New Aspect on Etiology and Therapy of Collagen Diseases and Inflammatory Bowel Diseases

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ABSTRACT
Many clinicians have considered that collagen diseases (or autoimmune diseases) may be at activated immune states and have therefore used steroid hormones and other immunosuppressive drugs for the treatment. However, these diseases seem to be worsened by the treatment after all. The immunologic state similar to collagen diseases is also seen in aging, showing the appearance of autoreactive T cells and autoantibody-producing B cells. In fact, conventional lymphocytes rather decrease in patients with collagen diseases or autoimmune diseases and inversely granulocytes increase. In other words, these patients are at immunosuppressive states, showing the increase of granulocytes, extrathymic T cells, and autoantibody-producing B cells. Infections or stress may induce these immunologic states in the patients. Patients with inflammatory bowel diseases also show granulocytosis and lymphocytopenia but clinicians use NSAIDs and steroid hormones which have the activating potential of granulocytes. I propose here that clinicians should select the treatments of immunopotentiation rather than those of immunosuppression for these patients with collagen diseases or inflammatory bowel diseases.

In a series of recent studies, we have revealed that the distribution of leukocytes are regulated by the autonomic nervous system (11, 12). Granulocytes bear adrenergic receptors on the surface: their number and functions are activated with sympathetic nerve stimulation. On the other hand, lymphocytes bear cholinergic receptors on the surface: their number and functions are activated with parasympathetic nerve stimulation. These responses may be beneficial for protection of the body of living beings. Granulocytes are important for protection of bacteria which invade the body at active life, whereas lymphocytes are important for processing foreign antigens and viral particles which invade the body (e.g., the intestine) at resting time or feeding time. However, living beings fall victims to diseases if the autonomic nervous system deviates too much to one direction. Overactivation of granulocytes is related to tissue damage via their superoxide production (4, 10). Inversely, overactivation of lymphocytes is related to allergic diseases. In this communication, we represent possible mechanisms and therapies of collagen diseases and some inflammatory bowel diseases.

PHYLOGENETIC DEVELOPMENT OF LEUKOCYTES
Proto-macrophages differentiated into blood cells as well as into cells of mesodermal origin in subsequent phylogenic development. These proto-macrophages are also important as the origin of self-defense cells, namely, leukocytes. In addition, proto-macrophages might give rise to granulocytes and
lymphocytes in phylogeny (Fig. 1). We consider that macrophages differentiated into granulocytes by refining their phagocytic function and acquiring numerous cytoplasmic granules. On the other hand, there was another direction of differentiation in macrophages. Some macrophages lost their phagocytic function and developed adhesion molecules on the surface. One family of such adhesion molecules is a product of the immunoglobulin gene superfamily. As a result, such macrophages differentiated into lymphocytes which have immune functions.

In the peripheral blood of humans, macrophages, granulocytes, and lymphocytes constitute approximately 5%, 60%, and 35% of the leukocytes, respectively. However, the ratio of these components among leukocytes varies by influence of the autonomic nervous system (11, 12).

**STRESS AND GASTRIC ULCERS**

We have reported that an H₂-blocker decreased the number of granulocytes while a proton pump inhibitor (PPI) suppressed the function (i.e., superoxide production) of granulocytes in humans and mice (5). In sharp contrast, the H₂-blocker did not suppress the function of granulocytes and the PPI did not decrease the number of granulocytes. In conjunction with recent results that granulocytes carry surface adrenergic receptors and are activated by sympathetic nerve stimulation (11), these results suggest that the accumulation of granulocytes in the gastric mucosa might be important in the pathogenesis of gastric ulcers. Sympathetic nerve activation in such patients might be induced by mental and physical stress or bacterial infection (e.g., H. pylori).

As shown in a mouse study in Fig. 2, granulocyte trafficking (the bone marrow→the circulation→the mucosa) is important for understanding the granulocyte theory. If stress continues chronically, granulopoiesis itself increases in the bone marrow and the levels of granulocytes in the periphery are elevated in various tissues, as well as in the blood. This study enables us to properly understand the therapeutic base of antiulcer agents, H₂-blockers, and PPIs. The proposed theory also explains why H₂-blockers and PPIs ameliorate the junction ulcers in patients with total gastrectomy who have lost all parietal cells for acid secretion.

Given the above, we propose a granulocyte theory of gastric ulcer formation (5). Granulocytes, which carry surface adrenergic receptors, are therefore acti-
vated by sympathetic nerve stimulation and subsequently induce gastric ulcers. In recent studies, *H. pylori* infection has been raised as one of the causes of gastric ulcer formation (1, 2, 6). However, this concept is incomplete. Thus, some gastric ulcer patients are free from this infection and many healthy individuals are infected with *H. pylori*. With the granulocyte theory of gastric ulcer formation, all cases of gastric ulcer can possibly be explained. In this case, *H. pylori* infection is important as a secondary factor that stimulates granulocytes. Sympathetic nerve stimulation is induced by many causes, including mental stress, overwork, bacterial infection, NSAID administration, and a combination of such factors.

**IMMUNOLOGIC STATES OF AUTOIMMUNE DISEASES**

Many investigators and clinicians have long believed that self-reactive forbidden clones, which evoke autoimmune diseases or collagen diseases, may be generated through failure in T-cell differentiation in the thymus. However, we have never encountered with such failure of intrathymic T-cell differentiation (3, 8).

In autoimmune prone NZB/W F₁ mice, severe thymic atrophy is rather induced at the onset of disease (Fig. 3). This result suggests that the mainstream of T-cell differentiation is arrested under autoimmune conditions. Inversely, the number of lymphocytes in the liver (consisting of extrathymic T cells) and that of peritoneal exudate cells (consisting of autoantibody-producing B cells or B-1 cells) increase. In other words, extrathymic pathway of T-cell differentiation, which primarily produces self-reactive forbidden clones without negative selection, is activated in autoimmune diseases or collagen diseases.

Reflecting these situation, we have always observed that lymphocytopenia (consisting of conventional T and B cells) and granulocytosis (results from sympathetic nerve stimulation) are accompanied in patients with rheumatoid arthritis (RA) (Table 1). Similar phenomena are also seen in patients with other autoimmune diseases or collagen diseases (e. g., SLE, Hashimoto’s disease, scleroderma, Behçet’s syndrome, etc.) (data not shown).

**DIRECT CAUSES TO THE ONSET OF AUTOIMMUNE DISEASES**

There are two major causes to induce autoimmune diseases or collagen diseases, namely, 1) infections and 2) mental or physical stress (Fig. 4). When viral or bacterial infection is severe, tissue damage is evoked by inflammation. Such tissue damage induces sympathetic nerve activation and results in granulocytosis and primitive lymphocytes (i. e., innate immunity). If tissue damage is continued, activated granulocytes produce superoxides as well as

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<td>Control</td>
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1) Infections → tissue damage → sympathetic nerve activation
2) Mental or physical stress → sympathetic nerve activation
   → tissue damage

Sympathetic nerve activation induces thymic atrophy and
the activation of innate immune system.

Fig. 4 Possible causes and mechanisms underlying the onset of autoimmune diseases. Tissue damage and sympathetic nerve activation influence with each other.

many inflammatory cytokines (e.g., TNFα, IFNg, and IL-6) and accelerate again tissue damage. At this time, primitive lymphocytes such as extrathymic T cells (i.e., CD56+T or CD57+T cells in humans) mediate autoreactivity against denatured self-tissue and B-1 cells produce autoantibodies.

These responses are sometimes beneficial for the elimination of denatured self-cells or are sometimes dangerous for the acceleration of tissue damage. Similarly, mental or physical stress becomes to be the cause of autoimmune diseases, because such stress induces sympathetic nerve activation. Such sympathetic nerve activation then induces granulocytosis and results in tissue damage. Indeed, we are often able to listen such mental and physical stress (e.g., overwork) at the onset of disease from patients with autoimmune diseases.

Sympathetic nerve activation also induces thymic atrophy which indicates the arrest of the mainstream of T-cell differentiation in the thymus (5). Inversely, primitive lymphocytes such as NK cells, extrathymic T cells, and autoantibody-producing B-1 cells (i.e., constituents of innate immune system) are activated. Again, these responses are not always harmful for our body, because these cells are responsible to the elimination of abnormal or denatured self-cells. A similar opinion is also proposed by other investigators (9).

NEW THERAPEUTIC APPROACH FOR AUTO-IMMUNE DISEASES OR COLLAGEN DISEASES

If we notice that the immunologic state of autoimmune diseases or collagen diseases is severe immunosuppression, new therapeutic approach for these diseases is raised. Up to the present, a conventional therapy for autoimmune diseases or collagen diseases has been the administration of immunosuppressants and steroid hormones. But the therapy should worsen the diseases. NSAIDs which reduce our body temperature are also dangerous for the treatment of these diseases: NSAIDs induce severe sympathetic nerve activation and result in granulocytosis (13, 14). This phenomenon is also related to the gastritis and gastric ulcers induced by the long-lasting administration of NSAIDs.

We always encounter the inflammations of autoimmune diseases or collagen diseases, showing pain, fever, redness, skin rash, diarrhea, etc. These inflammations should be considered as a result of acceleration of circulation which induces the recovery from tissue damage. Parasympathetic nerve activation is related to these symptoms. Actual factors include prostaglandins, histamine, serotonin, acetylcholine, leukotriens, etc.

Instead of the use of immunosuppressants, we have rather to select the therapy to increase circulation, inflammation, and immune functions. Such trials include mild exercise, taking bath, laughing, and the treatment of oriental medicine such as acupuncture and Chinese medicines. Especially, acupuncture and Chinese medicines induce the stimulation of parasympathetic nerves in patients (7).

EXPLOSION IN THE NUMBER OF PATIENTS WITH ULCERATIVE COLITIS IN JAPAN

In 1975, ulcerative colitis was indicated as one of specific diseases by the declaration of Japanese Government. The number of such patients increased thereafter (Fig. 5). We consider that an inappropriate therapy was fixed since that time, because the fixed therapy is the use of aminosalicylic acid (one of NSAIDs) and steroid hormones.

In this experiment (Fig. 6), we examined how NSAID (indomethacin, 0.5 mg/day/mouse 7 days in this experiment) stimulated sympathetic nerves. A prominent increase in the serum concentration of catecholamines was induced in mice. It is speculated that the suppression of prostaglandins by NSAIDs is related to this phenomenon. Thus, prostaglandine acts as the suppressive system against catecholamines.

Such sympathetic nerve stimulations induced by indomethacin then induced the infiltration of granulocytes into various sites of the digestive tract, show-
Fig. 5  Explosion in the number of patients with ulcerative colitis in Japan. In 1975, ulcerative colitis was indicated as one of the incurable diseases. From that time, the number of patients began to increase.

Fig. 6  The activation of sympathetic nerves by the continuous administration of NSAID. Indomethacin (0.5 mg/day/mouse) was administrated every day for 7 days. Serum levels of catecholamines were measured on day 8.
THERAPEUTIC PROPOSAL FOR ULCERATIVE COLITIS AND CROHN'S DISEASE

In light of these findings, we propose new therapeutic approaches for ulcerative colitis and Crohn's disease. It includes 1) Cessation of salazosulphapyridine (Salazopyrin), mesalazine (Pentasa), and steroid (Steroneam) which induce granulocytosis, 2) Care of mental stress and foods, and 3) Stimulate parasympathetic nerves (acupuncture, mild exercise, taking bath, laughing, etc).

ACCOMPANYING PAIN, REDNESS, FEVER, INFLAMMATION, AND DIARRHEA IN THE RECOVERY RESPONSE FROM DISEASES

When we encounter severe mental or physical stress, circulation failure and tissue damage are induced by sympathetic nerve activation (Fig. 9). To recover circulation failure and tissue damage, parasympathetic nerve reflex is suddenly induced, accompanying pain, redness, fever, inflammation, and diarrhea. As already mentioned, prostaglandins, acetylcholine, serotonin, histamine, etc are associated

Fig. 7 infiltration of granulocytes into the large intestine, but not the small intestine, by the continuous administration of NSAID. Experimental protocol is the same as Fig. 6. Two-color staining for Mac-1 and Gr-1 was conducted to identify granulocytes (Mac-1, Gr-1). IEL—intraepithelial leukocytes, LPL—lamina propria leukocytes

Fig. 8 Comparisons of the number of whole leukocytes and the proportion and number of granulocytes and lymphocytes between patients with ulcerative colitis (UC) and Crohn's disease (CD).

Fig. 9 Recovery responses from circulation failure and tissue damage. Irritable symptoms are rather recovery responses with parasympathetic nerve activation. The long-lasting administration of NSAIDs and steroid hormones suppress these valuable responses in our body.
with these responses.

If we used NSAIDs or steroid hormones, especially for a long time, all recovery responses are ceased. Therefore, the suppression of symptoms and the reappearance of recovery responses are repeated in the patients.

REFERENCES


