative value indicate reduction

Table (5) and figure (5) show that: **Pregnancy** occurred in 53 (53.0%) of the studied cases.

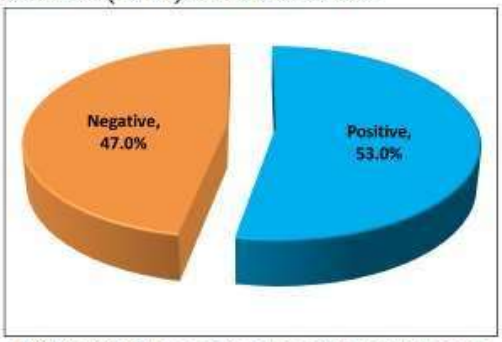


**Figure (5): Pregnancy among the studied cases**

Table (6) shows that: No significant differences according to pregnancy regarding BMI, duration of infertility and type of infertility. Age was significantly lower in cases with clinical pregnancy.

**Table (6): Comparison according to pregnancy regarding demographic characteristics**

| Variables | | Positive (N=53) | Negative (N=47) | P-value |
|---|---|---|---|---|
| Age (years) | | 28.7±3.8 | 30.3±4.0 | ^0.046* |
| BMI (kg/m$^2$) | | 25.7±4.0 | 26.5±2.7 | ^0.266 |
| Duration of infertility (years) | | 5.1±2.9 | 5.1±3.2 | ^0.991 |
| Type of infertility | Primary | 34 (64.2%) | 32 (68.1%) | #0.679 |
| | Secondary | 19 (35.8%) | 15 (31.9%) | |

^Independent t-test. #Chi square test

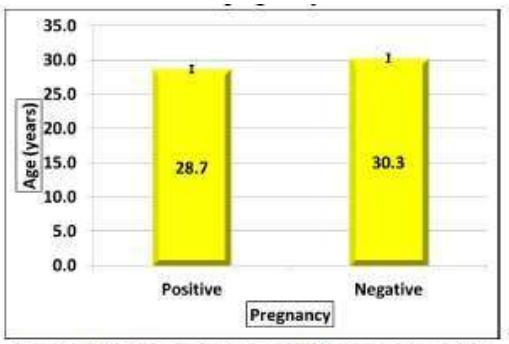


**Figure (6): Comparison according to pregnancy regarding age**

Table (7) shows that: No significant differences according to clinical pregnancy regarding Oocyte retrival and embryo transfer.

**Table (7): Comparison according to pregnancy regarding Oocyte retrival and embryo transfer**

| Characteristics | Positive (N=53) | Negative (N=47) | ^P-value |
|---|---|---|---|
| Oocyte retrival | 12.3±7.1 | 11.5±8.1 | 0.577 |
| Embryo transfer | 2.6±0.7 | 2.4±0.8 | 0.199 |

^Independent t-test.

Table (8) and figures (7) show that: No significant differences according to pregnancy regarding CRP at baseline and at Oocyte pickup as well as CRP change at oocyte pickup. CRP at embryo transfer and CRP change at embryo transfer were significantly higher in cases with positive pregnancy

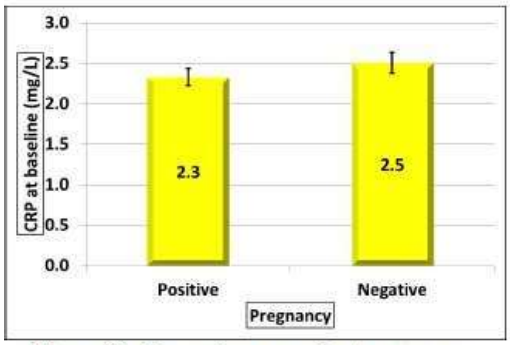


**Figure (7): Comparison according to pregnancy regarding CRP at baseline**

**Table (8): Comparison according to pregnancy regarding CRP (mg/L)**

| Time | | Positive (N=53) | Negative (N=47) | P-value |
|---|---|---|---|---|
| Baseline | | 2.3±0.8 | 2.5±0.9 | ^0.277 |
| Oocyte pickup | | 2.7±0.9 | 2.8±1.0 | ^0.429 |
| Embryo transfer | | 7.4±2.2 | 5.2±2.2 | ^<0.001* |
| Change at oocyte pickup | Mean±SD | 0.3±0.3 | 0.3±0.4 | ^0.692 |
| | Elevation | 46 (86.8%) | 35 (74.5%) | #0.117 |
| Change at embryo transfer | Mean±SD | 5.1±2.1 | 2.7±2.0 | ^<0.001* |
| | Elevation | 53 (100.0%) | 42 (89.4%) | §0.020* |

^Independent t-test. #Chi square test. §Fisher's Exact test. *Signifciant

Table (9) and figure (11): CRP at baseline and at Oocyte pickup as well as CRP change at oocyte pickup had no significant diagnostic performance in predicting pregnancy. CRP at embryo transfer and CRP change at embryo transfer had significant moderate diagnostic performance in predicting pregnancy; was higher in CRP change at embryo transfer

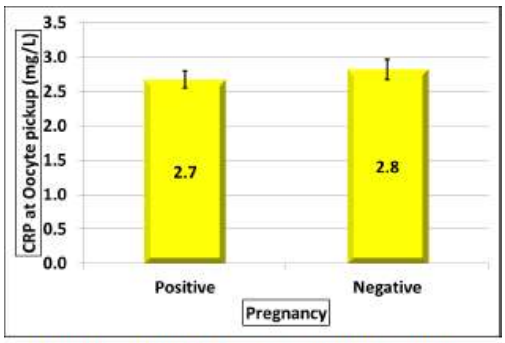


**Figure (8): Comparison according to pregnancy regarding CRP at Oocyte pickup**

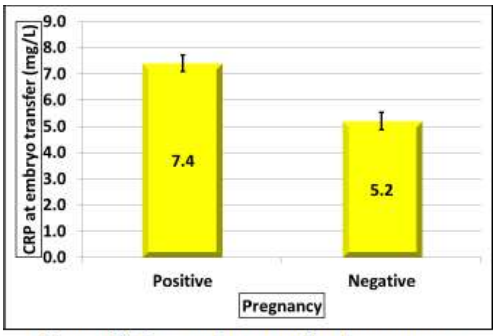


**Figure (9): Comparison according to pregnancy regarding CRP at embryo transfer**

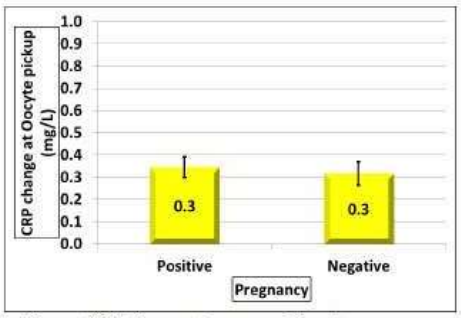


**Figure (10): Comparison according to pregnancy regarding CRP change at Oocyte pickup**

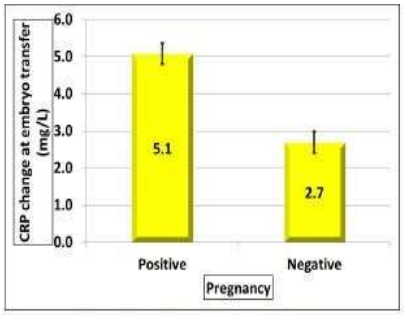


**Figure (11): Comparison according to pregnancy regarding CRP change at embryo transfer**

**Table (9): Diagnostic performance of CRP in predicting pregnancy**

| Time | AUC | SE | P | 95% CI | Cut off |
|---|---|---|---|---|---|
| Baseline | 0.556 | 0.058 | 0.335 | 0.441–0.671 | -- |
| Oocyte pickup | 0.551 | 0.059 | 0.377 | 0.437–0.666 | -- |
| Embryo transfer | 0.751 | 0.048 | <0.001* | 0.657–0.845 | ≥7.5 |
| Change at oocyte pickup | 0.552 | 0.058 | 0.367 | 0.439–0.666 | -- |
| Change at embryo transfer | 0.796 | 0.044 | <0.001* | 0.710–0.882 | ≥5.1 |

AUC: Area under curve, SE: Standard error, CI: Confidence interval, *significant

Table (10) and figure (12) show that: CRP at embryo transfer ≥7.0 and CRP change at embryo transfer ≥5.1 had high specificity and PPV, but low sensitivity and NPV. CRP change at embryo transfer ≥5.1 had higher diagnostic charactersitcis in predicting pregnancy.

**Table (10): Diagnostic charactersitics of CRP cutoff points in predicting pregnancy**

| Characters | At embryo transfer ≥7.5 | | Change at embryo transfer ≥5.1 | |
|---|---|---|---|---|
| | Value | 95% CI | Value | 95% CI |
| Sensitivity | 45.3% | 31.6%–59.6% | 56.6% | 42.3%–70.2% |
| Specificity | 89.4% | 76.9%–96.5% | 91.5% | 79.6%–97.6% |
| Diagnostic accuracy | 66.0% | 55.8%–75.2% | 73.0% | 63.2%–81.4% |
| Youden's index | 34.6% | 18.6%–50.7% | 48.1% | 32.5%–63.6% |
| PPV | 82.8% | 64.2%–94.2% | 88.2% | 72.5%–96.7% |
| NPV | 59.2% | 46.8%–70.7% | 65.2% | 52.4%–76.5% |
| LR+ | 4.26 | 1.77–10.26 | 6.65 | 2.53–17.48 |
| LR- | 0.61 | 0.47–0.80 | 0.47 | 0.34–0.65 |
| LR | 6.95 | 2.38–20.34 | 14.02 | 4.40–44.71 |
| Kappa | 0.337 | 0.176–0.498 | 0.470 | 0.311–0.629 |

CI: Confidence interval, PPV: Positive Predictive value, NPV: Negative Predictive value, LR+: Positive likelihood ratio, LR-: Negative likelihood ratio, LR: Diagnostic odd ratio

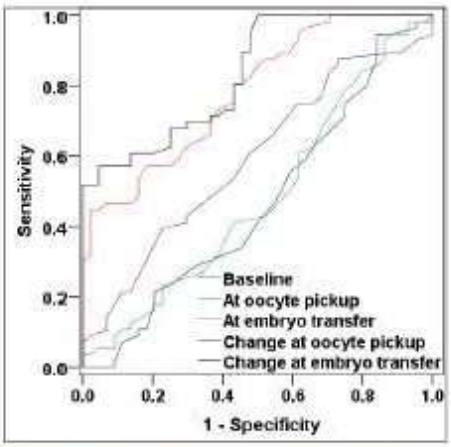


**Figure (12): ROC curve for CRP in predicting pregnancy**

## DISCUSSION

C-reactive protein (CRP) is an acute phase protein, produced by the liver. Its levels rise dramatically in the presence of infection or trauma. Small increases of serum CRP levels are believed to indicate low-grade inflammation (Seckin et al., 2012). Concentration of CRP, with a molecular weight of 110,000-120,000 KD, increases 1000 times after invasion and tissue damage. It activates complement and can attach to activated lymphocytes, invasive organisms, and damaged tissues. CRP can act as a non-specific opsonin to increase phagocytosis, removing cells and damaged, dead or dying organisms, reinforce innate immunity, and protection against tissue injury. Therefore, CRP by increasing the renovation speed, results in healing of damaged tissues. Studies have shown that CRP correlates with age and is higher in females (Arefi et al., 2010). As ovulation is considered to be an inflammatory process, the association between CRP levels and assisted reproductive technique (ART) cycles has been investigated by several studies (Orvieto et al., 2004, Levin et al., 2007, Arefi et al., 2009). This study was a Prospective cohort study to assess the curve of CRP changes in different stages of IVF/ICSI cycles; and the association between these changes with the rate of pregnancy. A total of 100 infertile women were included in this study. In this study there were no statistically significant differences according to clinical pregnancy regarding the sociodemographic data; BMI, duration of infertility and type of infertility, but Age was significantly lower in cases with clinical pregnancy, which coincides with the results of study reported by (Arefi et al., 2010, Seckin et al., 2012), but this studies showed no significant difference according to clinical pregnancy regarding age. Also, this study demonstrated There were no significant differences between pregnant and non-pregnant women according to clinical pregnancy regarding Oocyte retrieval and embryo transfer P value0.577, 0.199 respectively, which came in agreement with studies by (Seckin et al., 2012, Arefi et al., 2010, Elhalaby et al., 2020). Controlled hyperstimulation of the ovary and especially puncture of the ovaries in in-vitro fertilization (IVF) or intracytoplasmic sperm injection (ICSI) cycles is probably associated with changes in CRP concentrations. these changes may affect the success rate of IVF/ICSI, implantation, and pregnancy (Korhonen et al., 2016, Tasdemir et al., 2015). The level of CRP increases in ovulation stimulation cycle with standard long protocol. (Wunder et al., 2005) showed that leptin and CRP in ovulation stimulation cycle increase until the day of puncture, (Orvieto et al., 2004) showed the same results. This elevation from induction to puncture was 60% (Güdücü et al., 2013). (Levin et al., 2005) reported that the average CRP level in hyper stimulated patients was significantly higher than patients who aren't hyper stimulated or the control group. Ovulation induction is an inflammatory process leading to increased level of CRP, also, they determined CRP levels before commencing ovarian stimulation, during the stimulation phase and on the ovum pick-up day. They noted that higher hs-CRP levels before stimulation were associated with IVF treatment failure and also, they stated that patients who failed to conceive had significantly higher levels of CRP during all stages of the stimulation. (Almagor et al., 2004) took serial measurements of CRP from the day of oocyte retrieval, on embryo transfer day, during the implantation period and until βhCG testing day. They tested the assumption that the ratios of CRP (transfer/retrieval and day 5–7 of transfer/retrieval) rather than the daily concentration may significantly differ between pregnant and non-pregnant women. They demonstrated that the concentrations of CRP in blood increased during the first week following oocyte pick-up, and the CRP ratio of embryo transfer/ oocyte retrieval was significantly lower in conception cycles. They speculated that successful outcome was associated with a relatively small increment in CRP on the day of embryo transfer. Also, they found that serum CRP levels measured on day of embryo transfer to be correlated with the outcome of in-vitro fertilization (IVF) treatment patients, whose CRP level decreased on transfer day, had lower chance of pregnancy (Almagor et al., 2004). While patients whose CRP level elevated on embryo transfer day, applied treatment resulted in pregnancy. It has been suggested that its increase reflects a state of systemic inflammatory response due to ovarian stimulation, ovarian trauma and blastocyst-endometrium interaction (Arefi et al., 2009, Robinson et al., 2008). The half-life of CRP is approximately 19 hours, so it reflects ongoing inflammation (Seckin et al., 2012). The available results of the relationship between CRP concentrations and IVF success are controversial. Some studies have shown that there is no correlation between circulating levels of hs-CRP prior to ovarian stimulation and IVF outcome (Robinson et al., 2008, Wunder et al., 2005). In this study, CRP was measured by comparative ELISA method with designed antigen, which is quantitative and more precise than latex method. This study demonstrated that the level of CRP increases in ovulation stimulation cycle with long protocol, which agree with study by (Mohamed et al., 2019, Arefi et al., 2010). The current study showed no significant differences according to clinical pregnancy regarding CRP at baseline and at Oocyte pickup as well as CRP change at oocyte pickup, but CRP at embryo transfer and CRP change at embryo transfer were significantly higher in cases with positive pregnancy, which came in agreement with study by (Arefi et al., 2010), which showed The CRP level was not significantly different between pregnant women and those who did not become pregnant in any stages except for transfer day and study by (Mohamed et al., 2019), which showed statistically significant difference on comparing CRP levels between pregnant and nonpregnant cases on day of ovum pick up, while there was no statistically significant differenceon comparing CRP levels between pregnant and nonpregnant cases on day of embryo transfer, also, study by (Korhonen et al., 2016) found that patients whose CRP level decreased on transfer day, had lower chance of pregnancy while patients whose CRP level elevated on embryo transfer day, applied treatment resulted in pregnancy. On contrary, study by (Seckin et al., 2012) showed The mean serum hs-CRP levels were not significantly different between pregnant and non-pregnant women both on the day of initiation of ovarian stimulation (3.61 ± 2.86 and 3.24 ± 2.68 mg/L, respectively; p = 0.457) and on day 7 after embryo transfer (10.58 ± 11.35 and 9.14 ± 11.36 mg/L, respectively; p = 0.394). There was a significant rise in hs CRP levels at 7th day after embryo transfer as compared with the first day of gonadotrophin treatment in both conception and non-conception groups (p = 0.001). The mean CRP ratio was not significantly different between pregnant and non pregnant groups (3.71 ± 4.68 and 3.86 ± 7.24 mg/L, respectively; p = 0.995). (Seckin et al., 2012) concluded that the extent of the rise in hs-CRP levels did not appear to affect pregnancy rates in IVF and it cannot be used as a prognostic marker of IVF success. Also, other study by (Robinson et al., 2008) demonstrated that serum hs-CRP concentration is not a predictive marker of cycle or pregnancy outcome in women undergoing IVF treatment. There were no significant differences when comparing hs-CRP levels between the entry patients' group and any of the following groups - COH treatment (P=.513), normal stimulation (P=.526), successful fertilization (P=.718), successful division (P=.983) and clinical pregnancy (P=.595). In this study, CRP at baseline and at Oocyte pickup as well as CRP change at oocyte pickup had no significant diagnostic performance in predicting pregnancy. CRP at embryo transfer (P value 0.0001 and cutoff value ≥7.5) and CRP change at embryo transfer (P value 0.0001 and cutoff value ≥5.1) had significant moderate diagnostic performance in predicting pregnancy; was higher in CRP change at embryo transfer.

CRP at embryo transfer ≥7.5, and CRP change at embryo transfer ≥5.1 The receiver operating characteristic (ROC) curve showed that high specificity (89.4% and 91.5% respectively) and PPV (82.8% and 88.2% respectively), but low sensitivity (45.3% and 56.6% respectively) and NPV (59.2% and 65.2% respectively) and diagnostic accuracy (66% and 73% respectively). CRP change at embryo transfer ≥5.1 had higher diagnostic characteristics in predicting pregnancy. CRP was elevated in 86.8% (n=46) from women who got pregnant while women who did not get pregnant, CRP was elevated in 74.5% (n=35) at oocyte pick-up (P value = 0.11). And at embryo transfer, CRP was elevated in 100% (n=53) from women who got pregnant while women who did not get pregnant, CRP was elevated in 89.4% (n=42) (P value = 0.02) On contrary, study by (Mohamed et al., 2019) The receiver operating characteristic (ROC) curve showed that low specificity (SP) was 52.4%, high sensitivity (SV) was 82.8%, low positive predictive value (PPV) was 70.6%, high negative predictive value (NPV) was 86.8% and accuracy was 70 %. This study has many strengths point as using CRP level in ICSI and associate these changes with the rate of pregnancy and CRP test is simple method, cheap, valuable to predict the rate of pregnancy. There were many difficulties as this study was in corona outbreak so the IVF unit was locked down for more than seven months and this led to long time to recruit the cases for the study. Finally, this present study concluded that C-reactive protein (CRP) is a sensitive marker in inflammatory reactions. Controlled hyperstimulation of the ovary especially puncture of the ovaries in in-vitro fertilization (IVF) or intracytoplasmic sperm injection (ICSI) cycles is an inflammatory process leading to changes in CRP concentration, these changes may affect success rate of IVF/ICSI and clinical pregnancy. Patients whose CRP level decreased on transfer day, had lower chance of pregnancy, whereas patients whose CRP level elevated on embryo transfer day had high chance of pregnancy.

## CONCLUSION AND RECOMMENDATIONS

Controlled hyperstimulation of the ovary especially puncture of the ovaries in in-vitro fertilization (IVF) or intracytoplasmic sperm injection (ICSI) cycles is an inflammatory process leading to changes in CRP concentration, these changes may affect success rate of IVF/ICSI and clinical pregnancy. Patients whose CRP level decreased on transfer day, had lower chance of pregnancy, whereas patients whose CRP level elevated on embryo transfer day had high chance of pregnancy.

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**DOI:**10.31579/2642-9756/047