

Environmental exposure to endotoxin in rural and urban Egyptian school children and its relation to asthma and atopy

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Aim. The environmental exposure of farm children to micro-organisms in dust has been related to a reduced prevalence of asthma and atopy. The aim of this study was to evaluate the relation between settled dust endotoxin and development of asthma and/or atopy in rural and urban school children.

Methods. A comparative study was conducted on 40 rural and 40 urban school children (6-12 years). Parental self-reported allergic symptoms questionnaires were distributed. Forced expiratory volume in the first second (FEV1), forced vital capacity (FVC) were measured using MIR spirometry and FEV1% was calculated. Skin prick testing with the most common aeroallergens was performed. *Limulus amoebocyte lysate* endotoxin content was measured in settled dust samples.

Results. The rural group which has higher exposure to farm animals and feeding on farm milk has less allergic symptoms. Rural students showed highly significant FEV1, FEV1% and significant FVC versus urban students (110.9±19.7, 103.8±12.2, 105.8±24.3 vs. 92.3±24.2, 98.4±18.9, 92.7±23.2, respectively). Rural school dust contains significantly higher level of endotoxin (2-3 EU/mg) than urban school (0-0.1 EU/mg). Urban residence was associated with increase risk of asthma

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after age and sex adjustments (OR_{adj}), 5.16; 95% [CI], 0.95-28).

Conclusion. Our results support the hygiene theory, i.e., endotoxin exposure could be protective to asthma and atopy in school children.

Key words: Endotoxins - Asthma - Child - Dust.

Epidemiological trends of allergic diseases and asthma in children reveal a global rise in their prevalence over the past 50 years. Their rapid rise, especially in metropolitan locals, suggests that recent changes in modern environments and/or life styles underlie these trends.¹

Long-term and early-life (younger than one year) exposure to stables induces a strong protective effect against development of asthma, hay fever, and atopic sensitization.²

Why farm environment and animals could be protective to allergic symptoms in children? A possible explanation for this evolution is the so-called "hygiene hypothesis". Which was first formulated in 1989 by David Strachan.³ That naturally occurring microbial exposures in early life may

difference is highly significant except for dust exposure, it is not significant. On the other hand the number of urban students who have allergic signs and symptoms like sneezing 35/40 (87.5%), dyspnea 13/40 (32.5%), wheezing 37/40 (92.5%) and urticaria 12/40 (30%) are higher than the rural students (14/40 35%, 3/40 7.5%, 30/40 75% and 3/40 7.5% respectively). This difference is highly significant in the first two and significant in the last two. There is no statistical difference regarding: running nose, eczema, GIT symptoms and eye allergy between both groups (Table II).

Rural students showed better mean pulmonary functions than urban students. It is highly significant in Forced Expiratory

Volume first second (FEV1=110.9±19.7 *vs.* 92.3±24.2) and Forced Expiratory Volume first second percent (FEV1%=103.8±12.2 *vs.* 98.4±18.9) and significant in Forced Vital Capacity (FVC=105.8±24.3 *vs.* 92.7±23.2). NB: FEV1%=FEV1/FVC x 100 (Table III).

We skin prick tested both groups. Urban students were allergic to a wider variety of allergens and the reaction was more severe than in rural ones. The numbers of students who had skin sensitivity were higher in urban than rural schools. The difference in numbers was: highly significant regarding dust, cats' hair, sheep's wool, cows' milk, significant in dogs' hair, pigeons' feathers and not significant in dust mite antigens (Table IV).

TABLE II.—Comparison between history of exposure to farm environment and symptoms of asthma and atopy.

Item		Rural (%)		Urban (%)		P value
Contact to farm animal	P	40	100 %	7	17.5 %	<0.01
	N	0	0 %	33	82.5 %	
Exposure to dust	P	40	100 %	36	90 %	NS
	N	0	0 %	4	10 %	
Feeding on farm milk	P	40	100 %	6	15 %	<0.01
	N	0	0 %	34	85 %	
Sneezing	P	14	35 %	35	87.5 %	<0.01
	N	26	65 %	5	12.5 %	
Running nose	P	29	72.5 %	34	85 %	NS
	N	11	27.5 %	6	15 %	
Wheezing	P	30	75 %	37	92.5 %	=0
	N	10	25 %	3	7.5 %	
Dyspnea	P	3	7.5 %	13	32.5 %	<0.01
	N	37	92.5 %	27	67.5 %	
Urticaria	P	3	7.5 %	12	30 %	<0.01
	N	37	92.5 %	28	70 %	
Eczema	P	0	0 %	2	5 %	NS
	N	40	100 %	38	95 %	
GIT Symptoms	P	2	5 %	7	17.5 %	NS
	N	38	95 %	33	82.5 %	
Eye allergy	P	4	10 %	8	20 %	NS
	N	36	90 %	32	80 %	

Using Chi-square; P≤0.01 highly significant, P≤0.05 significant, P=0.05 borderline significant; NS: not significant (P>0.05); P: positive; N: negative.

TABLE III.—Comparison between pulmonary function tests in rural and urban school students.

Test	Rural Mean±SD (Range)	Urban Mean±SD (Range)	P Value
FEV1	110.9±19.7 (67-157)	92.3±24.2 (49-174)	<0.01
FVC	105.8±24.3 (60-163)	92.7±23.2 (45-149)	<0.05
FEV1 %	103.8±12.2 (58-112)	98.4±18.9 (51-113)	<0.01

Using Student's t test; P<0.01 highly significant; P<0.05 significant; FEV1% FEV1/FVC x 100.

TABLE IV.—Comparison between number of positive and negative skin test in rural and urban school students.

Tested item		Rural (%)		Urban (%)		P value
Dust	-ve	36	90 %	22	55 %	<0.01
	+1	3	7.5 %	13	32.5 %	
	+2	1	2.5 %	5	12.5 %	
Dust mite	-ve	28	70 %	20	50 %	NS
	+1	10	25 %	15	37.5 %	
	+2	2	5 %	5	12.5 %	
Dog's hair	-ve	39	97.5 %	30	75 %	<0.05
	+1	1	2.5 %	8	20 %	
	+2			2	5 %	
Cat's hair	-ve	39	97.5 %	25	62.5 %	<0.01
	+1	1	2.5 %	12	30 %	
	+2			3	7.5 %	
Pigeon's feather	-ve	37	92.5 %	29	72.5 %	<0.05
	+1	3	7.5 %	9	22.5 %	
	+2			2	5 %	
Sheep's wool	-ve	36	90 %	21	52.5 %	<0.01
	+1	4	10 %	13	32.5 %	
	+2			6	15 %	
Cow's milk	-ve	38	95 %	28	70 %	<0.01
	+1	2	5 %	10	25 %	
	+2			2	5 %	

Using Chi-square; $P < 0.01$ highly significant; $P < 0.05$ significant; -ve: negative; NS: not significant ($P > 0.05$).

TABLE V.—Assessment of risk factors for asthma after age and sex adjustment by logistic regression.

Risk factor	P	OR	CI
Age	> 0.05	0.802	0.47-1.3
Sex	> 0.05	0.525	
Residence	< 0.05	5.163	0.95-28

OR, odds ratio; CI, confidence interval; OR >2 risk.

The dust of the rural school contained significantly higher level of gram negative bacterial endotoxin (2-3 EU/mg) than the urban school (0-0.1 EU/mg) in different floors at different time. The dust sample of

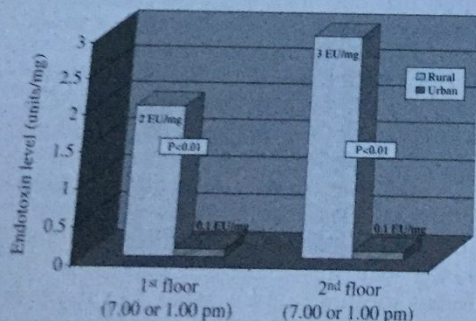


Figure 3.—Comparison between endotoxins levels in settled dust of rural and urban schools at different floors.

the second floor had higher endotoxin content than that of the first floor. There was no difference between samples taken at 7.00 a.m. and 1.00 p.m. in both rural and urban schools. There was no change of the dust content by time change (Figure 3).

Using logistic regression, urban residence was associated with increased risk of asthma after age and sex adjustments (adjusted odds ratio [OR_{adj}], 5.16; 95% confidence interval [CI], 0.95-28) (Table V).

Discussion

Because both rural and urban groups are age, weight and height matched, there is no statistical difference between means of both groups.

Data collected by parental self-reported

allergic symptoms questionnaires showed that the group which had higher exposure to farm animals and feeding on farm milk had less allergic symptoms. That was the rural group. On the contrary, the urban group which shows less exposure had higher allergic symptoms. So apparently the farm environment is more or less protective to the development of allergy.

The difference between the rural and the urban environments is that humans are living very close to farm animals and stables which increases the house dust.¹²

In the present study, the urban students are also exposed to dust, i.e. there is no significant difference between rural and urban students regarding dust exposure. So we attributed the difference between the two groups to the bacterial content of the dust. We suggested that, in the farm environment there is transfer of microorganisms from animal sheds to classrooms in the school by dust.

Group of researchers studied five different dust samples (mattress, cow-shed, swine-shed, chicken-shed and horse-shed). They found that the distinct farm environment dust reflected different PCR bacterial profiles.⁵

In this work, endotoxin - an intrinsic part of the outer membrane of gram negative bacteria - content of the dust proved to be significantly higher in rural dust than in urban dust. So, it seems that endotoxin exposure is protective against asthma and/or atopy.

Endotoxin levels were significantly higher in mattresses and dust from kitchen floors in farming families where children had regular contact to farm animals as compared to control non-farming families in the same rural environment.¹³

Homes without animals revealed a negative correlation between house dust and endotoxin. Central air condition decreases indoor endotoxin.¹²

In one study they found that regular contact to farm animals increases indoor home endotoxin concentrations, and might be protective against atopy.¹⁴ Another study done on the settled dust in classrooms con-

cluded that microbial exposure could be protective against asthma.^{15, 16}

To test whether this endotoxin can be protective from asthma and/or atopy, we compared between the pulmonary function and skin prick test. We noticed that the rural group has better performance of pulmonary function and at the same time expressed less skin reactions than the urban group. The difference is significant to highly significant. Based on these results we believe that endotoxin can be protective from asthma and atopy as well.

This is similar to the conclusion obtained by Reidler *et al.*²

Our explanation is that children raised in modern metropolitan life styles, relatively devoid of this natural microbial burden, may have under-stimulated immune systems in infancy, thereby allowing for the "allergic march" - a pattern of proallergic immune development and disorders that occur in early life.

On the contrary, others believe that higher levels of endotoxin in dust from the child's bed may be associated with increased severity of asthma in children.¹⁸ Others stated that increasing endotoxin exposure was associated with an increased atopy in children.¹⁹

While others believe that early endotoxin exposure may be a protective factor against atopy, it is still a risk factor for asthma in children.²⁰

In the present study, both groups are sensitive to dust mite (the difference is not statistically significant). It should be emphasized that dust mite exposure couldn't be the protective part of the dust against bronchial asthma but may be a risk factor.

Early (at the age of 2 -3 months) exposure to high levels of dust mite allergen is associated with increased risks of asthma and late-onset wheeze.²⁰

A limitation of our study is that we didn't put mite species and its allergen content into our consideration. There are many mite kinds with many allergen content in different locations and seasons. For example there are more than 30 species have been found in house dust in Poland.²¹ However,

our aim was primarily to explore endotoxin content of the dust and its protectiveness to asthma and atopy.

Finally, residence was found to be the most important risk factors for bronchial asthma or atopy after age and sex adjustment in this study.

Conclusions

In conclusion, our results support the "hygiene hypothesis", i.e. endotoxin exposure could be protective of atopy and asthma in school age children.

Riassunto

Obiettivo. L'esposizione ambientale ad un'endotossina in bambini rurali ed urbane dell'Egitto e sua correlazione con l'asma e l'atopia.

Obiettivo. L'esposizione ambientale dei bambini in campagna a micro-organismi nella polvere è stata messa in correlazione con una ridotta prevalenza di asma ed atopia. L'obiettivo di questo studio era di valutare la relazione tra una particolare endotossina della polvere e lo sviluppo di asma e/o atopia in bambini in età scolare dell'area rurale ed urbana.

Metodi. È stato condotto uno studio comparativo su 4 bambini in età scolare in area rurale e in 40 bambini in età scolare in area urbana (6-12 anni). Sono stati distribuiti ai genitori dei questionari relativi ai sintomi allergici. Sono stati misurati il volume espiratorio forzato nel primo secondo (FEV1), la capacità vitale forzata (FVC) mediante lo spirometro MIR spiromark ed è stata calcolata la FEV1%. I prick test cutanei sono stati effettuati con i più comuni aeroallergeni. Il contenuto di endotossina di *Limulus amoebocyte lysate* è stato misurato in determinati campioni di polvere.

Risultati. Il gruppo rurale che era maggiormente esposto ad animali da fattoria e si alimentava con latte di fattoria ha riportato meno sintomi allergici. Gli studenti dell'area rurale hanno riportato valori significativamente superiori di FEV1, FEV1% e di FVC versus gli studenti dell'area urbana (110,9±19,7, 103,8±12,2, 105,8±24,3 vs. 92,3±24,2, 98,4±18,9, 92,7±23,2, rispettivamente). La polvere nelle scuole di campagna contiene livelli significativamente maggiori di endotossina (2-3 EU/mg) rispetto alle scuole di città (0-0,1 EU/mg). La residenza urbana è stata associata ad un aumentato rischio di asma dopo l'aggiustamento per età e sesso (OR_{adj} 5,16; 95% CI, 0,95-28).

Conclusioni. I nostri risultati supportano la teo-

ria dell'igiene, secondo cui l'esposizione all'endotossina sarebbe protettiva nei confronti dell'asma e dell'atopia nei bambini in età scolare.

Parole chiave: Endotossine - Asma - Età pediatrica - Polvere.

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