The Effectiveness of Probiotic in The Treatment of Inflamed Acne

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ABSTRACT

Background: Prolonged use of oral antibiotic in inflamed acne may impair the beneficial microorganisms. Probiotic can reduce inflammatory mediators by improving the leaky gut, decrease IGF-1 level which is involved in follicular hyperkeratinization and also decrease substance P level effecting on sebocytes and sebum production.

Objectives: To evaluate the effectiveness of probiotic in the treatment of inflamed acne.

Methods: The prospective, experimental study follows the double-blinded, randomized controlled trial. Thirty female patients, aged between 20-40 years old, with mild to moderate acne vulgaris were enrolled. All participants were divided equally into two groups and randomly assigned either TS6 probiotics (100×10^9 CFUs/day) or placebo to be taken for 12 weeks. Inflamed lesion counts, comedones and total lesion counts were evaluated at baseline and during follow-up visits 2, 4, 8, and 12 weeks from the treatment initiation. The sebum score at forehead, both cheeks and chin were assessed by Sebumeter at baseline and 12^{th} week. Post-acne redness (PAR) were estimated by Mexameter MX18 at baseline and 12^{th} week. The statistical analysis within group used pair T-test whereas in between groups using T-test.

Results: Twenty-five patients completed the study. Both of the probiotic group and the placebo group showed significant decrease in inflamed lesion counts at 2^{nd} week ($p < 0.01$ and $p < 0.05$), significant comedones depletion at 8^{th} week ($p < 0.01$ and $p < 0.05$), and significant improvement in total lesion counts at 4^{th} week ($p < 0.01$ and $p < 0.05$) with no difference between two groups. Percentage reduction of inflamed lesion counts in the probiotic group was higher than in the placebo group in every follow-up visits (at 2^{nd} week were 35.3% and 28.6%, at 4^{th} week were 47.1% and 35.7%, at 8^{th} week were 70.6% and 50%, and at 12^{th} week were 76.5% and 64.3%, respectively). Percentage reduction of comedones in the probiotic group was higher than in the placebo group in every follow-up visits (at 2^{nd} week were 8.3% and -2.6%, at 4^{th} week were 14.6% and 10.3%, at 8^{th} week were 33.3% and 23.1%, and at 12^{th} week were 50% and 41%, respectively). Percentage reduction of total lesion counts in the probiotic group was higher than in the placebo group in every follow-up visits (at 2^{nd} week were 15.4% and 3.8%, at 4^{th} week were 23.1% and 17.3%, at 8^{th} week were 43.1% and 28.8%, and at 12^{th} week were 58.5% and 46.2%, respectively). At 12^{th} week, the sebum score of all forehead, both cheeks and chin areas in the probiotic group were reduced, though insignificantly. At 12^{th} week, the erythema index of the PAR in both groups reduced significantly ($p < 0.01$, $p < 0.01$) with no difference between two groups.

Conclusion: The treatment of mild to moderate acne by supplementing probiotics with the standard topical regimens have no significant difference in inflamed lesion counts, comedones and total lesion counts comparing to the topical treatment alone. Percent reduction of inflamed lesion counts, comedones and total lesion counts in the probiotic group were higher than the placebo group in every follow-up visits. However, further studies including the duration of treatment, an appropriate amount or specific microorganisms of probiotic should be conducted.

Keywords: probiotics, acne vulgaris, inflamed acne, TS6 probiotic

INTRODUCTION

Acne vulgaris is a common skin condition that affects most of the population. The pathogenesis of acne is starting from the follicular hyperkeratinization and increased sebum production by sebaceous gland hyperplasia, so that the lesions become closed or open comedones. Moreover bacterium named *Propionibacterium acnes* secret chemotactic and inflammatory mediators leading to inflamed lesions, such as papule, pustules and nodulocystic lesions.

Acne treatment, consisting of topical and systemic medications, is depending on the acne severity. Prolong use of oral antibiotic may destroy beneficial microorganisms in the body causing other conditions. Probiotic supplementation can reduce systemic markers of inflammation, decrease IGF-1 level which is involved in follicular hyperkeratinization and also decrease substance P level effecting on sebocytes and sebum production. In combination with topical acne medications, probiotic may be favorable in acne treatments and are concurrently beneficial to the gut.

LITERATURE REVIEW

Strokes and Pillsbury (1930) provided a theoretical and practical consideration of gastrointestinal mechanism to explain how the skin is influenced by emotional and nervous states. The emotional states such as depression, worry and anxiety alter gastrointestinal tract function and microbiota, which the authors theorised, in turn promotes local and systemic inflammation. Later in 1983, a study involving 80 acne patients showed the presence of lipopolysaccharide (LPS) endotoxins from *Escherichia coli* in the serum of acne patient (Juhlin *et al*, 1983). The study of 13,000 adolescents showed that those with acne were more likely to experience gastrointestinal symptoms including constipation, halitosis, and gastric reflux. More specifically, abdominal bloating was 37% more likely to be associated with acne and other seborrheic diseases (Zhang *et al.*, 2008).

Previous studies enquiring into the potential link between diet and acne vulgaris have shown controversial results. The underlying mechanism of dietary effect in acne vulgaris formation might be the role of insulin-like growth factor-1 (IGF-1) in facilitating cell proliferation involved in acne pathogenesis (Cordian, 2005). Acute hyperinsulinemia due to consumption of high glycemic load diet would cause and increase in IGF-1/insulin-like growth factor binding protein-3 (IGFBP-3) ratio thus enhancing the effects of IGF-1(Cordian *et al*, 2002). Hyperinsulinemia resulting from high glycemic load diet would also increase circulating androgens and decrease sex hormone binding protein, leading to increase sebum synthesis, which are crucial in acne development. Moreover, the loss of *bifidobacteria* by poor dietary choices - high fat, sugar - leads to increase intestinal permeability, encroachment of LPS endotoxins through the intestinal barrier, which in turn leads to low grade inflammation, oxidative stress, insulin resistance and sickness behavior (Cani & Delzenne, 2009)

Substance P induces, both directly and indirectly, inflammation by modulating the release of proinflammatory cytokines and chemokines (also in the skin). This neuropeptide affects the activity of the pilosebaceous unit by stimulating proliferation and differentiation of sebaceous glands, lipid synthesis and induction of neutral endopeptidase expression in sebaceous cells and of E-selectin in perifollicular vessels (Makrantonaki *et al*, 2011). 40 of 80 participants with acne vulgaris presented a higher average substance P than the controls (Rokowska-Waluch *et al*, 2016).

Probiotic, such live microorganisms, have been used for decades to help abdominal symptoms by reducing endotoxins and inflammatory cytokines when administered in adequate amounts (FAO/WHO). *Bifidobacteria* and *Lactobacilli* are lactic-producing bacteria normally found in the gut that may assist in the treatment of inflammatory skin diseases, such as acne (Hacici-Rachinel, 2009). Probiotics can prevent the deleterious effects of TNF-α and interferon-Ɣ on the

intestinal epithelium, a finding that may be helpful for other inflammatory conditions (Resta-Lenert, 2016). The orally administered *Bifidobacterium lactis* can improve fasting insulin levels and glucose turnover rates, even in the presence if a high-fat diet (Burcelin, 2010). Probiotics influence the release of substance P in the intestinal tract and the skin (Bowwe, 2014). The ability of probiotics to help attenuate substance P release in the skin and intestinal tract (Gueniche *et al* , 2010) is also of relevance in the pathway between the nervous system, gut, and skin.

The study in 1961 gave probiotic tablets containing both *L. acidophilus* and *Lactobacillus bulgaricus* to 300 patients for 16 days with an interim two-week washout after the first eight days. The author reported 80% if patients with acne had some degrees of clinical improvement, with the greatest improvement in those with severe inflammatory acne. Unfortunately, the study did not have controls and the rationale for such a dosing regimen is unclear (Siver, 1961). One study tasted an oral supplement composed of lyophilised *L. acidophilus* and *B. bifidum* in 40 patients as an adjuvant to standard antibiotics in the half of the group. The author reported patients treated with a probiotic had improved clinical outcome and reported fewer side effects from the standard antibiotics (Marchetti, 1987).

The purpose of this study was to evaluate the anti-inflammatory effects of the probiotic supplementation on the inflamed acne, and other beneficial skin conditions, such as, sebum production and post-acne redness.

METHODS

The study was approved by the Ethic committee, Faculty of Regenerative Medicine, Dhurakij Pundit University (No.005/60). All patients signed informed-consent forms before inclusion.

From January to April 2018, 20 to 40 years old of healthy female patients enrolled in this prospective, randomized, double-blinded study. Only subjects with mild to moderate acne vulgaris were included (comedones less than 10 and/or inflamed lesions less than 10 were classified as mild acne; comedones 10 to 40 and/or inflamed lesions 10 to 40 and/or cystic lesions 1 to 3 were classified as moderate acne). Patients were excluded from trial if they had history of acute/chronic illness or other facial skin diseases. Pregnant or breastfeeding patients were also excluded from the study. Patients who were concurrently using isotretinoin or oral contraceptive pill within the past 6 months, and oral antibiotic within 4 months were also excluded in the trial.

Subjects were randomly assigned to two groups. The probiotic group received "TS6 probiotic" twice a day (100×10^9 CFUs/day), while the placebo group received identically sachet without probiotic for 12 weeks. TS6 probiotic is the synbiotic containing 6 species (*Lactobacillus acidophilus, Lactobacillus casei, Bifidobacterium longum, Bifidobacterium infantis, Bifidobacterium bifigum, Lactococcus lactis*) and one prebiotic (Oligosaccharide). Both groups were also received topical 2.5% benzoyl peroxide (Benzac®) and 1% clindamycin lotion (Clinda-M®) applied twice daily.

Lesion count (inflamed lesions, comedones, and total lesions) and digital photograph were performed at baseline and during follow-up visits 2, 4, 8, and 12 weeks from the treatment initiation. The sebum score at forehead, both cheeks and chin were assessed by Sebumeter at baseline and 12th week. Post-acne redness (PAR) were estimated by Mexameter MX18 at baseline and 12th week. The statistical analysis within group used pair T-test whereas in between groups using T-test.

FINDINGS

Twenty-five female patients completed the study. Thirteen patients were in the probiotic group (mean age 29.2 ± 4.0) and twelve patients were in the placebo group (mean age 27.8 ± 4.5).

Table 1

Subject characteristics at baseline

n=25	Probiotic (n=13)	Placebo (n=12)	p-value > 0.05
Age (years)	29.2 ± 4.0	27.8 ± 4.5	0.45
Inflamed lesion count	17	14	0.33
Comedone count	48	39	0.50
Total lesion count	65	52	0.39
Mild acne, n (%)	5 (38.5%)	5 (41.7%)	
Moderate acne n, (%)	8 (61.5%)	7 (58.3%)	
Sebum score			
- Forehead	79	60	0.34
- Rt. Cheek	59	41	0.09
- Lt. Cheek	63	40	0.02
- Chin	76	93	0.37
Erythema index	400	393	0.81

Both of the probiotic group and the placebo group showed significant decrease in inflamed lesion counts at 2nd week (p = 0.002 and p = 0.015) with no difference between two groups (p = 0.31). Percentage reduction of inflamed lesion counts in the probiotic group was higher than in the placebo group in every follow-up visits (at 2nd week were 35.3% and 28.6%, at 4th week were 47.1% and 35.7%, at 8th week were 70.6% and 50%, and at 12th week were 76.5% and 64.3%, respectively).

Table 2

Comparison of % reduction of inflamed lesion counts between the probiotic and the placebo.

Inflamed lesion counts / week	Probiotic			Placebo			p-value intergroup
	% Reduction	p-value*	p-value**	% Reduction	p-value*	p-value**	
2	35.3%	**0.002**	**0.002**	28.6%	**0.015**	**0.015**	0.31
4	47.1%	**0.000**	0.071	35.7%	**0.004**	0.240	0.40
8	70.6%	**0.000**	**0.004**	50.0%	**0.003**	0.109	0.18
12	76.5%	**0.000**	0.088	64.3%	**0.000**	0.096	0.19

(*) compared to baseline ; (**) compared to previous visit ; **bold text** showed significant (p-value < 0.05)

Both of the probiotic group and the placebo group showed significant comedones depletion at 8th week (p = 0.004 and p = 0.048) with no difference between two groups (p = 0.40). Comedones in the placebo group increased in first 2 weeks (2.6%) and gradually decreased by week 4 to week 12, whereas comedones in the probiotic groups have reduced in every weeks since the beginning. Percentage reduction of comedones in the probiotic group was higher than in the placebo group in every follow-up visits (at 2nd week were 8.3% and -2.6%, at 4th week were 14.6% and 10.3%, at 8th week were 33.3% and 23.1%, and at 12th week were 50% and 41%, respectively).

Table 3

Comparison of % reduction of comedones between the probiotic and the placebo.

Comedones / week	Probiotic			Placebo			p-value intergroup
	% Reduction	p-value*	p-value**	% Reduction	p-value*	p-value**	
2	8.3%	0.226	0.226	-2.6%	0.299	0.299	0.37
4	14.6%	0.073	0.064	10.3%	0.118	0.058	0.29
8	33.3%	**0.004**	**0.002**	23.1%	**0.048**	**0.036**	0.40
12	50.0%	**0.002**	**0.008**	41.0%	**0.022**	**0.015**	0.47

(*) compared to baseline ; (**) compared to previous visit ; **bold text** showed significant (p-value < 0.05)

Both of the probiotic group and the placebo group showed significant improvement in total lesion counts at 4th week (p = 0.005 and p = 0.024) with no difference between two groups (p = 0.28). Percentage reduction of total lesion counts in the probiotic group was higher than in the placebo group in every follow-up visits (at 2nd week were 15.4% and 3.8%, at 4th week were 23.1% and 17.3%, at 8th week were 43.1% and 28.8%, and at 12th week were 58.5% and 46.2%, respectively).

Table 4

Comparison of % reduction of total lesion counts between the probiotic and the placebo.

Total lesion counts / week	Probiotic			Placebo			p-value intergroup
	% Reduction	p-value*	p-value**	% Reduction	p-value*	p-value**	
2	15.4%	0.051	0.051	3.8%	0.222	0.222	0.34
4	23.1%	**0.005**	**0.008**	17.3%	**0.024**	**0.046**	0.28
8	43.1%	**0.000**	**0.000**	28.8%	**0.008**	**0.016**	0.50
12	58.5%	**0.000**	**0.005**	46.2%	**0.005**	**0.017**	0.46

(*) compared to baseline ; (**) compared to previous visit ; **bold text** showed significant (p-value < 0.05)

At 12th week, sebum score of forehead, both cheeks and chin areas in the probiotic group were reduced, though insignificantly. Probiotic probably showed benefits on sebum production, but have to extend the course of treatment to prove the significant results.

Table 5

Comparison of sebum score of all forehead, both cheeks and chin area at baseline and 12th week between the probiotic and the placebo

Forehead	Baseline	12th week	Δ	p-value btw. group.	p-value intergroup
Probiotic	79	70	-9	0.30	0.33
Placebo	60	79	19	0.02	
Rt. Cheek	Baseline	12th week	Δ	p-value btw. group.	p-value intergroup
Probiotic	59	50	-9	0.20	0.10
Placebo	41	38	-3	0.35	
Lt. Cheek	Baseline	12th week	Δ	p-value btw. group.	p-value intergroup
Probiotic	63	47	-16	0.08	0.44
Placebo	40	45	5	0.22	
Chin	Baseline	12th week	Δ	p-value btw. group.	p-value intergroup
Probiotic	76	66	-10	0.26	0.22
Placebo	93	78	-15	0.10	

At 12th week, the erythema index of the PAR in both groups reduced significantly ($p < 0.01$, $p < 0.01$) with no difference between two groups. Implying that probiotic have no benefit on the PAR compared to the control group.

Table 6

Comparison of % reduction of post acne redness score by Mexameter MX18 between the probiotic and the placebo

Group	Baseline	12th week	% Reduction	p-value btw. group.	p-value intergroup
Probiotic	400	321	19.0%	< 0.01	0.48
Placebo	393	320	18.7%	< 0.01	

Five participants (38.5%) in the probiotic group experienced an increased in the frequency of defecation. No participants complaint about any side effects after probiotic administration.

DISCUSSIONS

Compare to Jung *et la* study, Forty five female patients with mild to moderate acne were divided into 3 groups. Group A received only probiotic supplementation (*L. acidophilus, L. delbrueckii spp., bulgaricus,* and *B. bifidum),* group B received only minocyclin, and group C received both probiotic and minocyclin. All participants applied 5% benzoyl peroxide cream in 12 weeks. The research found that group A (only probiotic) had significant reduction in comedones and total lesions at the 4th week, and had significant reduction in inflamed lesions at 8th week, whereas the TS6 study found that the inflamed lesions had significant reduction at 2nd week, comedones had reduction at 8th week, and total lesions had reduction at 4th week. This reason can be explained by the different in topical acne treatments applying of two studies. The TS6 study used two topical acne medications (2.5% benzoyl peroxide and 1% clindamycin lotion), but the Jung study applied only one medication (5% benzoyl peroxide). Clindamycin lotion had ability to kill *P.acne* and had anti-inflammation effect. When applying 1% clindamycin lotion together with 2.5% benzoyl peroxide in TS6 study, the inflamed lesions had the significant reduction faster than only 5% benzoyl peroxide cream in Jung study. And because of the concentration, 5% benzoyl peroxide using in Jung study had the significant reduction in comedones faster than 2.5% benzoyl peroxide using in TS6 study.

By supplementing TS6 probiotic with the standard topical regimens in mild to moderate acne vulgaris had no significant difference in inflamed lesion counts, comedones and total lesion counts comparing to the topical treatment alone. But percent reduction of inflamed lesion counts, comedones and total lesion counts in the probiotic group were higher than the control group in every follow-up visits. However, further studies including the duration of treatment, an appropriate amount of probiotic per day or the specific microorganisms of probiotic should be conducted.

RECOMMENADATIONS

1. The further trial should extend the duration of the study so that it may show the statistical difference in reduction of inflamed lesions, comedones, and total lesions between the probiotic group and the placebo group.

2. Because of the unknown amount of the probiotic using in acne treatment, the further study should increase amount of probiotic and compare with 3 sachets per day or 4 sachets per day to show the different results.

3. Should also compare to the other species of the probiotic treating in acne vulgaris.

4. The further study should administrate oral probiotic emphasized on the specific group, such as the moderate acne group or the severe acne group.

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