Commentary

Transforming Growth Factor β: The Good, the Bad, and the Ugly

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As is often the case with peptide growth factors and cytokines, the original activity discovered and identified as TGF-β merely represented the tip of the iceberg. Not only does it regulate growth, as evident by its name, but TGF-β mediates far-ranging biologic processes including inflammation and host defense, in addition to development, tissue repair, and tumorigenesis. Released locally from platelet stores early in an inflammatory response, TGF-β is then generated by inflammatory cells themselves as part of the cytokine network. Initially, in its defensive role, TGF-β mediates the egress of undifferentiated leukocytes and, subsequently, it facilitates resolution of inflammation and promotes tissue repair (1, 2). But this carefully choreographed series of events, which represents the "good" side of TGF-β, is dependent upon a critical balance of the growth factor. It is becoming increasingly evident that too much of a good thing can be bad; excess TGF-β within a lesion has been associated with unresolved inflammation and fibrotic events (1–3). Now, it appears, based on the study by Lowrance et al. (4) in the MRL/1pr autoimmune murine model, that TGF-β has an even uglier side, in that its accumulation in the circulation may predispose the host to serious and recurrent infections. Moreover, in humans, genetic and acquired diseases such as systemic lupus erythematosis (SLE) may mirror the MRL/1pr mouse.

Under the best of circumstances, TGF-β provides a link between each of the processes by which cells and/or tissues respond to infection or injury and initiate repair. By increasing adhesion molecules, generating a potent chemotactic gradient, and inducing itself and an array of other factors as part of the cytokine network (5–7), TGF-β orchestrates leukocyte recruitment and activation. TGF-β then downregulates these processes by inhibiting the functions of inflammatory cells once they are activated (2, 8) and facilitates healing by promoting fibroblast recruitment and matrix synthesis (3). As recently reviewed (2), this apparent contradictory influence of TGF-β on cells of the immune system, both stimulatory and inhibitory, is accounted for, in part by the differential effects of TGF-β on resting and activated cells. As a general, but by no means exclusive rule, resting, immature cells are stimulated by TGF-β, whereas activated representatives of the same cell populations may be inhibited by TGF-β. Thus, TGF-β serves as a conversion factor, converting an active inflammatory site into one dominated by resolution and repair. Upsetting the delicate balance of TGF-β that dictates these events may have pathologic consequences.

Indeed, persistent stimulation of immune/inflammatory events by internal or external challenges can overload the balance with chronic secretion of TGF-β. Not only might this local excess lead to unresolved inflammation (9), but also, TGF-β may reach the circulation. When elevated systemic levels of TGF-β occur, either endogenously during disease states (4, 10), or after exogenous administration (11–14), a plethora of additional cellular targets and consequently, diverse signaling pathways with different outcomes, may become operative. Although the source(s) of the endogenous levels of blood TGF-β is uncertain in the autoimmune MRL/1pr mice, the spleen may contribute to the circulating pool in this and perhaps other models characterized by immune malfunction (4, 15). Why TGF-β accumulates is unknown, but TGF-β autoinduces its own synthesis (7). In addition, dysregulation of its activity because of persistence of the enzymatic machinery essential for its activation or insufficient levels of inhibitors may be contributory.

Nonetheless, whereas the tightly controlled, localized production of TGF-β occurs as a necessary component of an active inflammatory process, the aftermath of excess TGF-β, now found in the circulation, is to inhibit the same immune and inflammatory pathways. It is not inconceivable that this built-in negative feedback loop, occurring in TGF-β excess, evolved for the protection of the host during bouts of rampant inflammation. The dichotomy between local and systemically disseminated TGF-β was first documented in an experimental arthritis model in which local secretion or administration of TGF-β was found to drive the inflammatory response, whereas systemic inoculation inhibited the same response (9, 13, 16) (Fig. 1).

By what mechanism(s) does circulating TGF-β promote immune suppression and can these pathways be used to clinical advantage? Recent evidence suggests that systemic TGF-β targets endothelial cells, where it inhibits E-selectin expression to block adhesion and targeting of leukocytes to the site of inflammation (17) (Fig. 2). Moreover, since leukocytes are normally sensitive to a concentration gradient of chemotactic signals emanating from the site of inflammation (5, 9), the presence of elevated TGF-β in the blood would destroy such an outward gradient (Fig. 2). Evidently, the effects of secreted or exogenous TGF-β extend far beyond the control of cell growth and differentiation to include a myriad of other regulatory activities converging on phagocytic cells and activated lymphoid cells of all lineages (reviewed in 1, 2). While it

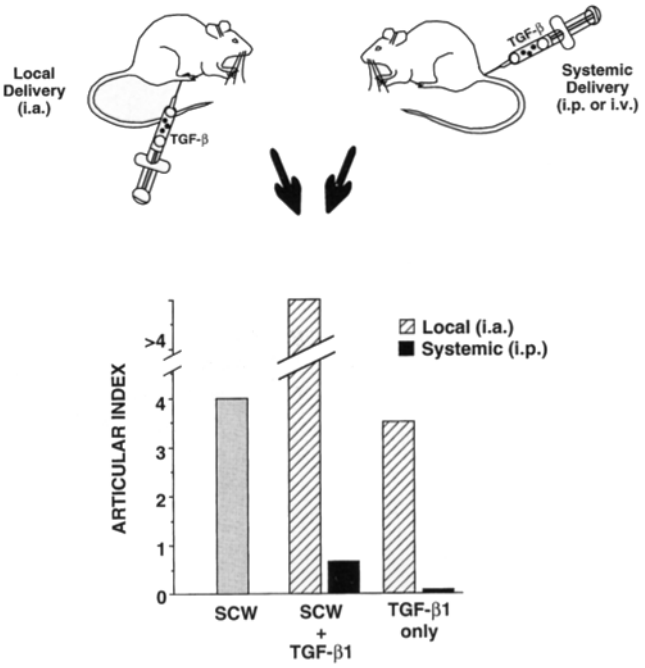

Figure 1. Effect of route of TGF-β administration on streptococcal cell wall (SCW)-induced arthritis. Arthritis was induced with group A SCWs (30 μg rhamnose i.p./100 gm rat) and TGF-β was injected systemically (i.p., 1 μg/daily) or locally (intraarticular [i.a.], 250 ng/joint, 1 injection only). In control (nonarthritic) rats, TGF-β alone was given systemically (1 μg daily) or locally (i.a., 3-daily 100 ng injections). Articular indices were determined as described (13, 16).

is these pathways that offer promise for TGF-β in immunotherapy, there can be a downside to inducing a state of immunologic unresponsiveness, in that overexpression or parenteral administration of TGF-β may convert a natural, self-limited infection into one with potentially lethal consequences (4, 18, 19).

The ability of endogenous or pharmacologically elevated circulating TGF-β in the MRL/1pr mice to suppress antimicrobial functions reflects TGF-β's reported inhibitory influence on reactive oxygen and nitrogen intermediates (8, 20). Increased nitric oxide (NO) generated from L-arginine and molecular oxygen is a pivotal mechanism by which macrophages inhibit a variety of pathogens, and downregulation of the inducible form of NO synthase has been associated with reactivation of bacterial and viral infections (21, 22). While the ability of TGF-β to dampen widely inducible NO generation may have evolved to protect the host from an overabundance of this autotoxic molecule (23, 24), excess TGF-β can have its own problems. Paradoxically, since TGF-β inhibits NO, it is somewhat surprising that augmented levels of NO and TGF-β coexist in the MRL/1pr mice (4, 25), and moreover, that increased NO does not appear to confer any survival advantage against the bacteria. These findings reiterate the unpredictable nature of a pleiotropic molecule such as TGF-β, and emphasize the impact that this peptide may have on host resistance.

A number of human genetic and acquired diseases are also accompanied by a nonspecific immune depression. The potent effects of TGF-β on cellular immune pathways resemble in many ways those accompanying such autoimmune and infectious diseases, and elevated circulating levels of TGF-β have been measured in SLE, the human equivalent of the MRL/1pr mouse, as well as in HIV-1 infection and arthritis (4, 10, 26). In the process of defending the host from persistent antigens or invading pathogens, the overzealous release of intended protective molecules such as TGF-β can, through its endocrine links, have an opposite effect. Under these circumstances, it may be appropriate to consider employment of cytokine antagonists.

Because TGF-β is a critical determinant of so many aspects of the host response to injury and infection and to the process of tissue repair, there is considerable interest in its potential therapeutic applications. Taking advantage of the good

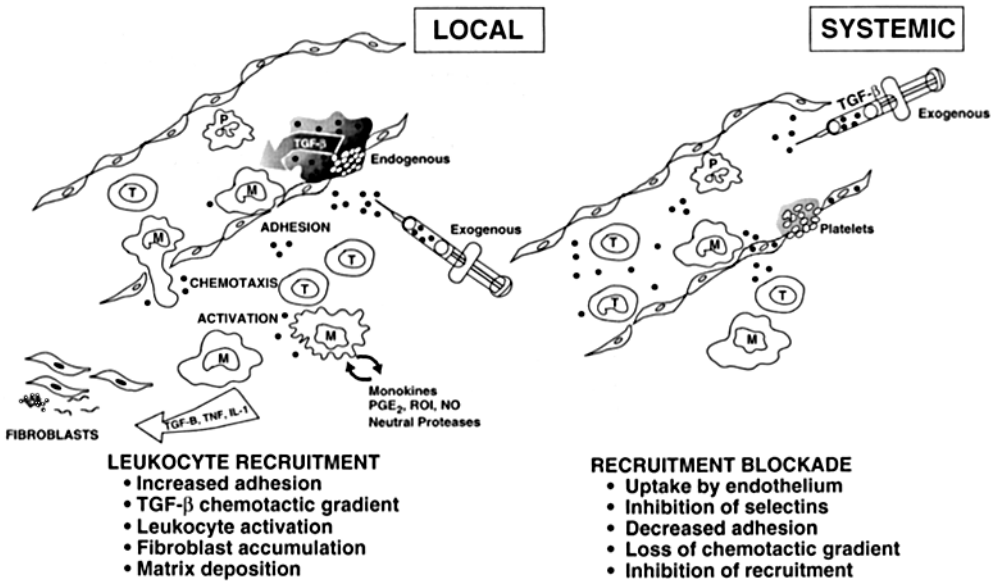

Figure 2. Differential regulation of inflammation by local and systemic TGF-β. Summary of differential effects of TGF-β on inflammatory cell recruitment and activation after local administration and administration through the circulation. T, T lymphocyte; M, monocyte/macrophage; P, polymorphonuclear leukocyte.

aspects of TGF-β's role in inflammation and wound healing, local application can be used to promote repair (27, 28) and with a single treatment, the bad aspects of overabundant TGF-β (excess inflammation and/or fibrosis) can be averted. Conversely, in conditions characterized by chronic inflammation and fibrosis (too much TGF-β), locally active TGF-β antagonists are finding experimental applications (1, 3, 9, 29). However, one of TGF-β's main therapeutic attractions remains its promise as an immunosuppressive agent, and to optimize these mechanisms requires systemic delivery. Under these conditions, TGF-β has disease-limiting properties in autoimmune and chronic inflammatory diseases, but the very mechanisms it suppresses are also those necessary for defense against infectious pathogens. Its potentially "ugly" side, predisposing the patient to uncontrolled infections, must be factored into the design of therapeutic strategies. The information presented in the MRL/1pr study suggests that not only may aberrant production of endogenous TGF-β contribute to the high risk of bacterial infections in these animals, and perhaps in lupus and other acquired and genetic diseases, but that exogenous TGF-β delivered by systemic routes, even in a single dose, may have similar manifestations. Ongoing studies in animal models and humans may ultimately enable us to manipulate the dose of TGF-β, the route of administration, the timing, and other clinically relevant parameters to either potentiate or suppress the immune response as best benefits the host. Adjunctive approaches may include manipulation of transcriptional and translational events involved in controlling the endogenous synthesis of the peptide and/or its natural inhibitors to sculpt the desired immunologic outcome. Regulated TGF-β synthesis during the induction of oral tolerance provides an example of such a directed response (30). Even though we know a lot about TGF-β, there is still a lot to learn.

We eagerly awaited the genetically engineered TGF-β loss-of-function mutation to answer all we ever wanted to know about TGF-β, including its role in inflammation and immune function, but we found that the TGF-β1 gene knockout has, as appears to be typical of this quixotic molecule, provided more questions than answers. Although the mice appear surprisingly normal at birth and for 2–3 wk thereafter, a marked leukocyte adhesion to endothelium (31) with infiltration into vital organs leads to organ failure, cachexia, and death (32–34). This was initially thought to be the obvious result of the absence of the requisite immunosuppressive actions of TGF-β1. However, continued analysis has revealed that the TGF-β1 knockouts, born to TGF-β heterozygous parents, were the recipients of maternally transferred TGF-β1 (35), confounding the interpretation of the incipient pathology. Again in this model, it is the systemic transport of TGF-β1, occurring pre- and postnatally, which yields an unpredictable outcome. Maternal TGF-β, serving as an endocrine source of this molecule, albeit temporary, appears to be sufficient for embryogenesis and birth of the knockouts, but likely also orchestrates their demise. The slacking off of the maternal supply of TGF-β1 appears to coincide with the onset of symptoms and hypothetically, at least, residual TGF-β1, in the absence of any circulating TGF-β1, may leach or be transported from tissue stores to generate a transient chemotactic gradient and initiate leukocyte migration (31). By whatever mechanisms this recruitment is triggered, once the inflammatory cells accumulate in the tissues, there is no possibility to generate de novo the TGF-β1 required to suppress leukocyte function as should occur in the normal resolution of inflammatory events (1, 2). Inexplicably, however, systemically delivered exogenous TGF-β does not reverse this process, but survival can be prolonged if inflammation is suppressed either genetically, by placement of the TGF-β1 targeted allele onto the severe combined immunodeficiency background (TGF-β1 mutant SCID) (Diebold, R., personal communication), or pharmacologically, by blocking adhesion and recruitment (31).

The TGF-β1 knockout and MRL/1pr models provide important paradigms to unravel the ramifications of too much or too little TGF-β on host defense. Paradoxically, both extremes appear to be associated with autoimmune-like phenomena. One thing is for sure, there is much to be done if we are to adequately understand the regulation of and by this amazing peptide, in order to develop therapeutic strategies to use the good, control the bad, and eliminate the ugly consequences of TGF-β.

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