Letter to the Editor

Measuring Soluble Adhesion Molecules

Sfikakis and Tsokos published a review (2) on a basic-immunology topic that is currently under very active investigation. Specifically, they focus on the clinical laboratory measurements of various soluble adhesion molecules (SAM) in autoimmune diseases.

The studies on SAM are providing important new insights into the immune response and mechanisms of pathogenesis of some chronic autoimmune inflammatory disorders. It would seem that factor analyses could (or even should) provide some exciting additions to the repertoire of diagnostic laboratory immunology tests, ones that would identify or monitor disease activity.

During the immune responses and accompanying inflammation, the gene expression and synthesis of SAM become upregulated. Surface SAM increase the affinity of interactions of the immune cells and also provide a localizing or targeting effect for populations of activated cells relative to certain tissue sites, including abnormal localization in the case of autoimmunity. The SAM are a very diverse family of molecules; in addition to adhesion properties, many of them possess peptide sequences that are homologous to known growth-promoting molecules.

It seems that it is mainly the cytokines elaborated during early immune activation that causes the upregulation of SAM. Successful therapeutic agents used in autoimmune diseases do the opposite and downregulate SAM synthesis.

These properties so vital to the diseases must have some diagnostic utility. Interestingly, SAM become solubilized from membranes and can be measured in plasma or other body fluids. This phenomenon is more pronounced during disease commercial kits are available to measure the soluble SAM. Can we now use these to monitor clinical disease activity in terms of real relevant biological products?

Sfikakis and Tsokos provided a table of data (Table 1 [2]) from numerous publications listing several SAM whose concentrations in plasma, in inflammatory disease, may be raised to levels over five times the “normal”.

Sfikakis and Tsokos point out that it is not totally clear what the concentration of the SAM is. Is it an index of ongoing increased cell activity in which the SAM further promotes the “inflammation,” or is it an index of a regulatory host response with the SAM functioning to modulate the response by tying up receptors and blocking cellular interactions?

Sfikakis and Tsokos lead us through a substantial body of literature on soluble and tissue-expressed adhesion molecules in various CT and vasculitis disorders. They conclude that raised SAM probably reflect cell activation, but the value of specific measurements remains uncertain. In the future, better-controlled studies are in order.

There are many pitfalls in measuring SAM: a lack of good reference standards, existence of isoforms with different antigenicities and other properties, tissue absorption effects of the SAM, and others. The imperfections in the technology are being addressed, however (1), and it should soon be possible to do really precise measurements on clinical samples—if this is worth doing.

In summarizing their conclusions about the investigations on cell adhesion molecules (CAM), Sfikakis and Tsokos point out some of the deficiencies in gathering data on clinical samples—poorly standardized techniques, sometimes a failure by investigators to uniformly process and study comparable material, and ignorance concerning normal values and their variations. These all lead to uncertainties regarding the potential value of future assays in clinical settings.

This is unfortunate, because measurements such as these deal with substances that are dynamically affected right at the heart of the immune response, becoming altered as part of pathogenesis and disease activity. The story of their actions is very convincing, and it is integral to the very precise current understanding of how immune responses work. In autoimmunity, disease-triggering factors activate the immune system, cells which elaborate cytokines, and CAM; these then promote and amplify the immune inflammatory mechanisms and later may modulate these. The CAM responses must be able to provide good surrogate markers for disease activation and activity (and possibly therapeutic drug efficacy)—if we can learn how and when to best measure specific factors. This is exactly the type of measurement that is needed clinically for many chronic immune-inflammatory diseases—a really good objective surrogate marker of the disease activity. Therapeutic decision making in such diseases can be very difficult on the clinical grounds alone. Reliable and practical laboratory markers would be extremely useful; serum cytokines and adhesion molecule measurements hold (held) the promise of being able to deliver this. If the measurements of these analytes have not yet delivered, is this because the basic science describing these substances and activities and our understanding of immune mechanisms are faulty, or is it because we have simply not learned how to do these tests in a meaningful way, taking sufficient care in sample processing and ensuring really precise standardized assays?

It might be nice to choose a few of the most promising CAM assays—one that reflects activation mainly of the responding immune-related cells and others that are more reflective of the target tissue cell activation—and then do further, intensive studies on measured values in the normal state and the disease state. Since it is likely that we will continue to be dependent on blood and plasma samples as the substrates for most of our clinical laboratory measurements for the assessment of the immune status in humans, we must learn how to handle and process these specimens and learn when to take them from the patient. This means overcoming technical problems.

The data can then be placed in the context of disease activity (or suppression) with a better understanding of what soluble CAM really signify. I remain very optimistic that measurements of these critically relevant molecules should expand our ability to adequately judge the integrity and activation status of a patient’s immune system.

REFERENCES


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Author’s Reply

Dr. Osterland offers a number of thoughtful remarks on measuring soluble adhesion molecules (SAM). These considerations extend the scope of our recent review (Clin. Diagn. Lab. Immunol. 4:241–246, 1997) and stress the potential importance of measuring SAM in clinical samples.

Prior to attempting to measure SAM in clinical samples, basic studies have usually suggested, or proven, that a molecule, usually expressed on the cell surface membrane, is important in cell activation and function. Subsequent studies invariably showed that this molecule exists in a soluble form either because it is shed off the cell membrane or because the cells produce an alternatively spliced molecule that lacks the transmembrane portion. With all this information in hand the clinical researcher should assume the leadership in establishing the clinical usefulness of measuring the levels of any given molecule in clinical samples. What we tried to convey in our review is that in many cases a “shortcut” had been taken to establish the clinical importance of measuring SAM in patient samples. Measurements of SAM have been performed and reported on samples collected in a cross-sectional manner.

Although there is no doubt, as Dr. Osterland points out, that the assays need to be standardized and the samples need to be collected properly in order to eliminate intra- and interexperimental and, when possible, interlaboratory variation, there are a number of issues that should be addressed prior to making the assays available to clinical practice.

(i) Does the assay offer help in establishing a specific diagnosis? Most, practically all, SAM reflect immune activation or modulation, and no disease or group of diseases seems to be preferentially associated with any given SAM. The answer to this question can easily be established in properly designed cross-sectional studies that include samples from patients with control diseases.

(ii) Does the assay have any value in determining disease activity? Such a question has to be answered properly prior to claiming the assay as a clinical tool. Cross-sectional studies cannot be used to answer this question, and showing differences between samples collected from patients with active disease and samples collected from patients with inactive disease is not sufficient to answer this question. The disease process leads to increased levels of SAM in body fluid samples and clinical manifestations. These two do not present necessarily at the same time point. Therefore, samples collected from patients with clinically inactive disease may have been misclassified because they were taken from patients in whom the disease flared shortly thereafter and vice versa. If SAM contribute to the disease pathogenic process then they should precede the expression of the clinical disease. Prospective and preferably controlled studies need to be done in order to show that a SAM can predict clinical activity. For instance, if soluble interleukin-2 receptor (sIL-2R) is elevated in patients with Graves’ disease, then serum sIL-2R levels should be measured prospectively in groups of patients that enter a randomized therapeutic trial with two or more modalities that are expected to have different effects on the disease activity. If sIL-2R can predict disease activity, then its levels in serum in the group of patients that achieved more clinical disease improvement should display, in a time-progressive manner, lower levels of sIL-2R. Similarly, if soluble ICAM-1 is elevated in patients with rheumatoid arthritis and if the levels of soluble ICAM-1 in serum predict disease activity, then, if measured in groups of patients who undergo clinical treatment with regimens that are expected to affect disease activity, its levels should be lower in the group of patients that achieved more pronounced clinical improvement than in the group that achieved less clinical improvement.

Proper studies are also needed to determine the time gap between the determination of an elevated value of a SAM in a clinical sample and the expression of clinical disease. In a disease that is typified by remissions and relapses, such as systemic lupus erythematosus, a candidate SAM, i.e., sIL-2R or soluble ICAM-1, measured prospectively on a set time basis, i.e., monthly, should permit the determination of the time that elapses between an elevated serum SAM level and clinical disease.

In conclusion, we believe that SAM present an opportunity to both learn more about the disease process and advance our clinical armamentarium. Careful clinical studies should not be avoided; instead, they should be carried out as early as possible in order to definitively address the question of clinical usefulness for any old or new SAM.

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