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ORIGINAL ARTICLE

The Effect of Probiotics on Interleukin- 8 and Intestinal Flora in Irritable Bowel Syndrome in Hospital of Zagazig University

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ABSTRACT

Background: *Irritable Bowel Syndrome (IBS) is a common functional Gastrointestinal disorder, characterized by recurrent abdominal pain or discomfort in association with change in stool form or stool frequency. These symptoms have a relapsing and remitting course. The pathophysiology of IBS is not yet well understood. A probiotic is a live organism that when ingested in enough amounts, causes a health benefit to the host. While probiotics have been used on an empiric basis to manage IBS symptoms, several recent researches provide a more logical basis for probiotics use in this field.* ***Objectives:*** *This study was performed to assess the effect of daily use of probiotic on IBS symptoms, pro-inflammatory cytokine (IL-8 level) and intestinal flora.* ***Methodology:*** *This study included total number of 90 patients complaining of IBS symptoms. These patients comprised 22 men and 68 women. They were divided into two groups: Group I included 45 patients complaining of IBS symptoms and treated with probiotics and group II included 45 patients complaining of IBS symptoms and treated with symptomatic treatment. All patients in the study were subjected to blood sampling for measurement of interleukin- 8 and stool sampling for stool culture. Patients of group1 were treated with probotic daily for 4 weeks (Lactobacillus delbruekii and Lactobacillus fermentum 10 billions twice daily).* ***Results:*** *There were statistical significance difference between Group I and Group II in abdominal pain improvement with increase number of improved cases among group1. Regarding improvement time it was statisticaly significant shorter in Group I comparing to group II. There was no statistical significance difference between Group I and Group II in base line IL-8 level. While there was statistical significance differences between them in its level after 1 month. Regarding comparison within the same group there was highly statistical significance differences between base line IL-8 levels and after 1 month results in group I. While there were statistical significance differences between Group I and Group II in IL-8 level after 1 month. There were no statistical significance difference between Group I and Group II regarding base line and after 1 month stool culture results. Regarding comparison within the same group there were highly statistical significance differences between base line stool culture results and after 1 month results in both groups with higher significance difference in group I .* ***Conclusion:*** *Probiotics are an effective treatment option for IBS patients. Daily use of probiotic will improve IBS symptoms, decrease IL-8 level and restore normal intestinal flora.*

INTRODUCTION

Irritable Bowel Syndrome (IBS) is a common functional Gastrointestinal disorder, characterized by recurrent abdominal pain or discomfort in association with change in stool form or stool frequency[1].

These symptoms have a relapsing and remitting course[2], with a prevalence about 20% in the general population, and is more common in women[3]. Patients with IBS are more likely to consume healthcare resources than healthy individuals, with up to 80% of patients consulting their primary care physicians as a result of symptoms[4].

Diagnosing IBS can be challenging due to overlap between gastrointestinal disordersr, which typically present with abdominal pain and changes in gut motility, such as chronic diarrhea and/or constipation[5].

The pathophysiology of IBS is not yet well understood. Recently, growing evidence has suggested an alteration in the immune system cell profile of IBS

patients and a close relationship between the immune and nervous system[6].

Furthermore, O'Mahony et al.[7] and Kajander et al.[8] have studied the relationship between probiotic intake and blood cytokine levels and changes in fecal microbiota.

A probiotic is defined as a live organism that, when ingested in enough amounts, exerts a health benefit to the host. The most commonly used probiotics are lactic acid bacteria and non-pathogenic yeast[9].

Cytokines are primarily synthesized by the immune cells. They regulate differentiation and activation of these immune cells, and are involved in their immunological functions[10].

Functionally, cytokines can be divided into pro-inflammatory (TNF-a, IFN-c, IL-1, IL-2, IL-6, IL-8, IL-12, IL-17, and IL-18) and anti-inflammatory (IL-4, IL-10 and TGF-b) cytokines[11].

While probiotics have been used on an empiric basis in the management of IBS, several recent researchs provide more logical basis for their use in this fieldb. These include the clear recognition that IBS may be induced by bacterial gastroenteritis (postinfectious IBS) and that qualitative changes in the flora, as well as immune dysfunction, may be prevalent in IBS, in general[12].

This study was carried out in an attempt to assess the effect of daily use of probiotic on IBS symptoms, pro-inflammatory cytokine(IL-8 level) and intestinal flora.

METHODOLOGY

This study was conducted in the Outpatient Clinic of Gastroenterology, Internal Medicine Department, Zagazig University Hospitals.

Patients:

It included total number 90 patients complaining of IBS symptoms with the following criteria (according to Rome III criteria): abdominal pain or discomfort and changes in stool frequency or consistency at least 3 days per month in the last 3 months, onset of symptoms at least 6 months prior to diagnosis and must be associated with two or three of the following:

- Improves with defecation.
- Onset of pain coincides with changes in stool frequency.
- Onset of symptoms is accompanied by changes in the form or appearance of stool.

These patients comprised 22 men and 68 women. They were divided into two groups:

- ***Group I :*** It included 45 patients complaining of IBS symptoms and treated with probiotics.
- ***Group II :*** This group included 45 patients complaining of IBS symptoms and treated with symptomatic treatment .

Inclusion criteria:

This study included:

- Patients ≥ 18 years.
- Patients of both sexes.
- Patients suffering from recurrent IBS symptoms according to Rome III criteria.

Exclusion criteria:

- Hepatic patients.
- Diabetic patients.
- Patients having inflammatory bowel diseases proved by colonoscopy.

Methods:

All patients in the study were subjected to the following:

- Detailed history taking.
- Complete general examination.
- Local abdominal examination.
- Review of drug history to exclude any drug that may affect GI motility.
- IL-8 measurement by ELISA using the AviBion human IL-8 kits ((Ani Biotech Oy, Orgenium Laboratories Business, FIN-07120 vantaa FINLAND) .
- Stool sampling for stool culture, stool sample was diluted by serial dilution method (fig:1) then cultured on blood agar aerobically and gentamicin blood agar anaerobically .
- Patients of group I were treated with probotics daily for 4 weeks (Lactobacillus delbruekii and Lactobacillus fermentum 10 billions) twice daily.
- Repeating blood sample and stool culture and re-assessment of IBS symptoms.

Ethical permission:

Written informed consents have been obtained from all patients participating in this study after informing them about the steps of study and capability to withdraw at any time after approval of Ethical Committee in Faculty of Medicine, Zagazig University.

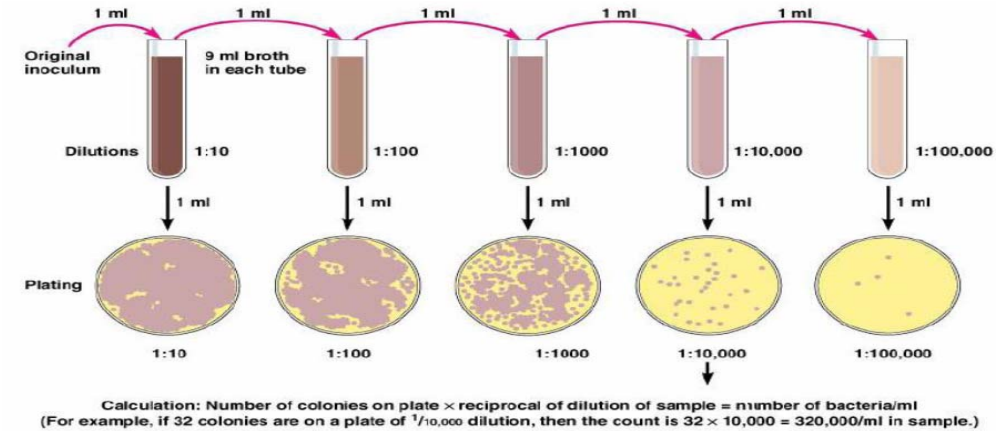

Fig. 1: Serial dilution method.

Statistical analysis:

The collected data were computerized and statistically analyzed using SPSS program (Statistical Package for Social Science) version 18.0. Qualitative data were represented as frequencies and relative percentages. Chi square test was used to calculate difference between qualitative variables. Quantitative data were expressed as mean ± SD (Standard deviation). Median is the middle value of data after arranging values in ascending or descending manner. Independent T test was used to calculate difference between quantitative variables in two groups in normally distributed data. Paired sample T test was used to calculate difference between quantitative variables in the same group pre and post-treatment in normally distributed data. Mann Whitney test was used to calculate difference between quantitative variables in two groups in not normally distributed data. Paired Wilcoxon test was used to calculate difference between quantitative variables in the same group pre and post-treatment in not normally distributed data. Pearson and spearsmann correlation coefficient was used to calculate correlation between quantitative variables. The significance Level for all above mentioned statistical tests done. The threshold of significance is fixed at 5% level (P-value).

***** P value of >0.05 indicates non-significant results.

***** P value of <0.05 indicates significant results.

RESULTS

Table 1 showed that 77.8% of group I were females and 22.2% were males While 73.3% of group II were females and 26.7% were males. There were no statistical significance differences between the two groups regards sex distribution.

Table 2 showed that there were statistical significance difference between groups I and group II in SGOT (p=0.03). But there were no differences between them in fasting blood sugar (FBS), Creatinine and SGPT.

Tables 3 showed that there was statistical significance difference between group I and group II in base line ESR 1st hour (p = 0.04) and ESR 1st and 2nd hour after 1 month (p < 0.001). But there were no differences between them in base line ESR 2nd hour. Regarding comparison within the same group there were highly statistical significance differences between base line and after 1 month readings in both ESR 1st hour and 2nd hour in both groups (p < 0.001).

Tables 4 showed that there was no statistical significance difference between group I and group II in base line IL-8 level. But there were statistical significance differences between them in its level after 1 month (p = 0.001). Regarding comparison within the same group there were highly statistical significance differences between base line and after 1 month in group using Probiotics (p < 0.001).

Table 5 showed that there was no statistical significance difference between group I and group II in base line and after 1 month stool total aerobic bacterial count. Regarding comparison within the same group there were highly statistical significance differences between base line and after 1 month in both groups with higher significance difference in group using probiotics (p = 0.001).

Table (6) shows that there were statistical significance differences between group I and group II in both baseline (p<0.001) and after 1 month results (p<0.001) of stool total anaerobic bacterial count. Also there was statistical significance difference in group I between baseline and after 1 month results(p <0.001) but no difference was found in group II.

Table 7 showed that there was statistical significance difference between group I and group II in abdominal pain improvement (p <0.001) with increase no of improved cases among group I. Regarding improvement time it was statistically significant shorter in group I comparing to group II (p = 0.003).

Figures (2) showed that there were statistical significance +ve correlation between base line and 1 month IL-8 level in group I .

Table 1: Sex distribution of the two studied group:

Variable	*Group I (n=45)*		*Group II (n=45)*		χ^2	*P*
	No	**%**	**No**	**%**		
Sex:						
Female	35	77.8	33	73.3	0.24	0.62
Male	10	22.2	12	26.7		NS

Table 2: Biochemical investigations of the two studied groups:

Variable	*Group I (n=45)*	*Group II (n=45)*	*t*	*p*
FBS (mg/dL)				
Mean ± SD	89.64 ± 8.74	88.09 ± 7.94	0.88	0.38
Range	73 - 109	75 - 107		NS
S.Creatinine (mg/dL)				
Mean ± SD	0.81 ± 0.13	0.82 ± 0.14	0.40	0.69
Range	0.6 – 1.05	0.6 – 1.1		NS
S.GOT (U/L)				
Mean ± SD	15.8 ± 3.35	17.24 ± 2.83	**2.21**	**0.03***
Range	10 - 25	13 - 25		
S.GPT (U/L)				
Mean ± SD	17.73 ± 5.29	17.42 ± 5.27	0.28	0.78
Range	9 - 31	10 - 28		NS

Table 3: ESR of the two studied groups :

Variable	*Group I(n=45)*	*Group II(n=45)*	*t*	*p*
ESR 1st hour (mm/h): (baseline)				
Mean ± SD	18.07 ± 3.77	16.73 ± 2.08	2.07	0.04*
Range	10 – 29	12 - 20		
ESR 1st hour(mm/h): (after 1 month)				
Mean ± SD	11.93 ± 3.19	15.36 ± 2.51	5.18	<0.001**
Range	6 - 22	10 - 21		
Paired t	23.53	4.84		
P	<0.001**	<0.001**		
ESR 2nd hour(mm/h): (baseline)				
Mean ± SD	34.16 ± 5.94	33.96 ± 4.84	0.18	0.86
Range	21 - 44	24 - 41		NS
ESR 2nd hour(mm/h): (after 1 month)				
Mean ± SD	26.73 ± 5.45	32.67 ± 4.58	5.12	<0.001**
Range	17 - 35	21 - 39		
Paired t	17.06	3.85		
P	<0.001**	<0.001**		

Table 4 : Base line IL-8 level and after 1 month in the two studied groups:

Variable	*Group I(n=45)*	*Group II(n=45)*	*MW*	*p*
Base line IL-8 (pg/mL):				
Mean ± SD	6.73 ± 2.77	5.15 ± 3.11		
Median	7.25	5.12	2.19	0.06
Range	0.99 – 11.2	0.59 – 10.73		NS
IL-8 After 1 month				
(pg/mL):*Mean ± SD*	2.19 ± 1.05	4.37 ± 2.83		
Median	2.1	3.88	3.37	0.001**
Range	0.35 – 4.2	0.58 – 10.2		
Paired Wilicoxon	5.51	1.75		
P	<0.001**	0.08 NS		

Table 5: Baseline Stool total aerobic bacterial count and after 1 month in the two studied groups :

Variable	*Group I(n=45)*	*Group II(n=45)*	*MW*	*p*
Base line ST culture : *Mean ± SD* *Median* *Range*	 $361.24x10^5 \pm 296.83x10^5$ $81x10^5$ $62.1x10^5 – 730x10^5$	 $321.6x10^5 \pm 291.9x10^5$ $71\ x10^5$ $71x10^5 – 691x10^5$	 0.57	 0.57 NS
ST culture After 1 month: *Mean ± SD* *Median* *Range*	 $437.7x10^5 \pm 350.99x10^5$ $615\ x10^5$ $63.7x10^5$ - $1000\ x10^5$	 $388.1\ x10^5 \pm 348.01x10^5$ $88\ x10^5$ $60.7x10^5$ - $825\ x10^5$	 0.70	 0.48 NS
Paired Wilicoxon	**3.34**	**2.41**		
P	**0.001****	**0.02***		

Table 6: Baseline Stool total anerobic bacterial count and after 1 month in the two studied groups :

Variable	*Group I(n=45)*	*Group II(n=45)*	*MW*	*p*
Base line ST culture : *Mean ± SD* *Median* *Range*	 $370.96x10^6 \pm 190.6x10^6$ $333\ x10^6$ $77\ x10^6 – 765\ x10^6$	 $226.89x10^6 \pm 178.12x10^6$ $119\ x10^6$ $66x10^6 – 650x10^6$	 **3.89**	 **<0.001****
ST culture After 1 month: *Mean ± SD* *Median* *Range*	 $405.31x10^6 \pm 205.08x10^6$ $375\ x10^6$ $99\ x10^6$ - $950\ x10^5$	 $233.69\ x10^6 \pm 182.86x10^6$ $130\ x10^6$ $65\ x10^6$ - $660\ x10^6$	 **4.22**	 **<0.001****
Paired Wilicoxon	**5.42**	**1.35**		
P	**<0.001****	**0.18**		

Table 7: Abdominal pain after 1 month among the two studied groups:

Variable	*Group I(n=45)*		*Group II(n=45)*		χ^2	*P*
	No	**%**	**No**	**%**		
Abdominal pain: ***Not improved*** ***Relapse & remission*** ***Improved***	 8 0 32	 20 0 80	 0 33 4	 0 89.2 10.8	 62.76	 <0.001**
Improvement time (weeks):					MW	p
Mean ± SD ***Range***	2.03 ± 0.97 1 - 4		3.75 ± 0.5 3 - 4		2.30	0.003**

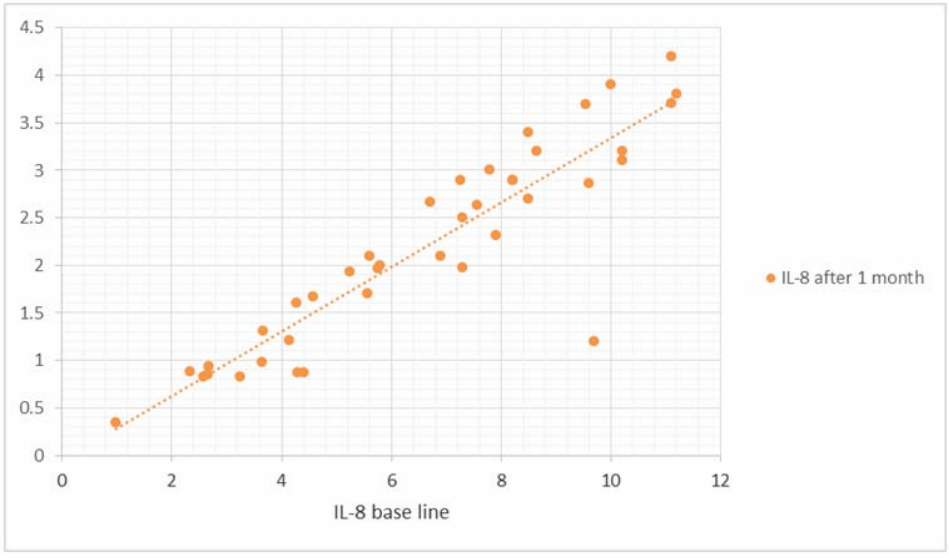

Fig. 2: Correlation between base line IL-8 and IL-8 after 1 month in probiotic group.

DISCUSSION

Irritable bowel syndrome (IBS) is a chronic functional gut disorder that affects about 8%-10% of the population in Western countries, mainly young and middle aged women. Although IBS, as with other functional gut disorders, is a benign disorder with a good long-term prognosis, it has an important impact on a patient's quality of life. IBS also produces a significant economic burden due to both direct health care-related costs and indirect costs due to impaired work productivity. In fact, IBS has been proposed as the second leading cause of absenteeism after the common cold[3].

Treating irritable bowel syndrome (IBS) can be challenging. Among the wide variety of treatment options, probiotics appear to be one of the best options[13]. Ortiz Lucas et al.[6] have suggested an alteration in the immune system cell profile of IBS patients and a close relationship between the immune and nervous systems. Furthermore, Kajander et al.[14] have studied the relationship between probiotic intake and blood cytokine levels and changes in fecal microbiota.

In our study , we assessed the effect of daily use of probiotic on IBS symptoms, pro-inflammatory cytokine(IL-8) level and intestinal flora. Our study included total number of 90 patients complaining of IBS symptoms . These patients were divided into two groups: Group I included 45 patients complaining of IBS symptoms and treated with probiotics and group II included 45 patients complaining of IBS symptoms and treated with symptomatic treatment only.

All patients in the study were subjected to detailed history taking, complete general examination and local abdominal examination. Patients were treated with probotics daily for 4 weeks (Lactobacillus delbruekii and Lactobacillus fermentum 10 billions) twice daily. Interleukin- 8 was measured for all patients.

Our results showed that there were statistical significance difference between group using probiotics (group I) and Group II in abdominal pain improvement with increase number of improved cases among group using probiotic. Regarding improvement time it was statisticaly significant shorter in Group I comparing to group II.

The different results concerning the efficacy of probiotics in improving distension could be explained by the presence or absence of different probiotic species in the mixture, as shown by Moayyedi et al.[15].

These results were in agreement with Moayyedi et al.[16] who have evaluated the role of probiotics in IBS therapy and concluded that probiotics appear to improve overall IBS symptoms.

Also, Ortiz-Lucas et al.[17] have assessed the efficacy of each specific probiotic species in alleviating characteristic IBS symptoms. They concluded that some probiotics are an effective therapeutic option for IBS patients, and the effects on each IBS symptom are likely species-specific.

Lorenzo-Zúñiga et al.[18] determined the dose-related effects of a novel probiotic combination, I.31, on irritable bowel syndrome (IBS)-related quality of life (IBS-QOL). They found that a new combination of three different probiotic bacteria was superior to placebo in improving IBS-related quality of life in patients with IBS and diarrhea. After 6 wk of treatment, the difference was evident in both high and low doses of bacteria, and the increment in quality of life was mainly due to an increment in the mental status domain and was associated with an improvement in gut related anxiety. Hence, this probiotic combination can be useful for the treatment of patients with IBS that impacts their quality of life.

In our study, there were no statistical significance difference between Group I and Group II in base line IL-8 level. But there were statistical significance differences between them in its level after 1 month. Regarding comparison within the same group there were highly statistical significance differences between base line and after 1 month in group using Probiotic.

Our results showed that there were statistical significance differences between Group I and Group II in IL-8 level after 1 month.

Timothy et al.[19] found that the proinflammatory cytokines (IL-6 and IL-8) were elevated in all IBS subgroups (diarrhea predominant, constipated and alternators).

Our results showed that there were no statistical significance difference between Group I and Group II in base line and after 1 month stool culture results. Regarding comparison within the same group there were highly statistical significance differences between base line and after 1 month in both groups with higher significance difference in group using probiotics.

Also, Yoon et al.[20] investigated the efficacy of treatment with multispecies probiotics on irritable bowel syndrome (IBS) symptoms and the alterations of gut microbiota in patients who have taken probiotics. They concluded that multispecies probiotics are effective in IBS patients and induce the alterations in the composition of intestinal microbiota.

Also, Schmulson et al.[21] have found that the gut microbiota in individuals with IBS is different from that in healthy subjects, but a common characteristic present in all the patients has not been established. The incidence and prevalence of post infectous IBS (PI-IBS) varies from 9-10% and 3-17%, respectively,and the latter decreases over time. Bacterial etiology is the most frequent but post-viral and parasitic cases have been

reported. A sub-group of patients has increased enterochromaffin cells, intraepithelial lymphocytes, and mast cells in the intestinal mucosa, but no differences between PI-IBS and non PI-IBS have been determined. Methanogenic microbiota has been associated with IBS with constipation.

Similarly, positive or negative results were found for constipation-IBS patients. No effects on the consistency of stools have been shown in diarrhea-IBS and constipation-IBS subgroups. Although the absence of adverse effects is an additional advantage of probiotic therapy, clinicians should consider the global state of the patient before prescribing them[22].

CONCLUSION

Probiotics are an effective treatment option for IBS patients and that the effects of probiotics on each IBS symptom are likely species-specific. Daily use of probiotic will improve IBS symptoms, decrease IL-8 level (pro-inflammatory cytokine) and restore normal intestinal flora.

Future researchs should focus more specifically on species, combinations, dose, duration, IBS subtypes, and IBS individual symptoms, while employing standardized measurement tools.

Although probiotics are a safe therapy, clinicians should consider other concomitant pathologies when prescribing them to their patients.

Finally, a better design and combination of probiotics will soon be available for the treatment of IBS and other pathologies involving intestinal and general immunity.

REFERENCES

1. Longstreth GF, Thompson WG, Chey WD, Houghton LA, Mearin F and Spiller RC. Functional bowel disorders. Gastroenterology 2006; 130:1480-1491.
2. Ford AC, Talley NJ, Spiegel BM, Foxx-Orenstein AE, Schiller L, Quigley EMM, et al. Effect of fibre, antispasmodics, and peppermint oil in the treatment of irritable bowel syndrome: systematic review and meta-analysis. BMJ 2008; 337: a2313.
3. Lovell RM and Ford AC. Global prevalence of, and risk factors for, irritable bowel syndrome: A meta analysis. Clin Gastroenterol Hepatol 2012; 10: 712-721.
4. Koloski NA, Talley NJ, Huskic SS and Boyce PM. Predictors of conventional and alternative healthcare seeking for irritable bowel syndrome and functional dyspepsia. Aliment Pharmacol Ther 2003; 17: 841-851.
5. Drossman DA, Camilleri M, Mayer EA and Whitehead WE. AGA technical review on irritable bowel syndrome. Gastroenterology 2002; 123: 2108-31.
6. Ortiz Lucas M, SazPeiro P and Sebastian Domingo JJ. Irritable bowel syndrome immune hypothesis: Part two: The role of cytokines. Rev Esp Enferm Dig 2010; 102: 711-7.
7. O'Mahony L, McCarthy J, Kelly P, Hurley G, Luo F, Chen K, et al. Lactobacillus and bifidobacterium in irritable bowel syndrome: Symptom responses and relationship to cytokine profiles. Gastroenterology 2005; 128: 541-51.
8. Kajander K, Myllyluoma E, Rajili-Stojanovi M, Kyrönpalo S, Rasmussen M, Järvenpää S, et al. Clinical trial: Multispecies probiotic supplementation alleviates the symptoms of irritable bowel syndrome and stabilizes intestinal microbiota. Aliment Pharmacol Ther 2008; 27: 48-57.
9. Quigley EMM. Gut microbiota and the role of probiotics in therapy. Current Opinion in Pharmacology 2011; 11: 593-603.
10. Resch K. Cytokines. In: Nijkamp FP, Parnham MJ, editors. Principles of immunopharmacology. Basel: Springer 2005; 46-61.
11. Sivalingam SP, Thumboo J, Vasoo S, Thio ST, Tse C and Fong KY. In vivo pro- and anti-inflammatory cytokines in normal and patients with rheumatoid arthritis. Ann Acad Med Singapore 2007; 36: 96-9.
12. Ivison SM and Steiner TS. Anti-falgellin antibodies in irritable bowel syndrome: Another attack on our commensals. Neurogastro-Enterol Motil 2008; 20: 1081-1085.
13. Enck P, Zimmermann K, Menke G and Klosterhalfen S. Randomized controlled treatment trial of irritable bowel syndrome with a probiotic E. coli preparation (DSM17252) compared to placebo. Z Gastroenterol 2010; 47:209-214.
14. Kajander K, Myllyluoma E, Rajili-Stojanovi M, Kyrönpalo S, Rasmussen M, Järvenpää S, et al. Clinical trial: multispecies probiotic supplementation alleviates the symptoms of irritable bowel syndrome and stabilizes intestinal microbiota. Aliment Pharmacol Ther 2008; 27:48-57.
15. Moayyedi P, Ford AC, Talley NJ, Cremonini F, Foxx-Orenstein AE, Brandt LJ, et al. The efficacy of probiotics in the treatment of irritable bowel syndrome: a systematic review. Gut 2010; 59:325-332.
16. Moayyedi, P; Quigley, EM; Lacy, BE; Lembo AJ, Saito YA, Schiller LR, et al. The effect of fiber supplementation on irritable bowel syndrome: a systematic review and meta-analysis. The American journal of gastroenterology 2014; 109 (9): 1367–74.
17. Ortiz-Lucas M, Tobias A, Saz P and Sebastián JJ. Effect of probiotic species on irritable bowel syndrome symptoms: A bring up to date meta-analysis. Rev Esp Enferm Dig 2013; 105: 19-36.

18. Lorenzo-Zúñiga V, Llop E, Suarez C, Alvarez B, Abreu L, et al. I.31, a new combination of probiotics, improves irritable bowel syndrome-related quality of life. World J Gastroenterol 2014; 20(26): 8709-8716.
19. Timothy GD, Eamonn MM, Salah MM, Scully P, O'Brien S, O'Mahony L, et al. Hypothalamic-pituitary-gut Axis dysregulation in irritable bowel syndrome: plasma cytokines as a potential biomarker? Gastroenterology 2006; 130:304-311.
20. Yoon JS, Sohn W, Lee OY, Lee SP, Lee KN, Jun DW, et al. Effect of multispecies probiotics on irritable bowel syndrome: A randomized, double-blind, placebo-controlled trial. J Gastroenterol Hepatol 2014; 29(1): 52-59.
21. Schmulson M, Bielsa MV, Carmona-Sánchez R Hernández A, López-Colombo A, López Vidal Y, et al. Microbiota, gastrointestinal infections, low-grade inflammation, and antibiotic therapy in irritable bowel syndrome: an evidence-based review. Rev Gastroenterol Mex (in Spanish; Castilian) 2014; 79 (2): 96–134.
22. Spiller R and Garsed K. Postinfectious irritable bowel syndrome". Gastroenterology 2009; 136 (6): 1979–88.