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ORIGINAL ARTICLE

# Innovative strategies for the rapid restoration of intestinal function in patients undergoing abdominal surgery: use of probiotics
## Pilot study of 15 patients

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ABSTRACT

BACKGROUND: The clinical value of probiotics in patients undergoing abdominal surgery, particularly colorectal surgery, remains uncertain despite their well-documented health benefits. This pilot randomized controlled trial aimed to assess the effects of perioperative and postoperative oral administration of two probiotics, *Clostridium butyricum* CBM588® and *Bifidobacterium longum* ES1, on immune function, systemic inflammatory response, postoperative infections, and recovery after colorectal surgery.
METHODS: Fifteen adult patients underwent colorectal resection, with two groups receiving probiotics and one acting as a control. Blood and fecal samples were collected, and clinical parameters were assessed.
RESULTS: Results showed the safety of probiotics, resistance to antibiotics and gastric acid, and potential benefits in reducing postoperative infections and intestinal inflammation.
CONCLUSIONS: Future trials should provide more conclusive evidence on the efficacy and safety of perioperative probiotic administration in colorectal surgery, aiming for improved patient outcomes and reduced healthcare costs.

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KEY WORDS: Probiotics; Intestines; Digestive system surgical procedures; Neoplasms.

The perioperative and postoperative oral administration of probiotics to patients undergoing colorectal surgery has emerged as a topic of considerable interest in the field of surgical interventions and gastrointestinal health. Colorectal surgery poses a risk of postoperative compli-

cations, including infections and disturbances in gut microbiota. Probiotics, live microorganisms with potential health benefits, have been investigated for their ability to modulate the gut microbiota and influence the immune system.[1-3]

Some clinical studies have demonstrated that prophylactic administration of probiotics, in some cases in combination with prebiotics (as symbiotics), to patients scheduled for abdominal surgery reduced the incidence of postoperative infections.[4, 5] Also, some reviews collecting data from different research[6, 7] have yielded results in that direction.

Therefore, the effects of combinations of probiotics and/or prebiotics on postoperative infections in patients is still under study, and further research involving the use of different species of probiotics for specific patient groups are needed to better define their efficacy. These findings align with the understanding that the delicate balance of the gut microbiota plays a crucial role in maintaining intestinal health and preventing surgical complications, also reducing the hospitalization after surgery.

Probiotics are believed to enhance the recovery of intestinal function by promoting the growth of beneficial bacteria and suppressing the proliferation of harmful pathogens. Additionally, they may exert anti-inflammatory effects, potentially reducing the overall inflammatory response associated with surgical trauma.[8, 9]

For example, *Clostridium butyricum* (CB) is a butyrate-producing, spore-forming anaerobic gram-positive, and obligate anaerobic rod bacterium. Butyrate producing bacteria could potentially be used in patients with Colorectal cancer (CRC).[10, 11] *Clostridium butyricum* may modulate the composition of the gut microbiome and promote the growth of beneficial microbes in the intestine, such as *Bifidobacterium* and *Lactobacillus*.[12, 13] *Clostridium butyricum* is effective against gastrointestinal infections caused by antibiotics-induced dysbiosis, it can inhibit the transmission of antibiotic-resistant genes and maintain homeostasis of the gut microbiome.[14] The treatment with *Clostridium butyricum* in association with specific immunotherapy was effective in ulcerative colitis and food allergy.[15]

Another relevant probiotic is the *Bifidobacterium longum*, a beneficial bacterial strain belonging to the *Bifidobacterium* genus. It aids in the fermentation of non-digestible fibers, producing short-chain fatty acids that promote intestinal well-being. It has been used also in addition to other probiotics in postoperative treatment for colorectal surgery[16] with significant positive results for example reducing the number of days to first defecation and the number of diarrhea episodes. *Bifidobacterium longum* was also able to suppress inflammatory reactions, maintain immune responses[17] and exert antitumor activity in case of colon tumor.[18] Laboratory studies also showed that, *Bifidobacterium longum* attenuates the production of inflammatory cytokines and the CD4+ T-cell mediated immune response in an animal model of gliadin-induced enteropathy.[19, 20]

The immunomodulatory properties of probiotics are particularly relevant in the context of colorectal surgery, where preserving immune function is essential for optimal recovery. Regarding the faster recovery of bowel movements after surgery coupled with a reduction in infections and inflammation after colorectal surgery, can also significantly contribute to a reduction in the cost of hospital stays through various interconnected mechanisms with the rapid overall recovery of the patients, reduced use of drugs including antibiotics.

While these factors collectively contribute to cost reduction, it is crucial to emphasize that the economic impact may vary based on individual patient characteristics, surgical procedures, and healthcare system nuances. Nonetheless, the relationship between expedited recovery of bowel movements, reduced infections, and inflammation is integral to improving patient outcomes and achieving cost-effective healthcare delivery.

The use of probiotics has been widely documented to benefit human health, but their clinical value in patients undergoing abdominal surgery remains unclear. The present pilot study is a randomized control trial designed to investigate the effects of perioperative and postoperative oral administration of two probiotics: *Clostridium butyricum* CBM588® (Butirrisan®, PharmExtracta S.p.A., Italy) and *Bifidobacterium longum* ES1 (GliadinES®, PharmExtracta S.p.A., Italy)

in patients undergoing colorectal surgery. Effects on immune function, systemic inflammatory response, postoperative infections and recovery after surgery of colon, rectum and anus have been evaluated.

## Materials and methods

### Study design and participants

A randomized control trial was performed in 2023 on 15 adult patients undergoing colorectal resection at the Azienda Ospedaliero Universitaria Città della Salute e della Scienza in Turin (Italy). Patients were excluded from the study if they were under radiotherapy, affected by autoimmune diseases, and had advanced stages of malignant tumors. Patients were informed about the protocol and gave individual consent to participate in the trial. All the 15 patients completed the whole trial. Patients were randomly allocated to three groups 30 days prior to the surgery. Two groups were treated with probiotics 30 days before and continued for another 30 days after surgery (leaving the day of surgery and two days after it without any supplementation). Specifically, one group (Group 1, N.=5) received a supplement containing *Bifidobacterium longum* ES1 (Table I), the second group (Group 2, N.=5) received a supplement containing *Clostridium butyricum* CBM588® (Table II). The last group (Control, N.=5) was used as control as no supplementation was given to the patients for the whole trial duration. Maltodextrins were not administered to prevent bias.

TABLE I.—*Supplement with* Bifidobacterium longum *ES1 (1 billion live cells). Dosage: 1 stick/day.*

| Ingredients | mg/stick | % |
|---|---|---|
| *Bifidobacterium longum* ES1 | 30 | 3 |
| Maltodestrine | 600 | 60 |
| Sorbitol | 267.6 | 26.76 |
| Rice starch | 100 | 10 |
| Silicon dioxide | 1.2 | 0.12 |
| Sucralose | 1.2 | 0.12 |
| Total | 1000 | 100 |

TABLE II.—*Ingredients of the supplement containing* Clostridium butyricum *CBM588® ≥4.5×10⁵ CFU (colony-forming unit) (30 mg) per tablet. Dosage: 2 tablets 3 times/day.*

| Ingredients | mg/tablet | % |
|---|---|---|
| *Clostridium butyricum* (CBM588®) | 30 | 10% |
| Lactose | 172.2 | 57.4% |
| Rice starch | 73.8 | 24.6% |
| Microcrystalline cellulose | 15 | 5% |
| Sucrose | 6 | 2% |
| Magnesium salt of fatty acid | 1.5 | 0.5% |
| Talc E553b | 1.5 | 0.5% |
| Total | 300 | 100% |

### Clinical observations and blood test

All patients received a short-term antibiotics treatment during the surgery. Antibiotic prophylaxis performed due to scheme with single dose during induction of anesthesia: * clindamycin 2 g (if the patient declare allergy use ciprofloxacin 400 mg or amoxiclavulanate 2.2 g), * metronidazole 500 mg (only in case of left hemicolectomy as a combination drug). No antibiotic therapy administered postoperatively. Anamnestic data and medical history were recorded. A questionnaire was developed and data were recorded by the same doctor during the trial (Supplementary Digital Material 1: Supplementary File 1).

Blood samples were collected 30 days before, 10 and 30 days after surgery (T-30, T+10, T+30) to assess some biomarkers related to the health condition of the patients monitoring infection and inflammation status (white blood cells-WBC, Hemoglobin-Hb, C-reactive protein-CRP). Fecal samples were also collected (T-30, T+10, T+30) for the calprotectin marker to assess intestine inflammation. The Bristol scale (scale 1-7)[20] was used to describe the shapes and types of stools 30 days before the surgery, then 30 days after (T-30, T+30). The number of defecations/day was recorded 30 days before and 30 days after the surgery (T-30, T+30), the mean number of defecation per day in a week were reported.

### Statistical analysis

The Kruskal-Wallis Test was used to compare the distributions of two or more independent groups. A low P value indicates that at least one of the groups differs from the others. ANCOVA was used to compare a dependent variable across groups, while controlling for continuous variables. A low P value indicates significant differences between the groups, once adjusted for the covariates.

The Friedman Test was used to compare more than two related groups across more than two repeated measures. A low P value indicates significant differences between the groups over the time points.

The Wilcoxon Test was used to compare two paired samples or repeated measures on a single sample. A low P value indicates significant differences between the pairs.

Python was used for analysis, with the following versions: version of Python: 3.11.8; pandas version: 2.1.4; scipy version: 1.12.0; statsmodels version: 0.14.0.

## Results

Background patients data are summarized in Table III. All patients except two (44 and 47) were 61-83 years old (mean age 68 years). Five female and 10 male patients were included in the study. Most of the patients were overweight or obese as shown by the BMI values. Several concomitant conditions were reported by patients and recorded. All except two of the pathologies for which the surgery was planned were malignant neoplasms. All surgical procedures was performed completely in laparoscopy. At Group 1 was performed three right hemicolectomy (AdenoCa pT3N0 G2 // AdenoCa pT4aN1b G2 // AdenoCa pT2N0 G2); one extended to transverse colon right hemicolectomy + wedge resection of mid part of jejunum + cholecistectomy for suspected tumor infiltration (AdenoCa pt4N0 G2 colon + NET pT1 G1 jejunum + gall bladder regular with area of nonspecific parietal fibrosis) and one left hemicolectomy (pseudotumor due to diverticular diseases with diverticulitis). At Group 2 was performed one right hemicolectomy + cholecistectomy (NET pT3N1 G2 + chronic calculous cholecystitis); four left hemicolectomy (AdenoCa pT1N0 G2 // AdenoCa pT2N0 G2 // AdenoCa pT3N0 G2 // diverticular inflammation with LGD). At Group 3 was performed two right hemicolectomy (AdenoCa pT2N0 G2 // serrated sessile adenoma LGD with focal HGD) and three left hemicolectomy (AdenoCa pT3N0 G2 // tubulo-villous adenoma HGD + intraepithelial neoplasia IEN // fibrotic stricture with focal HGD).

No patient developed fever (*i.e.* ≥38.0°).

Time of hospitalization (days after surgery): was in average 4.4 // median 4.

Time of bowel evacuation (days after surgery): was in average 3.4 // median 3.

No side effects in the two treated groups were reported.

Blood and fecal parameters by time points are summarized with mean and standard deviations in Table IV. Bristol scale data and number of bowel evacuation/day are represented in Figure 1.

For all parameters, pre-surgery the p-values were not statistically significant, indicating that there were no substantial differences between the groups at baseline (WBC P=0.454; HB P=0.104; PCR P=0.492; CALPROT P=0.336). The Kruskal-Wallis Test results confirm that for the ob-

TABLE III.—*Background main characteristics of patients belonging to the three groups, Group 1 received a supplement containing* Bifidobacterium longum *ES1, Group 2 which received a supplement containing* Clostridium butyricum *CBM588®, and the control.*

| Characteristic | Patients (N.=15) | | |
|---|---|---|---|
| | Control (N.=5) | Group 1 (N.=5) | Group 2 (N.=5) |
| Gender (M/F) | 2/3 | 4/1 | 4/1 |
| Age (mean, years) | 70 | 61 | 71 |
| Body Mass Index (BMI, kg/m²) | 29.9 | 27.8 | 25.4 |
| Hypertension | 4 | 3 | 5 |
| Diabetes | 3 | 2 | 0 |
| Cardiovascular diseases | 3 | 3 | 2 |
| Gastroenteric diseases | 1 | 2 | 2 |
| Histological findings | Adenocarcinoma with no lymph nodes involved (N.=4), diverticular disease (N.=1) | Adenocarcinoma with lymph nodes involved (N.=4), diverticular disease (N.=1) | Adenocarcinoma grading 2 (N.=4), cholecystitis (N.=1) |
| Surgery duration (minutes, mean) | 119 | 170 | 175 |

TABLE IV.—*Mean and standard deviation for the blood (white blood cells-WBC, Hemoglobin-Hb, C-reactive protein-CRP) and fecal marker (calprotectin-CALPROT) at the three time points (7 days before surgery, 10 days after and 30 days after surgery) for the three groups under study.*

| Parameter | Timepoint | Controls | Group 1 | Group 2 |
|---|---|---|---|---|
| WBC ($10^9$/L) | Pre-surgery | 7.1±2.4 | 7.1±1.5 | 6.6±1.1 |
| Normal range: 4.0-10.0 | 10 days | 6.3±2.3 | 6.6±1.6 | 8.4±0.8 |
| | 30 days | 6.4±1.2 | 5.5±0.8 | 5.9±1.2 |
| HB (g/dL) | Pre-surgery | 14.3±0.6 | 13.2±1.8 | 14.5±1.9 |
| Normal range: M 13.5-18; F 12.0-16.0 | 10 days | 13.2±0.9 | 12.4±0.9 | 13.7±1 |
| | 30 days | 14±0.3 | 14.1±1.5 | 14.3±0.8 |
| CRP (mg/L) | Pre-surgery | 4.4±1.5 | 3.1±1.8 | 5.4±1.4 |
| Normal range <5.0 | 10 days | 23.9±14.5 | 13.6±5.4 | 23.9±16.7 |
| | 30 days | 3.6±1.4 | 4.2±1.6 | 5±0.5 |
| CALPROT (mg/kg) | Pre-surgery | 52.2±42.8 | 88.6±56.9 | 58±26.9 |
| Normal range <50 | 10 days | 1012±1167.9 | 1877.4±612.9 | 1457.4±738.1 |
| | 30 days | 79.8±74.6 | 69±34.8 | 42±14.7 |

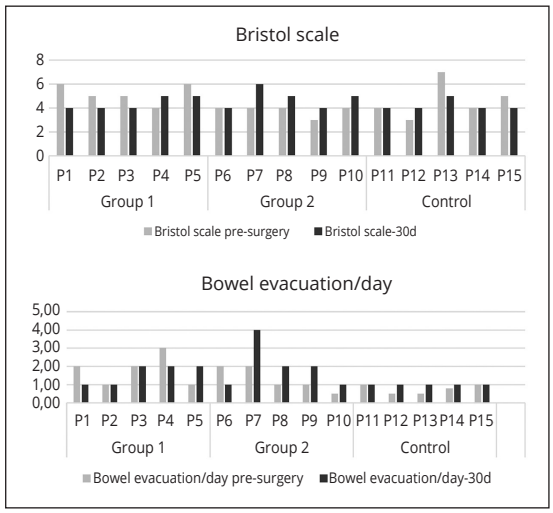


Figure 1.—Bristol scale data and mean number of bowel evacuation/day in a week at two time points and by group under study.

served variables, the three groups of patients were well randomized at the beginning of the study.

Based on the results from the ANCOVA, for all tested parameters (WBC, HB, PCR, CALPROT), the P values for groups, age, and BMI are not statistically significant, suggesting that, after adjusting for age and BMI, there are no significant differences in the baseline levels of the parameters between the groups. Table V represent the results of Wilcoxon Test.

## Discussion

This pilot study had the aim to test the effect of two probiotics: *Clostridium butyricum* CBM588® and *Bifidobacterium longum* ES1 on some of the main parameters relevant for colorectal surgery. The promising preliminary results of this study

TABLE V.—*Results of the Wilcoxon Test on white blood cells (WBC), hemoglobin (Hb), C-reactive protein (CRP) and the fecal marker calprotectin (CALPROT). Comparison were made between 10 days after surgery and 30 days after surgery, before the surgery and after 10 days, and before the surgery and 30 days after the surgery.*

| P value Wilcoxon | | Periods of comparison | | |
|---|---|---|---|---|
| Variable | Group | 10 days *vs.* 30 days | Pre-surgery *vs.* 10 days | Pre-surgery *vs.* 30 days |
| CALPROT | Group 1 | 0.0625* | 0.0625* | 0.0625* |
| | Group 2 | 0.125 | 0.0625* | 1 |
| | Group 3 | 0.0625* | 0.0625* | 0.625 |
| Hb | Group 1 | 0.0625* | 0.625 | 0.625 |
| | Group 2 | 0.125 | 0.1875 | 0.625 |
| | Group 3 | 0.0625* | 0.0625* | 0.125 |
| CRP | Group 1 | 0.0625* | 0.0625* | 0.625 |
| | Group 2 | 0.0625* | 0.0625* | 0.1875 |
| | Group 3 | 0.0625* | 0.0625* | 0.4375 |
| WBC | Group 1 | 0.0625* | 0.4375 | 0.0625* |
| | Group 2 | 0.8125 | 0.8125 | 1 |
| | Group 3 | 0.0625* | 0.1875 | 0.3125 |

*Statistically significant difference.

will help planning future trials in order to better understand the use of perioperative probiotics in case of colorectal surgery. This is further supported by several other studies[21] highlighting the need to perform trials on this topic as it would be an excellent help for the hospital and home management of these patients.

In this study, the two probiotics were considered safe and no side effects were reported by patients, this confirms previous studies on human patients with different conditions.[18, 22] This is a relevant result given the malignant pathologies and the fragile mental health status of the patients in Group 1 and 2 which at least did not see any additional discomfort linked to the products before and after the surgery.

Furthermore, it is noteworthy that the probiotics administered to the patients in our trial exhibited resistance to both the antibiotics administered during surgery and to gastric acid (hydrochloric acid). This aspect holds significant importance, as it ensures that the effects of these products remain unaltered and unaffected by the concurrent presence of antibiotics and gastric acid, thereby enhancing the reliability and integrity of our study findings.

Several studies reported beneficial effects of probiotics in patients undergoing GI surgery in terms of reduction of infection complications which are the most concerning post-surgery problems (abscesses, urinary infections, infection in the Trocar sites). The infection indexes are relevant to indirectly highlight any potential surgery complications which are very important especially in case of tumors. In a recent systematic review, authors stated that probiotics may result in reducing overall postoperative infectious complications after colorectal cancer surgery (RR: 0.45, 95% CI: 0.27 to 0.76; $I^2$=38%; low CoE).[22] The reduction of infections can guarantee a better and faster recovery to be back to a normal life.

In our study, it was not possible to see any significant difference in the variation of WBC in the different groups of patients at the different time points. Beside this, it has to be noticed that in Group 1 and Group 2 the malignant tumors affecting these patients were more severe and advanced than in the control group. This might have had an effect on blood results on WBC count as the two treated groups might have had a different immune response. Beside this the WBC values seemed decreased more than the control at T+30.

The stability of hemoglobin (Hb) levels throughout the entire trial serves as a reaffirmation of the overall successful surgical procedure. This consistent maintenance of Hb levels indicates the robustness of the surgery, reflecting a positive outcome in terms of patient well-being.

C-reactive protein (CRP) levels were comparable among the three groups, displaying an anticipated rise on the 10$^{th}$ day post-surgery, notably less pronounced in Group 1. Subsequently, CRP levels trended towards similarity at the conclusion of the trial. On the contrary, in a trial with patients undergoing radical colorectal resection an oral bifid triple viable probiotics was used and the treated group showed a reduction in CRP. So, this effect could also be modulated by different elements such as the type of pathology and the type/ duration of probiotic.[23]

Both groups of patients treated with probiotics in our study showed a reduction in the fecal Calprotectin levels 30 days post-surgery and the mean values in both groups were even lower at the end of the trial than at the beginning. On the contrary, the control group showed an increase of CALPRO level compared to the beginning of the study. This result is very relevant as it indicates a reduced intestinal inflammation in the treated groups compared to the control.

The study assessed the weekly bowel evacuation average, revealing that in Groups 1 and 2, the numbers were higher at the study's conclusion compared to the control group. This suggests a potential overall positive impact of probiotics on bowel movements during administrations, without inducing extreme effects such as diarrhea or constipation. Instead, the probiotics seemed to facilitate a more regular and frequent bowel movement, offering potential benefits for individuals with this condition. In the next study, it would be beneficial to evaluate the number of days before the first bowel evacuation after surgery and compare with the control group.

Data collected on Bristol scale scores gave not homogeneous results between and within groups

and it is difficult to interpret the results. P value around 0.0625 in a lot of Wilcoxon's tests, could suggest that would be beneficial to the possibility of statistically significant results by increasing the number of patients.

The reduction in infections and inflammation after colorectal surgery can be coupled with faster recovery of bowel movements after surgery, and it can also significantly contribute to a reduction in the cost of hospital stays through various interconnected mechanisms. In a recent review which includes several data on trials testing hospital stay in patients treated with probiotics, the use of probiotics resulted in a little to no difference in hospital stay after colorectal surgery.[21]

For example, shortened hospital stay directly translate to cost savings by reducing room charges, nursing care, and ancillary services; decreased postoperative complication rates such as infections and inflammatory responses minimize additional medical interventions, imaging studies, and extended monitoring; lower antibiotic usage not only lowers pharmaceutical costs but also reduces the risk of antibiotic-related complications and antibiotic resistance; less intensive care requirement; minimized readmissions due to postoperative issues; optimized resource utilization including hospital beds, staffing, and surgical suites; improved patient satisfaction which are more likely to adhere to postoperative care instructions, reducing the risk of complications and readmissions.

### Limitations of the study

The study's limitations are associated with certain confounding factors, such as the presence, type, and severity of the tumor, age group variations, overweight conditions, and diverse home diets that may influence the microbiota and consequently impact treatment outcomes. To enhance result standardization in the trial, a more homogeneous population and an increased number of participants would be beneficial. The optimal probiotic regimen, including specific strains, dosages, and duration of administration, remains an area of ongoing research. Variability in study designs and outcomes has led to challenges in establishing standardized guidelines for probiotic use in colorectal surgery. Despite numerous studies exploring the effects of probiotics on various parameters, clear conclusions remain elusive due to the presence of biases, a challenge shared with our study, acknowledging its compatibility with our identified limitations.

Future endeavors should prioritize well-designed, randomized controlled trials, specifically honing in on outcomes following invasive abdominal procedures. These trials are essential to furnish more precise and conclusive evidence pertaining to the efficacy and safety of perioperative and postoperative oral administration of probiotics in this particular context. Beyond physiological aspects, consideration of the psychological impact of neoplastic disease, coupled with surgical outcomes and the pace of recovery to resume normal life, warrants attention.

## Conclusions

The primary goals of this pilot study have been successfully accomplished, providing us with vital insights and promising preliminary data for a more comprehensive investigation into the effects of the same products. Subsequent trials could delve into the potential positive impacts of perioperative probiotic administration in colorectal surgery on a broader scale, involving interdisciplinary collaboration with other medical disciplines. This holistic approach aims to thoroughly assess the overall impact on patients' well-being.

## References

**1.** Madsen K, Kornish A, Soper P, McKaigney C, Jijon H, Yachimec C, *et al.* Probiotic bacteria anhance murine and human intestinal epithelial barrier function. Gastroenterology 2001;121:580–91.

**2.** Macfarlane GT, Cummings JH. Probiotics, infection and immunity. Curr Opin Infect Dis 2002;15:501–6.

**3.** Mizuta M, Endo I, Yamamoto S, Inokawa H, Kubo M, Udaka T, *et al.* Perioperative supplementation with bifidobacteria improves postoperative nutritional recovery, inflammatory response, and fecal microbiota in patients undergoing colorectal surgery: a prospective, randomized clinical trial. Biosci Microbiota Food Health 2016;35:77–87.

**4.** Nomura T, Tsuchiya Y, Nashimoto A, Yabusaki H, Takii Y, Nakagawa S, *et al.* Probiotics reduce infectious complications after pancreaticoduodenectomy. Hepatogastroenterology 2007;54:661–3.

**5.** Okazaki M, Matsukuma S, Suto R, Miyazaki K, Hidaka M, Matsuo M, *et al.* Perioperative synbiotic therapy in elderly pa-

tients undergoing gastroenterological surgery: a prospective, randomized control trial. Nutrition 2013;29:1224–30.

**6.** Peitsidou K, Karantanos T, Theodoropoulos GE. Probiotics, prebiotics, synbiotics: is there enough evidence to support their use in colorectal cancer surgery? Dig Surg 2012;29:426–38.

**7.** He D, Wang HY, Feng JY, Zhang MM, Zhou Y, Wu XT. Use of pro-/synbiotics as prophylaxis in patients undergoing colorectal resection for cancer: a meta-analysis of randomized controlled trials. Clin Res Hepatol Gastroenterol 2013;37:406–15.

**8.** Carlini M, Grieco M, Spoletini D, Menditto R, Napoleone V, Brachini G, *et al.* Implementation of the gut microbiota prevents anastomotic leaks in laparoscopic colorectal surgery for cancer:the results of the MIRACLe study. Updates Surg 2022;74:1253–62.

**9.** Marcellinaro R, Grieco M, Spoletini D, Troiano R, Avella P, Brachini G, *et al.* How to reduce the colorectal anastomotic leakage? The MIRACLe protocol experience in a cohort in a single high-volume centre. Updates Surg 2023;75:1559–67.

**10.** Pu W, Zhang H, Zhang T, Guo X, Wang X, Tang S. Inhibitory effects of Clostridium butyricum culture and supernatant on inflammatory colorectal cancer in mice. Front Immunol 2023;14:1004756.

**11.** Wang T, Cai G, Qiu Y, Fei N, Zhang M, Pang X, *et al.* Structural segregation of gut microbiota between colorectal cancer patients and healthy volunteers. ISME J 2012;6:320–9.

**12.** Ambalam P, Raman M, Purama RK, Doble M. Probiotics, prebiotics and colorectal cancer prevention. Best Pract Res Clin Gastroenterol 2016;30:119–31.

**13.** Jacouton E, Chain F, Sokol H, Langella P, Bermúdez-Humarán LG. Probiotic strain Lactobacillus casei BL23 prevents colitis-associated colorectal cancer. Front Immunol 2017;8:1553.

**14.** Ariyoshi T, Hagihara M, Takahashi M, Mikamo H. Effect of Clostridium butyricum on Gastrointestinal Infections. Biomedicines 2022;10:483.

**15.** Bin Lan, Yang F, Lu D, Lin Z. Specific immunotherapy plus Clostridium butyricum alleviates ulcerative colitis in patients with food allergy. Sci Rep 2016;6:25587.

**16.** Yang Y, Xia Y, Chen H, Hong L, Feng J, Yang J, *et al.* The effect of perioperative probiotics treatment for colorectal cancer: short-term outcomes of a randomized controlled trial. Oncotarget 2016;7:8432–40.

**17.** Akatsu H, Iwabuchi N, Xiao JZ, Matsuyama Z, Kurihara R, Okuda K, *et al.* Clinical effects of probiotic Bifidobacterium longum BB536 on immune function and intestinal microbiota in elderly patients receiving enteral tube feeding. JPEN J Parenter Enteral Nutr 2013;37:631–40.

**18.** Singh J, Rivenson A, Tomita M, Shimamura S, Ishibashi N, Reddy BS. Bifidobacterium longum, a lactic acid-producing intestinal bacterium inhibits colon cancer and modulates the intermediate biomarkers of colon carcinogenesis. Carcinogenesis 1997;18:833–41.

**19.** Laparra JM, Olivares M, Gallina O, Sanz Y. Bifidobacterium longum CECT 7347 modulates immune responses in a gliadin-induced enteropathy animal model. PLoS One 2012;7:e30744.

**20.** Lewis SJ, Heaton KW. Stool form scale as a useful guide to intestinal transit time. Scand J Gastroenterol 1997;32:920–4.

**21.** An S, Kim K, Kim MH, Jung JH, Kim Y. Perioperative Probiotics Application for Preventing Postoperative Complications in Patients with Colorectal Cancer: A Systematic Review and Meta-Analysis. Medicina (Kaunas) 2022;58:1644.

**22.** Stoeva MK, Garcia-So J, Justice N, Myers J, Tyagi S, Nemchek M, *et al.* Butyrate-producing human gut symbiont, Clostridium butyricum, and its role in health and disease. Gut Microbes 2021;13:1–28.

**23.** Zhang JW, Du P, Gao J, Yang BR, Fang WJ, Ying CM. Preoperative probiotics decrease postoperative infectious complications of colorectal cancer. Am J Med Sci 2012;343:199–205.

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*Conflicts of interest*

The authors certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

*Authors' contributions*

Elisabetta Radice: study design, data collection, manuscript writing and final approval. Galyna Shabat: corresponding author, participation at manuscript writing, manuscript final approval. Elisa Martello: data analysis and interpretation, manuscript writing and final approval. Olexii Potapov, Giorgia Meineri, Paolo Risso, Francesco Di Pietro: data analysis, manuscript final approval. Francesco Olandese: data collection, analysis and interpretation, manuscript final approval. All authors read and approved the final version of the manuscript.

*History*

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SUPPLEMENTARY DIGITAL MATERIAL 1

## Questionnaire and data collection template (in Italian)

**SCHEDA PER LA RACCOLTA DATI PAZIENTI**

Nome: _____ Cognome: _____

Indirizzo: _____

e-mail: _____

telefono: _____

Età: _____ Sesso: _____ peso: _____ altezza: _____ BMI: _____

Patologia Maligna: SI/NO

Sede: _____

Stadio: _____

Patologie concomitanti: _____

Farmaci preoperatori: _____

**SCHEMA TERAPEUTICO SOLO PER I DUE GRUPPI DI PAZIENTI CON IL SUPPLEMENTO/PROBIOTICO**

1) 30 gg prima dell'intervento:

Gruppo 1 Supplemento con *Bifidobacterium longum* ES1 cpr: 1x3 al di ai pasti per 30gg

Gruppo 2 Supplemento con *Clostridium butyricum* CBM588® orosolubile: 1 bustina al di (ore 10 lontano dai pasti) per 30 giorni

2) Sospensione del supplemento per 3 giorni (giorno dell' intervento e due giorni dopo)

3) Dopo 3 giorni dall'intervento:

Gruppo 1 Supplemento con *Bifidobacterium longum* ES1 cpr 1x3 al di per 30 giorni

Gruppo 2 Supplemento con *Clostridium butyricum* CBM588® orosolubile: 1 bustina orosolubile ore 10 e continuare per 30 giorni

**ESAMI DI LABORATORIO**

Esami pre-operatori (30 giorni prima dell' intervento):

Emocromo _____ PCR _____ Calprotectina fecale _____

Esami 10 giorni dopo l'intervento:

Emocromo _____ PCR _____ Calprotectina fecale _____

Esami 30 giorni dopo l'intervento:

Emocromo _____ PCR _____ Calprotectina fecale _____

**QUESTIONARIO pre-operatorio (30 giorni prima dell' intervento chirurgico)**

Scala di Bristol (PUNTEGGIO 1-7)


qualità di vita

dolore addominale: SI/NO

numero di evacuazioni : _____

gonfiore addominale: SI/NO

**QUESTIONARIO post-operatorio (dopo 30 giorni dopo l'intervento chirurgico)**

Scala di Bristol (PUNTEGGIO 1-7)

qualità di vita

dolore addominale: SI/NO

numero di evacuazioni : _____

gonfiore addominale: SI/NO

## Informed consent (Italian), PDF format attached