OPEN ACCESS Freely available online

SaBios: Rev. Saúde e Biol., v. 3, n.1 pp.33-39, Jul-Dez, 2008

Artigo de Revisão

NITRIC OXIDE IN THE IMMUNE RESPONSE MODULATION

Wander Rogério Pavanelli[1].

ABSTRACT

The signalling process by which nitric oxide (NO) acts in diverse cells is extremely complex and indirect. This process performs by generating reactive nitrogen oxide species that chemically modify enzymes, signalling proteins and transcription factors. Sometimes, immune interventions use as a strategic target the inducible nitric oxide synthase enzyme (iNOS) function that proceeds as a key mediator by inhibiting tissue damage observed in inflammatory diseases. In this way, excessive NO is necessary to limit destructive Th1 response and to favor Th2 response (immunomodulatory). In this case, the regulatory role of iNOS exceeds its cytotoxic function. Inhibition will exacerbate rather than suppress the disease. The role of NO might be different in early or late disease stages. For a given cell, the response to NO will depend on its reactivity state, microenvironment and tissue type. Therefore, Th1/Th2 balance influenced by NO has become normal apparent at a diverse population of immune cells. The two constitutively expressed isoforms (NOS-I and NOS-II), may also be up-regulated by immune response to release substantial amounts of NO. However, the contribution of these sources of NO production to immunoregulation in chronic immune responses remains to be shown. Thus, in vivo studies, mainly in humans, are necessary. In fact, new studies about mechanisms of action of NO in target molecules and cells are necessary for comprehending its role in infection and immunological diseases.

Palavras-chave: *nitric oxide (NO); immune response Th1/Th2; immunoregulation; nitric oxide synthase (NOS).*

THE IMPORTANCE OF OSTEOLOGICAL COLLECTIONS TO THE STUDY OF BIODIVERSITY

RESUMO

O processo de sinalização através dos qual o óxido nítrico (NO) atua em diversas células é extremamente complexo e indireto, agindo através da geração de espécie reativa de oxigênio, quimicamente modificando enzimas, sinalização de proteínas e fatores de transcrição. Algumas vezes, intervenções imune usam como estratégia-alvo a função da enzima óxido nítrico sintase induzível (iNOS), um mediador para inibir os danos provocados no tecido em doenças inflamatórias. Por esta via uma quantidade excessiva de NO é necessária para limitar o efeito destrutivo da resposta Th1 e favorecer a resposta Th2 (imunomoduladora). Neste caso o papel regulador da iNOS excede a função citotóxica, de forma que a inibição da iNOS pode exacerbar a supressão da doença. O papel do NO pode ser distinto nos estágios iniciais e tardios das doenças. Para uma determinada célula, a resposta do NO pode depender do estado de reatividade, do micro ambiente e tipo de tecido. Portanto o balanço entre Th1 e Th2 influenciado por NO tornou-se aparentemente normal em diversas células. As duas isoformas expressas constitutivamente (NOS-I e NOS-II) podem também serem reguladas pelas reposta imune para liberar NO. Contudo, a contribuição destas fontes de NO para a imunoregulação de resposta imune crônica permanece desconhecida. Desta forma, parece ser considerável o papel do NO em estudos in vivo, principalmente em humanos. De fato, são necessários novos estudos sobre o mecanismo de ação do NO em moléculas alvos e células para compreender seu papel nas infecções e doenças imunológicas.

Key words: *óxido nítrico (NO), resposta imune Th1/Th2, imunoregulação, óxido nítrico sintase (NOS).*

Nitric oxide (NO) is an important mediator of homeostatic processes. Host defense and changes in its generation or actions contribute to pathologic states. Nitric oxide discovery is not only important to our understanding of biology but it is also a base for development of new approaches for management and treatment of different diseases.

[1] Department of Biochemistry and Immunology, School of Medicine of Ribeirão Preto-Universidade de São Paulo, Ribeirão Preto, Brazil.

NO formation may have been originated as a first-line defense of metazoan cells against intracellular pathogens. It can be confirmed by the wide occurrence of enzyme responsible for NO production in these cells. NO-synthase (NOS) responses in several species ranging from invertebrates (Limulus polyphemus) (1) to insects (2,3), mammals, and non-vertebrates mammals (4-6). In mammals, NO response raises in response to infection by a wide gamma of intracellular pathogens such as bacteria, fungi, and parasites (6) (e.g. Leishmania spp. and T. cruzi). Transcription factors involved in NO synthesis in response to different stress situations seems to be related to evolutionary diversity.

Three isoforms of NO are described: neuronal (nNOS, NOS-I), endothelial (eNOS, NOS-III), and inducible (iNOS, NOS-II) (7,8). The first and second isoforms are constitutively expressed. However, NOS-I and NOS-III can be up-regulated to release substantial amounts of NO mainly in the central and peripheral nervous system and vascular endothelial cells, respectively. The inducible nitric oxide synthase (iNOS) is produced mainly in macrophages, but it also can be induced in a large variety of cells stimulated by cytokines and polysaccharides. NO biosynthesis is performed by one of NO synthases (NOS) in an oxidative reaction. This reaction mediates the incorporation of molecular oxygen (O2) into the unstable intermediate Nω-hydroxy-L-arginine, and subsequently into L-citrulline (9).

Data suggest that NO is involved in specific immunity, but its precise role is not yet clear. In addition, increasing evidence indicates that NO may act in acute and chronic inflammation. In fact, the treatment with inhibitors of NO syntase (NOS) reduces the inflammation in rats with acute inflammation or adjuvant arthritis (10-15). Tissue damage may be related to NO cytostatic or cytotoxic effects, not only for invading microorganisms but also for other cells. However, in some situations NO may interact with oxygen-derived radicals to generate molecules that could enhance its cytotoxicity. Reports suggest that inhibitors of NO synthesis and NO donors protect against some forms of injury, probably due to the dual nature of NO: cytotoxic and vasodilator (potentially protective) (16, 17).

NO has a multifaceted role in inflammatory reactions, ranging from improvement of vasodilatation and edema (by modulating sensory nerve endings and leukocyte activity) to tissue cytotoxicity (18-21). The damage to target cells by NO released from activated macrophages or endothelial cells has been confirmed in vitro. Necrotic and apoptotic pathways of cell death may be triggered by a high dose of NO (22-24).

The induction of apoptosis (programmed cell death) is important in regulation of T cell maturation in thymus as well as T cell growth in periphery. It seems that NO may regulate the apoptosis pathway. Researches have shown that low concentrations of NO protect cells from apoptosis by inactivating CPP32-like protease and by increasing Bcl2 protein expression (25). High doses of NO induce apoptosis in thymocyte as well as in splenic T cells (26). Low doses of NO protect thymocyte apoptosis induced by anti-CD3. The anti-apoptotic or pro-apoptotic effects of NO probably involve interaction of reactive-oxygen intermediates. These effects are also dependent on the redox state of cell (24, 27).

Interestingly, Th1 cells are more susceptible to apoptosis than Th2 cells. NO may regulates the Th1/Th2 balance by promoting or suppressing apoptosis at high/low doses (28). The cytoprotective properties of low/intermediate levels of NO might limit tissue damage during inflammation, independent of attenuating Th1 responses (24; 29-31).

NO down regulates the expression of selectins (P and E), vascular cell adhesion molecule, and intracellular adhesion molecule-1 (ICAM-1). These processes result in suppression of binding to respective ligands on the vessel wall (32,33). Consequently, rolling of leukocytes in endothelium is inhibited as well as migration of cells from vessels to the tissues. Recent studies have suggested that P and E-selectin mediate recruitment of Th1 (but not Th2) cells into inflamed tissues (34, 35). Due to P-selectin expression was found to be down regulated in the presence of NO, it is clear that NO preferentially down regulates the accumulation of Th1 cells at sites of chronic inflammation by interfering with the adhesion process (36, 37).

It has been demonstrated that concanavalin-A induces iNOS expression in macrophages and, consequently, the NO production decreases mitochondrial function and DNA synthesis in T cells. Thus, cell proliferation in certain 'low responder' rodents is suppressed (38). Recent studies invalidate

the concept of an exclusive nonspecific cytostatic effect of NO (39). Rather, specific impairment of Th1 cell was observed, while the Th2 cell function appeared to be unaltered. This also agreed with concomitant observation of suppressed IL-2 and IFN-γ (40).

In murine lymphocytes, the target for NO action is the IL-2 gene. Exposure to NO suppresses IL-2 gene expression at transcription level, consequently modulating the Th1/Th2 balance by favoring the Th2 response (40). Exogenous IL-2 can reverse the suppressive effect of NO on Th1 cells. In humans, NO may limit Th1 cell activity by supporting down regulatory IL-4 production. Recently, it has been shown that at high concentrations, NO inhibits IL-12 synthesis by activated macrophages. In this way, expansion of Th1 cells is indirectly suppressed (41). At low concentrations, NO selectively enhances the induction of Th1 cells and it has no effect on Th2 cells (30).

Various functions of human phagocytes are modulated by NO. In macrophages, NO induces transcription of IL-12 p40 gene but not of human IL-12 p35 gene (42). Due to IL-12 (p40) homodimer is an antagonist of IL-12; this might be a further indication for a fewer Th1 reactivity in the presence of NO (43). Similarly, it has been reported that iNOS expression contributes to desensitization of macrophages observed after exposure to a low concentration of lipopolysaccharide. NO inhibits the expression of major histocompatibility complex class II (MHC-II).

At low concentrations, NO selectively enhances the induction of Th1 cells and has no effect on the Th2 cells (40). NO exerts this effect in synergy with IL-12 during Th1 cell differentiation and has no effect on fully committed Th1 cells (30). In addition, CD4+ T cells seem to be directly affected by NO. This suggests an additional pathway by which NO may modulate the immune response (41). The iNOS activity has been found also to regulate chemokine production. Therefore, NO might serve to limit the extent of potentially dangerous local cellular immune responses. INOS is also expressed during chronic asthma, and it has been suggested that NO supports a Th2 partiality of immune reactivity in the lung (44, 45) limiting the damage to tissue provoked by exacerbated immune response.

NO may induce expression of Th2 cytokine IL-4 and IL-5, while Th1 cytokines, IFN-γ and IL-2 are suppressed. Moreover, the apoptosis-inducing activity of NO also affects Th1 cells. Thus, apoptosis is more common in Th1 cells than Th2 cells (40). The proposed role of iNOS is in accord with the observation that mice with a disrupted iNOS gene exhibit enhanced Th1 activity. It is important to note that this concept may be also applied to human immune system (46)..

There is now, sufficient evidence for NO production via iNOS enzyme activity in human tissue during inflammation. Smaller amounts of NO might be released in humans, and human cells are more resistant to the cytotoxic effects of NO. Thus, the cytotoxic action of NO towards autologous human immune or tissue cells might be less relevant when compared with its regulatory effect. In humans, the cytotoxic potential of NO is linked to the formation of peroxynitrite, which only occurs at the sites of simultaneous superoxide formation, such as in phagocytes.

Signaling processes by which NO acts to regulate cells are extremely complex. These indirect processes occur through generation of reactive nitrogen oxide species that chemically modify enzymes, signaling proteins and transcription factors. Sometimes, immune intervention strategies target iNOS as a key mediator for tissue damage in inflammatory diseases. Approaches of this type take into account that NO also serves to limit destructive Th1 responses. In those cases where the regulatory role of iNOS exceeds its cytotoxic function, inhibition of iNOS will exacerbate rather than suppress the disease. The role of NO might be different in early or late disease stages. For a given cell, the response to NO will depend on its reactivity state, the microenvironment and its tissue type.

Therefore, differences in Th1/Th2 balance promoted by NO will become more apparent at immune cell populations rather than at single clones level. The two constitutively expressed isoforms, NOS-I and NOS-II, may also be upregulated to release substantial amounts of NO. However, the contribution of these sources of NO production to immunoregulation in chronic immune responses remains to be shown. Thus, considerable gaps about the role of NO in vivo, particularly in humans, need to be explained. Future directions will focus on molecular action mechanisms of NO, its target molecules and cells and its role in infection and immunological mediated diseases.

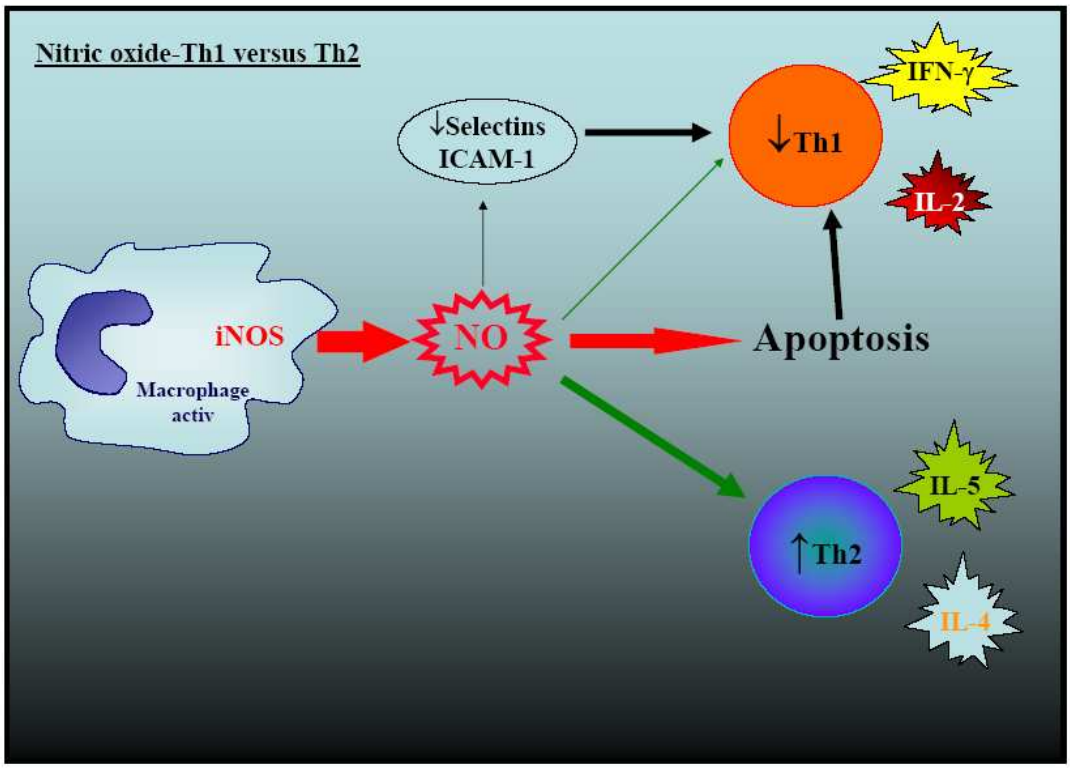

Figura.1 – Immunoregulation by nitric oxide. Activated cells, among them, Macrophages, produce Nitric oxide (NO) for the induction of Nitric oxide synthase enzyme (iNOs). The NO shows some important function: inhibition of molecular adhesion expression (selectins and ICAM-1), that reduces the migration of cells from Th-1 profile to infection site. Moreover, NO may directly inhibit these cells promoting a reduction in production of cytokines as IFN-□ and IL-2. Inflammatory mediator induces apoptosis (cellular death) in Th1 cells. In addition, NO may induces the differentiation of immune response for Th2 profile and the synthesis of cytokines as IL-4 and IL-5.

Wander Rogério Pavanelli
Endereço para correspondência: School of Medicine;
Av. Bandeirantes, 3900 –
14049-900
Ribeirão Preto, SP, Brazil
Tel. 55-16-3602-3231;
Fax: 55-16-3633-6840
E-mail: wanderpavanelli@usp.br

Recebido em 10/02/08
Aceito em 30/09/08

REFERENCES

(1) NATHAN, C.; XIE, Q.W. Regulation of biosynthesis of Nitric oxide. **Journal Biological Chemistry**. 1994, v.13 269, p. 13725-8.

(2) REGULSKI, M.; TULLY, T. Molecular and biochemical characterization of dNOS: a Drosophila Ca2+/calmodulin-dependent Nitric oxide synthase. **Proceedings of the National Academy of Sciences-USA**. 1995, v. 26, p. 9072-6

(3) RIBEIRO, J.M.; HAZZARD, J.M.; NUSSENZVEIG, R.H.; CHAMPAGNE, D.E.; WALKER, F.A. Reversible binding of Nitric oxide by a salivary heme protein from a blood sucking insect. **Science**, 1993, v. 260, p. 539-41.

(4) SUNG, Y.J.; HOTCHKISS, J.H.; AUSTIC, R.E.; DIETERT, R.R. L-arginine-dependent production of reactive nitrogen intermediate by macrophages of a uricotelic species. **Journal Leukocyte Biology**. 1991, v. 50 (1), p.49-56.

(5) LIN, A.W.; CHANG, C.C.; MCCORMICK, C.C. Molecular cloning and expression of an avian macrophage nitric-oxide synthase cDNA and the analysis of the genomic 5'-flanking region. **Biological Chemistry**. 1996, v. 271(20), p. 11911-9.

(6) HOLMQVIST, B.I.; OSTHOLM, T.; ALM, P.; EKSTROM, P. Nitric oxide synthase in the brain of a teleost. Neurological letters, 1994, v. 25; 171(1-2), p. 205-7. LIEW, F.Y.; WEI, X.Q.; PROUDFOOT, L. Cytokines and Nitric oxide as effector molecules against parasitic infections. **Philosophical Royal Society of London**, 1997, v. 29; 352(1359), p.1311-5.

(8) ALDERTON, W.K.; COOPER, C.E.; KNOWLES, R.G. Nitric oxide synthases: structure, function and inhibition. **Journal Biochemistry**, 2001, v. 1;357, p. 593-615.

(9) SALERNO, J.C.; FREY, C.; MCMILLAN, K.; WILLIAMS, R.F.; MASTERS, B.S.; GRIFFITH, O.W. Characterization by electron paramagnetic resonance of the interactions of L-arginine and L-thiocitrulline with the heme cofactor region of Nitric oxide synthase, **Journal Biological Chemistry**, 1995, v. 270, p.27423-27428.

(10) MCCALL, T.B.; BOUGHTON-SMITH, N.K.; PALMER, R.M.; WHITTLE, B.J.; MONCADA, S. Synthesis of Nitric oxide from L-arginine by neutrophils. Release and interaction with superoxide anion. **Journal Biochemistry**, 1989, v.261, p. 293-296.

(11) SALTER, M.; KNOWLES, R.G.; MONCADA, S: Widespread tissue distribution, species distribution and changes in activity of Ca(2+)-dependent and Ca(2+)-independent Nitric oxide synthases. **FEBS Lett**, 1991, v.291, p.145-149.

(12) OLKEN, N.M.; OSAWA, Y.; MARLETTA, M.A. Characterization of the inactivation of Nitric oxide synthase by NG-methyl-L-arginine: evidence for heme loss, **Biochemistry**, 1994, v. 33, p.14784-14791.

(13) HICKEY, M.J.; SHARKEY, K.A.; SIHOTA, E.G.; REINHARDT, P.H.; MACMICKING, J.D.; NATHAN, C.; KUBES, P. Inducible Nitric oxide synthase-deficient mice have enhanced

leukocyte-endothelium interactions in endotoxemia, **Faseb Journal**, 1997, v.11, p. 955-964.

(14) ICHIMORI, K.; STUEHR, D.J.; ATKINSON, R.N.; KING, S.B. Synthesis and evaluation of new sulfur-containing L-arginine-derived inhibitors of Nitric oxide synthase, **Journal Medicine Chemistry**, 1999, v. 42, p.1842-1848.

(15) STEFANOVIC-RACIC, M.; MEYERS, K.; MESCHTER, C.; COFFEY, J.W.; HOFFMAN, R.A.; EVANS, C.H. Comparison of the Nitric oxide synthase inhibitors methylarginine and aminoguanidine as prophylactic and therapeutic agents in rat adjuvant arthritis, **Journal Rheumatology**, 1995, v. 22, p. 1922-1928.

(16) CIFONE, M.G.; CIRONI, L.; MECCIA, M.A.; RONCAIOLI, P.; FESTUCCIA, C.; DE NUNTIIS, G.; D'ALO, S; SANTONI, A. Role of Nitric oxide in cell-mediated tumor cytotoxicity, **Adv. Neuroimmunol**, 1995, v.5, p.443-461.

(17) KRONCKE, K.D.; SUSCHEK, C.V.; KOLB-BACHOFEN, V. Implications of inducible Nitric oxide synthase expression and enzyme activity. **Antioxidante Redox Signal**, 2000, v. 2, p. 585-605.

(18) LASKIN, D.L.; HECK, D.E.; LASKIN, J.D. Role of inflammatory cytokines and Nitric oxide in hepatic and pulmonary toxicity. **Toxicological Letters**, 1998, v. 102-103, p. 289-293.

(19) GRIFFITHS, M.J.; LIU, S.; CURZEN, N.P.; MESSENT, M.; EVANS, T.W. In vivo treatment with endotoxin induces Nitric oxide synthase in rat main pulmonary artery. **American Journal Physiology**, 1995, v. 268, p. L509-518.

(20) BOGDAN, C. Of microbes, macrophages and Nitric oxide. **Behring**. Inst Mitt, 1997, p. 58-72.

(21) CHRISTOPHERSON, W.M. The significance of the pathologic findings in endometrial cancer. **Clinical Obstetrics Gynecologic**, 1986, v. 13, p.673-693.

(22) JOHANN, A.M.; BARRA, V.; KUHN, A.M.; WEIGERT, A.; VON KNETHEN, A.; BRUNE. B. Apoptotic cells induce arginase II in macrophages, thereby attenuating NO production. **Faseb Journal**, 2007, v. 21, p. 2704-2712.

(23) SHIBATA, T.; NAGATA, K.; KOBAYASHI, Y. Cutting edge: a critical role of nitrogen oxide in preventing inflammation upon apoptotic cell clearance. **Journal Immunology**, 2007, v.179, p.3407-3411.

(24) OKUDA, Y.; SAKODA, S.; SHIMAOKA, M.; YANAGIHARA, T. Nitric oxide induces apoptosis in mouse splenic T lymphocytes. **Immunology Letters**, 1996, v.52, p.135-138.

(25) NISHIO, E.; WATANABE, Y. No induced apoptosis accompanying the change of oncoprotein expression and the activation of CPP32 protease. **Life Science**, 1998, v.62, p. 239-245.

(26) OGURA, T.; TATEMICHI, M.; ESUMI, H. Nitric oxide inhibits CPP32-like activity under redox regulation. **Biochemistry Biophysics Research Communication**, 1997, v. 236, p.365-369.

(27) VON KNETHEN, A.; CALLSEN, D.; BRUNE, B. NF-kappaB and AP-1 activation by Nitric oxide attenuated apoptotic cell death in RAW 264.7 macrophage. **Molecular Biological Cell**, 1999, v.10, p.361-372.

(28) XIAO, B.G.; MA, C.G.; XU L.Y.; LINK, H.; LU, C.Z. IL-12/IFN-gamma/NO axis plays critical role in development of Th1-mediated experimental autoimmune encephalomyelitis. **Molecular Immunology**, 2008, v.45, p.1191-1196.

(29) CATTELL, V.; JANSEN, A. Inducible Nitric oxide synthase in inflammation. **Histochemistry Journal**, 1995, v.27, p.777-784.

(30) NIEDBALA, W.; WEI, X.Q.; PIEDRAFITA, D.; XU, D.; LIEW, F.Y. Effects of Nitric oxide on the induction and differentiation of Th1 cells. **European Journal Immunology**, 1999, v.29,p.2498-2505.

(31) DE GOUW, H.W.; MARSHALL-PARTRIDGE, S.J.; VAN DER, V. H.; VAN DEN, A.J.G.; HIEMSTRA, P.S.; STERK, P.J. Role of Nitric oxide in the airway response to

exercise in healthy and asthmatic subjects. **Journal Applicative Physiology**, 2001, v.90, p.586-592.

(32) GRIES, A.; BODE, C.; PETER, K.; HERR, A.; BOHRER, H.; MOTSCH, J.; MARTIN, E. Inhaled Nitric oxide inhibits human platelet aggregation, P-selectin expression, and fibrinogen binding in vitro and in vivo. **Circulation**, 1998, v.97, p. 1481-1487.

(33) ARMSTEAD, V.E.; MINCHENKO, A.G.; SCHUHL, R.A.; HAYWARD, R.; NOSSULI, T.O.; LEFER, A.M. Regulation of P-selectin expression in human endothelial cells by Nitric oxide. **American Journal Physiology**, 1997, v.273, p.740-746.

(34) MANGAN, P.R.; O'QUINN, D.; HARRINGTON, L.; BONDER, C.S.; KUBES, P.; KUCIK, D.F.; BULLARD, D.C.; WEAVER, C.T. Both Th1 and Th2 cells require P-selectin glycoprotein ligand-1 for optimal rolling on inflamed endothelium. **American Journal Pathology**, 2005, v.167, p.1661-1675.

(35) BONDER, C.S.; NORMAN, M.U.; SWAIN, M.G.; ZBYTNUIK, L.D.; YAMANOUCHI, J.; SANTAMARIA, P.; AJUEBOR, M.; SALMI, M.; JALKANEN, S.; KUBES, P. Rules of recruitment for Th1 and Th2 lymphocytes in inflamed liver: a role for alpha-4 integrin and vascular adhesion protein-1, **Immunity**, 2005, v.23, p.153-163.

(36) IGNARRO, L.J.; BYRNS, R.E.; WOOD, K.S. Endothelium-dependent modulation of cGMP levels and intrinsic smooth muscle tone in isolated bovine intrapulmonary artery and vein, **Circulation Research**, 1987, v.60, p.82-92.

(37) PALMER, R.M.; FERRIGE, A.G.; MONCADA, S. Nitric oxide release accounts for the biological activity of endothelium-derived relaxing factor. **Nature**, 1987, v. 327, p. 524-526.

(38) SASS, G.; KOERBER, K.; BANG, R.; GUEHRING, H.; TIEGS, G. Inducible Nitric oxide synthase is critical for immune-mediated liver injury in mice. **Journal Clinical Investigation**, 2001, v.107, p.439-447.

(39) LEWIS, E.; ROGACHEV, B.; SHAKED, G.; DOUVDEVANI, A. The in vitro effects of ketamine at large concentrations can be attributed to a nonspecific cytostatic effect. **Anesthesiology Analgesic**, 2001, v.92, p.927-929.

(40) TAYLOR-ROBINSON, A.W.; LIEW, F.Y.; SEVERN, A.; XU, D.; MCSORLEY, S.J.; GARSIDE, P.; PADRON, J.; PHILLIPS, R.S. Regulation of the immune response by nitric oxide differentially produced by T helper type 1 and T helper type 2 cells. **European Journal Immunology**, 1994, v.24, p.980-984.

(41) VAN DER VEEN, R.C. Nitric oxide and T helper cell immunity. **International Immunopharmacol**, 2001, v.1, p. 1491-1500.

(42) SALVUCCI, O.; KOLB, J.P.; DUGAS, B.; DUGAS, N.; CHOUAIB, S. The induction of nitric oxide by interleukin-12 and tumor necrosis factor-alpha in human natural killer cells: relationship with the regulation of lytic activity. **Blood**, 1998, v. 92, p. 2093-2102.

(43) PAHAN, K.; SHEIKH, F.G.; LIU, X.; HILGER, S.; MCKINNEY, M; PETRO, T.M. Induction of nitric-oxide synthase and activation of NF-kappaB by interleukin-12 p40 in microglial cells. **Journal Biological Chemistry**, 2001, v.276, p.7899-7905.

(44) KRONCKE, K.D.; FEHSEL, K.; SUSCHEK, C.; KOLB-BACHOFEN, V. Inducible nitric oxide synthase-derived nitric oxide in gene regulation, cell death and cell survival. **International Immunopharmacologic**, 2001, v.1, p.1407-1420.

(45) KRONCKE, K.D.; FEHSEL, K.; KOLB-BACHOFEN, V. Inducible nitric oxide synthase in human diseases. **Clinical Experimental Immunology**, 1998, v.113, p.147-156.

(46) SINGH, V.K.; MEHROTRA, S.; NARAYAN, P.; PANDEY, C.M.; AGARWAL, S.S. Modulation of autoimmune diseases by nitric oxide. **Immunology Research**, 2000, p.22:1-19.