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Scientists are working on a feline vaccine that could block the protein most commonly associated with allergic reactions to cats.

The race to deliver the hypoallergenic cat

Researchers are looking beyond allergen immunotherapy to help people whose pets make them sneeze. **By Amber Dance**

"Doctor, if you tell me to get rid of the cat, I'm getting rid of my kids first," said the parent of one young patient with an allergy to the family pet. Sandra Gawchik, an allergist in Chester, Pennsylvania, knew they were only half-joking – she had heard similar threats before. Another patient recounted being pushed out of the house by his wife and daughter over his cat allergy, saying they preferred the feline to him.

Sensitivity to pets (cat and dog allergies occur at similar rates) is typically the second most prevalent allergy in any region, after pollen or mites, says Joaquin Sastre, an allergist at the Jiménez Díaz Foundation University Hospital in Madrid. Up to 30% of people show sensitivity to cats in some regions.

The effects can be limited to sniffles and sneezes for some people. For others, however, the allergy can prompt dangerous asthma attacks. A US study estimated that, among cat-sensitive people, 47% of emergency hospital visits could be attributed to feline exposure[1]. And allergy is among the main reasons why owners send cats to shelters.

Small doses of cat allergens – the major one being a small, sticky protein called Fel d 1 – can help to build up tolerance over time. But treatments can cost US$800–1,000 annually, require up to 100 injections over 3–5 years, and are not always permanent or wholly effective.

Some researchers are trying to improve this immunotherapy, and others are trying a different angle by injecting Fel d 1 antibodies into people with allergies. Scientists are also seeking ways to neutralize Fel d 1 before it leaves the animal – or even stop cats from producing it entirely.

Sticky stuff

Cats secrete the Fel d 1 protein from their salivary and sebaceous glands, and spread it throughout their fur during regular tongue baths. Released in shed skin and hair, it floats in the air for hours, attaching to carpets, curtains and clothing. Some cat owners with allergy concerns invest substantial time in cleansing

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not only their houses but also their pets; however, the protein is difficult to eradicate. "You'd have to bathe the cat every other day," says Gawchik. And because Fel d 1 sticks to cat owners' clothing, it can show up in places where no feline has ever set paw.

Fel d 1's tiny dimensions also make it a tricky customer. At less than one-tenth the size of a single ribosome, it can slip deep into the lungs and trigger asthma, alongside itchy eyes and nasal congestion. Despite these difficulties, however, cat allergy could be seen as a low-hanging fruit for allergy researchers. Although at least eight different cat proteins can cause allergy, Fel d 1 is by far the major contributor. Tamping down a person's response to just this one protein, or reducing the presence of Fel d 1 in the cat and its environment, could minimize symptoms across a wide swathe of feline-sensitive people. Unlike in dogs, which produce several key allergens, scientists need only hit the bullseye once in cats.

People who are allergic to Fel d 1 react by creating immunoglobulin E (IgE) antibodies. These induce mast cells to spew out histamine and other chemicals that create sneezing, itching and congestion. A less unpleasant response to the protein would be one involving IgG antibodies – the type made in response to viruses – which are not generally associated with allergic reactions. People who had cats as children and who have an overall lower risk of allergy tend to develop this kind of response, notes Martin Chapman, chief executive of Indoor Biotechnologies in Charlottesville, Virginia, which specializes in products and laboratory services linked to indoor air quality, allergies and asthma.

Current immunotherapy for allergies trains the body to make IgG antibodies that grab antigens before IgE can lock on. The injections contain purified cat allergens, or a broader mix of cat proteins. Overall, immunotherapy is up to 70% effective, says Gawchik, but it only works "if the patient religiously gets shots, on a weekly basis", and some people return for booster injections years later because their allergies have returned. Sastre says that nearly one-third of his patients still need some form of anti-allergy medication, such as antihistamines, to keep their symptoms in check. And in some cases, the small doses of allergen cause the very reactions they are meant to stifle. That is why researchers see room for improvement.

The Immune Tolerance Network, a collaboration funded by the US National Institute of Allergy and Infectious Diseases, hopes to make immunotherapy work better and faster by interfering with an allergy-driving protein called thymic stromal lymphopoietin (TSLP). In a phase I and II trial, participants received weekly immunotherapy and a monthly infusion of antibodies against TSLP.

"We hope to induce a state of tolerance that would potentially occur more quickly, last longer and reach a higher level," says principal investigator Jonathan Corren, an allergist and immunologist at the University of California, Los Angeles. The group plans to publish its findings soon. "The results, at this point, do look promising," Corren says.

The antibody therapy would be costly, Corren adds, so would be best suited for people with severe allergies or asthma. But it needn't be restricted to those sensitive to cats. "The approach should be applicable to any number of allergens," he says.

Adiga Life Sciences in Huntingdon, UK, is trying a different approach, substituting Fel d 1 for Cat-PAD, which consists of smaller, synthetic peptides based on Fel d 1. The hope is that a synthetic allergen would be less likely to cause a reaction and could therefore be given in higher doses – potentially enabling people to achieve tolerance in just four treatments.

In a phase II trial, 11 participants who received Cat-PAD saw their symptoms reduced for up to 2 years[2]. But a 2016 phase III study failed when people receiving a placebo developed just as much tolerance to Fel d 1 as did those on the immunotherapy. Mark Larché, an immunologist at McMaster University in Hamilton, Canada, and a consultant for the company, speculates that part of the problem might have been that all the phase III participants had to be cat owners. As a result, even those in the placebo group would have daily exposure to Fel d 1, potentially promoting tolerance.

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Adiga is now planning a trial with non-cat-owners. Larché says that participants will be tested for allergy symptoms by sending them to cat cafes (theme cafes where patrons can watch or play with cats). The company is also pursuing peptide-based immunotherapy for allergies to grass[3] and dust mites, Larché says, and has designed peptides for birch allergy – a common problem in northern Europe and the northeastern United States.

All of these immunotherapies ultimately rely on patients generating "a very specific immune response to the allergen", says immunologist Jamie Orengo at Regeneron Pharmaceuticals, a biotechnology company in Tarrytown, New York. If a person cannot make a potent enough response, they will not build up tolerance. But Regeneron aims to circumvent this issue by dosing people directly with Fel d 1 IgG antibodies.

The treatment is based on two IgG antibodies that cover up key IgE binding sites on either end of the allergen. Together, they can block up to 83% of the IgE–Fel d 1 interactions responsible for most allergic reactions to cats. In a phase Ib test, people with a cat allergy received a single dose of the antibody combination[4]. Then, in four tests conducted between eight days and nearly three months later, researchers sprayed cat hair extract into the participants' nostrils. Symptoms dropped in 60% of participants, by an average of 60%, Orengo says. Regeneron is now running a phase II trial.

The advantage of the antibodies, says Orengo, is that they will work more quickly than immunotherapies that require the patient to build tolerance. However, because the patient will not be producing the antibodies themselves, regular boosters will be required, perhaps quarterly. The team is also working on a similar treatment against birch pollen, and is considering other allergens, too.

"This is certainly an interesting approach," says Martin Bachmann, an immunologist at the University of Bern. "But it's probably going to be kind of expensive."

Immune to the allergen

Bachmann is among the scientists who are targeting Fel d 1 in cats themselves. If the animals did not produce the protein, this should prevent allergic reactions in most people. But researchers are unsure how thoroughly the protein needs to be eliminated to have the desired effect, as well as what happens to a cat without Fel d 1.

No one knows what Fel d 1 does for felines. Some cats have a little and some have a lot, so large amounts don't seem to be necessary. And the cat's breed doesn't seem to make much difference; even hairless Sphynx cats make Fel d 1. On average, unneutered males have the most and females the least, suggesting the protein might carry hormones or pheromones. But it has also been hypothesized to protect cats' skin.

In 2013, Bachmann co-founded HypoPet, a spin-off company from the University of Zurich in Switzerland. The firm aims to create hypoallergenic cats by vaccinating them against their own Fel d 1, so that the cat's IgG antibodies block the protein before it is sniffed up by a human.

Bachmann built the vaccine out of recombinant Fel d 1, a bit of tetanus toxin peptide and the coat from a plant virus. "The immune system thinks it's a virus," he says, and accordingly responds with IgG (see 'Immunizing cats

IMMUNIZING CATS AGAINST THEIR OWN ALLERGEN

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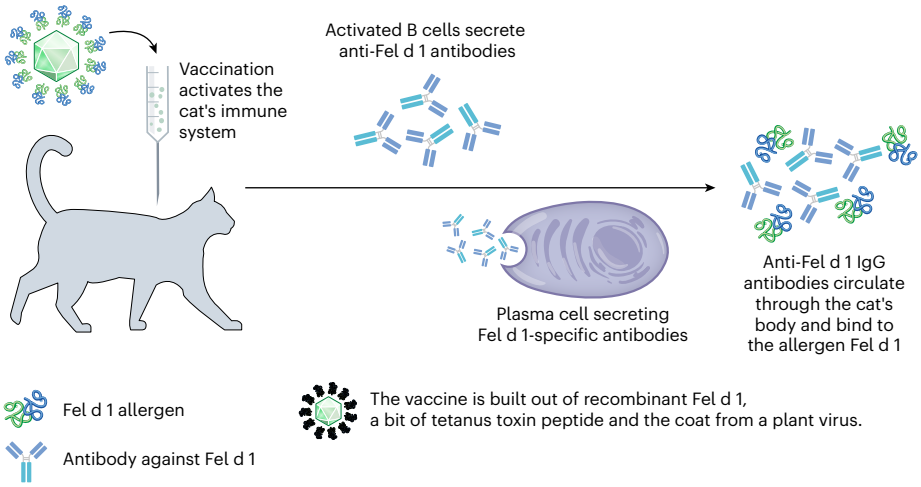

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against their own allergen'). In an early test, the level of Fel d 1 in the tears of 18 cats fell by more than half 42 days after vaccination[5]. "Every cat makes a good IgG response," he says, and the vaccine doesn't seem to cause them any problems.

The team then tested the vaccine in 13 cats owned by 10 people with cat allergies. They asked the owners to report on their symptoms, as well as how long they could stand to pet their cat before it became uncomfortable. Seven of the pet owners saw their symptoms decrease, and 8 of them were able to extend their petting time from an average of 16.9 minutes before the vaccine to 27.7 minutes after[6].

Bachmann isn't sure how long the vaccine's effect will last, and annual booster injections might be necessary. Another challenge HypoPet faces is to convince regulators to approve a vaccine that doesn't directly improve the cat's welfare. In the United States, a direct benefit to the cat is not a requirement, but in the European Union it is. Bachmann is speaking to authorities in both territories.

He also thinks that the vaccine could be used in people, and he tried an early version[7] on himself, with benefits for his cat allergies. He hopes to bring the vaccine to human clinical trials soon, along with another using the same approach against peanut allergy. He is also thinking about generating hypoallergenic dogs, although multi-component vaccines would be required to cover their multiple allergens.

Tasty solution

Just as Regeneron is exploring the possibility of delivering antibodies directly to patients, some are taking a similar approach to dealing with Fel d 1 in the cats themselves. Pet-food company Nestlé Purina in St Louis, Missouri, offers an antibody-coated product that is designed to neutralize the feline allergen. Known as Pro Plan LiveClear, it was released earlier this year in the United States, Europe and Russia, and is expected to launch in other territories in 2021.

Ebenezer Satyaraj, an immunologist at Nestlé Purina, was inspired by personal experience – his daughter broke out in a rash after visiting a cat-owning friend. When he and his colleagues started looking for a way to neutralize Fel d 1, they landed on chicken antibodies.

The chicken version of IgG is called IgY, says Satyaraj, and this can be found in the egg powder that has long been used to supplement animal foodstuffs. His team contracted with a company that exposed hens to Fel d 1, so that the chickens naturally made IgY against the cat protein, which was transferred to their eggs. The company provided egg powder loaded with these anti-Fel d 1 antibodies, and Purina coated its pet food with it.

More than two dozen cats that ate the food for six months appeared totally normal, when subjected to a battery of behavioural tests[8]. In another study of 105 domestic shorthair cats[9], it took about 3 weeks for the diet to have an effect on Fel d 1, eventually reducing the concentration in hair and fur by an average of 47%.

That might not be enough to help the most-sensitive allergy sufferers, says Gawchik, but it could make a difference for some. In an as-yet-unpublished study, researchers at Washington University in St Louis found that people with a cat allergy had fewer symptoms when exposed to skin flakes and fur from cats eating the special food than when exposed to skin and fur from cats eating a control diet.

Chapman hopes that his team at Indoor Biotechnologies will be able to make a truly hypoallergenic feline by eliminating Fel d 1 completely. They are in the early stages of developing a gene therapy against the two genes that encode the subunits of Fel d 1. Eliminating them from the salivary and sebaceous glands should create a hypoallergenic cat.

Chapman and his colleagues first had the idea in the 1990s, when the Fel d 1 genetic sequences were identified[10], but the rise of CRISPR gene-editing technology has provided an opening to try it. Chapman already holds a patent on the idea, but it will take a lot of work before allergen-free kitties are on offer.

Nicole Brackett, a postdoctoral researcher at the company, has analysed the Fel d 1 sequences of dozens of cats. From this, she was able to design guide RNAs that can be used to bring about a CRISPR-based disruption of the Fel d 1 genes. So far, she has managed to destroy the Fel d 1 sequences in an immortalized cat kidney cell line, with efficiencies of up to 55%.

The company plans to treat cats individually, rather than breed hypoallergenic kittens, so it needs a way to deliver the gene therapy to the glands in grown cats. Brackett is considering using a viral vector in an injected or topical solution. "We will definitely be watching this space," says Orengo. But she notes that even if Chapman and Brackett could eliminate Fel d 1 from a customer's cat, the owner would still be likely to encounter the ubiquitous allergen. Ideally, Bachmann says, allergy sufferers would take a multi-pronged approach. "You want to tackle it from both ends," he says. "You desensitize the allergic humans; at the same time you render the cat hypoallergenic."

Gawchik is intrigued that LiveClear cat food might allow her patients to take exactly that dual approach today. She plans to recommend it to patients and cat-sensitive members of her family, too. "It's worth a try," she says. Perhaps this, or other options, might one day mean cat lovers don't have to choose between their feline and human loved ones.

Amber Dance is a freelance science journalist in Los Angeles, California.

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