# INDOOR ALLERGENS IN SCHOOLS: A COMPARISON BETWEEN SWEDEN AND CHINA

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## ABSTRACT
There are few studies comparing indoor allergens schools from different parts of the world. We measured allergens levels in 23 classrooms in Uppsala, Sweden, and 30 classrooms in Shanghai, China. Dust was collected by vacuum cleaning, and analysed for allergens from cat (Fel d 1), dog (Can f 1), horse (Equ cx), house dust mites (Der p 1, Der f 1), cockroach (Bla g 1), and mould (*Alternaria alternata*, Alt a 1) by ELISA. All Swedish classrooms had cat allergen, and most had dog and horse allergens. In Shanghai, 13% had cat allergen, and 7% dog allergen, while none had horse allergen. House dust mite, cockroach and *Alternaria* allergen were not detected in any sample from either country. Pet allergy and current asthma were less common in Shanghai. Causative factors could be less furry pets at home, wearing of school uniforms resulting in reduced influx of allergens, and less fittings and textiles.

## INDEX TERMS
Asthma, Allergy, Pet allergen, Horse allergen, School environment

## INTRODUCTION
The school environment has been paid an increased attention. Poor ventilation, lack of maintenance and unsatisfactory cleaning are thought to be common in many countries. However, there are few studies available comparing the school environment in different parts of the world. In Uppsala, Sweden, we have undertaken a number of studies of health effects in the school environment, among school personnel and pupils (Elfman et al., 2000, Smedje et al., 1997a; Smedje and Norbäck, 2001a,b). The presence of allergens in schools, particularly cat allergen, may influence/increase allergy and asthma (Almqvist et al., 2001, Munir et al., 1993; Smedje and Norbäck, 2001b). The aim of this study was to compare allergens levels in settled dust in classrooms in Uppsala and Shanghai, and to compare the prevalence of allergies and asthma among pupils.

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## MATERIAL AND METHODS

### Study design

In Sweden, the compulsory school comprise 9 forms (6 in the primary school and 3 in the junior high school). The vast majority of Swedish children attend public schools. In the municipality of Uppsala, a city in central Sweden with approximately 160 000 inhabitants, there are about 60 compulsory schools. The main aim of this study was to study a cohort of all children aged 6-13 within one geographical area (Knivsta), and to follow their development of allergies and asthma. All 8 primary schools (1-6$^{th}$ form), belonging to Knivsta, a part of the rural outskirts of the municipality, were included in the study. In year 2000, the headmasters were contacted, and all schools agreed to participate.

In China, there is a 6 year primary school, followed by a 3 year junior high school. Shanghai is the largest city in China with approximately 16 million inhabitants, situated at the coast in mid-China. The main aim of the project was to form a cohort of pupils, and follow their development of allergies and asthma during a 2 year period, while the pupils remained in the same schools. The school administration in two people's commune in Shanghai were contacted. One commune was situated in central Shanghai, with totally 30 primary schools. The other was in the western part of Shanghai, with totally 24 primary schools. The western commune was in a more wealthy area than the central commune. Five schools were randomly selected from each of the two communes. The headmasters were contacted, and all schools agreed to participate.

### Questionnaire study

In Uppsala, Knivsta, all pupils in the eight primary schools (N=1482) received a postal questionnaire in May 2000, requesting information about allergies, asthma, and respiratory symptoms. Since the pupils were young (<14 years), the questionnaire were answered by the parents in co-operation with the children. Totally, 1014 participated (68%).The questionnaire included the core questionnaire from The International Study Group of Asthma and Allergies in Chindhood (ISAAC) (ISAAC, 1998). It also contained additional questions from the previous Swedish school study (Smedje et al., 2001a,b), and some questions from the European Community Respiratory Health Survey (ECRHS) (Janson et al., 2001). In Shanghai, three classes from the 1$^{st}$ form were selected from each of the 10 schools. All pupils in the 30 classes (N=1435) received a questionnaire in November 2000, containing similar questions as in the Swedish study. Totally, 1414 participated (99%). The questionnaires were distributed by the teachers, and was answered the same day. The questions were translated from Swedish or English to Chinese by one person, and back-translated by another person. Detailed data from the questionnaire studies will be presented elsewhere. This presentation deals only with self- reported allergy, cumulative prevalence of asthma, and current asthma. Current asthma was defined as having either current asthma medication, or having had an asthma attack during the last 12 months.

### Hygienic measurements

Measurements of allergens, and other pollutants were performed in November-December 2000 in both countries. Both in Sweden and Shanghai, each class had a home-classroom where the pupils spent most of their time. In Sweden, since all classes were included in the study, we selected 3 home-classrooms from each school. In doing so different parts of the school buildings and different grades were represented. The only exception was one school which had only two classrooms. In Shanghai, measurements were performed in the home-classroom of the selected classes. Totally, 23 classrooms in Uppsala and 30 in Shanghai were investigated. The buildings were inspected and details on construction, materials, amount of

open shelves and fabrics, and signs of building dampness were noted. Measurements of temperature, relative humidity, carbon dioxide ($CO_2$), and other air pollutants were performed (to be presented in a separate publication). Settled dust were collected with a vacuum cleaner fitted with a special dust collector (ALK Abello, Copenhagen, Denmark) equipped with a Millipore filter (pore size 6 μm). Vacuum cleaning was performed over desks, chairs, shelves and floor during totally 4 minutes per sample, 2 minutes on the floor and 2 minutes on other surfaces. Each classroom was divided into two halves, and two samples per classroom were taken.The samples from the part next to the corridor were analysed for allergens. The filters were sealed in plastic bags and stored at –20 °C until extraction.

**Analysis of allergen concentration**

The total amount of dust was weighed in each sample. Samples of settled dust (100 mg) were extracted in 2 ml of phosphate buffered saline containing 0.05% Tween 20 (1/20 W/v) by rotating mixing over night at +4-8 °C. Samples were then centrifuged at 4 500 rpm for 10 min followed by another centrifugation of the supernatant at 10 000 rpm for 10 minutes. The final supernatant was transferred to Micro tubes (Sarstedt, Germany) and stored frozen at –20 °C until analysed for the content of allergen. Allergen levels were determined using two-site sandwich ELISA for cat (Fel d 1), dog (Can f 1), houe dust mite (Der p 1 and Der f 1), cockroach (Bla g 1) and mould allergen (*Alternaria alternaria* ALT a 1) (INDOOR Biotechologies Ltd., USA), and horse allergen (Equ ex) (MABTECH, Stockholm, Sweden) (Emenius et al., 2001), using monoclonal antibodies. The assays were basically performed according to the protocols provided by the manufacturer except in the dog assay the horseradish peroxidase labelled goat anti-rabbit IgE was from DAKOPATTS, Denmark. The dust samples were diluted in phosphate buffered saline containing 0.05% Tween and 1% bovine serum albumin (BSA) in serial dilution starting with 1/5 and assayed in duplicates. Allergens concentrations were expressed as ng/g dust, except for horse allergen concentrations which were expressed as Units/g dust, where 1 Unit is equal to 1 ng protein of a horsehair and dander extract used as standard (Allergon, Välinge, Sweden and NIBSC, Hertfordshire, UK). Protein determination was performed on the standard with the micro BCA method (Pierce, Rockford, USA) using BSA as standard.

**RESULTS**

In Uppsala, Knivsta the mean age of the buildings was 46 years (range 1- 103 years). Mechanical ventilation, with supply and exhaust air systems, without air humidification, were found in 7 schools (88%), while one had natural ventilation only. Five of the buildings (63%) were made of wood, while the other 3 were of concrete constructions. All classrooms had hard floor coverings made either from poly-vinyl-chloride (PVC) or linoleum. None of the classrooms had visible signs of dampness, mould growth or mouldy odour. Most of the classrooms had daily floor cleaning, and the desks were wiped mostly by the students 1-5 times a week. In Shanghai, the mean age of the school buildings was 33 years (range 3-65 y). None had a mechanical ventilation system or air-conditioning, nor any heating system. All were concrete buildings with openable windows. One classroom (3%) had a wooden floor while the other classrooms (97%) did not have any floor covering but consisted of unpainted concrete. Only one school in central Shanghai had signs of building dampness (water leakage and visible mould growth). All schools had daily cleaning of floors and desks performed by the pupils. In one school, there were small table-cloths on the desks. Otherwise, none of the classrooms contained any textiles, fleecy materials or any book shelves.

All Uppsala/Knivsta classrooms had cat allergen and most had dog and horse allergens. The median concentration per gram dust was 1200 ng/g Fel d 1 (range 290-4700 ng/g), 1525 ng/g

Can f 1 (<200-3650), and 2050 U/g Equ cx (<120-8500 U/g). In Shanghai, cat allergen was found in only 4 (13 %), and dog allergen in 2 (7 %) of the classroom samples. The range was <200-440 ng/g Fel d 1, and <200-750 ng/g Can f 1. Horse allergen was not detected in any sample in Shanghai. Mite (Der p 1, Der f 1), cockroach (Bla g 1) and mould allergen (Alt a 1) were not detected (<200 ng/g) in any sample from either country. Total amount of dust per sample was almost twice as high in Uppsala/Knivsta (median 787 mg), compared to Shanghai (median 412 mg). Mean allergen levels per school is given in table 1.

**Table 1** Mean allergen concentration in settled dust in eight Swedish and ten Chinese schools.

| | Year of construction | Total dust (mg/sample) | Fel d 1 (ng/g) | Can fl (ng/g) | Equ cx (units/g) |
|---|---|---|---|---|---|
| **Uppsala, Knivsta** | | | | | |
| School 1 | 1955 | 1040 | 1230 | 1880 | 4070 |
| School 2 | 1983 | 1190 | 1240 | 430 | 2930 |
| School 3 | 1897 | 450 | 1670 | 1630 | 3270 |
| School 4 | 1910 | 790 | 860 | 820 | 940 |
| School 5 | 1959+1999 | 1640 | 2010 | 1670 | 2330 |
| School 6 | 1999 | 470 | 860 | 1830 | 1530 |
| School 7 | 1964 | 1240 | 1330 | 1140 | 940 |
| School 8 | 1962* | 530 | 3300 | 3380 | 2020 |
| **Central Shanghai** | | | | | |
| School 1 | 1964 | 1310 | ND | ND | ND |
| School 2 | 1964 | 280 | ND | ND | ND |
| School 3 | 1935+1974 | 520 | 200 | ND | ND |
| School 4 | 1958 | 1060 | ND | ND | ND |
| School 5 | 1957 | 460 | 350 | 240 | ND |
| **Western Shanghai** | | | | | |
| School 6 | 1963 | 670 | ND | ND | ND |
| School 7 | 1988 | 680 | ND | ND | ND |
| School 8 | 1963+1997 | 260 | ND | 380 | ND |
| School 9 | 1978 | 280 | ND | ND | ND |
| School 10 | 1964 | 710 | 220 | ND | ND |

*Reconstructed in year 1999 ND=not detected in any dust sample from the school

**Table 2** Age, gender, asthma and allergy among pupils in Uppsala, and Shanghai

| | Pupils in Knivsta | Pupils in Shanghai | 2-tailed p-value |
|---|---|---|---|
| Mean age (years) | 9.5 | 13.0 | <0.001 |
| Girls (%) | 51 | 50 | NS |
| Cat allergy (%) | 6.7 | 0.8 | <0.001 |
| Dog allergy (%) | 4.7 | 1.1 | <0.001 |
| Hay fever (%) | 7.1 | 4.5 | <0.01 |
| Ever had asthma | 7.4 | 10.5 | <0.01 |
| Current asthma | 5.4 | 3.1 | <0.01 |

The Shanghai pupils were, on average, 3.5 years older. Despite being older, cat and dog allergy and hay fever were less common among Shanghai pupils. The cumulative incidence of

asthma ("ever had asthma") was higher in Shanghai, while current asthma (last 12 months) was less prevalent, table 2.

## DISCUSSION

The study compares allergen levels measured in two school studies, during the same season. The sampling method was the same in both studies, the allergen analysis was performed at the same laboratory and with the same methodology. Thus the results should be comparable with respect to sampling and immunological analysis.Comparability of questionnaire data is, however, somewhat limited. The questions were identical, but information was collected in different seasons, with different methods for distribution, and the age of the pupils were different. Older pupils would, however, been expected to have a higher prevalence of asthma and allergies. Despite this, the Shanghai pupils reported less allergies, particularly pet allergy.

The demographic structure, and the degree of urbanisation was different. Knivsta is a rural part of the municipality of Uppsala, with a lot of the children keeping pets, and riding horses. Keeping pets in Sweden is, however, not restricted to the countryside: a majority of Swedish children keep furry pets in the home. Data from the county of Uppsala have shown that 56% of compulsory school children had furry pets at home, of which 34% had a cat, and 25% had a dog (Smedje et al., 2001b). Shanghai is the most wealthy part of China and has a rapid economical development. In urban China, pets are becoming more popular but are still rare as compared to Sweden. Ownership of dogs in the city of Shanghai is discouraged by the local authorities and dog owners must pay a relatively high annual fee. We have no information on the number of cats and dogs, or allergen levels in the homes in Shanghai. In a study of residential homes in Hong Kong, it was shown that cat allergen (Fel d 1), house dust mite allergen (Der p1), and cockroach allergen (Bla g 2) was found in most homes. The geometric mean level of Der p 1 in mattress dust was relatively high (8 800 ng/g dust), while levels of cat (300 ng/g dust Fel d 1), and cockroach (100 ng/mg Bla g 2) was low (Leung et al., 1998).

The high levels of cat and dog allergens in the classrooms in Knivsta, Uppsala is in agreement with data from previous studies in Sweden, but results are not directly comparative due to methodological differences (Munir et al., 1993; Smedje et al., 1997; Perzanowski et al., 1999). The presence of high levels of horse allergen in schools is a new but not unexpected finding. The absence of house dust mite allergens in the Swedish schools is in agreement with previous findings (Smedje et al., 1997). There is sparse information on allergen levels in Asian schools, including China. The main source of the allergen contamination in the classroom is influx of allergens from the cloths of the children (Smedje and Norbäck, 2001b, Perzanowski et al., 1999, Patchell et al., 1997). Textile materials and fitting may serve as a reservoir of allergens (Smedje and Norbäck, 2001a). Allergen levels in schools may be reduced if there is less furry pets at homes, and if there is less textiles and fleecy material in the classrooms. These conditions were better in Shanghai.

## CONCLUSION AND IMPLICATIONS

Allergen levels and pet allergy were higher in Uppsala than in Shanghai schools. Causative factors could be less furry pets at home, absence of textile fittings and book shelves, and daily desk cleaning in Shanghai.

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