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Indoor Environmental Quality within an Elementary School Classroom: Measurements of *Felis domesticus* I, *Dermatophagoides pteronyssinus* I, *Dermatophagoides farinae* I, and *Blatella germanica* in Carpeting

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Indoor Environmental Quality within an Elementary School Classroom: Measurements of *Felis domesticus* I, *Dermatophagoides pteronyssinus* I, *Dermatophagoides farinae* I, and *Blatella germanica* in Carpeting

by

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Dedication

This thesis is dedicated to my husband, Chris, my parents, Connie and Wayne, and my aunt, Jewel, with love. Your support every step of the way will always be appreciated and your belief in the importance of education has truly helped to push me to complete what I started.

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ABSTRACT

The United States Environmental Protection Agency acknowledges that indoor environments can impact the health of students and can affect concentration, attendance, and student performance (USEPA 2008). In a school year, children are required by the Florida Department of Education to be in school for a total of 180 days, which is 795 hours spent in a classroom (FLDOE 2008). In the elementary school setting, kindergarten classes typically spend a portion of the school day with floor-based activities. The American Lung Association (ALA) states that over 6.8 million children under the age of 18 are affected by asthma (ALA 2008). Allergic reactions and/or sensitization to allergens such as dust, animal dander, and cockroaches are among triggers associated with asthma cases. Literature suggests looking at the areas where individuals spend a considerable amount of time to identify potential exposure sources. Currently, many of these studies have been done regarding the home indoor environment; however, few specifically document the concentrations in carpeting in elementary schools and the exposure time associated with floor-based activities.

The objective of this research was to quantify the concentrations of cat (*Felis domesticus* I), dust mite (*Dermatophagoides pteronyssinus* I, *Dermatophagoides farinae*

I), and cockroach (*Blatella germanica)* allergens in carpeting in an elementary school kindergarten class and to document and quantify student group activities that are floor-based.

One Florida elementary school classroom was identified as the study site. A total of eight reservoir dust samples were collected during the school year to be analyzed for *Felis domesticus* I, *Dermatophagoides pteronyssinus* I, *Dermatophagoides farinae* I, and *Blatella germanica* allergens. The sampling reservoir was the carpeting used for group floor-based activities by the school children. Dust samples from the carpet were analyzed by The Johns Hopkins University Reference Laboratory for Dermatology, Allergy, and Clinical Immunology (DACI). The sample collection methodology followed the "Dust Analysis Instructions for Use" provided by the DACI laboratory, along with the "nozzle sock" sampling media. Following discussions with the kindergarten teacher regarding curriculum and scheduled classroom activities, group floor activities were identified. The kindergarten class was observed periodically throughout a school year to document and quantify classroom activities that were floor-based. The information documented includes: occupancy of classroom, occupied floor area, occupant density, and time spent on carpeting. *Felis domesticus* I (*Fel d* I) measurements ranged from 2,206 – 10,558 ng of allergen/g of settled dust, *Dermatophagoides pteronyssinus* I (*Der p* I) measurements ranged from 3,408 – 86,704 ng/g and *Dermatophagoides farinae* I (*Der f* I) measurements ranged from 704 – 6,720 ng/g, and *Blatella germanica* (*Bla g* I) measurements were below detection limits.

Based upon the DACI criteria, dust mite concentrations were moderate to high and cat concentrations were low to moderate. Kindergarten children spent approximately 38% of classroom time in floor-based activities.

INTRODUCTION

The management of immunoglobulin E (IgE)-mediated human allergic diseases, such as extrinsic asthma and rhinoconjunctivitis, involves a combination of four potential approaches: allergen avoidance, symptom-directed pharmacotherapy, allergen specific-directed immunotherapy, or hyposensitization and omalizumab (omalizumab is a monoclonal antibody used to reduce allergic hypersensitivity) (Hamilton, 2005). To facilitate avoidance of allergen exposure and document remediation actions in the allergic individual's personal and work or school environments, it is useful to document the actual level and location of the relevant allergens that can trigger symptoms. This is particularly important for areas where allergic individuals spend considerable time (Hamilton, 2005). Kindergarten children, ages 5 to 6 years old, spend 795 hours in a classroom. Allergy symptoms gradually intensify as indoor allergens accumulate and/or the patient's allergic sensitivity increases (Hamilton, 1992). Avoidance studies strongly suggest that continuing exposure plays a role in most cases of chronic asthma (Platts-Mills et. al., 1995). The suggested dust reservoir sampling method for allergens is vacuum sampling (HUD, 2004) and to date this is the best index of exposure to allergens. This study quantified the concentrations of cat (*Felis domesticus* I) (*Fel d* I), dust mite (*Dermatophagoides pteronyssinus* I, *Dermatophagoides farinae* I) (*Der p* I, *Der f* I), and cockroach (*Blatella germanica)* (*Bla g* I) allergens in carpeting and document and quantify classroom activities which are floor-based.

The elementary school and kindergarten class was chosen based on age of the school building, age of carpeting, and the willingness of the school administration and the teacher to participate. Prior to the start of the study, discussions with the kindergarten teacher identified curriculum and scheduled classroom activities that involved group floor time. This information provided the basis for estimating the exposure time of kindergarten children to allergens in the carpeting. The dust sampling area was determined by the location of the group floor based activities.

Strong documentation exist that asthma and allergies can be triggered and exacerbated by exposure to many environmental factors and sensitization to indoor allergens is a risk factor for the development of asthma in children and adults (ATSDR, 2007). Few exposure data exist for school children who are at greater risk based upon their scheduled classroom curriculum. This research provides needed information on student exposure time and concentration levels of allergens within carpeting, and information that furthers the knowledge of these topics.

Purpose

The purpose of this research was to quantify the concentrations of *Fel d* I, *Der p* I, *Der f* I, and *Bla g* I allergens in carpeting, and to quantify floor based classroom activity time.

The specific objectives of this research were:

1. To quantify and document the classroom activities which are floor-based;
   a. The location of group floor-based activities was determined.
   b. The group floor area was determined.
   c. The number of students during each classroom observation was determined.
   d. The duration of time students spent in the group floor-based activities was determined.
   e. The activities students performed while in groups on the floor were documented.

2. To quantify the allergen exposures to the kindergarten students using the DACI lab method;
   a. The concentrations of *Fel d* I.
   b. The concentrations of *Der p* I.
   c. The concentrations of *Der f* I.
   d. The concentrations of *Bla g* I.

LITERATURE REVIEW

Background

Dust is a heterogeneous mixture comprising a variety of inorganic and organic particles, metals, and fibers of different sizes (Elliott, Arbes Jr. et al. 2007). Common organic particles that may be found within dust include dust mites, cat allergen, and cockroach allergens. Dust mites *Dermatophagoides pteronyssinus* (*Der p* I) and *Dermatophagoides farinae* (*Der f* I) belong to phylum Arthropoda and subphylum Chilcerata (Macher, Ammann et al. 1999) *D. farinae and D. pteronyssinus* are found most frequently and are most widely distributed geographically (Macher, Ammann et al. 1999). A female *D. farinae* measures 425 µm in length and weighs between 10 and 16 µg, while the female *D. pteronyssinus* are only 300 to 350 µm long (Macher, Ammann et al. 1999). Indoor dust mites are commonly found in areas, such as, carpeting, stuffed animals, and upholstered furniture. Mites reportedly produce up to 200 times their own weight in allergen-rich fecal pellets of a mean average diameter of 20 microns during their 2 to 3.5 month life span (Hamilton 2005). Literature review suggests that there is a correlation between dust mite concentration, temperature, and relative humidity. Mite densities exhibit a seasonal cycle that parallels relative humidity changes, with the highest mite concentrations occurring during periods of high relative humidity (Macher, Ammann et al. 1999). Experiments have shown that mites feed sparingly at relative humidity (RH) levels less than 70%, producing little fecal material and associated

allergen (Macher, Ammann et al. 1999). Mites consume 75% less food (with a corresponding reduction in fecal pellet production) at 75% RH than at 85% (Macher, Ammann et al. 1999). All mites gradually dehydrate and die when held for more than 11 days at humidities below 50% (Macher, Ammann et al. 1999).

Felis domesticus, the domestic cat, has become the most common indoor pet in the United States as of 1989 (Hamilton, Chapman et al. 1992). All warm-blooded animals produce potential allergens in their dander, urine, feces, and saliva (German and Harper 2002). The sublingual mucous salivary glands and hair root sebaceous glands of the cat produce a potent 38-kD allergen (Hamilton 2005). It adheres tenaciously to fibers in carpets and dust particles from 2 to 10 microns in size that can be inhaled. The principal cat allergen, *Fel d* I is heat stable and thus steam cleaning of carpets has no added benefit over regular vacuuming in removing cat allergen from the home (Hamilton, Chapman et al. 1992).

Of the 50 varieties of cockroaches in the US, only 8 are considered important indoors species. *Blatella germanica (Bla g* I*)* is considered the most cosmopolitan cockroach (Hamilton, Chapman et al. 1992). Cockroaches can become abundant in any environment where sanitary practices are inadequate (Macher, Ammann et al. 1999). *Bla g* I is used as an indicator of the level of cockroach infestation (Hamilton, Chapman et al. 1992). The particles that carry cockroach allergen are relatively large (at least 10 µm in size) and remain airborne for a short period of time after disturbance (German and Harper 2002).

Exposure

Antigen exposures have increased due to people spending more time indoors and recent changes in homes and offices (e.g., higher mean indoor temperatures, reduced ventilation, laundering with cool wash detergents that may not remove allergens effectively, and widespread use of carpeting) (Macher, Ammann et al. 1999). The number of adult and children residents, type of dwelling, age of home, duration of habitation, prior occupancy, and urban or rural location are some factors that may influence aeroallergen burden in the home. In general, a greater number of inhabitants will deposit higher levels of skin and food particles throughout the home (Hamilton, Chapman et al. 1992). Literature suggests the same application may be applied to an elementary school setting. Cat allergens adhere to dust particles that range in size from 2 to 20 microns (Hamilton, Chapman et al. 1992). The commonality of the domesticated cat in households throughout the United States and the size of dust particles that the cat allergen adheres to allows for cross contamination between homes and schools. In the Abramson, et al. (2006) report cat allergen was identified despite the lack of domestic cats in the school setting. However, it is unclear to the amount allergens which are brought from home by the students to the school (Tranter 2005). The common source of cockroach antigens is in settled dust while their common breeding grounds include kitchens, basements, and bathrooms. The antigenic product of cockroaches is fecal particles, saliva, and dried body fragments. Proteins derived from cockroaches are associated with particles greater than 5 µm diameter and become airborne only when a room is disturbed (Macher, Ammann et al. 1999). To facilitate avoidance of allergen exposure and document remediation actions in the allergic individual's personal and work or school

environments, it is useful to document the actual level and location of relevant allergens that can trigger symptoms (Hamilton 2005). Several studies have documented a dose-response relationship between cumulative exposure to dust mite allergen and subsequent sensitization of exposed persons (Macher, Ammann et al. 1999). The case for a causal relationship would best be supported by evidence for a dose-response relationship between allergen exposure and symptoms (Platts-Mills, Sporik et al. 1995). Exposure to dust mite, cat, and cockroach allergens comes after carpeting becomes disturbed. However, the allergen concentration of dust mite, cockroach, and cat in dust does not reflect the direct measurement of the allergen entering the lungs. At this time, there is no measurement to directly assess the total allergen content entering the lungs based upon range of allergen particle size. Although measurements of reservoir dust may be rather distant from an actual measurement of allergen entering the lungs, nonetheless, for mite and cockroach allergens are the best index of exposure that is available (Platts-Mills, Sporik et al. 1995). For cat allergen it is clear that airborne allergen is carried on a wide range of particle sizes and that the relationship between airborne levels and floor samples is extremely variable (Platts-Mills, Sporik et al. 1995). Avoidance studies strongly suggest that continuing exposure plays a role in most cases of chronic asthma (Platts-Mills, Sporik et al. 1995). This is strong evidence that continuing exposure plays an important role in symptoms (Platts-Mills, Sporik et al. 1995). However, an equally important concept is that the dose response for symptoms may not be comparable to the dose response for sensitization (Platts-Mills, Sporik et al. 1995).

Health Effects

The immune response to inhaled cat (*Felis domesticus*), dust mite (*Der p* I, *Der f* I*)*, and cockroach (*Bla g* I*)* allergens yields many different symptoms which include allergic rhinitis, extrinsic asthma, allergic sinusitis, atopic dermatitis, bronchial hyperactivity (BHR), and hypersensitivity pneumonitis. The body's response depends on the source material, host factors (e.g. genetic factors and prior exposure), and the duration and intensity of exposure (Macher, Ammann et al. 1999). There are two phases which are considered with regard to inhaled allergens. The first is sensitization, in which, time is required for the body to develop an immunological sensitization (Macher, Ammann et al. 1999). The second is the production of chronic bronchial inflammation (Platts-Mills, Sporik et al. 1995; Gold 2000). Experiments suggest that continued allergen exposure is necessary to maintain inflammation which is the cause of BHR and is an important long-term cause of inflammation. For the first phase (i.e. sensitization) there is clear evidence for a dose-response relationship such that the higher the levels of allergen in the homes of a community, the larger the percent of children who will become sensitized. In contrast, the evidence for a dose-response relationship among sensitized individuals is indirect and unlikely to be demonstrated by cross-sectional studies of a population (Platts-Mills, Sporik et al. 1995). Sensitization and high exposure to cockroach allergen has been strongly associated with the risk of asthma in some studies (Tovey and Marks 1999). The measured concentrations can be compared with 'risk levels', which are approximate indicator levels based on concentrations in homes that consistently correlated with asthma exacerbation or sensitization (Table 1) (Tranter 2005).

Table 1: Risk levels for asthma were developed from home studies and have been applied to school research

Allergen	Risk levels [1, 4, 6, 22]	Alternatives levels of interest cited
Dust mite (*Der p* I, *Der f* I)	Sensitization (for atopic) >2,000 ng/g settled dust Symptoms >10,000 ng/g settled dust	'Increased level' >25 ng/m2 area sampled
Cat (*Fel d* I)	Sensitization >8,000 ng/g settled dust	'Moderate level': 1,000–8,000 ng/g settled dust, >8,000 ng/m2 area sampled
Dog (*Can f* I)	Sensitization >10,000 ng/g settled dust	'Moderate level': 1,000–10,000 ng/g settled dust
Cockroach (*Bla g* I *or* II)	Sensitization >2U/g (activity units of antigen per gram settled dust)	'Symptom threshold'>2, >8 U/g 'Sensitization threshold'>1, >5, >10 U/g 'Level of concern'>1U/g 'Low threshold'>5,000 ng/g settled dust

In a study where carpet allergen measurements were recorded between two elementary schools the dust mite concentrations ranged from non-detectable to 26 ng/g, cat allergens concentrations ranged from 710 ng/g to 1,710 ng/g, and cockroach 0.82 U/g to 9.00 U/g (Ramachandran, Adgate et al. 2005). Limited literature exists on concentrations of allergens in carpets among elementary schools in the United States. Table 2 shows the review of ten to eighteen studies in the United States on indoor allergens in settled school dust reported by D.C. Tranter.

Table 2: Allergen Concentrations in settled dust carpeting

Allergen	Location	School Concentration (ng/g)
Dust mite (*Der p* I, *Der f* I) 17 studies	Florida, USA, May-June	42-14,646 ng/g (arithmetic mean, *Der p* I 7,204 ng/g and *Der f* I 3,457 ng/g)
	Texas, USA, April-May	10-50,900 ng/g (median, 575 ng/g)
	North Carolina, USA, late spring	7,000 ng/g (geometric mean)
Cat (*Fel d* I) 18 studies	United States, 34% cat ownership among households	8-6,000 ng/g (geometric mean 6,000 ng/g)
Cockroach (*Bla g* I *or* II) 10 studies	North Carolina, USA Texas, USA	4,600 ng/g (geometric mean) 1.6–15.4 ng/g (median, 5.7 U/g)

Related Studies

Although studies identify that hard floors retain less allergen content and are recommended in avoidance studies, carpeting remains a common trend for homes, office, and school locations (Macher, Tsai et al. 2002; Causer, Lewis et al. 2004; Causer, Shorter et al. 2006). The popularity of carpeting seems to be because of the appearance, texture, insulating, and sound absorbing properties (Causer, Lewis et al. 2004). Additional literature suggests that the carpet construction, wear, and cleaning methods should be considered in the evaluation of allergen loading content.

In 2002 at the Indoor Air conference in Monterey, California the results of concentrations of *Fel d* I, *Der f* I, and *Der p* I allergens was reported for 93 of 100 U.S. office buildings. Samples were collected during the summer and winter months throughout the United States. Cat allergens were found in almost all BASE buildings even though cats seldom enter offices (Macher, Tsai et al. 2002). Dust mite allergens were detected in approximately half of the samples (Macher, Tsai et al. 2002). Possible

sources of these allergens again are the occupants and their belongings (IOM 1993; IOM 2002) but also colonization of suitable habitats in offices, such as carpets and upholstered furniture that provide mites with food (primarily human skin flakes), moisture, warmth, and protection (Macher, Tsai et al. 2002).

Floor covering construction which may affect the how allergens are inhaled and which may provoke an allergic response depends on the type of disturbance, environmental conditions, the level of contamination, and the nature of the floor covering and where the allergen resides within it (Causer, Shorter et al. 2006). The most common carpeting construction in the world is reportedly synthetic. Circumstantial evidence within the report suggests that a relationship between carpet construction and allergen content exists (Causer, Shorter et al. 2006). From the Causer study (2006) it appears that the extent of the pile flattening that occurs after carpet wear differs between carpet types, but neither pile height, pile conformation, pile density, nor yarn twist greatly influence how much *Der p* I collects within the carpet pile.

Vacuuming has been studied extensively as an intervention to improve hygiene and to decrease the frequency and severity of asthma, allergies, and other health outcomes (Corsi, Siegel et al. 2008). However, dry vacuum cleaning one to two times per week may be sufficient to remove soil from carpeting it will not be regular enough to reduce the allergen load in carpet to the point where a clinical improvement could be expected (Causer, Lewis et al. 2004). Causer (2004) found that the dust recovery rates were not proportional to allergen recovery rates, and that the ratio was influenced, to some extent, by the carpet construction. Carpeting constructed of low pile height and density appeared to have less of an allergen content because dry vacuuming was able to

recover a more significant amount dust. In the Causer (2004) study, carpet cleaning methods were evaluated based upon no vacuuming, wet extraction, and dry extraction. Each extraction was performed separately. The study concluded that vacuuming procedures must be performed on a regular basis, and techniques employed to avoid the increased exposure to airborne allergen that occurs during vacuuming. In addition, while unworn carpets removed allergens through vacuuming the efficiency of removal with worn carpeting was considerably reduced.

METHODS

Study Design

This research project was a study of the concentrations of cat (*Felis domesticus* I), dust mite (*Dermatophagoides pteronyssinus* I *and Dermatophagoides farinae* I*)*, and cockroach (*Bla g* I*)* allergens in carpeting in an elementary school kindergarten class. The hypothesis of this research was that kindergarten children increase their exposure to allergens as a result of the time spent on floor-based school activities and the concentration levels of allergens in the carpeting. This project was not designed to assess the allergic response in the students or teaching staff.

While literature is available describing sampling techniques for reservoir dust, dust collection devices, performance of vacuum in collecting allergen samples in residential buildings, limited literature is available for public school settings. At this time, there has been no standard developed for reservoir dust sampling. Therefore, the DACI Laboratory *Dust Analysis Method* was utilized for sampling technique, and sample duration.

The kindergarten class and teaching staff did not participate in any part of the research project. Therefore, the University of South Florida Institutional Review Board (IRB) waived further requirements.

Only one kindergarten class was observed during this research and reservoir dust samples were obtained from the same classroom throughout the duration of this project.

The limitations associated with this research include the age of the carpeting; custodial maintenance, building construction, and teaching curriculum, which varies in all other kindergarten classrooms or public schools. However, this data provides a foundation for further research and assessments.

Classroom Observation

In the elementary schools, kindergarten classes typically spend a portion of the school day with floor-based activities. Following discussions regarding curriculum and scheduled classroom activities with the kindergarten teacher, group floor activities where identified. The kindergarten teacher provided a curriculum plan. Tables 4 and 5 detail the classroom curriculum. Student group floor-based activities are conducted at the front of the classroom on the carpeting which is adjacent to the white board and the teacher's work desk and is a common area for people to walk through the classroom. In addition, there are several floor "centers" for individual student activities which are located throughout the classroom. The kindergarten class has a total of nineteen students enrolled. The kindergarten class was observed throughout a school year to document and quantify classroom activities which are floor-based. The information documented includes; occupancy of classroom, occupied floor area, occupant density, and time spent on carpeting. The data compares the time spent in the group floor setting to the total time spent in the classroom during a school day.

Students enter the classroom at 8:30 AM and place their belongings at the back of the classroom in open lockers. Students then return to the classroom and go to the carpeted area designated for the group. Group floor time lasts for approximately fifteen to twenty minutes. During the morning floor time the student announcements, Pledge of

Allegiance, and attendance are conducted. The group floor based activities of the students includes standing, sitting in place, and jogging in place. Students return to their assigned desks until approximately 10:00 AM at which time they return to the group floor area for a group comprehension lesson that extends until approximately 10:15 AM. Students leave the group floor area for other classroom instruction until lunch at which time they leave the classroom through the exterior door and walk to the cafeteria. Students return to the classroom at 11:52 AM and return to their desks. Recess is scheduled from 12:10 PM until 12:30 PM. Following recess students return to the carpeted area for approximately thirty minutes. Specials classes, such as fine art and physical education, occur at the end of the school day. Student dismissal occurs at 2:45 PM.

On the November 19, 2007 classroom observation of an adjoining classroom of third grade students assisted the kindergarten class in a special activity which involved sitting on the carpeted group area. The height of the students observed in the sitting position is approximately twenty-four inches from of the ground. On December 14, 2007, students who completed their desk work early were permitted to go to the carpeted floor area to read and were observed to sit, stand, or lay down with their faces against the carpet. Group floor activities do not include assigned seating which allows students to sit in different locations within the designated group floor area. Prior to the start of the classroom activities at 9:49 AM on January 22, 2008 was observed to include eating snacks at their student desks. Following the 9:49 AM group floor activity students subsequently moved to different individual floor “centers” and student desks for other classroom activities. The February 12, 2008 classroom observation involved special Valentine’s Day activities for the group floor area and additional group floor time was

planned due to the school campus participating in the Florida Comprehensive Aptitude Test (FCAT). On February 19, 2008, the teacher was in meetings for the day which required a substitute teacher who spent an extended period of the class time on the floor in group activities. During the various classroom observations on March 25; April 30; May 15; and May 21, 2008 involved students standing, sitting, and walking in place within the designated group floor area.

Although the curriculum agenda for the kindergarten class was identified by the teacher variations from this regularly occurred as a result of substitute teachers, special guest speakers, and special classroom activities, such as Valentine's Day and the FCAT. Therefore, the curriculum agenda was utilized to identify the potential duration of exposure.

Sampling Strategy

The Johns Hopkins University Reference Laboratory for Dermatology, Allergy, and Clinical Immunology (DACI) recommends school reservoir dust specimen collection site selection should target areas where large numbers of allergic workers or students congregate. The sample area was determined by the location of the group floor based activities. During a student group floor activity the carpeted area was marked with Duct Tape which outlined the location of the students. Subsequently, the area was measured and recorded. The area marked during the group floor activity remained in place throughout the duration of the research project. Figure 2 diagrams the classroom design.

Eight reservoir bulk dust samples were collected from the carpeted floor utilizing a "nozzle sock" sample media. The "nozzle sock," provided by the DACI Laboratory, is a vacuum cleaner adapter that is inserted into the base of the vacuum hose portion of the

vacuum cleaner. The “nozzle sock” is manufactured from a refined glass spun polyethylene fiber pulp media (Hamilton, Assessment). The “nozzle sock” was inserted into a Numatic International RSV 130 back pack vacuum cleaner, serial number 034413843, which is utilized by the custodial cleaning staff. The same RSV 130 back pack vacuum cleaner was utilized throughout the research project. In addition, three Numatic International low profile vacuum floor tools were purchased for the research project to ensure the reservoir dust samples collected obtained from the pre-designated sampling area did not have cross contamination of dust from other locations throughout the school. Following each sampling event, the low profile vacuum floor tools were cleaned utilizing microfiber cloths.

The DACI Laboratory sampling protocol requires vacuum samples be collected for a minimum of two minutes. Vacuum samples were timed using a standard stop watch for the duration of two minutes. The entire designated floor area was continuously vacuumed horizontally for one minute and vertically for one minute. Samples were collected following school dismissal. The “nozzle sock” was subsequently removed from the vacuum cleaner hose and placed into the plastic zip lock bag provided and mailed to the DACI Laboratory. Sample collection occurred monthly during the 2007-2008 school year, with the exception of December due to the shortened holiday month.

Sample Analysis

Sample analysis was performed by the DACI Laboratory. Upon receipt of the reservoir dust samples the DACI laboratory sieves crude dust through a 50-mesh metal sieve onto waxed laboratory weighing paper to allow dust particles smaller than 240 microns to pass through (Hamilton 2005). Following the sieving process the fine dust

particles are weighed on an analytical balance. Results are reported in mass quantities of allergen per gram of dust. One hundred milligrams of fine reservoir dust is extracted in 2 mL of filtered phosphate-buffered saline (PBS) containing protein, such as 1% bovine serum albumin (Hamilton 2005). Suspended samples are stored for approximately 12 to 16 hours and then centrifuged so that the solid may be collected and removed. The allergen extract is subsequently frozen at -20 ºC or lower.

The DACI laboratory utilizes two-site immunoenzymatetric assays (IEMAs) to quantify *Fel d* I, *Der p* I, *Der f* I, and *Bla g* I allergens. Each allergen IEMA is initiated by adsorbing the allergen-specific capture antibody on a plastic microtiter plate (Hamilton 2005). The interpolated levels of allergen are then corrected for the mass of the dust extracted, and final results are reported as µg or U/g of fine dust (Hamilton 2005). The DACI laboratory calibrates standards against various reference preparations.

Based upon literature research and discussions with Johns Hopkins Asthma and Allergy Center the DACI Laboratory was chosen for vacuum sample analysis. The Housing and Urban Development (HUD) Office of Healthy Homes and Lead Hazard Control in 2004 provided HUD's Health Homes Initiative Grantees a Background and Justification for Vacuum Sampling Protocol for Allergen in Household Dust which summarizes the various vacuums and dust collection devices available for reservoir dust sampling.

RESULTS

Classroom Observation

Twelve classroom observations of group floor activities were performed from November 17, 2007 through May 21, 2008. Table 3 presents the student occupancy, date of classroom observation, total amount of time students spent on the carpeted floor, and the activities the students performed while in their groups.

Table 3: Classroom Observation of Floor Based Group Activities

Date	Student Occupancy	Start Time	Stop Time	Total Minutes	Activities
11/19/07	18	8:59 AM	9:15 AM	16	sitting, standing, jogging in place
11/19/07	18	9:49 AM	10:03 AM	14	sitting, 3rd grade class participates
12/14/07	19	9:05 AM	9:35 AM	30	laying down on floor with face against carpet, sitting, standing in place
1/22/08	17	8:43 AM	9:13 AM	31	sitting, standing
1/22/08	17	9:49 AM	10:30 AM	38	sitting, standing, laying down on floor with face against carpet
2/12/08	18	12:14 PM	12:30 PM	16	sitting, standing, stomping in place
2/19/08	17	8:43 AM	9:46 AM	63	sitting, standing, walking in place
3/25/08	17	8:44 AM	9:24 AM	25	sitting, standing
4/30/08	18	8:41 AM	9:00 AM	19	sitting, standing
4/30/08	18	9:44 AM	9:52 AM	8	sitting, standing
5/15/08	18	12:40 PM	1:18 PM	38	sitting
5/21/08	17	8:36 AM	9:00 AM	24	sitting, standing, walking in place

Table 4 and 5 details the curriculum agenda. The curriculum agenda was altered during the month of April, 2008. As a result, additional group floor time activities were initiated.

Table 4: Curriculum Agenda August, 2007 - April, 2008

Time	Curriculum Plan	Total Time (min)
8:30 - 8:40 AM	Enter Classroom	10
8:40 - 9:15 AM	Welcome/Attendance (Group Floor)	35
9:15 - 10:00 AM	Student Desks	45
10:00 - 10:15 AM	Group Floor	15
10:15 - 10:30 AM	Centers Individual Floor Activities	15
10:30 - 11:00 AM	Centers Individual Floor Activities	30
11:00 - 11:15 AM	Student Desks	15
11:15 - 11:52 AM	Out of Classroom (Lunch)	37
11:52 - 12:10 PM	Student Desks	18
12:10 - 12:30 PM	Out of Classroom (Recess)	20
12:30 - 1:15 PM	Group Floor	45
1:15 - 2:00 PM	Specials - Out of Classroom	45
2:00 - 2:15 PM	Student Desks	15
2:15 - 2:45 PM	Student Desks	30
2:45 PM	Dismissal	
	Total Group Floor Time	95
	Total Individual Floor Time	45
	Total Carpet Time	140
	Total Classroom Time	273

Table 5: Curriculum Agenda April, 2008 - May, 2008

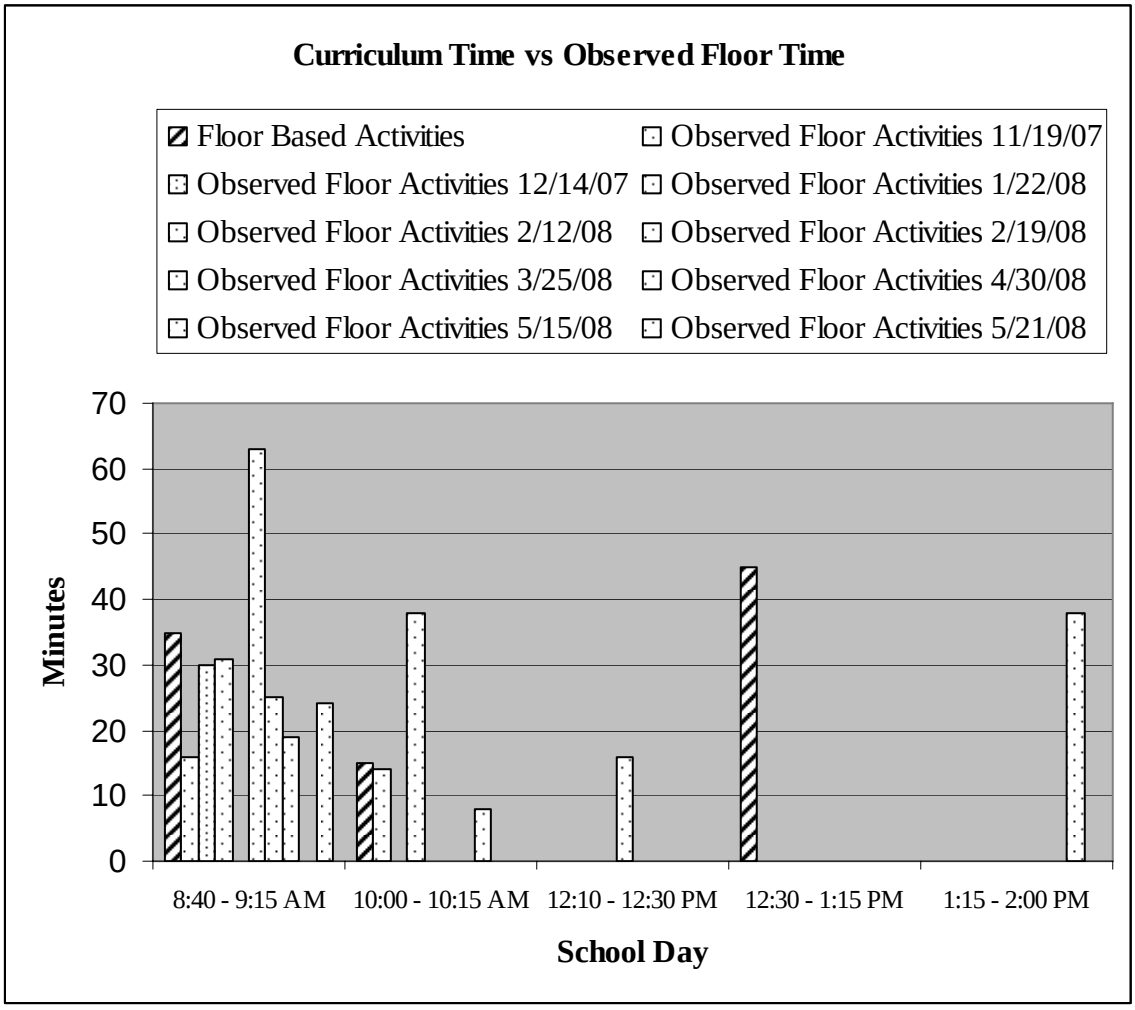
Time	Curriculum Plan	Total Time (min)
8:30 - 8:40 AM	Welcome/Enter Classroom	10
8:40 - 9:00 AM	Group Floor	20
9:00 - 9:10 AM	Group Floor	10
9:10 - 9:25 AM	Center Individual Floor Activities	15
9:25 - 9:35 AM	Student Desks	10
9:35 - 9:45 AM	Group Floor	10
9:45 - 10:00 AM	Center Individual Floor Activities	15
10:00 - 10:10 AM	Group Floor	10

Table 5 (Continued): Curriculum Agenda April, 2008 - May, 2008

10:10 - 10:25 AM	Center Individual Floor Activities	15
10:25 - 10:35 AM	Group Floor	10
10:35 - 10:50 AM	Center Individual Floor Activities	15
10:50 - 11:00 AM	Group Floor	10
11:00 - 11:20 AM	Student Desks	20
11:20 - 11:52 AM	Out of Classroom (Lunch)	32
11:52 - 12:10 PM	Group Floor	18
12:10 - 12:30 PM	Out of Classroom (Recess)	20
12:30 - 1:15 PM	Group Floor	15
1:15 - 1:55 PM	Specials - Out of Classroom	40
1:55 - 2:40 PM	Student Desks	45
2:40 - 2:45 PM	Student Desks	5
2:45 PM	Dismissal	
	Total Group Floor Time	103
	Total Individual Floor Time	60
	Total Carpet Time	163
	Total Classroom Time	253

The total time students spent in group floor activities ranged from 95 to 103 minutes. The total time students spent within their classroom each day ranged from 253 and 273 minutes. The percentage of the students classroom time spent on the designated group floor area was 38% of their day. Figure 1 shows the actual observed time students spent in group floor activities compared to the curriculum agenda.

Figure 1: Curriculum Agenda vs Observed Floor Time



Classroom Design and Classroom Maintenance

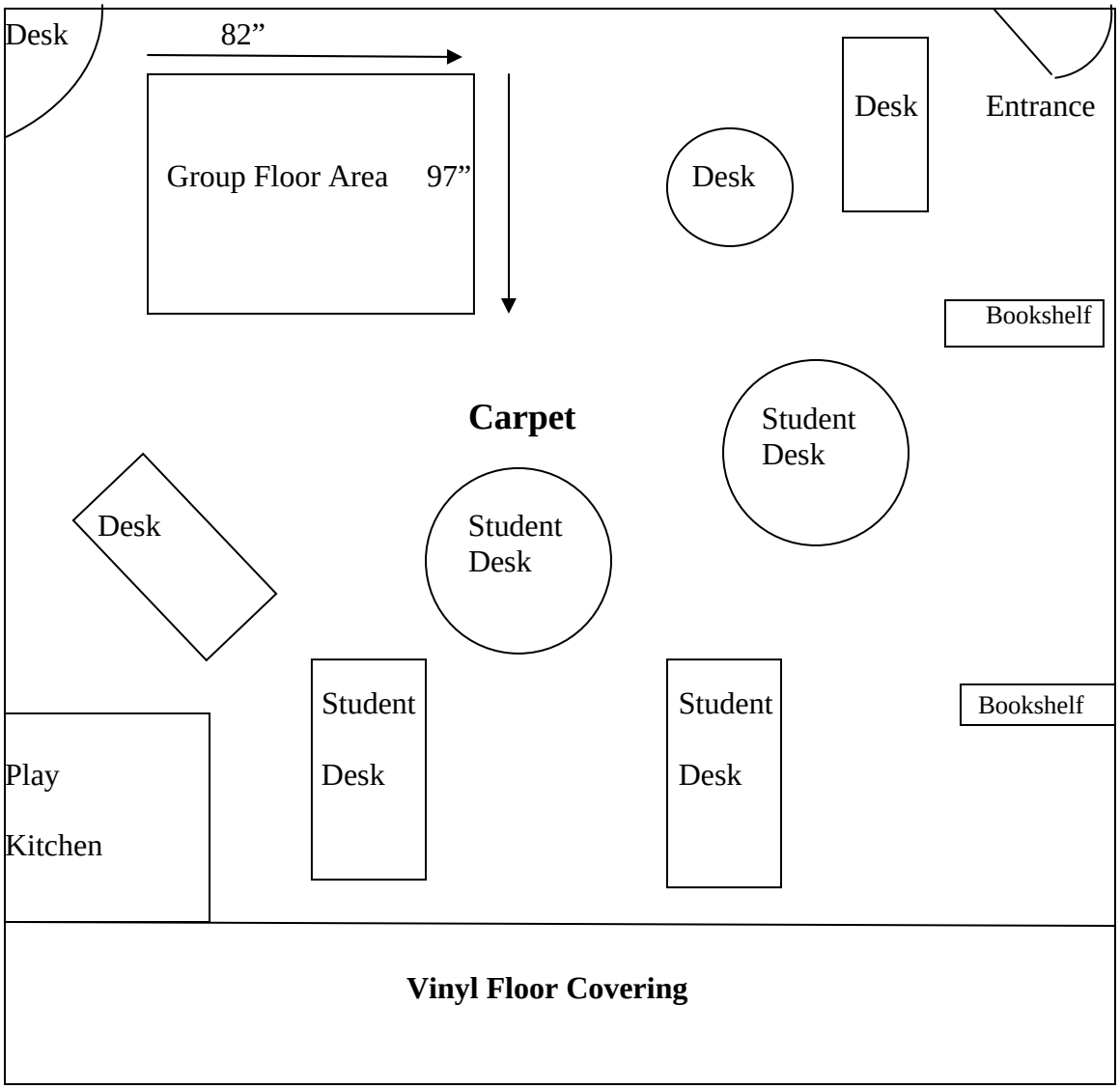
Figure 2 shows the interior lay-out of the kindergarten classroom. The designated group floor area was 82 inches long and 97 inches wide. The group floor area was positioned at the front of the classroom adjacent to the exterior wall where the white board and teacher work desk were located. Samples were collected within the same location throughout the study. The student desks, individual "centers", teacher desk, and bookshelves are carpeted. The back of the class where the classroom sink and restroom are located is vinyl floor covering. The carpeting in the classroom is grey and

approximately 18 years old. The kindergarten class enters the building through an exterior door.

The building design includes four classrooms which are connected to a central pod area where the teacher planning room and workroom are located. Each classroom has an individual exterior door. The building is constructed of concrete masonry block units and a brick façade. The ceilings are Tectum™ and wood trusses. The heating, ventilation, and air-conditioning unit is controlled by the teachers with a single twist timer. The air handling system operates on a chilled water system with plenum return.

Custodial maintenance of the classroom included vacuuming nightly utilizing the Numatic International RSV 130 back pack vacuum cleaner. No further carpet maintenance was performed throughout the duration of this study. Dusting within classrooms only occurs on open horizontal surfaces where bookshelves, top of computers, filing cabinets, etc. are not cluttered with personal affects, papers, or books. The frequency of classroom dusting depends on availability among custodial staff.

Figure 2: Classroom Design (Not to Scale)



Analytical Results

Reservoir dust sampling was performed from November 30, 2007 through July 7, 2008. A total of eight samples were collected approximately every month, with the exception of December, 2007. Two weeks in December the school district is closed for winter break, therefore, sampling did not occur during this month. Temperature and relative humidity within the classroom ranged from 70 ºF to 76 ºF and 38% to 56% during the school year. During the unoccupied mode of the classroom the temperature and relative humidity were 83ºF and 67%, respectively. Table 6 provides a summary of temperature and relative humidity within the classroom during sampling events.

Table 6: Temperature and Relative Humidity within the Classroom

Sample Date	**Temperature (ºF)**	**Relative Humidity (%)**
11/30/07	72	52
01/14/08	70	49
02/12/08	71	53
03/07/08	73	54
04/14/08	76	38
05/05/08	73	54
06/03/08	72	56
07/07/08	83	67

Specific sampling results are located in Appendix A. Tables 7 and 8 provide a summary of sample results throughout the duration of the study. Table 9 summarizes the sample results presented in Table 7 and 8.

Table 7: Sample Results November, 2007 - March, 2008

	Sample Date			
	11/30/07	01/14/08	02/12/08	03/07/08
Allergen	ng/g	ng/g	ng/g	ng/g
Der p I	5239	16889	3408	4438
Der f I	2651	5951	704	1574
Fel d I	3924	7858	3086	2206
Bla g I	<0.7 U/g	<0.4 U/g	<0.4 U/g	<0.4 U/g

Table 8: Sample Results April, 2008 - July, 2008

	Sample Date			
	04/14/08	05/05/08	06/03/08	07/07/08
Allergen	ng/g	ng/g	ng/g	ng/g
Der p I	86704	30656	15232	26186
Der f I	6253	2718	2735	6720
Fel d I	4135	3856	10558	5797
Bla g I	<0.4 U/g	<0.4 U/g	<0.4 U/g	<0.4 U/g

Table 9: Descriptive Summary of Sample Results

Allergen	Mean	Median	Standard Deviation	Maximum	Minimum
Der p I	23594	16061	27427	86704	3408
Der f I	3663	2727	2305	6720	704
Fel d I	5178	4030	2781	10558	2206

The sample location and total area sampled remained consistent throughout the study. The mean sampling time was 2 minutes 4 seconds. DACI laboratory recommends vacuuming for at least 2 minutes. The reservoir dust analysis “Instructions for Use” is located in Appendix B. The DACI laboratory risk range characterization for allergens is provided in Table 10.

Table 10: DACI Risk Range

Allergen	Low	Medium	High
Der p I	< 400 ng/g	400 – 2000 ng/g	> 2000 ng/g
Der f I	< 400 ng/g	400 – 2000 ng/g	>2000 ng/g
Fel d I	< 8000 ng/g	8000 – 80000 ng/g	> 80000 ng/g

Der f I results ranged from 704 ng/g on February 12, 2008 to 6,720 ng/g on July 7, 2008. *Der p* I results ranged from 3,408 ng/g on February 12, 2008 to 86,704 on April 14, 2008. Sample results for *Fel d* I ranged from 2,206 ng/g on March 7, 2008 to 10,558 ng/g on June 3, 2008. The risk range for cockroach allergen has currently been established and therefore the DACI laboratory considers any positive results to be significant. All *Bla g* I samples were below the detection limit throughout the study and therefore do not present a significant risk for sensitization or symptoms. Tables 11 and 12 provide the DACI laboratory risk range characterization of the reservoir dust sample results.

Table 11: DACI Risk Range for Sample Results November, 2007 - March, 2008

	Sample Date							
	11/30/07		01/14/08		02/12/08		03/07/08	
Allergen	ng/g	DACI	ng/g	DACI	ng/g	DACI	ng/g	DACI
Der p I	5239	High	16889	High	3408	High	4438	High
Der f I	2651	High	5951	High	704	Moderate	1574	Moderate
Fel d I	3924	Low	7858	Low	3086	Low	2206	Low
Bla g I	<0.7 U/g		<0.4 U/g		<0.4 U/g		<0.4 U/g	

Table 12: DACI Risk Range for Sample Results April, 2008 - July, 2008

	Sample Date							
	04/14/08		05/05/08		06/03/08		07/07/08	
Allergen	ng/g	DACI	ng/g	DACI	ng/g	DACI	ng/g	DACI
Der p I	86704	High	30656	High	15232	High	26186	High
Der f I	6253	High	2718	High	2735	High	6720	High
Fel d I	4135	Low	3856	Low	10558	Moderate	5797	Low
Bla g I	<0.4 U/g		<0.4 U/g		<0.4 U/g		<0.4 U/g	

Laboratory analytical results are found in Appendix C.

DISCUSSION AND CONCLUSIONS

The purpose of this research was to characterize kindergarten children's' exposures to *Fel d* I, *Der p* I, *Der f* I, and *Bla g* I allergens in carpeting and to quantify classroom time spent on the floor. The literature regarding the characteristics of the most commonly reported agents that are associated with allergic responses to carpeting are cat, dust mite, and cockroach antigens. The review indicated that there is strong evidence that continuous exposure to high risk levels of allergens plays a role in symptoms and sensitization. The review also indicated there is little literature on the duration of exposure to allergens and the locations of exposures within a classroom.

This study sought to quantify the concentrations of cat, dust mite, and cockroach allergens in carpeting and document and quantify classroom activities which are floor-based. Over the course of a calendar school year the concentrations of cat, dust mite, and cockroach allergens were measured within reservoir dust samples. Additionally, classroom observations were performed to identify the location of group floor-based activities, the number of students, the duration of time students spent in the group floor-based activities, and the activities students performed while in groups on the floor. The interior of the classroom and building design was described to put in relation to where the group floor based activities occurred compared to the rest of the classroom activities and how custodial cleaning activities were performed throughout the school year. This information is necessary to

understand the potential for reservoir dust accumulation and the efficiency of allergen removal associated with the cleaning method.

The research did have limitations. The study design focused on one elementary school and one kindergarten classroom. Only the location of group floor-based activities was targeted to be sampled for reservoir dust samples, however, other floor activities did occur throughout the classroom in individual centers. Actual time spent in group floor-based activities varied from the scheduled curriculum plan throughout the school year because of substitute teachers with different curriculum plans, special holiday programs, such as Valentine's Day, FCAT, and other classroom presentations by outside school district personnel. Other variables include, the age of the school, school floor plan, and age of carpeting.

Discussion

The analytical results of this study were consistent with other reported studies. Tranter (2005) reviewed forty-one papers on indoor allergens in settled school dust and reported the findings and significant factors. The statistical data reported within the D.C. Tranter review included the geometric mean, arithmetic mean, and the median from the compilation of papers. The cat allergen concentrations were higher (2,206-10,558 ng/g) than the reported other studies (8-6,000 ng/g). Tranter (2005) reported the geometric mean of cat allergen within the United States from eighteen school studies as 6,000 ng/g, while the arithmetic mean from Table 9 was 5,178 ng/g. Dust mites concentrations were similar (704-86,704 ng/g) among the other reviewed studies by Tranter (42-50,900 ng/g). The arithmetic mean from a Florida school study for *Der p* I was three times lower than Table 9 results (23,594 ng/g), while the Tranter

(2005) literature reported an arithmetic mean for *Der f* I as 3,457 ng/g which was comparable to Table 9 results of 3,663 ng/g. There were no measurable level of cockroach allergens in this study, however, the geometric mean reported from a North Carolina school was 4,600 ng/g. One aspect which made the research presented here unique was the complete assessment of the classroom for an entire calendar school year. Another important aspect of this study was the quantification of exposure to the carpeting in a specific higher risk class.

The group floor area measurements for *Der p* I and *Der f* I ranged from 704 ng/g to 86,704 ng/g. Dust mites are associated with seasonal changes. The sample results on January 14, 2008 and April 14, 2008 were 16,889 ng/g and 86,704 ng/g, respectively, were above the expected concentrations compared to the other surrounding month's measurements. A number of variables, such as the time of year samples were collected, temperature of the classroom, relative humidity of the classroom, and custodial maintenance of the classroom, may have contributed to the wide range of concentrations of dust mites and inconsistency of measurements each month. Low to moderate levels of *Fel d* I concentrations were measured within the classroom, 2,206-10,558 ng/g. Since there are no cats in the classroom the measurement levels are a direct result of transference by the occupants. The students of the classroom were not interviewed, so the number of pet owners is unknown. All *Bla g* I measurements were below detection limits of 0.4 U/g and 0.7 U/g. Although students were observed to eat in the classroom, sanitary practices within the classroom appeared to have been adequate.

Measurable concentration levels were compared to the DACI laboratory criteria for potential risk exposures among atopic or sensitized individuals from home studies. These values are also referred to as 'exposure risks,' 'levels of concern,' provisional standards,' and 'symptom/sensitization levels' (Tranter 2005). However, the DACI criteria provide a good benchmark for comparison of concentrations and risk.

The total amount of time students spent daily in their classroom was between 253 and 273 minutes. Of this, 95 to 103 minutes was spent in group floor based activities. The observations of the classroom group floor based activities revealed that the curriculum plan was more of a guideline and depending on the day, other school activities, and the time of year actual time spent on the floor varied. The total number of students enrolled in the class was 19. During the group floor based activities, up to 19 students participated in curriculum activities, which ranged from sitting, standing, walking in place, jogging in place, and reading quietly on the floor. Depending on the activity, student participation provided different levels of carpet disturbances which may allow for varying levels of allergens to become aerosolized. While, there is no direct method for analyzing the actual inhaled concentration of aeroallergens there is evidence that dose-response relationships occur with higher levels of allergens present in settled dust reservoirs.

The building heating, ventilation, and air conditioning system was manually controlled by the teachers, and on nice days, classroom doors would be left open. The building design also allowed for people to enter each classroom from an exterior door. The exterior entrance went directly onto the carpeting and in order to cross the

classroom, visitors, students, etc. had to cross through the group floor area of the carpeting. This allowed for moisture on shoes, dirt, and debris to be tracked into the room and into the carpeting. In addition, the carpet was approximately 18 years old, which has resulted in the carpeting becoming more compact over time and has more of a likelihood of higher accumulation of allergens.

Conclusions

Kindergarten children spent approximately 38% of their classroom day in floor based group activities. The students were observed performing activities during the time spent on the carpet, which resulted in varying levels of carpet disturbance. Classroom observations identified differences in the planned curriculum and actual time spent on the floor. Therefore, the planned curriculum timeline was used to provide the exposure duration to allergens during the group floor activities. The conclusion can be drawn, however, that in a kindergarten classroom floor-based activities play an important role in the curriculum teaching for the children.

The group floor area measurements included moderate to high levels of *Der p* I and *Der f* I, low to moderate levels of *Fel d* I, and below detection limits of *Bla g* I. Concentrations of the allergens measured ranged from 2,206 ng/g – 10,558 ng/g *Felis domesticus* I, 3,408 ng/g – 86,704 ng/g *Dermatophagoides pteronyssinus* I, 704 ng/g – 6,720 ng/g, *Dermatophagoides farinae* I, and below detection limits for *Bla g* I, respectively. The concentration measurements varied from month to month and therefore, were not consistent throughout the study. Based upon the DACI criteria for risk range, high allergen concentrations (*Der p* I 86,704 ng/g and *Fel d* I 10,558 ng/g) were recorded in between months with concentrations of 3 to 19 times lower. The

analytical results suggest that there are significant levels of allergens in the carpeting and many variables play an important role in the concentrations. Based upon the DACI laboratory criteria there is a moderate to high risk of increased sensitization or allergic symptoms as a result of the repeated and continued exposure to the carpeting.

Recommendations for Future Research

Based on the findings from this study, the following recommendations for future research are provided. These include:

- Expand the study and select more kindergarten classrooms with different age carpeting to better characterize concentration levels.
- Conduct a cross-sectional epidemiological study that uses surveys designed to measure current health problems of occupants and reservoir dust sampling to assess exposure.
- Develop sampling protocol for airborne measurements for *Der p 1, Der f 1, Fel d 1,* and *Bla g*. This will provide a better understanding of exposure risk and dose-response.

References Cited

ALA (2008). "Childhood Asthma Overview." Retrieved December, 2008.

Causer, S., R. D. Lewis, et al. (2004). "Influence of Wear, Pile Height, and Cleaning Method on Removal of Mite Allergen from Carpet." Journal of Occupational and Environmental Hygiene **1**: 237-242.

Causer, S., C. Shorter, et al. (2006). "Effect of Floorcovering Construction on Content and Vertical Distribution of house Dust Mite Allergen, Der p 1." Journal of Occupational and Environmental Hygiene **3**: 161-168.

Corsi, R. L., J. A. Siegel, et al. (2008). "Particle Resuspension During the Use of Vacuum Cleaners on Residential Carpet." Journal of Occupational and Environmental Hygiene **5**(4): 232-238.

Elliott, L., S. J. Arbes Jr., et al. (2007). "Dust Weight and Asthma Prevalence in the National Survey of Lead and Allergens in Housing (NSLAH)." Environmental Health Perspectives **115**(2): 215-220.

FLDOE (2008). "Accountability, Research, and Measurement." Retrieved December, 2008.

German, J. A. and M. B. Harper (2002). "Environmental Control of Allergic Diseases." American Family Physician **66**(3): 421-426.

Gold, D. R. (2000). "Environmental Tobacco Smoke, Indoor Allergens, and Childhood Asthma." Environmental Health Perspectives **104**(4): 643-651.

Hamilton, R. G. (2005). "Assessment of Indoor Allergen Exposure." Current Allergy and Asthma Reports **5**: 394-401.

Hamilton, R. G., M. D. Chapman, et al. (1992). "House Dust Aeroallergen Measurements in Clinical Practice: A Guide to Allergen-Free Home and Work Environments." Journal of Immunology and Allergy Practice **14**(3): 9-25.

IOM (1993). Magnitude and dimensions of sensitization and disease cause by indoor allergens; Agents, sources, source controls, and diseases; Engineering control strategies. Indoor Allergens, Washington D.C., Institute of Medicine.

IOM (2002). Indoor biologic exposures. . In Clearing the Air: Asthma and Indoor Air Exposures, Washington D.C., Institute of Medicine.

Macher, J., H. Ammann, et al. (1999). Bioaerosols Assessment and Control. Cincinnati, American Conference of Governmental Industrial Hygienist.

Macher, J., F. Tsai, et al. (2002). Concentrations of cat and dust mite allergens in 93 U.S. office buildings. Indoor Air Conference, Monterey, CA, International Academy of Indoor Air Sciences.

Platts-Mills, T. A. E., R. B. Sporik, et al. (1995). "Is there a dose-response relationship between exposure to indoor allergens and symptoms of asthma?" The Journal of Allergy and Clinical Immunology **96**(4): 435-439.

Ramachandran, G., J. L. Adgate, et al. (2005). "Indoor Air Quality in Two Urban Elementary Schools - Measurements of Airborne Fungi, Carpet Allergens, CO2, Temperature, and Relative Humidity." Journal of Occupational and Environmental Hygiene **2**(553-566): 553.

Tovey, E. and G. Marks (1999). "Methods and Effectiveness of Environmental Control." Journal of Allergy and Clinical Immunology **103**(2 pt 1): 179-191.

Tranter, D. C. (2005). "Indoor allergens in settled school dust: a review of findings and significant factors
" Clinical and Experimental Allergy **35**: 126-136.

USEPA (2008). "Indoor Air Quality." Retrieved December, 2008.

Appendices

Appendix A: Sample Information

Location/Site	Elementary School/Group Floor Area			
Method	DACI Dust Analysis			
Equipment	Numatic International Vacuum Cleaner, RSV 130, Serial #044613259			
Sample ID	R1623	R2172	R2602	R3005
Sample Date	11/30/07	01/14/08	02/12/08	03/07/08
Sample Time	2:45 PM	3:00 PM	2:45 PM	2:45 PM
Classroom Occupancy	19	19	18	19
Sample Duration	2 mn 5 sec	2 mn 7 sec	1 mn 56 sec	2 mn 8 sec

Location/Site	Elementary School/Group Floor Area			
Method	DACI Dust Analysis			
Equipment	Numatic International Vacuum Cleaner, RSV 130, Serial #044613259			
Sample ID	R3419	R3735	R4011	R4446
Sample Date	04/14/08	05/05/08	06/03/08	07/07/08
Sample Time	3:24 PM	3:04 PM	1:30 PM	11:40 AM
Classroom Occupancy	15	18	18	0
Sample Duration	2 mn 1 sec	2 mn 7 sec	2 mn 5 sec	2 mn 18 sec

Appendix B: DACI Laboratory Instruction for Use

THE JOHNS HOPKINS UNIVERSITY SCHOOL OF MEDICINE

REFERENCE LABORATORY FOR DERMATOLOGY, ALLERGY AND CLINICAL IMMUNOLOGY

DACI

DUST ANALYSIS

Dust Mite Allergen – Cat Allergen – Dog Allergen – Cockroach Allergen – Mouse Allergen – Rat Allergen
Mold Spore Colony Count

This easy test offers an assessment of exposure to specific allergens in your home or work environment

Instructions for Use

Enclosed you will find the following materials for collecting dust samples with your vacuum cleaner. Please check to see that the following items are enclosed in your packet.

1. Dust Collector
2. Identification Closure Label (small)
3. Identification Label (large)
4. Ziploc bag
5. Remittance sheet

Please follow the instructions below and refer to the diagram on the reverse side of this page. If you have any questions, our toll-free line **800/344-DACI (3224)** is available from 8:30 a.m. – 5 p.m. Monday-Friday.

1. Remove any attachment from end of hose.
2. Insert dust collector into hose with the collector filter material end first; be sure that all of the filter material is within the hose as well as ½" of plastic material.
3. Fold the plastic material down around the outside of the hose.
4. Attach vacuum tool, such as upholstery head, wand, etc. to hose over the folded-down plastic extended and vacuum using the following guidelines:

GENERAL VACUUMING GUIDELINES: Sites of dust collection should take into consideration especially where the patient spends most of his/her time. It is important that the quantity of fine dust you collect is sufficient for our laboratory analysis. No conscious effort should be made to avoid the collection of sand, human and animal hair, textile fibers, lint, human dander, or insect parts. For practical reasons, vacuum the mattress first, upholstered furniture second, and then the floors. If carpet is not present, vacuum the bare floor adjacent to a major item of upholstered furniture.

FOR A GENERAL HOUSEHOLD SURVEY: Vacuum for at least 2 minutes at each of the following sites. Vacuum at least a one square yard area, preferably carpeted.

BEDROOM: After removing sheets and cloth covers, vacuum upper mattress surface (approximately 5' x 5' or the entire mattress surface if smaller). A mattress is sampled even if it is encased with plastic; encasements should not be removed. Vacuum floor areas.
FAMILY/LIVING ROOM: Vacuum the carpet in a large, exposed area for two minutes.
KITCHEN: Vacuum the floor, especially in corners and ledges where insects may reside.
BATHROOM: Vacuum carpet, rugs, and corners.

FOR A SPECIFIC ROOM SURVEY: Follow order of vacuuming above for specific area—mattress, upholstered furniture, and then floors. (If vacuuming an office site, vacuum carpet, corners of room, bookshelves, windowsills, and ledges, etc.)

5. Remove vacuum tool and collector. Complete the information on the Identification Closure Label. Fold plastic extender over twice and seal and closure label.
6. Complete the Write-On label (be sure to mark your name, address, vacuum date, etc.). Place the collector into the ziploc bag and seal.
7. Insert ziploc bag with dust into the self-addressed, postage-paid envelope and drop in the mailbox.

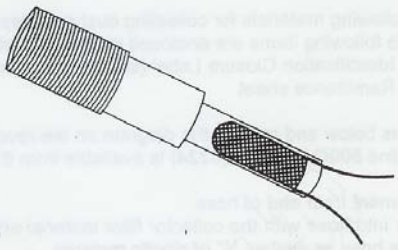
8/06

Room 1A20/5501 Hopkins Bayview Circle, Baltimore, Maryland 21224 410/550-2029, 800/344-3224, Fax 410/550-2030
Interstate License No. 19-1098 • Maryland State License No. 310 • Commonwealth of Pennsylvania No. 022620 • Florida Clinical Laboratory Law No. 80000 4946
CLIA 21D0649868 New York State Dept. of Health No. 4977

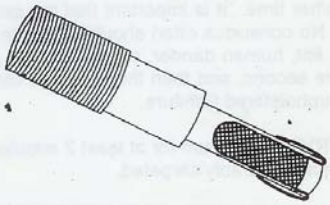
THE JOHNS HOPKINS UNIVERSITY SCHOOL OF MEDICINE
REFERENCE LABORATORY FOR DERMATOLOGY, ALLERGY AND CLINICAL IMMUNOLOGY

DACI

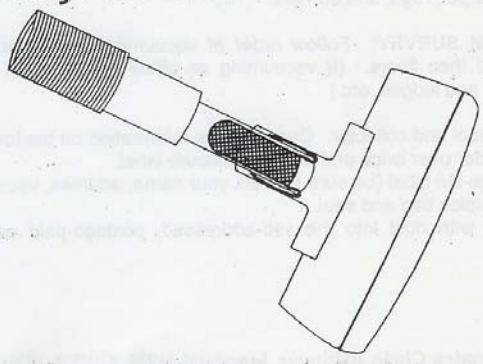
Step 1: Place collector into bottom of vacuum wand so that all of the filter media is within the wand.



Step 2: Fold the vinyl sleeve over the outside of the wand.



Step 3: Slide standard vacuum attachment over vinyl sleeve.



Room 1A20/5501 Hopkins Bayview Circle, Baltimore, Maryland 21224 410/550-2029 800/344-3224
Interstate License No. 19-1098, Maryland State License No. 310

Appendix C: Analytical Laboratory Results

Johns Hopkins University School of Medicine

DACI Reference Laboratory

for Dermatology, Allergy and Clinical Immunology

http://www.hopkins-allergy.org/services/daci.html

ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	DATE OF BIRTH GENDER AGE STATUS	 F BLDG 5 ROOM 003	SPECIMEN # SPEC DATE RECPT DATE PRINT DATE	R1623 11/30/07 12/4/07 12/17/07 3:37 pm

CLIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	REFERRING BILLING CODE CLIENT CODE DIAGNOSIS	NOT PROVIDED 14CC 1

PHONE 727-774-7949 FAX 727-774-7993 ACCT#

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	5239	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	2651	ng/G	HIGH
Cat (Fel d 1) Allergen (Dust)	3924	ng/G	LOW
Cockroach (Bla g 1) Allergen (Dust)	<0.7	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

Robert G. Hamilton, Ph.D., D.ABMLI
Director

These clinical tests were developed by the Johns Hopkins University DACI Reference Laboratory, which has determined their analytical performance characteristics. While the US Food and Drug Administration (FDA) has not cleared these tests, the FDA has determined that such clearance is not necessary. The Johns Hopkins University DACI Reference Laboratory is licensed under the Clinical Laboratory Improvement Amendments of 1988 (CLIA) to perform highly complex clinical testing.

Johns Hopkins University Asthma and Allergy Center, DACI Reference Laboratory, Room 1A20, 5501 Hopkins Bayview Circle Baltimore Maryland 21224 USA 410-550-2029 800-344-3224 Fax: 410-550-2030 www.hopkins-allergy.org/services/daci.html

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ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	DATE OF BIRTH GENDER F AGE STATUS 5.13m2		SPECIMEN # R2172 SPEC DATE 1/14/08 RECPT DATE 1/22/08 PRINT DATE 2/5/08 12:35 pm	
CLIENT NAME AND ADDRESS	PATIENT	REFERRING BILLING CODE CLIENT CODE DIAGNOSIS	NOT PROVIDED 14CC 1		
PHONE	FAX	ACCT#			

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	16889	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	5951	ng/G	HIGH
Cat (Fel d 1) Allergen (Dust)	7858	ng/G	LOW
Cockroach (Bla g 1) Allergen (Dust)	<0.4	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

Robert G. Hamilton, Ph.D., D.ABMLI
Director

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ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEWPORT RICHEY FL 34654	DATE OF BIRTH GENDER F AGE STATUS 54.97 SF		SPECIMEN # R2602 SPEC DATE 2/12/08 RECPT DATE 2/19/08 PRINT DATE 3/10/08 9:36 am	
CLIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	REFERRING BILLING CODE CLIENT CODE DIAGNOSIS	NOT PROVIDED 14CC 1		
PHONE 727-774-7949	FAX 727-774-7993	ACCT#			

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	3408	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	704	ng/G	MODERATE
Cat (Fel d 1) Allergen (Dust)	3086	ng/G	LOW
Cockroach (Bla g 1) Allergen (Dust)	<0.4	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

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Director

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MAR-10-2008 11:02 JHU DACI LAB 410 550 2030 P.001/001

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Johns Hopkins University School of Medicine

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ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREE BREEZE DR NEW PT RICHEY FL	DATE OF BIRTH GENDER AGE STATUS	F 54.97 SF CARPET	SPECIMEN # SPEC DATE RECPT DATE PRINT DATE	R3005 3/7/08 3/18/08 3/27/08 3:08 pm
CLIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	REFERRING BILLING CODE CLIENT CODE DIAGNOSIS	NOT PROVIDED 14CC 1		

PHONE 727-774-7949 FAX 727-774-7993 ACCT#

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	4438	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	1574	ng/G	MODERATE
Cat (Fel d 1) Allergen (Dust)	2206	ng/G	LOW
Cockroach (Bla g 1) Allergen (Dust)	<0.4	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

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MAR-27-2008 15:17 JHU DACI LAB 410 550 2030 P.001/001

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ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	DATE OF BIRTH GENDER F AGE STATUS 5.132M2		SPECIMEN # R3419 SPEC DATE 4/14/08 RECPT DATE 4/23/08 PRINT DATE 5/5/08 9:50 am	
CLIENT NAME AND ADDRESS	PATIENT	REFERRING BILLING CODE 14CC CLIENT CODE 1 DIAGNOSIS NOT PROVIDED			
PHONE	FAX	ACCT#			

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	86704	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	6253	ng/G	HIGH
Cat (Fel d 1) Allergen (Dust)	4135	ng/G	LOW
Cockroach (Bla g 1) Allergen (Dust)	<0.4	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

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MAY-05-2008 11:47 JHU DACI LAB 410 550 2030 P.001/001

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ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREE BREEZE DR. NEW PORT RICHEY FL 34654 727-774-7949	DATE OF BIRTH GENDER F AGE STATUS 5.132M2		SPECIMEN # R3735 SPEC DATE 5/5/08 RECPT DATE 5/16/08 PRINT DATE 6/9/08 10:24 am	
CLIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	REFERRING BILLING CODE 14CC CLIENT CODE 1 DIAGNOSIS	NOT PROVIDED		
PHONE 727-774-7949	FAX 727-774-7993	ACCT#			

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	30656	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	2718	ng/G	HIGH
Cat (Fel d 1) Allergen (Dust)	3856	ng/G	LOW
Cockroach (Bla g 1) Allergen (Dust)	<0.4	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

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JUN-09-2008 12:26 JHU DACI LAB 410 550 2030 P.001/001

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ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR. NEWPORT RICHEY FL 34654	DATE OF BIRTH GENDER AGE STATUS	F 5.13M2	SPECIMEN # SPEC DATE RECPT DATE PRINT DATE	R4011 6/3/08 6/9/08 6/13/08 10:47 am

CLIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	REFERRING BILLING CODE CLIENT CODE DIAGNOSIS	NOT PROVIDED 14CC 1

PHONE 727-774-7949 FAX 727-774-7993 ACCT#

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	15232	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	2735	ng/G	HIGH
Cat (Fel d 1) Allergen (Dust)	10558	ng/G	MODERATE
Cockroach (Bla g 1) Allergen (Dust)	<0.4	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

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AUG-28-2008 11:33 JHU DACI LAB 410 550 2030 P.001/001

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ENVIRONMENTAL DUST ALLERGENS

PATIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR. NEWPORT RICHEY FL 34654 727-774-7949	DATE OF BIRTH GENDER U AGE STATUS CLASSROOM		SPECIMEN # R4446 SPEC DATE 7/7/08 RECPT DATE 7/11/08 PRINT DATE 7/21/08 2:54 pm	
CLIENT NAME AND ADDRESS	FOWLER, JENNIFER 11815 TREEBREEZE DR NEW PT RICHEY FL 34654	REFERRING BILLING CODE CLIENT CODE DIAGNOSIS	NOT PROVIDED 14CC 1		
PHONE 727-774-7949	FAX 727-774-7993	ACCT#			

ALLERGEN (CPT 86849)	RESULT	UNITS	INTERPRETATION
Dust Mite (Der p 1) Allergen (Dust)	26186	ng/G	HIGH
Dust Mite (Der f 1) Allergen (Dust)	6720	ng/G	HIGH
Cat (Fel d 1) Allergen (Dust)	5797	ng/G	LOW
Cockroach (Bla g 1) Allergen (Dust)	<0.4	U/G	NEGATIVE

QNS = Quantity Insufficient

DUST MITES (Der p 1, Der f 1)	CAT (Fel d 1)	MOLD (Colony Count)	Risk ranges for these allergens have not been established:
<400 ng/G = Low 400 - 2,000 ng/G = Moderate >2000 ng/G = High	<8000 ng/G = Low 8,000 - 80,000 ng/G = Moderate >80,000 ng/G = High	<10,000 = Low 10,000 - 25,000 = Moderate >25,000 = High	DOG (Can f 1) COCKROACH (Bla g 1, Bla g 2) * MOUSE (Mus m 1), RAT (Rat n 1)* * Any positive result is considered significant.

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