PIVOTAL ROLES OF GLUCOCORTICOIDS IN THE PATHOGENESIS AND TREATMENT OF INFLAMMATORY RHEUMATIC DISEASES

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ABSTRACT

The glucocorticoid-mediated response plays a pivotal role in the modulation of immune and inflammatory processes. Deregulations at these levels have been implicated in a variety of disease conditions: excessive concentration of glucocorticoids may enhance the susceptibility to or severity of infectious diseases, whereas insufficient concentrations are associated with increased susceptibility and activity of inflammatory diseases.

Due to their immunosuppressive effects, glucocorticoids have been used for more than 50 years in the treatment of multiple inflammatory diseases. Particularly in rheumatoid arthritis (RA) glucocorticoid therapy has been proved to be very effective in ameliorating objective inflammatory markers, improving the general well being of patients and decreasing joint damage.

This review explores the mechanisms beneath the positive therapeutic impact of glucocorticoids in immune–mediated rheumatic diseases, with emphasis in RA.

Key-Words: Glucocorticoids; Rheumatoid Arthritis; Inflammation; Neurondocrine System.

RESUMO

A resposta mediada por glicocorticóides desempenha um papel fundamental na modulação dos processos imunológicos e inflamatórios. Perturbações nesta resposta foram implicadas numa variedade de doenças: concentrações excessivas de glicocorticóides podem afectar a susceptibilidade a doenças infecciosas, bem como a sua gravidade; concentrações insuficientes estão associadas a um aumento da susceptibilidade e actividade de doenças inflamatórias.

Devido aos seus efeitos imunosupressores, os glicocorticóides são usados há mais de 50 anos no tratamento de múltiplas doenças inflamatórias. Especificamente na terapêutica da artrite reumatóide, têm demonstrado a sua eficácia na melhoria da actividade inflamatória, no bem-estar dos doentes e ainda na diminuição da destruição articular.

Este artigo de revisão aborda os mecanismos que estão subjacentes a este impacto terapêutico positivo dos glicocorticóides em doenças reumáticas de base imunológica, com destaque para a artrite reumatóide.

Palavras-Chave: Glicocorticóides; Artrite Reumatóide; Inflamação; Sistema Neuroendócrino.

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Introduction

Glucocorticoids are very effective in the treatment of a variety of inflammatory and auto-immune conditions, including rheumatoid arthritis (RA). Not only do they improve clinical and biological markers of inflammation but they also contribute for a subjective feeling of well being in these patients. The authors review the mechanisms beneath these effects, highlighting the classic anti-inflammatory role and exploring the potential role of glucocorticoids as hormone replacement therapy in the context of a defective hypothalamus-pituitary-adrenal (HPA) axis in these conditions.

Inflammation and the neuroendocrine system

In the most recent theoretical models of inflammatory rheumatic diseases, it is proposed that multiple genetic and environmental factors operate in susceptible individuals, inducing chronic physiologic perturbations of homeostatic processes that eventually lead to the typical features of these disease states[1-3]. Following this model, identification of risk factors and normalization of the physiologic imbalances during the premorbid phase should allow to delay or even abort further development of the disease process[1]. Neuroendocrine-immune interactions play a central role in the modulation of the immune system and may, thus, be placed at the very core of the regulatory mechanisms able to promote or prevent chronic inflammatory diseases[4,5]. That is why it is so important to understand them and to face the remarkable difficulties in unravelling their complex and intricate nature.

Local inflammatory responses involve an enormous variety of substances, including cytokines, prostaglandins, free radicals and locally produced hormones. The cytokines interleukin 1β (IL-1β), IL-6 and tumour necrosis factor α (TNF-α) induce the production of chemotactic factors, responsible for further recruitment of inflammatory cells from the blood stream. This process is highly facilitated by the increased expression of adhesion molecules and selectines in endothelial cells, induced by the same cytokines.[6,7] Furthermore, IL-1, IL-6 and TNF-α start a cascade phenomenon amplified by a second wave of pro-inflammatory cytokines, such as IL-8, IL-12, IL-15, IL-17, IL-18, interferon γ (IFN-γ) and IFN-β.

T-helper (Th) cells play a major role in the amplification of the immune response, presenting a clear polarization between Th1 and Th2 types, traduced in differential patterns of cytokine expression. Th1 cells are involved in the cellular immune responses producing mainly pro-inflammatory cytokines such as IL-2 and IFN-γ, whereas Th2 cells are believed to ensure protection against extracellular pathogens through a humoral response, with the secretion of anti-inflammatory cytokines such as IL-4, IL-10, IL-11 and IL-13.[8,9]

Neuropeptides and hormones can modulate the activity of immune cells. Prolactin (PRL), corticotrophin-releasing hormone (CRH), arginine vasopressin (AVP) and substance P (SP)[5,10-14] are all produced and released at the inflammatory site where they have been demonstrated to play a potent pro-inflammatory role. Pro-inflammatory effects of local CRH have been similar, in potency, to those of TNF-α. Other neuropeptides and hormones, like adrenocorticotrophic hormone (ACTH), β-endorfin, somatostatin and α-melanocyte-stimula-

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ting hormone (α-MSH) seem to counterregulate these effects, exerting an anti-inflammatory influence.[15]

Conversely, IL-1β, TNF-α and IL-6 released from the inflammatory site, activate the production of α-MSH, CRH and AVP in the hypothalamus, resulting in the release of ACTH by the pituitary gland,[16] and finally increased cortisol production in the adrenal cortex.[17]

Anti-inflammatory role of glucocorticoids

Glucocorticoids exert potent anti-inflammatory effects at virtually every step of the inflammatory and immune process, suppressing cell adhesion, margination and migration, macrophage activation, antigen presentation, T cell receptor expression, T lymphocyte activation, proliferation, differentiation and mature cell function, including cytotoxicity, and B cell function including antibody production. They suppress cell-mediated immune responses in several ways, with emphasis on the inhibition of IL-2 and IL-2 receptor production, which leads to diminished lymphocyte proliferation and activation.[18] The production of almost all pro-inflammatory cytokines (e.g. IL-1β, IL-2, IL-6, IL-8 and TNF-α) is inhibited by glucocorticoids.[19-22] Conversely, glucocorticoids enhance the production of anti-inflammatory cytokines, such as IL-4, IL-5, IL-10, IL-13, and transforming growth factor β (TGF-β).[23] Finally, glucocorticoids lead to impaired synthesis of collagenase, elastase and tissue plasminogen activator thus reducing the degradation of cartilage, soft tissue and bone.[24]

Glucocorticoids achieve these effects through a variety of molecular mechanisms, including gene expression, transcription, translation, post-translational processing, protein secretion and cell progenitor proliferation and differentiation.[25,26] For example, they directly decrease transcription of the IL-6 and IL-1β genes, decreasing their production by immune cells. Other immunosuppressive effects are indirectly mediated through inhibition of pro-inflammatory transcription factors such as NF-kB and AP-1.[27] These factors promote the expression of genes for many pro-inflammatory cytokines, enzymes and adhesion molecules in inflammatory diseases. Conversely, cytokines such as TNF-α and IL-1β further activate NF-kB, leading to perpetuation of local inflammatory responses.[28]

At first, glucocorticoids were thought to have only immunosuppressive effects.[29,30] This still holds true for most pharmacologic effects. However, recent studies indicate that, at physiological concentrations, glucocorticoids have an immunomodulatory, rather than immunosuppressive effect, causing a shift from a Th1 to a Th2 cytokine pattern.[31-33] DeRijk *et al* have demonstrated that physiological changes in circulating glucocorticoid levels are associated with shifts in patterns of immune cytokine production.[34,35] Glucocorticoids were found to be especially efficient in diminishing the circulating levels of TNF-α, followed by IL-1β. IL-6 levels seem to be less responsive to these hormones.[35]

The entire synthetic enzyme machinery for glucocorticoid production has been identified in the thymus, as have glucocorticoid hormones and their precursors.[36] Studies have shown that these glucocorticoids play an important role in thymic T cell selection: depending on the concentration of glucocorticoids produced relative to the concentration of antigen, T cells undergo negative or positive selection, that is to say, they may be shunted to the death pathway and die by apoptosis or they may undergo clonal proliferation.

The increased production of corticosteroids constitutes an integral part of the systemic response to inflammation and a crucial step in preventing the progression of uncontrolled immune response or inflammation .[1]

Altogether, these neuroendocrine-immune loops play a pivotal role in the adequate modulation of the inflammatory response. Significant anomalies in these interactions may have a decisive contribution to the generation and progression of inflammatory rheumatic diseases.[15]

Dysfunction of the hypothalamus-pituitary-adrenal axis in rheumatoid arthritis?

Data generated from studies in animal models in the late 80´s led to the exciting hypothesis that RA and other human inflammatory and autoimmune diseases could be due to a blunted HPA axis response to immune challenge, leading to unchecked damaging inflammation. A series of elegant experiments by researchers from the NIH clearly demonstrated that the inherent high susceptibility for auto-immune disease of the Lewis rats (LEW/N),[37] is due to a defect in CRH production by

the hypothalamus, leading to an impaired corticosterone response to a variety of immune challenges.[38] The susceptibility in Lewis rats can be reversed by administration of glucocorticoids within the physiological range. Conversely the histocompatible Fischer rats, which are resistant to a variety of experimental auto-immune diseases can be made susceptible through glucocorticoid receptor blockade.[37] Interestingly, blunted plasma corticosterone responses were also described in lupus-prone mice, prior to the development of clinical symptoms.[39] Several studies in animal models also demonstrate that interrupting HPA axis adaptative response increases disease severity in animal models of arthritis[40] and mortality from septic shock.[41-43] Overall, serious disruption of the HPA axis at any level, whether in a genetic basis, through surgical means such as adrenalectomy or hypophysectomy, or with pharmacological interventions such as the treatment with the glucocorticoid receptor antagonist RU 486, can render an inflammatory resistant host into a susceptible one.[4,44] Conversely, transplantation of fetal hypothalamic tissue from inflammatory resistant Fisher rats into the third ventricle of inflammatory susceptible Lewis rats corrects the Lewis blunted HPA axis responses to bacterial lipopolysaccharide (LPS) and reverses its susceptibility to auto-immune disease.[45] Predisposition for autoimmune/inflammatory diseases has also been associated with abnormal glucocorticoid receptor[46] in experimental animal studies.[47]

A considerable effort has been employed in the search for evidence supporting the existence of a defective HPA axis in RA patients. Despite some inconsistency,[48-50] studies in untreated RA patients have mostly shown an association of normal serum and 24-hour urine cortisol with high ACTH serum levels. These results suggest a defective adrenal gland function.[51] A number of other studies have indicated that untreated individuals with RA have hypersecretion of ACTH without increased levels of cortisol, again suggesting a normal central response to inflammation, but impaired adrenal function.[51,52,53] In addition, there is evidence suggesting that RA patients have a loss of the normal circadian cortisol variance,[54] which is in the lower limit of normality during the day and does not increase in response to stress (surgery, for example).[55] Overall, these observations suggest a defect in the response to ACTH, again placing the HPA axis in the center of potentially crucial pathogenetic mechanisms.

Conclusion

The glucocorticoid-mediated response to inflammation must be stringently regulated, since both excessive and insufficient hormone secretion are associated with disease: chronic HPA axis over-activation, as occurs during stress, can affect susceptibility to or severity of infectious diseases, due to the immunosuppressive effects of glucocorticoids. On the other hand, blunted HPA axis responses are associated with increased susceptibility and activity of inflammatory diseases. In addition to the well known anti-inflammatory properties of corticosteroids the evidence for a defective HPA axis in RA offers new avenues to understand the spectrum of therapeutic benefits of glucocorticoids in RA and other immune mediated rheumatic diseases.

References

1. Masi AT, Bijlsma JW, Chikanza IC, Pitzalis C, Cutolo M. Neuroendocrine, immunologic, and microvascular systems interactions in rheumatoid arthritis: physiopathogenetic and therapeutic perspectives. Semin Arthritis Rheum 1999; 29: 65-81.
2. Masi AT. Hormonal and immunologic risk factors for the development of rheumatoid arthritis: an integrative physiopathogenetic perspective. Rheum Dis Clin North Am 2000; 26: 775-803
3. Masi AT. Neuroendocrine immune mechanisms in rheumatic diseases. An overview and future implications. Rheum Dis Clin North Am 2000; 26: 1003-1017
4. Sternberg EM. Emotions and disease: from balance of humors to balance of molecules. Nat Med 1997; 3: 264–267.
5. Chikanza IC, Grossman AB. Neuroendocrine immune responses to inflammation: the concept of the neuroendocrine immune loop. Baillieres Clin Rheumatol 1996; 10: 199-225.
6. Mackay CR, Imhof BA. Cell adhesion in the immune system. Immunol Today 1993; 14: 99-102.
7. Hirano T, Akira S, Taga T, Kishimoto T. Biological and clinical aspects of interleukin-6. Immunol Today 1990; 11: 443-449.
8. Kidd P. Th1/Th2 balance: the hypothesis, its limitations, and implications for health and disease. Altern Med Rev 2003; 8: 223-246.
9. Sternberg EM. Neuroendocrine regulation of autoimmune/inflammatory disease. J Endocrinol 2001; 169: 429-435.
10. Weinstock JV, Elliott D. The substance P and somatostatin interferon-gamma immunoregulatory circuit. Ann N Y Acad Sci 1998; 840: 532-539.
11. Cutolo M, Castagnetta L. Immunomodulatory mechanisms mediated by sex hormones in rheumatoid arthritis. Ann N Y Acad Sci 1996; 784: 237–251.
12. Masi AT, Da Silva JAP, Cutolo M. Perturbations of the

hypothalamic-pituitary-gonadal (HPG) axis and adrenal androgen (AA) functions in rheumatoid arthritis. Baillieres Clin Rheumatol 1996;10: 295–331.
13. Cerinic MM, Konttinen Y, Generini S, Cutolo M. Neuropeptides and steroid hormones in arthritis. Curr Opin Rheumatol 1998; 10: 220–235.
14. Cutolo M. Do sex hormones modulate the synovial macrophages in rheumatoid arthritis? Ann Rheum Dis 1997; 56: 281-284.
15. Chikanza IC, Grossman AB. Reciprocal interactions between the neuroendocrine and immune systems during inflammation. Rheum Dis Clin North Am 2000; 26: 693-711.
16. Trainer PJ, Faria M, Newell-Price J, et al. A comparison of the effects of human and ovine corticotrophin-releasing hormone on the pituitary-adrenal axis. J Clin Endocrinol Metab 1995; 80: 412-417.
17. Eijsbouts AM, Murphy EP. The role of the hypothalamic-pituitary-adrenal axis in rheumatoid arthritis. Baillieres Best Pract Res Clin Rheumatol 1999; 13: 599-613.
18. Ross JS, Bacon KB, Camp RD. Potent and selective inhibition of in vitro lymphocyte migration by cyclosporine and dexamethasone. Immunopharmacol Immunotoxicol 1990; 12: 439-455.
19. Snyder DS, Unuanue ER. Corticosteroids inhibit murine macrophage 1a expression and interleukin-1 production. J Immunol 1982; 129: 1803-1805.
20. Waage A, Slupphaug G, Shalaby R. Glucocorticoids inhibit the production of IL-6 from monocytes, endothelial cells and fibroblasts. Eur J Immunol 1990 ; 20: 2439-2443.
21. Mier JW, Vachino G, Klemper MS, et al. Inhibition of interleukin-2-induced tumor necrosis factor release by dexamethasone: prevention of an acquired neutrophil chemotaxis defect and differential suppression of interleukin-2 associated side effects. Blood 1990; 76: 1933-1940.
22. Mukaida N, Zacheriae CC, Gusella GL, Matsushima K. Dexamethasone inhibits the induction of monocyte chemotactic-activating factor production by IL-1 or tumor necrosis factor. J Immunol 1991; 146: 1212-1215.
23. Elenkov IJ. Glucocorticoids and the Th1/Th2 balance. Ann N Y Acad Sci 2004; 1024: 138-146.
24. Werb Z. Biochemical actions of glucocorticoids on macrophages in culture. Specific inhibition of elastase, collagenase, and plasminogen activator secretion and effects on other metabolic functions. J Exp Med 1978; 147: 1695-1712.
25. Buttgereit F, Straub RH, Wehling M, e col. Glucocorticoids in the treatment fo rheumatic diseases. An update on mechanisms of action. Arthritis Rheum 2004; 50: 3408-17.
26. Bijlsma JWJ, Saag KG, Buttgereit F, da Silva, JAP. Developments in glucocorticoid therapy. Rheum Dis Clin N Am 2005; 31: 1-17.
27. Barnes PJ. Anti-inflammatory action of glucocorticoids: molecular mechanisms. Clin Sci (Lond) 1998; 94: 557-572.
28. Barnes PJ, Karin M. Nuclear factor–kappaB: a pivotal transcription factor in chronic inflammatory diseases. N Engl J Med 1997; 336: 1066-1071.
29. Hench PS, Kendall EC, Slocumb CH, Polley HF. Effects of cortisone acetate and pituitary ACTH on rheumatoid arthritis, rheumatic fever and certain other conditions. Arch Med Interna 1950; 85:545–666.
30. Cupps TR, Fauci AS. Corticosteroid-mediated immunoregulation in man. Immunol Rev 1982; 65:133–155.
31. Elenkov IJ, Chrousos GP. Stress Hormones, Th1/Th2 patterns, Pro/Anti-inflammatory Cytokines and Susceptibility to Disease. Trends Endocrinol Metab 1999; 10: 359–368.
32. Ashwell JD, Lu FW, Vacchio MS. Glucocorticoids in T cell development and function. Annu Rev Immunol 2000; 18: 309–345.
33. Franchimont D, Galon J, Gadina M, Visconti R, Zhou Y, Aringer M, Frucht DM, Chrousos GP, O'Shea JJ. Inhibition of Th1 immune response by glucocorticoids: dexamethasone selectively inhibits IL-12-induced Stat4 phosphorylation in T lymphocytes. J Immunol 2000; 164: 1768–1774.
34. DeRijk RH, Petrides J, Deuster P, Gold PW, Sternberg EM. Changes in corticosteroid sensitivity of peripheral blood lymphocytes after strenuous exercise in humans. J Clin Endocrinol Metab 1996; 81: 228–235.
35. DeRijk R, Michelson D, Karp B, et al. Exercise and circadian rhythm-induced variations in plasma cortisol differentially regulate interleukin-1 beta (IL-1 beta), IL-6, and tumor necrosis factor-alpha (TNF alpha) production in humans: high sensitivity of TNF alpha and resistance of IL-6. J Clin Endocrinol Metab 1997; 82: 2182–2191.
36. Vacchio MS, Ashwell JD. Thymus-derived glucocorticoids regulate antigen-specific positive selection. J Exp Med 1997; 185 2033–2038.
37. Sternberg EM, Hill JM, Chrousos GP, Kamilaris T, Listwak SJ, Gold PW, Wilder RL. Inflammatory mediator-induced hypothalamic-pituitary-adrenal axis activation is defective in streptococcal cell wall arthritis-susceptible Lewis rats. Proc Natl Acad Sci USA 1989; 86: 2374-2378.
38. Sternberg EM, Young WS 3rd, Bernardini R et al. A central nervous system defect in biosynthesis of corticotropin-releasing hormone is associated with susceptibility to streptococcal cell wall-induced arthritis in Lewis rats. Proc Natl Acad Sci USA 1989; 86: 4771–4775.
39. Lechner O, Hu Y, Jafarian-Tehrani M et al. Disturbed immunoendocrine communication via the hypothalamo-pituitary-adrenal axis in murine lupus. Brain Behav Immun 1996; 10: 337–350.
40. Sarlis NJ, Chowdrey HS, Stephanou A, Lightman SL. Chronic activation of the hypothalamo-pituitary-adrenal axis and loss of circadian rhythm during adjuvant-induced arthritis in the rat. Endocrinology 1992; 130: 1775–1779.
41. Kanik KS, Chrousos GP, Schumacher HR, Crane ML, Yarboro CH, Wilder RL. Adrenocorticotropin, glucocorticoid, and androgen secretion in patients with

new onset synovitis/rheumatoid arthritis: relations with indices of inflammation. J Clin Endocrinol Metab 2000; 85: 1461-1466.
42. Mason D, MacPhee I, Antoini F. The role of the neuroendocrine system in determining genetic susceptibility to experimental allergic encephalomyelitis in the rat. Immunology 1990; 70: 1–5.
43. Edwards CK 3rd, Yunger LM, Lorence RM, Dantzer R, Kelley KW. The pituitary gland is required for protection against lethal effects of Salmonella typhimurium. Proc Natl Acad Sci USA 1991; 88: 2274–2277.
44. Sternberg EM. Neural–immune interactions in health and disease. J Clin Invest 1997;100: 2641–2647.
45. Misiewicz B, Poltorak M, Raybourne RB, Gomez M, Listwak S, Sternberg EM. Intracerebroventricular transplantation of embryonic neuronal tissue from inflammatory resistant into inflammatory susceptible rats suppresses specific components of inflammation. Exp Neurol 1997; 146: 305–314.
46. O'Connor TM, O'Halloran DJ, Shanahan F. The stress response and the hypothalamic-pituitary-adrenal axis: from molecule to melancholia. QJM 2000; 93: 323-333.
47. Kellendonk C, Tronche F, Reichardt HM, Schutz G. Mutagenesis of the glucocorticoid receptor in mice. J Steroid Biochem Mol Biol 1999; 69: 253–259.
48. Kanik KS, Wilder RL. Hormonal alterations in rheumatoid arthritis, including the effects of pregnancy.Rheum Dis Clin North Am 2000; 26: 805-823.
49. Harkness JA, Richter MB, Panayi GS, et al. Circadian variation in disease activity in rheumatoid arthritis. Br Med J (Clin Res Ed) 1982; 20: 551-554.
50. Mirone L, Altomonte L, D'Agostino P, Zoli A, Barini A, Magaro M. A study of serum androgen and cortisol levels in female patients with rheumatoid arthritis. Correlation with disease activity. Clin Rheumatol 1996; 15: 15-19.
51. Gudbjornsson B, Skogseid B, Oberg K, Wide L, Hallgren R. Intact adrenocorticotropic hormone secretion but impaired cortisol response in patients with active rheumatoid arthritis. Effect of glucocorticoids. J Rheumatol 1996; 23: 596-602.
52. Crofford LJ, Kalogeras KT, Mastorakos G, et al. Circadian relationship between interleukin (IL)-6 and hypothalamus-pituitary-adrenal axis hormones: Failure of IL-6 to cause sustained hypercortisolism in patients in early untreated rheumatoid arthritis. J Clin Endocrinol Metab 1997; 82: 1279-1283.
53. Hall J, Morand EF, Medback S, et al. Abnormal hypothalamic-pituitary-adrenal function in human rheumatoid arthritis. Effects of nonsteroidal anti-inflammatory drugs and water immersion. Arthritis Rheum 1994; 37: 1132-1137.
54. Neeck G, Federlin K, Graef V, Rusch D, Schmidt KL. Adrenal secretion of cortisol in patients with rheumatoid arthritis. J Rheumatol 1990; 17: 24-29.
55. Chikanza IC, Petrou P, Kingsley G, Chrousos G, Panayi GS. Defective hypothalamic response to immune and inflammatory stimuli in patients with rheumatoid arthritis. Arthritis Rheum 1992; 35: 1281--1288.

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