



A Technical Overview of the LRA by ELISA/ACT® Procedure

At least one out of every six Americans, or 40 million people in the United States, suffers from one or more of the autoimmune diseases. In response, Americans spend over \$300 billion per year directly for autoimmune syndrome treatments.¹ Many of these hypersensitive delayed allergy reactions are caused and/or aggravated by foods and chemicals. Autoimmune and immune dysfunction illnesses include chronic nasal congestion, asthma, migraine headache, pain syndromes, weight management, personality changes, unexplained depression, thought disorders, and metabolically based mood disorders.

The **Lymphocyte Response Assay (LRA) by ELISA/ACT®** procedure provides a unique and comprehensive solution for patients with signs of immunologic overload and its neuroendocrine consequences. Environmental physicians report that about 75% of all allergic/immunologic symptoms are caused by delayed allergy reactions.

A breakthrough in the complex world of clinical immunology, the **LRA by ELISA/ACT** tests uniquely measure all three delayed allergy pathways at the same time. With a simple blood draw, the “foreign invader” immunologic burden can be identified. The **LRA by ELISA/ACT** program enhances tissue repair and reduces interstitial (“third space”) swelling or “bloating.” The program also increases immunologic defenses in order to lighten the burden for patients suffering from a number of immunologic conditions. The net result is reduction in stubborn, chronic inflammatory and/or painful repair deficit conditions.

AN OVERVIEW OF DELAYED ALLERGIES

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Identifying the reactants that cause these symptoms is not an easy task. Physicians often find it difficult to prove they are due to immune-mediated reactions, intestinal-enzyme deficiencies, toxins or infections, or even neurological/psychological reactions. Historically, physicians have attempted to determine the presence of “food allergies” with a few tools: a detailed allergy history, a physical examination, and laboratory tests (primarily intradermal injections and blood tests). While these procedures may indicate that the patient has allergies, they are of little help in identifying their causes.² For example, an elevated total blood serum IgE indicates a patient has acute allergies but does not identify the source of the problem. The intradermal skin tests with commercial allergens used by traditional allergists are of limited value because of the large number of false-positive reactions in normal persons. None of the tests used by most allergists are of much help for delayed or hidden (Type II, III, and IV) hypersensitivities. Delayed food and chemical hypersensitivities are caused by the immune system’s inappropriate response to ingested or inhaled foods or chemicals and their metabolic consequences. Although an antigenic substance may seem harmless, the white blood cells identify it as an intruder, even if it is a potential nutrient, and will attempt to fight it off. When the total load of foreign material is excessive or when the white cells are already weak, attempts to break down and neutralize this intruder can damage white cells. Once damaged, white cells excrete powerful, potentially damaging, reactive materials into the bloodstream. If enough reactive excretion occurs, the weaker organs or systems in the body are symptomatically affected. Individuals suffering from recurrence of any of the symptoms listed in the following chart may be affected by delayed hypersensitivities.

¹ Based on data compiled by NIH (1997) and HCFA (1996).

² See *The Type I/Type 2 Allergy Relief Program* by Alan Scott Levin and Maria Zellerback for more technical detail about this difference.

Body Part	Symptoms That May Be Caused by Delayed Allergy Reactions
Head/Brain	Migraine ("allergic") and multiple sclerosis
Eyes	Blurring of vision due to retinitis
Ears	Ringing in the ears and earaches due to autoimmune meniere's syndrome
Nose	Rhinitis from chronic sinusitis
Throat	Cold and flu symptoms (recurrent due to weak/burdened immune system)
Heart rate	Palpitations, arrhythmia, and increased heart rate due to autoimmune myocarditis
Lungs	Asthma due to hypersensitivity
Intestines	Nausea, vomiting, diarrhea, and constipation due to inflammatory bowel conditions
Skin	Eczema and psoriasis
Muscle/Joint	Chronic muscle/joint aches and swelling, rheumatoid arthritis, and fibromyalgia
Bladder/Kidney	Unexplained salt/water retention due to immune complex activity

Some doctors recommend elimination diets for which patients eliminate suspected foods, keep a detailed food diary, and record results. Unfortunately, this procedure is both cumbersome to perform and imprecise. If a person is very hypersensitive, he/she can appear to be reacting to everything, and many people can then perceive themselves to be universal reactors. This, however, is usually due to nutrient and metabolic factors as well as hypersensitivities that provoke symptoms. These symptoms are often misinterpreted as only immune hypersensitivities until a comprehensive program like the LRA by ELISA/ACT treatment protocols are applied.

THE LRA BY ELISA/ACT PROCEDURE (ADVANCED CELL TEST)

Developed by Dr. Russell Jaffe in 1984, and now only available through ELISA/ACT Biotechnologies LLC (EAB), the LRA by ELISA/ACT, also known as the Advanced Cell Test, is five times more accurate than the best of previous tests. Using an average test result from 30-40,000 cells cultured, the LRA by ELISA/ACT procedure provides <.1% false-positives and <1% false-negatives. The LRA by ELISA/ACT has a low 3 percent day-to-day variance. Appendix 1 provides technical answers to method-related questions most often asked concerning the LRA by ELISA/ACT. Appendix 2 provides a comparison of various ELISA (serum) techniques with those of the LRA by ELISA/ACT. This figure not only illustrates how the LRA by ELISA/ACT procedure is processed, but also shows how the excellent accuracy is obtained. Appendix 3 shows a diagram of the four types of immune system responses and the types of diagnostic tests available to measure these reactions. It is important to note that *only* the LRA by ELISA/ACT measures all three delayed hypersensitivity reactions, thus providing a complete profile of the foods and chemicals a person should avoid.

The LRA by ELISA/ACT procedure is relatively simple for the patient. It requires a 12-hour fast³, followed by a one-ounce blood draw. Then, using fewer damaged cells (different from other tests) because of a specifically chosen blood draw procedure and a preservative uniquely formulated by EAB, the laboratory measures the reaction of the specially prepared white blood cells to as many as 400 items from the following general categories:

- Foods
- Additives/Preservatives
- Environmental chemicals and Toxic minerals
- Molds
- Danders, hairs, and feathers
- Medications
- Therapeutic herbs
- Food colorings

³ Interfering medications such as steroids, cortisone, aspirin, and antihistamines must also be avoided for four (4) days [steroids and cortisone] or two days [aspirin and antihistamines] as they inhibit important cell reactions and thus can mask hypersensitive reactivity. Call EAB at 800.553.5472 for a "medication bridge" if needed.

Upon receiving the test results, patients can be virtually certain of which of the 400 substances tested cause hidden problems and sometimes, even more importantly, which do not. For the first time, patients can accurately avoid substances that will allow the immune system to rebuild itself. To hasten this process, the test results may, by choice, include an individualized program combined with nutritional support to help the body heal itself.

The substances identified by the test are divided into three groups: strong reactions, moderate reactions, and nonreactions. Patients are advised to avoid substances causing strong reactions for at least six months and to avoid substances causing moderate reactions for at least three months.

After six months, a monitored reintroduction of the previously reactive foods can be offered to the patient. The patient may also choose to retake the test to document the degree to which his/her body has been able to heal itself. By retaking the test, the patient will be able to identify any new reactions that may have developed.

APPENDIX 1

FAQ ON TECHNICAL AND STATISTICAL INFORMATION SUPPORTING LRA BY ELISA/ACT TESTS

1. What does LRA by ELISA/ACT testing measure?

LRA by ELISA/ACT testing measures Type II, III, and IV delayed sensitivities, which include all Delayed Type Hypersensitivities (DTH) pathways.

NOTE: LRA by ELISA/ACT is the only assay that measures all delayed reactions in one sample at one time -- clinically significant humoral antibodies, immune complexes, and direct cell activation.

2. How are the LRA by ELISA/ACT results measured?

EAB uses an LRA-based, enzyme-enhanced ELISA/ACT technique.

3. Where do the antigens come from?

EAB isolates, prepares, and purifies its own antigens using the most advanced laboratory technique. This improves clinical success of LRA by ELISA/ACT testing and treatment programs.

4. How does LRA by ELISA/ACT testing compare with the RAST assay system?

The RAST assay system only tests for Type I [IgE] or acute reactions.

5. How does the precision of LRA by ELISA/ACT testing compare with other assay systems?

<u>Assay System</u>	<u>What is Measured</u>	<u>Method Variance</u>
LRA by ELISA/ACT	Type II, III, & IV (All delayed reactions)	≤ 3%
RAST [IgE]	Type I [IgE]	5-10%
ELISA [IgG]	Type II [IgG ₄ only]	8-15%
ELISA [IgG ₄]	Type II [IgG ₄ only]	10-20%
Immune Complex	Type III	10-20%

6. How does the LRA by ELISA/ACT compare with other ELISA assays?

<u>Assay System</u>	<u>What is Measured</u>	<u>Expected Correlation</u>
LRA by ELISA/ACT	Type II, III, & IV (All delayed reactions)	-100% DTH reactions
ELISA [IgG]	Type II [IgG ₄ only]	-25% DTH reactions
ELISA [IgG ₄]	Type II [IgG ₄ only]	-40% DTH reactions
Immune Complex	Type III	10-50% DTH reactions

Conclusion: LRA by ELISA/ACT provides the most clinically useful information with the fewest false-positive and false-negative results by using autologous cell-rich plasma (not serum).

7. What contributes to the precise results of LRA by ELISA/ACT tests?

EAB's unique, five-step quality control program contributes to the precise results of LRA by ELISA/ACT tests as outlined below:

- A. ANTIGENS purified for procedure to make "smart plastic microtiter plates"
- B. ANTICOAGULANT developed specifically for this assay to put cells in suspended animation
- C. CUSTOMIZED VACUTAINER technique for phlebotomy reduces artifacts from the blood draw, consumption of complement, prothrombin, kinyogen, Ig, etc. [prevents non-specific destruction]
- D. INCUBATION under physiologic conditions [*ex vivo*]
- E. REACTION ON CELL SURFACE by the ELISA/ACT technique. This allows simultaneous measurement of:
 - Humoral antibodies [immunoglobulin]
 - Immune complexes [multiple IgM, anti-IgG with complement] to use their native, physiologic surface to show action
 - Direct cell activation [lymphocyte]

APPENDIX 1, CONTINUED

8. How stable is the blood sample?

The blood sample is stable for at least 72 hours, assuming the sample is kept according to EAB's quality control requirements as detailed below:

- Between 4-10 C°
- In an all-plastic environment
- In a suitable shipping environment (provided by EAB)

9. How do healthy people respond?

Healthy people with <5 symptoms (according to the *Cornell Medical Index*) respond with just 3 ± 1.5 test positives out of 234 items tested. In a small group of asymptomatic, healthy people there were 0.1 ± 0.5 test positives out of 234 items tested.

10. How do chronically ill people respond?

Chronically ill people with >25 symptoms (according to the *Cornell Medical Index*) respond with 18 ± 3.2 test positives out of 234 items tested.

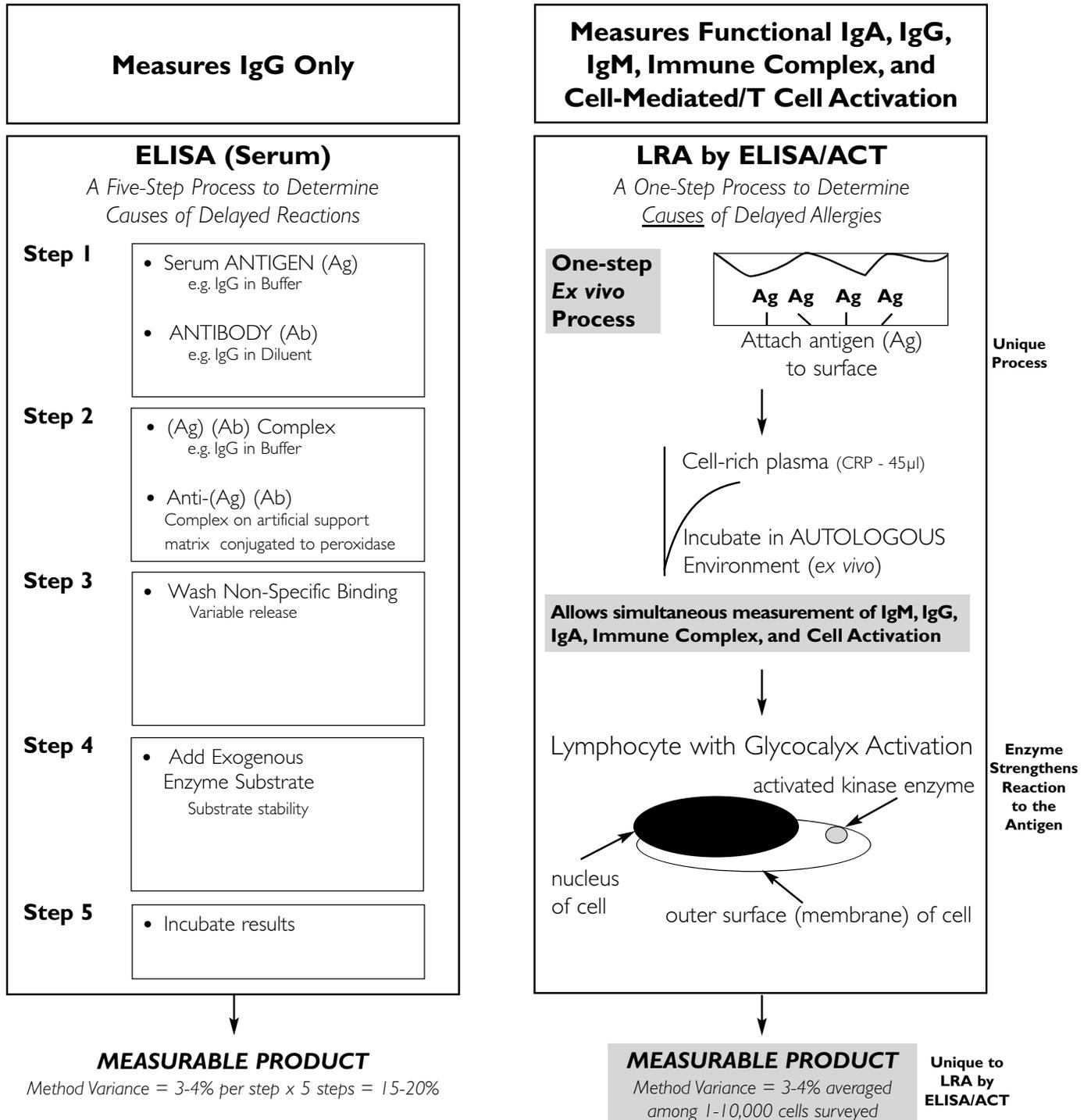
11. What clinical conditions is the LRA by ELISA/ACT procedure suitable for?

Below is just a sampling of the approximately 900 now recognized autoimmune conditions for which the LRA by ELISA/ACT is suitable.

- Autoimmune conditions
- Hashimoto's thyroiditis
- Inflammatory bowel disease (IBD)/Ulcerative colitis
- Fibromyalgia
- Migraine or related pain syndromes
- Myofascitis
- Diabetes (Adult and Insulin Dependent)
- Multiple sclerosis
- Sjogren's rheumatoid syndromes
- Lupus erythematosus (SLE)
- Psoriasis and psoriatic arthritis
- Treponemal hypersensitivity, 4° syphilis
- Glomerulonephritic syndromes
- Chronic viral syndromes (CMV, EBV, Herpes, etc.)
- Chronic fatigue/CFIDS
- Thyroiditis

APPENDIX 2

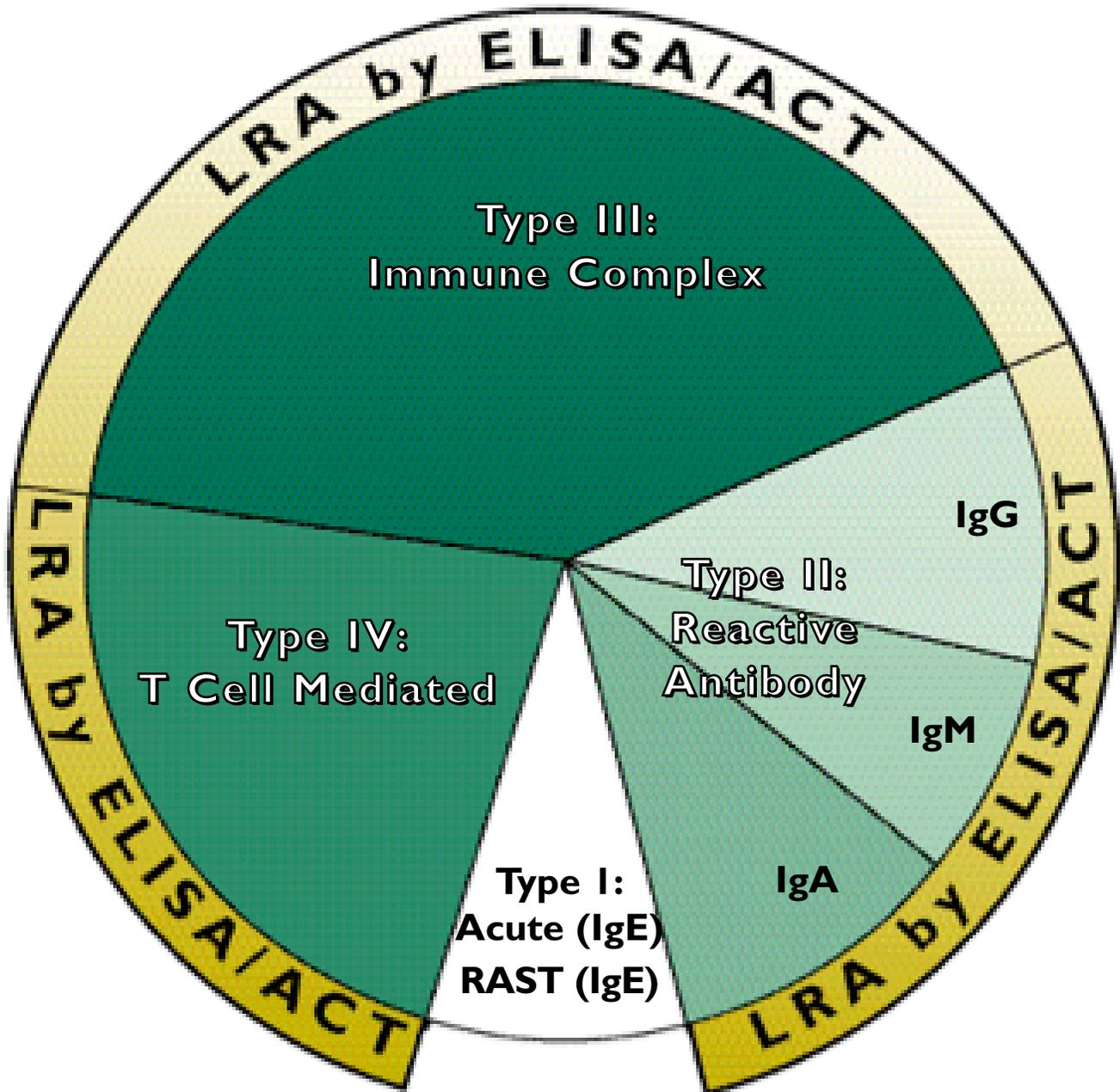
ELISA TECHNIQUES: A COMPARISON



The LRA by ELISA/ACT one-step, integrated method yields precise, reliable results in the analysis of delayed hypersensitivity. It is the only system able to measure concurrently all causes of delayed hypersensitivity. The LRA by ELISA/ACT test should not be confused with the ELISA IgG tests because *only* the LRA by ELISA/ACT is full spectrum, full function, and fully supported by clinical and basic science expertise “in house.”

APPENDIX 3

Functional lymphocyte response assays (LRA) are able to measure all delayed allergy responses.



LRA by ELISA/ACT® is a true cell culture. Comprehensive, ex vivo, functional procedures have been proven in clinical outcome studies to provide superior, sustained improvements and long-term remissions in autoimmune and immune dysfunction conditions.

SCIENTIFIC REFERENCES

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Note: For additional literature, please contact EAB's Client Services Department at 800.553.5472.



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