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# Fetal Plasma Tumor Necrosis Factor Concentration in Normal Pregnancy

**Key Words**
Cordocentesis
Fetal blood
Tumour necrosis factor
Fetal immunology

**Abstract**
The aim of the study was to investigate changes with gestation of fetal plasma tumor necrosis factor alpha (TNF-α) concentration. In a cross-sectional study, enzyme-linked immunoassay was used to measure plasma TNF-α concentration in 40 fetal blood samples obtained by cordocentesis (n = 25), cardiocentesis (n = 5) or at elective caesarean section (n = 10) at 12–38 weeks gestation. The fetal plasma concentration of TNF-α increased from a mean of 13.5 pg/ml at 12 weeks gestation to 37.5 pg/ml at 38 weeks ($r = 0.59$, $p < 0.0001$), and was significantly associated with the monocyte count ($r = 0.56$, $p < 0.001$). TNF-α is present in the fetal circulation from at least 12 weeks and the changes in plasma TNF-α concentration with gestation coincide with the development of the fetal monocyte-macrophage system.

## Introduction

Tumor necrosis factor alpha (TNF-α), which is primarily produced by the monocyte-macrophage system, plays a major role in host defence against infections and tumors [1]. TNF has also been implicated in regulation of growth and feto-placental development [2, 3]. Jäättelä et al. [2] measured TNF-α in amniotic fluid samples and reported that the concentration is higher in the second than in the third trimester of pregnancy. In contrast, Chen et al. [3] who measured TNF-α in the placenta and decidua

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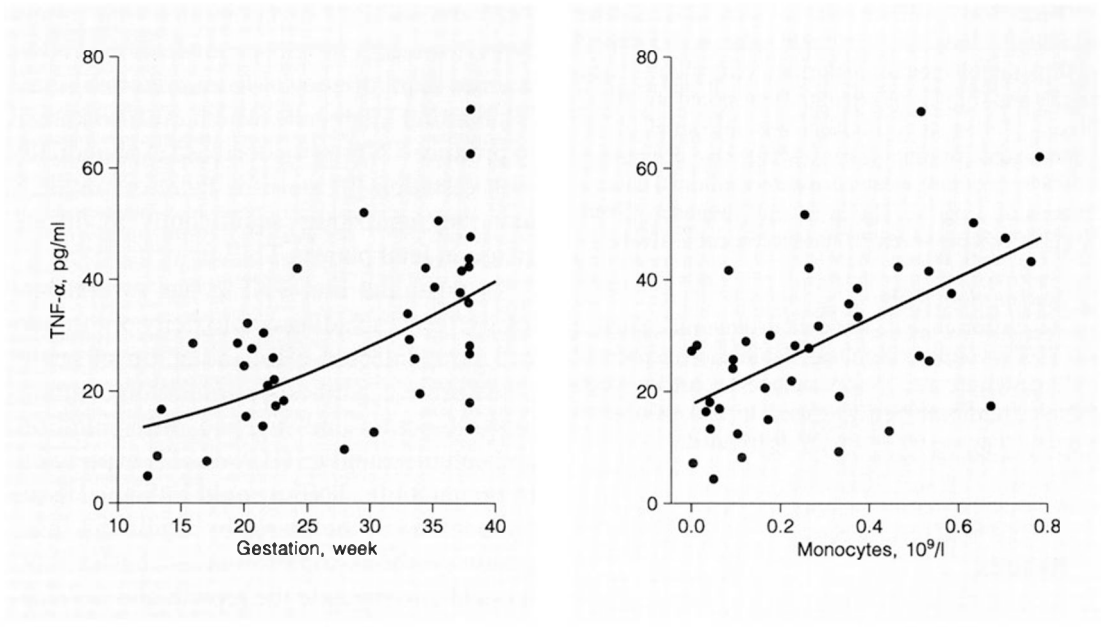


**Fig. 1.** Relationship of fetal plasma TNF-α concentration with gestation (**a**) ($r = 0.59$, $p < 0.0001$) and fetal monocyte count (**b**) ($r = 0.56$, $p < 0.001$).

found the levels to be higher in late than in early gestation.

The aim of this study is to determine whether TNF-α is present in the fetal circulation, and if so, to define its relationship to the developing fetal monocyte-macrophage system.

## Patients and Methods

In a cross-sectional study of 40 pregnancies, fetal blood samples were obtained either by (1) cordocentesis, at 18–37 weeks gestation (n = 25); (2) fetal cardiocentesis, from women undergoing elective terminations of pregnancy for social indications at 12–17 weeks (n = 5), and (3) from umbilical cord blood obtained at elective caesarean section at term for either breech presentation or previous caesarean section (n = 10). The indications for cordocentesis were fetal karyotyping because of advanced maternal age (n = 4), determination of fetal blood group in red blood cell isoimmunized pregnancies and where the fetus was subsequently found to be Coomb's negative (n = 2), and karyotyping for fetal malformations, such as hydronephrosis (n = 19). In all cases, the fetal abdominal circumference, haemoglobin and white blood cell concentrations were within the appropriate reference range for gestation and the fetal karyotype was normal [4, 5].

In each case, gestation was determined from the menstrual history and confirmed by an ultrasound scan in early pregnancy. Cordocentesis was performed without maternal sedation or fetal paralysis, and in all cases umbilical venous blood was obtained. Fetal cardiocentesis was performed under general anaesthesia and immediately before termination of pregnancy. Kleihauer-Betke testing confirmed that all blood samples contained only fetal blood. Fetal blood samples (180 µl) were collected into 20 µl of isotonic edetic acid solution (0.5 mmol/l in 0.15 mmol/l sodium chloride) and the full blood count was determined using a Coulter S-Plus counter (Coulter Electronics, Luton, UK). Blood films were stained by the May-Grünwald-Giemsa method for the differential white blood cell count.

Fetal blood samples (400 µl) were also collected into 20 µl of isotonic edetic acid solution (0.5 mmol/l in 0.15 mmol/l sodium chloride), centrifuged immediately and the plasma separated and stored at −20 °C. Plasma TNF-α concentration was measured by enzyme-linked immunoassay (Medgenix diagnostics, Brussels, Belgium), which can detect minimal concentrations of 3 pg/ml. The intra- and interassay coefficients of variation were 3.7 and 8.0%, respectively.

*Statistical Analysis*

Regression analysis was used to determine whether the TNF-α concentration was significantly associated with gestation and blood monocyte count. Logarithmic transformation was used to make Gaussian any data that was not normally distributed.

## Results

The concentration of TNF-α in fetal plasma increased significantly from a mean of 13.5 pg/ml at 12 weeks gestation to a mean of 37.5 pg/ml at 38 weeks (fig. 1, $r = 0.59$, $p < 0.0001$), and was significantly associated with the blood monocyte count (fig. 1, $r = 0.56$, $p < 0.001$).

## Discussion

This study has demonstrated that TNF-α is present in the fetal circulation from at least 12 weeks, and that the plasma concentration increases with advancing gestation. Since none of our patients had a history of infection and since none of the fetuses had a tumour, the observed alterations with gestation in fetal plasma TNF-α concentration are likely to reflect physiologic changes with fetal development.

The most likely source of TNF-α in the fetal circulation is the fetal monocyte-macrophage system and T lymphocytes, which also increase with advancing gestation [5, 6]. In vitro studies have demonstrated that monocytes from term infants produce more TNF-α activity than monocytes from preterm infants [7]. Natural killer cells, which are also known to produce TNF-α [8], decrease exponentially with gestation [9] and it is therefore unlikely that they contribute significantly to the increase in fetal plasma TNF-α.

In postnatal life, TNF-α has several biologic effects including cytotoxicity of tumour- and virus-infected cells, induction of acute-phase protein synthesis, stimulation of granulocyte function and myeloid differentiation, and enhancement of HLA class II expression. In prenatal life, TNF-α could influence tissue remodelling in the uterus by regulating collagenase synthesis and promoting angiogenesis. It could also regulate the growth and development of the feto-placental unit by inducing cell differentiation, controlling the energy requirements due to its catabolic effect, and promoting the secretion of other cytokines such as interleukin-1, γ- and β-interferon and haematopoietic colony-stimulating factors [1–3].

This study has demonstrated that TNF-α is present in the fetal circulation from at least 12 weeks. The changes in plasma TNF-α concentration with gestation coincide with the development of the fetal monocyte-macrophage system.

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