New peanut allergy test goes beyond scratching the surface

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Professor James Rusling (left) and Associate Professor Mark Peczuh, both in the Department of Chemistry, are pictured with the object of their research. Credit: Peter Morenus/UConn Photo

Current peanut allergy tests are not very reliable when it comes to diagnosing the severity of an individual's allergic reaction, which can range from hives to life-threatening anaphylactic shock.

With an estimated three million people in the United States allergic to

peanuts and tree nuts, having a more precise and reliable allergy test could prevent hospitalizations and allow for better monitoring of individuals suffering from peanut allergies.

Three chemists at the University of Connecticut (UConn) are developing a more advanced peanut allergy test that, based on initial results, is many times more sensitive than current procedures. The new test is capable of determining the potential intensity of a patient's allergic reaction through just a few drops of blood.

Understanding how the new test works requires a basic understanding of how allergic reactions happen. When an allergic person eats peanuts, their immune system releases an antibody protein known as immunoglobulin E or IgE. These antibodies fight off peanut allergen molecules by binding to them and flushing them out of the body. But the release of the antibodies causes tissue cells in the body to produce histamine, which in turn generates a variety of allergy symptoms such as itchy skin, runny nose, coughing, or wheezing. The more antibodies that are released, the more histamine is generated, the stronger the person's allergic response.

"A patient who has a serious allergy and gets exposed to an allergen protein will form antibodies in their body that should stay there for awhile," says UConn Professor James Rusling, who specializes in detecting protein biomarkers and used a similar process to detect proteins linked to cancer. "Our theory is that the level of those antibodies can be used to predict how severe a patient's allergy is at any one point in time."

While existing peanut allergy tests can generally measure IgE antibodies found in a blood sample, the presence of other biomolecules can distort the results and they are not always accurate.

The allergy test designed by Rusling, Mark Peczuh and Challa Vijaya Kumar screens out other biomolecules and measures the presence of antibodies that bind to very specific protein fragments, called peptides, and carbohydrate residues found in peanuts.

"The traditional method of measuring these antibodies uses a mixture of all the peanut proteins, not individual parts," says Peczuh, a specialist in carbohydrate synthesis whose daughter has a peanut allergy. "But some of the stuff in the mixture can lead to readings that a patient is allergic when she or he is not. And the converse can be true, where the results show someone is not allergic when they actually are."

In the study of their new system, the UConn chemists tested three components from the most potent peanut allergen. One sample was a protein peptide, another a carbohydrate residue, and the third was a positive control.

The chemists then injected blood serum from patients known to have peanut allergies into the array. As the blood serum floated over the samples, IgE antibodies were pulled down by the allergens and bound by them. They could then measure the quantity of antibodies to determine how strong a reaction a person would have to peanuts. To further refine the system, the team attached magnetic beads to the allergen samples. The beads captured the IgEs and amplified the final measurements, allowing them to detect concentrations of antibodies as low as 0.5-1 picogram per milliliter.

The test results correlated with the patients' known allergy levels from other tests and the team was encouraged to pursue further development of the approach. UConn Graduate student Amit Joshi performed most of the experiments in the initial test.

While the trial test was limited to just a few allergic components from

peanut glycoproteins, Rusling says it could be expanded to screen for more than 20, allowing for even more selective results.

Although the initial results are promising, the time frame for any clinical use of the test is still years away.

"Eventually, we'd like to use maybe five different peptides and carbohydrate samples to see how these IgEs bind to them," says Rusling. "That way, we could determine a clear fingerprint of a patient's susceptibility to a specific allergen."

There has been some debate over the role carbohydrates play in allergies. Because the UConn test has the capacity to test both protein peptides and carbohydrate residues, the researchers hope it can be used to learn more about how specific protein and carbohydrate epitopes bind to antibodies to gain a better understanding of how allergies are induced.

"Our hope is that this could be used as an analytical tool to investigate the actual biology of the allergic response to peanuts and other food items in general," says Rusling. "People have noted that certain carbohydrates may be involved in allergies and we'd like to determine whether they are involved or not."

More information: "Ultrasensitive carbohydrate-peptide SPR imaging microarray for diagnosing IgE mediated peanut allergy." *Analyst*. 2014 Nov 21;139(22):5728-33. DOI: 10.1039/c4an01544d.

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