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Letter

Lactobacillus reuteri DSM 17938 plus vitamin D_3 as ancillary treatment in allergic children with asthma

Asthma is a heterogeneous disease usually characterized by chronic airway inflammation.[1] Allergy and asthma prevalence has significantly increased worldwide during the past decades,[2] and the hygiene hypothesis suggests that reduced exposure to microbial agents could increase the risk of allergic diseases.[3] This theory has been recently revisited with an emphasis on the role of microbiota dysbiosis that promotes impaired immunologic tolerance to allergens.[4] In addition, vitamin D_3 deficiency has been associated with asthma and allergy inception and worsening.[5] Therefore, many attempts of restoring enteric microbiota with probiotic supplementation and correcting vitamin D_3 deficiency have been performed, but with conflicting results.[6]

Lactobacillus reuteri DSM 17938 reduced bronchial inflammation and significantly increased interleukin 10 in children with asthma.[7] The combination of *L reuteri* DSM 17938 plus vitamin D_3 improved allergen immunotherapy efficacy.[8] On the other hand, Kerley and colleagues[9] recently reported that 15-week vitamin D_3 supplementation in Irish children with asthma decreased schooldays missed alone, but there were no other advantageous changes in asthma parameters compared with placebo. Therefore, the aim of this pilot study was to evaluate the effect of a food supplement with *L reuteri* DSM 17938 (10^8 CFU) plus vitamin D_3 (400 IU) on airways inflammation, asthma control, and lung function in children with mild persistent asthma and allergic to house dust mites (HDMs).

This study was a randomized, double-blind, placebo-controlled trial. In total, 32 children (age range, 6–14 years) were recruited. Inclusion criteria were as follows: (1) diagnosis of mild persistent asthma (documented in the medical records) under good control; (2) monoallergy to HDMs; (3) regular treatment with montelukast (5 mg/d); and (4) serum vitamin 25-hydroxyvitamin D level less than 30 ng/mL. Exclusion criteria were as follows: (1) current (or in the past 8 weeks) therapy with inhaled or oral glucocorticosteroids and/or vitamin D supplement; (2) previous or current allergen immunotherapy; (3) normal serum vitamin D level; (4) chronic intestinal disorders that may interfere with the vitamin D_3 and probiotic pathways; and (5) chronic respiratory disorders.

Patients and their parents signed the informed consent form. The research was approved by the Ethical Committee of the Second Naples University (clinicaltrials.gov Identifier: NCT02734446). The study started in September 2015.

At baseline (T0), patients were visited and the parameters were recorded, including serum vitamin D assay. At randomization (with a ratio 1.1 according to a masked code), children were subdivided in 2 groups. Group A was treated with a solution that contained *L reuteri* DSM 17938 (10^8 CFU) plus vitamin D_3 (400 IU) 5 drops per day, and group B was treated with a solution that contained placebo (5 drops per day) for 90 days. Children were revisited at the end of treatment (T1) and after a 30-day follow-up (T2). Montelukast treatment was continued throughout the study. Salbutamol spray could be used as symptomatic treatment.

The primary outcome was bronchial inflammation assessed by fractional exhaled nitric oxide. Secondary outcomes were asthma control measured by questionnaire (Childhood Asthma Control Test [C-ACT]) and lung function evaluated by spirometry, considering bronchial reversibility to bronchodilation testing. In detail, forced expiratory volume in 1 second change was expressed as the percentage of the difference between pre–bronchodilation testing predicted values and post–bronchodilation testing values. These parameters were evaluated at all times, but the C-ACT was evaluated at T0 and T1, and serum vitamin D_3 levels were assayed at T0 and T1. Adherence to intervention therapy and montelukast was considered as percentage of taken doses. The Mann-Whitney test and Wilcoxon test were used (STATISTICA, StatSoft Italia, Padova, Italy).

All children completed the study, except 1 in group A and 2 in group B. The 2 groups were statistically homogeneous and comparable at baseline (Table 1). The adherence to intervention therapy and montelukast therapy was high in all subgroups (ranging from 85% and 100%). The variables, observed at all times, are reported in Table 2.

Serum vitamin D_3 significantly ($P < .001$) increased in group A, whereas there was no change in group B. The intergroup comparison revealed a significant difference at T1 ($P < .001$). Group A had a significant reduction of fractional exhaled nitric oxide between T0 and T1, T1 and T2, and T0 and T2 ($P < .001$ for all times); Group B had no change during the study (Fig 1). The intergroup comparison revealed a significant difference at T1 and T2 ($P < .001$ for both). Group A had a significant reduction in change in forced expiratory volume in 1 second between T0 and T1 and T0 and T2 ($P < .05$ for both) but not between T1 and T2. Group B had no change during the study. The intergroup comparison revealed a significant difference at T1 and T2 ($P = .02$ and $P < .05$, respectively). Group A had no significant change in the mean C-ACT score between T0 and T1. Group B had a significant reduction ($P = .03$) between T0 and T1. The intergroup comparison revealed a significant difference at

Table 1
Demographic Data in Group A and B

Variable	Group A (n = 15)	Group B (n = 17)
White ethnicity	15	17
Age, mean (SD), y	8.7 (1.7)	8.5 (1.9)
Males, No. (%)	6 (40)	7 (41)

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Table 2
Clinical and Functional Data in Groups A and B at T0, T1, and T2[a]

Variable	T0			T1			T2		
	Group A	Group B	*P* Value[b]	Group A	Group B	*P* Value[b]	Group A	Group B	*P* Value[b]
Serum vitamin D, ng/mL	18.3 (5)	17.8 (4)	NS	35.8 (8)	16.9 (7)	<.001			
FeNO, ppm	42.5 (3.9)	44 (3.6)	NS	13 (7)	51 (40)	<.001	12.9 (6.7)	55 (47)	<.001
ΔFEV_1, % of predicted	9.8 (6)	8.7 (7)	NS	6.1 (4)	8.5 (5)	.02	5.6 (3.6)	9.3 (5)	<.05
C-ACT	23 (2)	21 (3.6)	NS	24 (4)	16 (4.6)	<.001			

Abbreviations: C-ACT, Childhood Asthma Control Test; FeNO, fractional exhaled nitric oxide; NS, nonsignificant; T0, baseline; T1, end of treatment; T2, 30-day follow-up.
[a]Data are expressed as mean (SD).
[b]*P* values concern intergroup analysis.

T1 ($P < .001$). Both treatments were well tolerated, and there were no clinically relevant adverse effects in children of either groups.

The present pilot study found that the food supplement that contained *L reuteri* DSM 17938 (10^8 CFU) plus vitamin D_3 (400 IU) was effective in reducing bronchial inflammation. In addition, there was a reduced response to bronchodilation in actively-treated children. These findings were associated with significant increase in serum vitamin D_3 concentration. Moreover, we treated children with low serum vitamin D_3 values, which delineates the indication only to those with vitamin D_3 deficiency. On the contrary, it was previously reported that *L reuteri* ATCC 55730 was not able to prevent asthma onset.[10] Thus, the probiotic's effects might be strain specific, even though appropriate studies should be performed to address this issue. In addition, there is the need to obtain more information supported by evidence-based medicine studies on this topic.[11] Regarding the role of vitamin D_3 in asthma treatment, the evidence on the benefits of vitamin D supplementation for asthma-related outcomes in children is still either limited or inconclusive.[12] However, the current study is, from our knowledge, the first study that evaluated the combination of probiotic plus vitamin D_3 in asthma treatment.

The present study has some limitations: it was conducted in a small sample of patients, immunologic parameters were not assessed, and it was conducted only in patients with mild to persistent and controlled asthma (to consider children with severe or poorly controlled asthma could have a larger impact). In addition, we excluded children taking low doses of inhaled corticosteroids because this therapy could interfere with bronchial inflammation measurement, so further studies should be designed to address these issues. Therefore, the safe use of *L reuteri* DSM 17938 plus vitamin D_3 could open new future frontiers in the treatment of allergic asthma, leading to a reduction of the corticosteroids use.

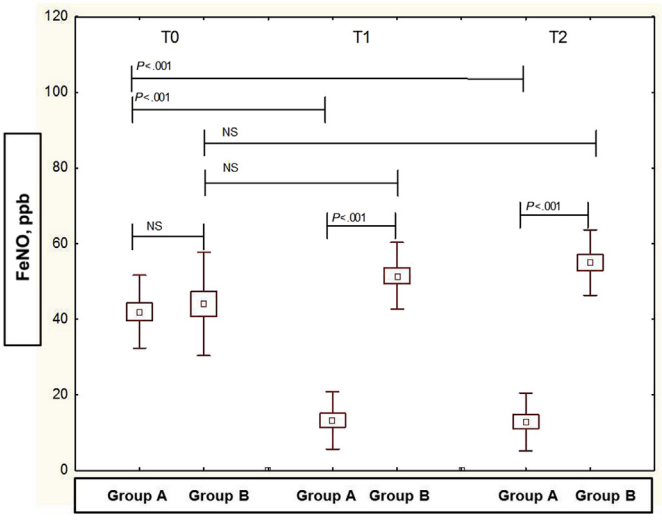

Figure 1. Fractional exhaled nitric oxide (FeNO) values in group A (active treatment) and group B (placebo) at baseline (T0), after treatment (T1), and after follow-up (T2). Data are expressed as mean, SE, and SD. NS indicates nonsignificant.

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