Original Article

# Role of IL-6 Levels in Predicting the Severity of Pregnancy Induced Hypertension and Fetomaternal Outcome

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## Abstract

*Background:* The hypertensive diseases complicating pregnancy still remains the major problem in developing countries that threatens the maternal health and foetal viability adding to maternal and neonatal mortality and morbidity. There are many screening tests available but none of them proved to be effective in predicting PIH and preeclampsia. Present study designed to assess maternal serum IL-6 levels in patients with pregnancy induced hypertension, preeclampsia and eclampsia and their correlation to fetomaternal outcome. *Method:* A case-control study on 50 normal pregnant women >20 weeks gestation as control and 50 women with SBP ≥ 140 mm of Hg and/or diastolic pressure ≥ 90 mm of Hg of pregnancy with >20 weeks gestation were recruited as study groups. All medical history, physical examination along with IL-6 levels were analysed. Outcome measures of newborns were assessed for maturity, APGAR scores, birth weight, and neonatal complications. *Results:* Mean SBP was 161.4 mm of Hg and mean DBP was 106.5 mm of Hg in study group. Mean IL-6 levels observed as 92.8 pg/ml in study group and 26.50 pg/ml in control group with p value of < 0.00001. In the present study, 21 (42%) babies were of APGAR < 7 in study group and 12 (24%) of babies were of APGAR < 7 in control group. The overall perinatal mortality was 36% in the study group, with 12 (24%) early neonatal deaths and it was only 2% in control group. *Conclusion:* The study results indicate that IL-6 concentrations were increased in the circulation of preeclamptic patients compared with control women. It supports the hypothesis of immune activation involved in preeclampsia and that IL-6 may participate in the abnormal immune response. Hence, it can be used as an indicator for assessing the severity of the disease and fetomaternal outcome.

**Keywords:** Preeclampsia; Interleukin-6; Pregnancy Induced Hypertension; Fetomaternal Outcome.

## Introduction

Pregnancy is the privilege of experiencing God's miracles on earth. Pregnancy and motherhood are the most beautiful and are really challenging time for a woman, both mentally and physically which is overwhelmed by many potential challenges of which one of the most common is preeclampsia (PE), a pregnancy specific multi system involving syndrome characterized by new onset of hypertension and proteinuria after twenty weeks of gestation [1].

Hypertensive disorders of pregnancy affects 5 – 20% of pregnant women [2]. It is a major cause of maternal mortality (15–20%), morbidity, perinatal mortality (20 - 25%), preterm births (15%), and intrauterine growth restriction (25%) in India. Preeclampsia is common below 25 years [3] and above 40 yrs of age. The incidence is gradually increasing over last few decades.

The key pathological factors of

preeclampsia are endothelial dysfunction and vasospasm. The hypoperfused placenta is a potential source of reactive oxygen species (ROS) as well as an inflammatory response. In response to inflammatory stimulus, there are increasing concentrations of pro inflammatory cytokines like IL-6, CRP, TNF- α etc.

IL-6 is a proinflammatory cytokine produced by mononuclear phagocytes, T-lymphocytes, B - lymphocytes and endothelial cells. Increasing concentrations of IL-6 play a significant role in pathogenesis of PIH. Thus IL-6 can be potential marker for assessing severity of preeclampsia and fetomaternal outcome.

## Material and Methods

This is a randomized case-control hospital based study done in the department of Obstetrics and Gynecology, Narayana Medical College Hospital, Nellore for a period of 2 years (October 2013 - September 2015). 50 normal pregnant women above 20 weeks were recruited as the control group and 50 women with Hypertensive disorders of pregnancy above 20 weeks were recruited as study group, after their consent for participation.

### Inclusion Criteria

All women with hypertensiv disorders of pregnancy with SBP ≥ 140 mm of Hg and or diastolic pressure ≥90 mm of Hg measured on two occasions atleast 6 hours apart with or without proteinuria.

### Exclusion Criteria

1. Preexisting medical conditions such as chronic inflammatory diseases (SLE, RA, Inflammatory bowel disease etc).

2. Acute inflammatory diseases (tonsillitis, UTI, chorioamnionitis, etc).

3. Cardiovascular disease.

4. Diabetes mellitus.

5. Renal disease.

6. Chronic hypertension.

### Patient Analysis

100 pregnant women attending the antenatal clinic at Narayana Medical College Hospital were recruited after a detailed history including the demography, complaints, period of gestation. Emphasis was made on previous obstetric history and medical history.

Complete general physical examination was carried out along with examination of the cardiovascular system and respiratory system. Obstetrical examination was carried out. Details regarding gestational age at the time of delivery, mode of delivery and any other significant intrapartum events were noted. Newborns were assessed for maturity, APGAR scores, birth weight, and neonatal complications.

### The Study was Divided into 2 Parts

*Part A:* Testing of IL-6 levels among the recruited study group and control group. Pregnant women with hypertensive disorders of pregnancy were taken as cases. A similar number of normal pregnant women were considered as controls.

*Part B:* The cases were analyzed and outcome was compared with controls.

### IL-6 Analysis

2ml blood sample was collected from the anticubital vein under strict aseptic precautions. The blood was allowed to clot for 30 minutes at room temperature and centrifuged at 3000 rpm for 15 minutes. The serum was then pipetted and placed in sterilized vials free of endotoxins at - $20^0$C until analysis.

### Biochemical Analysis

IL-6 was assayed by Diaclone Human IL-6 ELISA (Enzyme Linked Immunosorbent Assay) kits (France) which employs an antibody specific for human IL coated on a 96 well plate.

All pre and outcome measures were expressed in mean ± SD. Statistical analysis carried out using SPSS ver 16.0.

## Results

### Pregnancy Induced Hypertension (PIH) – Incidence

A total of 3504 pregnancies were delivered during the study period, of which 280 cases of PIH were diagnosed, giving an incidence of 8%. Majority of women (70%) were unbooked and (54%) were in the age group of 21-25 years. Most of them (74%) belonging to the low socioeconomic status. In present study, Pregnancy Induced Hypertension was more common in primigravida (54%) with average BMI of 30 kg/ $m^2$. The mean gestational age at presentation was 33.6 weeks. On systemic examination, mean systolic blood pressure was 161.4 mm of Hg and mean

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diastolic blood pressure was 106.5 mm of Hg in study group whereas in control group, mean systolic blood pressure was 106.4 mm of Hg and mean diastolic blood pressure was 72.6 mm of Hg (Table 1).

**Table 1:** Comparision of characteristics related to BP, Gestation and BMI.

| Charateristics | Normotensive (n=50) | Preeclamptic (n=50) |
|---|---|---|
| Age (Years) | 24.94 | 23.7 |
| Blood pressure(mm of Hg) | | |
| Systolic | 106.5 | 161.4 |
| Diastolic | 72.6 | 112.2 |
| Gesational age (weeks) | | 33.6 |
| Body Mass Index (kg/m²) | 28.16 | 29.99 |

*Laboratory Report*

In the present study, on haematological examination, mean haemoglobin was 10.55 gm/dl in the study group and 9.8 gm/dl in the control group. Mean platelet count in study group was 2.01 lakh/mm³ and in control group was 2.61 lakh/mm³. Mean serum urea was 22.4 mg/dL in study group and was 13.26 gm/dL in control group. Mean serum uric acid was 5.17 mg/dl in study group and 4.14 gm/dL in control group. Serum creatinine was 0.81 mg/dL in study group and in control group was 0.61 mg/dL. In the present study, mean IL-6 levels in the study group was 92.8 pg/ml and in control group was 26.50 pg/ml, with p value of < 0.00001 which is significant (Table 2).

**Table 2:** Comparision of lab parameters between normotensive pregnant women and preeclampsia

| Lab parameters in mean | Normotensive (n=50) | Preeclamptic (n=50) | *p* value |
|---|---|---|---|
| Hemoglobin (g/dL) | 9.6 | 10.55 | 0.005 |
| Platelet count (lakh/mm³) | 2.61 | 2.01 | 0.07 |
| Serum uric acid (mg/dL) | 4.14 | 5.17 | 0.00001 |
| Serum creatinine (mg/dL) | 0.61 | 0.81 | 0.00001 |
| Serum urea (mg/dl) | 13.26 | 22.4 | 0.00001 |
| IL 6 (pg/ml) | 92.8 | 26.50 | 0.00001 |

In the present study group of 50 women with Pregnancy Induced Hypertension, 11(22%) women were diagnosed as gestational hypertension, 13 (26%) women had mild pre - eclampsia, 22(44%) women had severe pre- eclampsia and 4 (8%) women had eclampsia.

*Delivery Mode & Fero-Maternal Outcome*

30 (60%) women in the study group were delivered by caesarean section whereas 15 (30%) were delivered by caesarean section group in the control group. mean birth weight in preeclamptic women was 1.8 kg and in normotensive women was 2.65 kg. In the present study, 21 (42%) babies were of APGAR < 7 in study group and 12 (24%) of babies were of APGAR < 7 in control group. The overall perinatal mortality was 36% in the study group, with 12 (24%) early neonatal deaths and it was only 2% in control group (Table 3).

**Table 3:** Delivery mode, fero-maternal outcome measurements

| Charateristics | Normotensive (n=50) | Preeclamptic (n=50) |
|---|---|---|
| Caesarean section | 30% | 60% |
| Mean birth weight | 2.65 kg | 1.8 kg |
| Apgar score at 5″ (< 7) | 24% | 42% |
| Perinatal mortality | 2% | 36% |

## Discussion

There are various classifications existing for hypertensive disorders of pregnancy. According to the classification of the Working Group of the (NHBPEP) National High Blood Pressure Education Program (2000) which is currently accepted worldwide.

1. Gestational hypertension — formerly termed pregnancy induced hypertension (PIH).

2. Preeclampsia and eclampsia syndrome.

3. Chronic hypertension superimposed on preeclampsia.

4. Chronic hypertension.

Preeclampsia is defined as hypertension

associated with proteinuria, greater than 0.3 g/L in a 24 - hr urine collection or 1+ by qualitative urine examination, after 20 weeks of gestation. Eclampsia is defined as seizures that cannot be attributed to other causes in women with preeclampsia.

Preeclampsia often affects young and nulliparous women, whereas older women are at greater risk for chronic hypertension with superimposed preeclampsia. The incidence is markedly influenced by race, ethnicity and thus by genetic predisposition. Other factors include environmental, socioeconomic, and even seasonal influences.

The hypothesis of preeclampsia syndrome is a two stage disorder is derived from observation that abnormal interphases between maternal, paternal and fetal tissues may cause preeclampsia.

*Preeclampsia as a Two-stage Disorder*
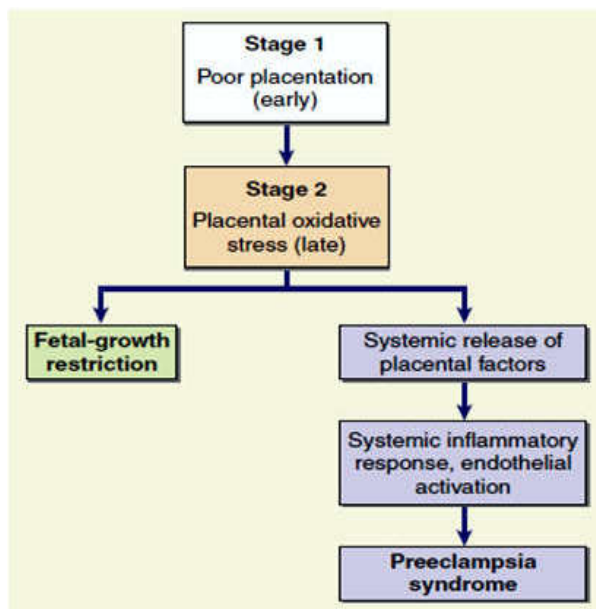


Schematic outlines the theory that the preeclampsia syndrome is a "two stage disorder ". Stage-1 is preclinical and characterized by faulty trophoblastic vascular remodeling of uterine arteries that causes placental hypoxia. Stage-2 is caused by release of placental factors into the maternal circulation causing systemic inflammatory response and endothelial activation. (Adapted from Borzychowski, 2006 [4], and Redman, 2009 [5], and their colleagues).

Cytokines play critical, essential roles in signalling between cells of the immune system with a wide a range of regulatory activities including the recruitment, activation, stimulation, killing, and suppression of immune and nonimmune cells. One of those cytokines is IL-6.

IL-6 is an interkeukin that acts as both proinflammatory cytokine and anti inflammatory cytokine. It is encoded by gene IL-6. It is produced by mononuclear phagocytes, T and B lymphocytes, fibroblasts, endothelial cells, hepatocytes and others to stimulate immune response.

*The Role of IL- 6 in PE*

1.   Increases vascular permeability by altering the ultrastructural distributions of tight junctions [6].

2.   Induces excessive or abnormal apoptotic and necrotic death of trophoblastic cells [7].

3.   Induces endothelial activation [7].

4.   Activates the renin angiotensin system [8].

5.   Contributes to an excess of decidual macrophages implicated in shallow extravillous trophoblast invasion of deciduas [9].

6.   Inhibits vascular prostacyclin production by downregulating cyclooxygenase Expression [10].

7.   Induces aberrant angiogenesis [11].

8.   Induces cell proliferation [12].

In the present study incidence of Hypertensive disorders of pregnancy is approximately 8% of all reported pregnancies which is correlating to study done by Dr. G. B. Doddamani et al [13]. In the present study, unbooked cases were 70% and 74% of women belong to low socioeconomic status Whereas it is 58% and 49% in the study done by Mehul. T. Parmar et al [14]. The mean BMI in study group was 29.99 and in control group was 28.16 which is correlating with Madhur Gupta et al [15] and A. Ozler et al [16].

The mean age in study group was 23.7 years and mean age in control group was 24.94 years which correlates with studies done by Dr. M. A. Sameer et al [17] and Magna Manjareeka et al [18]. In our study, more than half of the women (54%) were between 20 – 25 years of age group. This may be because most of the women in our study belong to rural areas and they start their reproductive career at an early age.

In the present study, mean gestational age at presentation was 33.68 weeks which correlates to the study conducted by N.Vitoratos et al [19] (32.71) and Magna Manjareeka et al [18] (34).

In the present study, mean systolic blood pressure in study group was 161.4 mm of Hg and in control group was 112.2 mm of Hg, which correlates with studies done by Kanika Mandi Choudhury et al [20] and Ghosh TK et al [21]. The p value was < 0.00001.In the present study, mean diastolic blood pressure in study group was 106.5 mm of Hg and in control group

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was 72.6 mm of Hg which is close to the studies done by Dr. Asia Naaz et al [22] and Attiya Ayaz et al [23]. The p value is < 0.00001.

In the present study, mean serum uric acid level was 5.17 mg/dl in study group and 4.14 mg/dl in control group which correlates with the study done by Cristina catarino et al [24] and Magna Manjareeka et al [18]. The mean serum creatinine level in study group was 0.81 mg/dL and in control group was 0.61 mg/dL which is close to study done by Magna manjareeka et al [18]. The mean serum urea in study group was 22.4 mg/dL and in control group was 13.26 mg/dL which is closely related to study done by Gearaldine et al [25]. The mean serum urea in study group was 22.4 mg/dL and in control group was 13.26 mg/dL which is closely related to study done by Gearaldine et al [25].

In the present study, mean IL-6 levels in study group was 92.8 pg/ml and in control group was 26.50 pg/ml. The p-value was < 0.00001. This proves that there is significant association of PE with IL-6 and it correlates with the studies done by Madhur Gupta et al [15] and Cristina et al [24] whose p value is < 0.001 and < 0.013 respectively.

In the present study, mean birth weight in study group was 1.8 kg and in control group was 2.65 kg which is close to the study conducted by N. Vitoratos et al [19]. In the present study, out of 44 live births, 30% of neonates had APGAR below 7 and 58% had APGAR ≥ 7 which correlates with study done by Dr. G. B. Doddamani et al [13]. In the present study, perinatal mortality was 36% which is closely related to study done by Dr. M. A. Sameer et al [17] in which perinatal mortality was 28.72%.

## Conclusion

The study results indicate that IL-6 concentrations were increased in the circulation of preeclamptic patients compared with control women. It supports the hypothesis of immune activation involved in preeclampsia and that IL-6 may participate in the abnormal immune response.

In present study, IL – 6 levels were also increased with increasing severity of the disease. Hence, it can be used as an indicator for assessing the severity of the disease and fetomaternal outcome.

The early detection of the markers of preeclampsia helps in the institution of prompt treatment. This has a definite role in reducing the morbidity and mortality in both mother and fetus. Further research is required to confirm the usefulness of IL – 6 in screening and monitoring of preeclampsia in large scale.

## References

1.  Sibai BM. Hypertensive Disorders in Women, Philadelphia, Saunders, 2004.

2.  Shakuntala Chhabra. Hypertensive Disorders with Pregnancy – An Unsolved Problem. FOGSI Focus 2007 May; P.1.

3.  Shruti S. Dubnashi, Wani RJ, Priti Chikhal, Hegde CV. PIH – Confounding Situations, Management Dilem-mas andSevere Consequences: Does Antenatal Care have a role. Bombay Hospital Journal; 2008; 50:45-49.

4.  Borzychowski AM, Sargent IL, Redman CW. Inflammation and pre- eclampsia.Semin Fetal Neonatal Med 2006; 11(5):309.

5.  Redman CWG, Sargent IL, Roberts JM: Immunology of abnormal pregnancy and preeclampsia. In Lindheimer MD, Roberts JM,Cunningham FG (eds): Chesley's Hypertensive Disorders of Pregnancy, 3rd ed. New York, Elsevier, In press, 2009.p.129.

6.  Desai TR, Leeper NJ, Hynes KL, Gewertz BL. Interleukin-6 causesendothelial barrier dysfunction via the protein kinase C pathway. J Surg Res 2002, 104:118–123.

7.  Chen LM, Liu B, Zhao HP, et al. IL-6, TNFalpha and TGFbeta promote nonapoptotic trophoblast deportation and subsequently causesendothelial cell activation. Placenta 2010; 31: 75–80.

8.  Lamarca BD, Ryan MJ, Gilbert JS, et al. Inflammatory cytokines in the Pathophysiology of hypertension during pre-eclampsia. Curr Hypertens Rep 2007; 9: 480–485.

9.  Charles J. Lockwood, Chih- Feng Yen, Murat Basar, Umit A. Kayisli, Maritza Martel, Irina Buhimschi, Catalin Buhimschi, S. Joseph Huang, Graciela Kirkun, and Frederick Schatz. Preeclampsia - related inflammatory cytokines regulate interleukin – 6 expression in human decidual cells. The American journal of pathology, 2008 June; 172(6).

10. Maruo N, Morita I, Ishizaki Y, Murota S. Inhibitory effects of interleukin 6 on prostaglandin I2 production in cultured bovine vascular endothelial cells. Arch Biochem Biophys 1992; 292:600–604.

11. Nilsson MB, Langley RR, Fidler IJ. Interleukin-6, secreted by human ovarian carcinoma cells, is a potent proangiogenic cytokine. Cancer Res 2005, 65:10794– 10800.

12. Tosato G, Pike SE. A rnonocyte-derived B-cell growth factor identified is IFN-beta-2 BSF-2RL-6.Ann N Y Acad Sci 1989; 557:181-91.

13. Dr. G. B. Doddamani, Dr. Usha. G. Doddamani. Perinatal outcome in pre-eclampsia: A prospective study. sch. J. Med. Sci; 2014: 2(1c):291–293.

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14. Mehul T Parmar, Harsha M solanki, Vibha V Gosalia. study of risk factors of perinatal death in pregnancy induced hypertension (PIH). National Journal of Community Medicine, 2001 Oct–Dec; 3(4).

15. Madhur Gupta and Suresh Chari. Predictive value of inflammatory Cytokines in Preeclampsia. IJBAR (2015); 6(04):334-338.

16. A. Ozler, A. Turgut, M. E. Sak, M. S. Evsen, H. E. Soydinc, O. Evliyaoglu,T. Glu. Serum levels of neopterin, tumor necrosis factor-alpha and Interleukin- 6 in preeclampsia: relationship with disease severity. European Review for Medical and Pharmacological Sciences 2012; 16:1707- 1712.

17. M. A. Sameer, D. P. Meshram , S. A. Deshpande, D. Sadhu, Pandit. Role of Platelet count as prognostic marker in Pregnancy Induced Hypertension. IOSR – JDMS. 2014; 13(4) Ver. III: 39–43.

18. Magna Manjareeka, Sitikanta Nanda. Elevated lvels of serum uric acid, creatinine or urea in preeclamptic women. International Journal of Medical Science and Public Health. 2013; 2(1).

19. N. Vitoratos, E. Economou, C. Iavazzo, K. Panoulis, and G. Crestas. Maaternal Serum Levels of TNF-Alpha an IL-6 long after delivery in Preeclamptic and Normotensive Pregnant women. Hindawi Publishing Corporation Mediators of Inflammation Volume 2010; Article ID 90864.

20. Kanika Mandi Choudhury, Munnam Das, Sulekha Ghosh (sarkar), Debasis Bhattacharya, Tapan Kumar Ghosh. Value if Serum â-hcG in Pathogenesis of Preeclampsia. J Clin Gynecol Obstet. 2012; I(4-5): 70-75.

21. Ghosh TK, Ghosh s, Bhattacharjee D. C – reactive protein levels in women with pregnancy induced hypertension. Bangladesh Journal of Medical Science 2011 Jul; 10(3).

22. Asiya Naaz, P. sushma, Mohammed Meraj Ahmed, DVHS Sarma, Amer Ali Baig, K. Eswar Reddy. A study on fibrinogen levels and platelet count in pregnancy induced hypertension. International Journal of scientific and Research Publications, 2015 Mar; 5(3):11-09.

23. Attiya Ayaz, Taj Muhammad, Shaheryar A Hussain, Sadia Habib. Neonatal outcome in pre-eclamptic patients. J Ayub Med Coll Abbottabad 2009; 21(2).

24. Cristina Catarino, Alice Santos-Silva, Luis Belo, Petronila Rocha-Pereira, Susana Rocha, Belmiro Patricio, Alexandre Quintanilha, and Irene Rebelo. Inflammatory disturbances in Preeclampsia: Relationship between maternal and umbilical cord blood. Hindawi publishing Corporation Journal of Pregnancy Volume 2012; Article ID 684384.

25. Gearaldine Monteiro, Subbalakshmi N.K. & Sheila R. Pai. relevance of measurement of haematological parameters in subjects with pregnancy induced hypertension.NUJHS 2014 Mar; 4(1). ISSN 2249-7110.