THE EFFECT OF PROBIOTICS ON CHILDREN WHO SUFFERING FROM FOOD ALLERGY

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ABSTRACT: *This study was carried out on 90 children aged 2 to 9 years old followed as:*
1-Conterol group(CG) (45 cases) : were kept under close observation while receiving the standard treatment of allergy and expermental diet every day till improvement of disease outcome.
2-Experimental group(EG) (45 cases): were kept under close observation while receiving the standard treatment of fooallergy and expermental diet including probiotic(Lactobacillus bacteria 10^9 live cells/ml) added to milk (250 ml) every day till improvement of disease outcome.
The main value of Hemoglobin of EG was higher than CG and significantly increased after dietary intervention from 12.113 ± 1.68 to 13.633 ±0.657,while mean CG increased from 12.291 ± 0.667 t0 12.931 ± 0.595 at (p< 0.001).
The main value of red cell count of EG was higher than CG and significantly increased after dietary intervention from 4.451 ± 0.616 t0 5.056 ± 0.222, while mean CG increased from 4.400 ± 0.275 to 4.667 ± 0.268 at (p < 0.05) .
After dietary intervention, The main value of White cell count of EG was higher than CG and significantly increased after dietary intervention from 9.858 ± 1.717 to 10.702 ± 1.506 ,while mean CG increased from 7.989 ±1.013 to 8.389 ± 1.015 at (p<0.01) .
Moreover , the The main value of IgE of EG was higher than CG and significantly increased after dietary intervention from 81.784 ± 18.335 to 50.284 ± 4.315 .while mean CGincreased from 77.402± 5.680 to 64.133 ± 5.186 at (p<0.05) .
conclusion: experimental group much more improved than conterol group.

Key words: *Probiotics, food allergy, children.*

INTRODUCTION

Probiotics *(Bifidobacterium, Lactobcilli)* are definded as live microorganisms with some nutritional and health application for human and animals as food supplements, which beneficially affect the health mainly by improving the intestinal microbial balance . Antibiotics and other related compounds are not included in this defintion (Schreenmeir and de Vere 2001).

Bifidobacterium, Lactobcilli are commonly used as probiotics, either as single species or in mixed culture with other bacteria . Other genera that have been used are *Bifido bacterium* . These are the two most widely researched and documented bacterial genera which applied in human nutrition .

Probiotcs culture has been used for thousands of years in food and alcoholic fermenations,and in the past century has undergone scientific scrutiny for their ability to revent and cure a variety of human diseases. The first clinical trials in the 1930,focused on the effect of probiotics on constipation ,and research has steadily increased since then "Sanders 2000".

Probiotics did not become popular until the public enbraced the idea of functional foods "foods that provide physiological benefits or reduce the risk of chronic diseases,over and above their basic nutritional value"during the last decade of the 20th century ,(Azcarat *et al.,* 2011).

Vanderhoof, *et al.,* (2008), studied probiotics in allergy management and they discovered that: probiotics are effective in the treatment and/or prevention of anumber of conditions, including diarrhoea, irritable bowel syndrome and atopic dermatits,and

the product used should be selected based on the particular indication.

Significant irovement has beennotedininfants recevinglactobacillus GG in their extensively hydrolyzed formula "vanderhoof, *et al.,* (2003)"

Savilahti, *et al.,* (2008), studied pre and robiotics in the revention and treatment of food allergy and they regarded that: modulatio of commensal bacteria of the gut with probiotics has been shown to modulate the immune system and to have an effect on both the prevention and treatment of food allergy. The effects have been highly variable depending on the mode of treatment and the optimal treatment remains unsettled at present.

We can reported that cosistent quality research is the prime driver of growth in the pribiotics industery . In addition to exploring other aspects of these microorganisms , the focus of the science of probiotics is on identifying and isolating specific probiotic strains ,and exploring the mechanisms through which each strain protects the host. Ongoing research shows that probiotics differ in terms of their characteristtics ,and the nature of the associated health benefits, including the finding that some beneficial effects may be strain-specific.

The early status of lif's food milk products with probiotics has been termed as the goal of bacteria in 21th century .

MATERIALS AND METHODS
Materials

This study was carried out on 90 children aged 2 to 9 years old . This study was carried out at Shebin-El-Kom, Minufyia University Hospital, this study was conducted during period from September 2012 to March 2013. Children who admitted to the hospital for treatment and meet the following criteria were enrolled into the study.

Inclution criteria :

1- Age : from 2 to 9 years old .
2- Children with foodallergy .

Methods :
A-Duration of the intervention :

The dietary intervention for this group was continued for 90 following days.

B-Diagnosis of children who suffering from foodallergy :

Food allergy cases were diagnosed by prof. Mohamed Bahbah (Professor and Head Dept of Children, University Hospital. Faculty of Medicin Minufiya University) and biochemical in pediatric department of minufiya university hosbital .

C-Experimental design :

This study was carried out on ninty child. All of them were suffering from food allergy. Half of patients were free living,hospitalized and received their normal diet with drugs without any modification(conterol group).The other patients were free living,hospitalized and receive their normal diet with drugs and received probiotics modification (*Lactobacillus bacteria* 10^9 live cells/ml) added to milk (250 ml) (experimental group) for 90 following days.

D-Administration of selected foods.

Milk with probiotics: we used (*Lactobacillus bacteria* 109 live cells/ml) added to milk 250ml/day. Every patient has given 1 bottle daily in breakfast and dinner.

E-Personal data:

The following data were collected from each child.
1- Socio demographic data: including sex, residency, parent's education, parent's jobs, family income and house state.
2- Health status: including disease definition, etiology of the disease, other diseases prevelance of other diseases in blood such as Animea, Redin eyes, friction in skin and other symptoms of illness.
3- Anthropometric measurments : including body weight and height. Body weight(kg) was measured by weighting subjects wearing indoor clothing, without shoes.

The measured were compared with CDS (2000).

4- Food habits: including meals number, skipped meal, kind of snacks between meals, tea concentration, methods of vegetables cooking,.......etc.

F-Biochemical analysis:

Blood samples were collected from subjects; at baseline and after dietary intervention period and the following parameters were determined.

1-IgE: immunglobin E was determined according to Verbruggen (1975).

2-White blood cell (WBCs) count : WBCs were determined sccording to the method described by Koda, Kimbel, *et al.,* (2001).

3-Red blood cell (RBC) count : red blood cell were estimated according to the method described by Decie and Lewis, (1998).

4-Hemoglobin (Hb) (g/dl) : Hemoglobin were estimated according to the method described by Decie and Lewis, (1998).

5-BCV (%) packed cell volume: BCV were estimated according to the method described by Decie and Lewis, (1998).

6-Mean corpuscular volume (MCV) (ml) : the MCV was calculated by diving the hematocried value by the RBc count according to Lee and Nieman (1996).

7-Mean corpuscular Hemoglobin (MCH) (pg): the MCH was calculated by diving the hematocried value by the RBc count according to Lee and Nieman (1996).

8-Mean corpuscular Hemoglobin concentration (MCHC) (%): the MCHC is the cocentration of hemoglobin (weight per volume) in the average red cell of the sample of blood. MCHC calculated by by diving the hematocried value by the RBc count according to Lee and Nieman (1996).

9-Platelet count.

G-Follow up :

1- Conterol group (45 case) : were kept under close observation while receiving the standard treatment of food allergy and expermental diet every day till improvement of disease outcome.

2- Experimental group (45 case): were kept under close observation while receiving the standard treatment of food allergy and expermental diet including probiotic every day till improvement of disease outcome.

H-Statical analysis:

Statical analysis were performed by using computer program statistical package for social science (SPSS) version 10 , 1998.

Results reported ± SD, also the significant difference between two groups were calculated by paired sample (T-test) for verifying difference between valued before and after inter

RESULTS AND DISCUSSION

This study was carried out on forty-five children who were suffering from food allergy. Their age ranged from (2 to 9) years old. These subjects were selected from Shibin-El–Kom university hospital.

Subjects were classified into two steps:

Step 1: all of patients done medical analysis such as, Hemoglobin, red cells counts, PCV, MCV, MCH, MCHC, Platelet count, White cell count and IGE before they have taken a diet with probiotics.

Step 2: patients fed a diet with probiotic in milk and yoghurt (250ml/daily) for 3 months and then they have done again these medical analysis then we regulated the results.

- From Table (1) most of study subjects were from rural (86.6 %) more than urban (13.3 %)

- These results agreed with (Josef thalhamer., 2000) found that cases was more prevalent among boys than children and 75% of children who suffering from food allergy in their study were from rural areas . These results indicated that, the environmental factors may play an important role in prevalence of children who suffering from food allergy.

Also ,the majority of subjects were classified as low socioeconomics class resprents (22%) . From Table (2) regarding methods of vegetables cooking the majority of studied subjects were prefer stwed was (93.4 %) more than by cooked (60%), while the rate by uncooked were (20%)

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Table (1): Distribution of the study sample in accordance to Economic and social situation depending on the (sex - Religion - Residency)   N=90

Variables		Male		Female		Total	
		No	%	No	%	No	%
Sex		52	57.778	38	42.222	90	100.000
Religion	Muslim	50	55.556	38	42.222	88	97.778
	Christian	2	2.222	0	0.000	2	2.222
Residency	Rural	48	53.333	30	33.333	78	86.667
	Urban	4	4.444	8	8.889	12	13.333

Table (2): Distributionof the study sample according to food habits depending vegetable cooking way for cases of male, female

The way of cooking vegetables	Male		Female		Total	
	No	%	No	%	No	%
Uncooked	5	11.111	4	8.889	9	20.000
Cooked	18	40.000	9	20.000	27	60.000
Stewed	24	53.333	18	40.000	42	93.333
Other	0	0.000	0	0.000	0	0.000

From Table (3) debending on the way cooking meat and poultry and fish cooked in away (stewed) of the sample is (95.6 %). more than who cooked in away (frying) were (77.8 %). while they cooked in away (cooked) way were (44.5 %). Than away with (agrilled) was (40%).

From Table (4) the rate of children who suffering from allergy from milk products was (51.2%) more than who suffering from fruit allergy (37.8%) more than who suffering from allergy from egg were (15.6%), while who suffering from allergy of fish were (11.2 %) .

This results agree with Bruni, *et al.,* (2009), who studied Cow's milk allergic on children and can present sensitization to probiotic and they suggested that: Probiotic use in patients with cow's milk allergy has to be limited to products that do not contain milk. This should be clearly reported in the label. In selected patients, it is advisable to perform a screening SPT with the product to evaluate its potential contamination with milk.

From Table (5) the Distribution of the study sample in accordance with (health history) depending on the type of allergies of cases (male, female) were follows as :that the proportion of cases that were infected with (skin allergies) of the sample is (86.67%) distributed respectively, (53.34%,

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33.34%) of cases (male, female).But the proportion of cases that were infected with (Eye allergies) of the sample is (26.67%) distributed respectively (24.45%, 2.23%) of cases (male, female).but the proportion of cases that were infected with (Nose and chest allergies) of the sample is (37.78%)

distributed respectively (20.00%, 17.78%) of cases (male, female).and the proportion of cases that were infected with (other) of the sample is (2.23%) and distributed respectively (22.23%, 0) of cases (male, female).

Table(3): Distribution of the study sample in accordance to food habits depending on the way of cooking meat, Poultry and fish of the cases of male, female

The way of cooking meat, Poultry and fish	Male		Female		Total	
	No	%	No	%	No	%
Stewed	25	55.556	18	40.000	43	95.556
Cooked	16	35.556	4	8.889	20	44.444
Frying	21	46.667	14	31.111	35	77.778
Grilled	12	26.667	6	13.333	18	40.000
Other	1	2.222	0	0.000	1	2.222

Table (4): Distribution of the study sample according to (date of injury depending on The diet of the allergenic) of cases (male, female)

Reasoned of Allergists	Male		Female		Total	
	No	%	No	%	No	%
Milk	11	24.444	12	26.667	23	51.111
Eggs	4	8.889	3	6.667	7	15.556
Fish	3	6.667	2	4.444	5	11.111
Beans	3	6.667	2	4.444	5	11.111
Fruit	13	28.889	4	8.889	17	37.778
Dust	6	13.333	0	0.000	6	13.333
Chocolate	0	0.000	1	2.222	1	2.222
Ibuprofen	1	2.222	0	0.000	1	2.222

Table (5): Distribution of the study sample in accordance to health history depending on the type of allergies of cases of male, female

Type of allergies	Male		Female		Total	
	No	%	No	%	No	%
Skin allergies	48	53.333	30	33.333	78	86.667
Eye allergies	22	24.444	2	2.222	24	26.667
Nose and chest allergies	18	20.000	16	17.778	34	37.778
Other	2	2.222	0	0.000	2	2.222

From Table (6) as for food habits, the distribution of the study sample in accordance with (dietary habits) depending on the number of daily meals of cases (male, female) for the sample of the study and their percentages were as follows:that the proportion of cases which the number of daily meals (three meals) of the sample is (2.23%) are distributed in a row (2.23%, 0%) of cases (male, female).and the proportion of cases which the number of daily meals (four meals) of the sample is (20.00%) distributed respectively (15.56%, 4.45%) of cases (male, female). but the proportion of cases which the number of daily meals (five meals) of the sample is (35.56%) distributed respectively (15.56%, 20.00%) of cases (male, female).also the proportion of cases which the number of daily meals (six meals) of the sample is (42.23%) distributed respectively (24.45%, 17.78%) of cases (male, female). It is also clear that the research sample has no cases are deleted any meal of food and daily lunches.

Pelto *et al.,* (1997) noticed that probiotic bacteria were shown to modulate phagocytosis differentially in healthy and allergic subjects; in healthy was an immunostimulatory effect, whereas in allergic persons, down –regulation of the inflammatory response was detected and was high in skin than any other places.

Table (6): Distribution of the study sample in accordance to food habits depending on the number of daily mealsOf cases of male, female   N=90

Number of daily meals	Male		Female		Total	
	No	%	No	%	No	%
Three meals	2	2.222	0	0.000	2	2.222
Four meals	14	15.556	4	4.444	18	20.000
Five meals	14	15.556	18	20.000	32	35.556
Six meals	22	24.444	16	17.778	38	42.222
Total	52	57.778	38	42.222	90	100.000

From Table (7) on other hand they were skip meals especially breakfast regarding kind of snacks eaten between meals were (57.7%) and (68.9%) were drink fruit juice . while were eat sweets were (28.9%) .

From Table (8) the main value of Hemoglobin of EG was higher than CG groups and significantly increased after dietary intervention from 12.113 ± 1.68 to 13.633 ±0.657.while CG increased from 12.291 ± 0.667 t0 12.931 ± 0.595 at (p< 0.001).

We can reorted that The main value of red cell count of EG was higher than CG and significantly increased after dietary intervention from 4.451 ± 0.616 t0 5.056 ± 0.222. while CG increased from 4.400 ± 0.275 to 4.667 ± 0.268 at (p < 0.05).

Also, the main value of PCV of EG was higher than CG and significantly increased after dietary intervention from 40.549 ± 1.943 to 43.407 ± 1.350. while CG increased from 38.133 ± 1.408 to 39.489 ± 1.576 at (p <0.05) .

It could be noticed that the main value of MCV of EG was higher than CG and significantly increased after dietary intervention from 84.800 ± 3.634 to 87.933 ±

3.467 . while CG increased from 80.133 ± 1.618 t0 83.311 ± 2.521 at (p < 0.05) .

After dietary intervention the main value of MCH of EG was higher than CG and significantly increased after dietary intervention from 27.267 ± 1.587 to 28.111 ± 0.775 .while CG increased from 26.889 ± 0.935 to 28.111 ± 0.775 at (p < 0.05) .

Moreover , the main value of MCHC of EG was higher than CG and significantly increased after dietary intervention from 31.882 ± 0.984 t0 32.978 ± 0.753 .while CG increased from 31.489 ± 0.506 to 32. 489 ± 0.506 at (p<0.01) .

It could be noticed that the main value of Platelet count of EG was higher than CG and significantly increased after dietary intervention from 278.622 ± 73.288 to 321.067 ± 58.950 .while CG increased from 245.111 ±46.251 to 277.556 ± 42.914 at (p< 0.01) .

Data reported that the main value of White cell count of EG was higher than CG and significantly increased after dietary intervention from 9.858 ± 1.717 to 10.702 ± 1.506 .while CGincreased from 7.989 ±1.013 to 8.389 ± 1.015 at (p<0.01) .

Table (7): Distribution of the study sample in accordance with (food habits) depending on the type of light snacks eaten between mealsof cases (male, female)

Type of light snacks eaten between meals	Male		Female		Total	
	No	%	No	%	No	%
Fruit juice	16	35.556	15	33.333	31	68.889
Soda	6	13.333	1	2.222	7	15.556
Chocolate	7	15.556	4	8.889	11	24.444
Snacks	14	31.111	12	26.667	26	57.778
Sweets	8	17.778	5	11.111	13	28.889
Other	1	2.222	0	0.000	1	2.222

Table (8): Significance differences between blood analyses indicators posttest analysis of the two group of study sample "experimental - control"N=90

Blood analysis indicators	experimental group		control group		Differences mean	"T" Value
	Mean	Standard deviation	Mean	Standard deviation		
hemoglobin	13.633	0.657	12.931	0.595	0.702	5.253***
red cell count	5.056	0.222	4.667	0.268	0.389	7.411***
PCV	43.407	1.350	39.489	1.576	3.918	12.523***
MCV	87.933	3.467	83.311	2.521	4.622	7.153***
MCH	28.889	0.745	28.111	0.775	0.778	4.797***
MCHC	32.978	0.753	32.489	0.506	0.489	3.574***
Platelet count	321.067	58.950	277.556	42.914	43.511	3.958***
White cell count	10.702	1.506	8.389	1.015	2.313	8.450***
IgE	50.284	4.315	64.133	5.186	13.849	13.618***

(***): refer to that the value of "T" tabular bigger than "T" calculated .

The value of "T" tabular when degree of freedom (88) , the standard level (0.05) = 2.000
The value of "T" tabular when degree of freedom (88) , the standard level (0.01) = 2.660
The value of "T" tabular when degree of freedom (88) , the standard level (0.001) = 3.460

Moreover , the main value of IgE of EG was higher than CG and significantly increased after dietary intervention from 81.784 ± 18.335 to 50.284 ± 4.315 .while CG increased from 77.402 ± 5.680 to 64.133 ± 5.186 at ($p < 0.05$)

*It could be noticed that these results were agreement with these reported Baahn,etal., (2008), who found that reflections on current food allergy controversies: specific IgE test application, patch testing, eosinophilic esophagitis, and probiotics.and they found that : First, the diagnostic proficiency of specific IgE (sIgE) measurement. sIgE testing has been a useful screening test; the higher the level, the more likely to be clinically relevant. However, published predictive values varied from one study to another. Levels derived from data on certain groups of patients reflect probabilities that can not be applied with certainty to individual patients. Several factors need to be considered in interpreting the value of sIgE in any particular patient. Definitive decisions require well-designed challenge testing. Second, a few studies suggested the usefulness of including patch testing in food allergy evaluation. It may reveal positive results that may or may not be clinically relevant. At present, its use is not generally accepted because of inconsistency in the reported findings and the lack of standardization of test materials and interpretation. It may possibly have a role in evaluating eosinophilic esophagitis (EE) more than in atopic dermatitis. Third, EE does not seem to be a new disease or is causing a miniepidemic. Its increasing diagnosis is probably because of a greater

awareness and better biopsy assessment. Fourth, the usefulness of probiotics on allergy prevention .

*Also it agreement wih Sandra Sudo *et al.,* (1996).investigated the role of intestinal micro flora in oral tolerance induction to the IgE response in germ free mice Obtained results indicated that probiotics ingestion had clear and pronounced role in elevation of IgE within very short time after intake

CONCLUTION

Supplemention with probiotics added to milk products tends to shorten the recovery time from food allergy among children from (1-9) years old .

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دراسة تأثير البروبيوتيك على الاطفال اللذين يعانون من الحساسية الغذائية

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الملخص العربي

تمت الدراسة على ٩٠ طفل مصابين بالحساسية الغذائية وتم تقسيم الأطفال إلى مجموعتين :

أ- **المجموعة الضابطة**(٤٥ طفل) : وهى التي تتناول الوجبة الغذائية مع العقاقير الطبية المضاد للحساسية الغذائية

ب- **المجموعة التجريبية** (٤٥طفل): وهى التي تتناول الوجبة الغذائية مع العقاقير الطبية المضادة للحساسية الغذائية مع إضافة (بكتريا لاكتوباسلس١٠⁹خلية حية الى ٢٥٠مل من اللبن) يوميا لمدة ثلاث شهر مقسمة في وجبتين القطار والعشاء .وتم أيضا عمل تحليل معملي لعينة دم من قبل وبعد التدخل الغذائي لصورة الدم كاملة وتحليل المناعة (IgE).

وتمت متابعة الأطفال إثناء تناولهم للعلاج الدوائي للحساسية الغذائية بالاضافى إلى الغذاء المختبر . وتم تحليل النتائج إحصائيا لإيجاد الانحراف المعياري و الفروق المعنوية باستخدام برنامج (SPSS).

أدى الإمداد بالوجبات التجريبية إلى انخفاض في تحليل (IgE)(P<0.001) بالنسبة للمجموعتين ولكن كان الانخفاض أكثر في المجموعة التجريبية عن الضابطة (١٨,٣٤±٨١,٧٩ إلى ٤,٣٢±٥٠,٢٩) (٥,٦٩±٧٧,٤١ إلى ٥,١٩±٦٤,١٤)على التوالي .

ومن الملاحظ أيضا إن استخدام اللبن اثر معنويا على مستوى الهيموجلوبين بالنسبة للمجموعتين ولكن كانت الزيادة أكثر في المجموعة التجريبية عن الضابطة (١,٦٩±١٢,١٢ الى ٠,٦٥٧±١٣,٦٤) (١٢,٣±٠,٦٧الى١٢,٩٤±٠,٦٠) على التوالي . وأيضا كرات الدم البيضاء زادت في المجموعة التجريبية عن الضابطة (٩,٨٦± ١,٧٢ إلى ١٠,٧١ ±١,٥٦) (٧,٩٩± ١,٠٢الى٨,٣٩ ± ١,٠٢) على التوالي .

إما بالنسبة لـ MCV,MCH,MCHC,PCV,WC,PLATELEST اثر استخدام اللبن معنويا بعد التدخل الغذائي في المجموعة التجريبية عن الضابطة التي اعتمدت على الوجبة الغذائية والعقاقير الطبية فقط .

وقد أوضحت النتائج ما يلي :

-ان نسبة التحسن فى المجموعة التجريبية زادت بنسبة كبيرة عن نسبة التحسن فى المجموعة الضابطة وذلك لاستخدام الميكروبيات النافعة (البروبيوتيك).