

Relationship between Probiotic Consumption and IL-10 and IL-17 Secreted by PBMCs in Overweight and Obese People

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Epidemiological evidence has linked obesity to pro-inflammatory conditions such as cancer, asthma and autoimmune diseases. T lymphocytes, macrophages, and the proinflammatory cytokines produced by these cells play key roles in the immunological disturbances accompanying obesity.¹ T lymphocytes exhibit pro-inflammatory or anti-inflammatory properties, depending on their subtypes. Obesity is associated with state of chronic low-grade inflammation, as demonstrated by increased levels of acute-phase proteins and pro-inflammatory mediators in serum of obese individuals compared with lean subjects.² In obesity, apart from the immune cells, adipose tissue contributes as a significant source of circulating serum interleukin(IL)-6. In other words adipocytes secrete cytokines such as immune cells. Moreover, IL-6 is essential for the differentiation of naïve CD4 T cells into the T helper (Th)17 subset and IL-17 secretion.³ IL-17, also referred to IL-17A, is a proinflammatory cytokine which has a pivotal role in autoimmune diseases.⁴

Unlike IL-17, IL-10 is a cytokine with pleiotropic effects in immunoregulation and inflammation. IL-10

is capable of inhibiting synthesis of pro-inflammatory cytokines such as Interferon (IFN)- γ , IL-2, IL-3, and Tumor Necrosis Factor (TNF)- α . This cytokine is mainly secreted from the T regulatory (Treg) cells.⁵ obesity is associated with a reduction of IL-10 levels.⁶

Probiotics can influence the immune system by their metabolites, cell wall components and DNA. Products of Probiotic are recognized by host cells with pattern recognition receptors.⁷ In addition, recent experimental studies have demonstrated the preventive effects of some bacterial strains on obesity.⁸ Probiotics are specified as live microorganisms which, when prescribed in adequate amounts, confer a health benefit on the host. The intestinal microbiota plays a fundamental role in maintaining immune homeostasis.⁹

The present randomized doubled-blind controlled clinical trial was performed recruiting 75 individuals with body mass index (BMI) 25-40, who were randomly assigned to the following three groups: Group 1 (n=25) who consumed regular yogurt as part of a low calorie diet [RLCD], group 2 (n=25) who received probiotic yogurt with a LCD [PLCD] and group 3 (n=25) who consumed probiotic yogurt without any low calorie diet [PWLCD] for 8 weeks. Participants in PLCD and PWLCD groups received 200 g/day yogurt containing Lactobacillus acidophilus La5, Bifidobacterium BB12 and lactobacillus casei DN001 108 CFU/gr. To evaluate the effects of BMI on

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concentrations of the cytokines, we conducted the assessment based on three BMI categories (BMI=25-29.9, BMI=30-34.9, BMI=35-39.9). Peripheral blood mononuclear cells were separated from blood and cultured for 72 hrs. IL-10 and IL-17 were measured in culture supernatants by Enzyme-Linked Immunosorbent Assay (ELISA) method.

All numeric variables were tested for normality of distribution by the Kolmogorov-Smirnov test and, if necessary, subjected to logarithmic transformation before applying parametric tests (IL-17).

There were no significant differences among the three groups for means of energy and nutrients intake at baseline. Since there was no change in the diet of probiotic yogurt group during the study period, based on the repeated test at the end of the study, there was no significant difference on the components of 24-hr food recall in this group. However, as expected, energy and nutrient intake were significantly decreased in the groups with LCD, at the end of the study compared to the start of the evaluation.

Table 1 shows that based on BMI, in groups with BMI=25-29.9 and BMI=30-34.9, the difference of IL-10 concentration, was statistically significant between the three intervention groups at the end of study.

In all three BMI groups, people in PLCD and PWLCD intervention groups, had significantly increased concentration of IL-10. Among three BMI groups, increase in concentration of IL-10 was more in group with BMI=30-34.9 ($p < 0.01$).

Regarding IL-17, Table 2 shows that only in people with BMI=30-34.9, the difference of IL-17

concentration was statistically significant between the three intervention groups at the end of study. Reduction of this cytokine in the supernatant of PBMCs in individuals with BMI=25-29.9 was significant in PLCD intervention group. About people with BMI=30-34.9, both PLCD and PWLCD intervention groups, had significantly decreased IL-17 concentrations after the intervention and in individuals with BMI=35.39.9, IL-17 was significantly decreased in PLCD and RLCD groups after the intervention.

Zeuthen, et.al showed that lactobacillus bacteria stimulate the production of IL-10 and thus inhibit Th1 and their inflammatory cytokines¹⁰ which is compatible with our results.

Winer, et.al showed that in obese rats on high fat diet, increase of IL-6 due to obesity caused maturation of Th17 and secretion of IL-17. This could inhibit IL-10 secretion and Treg development, which in turn intensified inflammation in the body.¹¹ A study was conducted by Sumarac-Dumanovic, et. al on obese women showed that high concentration of leptin and Macrophage migration Inhibitory Factor (MIF) could lead to Th17 maturation and production of IL-17.¹² Reduction in leptin level achieved by LCD removed the inhibitory effects on Treg and increased the proliferation of these cells.¹³

In conclusion, consumption of probiotic yogurt appears to be effective in reducing inflammation in overweight and obese people. Our findings can suggest the consumption of probiotics as an immunomodulator may solve some chronic inflammatory complications in obesity particularly if accompanied by weight loss diet.

Table 1. IL-10 concentration (pg/ml) in PBMCs based on Body Mass Index in three study groups.

BMI	Study groups	Week 0	Week 8	p-value*	p-value**
		Mean ± SD	Mean ± SD		
25-29.9 (n=28)	PLCD	988.26±236.68	1041.15±510.13	0.04	0.05
	RLCD	779.37±287.19	743.86±308.15	0.62	
	PWLCD	749.87±186.87	982.16±194.61	<0.001	
30-34.9 (n=24)	PLCD	743.05±268.83	1023±440	0.01	0.012
	RLCD	733.66±308.15	848.89±239.74	0.121	
	PWLCD	750±341.22	1004.42±370.79	0.004	
35-39.9 (n=23)	PLCD	683.78±277.63	939.44±161	0.009	0.611
	RLCD	795.63±275.02	998.64±296.6	0.091	
	PWLCD	904.37±276.7	1098±307.65	<0.005	

* paired sample T test (In each three intervention groups)

**ANCOVA (Between three intervention groups)

PLCD: Probiotic yogurt+ Low Calorie Diet

RLCD: Regular yogurt +Low Calorie Diet

PWLCD: Probiotic yogurt without Low Calorie Diet

Table 2. IL-17 concentration (pg/ml) in PBMCs based on BMI in three study groups.

BMI	Study groups	before	after	<i>p</i> -value*	<i>p</i> -value**
		Mean ± SD	Mean ± SD		
25-29.9 (n=28)	PLCD	1196.92±891.34	480.86±513.81	0.012	0.142
	RLCD	1786.47±990	919.29±546.82	0.111	
	PWLCD	1241.15±1199	862.05±675.5	0.131	
30-34.9 (n=24)	PLCD	1478.06±275.77	980.29±235.17	0.001	0.03
	RLCD	1543.47±396	858.1±216.68	0.312	
	PWLCD	1690.17±516.45	1011.56±621.17	0.020	
35-39.9 (n=23)	PLCD	1835.76±2269	817.8±1022.68	0.041	0.412
	RLCD	1611.34±696.17	1111.72±592.64	0.03	
	PWLCD	2144.67±1734.72	1795.43±2151.74	0.691	

* paired sample T test (In each three intervention groups)

**ANCOVA (Between three intervention groups)

PLCD: Probiotic yogurt+ Low Calorie Diet

RLCD: Regular yogurt +Low Calorie Diet

PWLCD: Probiotic yogurt without Low Calorie Diet

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