AllerGen

Innovation from cell to society

Fall 2010

Success Stories

Networks of Centres of Excellence

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Making a Difference from Cell to Society

AllerGen NCE Inc. (AllerGen), the Allergy, Genes and Environment Network, is pleased to present the first in an ongoing series of publications featuring significant research accomplishments of leading Canadian allergy, asthma, anaphylaxis, genetics and environment researchers and their partner organizations.

In this inaugural issue of *AllerGen Success Stories*, we are delighted to highlight five cutting-edge research and development initiatives from across Canada.

Featured projects illustrate:

- How products found within many Canadian homes impact childhood asthma
- Parallels between allergic reactions in horses and humans
- Global best-practices for Phase II trials for new asthma drugs
- Diagnostic tests to identify peanut allergy
- The impact of maternal stress in the development of asthma in young children.

AllerGen's research, networking, commercialization, knowledge mobilization and capacity building activities are aimed at reducing the morbidity, mortality and socio-economic impacts of allergic diseases nationally and globally. Since 2005, AllerGen NCE has fostered the development of a multi-disciplinary national and international network of research experts in allergic and related immune diseases.

AllerGen's invests in research about:

1. Gene-Environment Interactions

Strategic Focus: Genetics, environmental exposures and gene-environment interactions in allergy and asthma

2. Diagnostics and Therapeutics

Strategic Focus: Biomarkers, immune monitoring and drug development/discovery

3. Public Health, Ethics, Policy and Society

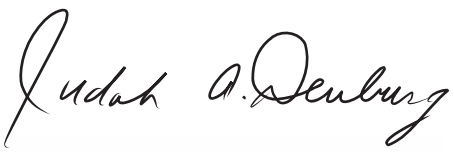
Strategic Focus: Allergy and immune disease prevalence, surveillance and management, evidence-informed public policy and knowledge translation.

AllerGen NCE also supports four cross-programmatic, multi-disciplinary research teams:

i. The Canadian Healthy Infant Longitudinal Development (CHILD) Study (a national birth cohort study)
ii. Food Allergy and Anaphylaxis – the Canadian Group on Food Allergy Research or CanGoFAR team
iii. Mind-Body Interactions and Allergic Disease
iv. Occupational and Work-related Allergy and Asthma.

Through its network of allergy and immune disease research experts and partner organizations from across sectors, and with core support from the Networks of Centres of Excellence (NCE) – a program of Industry Canada, AllerGen NCE is proudly supporting excellence in science and fostering innovation that advances knowledge about allergy, asthma and related immune diseases that contributes to improving the health and productivity of Canadians.

It is our hope that these stories will accelerate dissemination, discussion and translation of AllerGen research results and provide allergy, asthma and related immune disease sufferers, as well as their families and friends, with practical information about the forward-thinking research that is being conducted in Canada to reduce the global burden of allergy, asthma and related immune diseases.



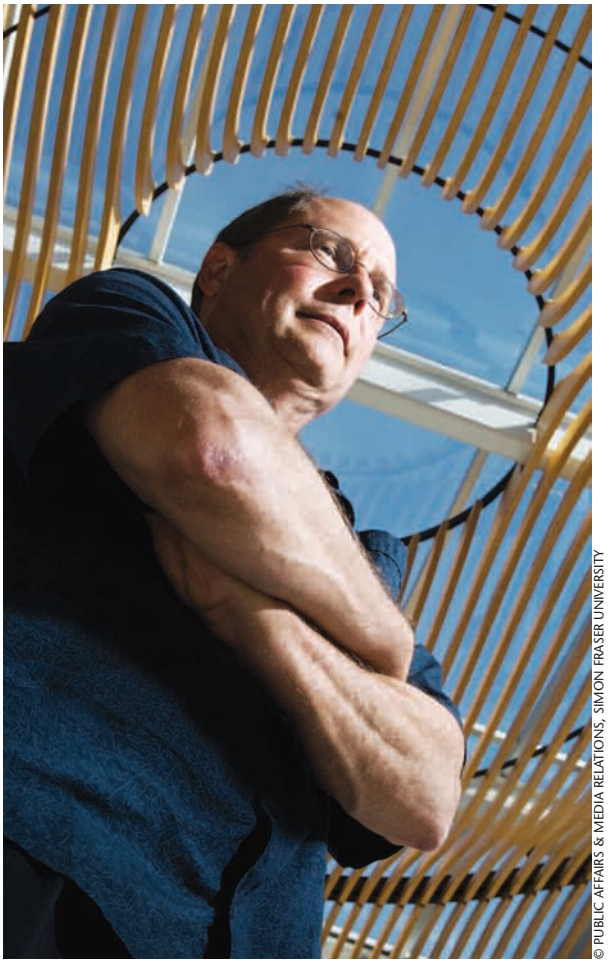
Judah Denburg, MD, FRCP(C), Scientific Director and CEO

Diana Royce, EdD, Managing Director and COO

The National Health and Nutrition Examination Survey (NHANES) found that over 95% of the entire population has phthalate residues in their urine — which means that these chemicals are getting into our bodies — through our noses, skin and mouths.

Babies' Exposure to Phthalates in the Home: Developing a Profile of Allergy and Asthma

How can you detect the presence of industrial chemicals called 'phthalates' in babies without taking a urine sample and ordering an expensive laboratory analysis? A multi-disciplinary team of researchers led by Dr. Tim Takaro, an Associate Professor at Simon Fraser University, discovered that you can predict part of the body burden of phthalates in a baby's urine by simply asking his or her parent's questions about the baby products that they use and the toys the baby plays with.



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Dr. Tim Takaro, Associate Professor, Simon Fraser University.

This research is an example of how collaborations across Canadian universities and partnerships between academia and industry breed success — which is why AllerGen NCE actively promotes broad professional relationships through networking and funding. On this multi-disciplinary team are researchers from Simon Fraser University (Drs Takaro and Ryan Allen); University of British Columbia (Drs Michael Brauer and Stuart Turvey); University of Toronto (Dr. James Scott); and Environment Canada (Dr. Jeff Brook). This team of environmental scientists and clinician-scientists in paediatrics, allergy/immunology and respiratory medicine, also partnered with the Canadian Housing and Mortgage Corporation on this project.

This study found a simple way of assessing babies' exposure to phthalates. The next step is the most wide-ranging investigation ever done in Canada on babies and the development of allergy and asthma, referred to as the Canadian Healthy Infant Longitudinal Development (CHILD) Study. This research team and other AllerGen-affiliated researchers are concerned with the larger question of what causes childhood allergy and asthma — chronic diseases that are rising in Canada at alarming rates. The widespread presence of phthalates in Canadian homes is just one factor being examined by the team.

What Are Phthalates?

Phthalates are industrial chemicals that are used to make an astounding number and variety of products found in the average Canadian home, including: personal care products for adults and kids; cosmetics; perfumes; cleaning and laundry detergents; lubricants; packaging; food; food containers and wrappers; drugs; fabrics; shower curtains; vinyl flooring; electronic equipment; paints; building materials; baby and toddler pacifiers; plastic feeding bottles; and children's toys. This is by no means a complete list.

Phthalates crept into widespread use over the last several decades because of their ability to increase a product's flexibility, transparency, durability and longevity. The problem is that phthalates do not chemically bind to the other materials in a product, so they continually leach out into the surrounding environment — like your home or baby bottle. In the United States, the *National Health and Nutrition Examination Survey* (NHANES) found that over 95% of the entire population has phthalate residues in their urine — which means that these chemicals are getting into our bodies — through our noses, skin and mouths.

Significant evidence exists suggesting that phthalates have a negative impact on human health. Scientists also wonder if phthalates have the ability to exacerbate allergic reactions to an allergen — a possible clue as to why allergy rates have exploded in industrialized nations around the world.

Looking for New, Inexpensive Ways of Assessing Phthalates Exposure

Canadian researchers like Dr. Takaro are trying to understand the causes of childhood asthma — a chronic and, therefore, lifelong disease in which the airways of the lungs are often inflamed, even when the patient is not experiencing breathing difficulties. According to the Asthma Society of Canada, currently one in eight Canadian children suffers from this disease and it is the leading reason for Canadian children missing school or being admitted to hospital.

Dr. Takaro and his colleagues are working towards developing a profile of children with allergy and asthma in Canada within the CHILD Study. One of the research questions that this study asks is "what is the possible role phthalates play in allergy and asthma?" The first step towards answering this question was to find a new, relatively quick and inexpensive way of assessing babies' exposure to phthalates. Exposure to phthalates is usually ascertained by analyzing a urine sample. However, it is quite expensive to undertake this laboratory analysis and it can also be difficult to collect a urine sample from very small children. Given the size of the CHILD Study, which is tracking the health of 5,000 Canadian babies, practical considerations such as cost and ease of assessing phthalate exposures become important.

Phthalates crept into widespread use over the last several decades because of their ability to increase a product's flexibility, transparency, durability and longevity. The problem is that phthalates do not chemically bind to the other materials in a product, so they continually leach out into the surrounding environment — like your home or baby bottle.

As a result, Dr. Takaro and his team participated in a 'mini-study' in Vancouver, involving 94 babies, all around three months of age. At this young age, children do not have asthma — although approximately 12% of Canadian children will eventually develop this disease.

The team collected a lot of information about the babies (beyond phthalate exposure) using four methods: 1) a questionnaire about the home environment and products found in the home, which was completed by parents; 2) home inspections by trained assessors looking for potential asthma triggers; 3) collection and analysis of dust samples from the floor; and 4) collection and analysis of urine samples for levels of phthalates.

Three month old infants from Vancouver had a range of detectable phthalates in their urine at levels comparable to recently surveyed U.S. children. For some phthalates, questionnaires could predict risk of phthalates in a baby's urine by know-

ing what products went directly on his or her skin (such as lotion and shampoo) and directly into the baby's mouth (such as pacifiers, baby bottles and toys made of soft plastic). Further analyses are ongoing, but it does appear that questionnaires about baby products in the home, along with home inspections by trained assessors, are a cheap and effective alternative to urine tests for assessing phthalate exposures.

The CHILD Study — On the 'Cutting Edge' of Canadian Research

The CHILD Study is a pioneering study of how genetic susceptibility and environmental exposures combine (gene-environment interactions) in the development of childhood asthma. "This collaborative Canadian initiative has united experts from AllerGen NCE's research program with public and private sector organizations," states AllerGen NCE CEO and Scientific Director, Dr. Judah Denburg. "It is a Canadian first in allergic disease research."

Takaro's 'mini-study' of 94 babies living in Vancouver proved to be a useful 'pilot' study for the CHILD Study — a pilot study is a small scale preliminary study conducted before the main research is undertaken in order to improve the final research design. Dr. Takaro and his team were successful in finding a relatively inexpensive and reliable method for assessing phthalate exposures in babies in the form of questionnaires and home visits. They developed several research guidelines for exposure assessment in the CHILD Study. They trained a group of people and put together a manual in the methods for doing effective home assessments and for collecting dust and urine samples. They also collected data for future analysis on other indoor factors that could contribute to allergy and asthma — like dust mites, cockroaches, rodent droppings and traffic pollution entering the home. Allergy and asthma experts realize that in analyzing the health effects of indoor contaminants, they cannot focus on just one thing (like phthalates) because contaminants likely interact with one another.

The CHILD Study also collects data on other potentially important factors in the development of allergy and asthma besides indoor and outdoor contaminants, such as: nutritional history, infectious exposures, psycho-social stress, immune system function and genetics.

The challenge for researchers is to develop a profile of allergy and asthma that can successfully understand and predict which children will develop asthma and which will not and then to design and deliver public health strategies and policies to prevent asthma. Dr. Takaro anticipates that, "In 7 years from now, the CHILD Study will pay off. We will be able to say that of the 5,000 children studied who developed asthma, this is their profile: they have these kinds of homes; they have these sorts of exposures; they live this far from a freeway — they have these sorts of behaviors in their childhood that increase their phthalates exposure — they have this interaction between diesel and dust mites or mould and phthalates — and people with gene X, Y and Z, are much more susceptible to these exposures than people who have genes A, B and C."

Dr. Takaro describes the CHILD Study as "cutting-edge" research because of the techniques being used, because it considers multiple exposures such as phthalates and traffic pollution, which hasn't been done before, and because it involves researchers with different and varied areas of expertise working together towards a common goal. Dr. Takaro credits AllerGen NCE with bringing multi-disciplinary teams together through its networking efforts, and through its conferences and annual meetings where researchers can present and discuss their work. He adds that, "Part of the cohesiveness of this group is the presence of funding (by AllerGen NCE) that places a high value on interdisciplinary work. Not all funding does that."

Beyond collaborations across Canada, there may be future opportunities between the Canadian researchers working on the CHILD Study and international research teams studying birth cohorts. Dr. Takaro is excited about the future possibilities, "You need a body of work — more than one study — to support a new conclusion about gene-environment interactions, or about [the cause of a] disease… Every study that contributes to that increases the confidence that scientists have in the results."

If AllerGen NCE and CHILD Study researchers have their way, future generations of Canadian children will be spared the burdens of living with allergies and asthma. Dr. Takaro states that, "There is evidence that if you intervene early, you can prevent asthma from occurring. Asthma is rarely diagnosed before the age of five." Profound changes are needed in how we diagnose, treat and manage allergy and asthma, and Takaro's science is helping to guide us in the right direction.

"Wherever man has left his foot-print in the long ascent from barbarism to civilization we will find the hoof-print of the horse beside it."
— John Moore

A Tale of Horses and Humans: Lung Health Parallels

Since the dawn of mankind, horses have helped humans not only survive, but to thrive. As human society evolved over the centuries, so did our relationship with these beautiful animals.

Dr. Jean-Pierre Lavoie, a Professor and researcher at the University of Montreal, as well as a practicing veterinarian, is passionate about horses and is an expert in horse internal medicine. For the past twenty years, Dr. Lavoie has been researching 'heaves,' a life-long and debilitating respiratory disease that is estimated to affect 15% of all horses in Canada. The first step in the research process was to develop techniques to measure lung function in horses. Dr. Lavoie sought advice from medical experts on the already well-established techniques for measuring human lung function. A particularly valuable collaboration was established at that time with McGill University's expert in lung mechanics, Dr. Jason Bates. These collaborations helped Dr. Lavoie to adapt the techniques for use in horses and led to the realization that there are startling similarities between heaves and human asthma. This AllerGen-supported research on heaves will benefit not only horses afflicted with this disease, but it also has the potential to be useful in helping people who suffer from asthma.

Dr. Jean-Pierre Lavoie, Professor and researcher, University of Montreal.

'Heaves' and Asthma — Parallel Airway Diseases

'Heaves' is a chronic (meaning life-long) condition. It is an allergic reaction in horses to dust and mold spores present in the barn or stable, particularly in the hay and straw that they eat and sleep on. It is characterized by wheezing, coughing and laboured breathing, and can limit the horse's ability to be ridden or worked.

'Heaves' is common in the northern hemisphere, but rare in the southern, probably because northern horses are more likely to be over-wintered in stables or barns and, therefore, are more likely to become sensitive to mold and dust. There is also a genetic component to heaves — horses born to parents with heaves are much more likely to develop the disease.

Asthma in humans is also a chronic condition. The symptoms of an asthma attack are the same as those for heaves — wheezing, coughing, chest tightness and shortness of breath. Asthma attacks may be triggered by allergens like household dust. However, for some people with asthma, attacks are brought on by other things like exercise or substances found in the workplace. There is also a genetic component to asthma.

At a fundamental level, heaves and asthma are similar in the responses that take place in the lungs when antigens like dust and mold are present. During an attack of heaves or asthma, the smooth muscle cells lining the air passages to the lungs constrict, which narrows the airways and makes breathing difficult. The resulting inflammation process affects the different types of cells present in the airways and lungs — for example, it increases the number of airway smooth muscle cells and mucus-producing cells that are present. These changes cause damage within the lung tissue, which is eventually repaired. Over time, however, the cycle of tissue damage and repair leads to changes in the airways. This process is called 'remodeling' and results in thickening of the airway

walls and narrowing of the airway passages, which makes symptoms progressively worse.

Researching the Inflammation Process and the Reversibility of Remodeling

Dr. Lavoie is one of the few researchers in the world studying heaves. He wants to understand why some horses in Canada will get sick with heaves when exposed to dust and mold while most horses do not. Dr. Lavoie also wants to know if remodeling can be reversed and if so, what are the best therapies to accomplish this.

He has teamed up with equine veterinary surgeons led by Drs Marcel Marcoux and Jacques Lussier, a molecular biologist, both at the University of Montreal. These collaborations were made possible because of partnered funding from AllerGen NCE and the Canadian Institutes of Health Research (CIHR) Institute of Infection and Immunity. This funding enabled Dr. Lavoie to recruit and retain nine outstanding graduate students, as well as a valuable research assistant.

Given the similarities between the inflammation and remodeling processes that occur in heaves and in asthma, Dr. Lavoie is also collaborating with world renowned asthma experts at McGill University and the Meakins-Christie Laboratories — Dr. James Martin, whose primary research interest is exploration of asthma through the use of animal models; and Dr. Qutayba Hamid, who is particularly interested in how asthma develops at the molecular level. AllerGen NCE funding also enabled Dr. Lavoie to bring on board geneticist Dr. Catherine Laprise (Université du Québec à Chicoutimi) who specializes in the genetics of asthma and allergies.

In the first phase of this innovative research project, horses with a history of heaves and horses with no history of the disease were kept in an allergen-free environment. Blood and tissue samples were taken from all horses using techniques that Dr. Lavoie and his collaborators had developed. In the second phase, the horses were kept in a barn and exposed to dusty and moldy hay for 30 days. Samples were again collected. Predictably, analysis of the samples from the horses with a history of heaves showed that they had developed an inflammatory response within the lungs. The question was why didn't all the horses become ill? The answer, it seems, lies in genetics.

This team has identified more than 160 genes that are different in the lung tissues of heaves-afflicted horses. They also have identified certain genes in horses that are potentially important in human asthma. The long-term hope is that further study of these genes will lead to the development of new and innovative treatments for both heaves and asthma.

As to whether or not remodeling can be reversed in people who suffer from asthma, the jury is still out. What is clear is that knowledge of the inflammatory and remodeling process in our four-legged friends can pave the way to answering this question. Dr. Lavoie and his team are convinced that the horse is a particularly useful animal model for understanding the reversibility of chronic remodeling in people with asthma for a variety of reasons.

The other question being addressed by this research team is whether or not long-term lung damage (*via* airway remodeling) can be reversed. They tried to reverse the airway remodeling in horses with heaves using two different strategies, the results of which they then compared. The common-sense strategy was to take the heaves-afflicted horses out of the dusty and moldy barn. Six months later, analysis of the tissue samples showed that there was not much reversal in the remodeling of the damaged tissue. One year later, these horses showed, on average, a 30% improvement. The other strategy tried by Dr. Lavoie and his team was to administer powerful drugs called corticosteroids (like those given for asthma). The horses that received the corticosteroid treatment

also showed, on average, a 30% improvement when checked six months later. Regardless of treatment strategy applied, Lavoie and his team were unable to reverse airway remodeling to an extent that resulted in improvements in lung function greater than 30%.

To date, this research project has shown that removing the horses from the stable or barn is just as effective as giving corticosteroids (though the latter approach does act more quickly). This is a useful finding, given the prohibitive cost of a course of corticosteroid treatment. The research also seems to suggest that once lung remodeling occurs, it is difficult to significantly reverse. However, Dr. Lavoie is keen to try other therapies to see if a reversal of airway remodeling is achievable.

As to whether or not remodeling can be reversed in people who suffer from asthma, the jury is still out. What is clear is that knowledge of the inflammatory and remodeling process in our

four-legged friends can pave the way to answering this question. Dr. Lavoie and his team are convinced that the horse is a particularly useful animal model for understanding the reversibility of chronic airway remodeling in people with asthma for a variety of reasons:

1. Heaves is similar to human asthma and occurs naturally in horses (laboratory rats and guinea pigs are artificially stimulated to produce respiratory symptoms).
2. Heaves is a life-long disease, like asthma, which allows researchers to study the natural progression of disease.
3. The size of the horse makes it easier for researchers to take samples from the outside perimeter (*i.e.*, small airways) of the lungs. Samples from this area are helpful in understanding asthma because it is believed that this is where most of the remodeling occurs. It is invasive and potentially dangerous to do outer-lung biopsies in humans.
4. Horses live much longer than laboratory animals like rats and guinea pigs, making it easier to measure a treatment's long-term effectiveness.

Benefits of this Research for Horses and Humans

'Heaves' represents a huge financial cost to Canada's multi-million dollar equine industry. There is also an emotional toll on owners who hate to see these intelligent animals in respiratory distress. This research has reaffirmed that removing the affected horse from the barn or stable is key. It has also identified the genes involved in heaves, which will likely lead to the development of more effective therapies and preventive measures for this debilitating disease.

This research is also set to propel asthma research forward. Using the horse as the animal model for future studies could be the missing link in scientists' search to fully understand asthma's inflammation and remodeling processes. Research that helps improve the day-to-day lives of people living with asthma will have a big impact in Canada. The Asthma Society of Canada reports that 12% of Canadian children and 10% of Canadian adults suffer from this disease.

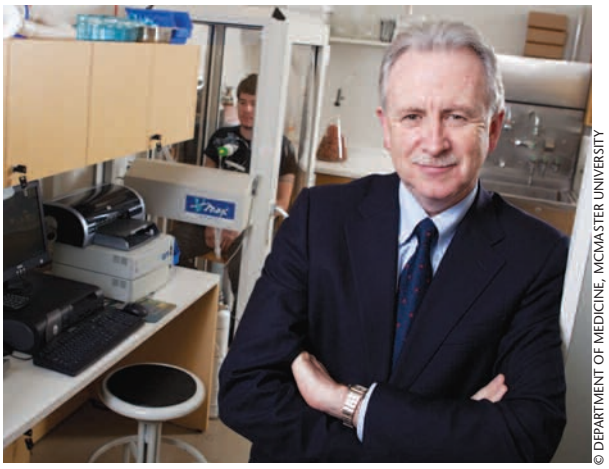
AllerGen NCE has played an important role in the success of this project. AllerGen funding fostered academic collaborations, which Dr. Lavoie says were both productive and personally meaningful. It enabled veterinary students to develop both clinical and research skills. He anticipates that some of the students will go on to become clinician-researchers, which are greatly needed. Dr. Lavoie also points out that AllerGen NCE actively promotes interest in research as a career by inviting graduate students to participate in meetings, "It has been very good for my students ... it makes them feel part of something important." He adds that Canada can never have enough talented researchers. "We need to raise the bar of expertise. We need to go for excellence."

At the end of the day, Dr. Lavoie hopes that this research opens people's minds to the benefits of comparative studies. By helping horses, we may also be helping ourselves.

In 2004, AllerGen NCE Inc., the Allergy, Genes and Environment Network, was established in response to the fact that nearly one in three Canadians now has allergic disease and more research is needed, both in terms of understanding and treating the disease.

International Drug Companies Line Up at AllerGen CIC's Doorstep

Developing new drugs is a very expensive business. Pharmaceutical and biotechnical companies dread the thought of investing millions upon millions of dollars into developing a drug that ultimately turns out to be a "lemon." They have heard that Canada has a group of researchers called the AllerGen 'Clinical Investigator Collaborative' (CIC) that is the best in the world at predicting if a potential new drug for asthma will work. They are now coming to Canada in droves to see for themselves.



Dr. Paul O'Byrne, the principal investigator for the AllerGen CIC and Chair of Medicine at McMaster University

Formation of the CIC

When asked about how the AllerGen CIC came to be, Dr. Paul O'Byrne, principal investigator for the AllerGen CIC and Chair of Medicine at McMaster University modestly replies, "It was a serendipitous series of events, as these things often are." But as the Roman philosopher Seneca famously said, "Luck is what happens when preparation meets opportunity."

When Dr. O'Byrne began training nearly three decades ago at McMaster University, he was part of a research group that used a particular clinical model, or research design, to study how allergic disease works in causing asthma. The group developed a clinical model that is able to predict with amazing accuracy whether or not a new drug will effectively treat allergies and asthma. "It was a very successful collaboration," recalls Dr. O'Byrne.

In 2004, AllerGen NCE was established in response to the fact that nearly one in three Canadians now has allergic disease and more research is needed, both in terms of understanding and treating the disease.

With the creation of a formal research network, Dr. O'Byrne saw the opportunity to take this already successful research collaboration, of which he was a part, to new heights. He pitched the idea that there should be centres across the country that all use the same standard procedures and clinical model to evaluate potential asthma medications. He was sure that pharmaceutical and biotechnology companies would jump at the chance to know early on in the drug development process if a given new compound was worth pursuing. With AllerGen NCE funding, the CIC was launched in 2005.

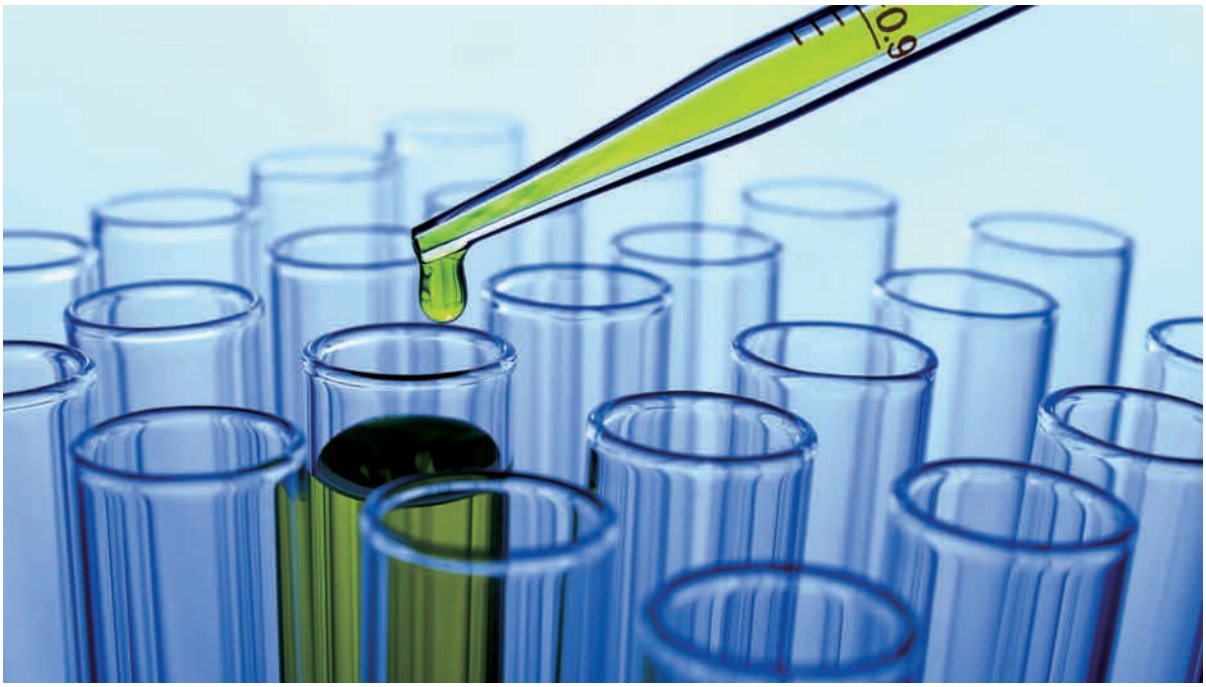
International Drug Companies Compete to Become a CIC Project

Dr. O'Byrne's belief that drug companies would be extremely interested in what the CIC has to offer proved to be correct. "In the five years that we have been doing this, the response from large pharmaceutical and small biotech companies has been phenomenal," says Dr. O'Byrne.

Usually research groups that specialize in doing drug studies compete against each other for contracts with big companies. The AllerGen CIC is in the unique position of having drug companies come to them, asking to be considered as future research partners.

Every year in November, the CIC holds a workshop where drug companies have an opportunity to make their pitch to AllerGen CIC site leaders. The CIC team decides which potential asthma drugs it is most interested in researching. Each year, the CIC receives six to eight proposals from drug companies, but it typically selects no more than three for intensive study. To date, the CIC has completed ten studies. "All of them [were] successfully done, and [the CIC] is working very well," states Dr. O'Byrne.

The CIC has expanded since its inception — it began with four centres in Canada and is now offering services at six academic institutions — McMaster University, Université Laval, University of Saskatchewan, University of Alberta, University of British



"Every study [the CIC does] is useful," says Dr. O'Byrne. "If the drug works, then the company gets enthusiastic about developing it and planning future studies. If it doesn't work, then they have to stand back and ask: is the drug wrong; is the concept wrong; is the way we give it wrong; is the dose wrong?" He adds, "The big advantage to industry is that we have managed to get every study done on time in a very cost effective way."

Columbia and the University of Calgary. Future CIC collaborating centres include international centres in Stockholm and Rotterdam.

The Secret of the CIC's Success

Drug research and development (R&D) begins with discovering new chemical compounds. Compounds occur either naturally (called 'biologics') or are made synthetically and potentially bind to and modify molecules in your body that play a key role in a particular disease. Given that the molecules of interest are often proteins, many drug therapies focus on inhibiting or stimulating protein activity.

Once promising chemical compounds are identified, actual drug development begins and pharmacological studies are carried out. In Phase I trials, the safety of the candidate drug is tested on healthy volunteers. Once the safety screening is complete, testing begins on a population of patients who have the disease. Phase II trials measure the effectiveness of the drug on a small number of people. Phase III trials are basically expensive, large-scale versions of Phase II trials.

The CIC offers one of the best teams in the world when it comes to conducting Phase II trials for new asthma drugs. The key to their success is the quality of the research. The CIC stringently ensures that each centre uses identical research methods with highly developed 'Standard Operating Procedures' and measures for quality assurance. Selecting the right patients to include in Phase II trials is also critical. The CIC is known for its efficient and effective patient screening process. Dr. O'Byrne explains, "We spend a lot of time and energy making sure that the subject is appropriately characterized. Once they are in the

study, we are confident that these individuals are precisely what we need to complete the study and generate the data that we seek. CIC studies need comparatively fewer patients because we excel at selecting the right patients to include from the outset of the study."

Both Corporations and Canadians Win

Because the CIC offers cost effective, timely and highly reliable studies, several small biotechnology companies, many of which are Canadian, have had an opportunity to study their drug's effectiveness, an opportunity that they might not otherwise have had. These companies have benefited from CIC's studies enormously. "Every study [the CIC does] is useful," says Dr. O'Byrne. "If the drug works, then the company gets enthusiastic about developing it and planning future studies. If it doesn't work, then they have to stand back and ask: is the drug wrong; is the concept wrong; is the way we give it wrong; is the dose wrong?" He adds, "The big advantage to industry is that we have managed to get every study done on time in a very cost effective way."

It is challenging to get venture capital funding to develop a new drug. But with a CIC study in hand that shows that the drug is promising, small biotechnology companies — including new Canadian 'start-ups' — are able to go to a large biotechnology or pharmaceutical company and get the funding needed to further their drug's development.

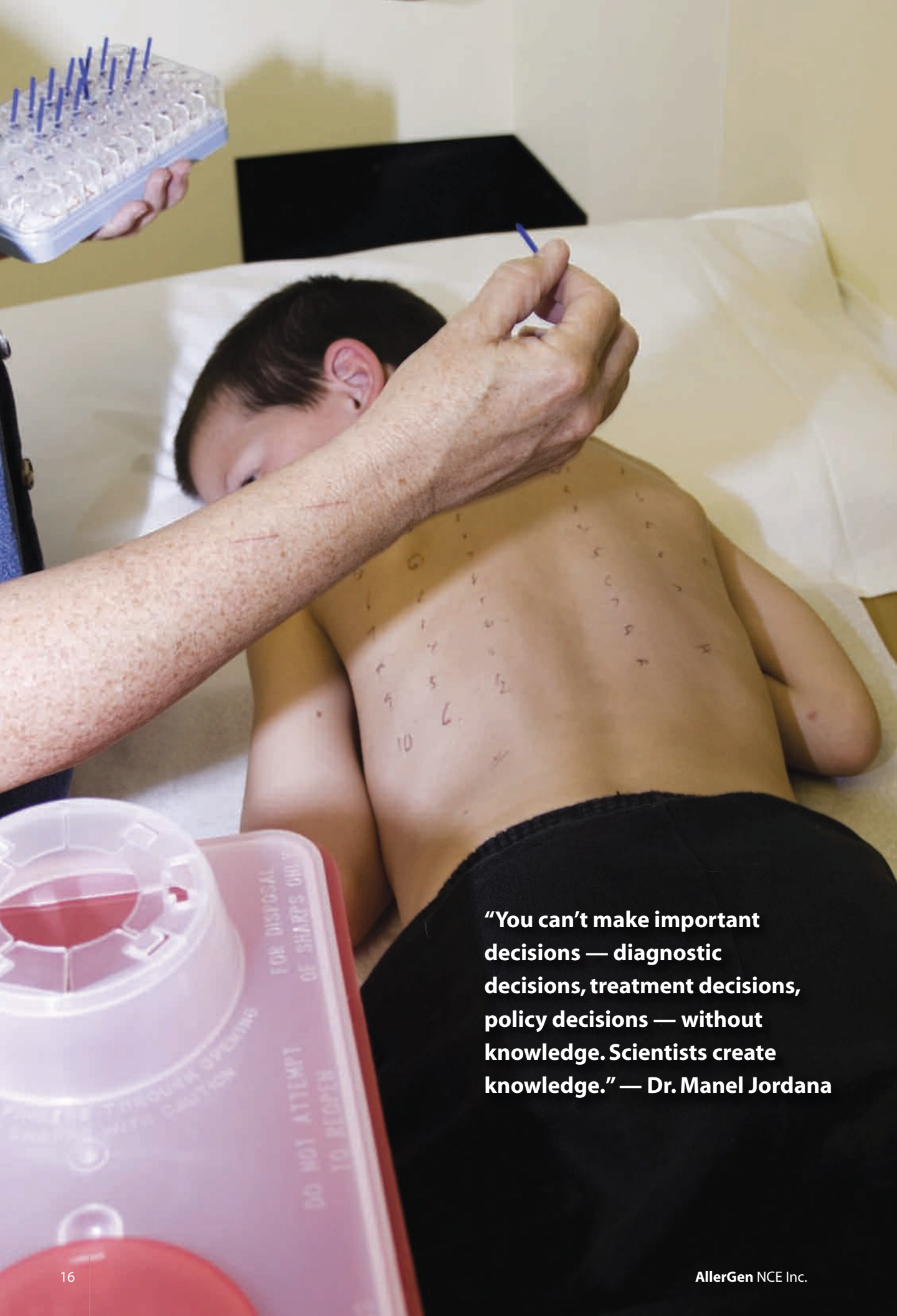
Why, to average Canadians, is what the AllerGen CIC does important? Chances are that you, or people you care about, suffer from asthma. Studies also suggest that the number of asthma patients is steadily rising. This disease costs Canadians close to a billion dollars every year in terms of direct costs, medical care, lost workplace productivity and indirect costs related to a reduced quality of life.

Canada clearly needs asthma research to reduce the symptoms for asthma sufferers and improve their quality of life. Unfortunately, large drug companies do not do a significant amount of R&D here in Canada. They tend to have their R&D operations in the US or in Western Europe. The AllerGen CIC has shifted this trend, demonstrating how excellence in Canadian research contributes to global pharmaceutical innovation. In addition, drug companies agree to share proprietary information about the molecule they developed with all AllerGen NCE-affiliated researchers — a real benefit for Canadian research. These molecules often work in very precise ways, which helps researchers 'tease apart' the mechanisms of the disease. "We as a research community in Canada are benefiting enormously from access to molecules that we normally wouldn't have access to," points out Dr. O'Byrne.

The AllerGen CIC's efficiency in completing Phase II studies for drug companies, along with the fact that Canadian researchers now have access to novel molecules, leads us to be hopeful that new and improved asthma treatments will be available sooner. Dr. O'Byrne says, "This has been a very successful collaboration between academic centres and industry, where both really benefit. We get a lot of information on the pathobiology of the disease by doing these studies. From our point of view, that's why we do them. We want to understand what is happening in these patients so we can try and treat them better. From the companies' point of view, they get access to a very well designed study that functions at a high level, so that they can quickly complete the studies that are needed for decision-making."

While AllerGen NCE over the past five years has invested a little over $2 million in the CIC, the CIC has brought in five times that amount — $10 million plus — through its contracts with drug companies. AllerGen NCE's investment is clearly worthwhile.

Since the CIC came into existence five years ago, it has been a resounding success. Every year, twice as many companies vie to have Phase II studies done by the CIC than is possible for the CIC to take on. This is because the AllerGen CIC has a proven track record of doing asthma drug studies of the highest quality and in a timely fashion, which is no small feat. The CIC also ensures that Canada isn't a 'backwater' in the international research community. Our scientists now have access to cutting-edge molecules developed by private companies. AllerGen NCE is proud of the CIC's success, much like a parent that has raised a superstar. In turn, the CIC gives AllerGen NCE well-deserved credit. "The CIC wouldn't have happened if AllerGen weren't funded and didn't exist," says Dr. O'Byrne. "We would have continued with our informal collaborations on an *ad hoc* basis, but AllerGen NCE has raised us to a completely new level."



"You can't make important decisions — diagnostic decisions, treatment decisions, policy decisions — without knowledge. Scientists create knowledge." — Dr. Manel Jordana

Canadian Physicians and Basic Research Scientists Successfully Partner on Innovative Diagnostic Test to Predict Peanut Allergy

What do peanuts, research and new methodologies all have in common? They are part of a Canadian success story with national and international implications. Canadian physicians and basic research scientists, supported by AllerGen NCE in partnership with Canadian research agencies and universities, have successfully identified a number of unique traits of truly peanut allergic patients, and are well on their way to developing a simple diagnostic test for predicting peanut allergy. This significant discovery will benefit patients' health outcomes, improve their quality of life as well as that of their families, and may help inform patient referral to specialists for oral peanut challenges.

Dr. Susan Waserman, a practicing Clinical Immunologist/ Allergist and Professor at McMaster University

Allergy to peanuts is one of the most serious allergies, and is estimated to account for as many as 80% of allergic reactions that are fatal or near fatal. In addition to this alarming statistic, there is evidence to suggest that the prevalence of peanut allergy has doubled over the last decade.

Physicians have been stumped by a group of patients (typically children), who test positively for peanut on either skin or blood tests, but who have never knowingly eaten them because their parents promote a peanut-free diet. Children avoid peanut because a member of the family is allergic, or because parents mistakenly believe that avoiding peanut prevents allergy. A positive skin test (a prick and a drop of peanut extract placed on the skin resulting in swelling and redness at the site), or a blood test which shows anti-peanut Immunoglobulin E (IgE) antibody, does not necessarily mean that the patient will have a harmful reaction after actually eating peanuts; some people have positive tests but eat peanuts without a problem. Generally, a firm diagnosis of peanut allergy can only be made when there is a history of allergic symptoms after ingestion. Those who are peanut-sensitized, but have no previous peanut exposure must, therefore, be fed peanut under supervised conditions to assess their response. This can potentially lead to serious symptoms of an allergic reaction ranging from upset stomach and skin rash, to difficulty breathing, low blood pressure, loss of consciousness and death. It is, therefore, especially desirable to have a safer diagnostic test to help predict who is truly peanut allergic.

A "Bedside to Bench" Collaboration is Born

Dr. Susan Waserman, a practicing Clinical Immunologist/ Allergist and Professor at McMaster University, was frustrated by being limited to dispensing the broad (and potentially unnecessary) advice of "continue avoiding peanuts" to parents of children whose peanut allergy status is ambiguous. As a result, she embarked on a mission to create a more sophisticated diagnostic test.

With support from AllerGen, and by leveraging its network of contacts, Dr. Waserman and Dr. Manel Jordana, an Immunologist and Professor of Pathology and Molecular Medicine at McMaster University, assembled a multi-disciplinary research team comprising clinical and basic researchers, statisticians, research nurses and graduate students from McMaster University, McGill University, the University of Manitoba and the Manitoba Institute for Child Health. The recruitment of patients for the

study and blood work was handled by Dr. Waserman, while the laboratory analysis was overseen by Dr. Jordana.

In order to create a diagnostic test, the team had to identify features that were unique to patients who had a true peanut allergy. Demographic profiles and blood work results were compiled for three groups of patients whose peanut allergy status was already known: 1) truly allergic children (positive skin test and definite history of reacting to peanuts); 2) "false-positive" children (positive skin test, but do not react to eating peanuts); and 3) non-allergic children (negative skin test and no history of reaction) who were the "control" group. Data gathered on the patients included their age, medications, and other allergic co-morbidities. Since a peanut allergy is essentially an abnormal response in which the body's immune system reacts as if the peanut protein is a threat, blood samples were examined for peanut specific IgE, as well as total IgE. They collected data on lymphocytes, a type of white blood cell that plays a vital role in the body's defence system. Finally, they examined other markers for immune system response, namely chemokines, which are proteins produced by cells in response to inflammation that attract other cells to the inflamed site.

The demographic and blood work data were provided to the team's statisticians who conducted a highly sophisticated mathematical analysis. According to Drs Waserman and Jordana, "The analysis was actually able to isolate four variables that gave us a high degree of predictability as to who was truly allergic and who was not." For instance, it turns out that total IgE is important, as is the age of the child. The "algorithm" (or set of conditions) for predicting peanut allergy has been patented, and could lead to a minimally invasive, technically simple, safe and accurate test kit in the near future. This will not totally eliminate the need to perform oral peanut challenges, but it will allow physicians to feed those who are likely not allergic, a safer approach than having to feed everyone. It is understood that these decisions will also be made in conjunction with the person's allergist.

Improving Quality of Life — No Longer A Tall Order

Given the increasing prevalence of peanut allergy, and its potential severity, a test kit that could quickly inform physicians whether or not the patient is truly peanut allergic would fill not just a Canadian need, but an international one.

A family physician would be able to ensure that "at risk" patients are appropriately prescribed an epinephrine auto-injector, a device that administers epinephrine (also known as adrenaline), the treatment of choice for anaphylaxis, and referred to an allergy specialist. Knowing one's true peanut allergy status creates an incentive to take peanut avoidance seriously, and ensures that patients are prepared to deal with an emergency — both of which are life-saving.

"Having a peanut-free diet is 'no picnic.' Children and their families become "slaves" to reading food labels and scanning every environment they enter into for the presence of peanuts. Even a simple activity like having a slice of a friend's birthday cake is not without risk...."
— Dr. Waserman

Those children and adolescents who turn out not to be allergic to peanuts, experience a dramatic improvement in their quality of life, because it is no longer necessary for them to avoid peanut. Having a peanut-free diet is, in the words of Dr. Waserman, "no picnic." Children and their families need to be meticulous about reading food labels and must always scan their environments for the presence of peanuts. Even a simple activity like having a slice of a friend's birthday cake is not without potential risk. While peanuts may not be part of the recipe, there is still risk of contamination since peanuts and peanut-based products are present in most household and commercial kitchens. Given the natural affinity between socializing and eating, not only does avoiding peanuts restrict a person's diet, it can restrict opportunities for enjoying social activities. A recent study found that the quality of life of children with peanut allergy was significantly poorer than that of children

with insulin dependent diabetes. A test kit that could rule out true peanut allergy would spare children and adolescents, their families, and other caregivers (like teachers and athletic coaches) much unnecessary stress.

From an economic standpoint, AllerGen's investment in this project will result in handsome dividends for Canadians. Besides the obvious international commercial potential of a diagnostic test kit for predicting peanut allergy, it will help determine which patients need to be urgently referred to the allergist, and may also help determine which individuals still need an oral peanut challenge. A unique component of Drs. Waserman's and Jordana's research, undertaken as part of an AllerGen-supported research team called the Canadian Group for Food Allergy Research (CanGoFAR), is that it intimately combines clinical and basic research. Dr. Jordana's laboratory

has established experimental models of peanut-induced allergy and anaphylaxis in mice that mimic many hallmarks of the human disease. This, combined with the research of other members of AllerGen's CanGoFAR team allows a careful dissection of many fundamental aspects (cells, molecules and tissues) of the disease process. An exciting outcome of this ground-breaking research is the discovery that certain drugs, when combined, are effective in significantly reducing the severity of an anaphylactic reaction, and in fact fully prevented the development of convulsions and death. This discovery (patent pending) was possible because the CanGoFAR team studied in-depth the roles various molecules play in peanut anaphylaxis. Drs Waserman and Jordana are currently exploring ways to translate this discovery into the clinic. When asked why basic research is important, Dr. Jordana replied, "You can't make important decisions — diagnostic decisions, treatment decisions, policy decisions — without knowledge. Scientists, both clinical and basic, create knowledge."

Canada Could Lead the Way in Food Allergy Diagnosis and Management

Until recently there was little research on food allergy being conducted in Canada. Now, within Canada there is a flurry of research activity with respect to the growing problem of food allergies.

This AllerGen NCE-funded project afforded the opportunity for Drs Waserman and Jordana, who are part of the AllerGen-supported CanGoFAR research team, which conducts basic and applied food allergy research, to not only build partnerships that can be drawn upon for future research opportunities, but also to hone Canadian expertise in diagnosing and managing food allergies. The clinical team developed expertise in performing oral peanut challenges (a risky procedure that involves feeding peanuts to potentially allergic people to see how they react), which is the "gold standard" for determining peanut allergy. With any diagnostic test, even the "gold standard" results may not be straightforward. Also, there currently are not enough allergy specialists or the infrastructure in Canada to routinely perform oral challenges in all centres. Interestingly, in performing these challenges, Dr. Waserman observed that the earliest signs of a peanut allergic reaction in children were abdominal pain and a change in personality; not hives or skin rash, which is what is taught.

The research team also includes seven students. Working on this project will undoubtedly help these professionals-in-training become first rate scientists and highly skilled clinicians, as the project drew on multidisciplinary competencies — research methodology, epidemiology, immunology, and adult and pediatric allergy. As for the statisticians, they have developed expertise to apply a sophisticated mathematical program to a complex clinical problem, a skill that can be called upon for future research.

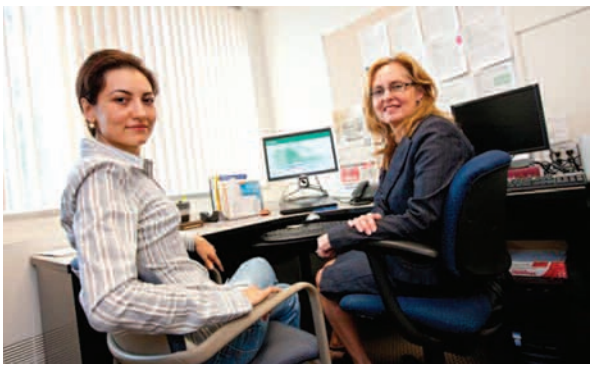
"The deployment of this comprehensive, multidisciplinary research team in food allergy is the most amazing thing that has happened in the area of food allergy in a long time," Dr. Waserman states with palpable excitement. "Without funding from AllerGen NCE, these projects would not have gone ahead." Drs Waserman and Jordana's research team is progressing with procurement of additional funding from Canadian and US-based organizations. On the one hand, the team is busy preparing the launch of the first immunotherapy trial for peanut allergy in Canada; on the other, they are preparing for publication of exciting new discoveries concerning key immunological pathways underlying peanut allergy which they hope, may be rapidly translated into clinical practice. "Our only limitation to becoming a world class centre in food allergy research is world class funding on a sustained basis," say Drs Waserman and Jordana.

Dr. Kozyrskyj and her team set out to determine the role of maternal depression and anxiety in the development of childhood asthma using the existing SAGE database. They also wanted to know whether or not the association is dependent upon the length of time that the mother is in emotional and mental distress.

Children with Stressed-Out Mothers at Risk for Developing Asthma

Mothers today juggle family and employment responsibilities like high-wire performers in Cirque du Soleil. Difficulty coping can lead to clinical depression or anxiety, either short-term or long-term.

Research has shown that the stress level of mothers in industrialized countries like Canada has increased over the years. There has also been a parallel increase in the number of children who are diagnosed with asthma. Dr. Anita Kozyrskyj, an epidemiologist and health services researcher at the University of Alberta, wondered if the two trends were linked. With funding from AllerGen NCE, she set out to investigate if maternal depression or anxiety is an independent risk factor for childhood asthma. What she found should have health care providers — from government policy makers to family physicians — sitting up and taking note.



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Salma Bahreinian, MSc student, University of Alberta and Dr. Anita Kozyrskyj, Associate Professor and Research Chair, University of Alberta

Looking at the 'Big Picture' of Asthma, Genes and the Environment

Dr. Kozyrskyj has always been interested in the impact that a person's social environment and lifestyle has upon his or her health. She is especially interested in the person's early childhood because studies show that social environment and lifestyle in the formative years have an enormous impact upon future health.

Early on in her career, Dr. Kozyrskyj, along with her colleagues Drs Allan Becker and Kent HayGlass at the University of Manitoba, undertook the novel *Study of Asthma, Genes and the Environment* — referred to as "SAGE." This project, funded by the CIHR, is a long-term study of 14,000 children born in Manitoba in 1995 (a 'birth cohort') and is primarily based on the anonymized health care records of the children and their mothers held in provincial databases. Within the study, the health of 723 children has been looked at in more detail. This group includes both children who have been diagnosed with asthma and children with no history of the disease. The detailed investigation includes confirmation of the asthma diagnosis by a pediatric allergist and sampling of the the child's home for dust and pet allergens.

The initial research question that Dr. Kozyrskyj focused on within the SAGE Study was the role of antibiotics taken during infancy in the development of asthma. While looking at this research question, she came across several publications about parental stress and childhood asthma. Dr. Kozyrskyj realised that the SAGE database could also be used to investigate the connection between a mother's mental health, in terms of depression and anxiety, and her child's respiratory health, in terms of asthma.

Maternal Stress in Early Childhood and the Development of Asthma

AllerGen NCE provided Dr. Kozyrskyj and her team, which includes Drs Becker, HayGlass and Brian MacNeil (University of Manitoba), as well as Dr. Patrick MacNeil (Dalhousie University), with funding recognizing that more studies are needed on the roles that social environment and lifestyle play in asthma. With AllerGen NCE funding in hand, the team was able to obtain matching funds from the Manitoba government. "AllerGen funding let me do the research," says a grateful Dr. Kozyrskyj.

Dr. Kozyrskyj and her team set out to determine the role of maternal depression and anxiety in the development of childhood asthma, using the existing SAGE database. They also wanted to know whether or not the association is dependent upon the length of time that the mother is in emotional and mental distress. The SAGE database is currently the only one in

the world that is population-based (and, therefore, large enough to identify trends) and which reflects 'real life' in that it does not exclude children traditionally considered to be at low risk for developing asthma.

From the healthcare records of the 14,000 Manitoban children, the researchers identified which children had been diagnosed with asthma by the age of seven, meaning that they had either seen a doctor at least twice for asthma, had been hospitalized at least once, or were prescribed asthma medications on at least two occasions. The results were validated against a physician diagnosis for asthma, made when the smaller group of 723 children were seen by a pediatric allergist. Some had allergic asthma, some had non-allergic asthma and some were completely healthy.

The SAGE database was also searched for codes that indicated that mothers had seen a physician on at least one occasion for depression or anxiety or had been prescribed a medication for these conditions. The length of time that the mother was in emotional and mental distress was also considered. Was it only during the first year of her child's life? Did it continue after her child's first year, but resolve by the time her child was five years of age? Or was the depression or anxiety chronic, meaning that it persisted beyond her child's seventh birthday? In addition to searching the SAGE database, all mothers of the 723 children completed questionnaires about their current emotional and mental states.

If a mother's depression or anxiety resolves within the first four years of her child's life, her child's odds of developing asthma are around the same as those for children with mothers who are doing well. If a mother continues to struggle with depression and anxiety past the child's fourth birthday, then the odds of her child developing asthma go up, 1.3 times, to be exact. "This is a statistically significant finding," says Dr. Kozyrskyj.

Having ascertained the odds of a child developing asthma, and the odds of a mother being treated for depression or anxiety (either short-term or long-term), the researchers could not simply 'mash' the two odds together and then make a pronouncement about the likelihood of developing asthma if your mother suffers from anxiety or depression. They had to take into account a number of factors that complicated the picture.

For example, Dr. Kozyrskyj and her team had to take into account the children's gender, because it is well known that boys are more likely to develop asthma than girls. As another example, they factored in the number of siblings the child had. The more siblings you have, the less likely you are to develop asthma, a phenomenon that has been attributed to getting infections from brothers and sisters, which challenges and strengthens the immune system.

In the second part of this study, Dr. Kozyrskyj and her team took blood samples from the smaller group of 723 children. They wanted to see if a mother's distress triggered a stress

response in her child that resulted in an abnormal serum cortisol level.

Why Mother's Happiness Matters

Dr. Kozyrskyj and her team found that an astounding 18.9% of the 14,000 mothers they studied had experienced either depression or anxiety after their child was born. But what is critical in terms of the development of childhood asthma is how long that depression or anxiety lasts.

If a mother's depression or anxiety resolves within the first four years of her child's life, her child's odds of developing asthma are around the same as those for children with mothers who are doing well. If a mother continues to struggle with depression and anxiety past the child's fourth birthday, then the odds of her child developing asthma go up, 1.3 times, to be exact. "This is a statistically significant finding," says Dr. Kozyrskyj.

Unsurprisingly, the research shows that lower-income women experience higher levels of post-partum depression. Other studies have found that asthma is more prevalent in inner city children, who typically live in highly stressed families.

What did surprise the researchers is finding that women who have a history of asthma themselves are more likely to have post-partum depression. "This was a totally unexpected finding," says Dr. Kozyrskyj. The reason for the association is unclear.

Dr. Kozyrskyj and her team also found that a mother's repeated bouts of depression or anxiety do, in fact, trigger a stress response in her child that results in an abnormal serum cortisol level. What is particularly interesting is that a mother's distress increases the cortisol level in a child who doesn't have asthma, but reduces the cortisol level in a child who does.

Why there is a link between a mother's state of mind and junior's asthma is the million-dollar question. Researchers don't have an exact answer yet. Some of the possible explanations include: First, depressed mothers are less likely to breastfeed and more likely to smoke, both of which increase the odds of childhood asthma and secondly, depressed mothers interact less with their children, which stresses these children and affects their immune systems in some way.

By no means is Dr. Kozyrskyj blaming depressed or anxious mothers for their kids' asthma. She underscores the fact that asthma is a complex disease and that there are many factors behind why a child develops it. She adds that while the research shows that stress "does have an important role to play, we aren't there yet to say definitely what that role is." Dr. Kozyrskyj does encourage mothers who are struggling to seek support from their family and friends. "Social support is really important," she says.

Canadian Research Sparks International Attention

Not surprisingly, given that this is the first study of its kind in the world, the media has been buzzing about this study's demonstration of a link between maternal depression and childhood asthma.

This study was accepted for publication by a highly prestigious journal, *The American Journal of Respiratory and Critical Care Medicine*, as well as discussed by the editor (a field expert) on the editorial page. As a result of this study, there are now similar studies underway in other countries, for example, the U.K. "Scientists in other countries are now asking the same questions," states Dr. Kozyrskyj.

Interest in this study extends beyond parents, respiratory health experts and scientists, to government policy makers. Manitoba, which already has an impressive province-wide screening program for post-partum depression, has approached Dr. Kozyrskyj to help it ensure that its public health nurses ask new mothers all the right questions. "Because our research shows a link between asthma and maternal depression, the Manitoba government is even keener to ensure that all mothers are screened for depression and anxiety," says Dr. Kozyrskyj.

An important advantage of AllerGen NCE funding is that it encourages a multi-disciplinary approach. "As a result of collaborating with people from different areas of expertise, I was able to ask more interesting questions. For example, I would have never ventured into the territory of looking at cortisol levels," states Dr. Kozyrskyj.

AllerGen NCE-supported multi-disciplinary teams, such as its Mind-Body cross-programmatic initiative, are continuing to work on finding answers about the role of the brain and behaviour in the development of allergies and asthma. Following Dr. Kozyrskyj's lead, these teams are poised to conduct more research looking at the role that social environment, behaviour and lifestyle play in the development of these conditions.

Success Stories

Networks of Centres of Excellence

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