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Daily production of human tumor necrosis factor in lipopolysaccharide (LPS)-stimulated ex vivo blood culture assays.

Abstract

Tumor necrosis factor (TNF) is a pleiotropic cytokine with immunological and neuroendocrine activities. A useful tool for studying TNF is the measurement of its in vitro and/or ex vivo over-expression, induced by a variety of stimuli on isolated peripheral mononuclear cells or whole blood, respectively. The capacity to over-express TNF, in ex vivo LPS-stimulated whole blood from 18 normal

individuals, showed inter-individual variations ranging from high (3 ng/ml) to low (0.7 ng/ml) producers. Although at a lower level, a similar situation was observed in the spontaneous production of the cytokine. In order to detect cyclic effects in these variations, blood samples were taken at 08:00, 12:00, 16:00 and 20:00 hours, from nine healthy volunteers, and cultured in the ex vivo system. TNF and cortisol were measured by immunometric assays. Both, LPS-stimulated whole blood and plasma showed important, individual variations in TNF levels. Although cortisol levels presented a normal circadian cycle, these individual patterns in TNF production were basically conserved during the day ($p > 0.05$), and no correlation was observed between the levels of the hormone and those of the cytokine. When total TNF levels were determined at 20:00 hours, a moderate, temporary variation pattern of the cytokine production was found. These results suggest that cortisol does not play a predominant role in determining the ex vivo capacity of blood to produce TNF. Presumably, the variable capacity to produce the cytokine may have a strong genetic component.

Full text

INTRODUCTION

In physiological concentrations of pg/ml of plasma, cytokines mediate a series of metabolic and biochemical changes in responding cells. Inflammatory and immune responses, resulting from autocrine, paracrine and endocrine cytokine actions may follow. Massive tissue damage, as a result of a variety of traumas, including sepsis, precipitates the loss of control of cytokine release, leading to a series of reactions involving damage to many organs. Disseminated micro-coagulation and vascular collapse may follow, culminating in a systemic inflammatory response syndrome or septic shock [1, 2].

Among the most important cytokines in this process, is tumor necrosis factor or TNF, a 17 kDa polypeptide released by monocytes and macrophages, which shares a wide range of beneficial and deleterious effects with other cytokines. TNF is the first factor involved in the cytokine cascade, in response to sepsis [3-6]. It also participates in a wide variety of sub-acute and chronic illnesses of non-infectious origin, such as burns, trauma and haemorrhagic shock [7], rheumatoid arthritis [8, 9], systemic lupus erythematosus, and diabetes mellitus [10]. The quantification of TNF is difficult due to its short (6 to 20 min) half-life, the existence of two active forms (seric and membrane-associated), and to the

presence of soluble receptors [11]. A useful tool for the study of TNF is the measurement of its *in vitro* and/or *ex vivo* over-expression, by peripheral mononuclear cells, isolated [12-14] or present in whole blood [15], respectively.

The observed variations in individual plasma TNF levels have been attributed to multiple factors. Among them, probably major histocompatibility complex (MHC)-associated genes are the most important. Thus, cells from HLA-DR2 individuals produce lower TNF levels than individuals bearing HLA-DR3 or HLA-DR4 haplotypes [16, 17]. In addition, a fundamental participation of TNF gene promoter region polymorphism, in the inter-individual differential expression of the cytokine, has recently been demonstrated. Specifically, these polymorphisms include nucleotide substitutions (G = › A or G = › C), at positions -238, -244, -308, -376 and -419 [17-20].

Mononuclear cells from different human individuals display a wide range of ability to produce TNF in *ex vivo* culture systems in response to a lipopolysaccharide (LPS) stimulus [13]. Circadian rhythms in immune system functions [21], the inhibitory capacity of glucocorticoids on *in vivo* and *in vitro* TNF production [22, 23], and genetic components, may influence the *ex vivo* capacity to produce the cytokine. Based on this, the aim of our study was to determine daily variations in TNF levels, both in plasma and in *ex vivo* cultures, and to investigate any association of these levels with daily cortisol fluctuations.

METHODS

Volunteers. For the analysis of the inter-individual variations in the over-expression of TNF, in the *ex vivo* assay of LPS-stimulated whole blood, 18 normal individuals (9 women and 9 men, average age 30.3) were enrolled. For the study of daily TNF variations, 9 healthy volunteer donors (4 women and 5 men, average age 24.5 yr) were recruited. In both groups, medical and dental status were determined. In addition, haemogram, urine sediment, and HIV (ELISA, ABBOTT, South Pasadena, Ca, USA) analyses were performed. In the second group, each individual participated in an experimental session between 08:00 AM and 08:00 PM, donating 1 blood sample every 4 hours. Volunteers were secluded in a comfortable, isolated environment. Light, food and water were provided *ad libitum*.

Bacterial LPS. LPS from *Escherichia coli*, serotype 026:B6 (SIGMA, St. Louis, Mo, USA) was used. All preparations were sonicated in three 15 s cycles, dissolved in endotoxin-free water to 100 mug/ml, then sonicated again and diluted out in cell culture medium, as required.

Anti-human recombinant TNF monoclonal antibody (anti-hrTNF Mab). This was generated in our laboratory by standard hybridoma technology [24]. Briefly, 6 to 8 week old BALB/c female mice (Jackson Laboratories, Bar Harbor, Ma., USA), were immunized with a 3 mug intra-splenic injection of the pure recombinant cytokine (Genetech, San Francisco, Ca., USA). The immune response was monitored by a solid phase immunoradiometric assay (IRMA). The mouse was boosted i.p. with 3 mug of the recombinant protein in PBS. Four days later, spleen cells were isolated and fused with NS02 myeloma cells. A monoclonal IgM antibody (E8), was expanded *in vitro* and *in vivo* and was purified by ammonium sulfate precipitation and size exclusion chromatography. Its biological activity was verified by immunowestern blot against the recombinant antigen.

Anti-human recombinant TNF polyclonal rabbit sera (anti-hrTNF PoAb). 2-4 month old female New Zealand rabbits were immunized four times subcutaneously in the dorsal region, with 10 mug of hrTNF, at weekly intervals. The first injection contained complete Freund ′s adjuvant, and the following, the incomplete reagent.

Immunoradiometric assay (IRMA) for TNF. PVC plates were coated with 50 mul of the anti-hrTNF Mab at 3 mug/ml in bicarbonate buffer, pH 9.6, for 12 hours at 4° C. The plates were then washed and blocked with 1% w/v BSA in PBS, for 2 hours at 4° C. After washing, 50 mul of the WBCS supernatants were added per well, in triplicate and incubated for 2 hours at 4° C. The plates were washed and 50 mul/well of the anti-hrTNF PoAb were added, in a 1/1,000 dilution, and incubated for 2 hours at 4° C. After washing, 25 mul/well (100,000 cpm) of a ^{125}I-labelled affinity purified IgG goat anti-rabbit IgG were added, followed by incubation for 2 hours at room temperature. Finally, the radioactivity was quantified in cpm. Each measurement was interpolated into a standard curve built with hrTNF dilutions (7-8,000 pg/ml).

Whole blood culture system (WBCS) for TNF detection. Sterile conditions were maintained during the whole procedure, as originally described [15], with minor modifications. Briefly, blood samples were diluted 5 times in RPMI-1640 medium,

supplemented with 200 mM L-glutamine, 200 IU/ml penicillin, 200 mg/ml streptomycin and 10 IU/ml heparin. After 4 hours incubation of 900 ml diluted blood, at 37° C in 5% CO_2, LPS was added to each culture well to a final concentration of 10 mug/ml. After an additional incubation of 12 hours, the content of each culture well was transferred to Eppendorf tubes, centrifuged, and TNF was measured by an antigen capture IRMA using an anti-human recombinant TNF (hrTNF) Mab.

Detection of plasma TNF and cortisol levels at different times during the day. Plasma TNF and cortisol were measured by ELISA (Biotrak▒, Amersham▒, Little Chalfont Buckinghamshire, England; quantification limit 0.1 pg/ml) and RIA (Coat-A-CountR cortisol, Diagnostics Products Corporation, Los Angeles, Ca, USA; quantification limit 0.2 mug/dl), respectively.

Statistical analysis. Variations in TNF levels both in *ex vivo* culture and plasma, throughout the day were analyzed by a double batch-divided design. For comparison of means, a Student-Newman-Keuls test was used. To determine differences between spontaneous and LPS-induced TNF production, both in women and men, a double tail Student's t test for independent samples was applied [25]. The induced and basal production of TNF and the association between plasma and *ex vivo*-expressed TNF with cortisol concentrations, at different times during the day, were correlated by linear regressions. The data were interpolated to standard curves and expressed as pg of TNF/ml of diluted blood.

Ethical considerations. Informed written consent was provided by volunteer human blood donors. Venipuncture was carried out by trained medical personnel.

Bioethical considerations. Mice and rabbits used were kept in our animal facility under veterinary supervision, following the NIH guidelines for the care and use of laboratory animals [26].

RESULTS

Inter-individual variations in the over-expression of TNF are shown in Figure 1. TNF levels, measured in the WBCS of 18 individuals, are significantly higher when induced with LPS, as compared to the spontaneous production of the cytokine

($p < 0.01$). Large inter-individual differences (0.7-3 ng/ml) were observed among the individuals. In 7 out of the 18 determinations shown here, the spontaneous production was above 500 pg/ml, a level seldom observed in physiological *in vivo* conditions. No significant differences between spontaneous or induced TNF levels in men and women ($0.1 > p > 0.05$), or correlation ($p > 0.1$) between basal and LPS-induced cytokine levels were found. Two individuals showed undetectable (less than 30 pg/ml) TNF levels.

The *ex vivo* levels of TNF, in response to an LPS stimulus, and its plasma levels, fluctuated throughout the day (blood samples obtained at 08:00, 12:00, 16:00 and 20:00 hours), as shown in Figure 2. We observed an important inter-individual variation in cytokine production. No correlation was observed between spontaneous and LPS-induced TNF levels ($p > 0.05$, $r = 0.69$), even when the analysis was performed separately for men and women ($p > 0.05$). On the other hand, LPS-induced TNF levels, were higher in men than women ($p < 0.05$).

As is shown in Figure 3, a significant difference ($p < 0.05$) for the total TNF levels found in the sample obtained at 20:00 hours was observed. This situation was consistent with the symmetrical linear regression analysis observed for *ex vivo* TNF levels at 08:00 hours, both basal and LPS-induced, simultaneously compared with those obtained at other times (12:00, 16:00 and 20:00 hours), as shown in Figure 4. The individual comparisons between 8:00 hours and other individual times, both for basal and LPS-induced TNF levels, were statistically significant, the highest level being obtained when 8:00 and 20:00 hours were compared (results not shown).

Surprisingly, no association was found between the endogenous plasma cortisol concentration and the TNF levels in either the *ex vivo* culture system or in plasma (Table 1).

DISCUSSION

Although a previous study [15] suggests that individual variations in TNF levels are minor in the absence of stimulus, Figure 1 shows a large variation in both spontaneous (*i.e.* individual number 15 spontaneously produces about 6 times more TNF than individuals 7 and 13) and LPS-induced TNF production (*i.e.* individual 18 produces about four times more TNF than individuals 2, 3, 6 and 13)

among human blood donors, when using the *ex vivo* system. In 7 out of the 18 determinations shown here, the spontaneous production was above 500 pg/ml, a level seldom observed in physiological *in vivo* conditions. The absence of correlation between these levels and those induced by LPS, may be due to the different nature of the stimulus (*i.e.* plastic, manipulation, contamination with undetectable levels of another type of LPS, etc.).

Considering the inhibitory capacity of glucocorticoids on *in vivo* and *in vitro* TNF production [22, 23], the daily rate of cortisol production should correlate with TNF levels. Although a circadian rhythm for cortisol levels was found (results not shown), we did not find an association between endogenous cortisol concentration and TNF produced in either the *ex vivo* system, and plasmatic TNF concentrations (Table 1). Since the biological effects of these hormones are determined by both the concentration and number of specific cellular receptors [27], it should be pointed out that the *ex vivo* blood culture, contains B and T, and other immunocompetent cells, in addition to monocytes, which all display receptors for glucocorticoids and which could sequestrate the hormone present in the culture. Thus, our results are not necessarily inconsistent with the known suppressive role of glucocorticoids on cytokine production and inflammation [28].

The higher LPS-induced TNF levels found in men (Figure 2) may indicate that female sexual hormone concentrations may modulate the cytokine levels. Thus, several studies indicate that, *in vitro*, both progesterone and estradiol reduce the expression of TNF mRNA in monocytes from peripheral blood during the luteal phase of ovarian cycle [29]. In addition, it has been demonstrated that *in vivo* estrogen treatment decreases the TNF and IL-1 production in human monocytes [30].

The data shown in Figure 3 represent the mean of both plasma and LPS-induced TNF levels for defined times. Although, the software used in the statistical analysis (double batch divided design) does not provide an estimation of the errors (presumably large, given the nature of the comparisons), the results show that there is an overall statistically significant increase in TNF levels at 20:00 hours. However, the general pattern of basal and LPS-induced TNF levels (high or low), in most of the individuals, is statistically constant along the day. This fact is consistent with the symmetrical linear regression analysis observed

among *ex vivo* TNF levels at 08:00 hours, both basal and LPS-induced, compared with those obtained at other times (Figure 4).

Although the *ex vivo* assay, used in this study, detects the highly variable capacity to express TNF, when the cultures were stimulated with LPS, the system does not provide information regarding the functional activity of the cytokine present in the sample. In other words, allelic polymorphism of TNF, at the gene or protein level [31] could have functional consequences. Likewise, equimolar concentrations of TNF from high and low TNF producers (such as individuals 18 and 3, respectively, Figure 1) may not have equivalent functional activities. Following a similar reasoning, it can be proposed that TNF coded for by different HLA haplotypes, may have different functional capabilities.

The large variations observed in induced and spontaneous TNF secretion, may have a genetic basis. Thus, the TNF secretory response can be regulated at a transcriptional, post-transcriptional and translational level [32]. Moreover, *in vitro* TNF levels have been associated with certain TNF alleles and microsatellite regions, as well as with certain HLA haplotypes [16, 17].

Since the TNF gene is located within the MHC, the most polymorphic system known in vertebrates (some loci may contain over 100 alleles) [33], emphasis has been made in studying the association of these alleles with certain autoimmune diseases. Thus, the glomerulonephritis associated with systemic lupus erythematosus is more frequent in the low TNF producing haplotype $DR2^+$ [16, 34]. Moreover, studies in insulin-dependent diabetes mellitus have associated the presence of DR2 haplotype with resistance to the disease [35-37]. When peripheral blood samples are stimulated with LPS, HLA-DR2 and $DQw1^+$ individuals produce less TNF than those samples from HLA-DR3 and $DR4^+$ individuals [13, 14, 16, 36, 38]. Given its location, the TNF gene shares characteristics with the MHC, such as the high level of conservation among species and reduced genetic recombination, which results in a linkage desequilibrium between MHC class II and the TNF genes within the population [36].

More important in the TNF inter-individual quantitative variations is the polymorphism affecting the TNF gene promoter region, at position -308. This polymorphism defines the TNF1 (presence of G) and TNF-2 (presence of A) genotypes, both low and high TNF producers, respectively [17-19]. In conditions

such as: cerebral malaria [39], mucocutaneous leishmaniasis [40], Langerhans cell histiocytosis [41], and rheumatoid arthritis [42], it has been noted that individuals homozygous for the TNF-2 allele, are at greater risk of during, or have a higher susceptibility to damage.

TNF production depends on many factors. Some studies have reported that psychological stress stimulates TNF, IFN-gamma, IL-6, and IL-1Ra production, while IL-10 and IL-4 production is decreased, thus evoking a Th1 response [43]. However, the main factors seem to be of a genetic nature, including: variations in the expression level of CD14, associated with the polymorphism of the promoter region of its coding gene at position -206 (C => T) [44]; MHC haplotypes, and the polymorphism affecting the promoter region of the TNF gene [17-20].

CONCLUSION

Finally, we can conclude that humans display a wide variation in both spontaneous and LPS-induced TNF production, when using the *ex vivo* short-term blood culture system. The higher TNF concentration at 20:00 hours may indicate the existence of a moderate, temporary variation pattern in cytokine production. However, the general pattern of basal and LPS-induced TNF levels (high or low), in most of the individuals, is statistically constant throughout the day. Although cortisol levels presented a normal circadian cycle, the individual patterns in TNF production were basically conserved during the day ($p > 0.05$) and, interestingly, no correlation was observed between the levels of the hormone and those of the cytokine. These results suggest that cortisol does not play a predominant role in determining the ability of *ex vivo* blood to produce TNF.

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Figures

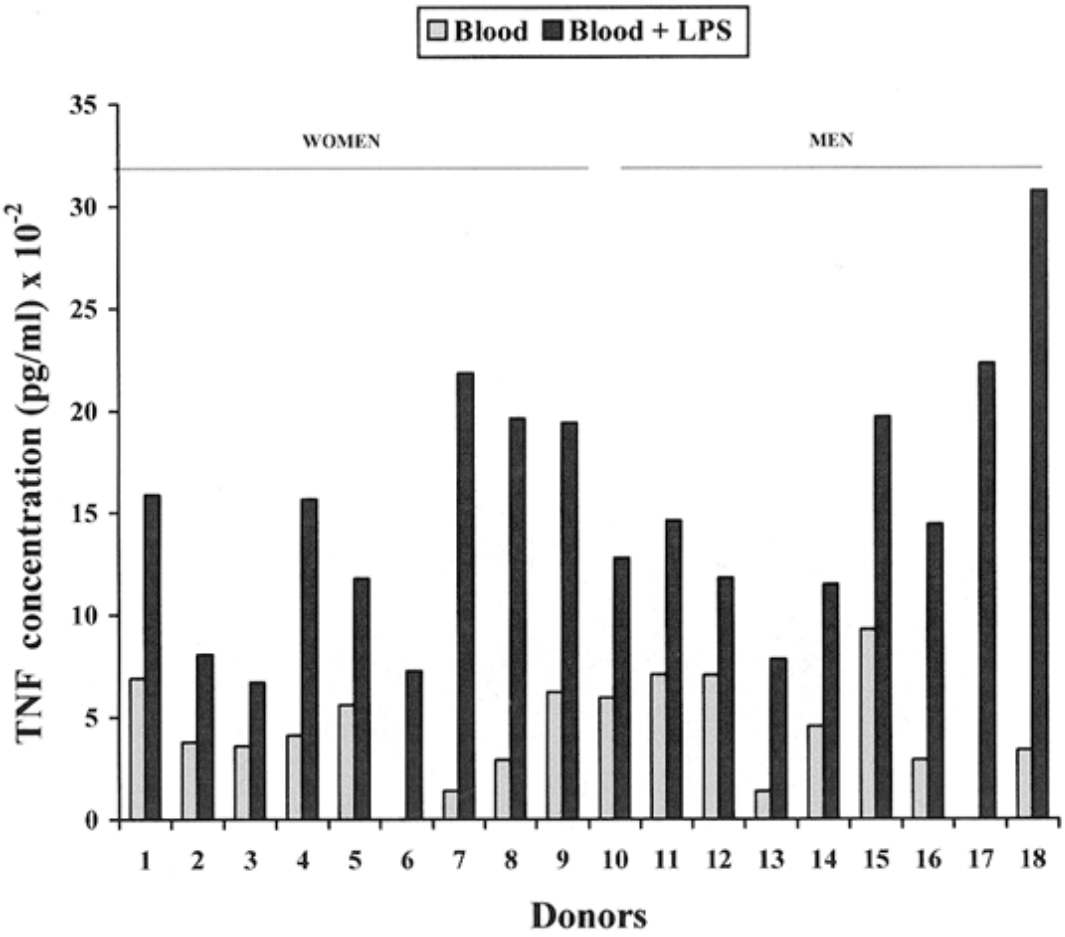


Figure 1

TNF levels in a whole human blood *ex vivo* culture are highly variable.

TNF levels in the *ex vivo* post-LPS stimulation and non-stimulation, measured in 18 individuals. Individuals 1-9, women; 10-18, men. Each value represents the average of nine observations, from one sample of blood. TNF was measured using a standard sandwich immunoradiometric assay.

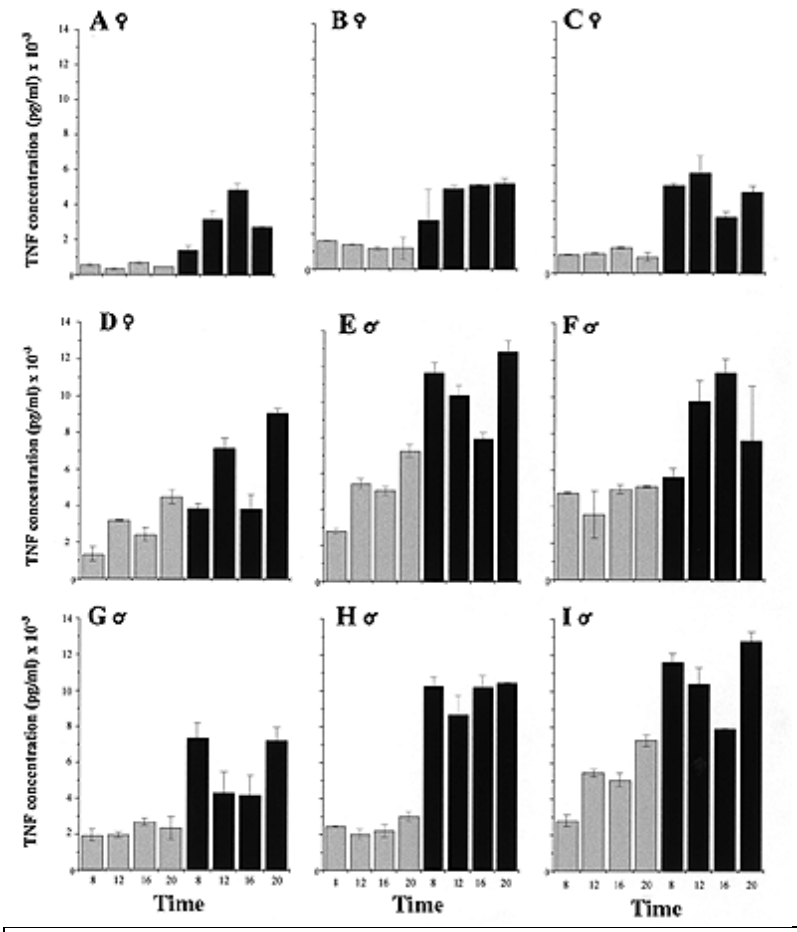


Figure 2

Individual TNF levels obtained in a whole blood *ex vivo* culture are relatively stable during the day.

The first and second set of bars indicates the presence and absence of LPS induction, respectively. Each group of 4 bars corresponds to samples obtained at 8:00, 12:00, 16:00, and 20:00 hours, respectively. Figures A-I represent results for women (A-D) and men (E-I). TNF levels are statistically higher ($p > 0.01$) in samples stimulated with LPS. No correlation ($p > 0.05$; $r = 0.69$; 34 degrees of freedom) between spontaneous and LPS-induced TNF levels was observed. A similar result was obtained when this comparison was applied to women and men ($p > 0.05$). However, LPS-induced TNF levels were higher ($p < 0.05$) in men. Each bar represents the

mean of two observations and standard deviations are shown with lines at the top.

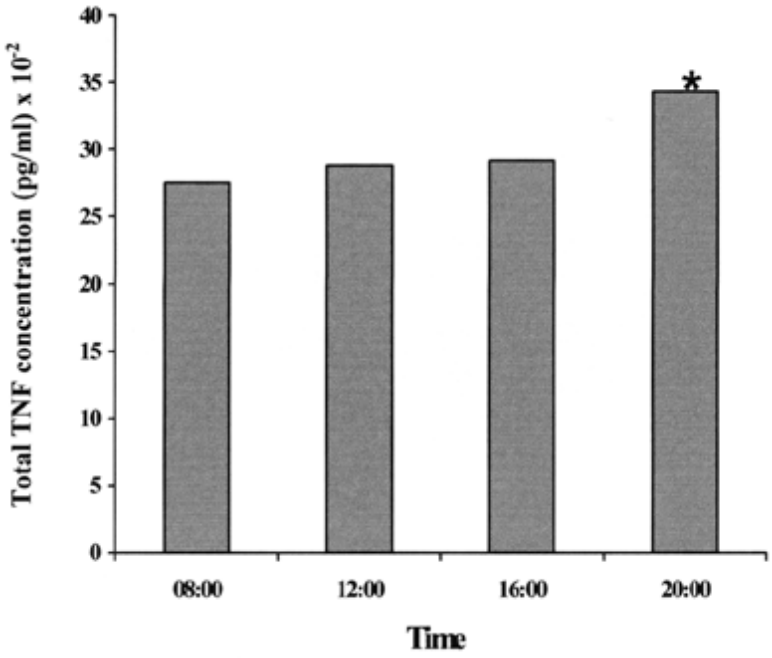


Figure 3

Total individual TNF levels increase at 20:00 hours.

Each bar represents both plasma and LPS-induced TNF for the times defined (8:00, 12:00, 16:00, and 20:00 hours). A significant difference ($p < 0.05$) for the TNF levels corresponding to the sample obtained at 20:00 hours was observed.

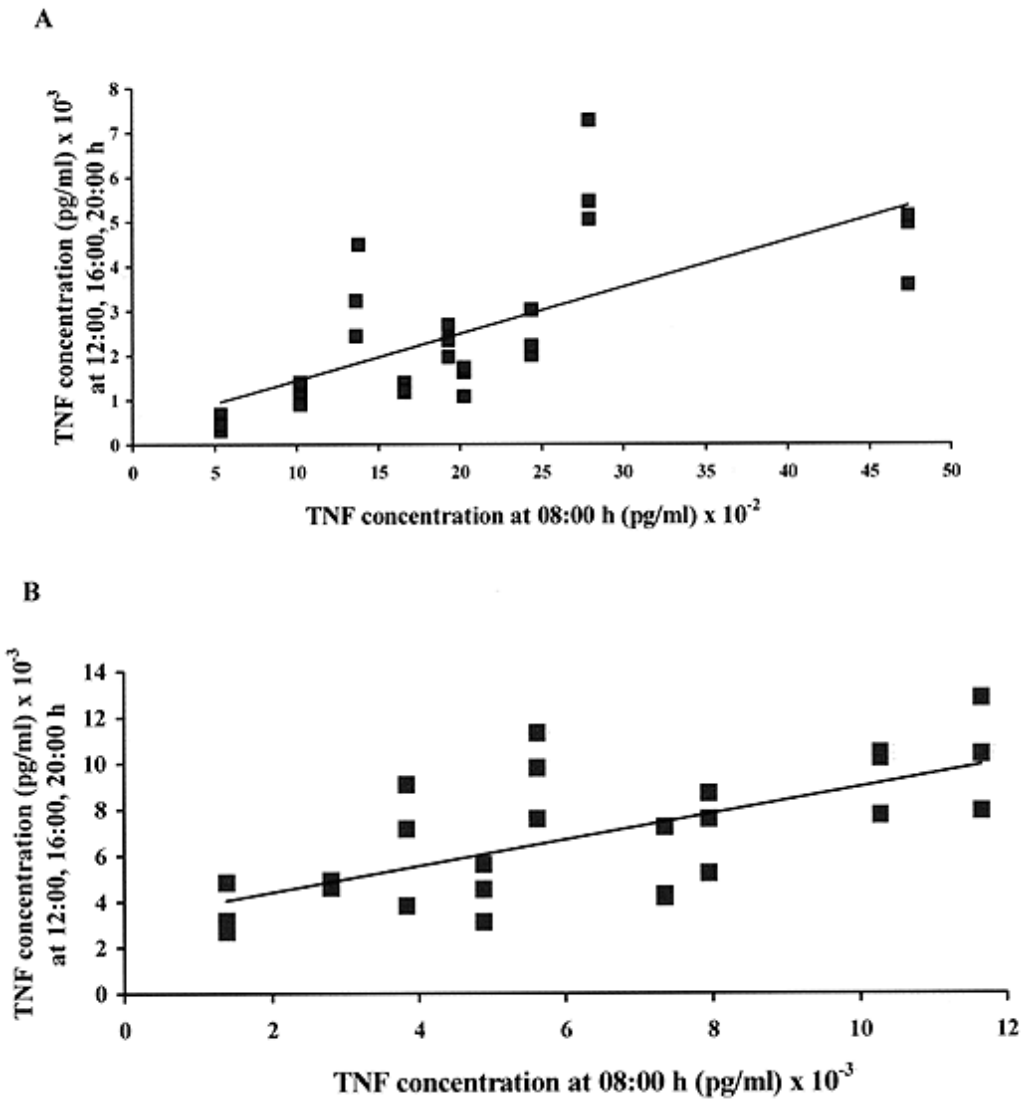


Figure 4

Correlation between *ex vivo* TNF levels at 8:00 hours and those obtained at 12:00, 16:00 and 20:00 hours, both basal (A) and LPS-induced (B).

The TNF levels determined at 08:00 hours were correlated, by linear regression (A: $r = 0.68$, $p < 0.01$; B: $r = 0.67$, $p < 0.01$; d.f. = 34, in both cases), with those obtained at other times.

Table 1
Correlation of individual *ex vivo* and plasma TNF levels, with the cortisol concentration

Observations were correlated by regression linear analysis. Data shown correspond to calculated r values. For significance degrees lower than 0.05 and 0.01, the probable r values (2 f.d.) are 0.95 and 0.99, respectively.

Volunteers	*Ex vivo* assay		Plasma
	LPS (–)	LPS (+)	
1	0.74	0.20	0.27
2	0.94	0.89	0.50
3	0.03	0.53	0.60
4	0.36	0.23	0.60
5	0.81	0.68	0.53
6	0.70	0.16	0.43
7	0.78	0.95	0.59
8	0.34	0.06	0.68
9	0.70	0.40	0.78

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In this issue

Regulation of interferon-g receptor (INF-gR) chains: a peculiar way to rule the life and death of human lymphocytes.

P. Bernabei, A. Allione, L. Rigamonti, M. Bosticardo, G. Losana, I. Borghi, G. Forni, F. Novelli

human, IFN-g, IFN-g receptor, signal transduction, cellular proliferation, apoptosis.

The interleukin-6 cytokine system in embryonic development, embryo-maternal interactions and

cardiogenesis.

P. Seiler, G. Plenz, M.C. Deng
interleukin-6, embryo, blastocyst, endometrium, heart, gp130.

Role of cytokines in the modulation of nitric oxide production by cyclic AMP.

Z. Zidek
cyclic AMP, prostaglandins, nitric oxide, interleukin-10, tumor necrosis factor-a, interferon-g.

The 3' untranslated region of tumor necrosis factor-a is highly conserved in idiopathic pulmonary fibrosis (IPF).

R.W. Freeburn, H. Kendall, L. Dobson, J. Egan, N.J. Simler, A.B. Millar
TNF-a, idiopathic pulmonary fibrosis, polymorphism.

Endogenous interferon-g impairs bacterial clearance from lungs during Pseudomonas aeruginosa pneumonia.

M.J. Schultz, A.W. Rijneveld, P. Speelman, S.J.H. van Deventer, T. van der Poll
interferon-g, cytokines, host defense, Pseudomonas aeruginosa, pneumonia.

Development of allergic contact dermatitis requires activation of both tumor necrosis factor-receptors.

F.M. Becke, T. Hehlgans, G. Brockhoff, D.N. Männel
allergic contact dermatitis, p75TNF receptor, p55TNF receptor, Langerhans cells, dendritic cells.

Transforming growth factor beta-1 and interferon-alpha in the aids dementia complex: possible relationship with cerebral viral load?

O. Perrella, P.B. Carreiri, A. Perrella, C. Sbreglia, F. Gorga, D. Guarnaccia, G. Tarantino
transforming growth factor-beta1, HIV-RNA, AIDS-dementia complex, interferon-alpha.

Interferon-beta (INF-b) antibodies in interferon-b1a- and interferon-b1b-treated multiple sclerosis patients. Prevalence, kinetics, cross-reactivity, and factors enhancing interferon-b immunogenicity in vivo.

P. Perini, A. Facchinetti, P. Bulian, A.R. Massaro, D. De Pascalis, A. Bertolotto, G. Biasi, P. Gallo

interferon-beta, natural autoantibodies, multiple sclerosis.

The promoter polymorphism of the interleukin-6 gene regulates interleukin-6 production in neonates but not in adults.

S. Kilpinen, J. Hulkkonen, X.Y. Wang, M. Hurme
interleukin-6, cord blood, gene polymorphism, inflammatory mediators.

The hepatitis C virus (HCV) induces a long-term increase in interleukin-10 production by human CD4+ T cells (H9).

O. Delpuech, D. Buffello-Le Guillou, E. Rubinstein, C. Féray, M.A. Petit
HCV-RNA, interleukin-10, H9 T cells, CD81, capsid mutation, protein kinase C.

Retrovirus-mediated gene transfer of the cytokine genes interleukin-1b and tumor necrosis factor-a into human neuroblastoma cells: consequences for cell line behavior and immunomodulatory properties.

C. Coze, T. Leimig, M.T. Jimeno, P. Mannoni
neuroblastoma, immunotherapy, gene therapy, cytokine, IL-1b, TNF-a.

Characterisation of cytokine mRNA expression in tumour-infiltrating mononuclear cells and tumour cells freshly isolated from human colorectal carcinomas.

A. Csiszar, T. Szentes, B. Haraszti, W. Zou, D. Emilie, G. Petranyi, E. Pocsik
colorectal cancer, tumour-infiltrating lymphocytes, cytokines, gene expression, RT-PCR.

Analysis of interleukin-18, interleukin-1 converting enzyme (ICE) and interleukin-18-related cytokines in Crohnís disease lesions.

F. Pagès, V. Lazar, A. Berger, C. Danel, S. Lebel-Binay, F. Zinzindohoué, P. Desreumaux, C. Cellier, N. Thiounn, D. Bellet, P.H. Cugnenc, W.H. Fridman
intestinal mucosa, Crohn's disease, interleukin-18, caspase-1/ICE, immunohistochemistry, reverse transcription polymerase chain reaction.

Daily production of human tumor necrosis factor in lipopolysaccharide (LPS)-stimulated ex vivo blood culture assays.

J.C. Aguillon, A. Escobar, V. Ferreira, A. Aguirre, L. Ferreira, M.C. Molina, A. Ferreira
TNF, cytokine, LPS.

Inducible nitric oxide synthase attenuates chronic colitis in human histocompatibility antigen HLA-B27/human b2 microglobulin transgenic rats.

H.S. Blanchard, E. Dernis-Labous, D. Lamarque, J. Tran van Nhieu, Z. Szepes, J.F. Fléjou, E. Wollman, B.J.R. Whittle, M. Breban
rodent, inflammation, nitric oxide, cytokines, in vivo animal models.

Up-regulation of mucin secretion in HT29-MTX cells by the pro-inflammatory cytokines tumor necrosis factor-a and interleukin-6.

M.G. Smirnova, S.L. Kiselev, J.P. Birchall, J.P. Pearson
MUC5AC, MUC5B, mucus-secreting cell line HT29-MTX.

A Jurkat T cell variant resistant to death receptor-induced apoptosis. Correlation with heat shock protein 27 and 70 levels.

J.E. Ricci, L. Maulon, V. Battaglione-Hofman, C. Bertolotto, F. Luciano, B. Mari, P. Hofman, P. Auberger
apoptosis, caspases, Hsp27, Hsp70, T lymphocytes.

Increased cell surface expression of histocompatibility class II IAd in c-FOS transfected clones of the P388D1 murine macrophage cell line.

Y. Le Garrec, E. Rouer, F. Conti, P. Michel, A. Pompidou
MHC class II antigens, murine macrophages, c-Fos.

Transforming growth factor-b inhibits interleukin-10 synthesis by human monocytic cells.

S. Suberville, A. Bellocq, I. Peguillet, O. Lantz, P. Stordeur, B. Fouqueray, L. Baud
transforming growth factor-b1, interleukin-10, U. 937 cells.

Evidence for tumor necrosis factor receptors (TNFRs) in human MRC5 fibroblast cells.

D. Derouich-Guergour, H. Pelloux, D. Aldebert, P. Demenge, P. Ambroise-Thomas
MRC5, fibroblasts, TNF receptor, RT-PCR, binding, flow cytometry.

Interleukin-9 induces 24P3 lipocalin gene expression in murine T cell lymphomas.

C. Orabona, L. Dumoutier, J.C. Renauld
cytokines, lipocalin, IL-9, 24P3.

Contrasting effect of interleukin-4 on the expression of cytosolic phospholipase A2, 5-lipoxygenase and 5-lipoxygenase-activating protein in peritoneal macrophages from control and ovalbumin-sensitized rats.

L. Escoubet-Lozach, C.F. M'Rini, A. Rey, M. Béraud, J.C. Lepert, J. Bernard, J.R. Mathieu, B. Pipy
cytokines, lipid mediators, macrophages, rat.

Fibrinogen cooperates with cytokines to induce interleukin-6 receptor mRNA expression in human hematopoietic CD34+ progenitors.

A. Hatzfeld, B. Panterne, N.O. Fortunel, M.N. Monier, J.A. Hatzfeld
human CD34+ cells, fibrinogen, competence factor, IL-6 receptor.

Balance between autocrine interleukin-1b and caspases defines life versus death of polymorphonuclear cells.

P. Bossù, E. del Grosso, M.P. Cesaroni, G. Maurizi, N. Balestro, A. Stoppacciaro, E. del Giudice, P. Ruggiero, D. Boraschi
apoptosis, interleukin-1, caspases, polymorphonuclear leukocytes, ICE.

Rat interleukin-10: production and characterisation of biologically active protein in a recombinant bacterial expression system.

V. Ball, S. Vigues, C.K. Gee, S. Poole, A.F. Bristow
IL-10, recombinant, rat, cytokines.

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