Role of probiotic in modulating immune response and prevention of constipation in critically ill patients; A randomized, double – blind, placebo – controlled trial

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ABSTRACT

Objectives : To assess the efficacy of probiotic in 1)the probiotic effect in enhancing immune responce 2) prevention of constipation in critically ill patient.

Design : A single centre, randomised, double blind, placebo controlled study that was conducted in the Intensive Care Unit, Universitiy Malaya Medical Centre between June 2016 to December 2016. A total of 36 patients who required mechanical ventilation and enteral nutritional feeding for atleast 3 days were randomised into two group : the intenvention group who received Hexbio, a probiotic preparation or the control group who received placebo. The patient were monitored for 5 consecutive days. Blood sample taken on 1,3,5 days of ntervention,min reflo reading with total usage of intravenous insulin, timing for first bowel movement, stool frequency and consistency were recorded using King's stool chart.

Result :

Primary outcome : Serum analysis for IL-6, IL-10 and hemeoxygenase still in progress. Therefore unable to modulate the outcomes.

Secondary outcomes: The intervention group showed shorter time for first bowel opening of 3.2 days compared with 4.6 days in placebo group (p=0.03). It is also showed higher mean frequency 3.75 times perday compared with 0.35 times perday (p=0.09). Beside that, they also presented with lower gastric aspirates with p value 0.0003. Other parameter showed indirect beneficial effect of probiotic in reducing morbidity in critically ill by reduce lenght of ICU stay, reduce number of days required ventilator support and better min blood sugar level.

Conclusion : Probiotics could reduce inflammation in critically ill-patients and might be considered as an adjunctive therapy in the treatment if critically ill-patients as it help in reduce morbidity . Probiotics also have beneficial effect in preventing contipations as statiscally showed significant difference in first bowel opening and reduced mean gastric contents daily. Other that that, probitotic promote antioxidant effect which suqsequently help in controlling blood sugar level in critical ill patients.

Abstrak

Objektif : untuk mengkaji kesan probiotik 1) memperbaiki immunisasi tubuh badan di kalangan pesakit ICU 2) kesan profilaktik terapi probiotik dalam mengurangkan kejadian sembelit dikalangan pesakit kritikal.

Kaedah : Ini adalah sebuah double-blind, placebo controlled study ke atas 36 pesakit kritikal yang dimasukkan ke Unit Rawatan Rapi Pusa tPerubatan Universiti Mlaya antara bulan Jun 2016 hingga Disember 2016. Pesakit telah dibahagikan secara rambang kepada dua kumpulan . Kumpulan pertama menerima Placebo (A) and kumpulan kedua menerima Hexbio yang merupakan probiotik yang mengandungi 6 jenis strain Lactobacillus dan Bifidobakteria, manakala kumpulan kawalan menerima placebo. Pesakit dipantau selama 5 hari. Ujian darah dimabil pada hari pertama, ketiga and kelima probiotik and masa pertama membuang air besar, kekerapan and konsisten direkodkan sebagai skor harian selama 5 hari.

Keputusan :

Perolehan pertama : ujian darah untuk mengkaji kesan probiotik dalam memperbaiki immunisasi masih dalam pemprosesan.

Perolehan kedua : kumpulan yang menerima probiotik menunjukkan masa yang lebih singkat 3.2 hari berbanding kumpulan kawalan 4.6 hari(p= 0.03). DIsamping itu, kadar kandungan dalam perut berkurangan di kumpulan yg menerima probiotik berbanding kumpulan kawalan.

Kesimpulan : Beberapa kajian klinical telah dijalankan menunjukkan keberkesaan probiotik dalam memperbaikki system immunisasi tubuh badan. Disamping itu, dalam kajian ini menunjukkan keputusan statistic yang positif dalam kesan probiotik untuk mengurangkan kadar sembelit dikalangan esakit kritikal.

ACKNOWLEDGMENT

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Not forgetting to Bcrobes for their generosity to provide samples of probiotic and placebo for the purpose of this study.

Finally,but never the least, a special thanks to my wonderful husband, Mohd Afzarizul Bin Othman Fuad for his understanding and kind words of wisdom through tough times and thiswhole journey.Without all your help and support, this project would not have been materialized.

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INTRODUCTIONS

Sepsis and its subsequent complications are the most common cause of death in IntensiveCare Units (ICUs).[5,6] The gastrointestinal tracts plays important role in _the pathogenesis of multiorgan dysfunction due to breakdown of intestinal barrierfunction and increasetranslocation of bacteria and bacteral components intosystemic circulation. During critical illness, alteration in gut flora are due to several factors that include changes in circulating stress, gut ischaemia, immunosuppresion,the use of antibiotics and other drugs, possible bacterial translocation and malnutritient.

Probiotics bacteria are live microorganism which provide health benefit the the host. They help to maintain the integrity of the intestinal barrier function by modulating the mucosal and systemic immune response of the host. Multipe probiotic studies have been included in treatment of critical ill patient to assess immunological response and prevention of constipation and subsequently reduce multiorgans dysfunction incidents. Numerous studies have shown that certain strains of lactobacilli and bifidobacteria can modulate the production of cytokines (mediators produced by immune cells) that are involved in the regulation, activation, growth, and differentiation of immune cells.

Endogenous probiotic bacteria of the gut such as Lactobacillus and Bifidobacterium play a vital role in maintaining the intestinal mucocal barrier and enhancing immune response is becoming evident from numerous study(10). In vitro studies have shown certain Bifidobacterium strains to release a proteinaceous factor that direcltly influence epithelial premeability and prevent invasions by potential pathogens(19).

Interleukin-6 (IL-6) is a multifunctional cytokine that plays a central role in host defense due to its wide range of immune and hematopoietic activities and its potent ability to induce the acute phase response.Probiotic shown to improve prolifeation of immune cells (De Simone et al. 1993) and prompt productions of proinflammatory cytokines such as tumor necrosis factor and interleukin 6 (Miettinen et al. 1996)Interleukin 6 (IL-6) is an inflammatory biomarker with a diagnostic and prognostic value in patients with sepsis which is used in the

prediction of mortality in patients with severe sepsis.[,7,8,9 Probiotics could reduce inflammation in critically ill-patients and might be considered as an adjunctive therapy in the treatment of critically ill-patients as its shown significant decrease in serum IL-6

Interleukin-10 (IL-10) is well characterized as an anti-inflammatory cytokine, with potent suppressive effects in preventing autoimmune disease.1 In the absence of IL-10, spontaneous mucosal autoimmunity develops, while the effect on systemic autoimmunity is far more muted. Some study shown Interleukin 10 (IL-10) is a cytokine of particular therapeutic interest in Inflammatory bowel disease since it plays a key role in the control of inflammatory responses to intestinal antigens and can restoretolerance of T cells to resident intestinal bacteria.[21

Properties of probiotics

Probiotics are, according to the FAO/WHO " live bacteria which when administered in adequate amounts confer the health benefit to the host. These bacteria do not contain any virulence properties or antiobiotic resistance cassettes. Probiotics, most commonly Lactobacillus and Bifidobacterium,can enhance nonspecific cellular immune response characterized by activation of macrophages, natural killer (NK) cells, antigen-specific cytotoxic T-lymphocytes, and the release of various cytokines in strain-specific and dose-dependent manner (1)

Probiotics are of beneficial effects in the treatment of a wide range of GI disease such as different types of diarrhea,[13,14] inflammatory bowel diseases,[15] irritable bowel syndrome[15] and pouchitis.[16,17] Since they play a role in reduction or elimination of pathogens and toxins, releasing of nutrients, antioxidants and growthfactors to stimulate intestinal motility, and regulation of the immune defence mechanisms by changing the

intestinal flora, probiotics seem to have beneficial effects in the improvement of critically ill-patients.[11,12]

OBJECTIVES OF STUDY

Primary outcome : to assess the efficacy of probiotic t in enhancing immune responce by measuring serum Interleukin 6 (IL-6), Interluekin 10 (IL-10) and hemeoxygenase level . Blood sample taken on Day 1, 3 and 5 of intervention from peripheral or central to be sent for analysis of the levels using ELISA.

Secondary outcomes:

1. Faecal output : Nursing staff will refer to the King's Stool Chart validated for use on ICU patients. The chart will aid in the characteristic of faecal frequency, consistency, and weight, which are then summarised into a daily stool score. The primary measurements would be the timing for the first bowel movement, frequency of bowel movement/week and stool consistency

2. Other parameters : Length of ICU stay, total numbers on ventilatior, timing and tolerance of nutrition, total use of prokinetic agents required for first bowel movement to occur, daily gastric volume from Day 0 to Day 5 , min blood sugar level and total insulin usage to control blood sugar in ICU patients.

METHODOLOGY

Recuitment process

Patients admitted between June and December 2016 to ICU of University Malaya Medical Centre were eligible for the study. The following are inclusion and exclusion criteria for this study

Inclusion criteria :

- Adult patients, 18 years and above
- Expected to have minumum stay 3 days in ICU
- Only patient who require mechanical ventilator support.

Exclusion criteria:

- Recent abdominal surgery within 7 days prior to ICU admission
- Patient with ileostody or colostomy
- Patients with prebiotic agents, for example lactulose
- Patients who are severely immunocompromised such as those with acute pancreatitis, HIV positive, neutropenia, advance lymphoma, steroid therapy and ongoing chemotherapy.
- Pregnancy

Consent will be obtained from their legal representative who will be briefed regarding the objectives of the study.

Any subject may withdraw at any time for any reason. These reason may include but are not limited to :

- Subject's or legal representative request
- Adverse event
- A concomitant therapy which could interfere with the results of the study.

Sample size

The sample size calculation is based on detecting difference level of IL-6, IL-10, Hemeoxygenase between the intervention and control groups by $1\log^{10}$ cells/g, in line with the previous study investigating on interleukin level in critically ill patients receiving enteral feeding. In order to achieve 90% power with $P< 0.05$ and 20% drop out value, 20 subjects were required in each group.

Study design :

This is a single centre, single centre, randomised, double blind, placebo controlled study. Enteral feeding was provided to study patient within 48 hours of ICU admission. By protocol, energy requirement were calculated using Indirect Calorimetry by ICU dietitian and progress accoding to Universitiy Malaya ICU protocol. All patients will be given standard fibre-free enteral feeding via feeding tube. When study patients started with enteral feeding, the study treatment and placebo preparation were dispensed in identical packaging and administered to the patient via feeding tube.

Patient will be randomly assigned by a computer generated system into either :

- Intervention Group ; will receive a sachet of Hexbio preparations, which a fermented milk product in granules forn with a mixed culture of six strains of Lactobacillus and Bifidobacterium, to be taken twice daily (every 12 hours). Hexbio is licensed as nutritional supplement by the National Pharmaceutical Control Bureau, Ministry of Health.

- Control group : will receive a placebo and same administration as in the intervention group.

Both group will received study treament for 5 days. . Blood sample taken on Day 1, 3 and 5 of intervention group from peripheral or central to be sent for analysis of the levels using ELISA.

A diagnosis of constipation is made when there is no bowel movement for 3 consecutive days. Upon diagnosis,stimulant laxative, ravin enema will be prescribed to the patients. All study received concomittent therapy including sedation and antiobiotic as considered appropriate by attending physician.

PROBIOTIC PROTOCOL

Objectives : to assess effect of probiotic in modulating immune response and prevention of contipation in critically ill patient

Recruitment : adult >18 years old, ventilated atleast 24-48 hours minimum stay ICU 3 days, allow NG feeding withitn 48 hours ICU admission

Exclusion : recent abdominal surgery, pancreatitis, pt on lactulose/ immunocompromised ,Subject refuse/withdrawal.

Start NG feeding OSMOLITE as calculated via INDIRECT CALORIMET(Novasource for fluid restrict, glucerna in poorly controlled DM)

PROBIOTIC to be given twice a day x 5/7 with IV maxolon 10 mg tds

Blood taking on D1, D3, D5

If patient no bowel opening for 3 days, to give laxative and continue

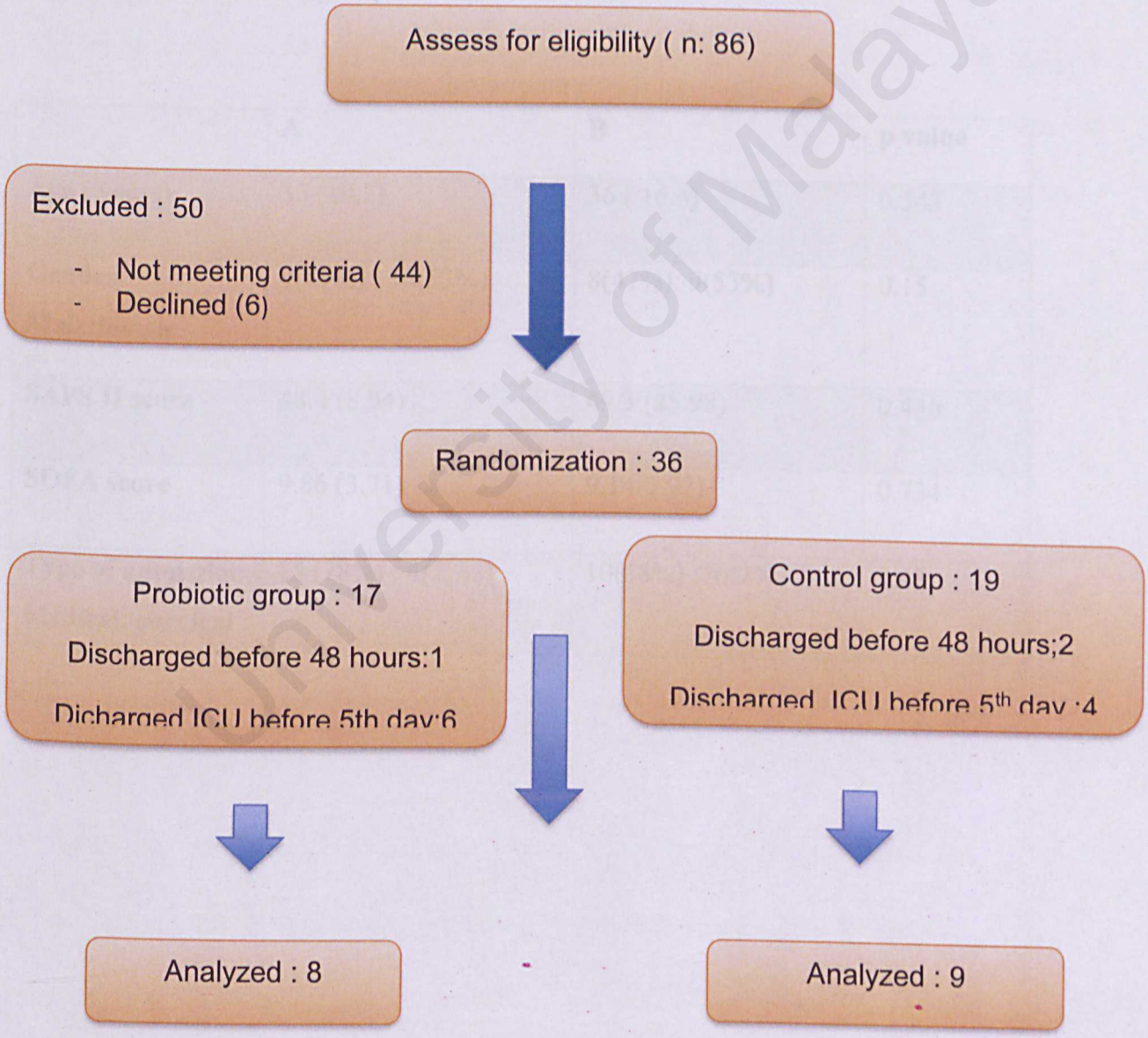
If NG aspirates <300 – return all aspirate and continue feeding.

If NG aspirates >300, reduce feeding 50% from current rate and increase accordingly

RESULTS

36 patients patient were enrolled into the trials, N = 19 received A preparations and N = 17 received B preparations . The demographic data of patients are summarized in Table 1. However, only 17 patients able tocomplete the 5 daysintervention, 11 patient completed 3 days intervention and 5 patients died during intervention and another 3patients no follow up done. No patients in the probiotic group developed *Lactobacillus*-induced sepsis.

Enrollment



Nutritional variables

All patients were started on standard fibre free enteral feeding via nasogastric tube continuesly within 24 hours of ICU admission. The patiens were started on Osmolite, no Novasourse if they require fluid restriction according to indirect calometry calculations by ICU dietitian recommendation. Administration rate was started at 20 mls/H and then increasd gradually as tolerated until each individual requirement was met. Intravenous Metoclopramide 10mg three times daily was administered as prokinetic agent to every patients.

Table 1 : Baseline demographic) (mean SD or proportion %)

	A	B	p value
Age (years)	35 (19.2)	36 (16.4)	0.543
Gender Male;female	9(47%) : 10(53%)	8(47%): 9(53%)	0.15
SAPS II score	48.4 (8.94)	49.3 (25.98)	0.436
SOFA score	9.86 (3.71)	9.14(3.97)	0.734
Type of admission Medical: surgical	15 (78%) : 4(22%)	10(58%) :7(42%)	0.43

Primary outcomes : Immunology enzymes level

During this course of study, serum analysis for enzymes level still in progress and unable to obtained the IL-6, IL-10 and hemeoxygenase level.

Secondary outcomes

Daily stool analysis

The timing of first bowel movement and mean frequency of bowel per day in both groups is summarized in Table 2. The king stool chart was used as the scoring system to record the stool consistency and stool weight of each patient on daily basis. The need for use of stimulant laxatives also recorded.

Table 2 : stool score (mean SD)

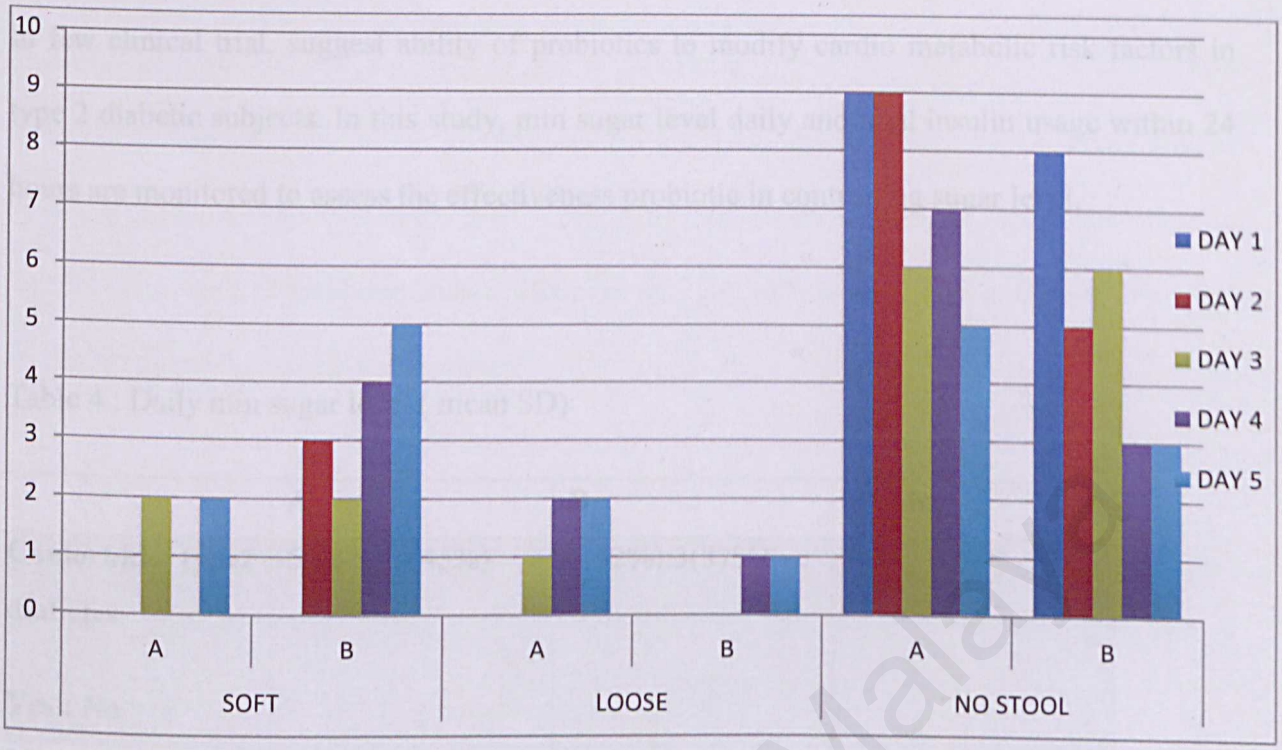
	A	B	p value
Timing of first bowel opening	4.6(1.4)	3.2(1.1)	0.03
Frequency bowel opening perday	0.35	3.75	0.09
Use of laxatives Yes : No	5(55%);4(45%)	2(25%);6(75%)	0.415

As the result above, the is significant difference between intervention group as they have shorter time for first bowel opening at 3.2 days compared with 4.6 days for placebo group (p value 0.03).

Daily stool chart (based on King's stool chart) n= no of patients

	A	B
Day 1		
Soft and formed	0	0
Loose and unformed	0	0
No stool	9	8
Day 2		
Soft and formed	0	3
Loose and unformed	0	0
No stool	9	5
Day 3		
Soft and formed	2	2
Loose and unformed	1	0
No stool	6	6
Day 4		
Soft and formed	0	4
Loose and unformed	2	1
No stool	7	3
Day 5		
Soft and formed	2	5
Loose and unformed	2	1
No stool	5	3

DAILY STOOL CONSISTENCY CHART



Other parameters

Apart from daily stool analysis, other parameters were monitored including gastric residual volume mls/day, no of days ventilator and length of stay at ICU showed at Table 3

Table 3 : gastric residual volumes, number of ventilator days and ICU stay (mean SD)

	A	B	p value
Gastric residual volumes(mls/day)	92(134)	14(38)	0.0003
Numbers of days ventilator	7.4(3.3)	5.7(3.32)	0.16
Lengths of ICU stay	9.6(4.9)	7(4.2)	0.13

Effect on sugar level in critical ill patients

In few clinical trial, suggest ability of probiotics to modify cardio metabolic risk factors in type 2 diabetic subjects. In this study, min sugar level daily and total insulin usage within 24 hours are monitored to assess the effectiveness probiotic in controlling sugar level.

Table 4 : Daily min sugar level (mean SD)

	A	B	p value
Comorbid type2 diabetes **Yes : No**	5(55%);4(45%)	5(62%):3(37%)	0.34
Min sugar level/day	9.37 (2.8)	7.4(1.53)	0.5
Total insulin in 24 hours	45.8(50.3)	42.4(78)	0.45

DISCUSSION

Probiotic bacteria are live microorganisms which confer to health benefits of the host. They help to maintain the integrity of the intestinal barrier function by modulating the mucosal and systemic immune response of the host. These bacteria have proven their beneficial effect in several conditions of ulcerativecolitis. More recently probiotics/synbiotics have been included in the treatment of critically ill patients.

The present study used a double-blind, placebo-controlled and randomized design to determine the effects of probiotics on critically ill in immunological booster effect. However, the serum analysis for immunological enzymes IL-6. IL-10 and hemeoxygenase level still in progress and unable to obtain the enzymes level for now.

The demographic data of patients in both groups were similar which means they were comparable as there was no significant difference between the mean age,gender, Simplified Acute physiology II (SAPS II) and Sepsis related Organ failure assessment (SOFA). Simplified Acute physiology II (SAPS II) score is a tool to measure the disease severity. An increasing score is closely correlated with the subsequent risk of many common diseases and hospital death. SOFA score is a simple and objective score that allows calculation of both the number and the severity of organs' dysfunctions and during the first few days of ICU admission is a proper indicator of prognosis. Independent of the initial score, an increase in SOFA score during the first 48 h in the ICU predicts a mortality rate of at least 50%.

Primary outcome : immunological response

Often credited as the first advocate for probiotics, ElieMetchnikoff, the father of immunology, investigated intestinal microbes as causative agents in aging, a process he called "autointoxication." He made the observation that lactic fermentation of milk products arrested putrefaction and suggested that the consumption of those products might offer the same protection to humans .

In previous clinical studies, IL-6 appears to be a good indicator of activation of the cytokine cascade and predicts subsequent organ dysfunction and mortality.[30] be associated with inflammation and sepsis IL-6 is a 21 kDa glycoprotein produced by many cell types including lymphocytes, fibroblasts and monocytes. It has many systemic effects including induction of acute phase protein production in the liver. It was reported by McNaught *et al.* who showed that enteral administration of ProViva, an oatmeal-based drink containing *L. plantarum* 299 v to critically ill patients resulted in significantly lower levels of IL-6 in the probiotic group compared with the controls.

IL-10 has important regulatory effects on immunological and inflammatory responses because of its capacity to downregulate class II MHC expression and to inhibit the production of proinflammatory cytokines by monocytes.

At the cellular level, hemeoxygenase positively affects a variety of substances that are involved in gut function. This is important to ensure the correct absorption of nutrients and to maintain barrier functionality.

However in this study, serum analysis for IL-6, IL10 and hemeoxygenase level still in progress. Therefor unable to analyse the effect of probiotics in immunological response in critically ill patients.

Probiotic effect in prevention of constipation in critical ill patients.

Probiotic , most commonly Lactobacillus and Bifidobacterium, have been shown to exert preventive effects in various disease, such as antibiotic induce diarrhea, constipation, necrotizing enterocolitis and campylobacter induced enteritis. Constipation is common critical ill patients with ICU stay more than 48 hours. An increase in the number of bowel movements or a decrease in transit time has been reported in controlled studies that employed probiotics for treating constipation [71,72]. The widely used laxative lactulose is a prebiotic, as it is not attacked by human disaccharidases and is substrate for the bifidobacteria in the colonic flora, that catabolise it to smaller molecules, creating an osmotic effect. However, furtherclinical trials require as there is a need for larger controlled studies using probiotics and prebiotics other than lactulose.

In our study showed there is significant differences between intervention group and placebo group as first bowel opening for intervention group is 3.2 and placebo group is 4.6 (p= 0.03). There is also significant differences in gastric residual volume daily as intervention group produced 14 mls/day and placebo group 92 mls per day (p = 0.0003). Therefore it is proven beneficial effect of probiotic in enhancing bowel motility in critically ill patients.

Probiotics in improving blood glucose level.

Oxidative stress plays a major role in the pathogenesis and progression of diabetes. Among various functional foods with an antioxidant effect, probiotic foods have been reported to repress oxidative stress and subsequently improving the blood glucose level . It was reported consumption of probiotic improved fasting blood glucose and antioxidant status in type 2 diabetic patients. The intervention group showed lower min blood sugar level 7.4 compared to placebo group 9.37. Although the results were

statisticallynsignificant, it has positive outcome.

Sepsis in critically ill patients

Septic complications in surgical and intensive care patients arecommon. Large majorities of nosocomial infections are caused by intestinallyderived organisms such as E. coli and the gut origin of sepsis hypothesis relies upon the phenomenon of bacterial translocation. There is therefore a sound theoretical basis for proposing that alterations in GI microflora might influence translocation and subsequent septic morbidity. In this study showed length of ICU stayed and number of days required of ventilators support in intervention group are less compared to placebo group. Although statistically insignificant ($p=0.13$) it can be concluded the beneficial effect of probiotic in enhancing immune response subsequently reduce morbidity in critically ill patients.

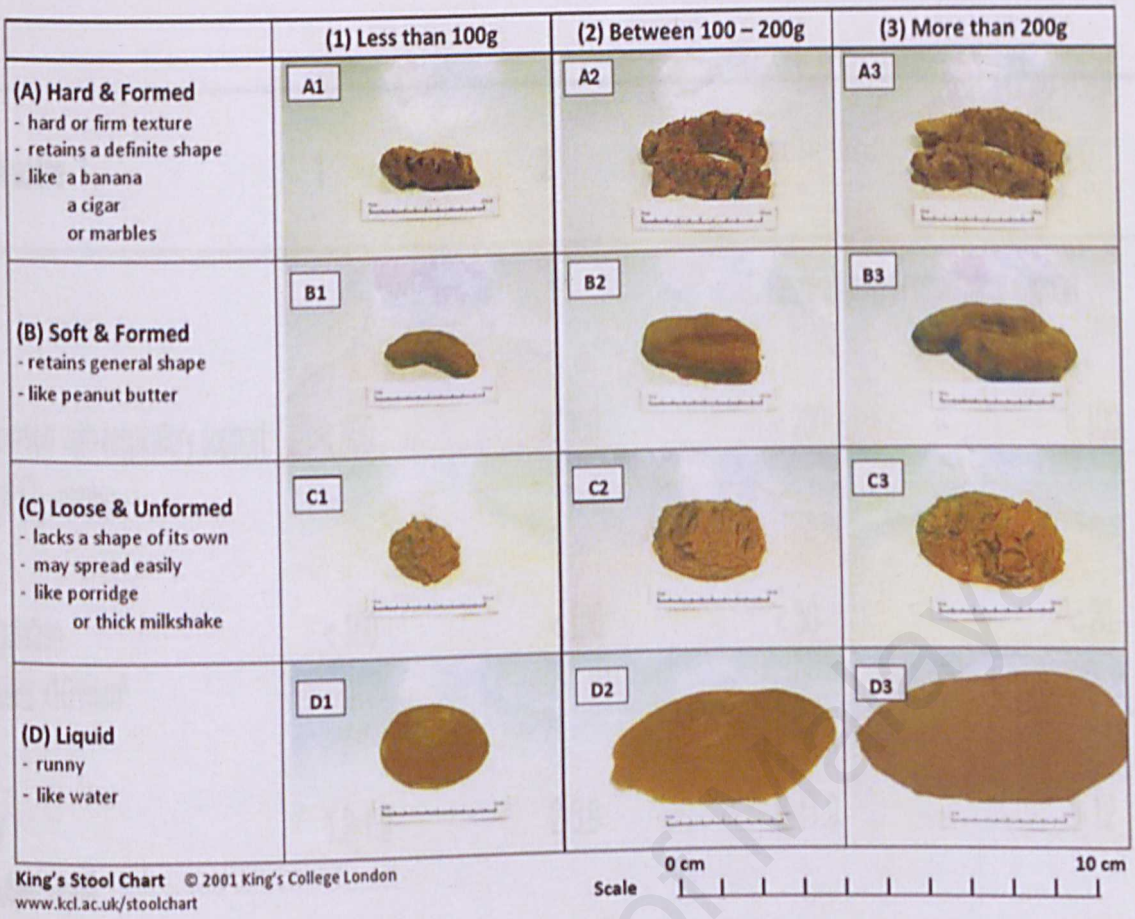
Limitation of the study

Our study was a single-center study with 36 patients included. Therefore, future multi-center trials with larger sample sizes of critically ill patients are required to achieve more solid results. Furthermore, further trials are needed to define the best dosage and optimal duration of therapy in these patients.

Based on previous clinical trial done, probiotics have beneficial effect in reducing inflammatory level in critically ill-patients and might be considered as an adjunctive therapy in the treatment of critically ill-patients. However, further studies with larger sample size are required to clarify their usefulness in this group of patients.

CONCLUSION

Probiotics could reduce inflammation in critically ill-patients and might be considered as an adjunctive therapy in the treatment of critically ill-patients as it help in reduce morbidity . Probiotics also have beneficial effect in preventing contipations as statiscally showed significant difference in first bowel opening and reduced mean gastric contents daily. Other that that, probitotic promote antioxidant effect which suqsequently help in controlling blood sugar level in critical ill patients.



King's Stool Chart

www.kcl.ac.uk/stoolchart

Instructions for use

1. First, consider the **consistency** of the faecal sample by comparison with both the verbal and photographic descriptors (A, B, C, D).

2. Then, consider the **weight** of the faecal sample by comparison with the photographic descriptors (1, 2, 3). Compare the size of the sample, using the life size 10cm scale, and compare it to the 10cm scale on each photographic descriptor.

3. Record the **frequency** of faecal output over a 24 hour period.

4. Any **other characteristics** of faecal output which are considered to be important must also be recorded e.g. incontinence, colour etc.

Whelan K, Judd PA, Preedy VR, Taylor MA. Covert assessment of concurrent and construct validity of a chart to characterize fecal output and diarrhea in patients receiving enteral nutrition. *Journal of Parenteral and Enteral Nutrition*. 2008; 32: 160-168.

Whelan K, Judd PA, Taylor MA. Assessment of faecal output in patients receiving enteral tube feeding: validation of a novel chart. *European Journal of Clinical Nutrition* 2004; 58: 130-137.

SOFA score	1	2	3	4
			Respiration with respiratory support	
Respiration with respiratory support PaO_2/FiO_2, mmHg	< 400	< 300	< 200	< 100
Coagulation Platelets $x10^3/mm^3$	< 150	< 100	< 50	< 20
Liver Bilirubin, mg/dl	1.2-1.9	2-5.9	6-11.9	> 12
Cardiovascular Hypotension >15 or (doses in ug/kg·min) cathecolamines> 0,1	MAP < 70mmHg	Dopamine ≤ 5 or Dobutamine (any dose)	Dopamine > 5 or cathecolamines ≤ 0.1	Dopamine
Neurologic Glasgow Coma Score	13-14	10-12	6-9	< 6
Renal Creatinine mg/dl or Urine output ml/zi	1.2-1.9	2-3.4	3.5-4.9 (200-500)	> 5 (< 200)

SAPS II Score

Parameter	Value (score)						
HR			<40 (11)	40-69 (2)	70-119 (0)	120-159 (4)	>160 (7)
SBP			<70 (13)	70-99 (5)	100-199 (0)	>200 (2)	
Temp					<39°C (0)	>39°C (3)	
PaO_2/FIO_2	<100 (11)	100-199 (9)	>200 (6)				
UO (ml)		<500 (11)	>500 (4)		>1000 (0)		
S. Urea					<28 (0)	28-83 (6)	>84 (10)
TLC (10^3/cc)				<1 (12)	1-20 (0)	>20 (3)	
K				<3 (3)	3-4.9 (0)	>5 (3)	
Na				<125 (5)	125-144 (0)	>145 (1)	
Bicarb			<15 (6)	15-19 (3)	>20 (0)		
Bil					<4 (0)	4-5.9 (4)	>6 (9)
GCS	<6 (26)	6-8 (13)	9-10 (7)	11-13 (5)	14-15 (0)		

Age -score

<40 → 0
40-59 → 7
60-69 → 12
70-74 → 15
75-79 → 16
≥80 → 18

Chronic disease:

Metastatic cancer → 9
Hemat.malig → 10
AIDS → 17

Type of admission:

Sched. Surgical → 0
Medical → 6
Emer.surgical → 8

JAMA 1993;270(24):2957-2963

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References

1. Ashraf R1, Shah NP. Immune system stimulation by probiotic microorganisms 2014;54(7):938-56.NCBI

2. B. Sheil,J. MacSharry,L. O'Callaghan,A. O'Riordan,A. Waters,J. Morgan,J. K. Collins,L.O'Mahony,F. ShanahanRole of interleukin (IL-10) in probiotic-mediated immune modulation: an assessment in wild-type and IL-10 knock-out mice , The journal of translasional immunology

3. .Sanaie S1, Ebrahimi-Mameghani M2, Hamishehkar H3, Mojtahedzadeh M4, Mahmoodpoor A5. : Effect of a multispecies probiotic on inflammatory markers in critically ill patients: A randomized, double-blind, placebo-controlled trial. 2014 Sep;19(9):827-33 NCBI

4. Yan F, Polk DB. Probiotics as functional food in the treatment of diarrhea. Curr Opin Clin Nutr Metab Care. 2006;9:717–21. [PubMed]

5. Centers for Disease Control (CDC). Increase in National Hospital Discharge Survey rates for septicemia – United States, 1979-1987. MMWR Morb Mortal Wkly Rep. 1990;39:31–4. [PubMed]

6. Martin GS, Mannino DM, Eaton S, Moss M. The epidemiology of sepsis in the United States from 1979 through 2000. N Engl J Med. 2003;348:1546–54. [PubMed]

7. . Haasper C, Kalmbach M, Dikos GD, Meller R, Müller C, Krettek C, et al. Prognostic value of procalcitonin (PCT) and/or interleukin-6 (IL-6) plasma levels after multiple trauma for the development of multi organ dysfunction syndrome (MODS) or sepsis. Technol Health Care. 2010;18:89–100. [PubMed]

8. 7. Hamishehkar H, Beigmohammadi MT, Abdollahi M, Ahmadi A, Mahmoodpour A, Mirjalili MR, et al. Identification of enhanced cytokine generation following sepsis. Dream of magic bullet for mortality prediction and therapeutic evaluation. Daru. 2010;18:155–62. [PMC free article] [PubMed]

9. 8. Farhoudi M, Najafi-Nesheli M, Hashemilar M, Mahmoodpoor A, Sharifipour E, Baradaran B, et al. Effect of IMOD™ on the inflammatory process after acute

ischemic stroke: A randomized clinical trial. Daru. 2013;21:26. [PMC free article] [PubMed]

10. Alberda C, Gramlich L, Meddings J, Field C, McCargar L, Kutsogiannis D, et al. Effects of probiotic therapy in critically ill patients: A randomized, double-blind, placebo-controlled trial. Am J Clin Nutr. 2007;85:816–23. [PubMed]

11. Morrow LE. Probiotics in the intensive care unit. Curr Opin Crit Care. 2009;15:144–8. [PubMed]

12. 25. Ebrahimi-Mameghani M, Sanaie S, Mahmoodpoor A, Hamishehkar H. Effect of a probiotic preparation (VSL#3) in critically ill patients: A randomized, double-blind, placebo-controlled trial (Pilot Study) Pak J Med Sci. 2013;29:490–4. [PMC free article] [PubMed]

13. Delia P, Sansotta G, Donato V, Messina G, Frosina P, Pergolizzi S, et al. Prevention of radiation-induced diarrhea with the use of VSL#3, a new high-potency probiotic preparation. Am J Gastroenterol. 2002;97:2150–2. [PubMed]

14. Hedin C, Whelan K, Lindsay JO. Evidence for the use of probiotics and prebiotics in inflammatory bowel disease: A review of clinical trials. Proc Nutr Soc. 2007;66:307–15. [PubMed]

15. 21. Guandalini S, Magazzù G, Chiaro A, La Balestra V, Di Nardo G, Gopalan S, et al. VSL#3 improves symptoms in children with irritable bowel syndrome: A multicenter, randomized, placebo-controlled, double-blind, crossover study. J Pediatr Gastroenterol Nutr. 2010;51:24–30. [PubMed]

16. 22. Gionchetti P, Rizzello F, Venturi A, Brigidi P, Matteuzzi D, Bazzocchi G, et al. Oral bacteriotherapy as maintenance treatment in patients with chronic pouchitis: A double-blind, placebo-controlled trial. Gastroenterology. 2000;119:305–9. [PubMed]

17. 23. Gionchetti P, Rizzello F, Helwig U, Venturi A, Lammers KM, Brigidi P, et al. Prophylaxis of pouchitis onset with probiotic therapy: A double-blind, placebo-controlled trial. Gastroenterology. 2003;124:1202–9. [PubMed]

18. 24. Morrow LE. Probiotics in the intensive care unit. Curr Opin Crit Care. 2009;15:144–8. [PubMed]

19. Madsen K, Cornish A, Soper P, et al. Probiotic bacteria enhance murine and human intestinal epithelial barrier function. Gastroenterology 2001;121:580–91. CrossRefMedline

20. Sarvin Sanaie, Mehrangiz Ebrahimi-Mameghani,1 Hadi Hamishehkar,2 Mojtaba Mojtahedzadeh,3 and Ata Mahmoodpoor4 : Effect of a multispecies probiotic on inflammatory markers in critically ill patients: A randomized, double-blind, placebo-controlled trial.

21. Steidler, L., Hans, W. and Schotte, L. et al. (2000) Treatment of murine colitis by Lactococcus lactis secreting interleukin 10. Science 289, 1352^1354.

22. Koebnick C, Wagner I, Leitzmann P, Stern U, Zunft HJ. Probiotic beverage containing Lactobacillus casei Shirota improves gastrointestinal symptoms in patients with chronic constipation. Can J Gastroenterol 2003; 17:655-9

23. Ouwehand AC, Lagstrom H, Suomalainen T, Salminen S.. Effect of probiotics on constipation, fecal azoreductase activity and fecal mucin content in the elderly. Ann Nutr Metab 2002; 46:159–62.

24. Rayes N, Seehofer D, Hansen S, Boucsein K, Muller AR, Serke S et al. Early enteral supply of lactobacillus and fiber versus selective bowel decontamination: a controlled trial in liver transplant recipients. Transplantation.2002;74:123-7

25. Rayes N, Hansen S, Seehofer D, Muller AR, Serke S, Bengmark S, et al. Early enteral supply of fibre and lactobacilli vs parenteral nutrition—a controlled trial in major abdominal surgery patients. Nutrition 2002;18: 609–15.