KNOWLEDGE AND ACCEPTANCE OF FUNCTIONAL FOODS: A PRELIMINARY STUDY ON INFLUENCE OF A SYNBIOTIC FERMENTED MILK ON ATHLETE HEALTH

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ABSTRACT: Functional foods (FFs) provide benefits beyond basic nutrition and play a role in reducing the risk of certain diseases. FFs may also be designed to improve performances and healthy status of athletes. This study investigated young Italian athletes awareness and consumption of FFs, and the effect of a synbiotic fermented milk enriched with probiotics and oat bran, on gut microbiota and on upper respiratory tract. Data collection from the knowledge and acceptance study was performed through a questionnaire, developed and administered on 164 athletes. The effects of a synbiotic functional food were studied by a preliminary intervention study on 10 healthy volunteers following an intense gym-training program. Faeces were collected for microbiological and molecular analysis before and at the end of the 4 weeks consumption, while saliva and spit were collected at the same time points for lipid peroxidation and secretory IgA detection. Data obtained from the knowledge and acceptance study indicated that only 17% of the young Italian athletes were familiar with the term “functional food” although the respondents were aware of the link between diet and health. In the human trial, the synbiotic group showed a significant increase of Lactobacillus spp. and Bifidobacterium spp., a significant improvement in intestinal regularity, ease of defecation and in upper respiratory symptoms after the consumption. The data support the hypothesis that an adequate synbiotic supplementation can improve gut health, oxidative status and mucosal immunity with beneficial effects on wellbeing of active people.

KEY WORDS: Athletes, Functional Food, Gut Health, Immune Markers, Prebiotics, Probiotics

INTRODUCTION

Nowadays, health care costs of some chronic diseases that are associated with diet and lifestyle raise an important interest to governments and consumers. Among other initiatives, this prompt interest on dietary improvements includes functional foods. Beside the basic nutritional needs, functional foods provide also additional physiological benefits. The beliefs that “eating healthy/right/well” improves life quality, supported by nutritional science development, have increased the interest in functional foods (Ding et al., 2015). Consumer interest in the relationship between diet and health has increased the demand for information on functional foods. It has been observed that modern consumers are increasingly interested in their personal health, expecting the food they eat to be tasty and healthy, and in the same time capable of preventing illnesses and reducing the diseases risk.

The market for functional foods is expanding, illustrating the need for better understanding of consumers’ decision-making for these foods to guide food policy and financial decisions. Functional foods/foods for health are an important part of an overall healthful lifestyle that includes a balanced diet and physical activity. The newest definition for “functional foods” was revised to: “Natural or processed foods that contain known or unknown biologically-active compounds; which, in defined, effective non-toxic amounts, provide a clinically proven and documented health benefit for the prevention, management, or treatment of chronic diseases” (Martirosyan and Singh, 2015).

The food market gives the consumer innumerable choices
of food from all over the world. In the last years, scientists have changed the consideration of food as source of energy and nutrients into the action of biologically active food components on human health. Also the consumer interest about the active role of food in well-being and life-prolongation has been increased. Recently, a wide range of functional foods/ingredients was designed and produced in all over the world; these new products include probiotic, prebiotic and synbiotic foods, but also foods enriched with antioxidants, carotenoids, fatty acids, flavonoids, phytoestrogens, phenolic acids, minerals, vitamins and fat, sugar and salt-reduced foods (Coman and Cresci, 2014; Homayouni et al., 2012).

The wide number of health objectives of functional foods include enhancing weight loss, improving joint health, increasing muscle and bone strength, decreasing risk factors for non-communicable diseases (autoimmune and heart diseases, stroke, cancers, diabetes, obesity, osteoporosis, Alzheimer’s disease), enhancing gastrointestinal functions, modulating redox and antioxidant systems, decreasing wrinkles and improving mood/behaviour and physical performances (Aiello, 2011).

Probiotics are one of the most accepted functional foods in form of fermented milk and they are widely produced. The uses of prebiotics and probiotics together, known as synbiotics, have a positive effect, since prebiotics promote the growth and activities of probiotics. It is clear from literature that new kinds of fermented milks containing various nutrients are being tested as curatives for specific diseases and are approaching medical food effectiveness in conventional food format and will continue to be introduced to the food supply. Functional dairy products offer requirements, benefits to health that are strengthened by the addition of probiotics as well as by certain types of soluble fibers known as prebiotics (Coman et al., 2013). Suggested and demonstrated health benefits that have been associated with the consumption of probiotics and/or prebiotics include improvement of intestinal tract health enhancing the epithelial barrier, enhancement of immune system and bioavailability of nutrients, alleviation of allergic symptoms, reduction of diarrhea incidence, lactose intolerance, cholesterol level and risk of specific cancers (such as colon cancer) (Coman and Cresci, 2014; Roberfroid et al., 2010).

Studies on athletes and probiotics/prebiotics effect on sport performance are extremely limited and indecisive, no evidence on performance benefits having been found so far. Reasons given for using probiotics and prebiotics are to maintain and promote health, feel better, reduce the risk of disease, and treat a specific medical condition such as gastroesophageal reflux disease, irritable bowel syndrome, and constipation (Nichols, 2007), but the incidence of probiotic use by athletes is not known. In addition, athletes practicing different types of sports need special nutrition before, during and after training to be in a healthy state together with complete fitness and performance. Supplementation of special food products formulated by nutritionists may be the solution.

Different health risks for athletes have been reported that may be mainly related to oxidative stress, dehydration and gastrointestinal tract (GIT) effect during strenuous exercises (Al-Okbi et al., 2014).

Production of innovative functional food is crucial for athletes and this required first studying nutritional status, physiological and biochemical changes occur due to practicing different sports and training. Also, suitable functional foods must be designed to be suitable before, during and after training. Athletes undertaking severe and intense exercises may suffer from an increased risk of upper respiratory tract infections (URTIs) (exercise-induced perturbations in cellular and humoral immunity, with acute and chronic changes in secretory IgA) and gastrointestinal symptoms (disturbed mucosal surfaces with an increased risk of common gut problems such as nausea, bloating, cramping, pain, diarrhea or bleeding) (West et al., 2009; Pyne et al., 2015). It is also known that intensive exercise and strenuous physical training causes a rise in chronic exhaustion and a decrease in athlete’s efficiency (Salarkia et al., 2013). For these reasons there is a strong interest among athletic community on probiotic’s benefits to reduce susceptibility to URTIs and GI illness.

Some probiotic strains may confer benefits to athletes and may represent an effective supplementation for them furnishing an appropriate antioxidant barrier essential for preventing dangerous levels of oxidative stress (West et al., 2009; Gleeson et al., 2011; Martarelli et al., 2011; Lamprecht et al., 2012). In order to increase the chances of success in functional foods market, consumer acceptance has been identified as the decisive factor. Increasing awareness of functional foods would have many health benefits such as reducing the incidence of non-communicable diseases. The first aim of this study was to investigate consumer awareness and consumption of functional foods among young Italian athletes. Nutritional practices that promote good health and optimal athletic performance are of interest to athletes, coaches, exercise scientists and dieticians. Probiotics and prebiotics may benefit athletic performances indirectly by maintaining GI function and health, preventing the immunosuppressive effects of intense exercise, reducing susceptibility to illness and increase antioxidant levels and neutralize the effects of reactive oxygen species (West et al., 2009; Martarelli et al., 2011). Related to this matter, the second aim of this study was to explore the effect of symbiotic functional foods, fermented milk enriched with probiotics and oat bran as prebiotic substrate, on gut health, incidence of URTIs and immune markers in athletes.

**MATERIALS AND METHODS**

**Survey on functional foods**

**Subjects and data collection**

Data collection was performed through a questionnaire, developed and administered to a sample of 164 young athletes aged 6 - 17 years, afferent to the Department of Sport Medicine at Jesi Hospital (AN, Italy). The sample thus included
individuals of both sexes that were undergone to sporting visit and resident to Jesi or nearby. Survey was administrated by previously trained interviewers (undergraduate students in nutrition). The interviewers used a questionnaire tested in a pilot study.

**Ethical aspects, recruitment and randomization**

All subjects provided written informed consent given by the parents because they are all minors prior to participating in this investigation. This study was conducted according to the guidelines of the Declaration of Helsinki for Research on Human Subjects 1989 and the General Direction of Area Vasta 2 approved the study protocol.

**Questionnaire**

The questions were partly based on knowledge and consumption of conventional and functional foods (FFs). The questionnaire was designed with eight sections, the respondents were asked about their food behaviours, the influence of the media to purchasing a food product, their knowledge on functional foods, the grade of consumption, the health benefits, and statements about functional and conventional foods. The answers were multiple choices.

**Intervention study**

**Subjects**

A total of 10 healthy adult human volunteers following an intense gym-training program (7 females and 3 males) with ages ranging from 20 to 45 years were included in the study. Inclusion criteria were healthy persons (chronic diseases controlled with proper medications were allowed), age and acceptance of the study protocol. Antibiotics were not allowed two months before and during the intervention. Background information was collecting from the volunteers through screening interviews, regarding general health, medications, and manner of living. The subjects followed their habitual diet during the study. The exclusion criteria were critical illness, inflammatory bowel disease, lactose intolerance, frequent GI disorders or metabolic disease. They provided written informed consent and the guidelines of Declaration of Helsinki for Research on Human Subjects 1989 was followed during the trial.

**Study food product**

The food product used in the study was a synbiotic fermented milk containing the mixture of two probiotic strains (1:1, about 10^9 CFU per strain, per portion), *Lactobacillus rhamnosus* IMC 501^*T* and *Lactobacillus paracasei* IMC 502^*T* (Synbiotec S.r.l., Camerino, Italy) (Verdenelli et al., 2009) in the presence of oat bran fibre as prebiotic source (Coman et al., 2013).

The fermented milk samples were produced in a YO&MI YZ-150 yogurt machines (Plastitalia Group, Firenze, Italy), using pasteurized whole milk, starters, probiotics and oat bran flour. The control product consisted of fermented milk without the probiotic and prebiotic enrichment. During the intervention period the subjects were instructed to consume during the day one portion of approximately 200 ml of fermented milk. The use of test products was recorded weekly, and the investigator checked the records at each visit. Volunteer compliance was determined by verbal assessment by the investigator.

**Study design**

Volunteers were randomly distributed into two groups: control group (n=5) and synbiotic group (n=5). Both study subjects and investigators were blinded to the nature of the products. The study was performed by a 4 weeks intervention period. During the intervention period, the synbiotic group consumed daily a synbiotic-fermented milk (200ml), while the control group consumed during the 4 weeks fermented milk without prebiotics and probiotics.

**Samples collection**

Faeces were collected at the morning and frozen immediately in a portable -20°C freezer, for microbiological and molecular analysis, before and at the end of the 4 weeks intervention. At the same time points buccal cells and spit were also collected. The buccal cells were took with a swab placed between the cheek and teeth for up to five minutes for the lipid peroxidation, while the spumt was took into a sterile container and centrifuged for 5 minutes at 800×g and frozen at -80°C until secretory IgA analysis were performed.

**Microbial analysis**

**Stool DNA Extraction Protocol**

DNA extraction from faecal samples was performed using a modified benzyl chloride method (Zhu et al., 1993) after a cleaning step in order to remove the undigested particles from the faeces.

**Quantitative Real-Time PCR**

A Real-Time quantitative PCR (qPCR) procedure, using specific primers as reported by Nasuti et al. (2016), was used for the quantification of selected bacterial groups: *Lactobacillus* spp., *Bifidobacterium* spp., Enterobacteriaceae, *Clostridium cocoides-Eubacterium rectale* group, *Staphylococcus* spp., *Bacteroides-Prevotella-Porphyromonas* spp. All PCR experiments were carried out in triplicate.

**Lipid oxidation in buccal cells**

The samples of saliva cells treated as previously described, at a protein concentration of 0.1 mg/ml, were homogenized in PBS and used for fluorescence measurements with diphenyl-1-pyrenolphosphine (DPPP). Methanol solubilised DPPP was used at a final concentration of 1μM. The cell suspension was incubated at 37°C for 3 min in the dark. Cells were washed three times with PBS and suspended in the same buffer. At the times indicated, fluorescence intensities of the samples were measured on a Hitachi 4500 spectrofluorometer using 351 and 380 nm as excitation and emission wavelengths, respectively.
**New functional foods for athlete health**

**Determination of sIgA in saliva**

Secretory IgA (sIgA) consist of two IgA monomers joined by the J-chain and an additional secretory component. It is secreted in plasma cells located in the lamina propria of mucosal membranes. This means that lack of serum IgA does not necessarily correlate with a lack of sIgA. It plays a major role in the alternative complement pathway and in activating inflammatory reactions. The determination was performed in duplicate using a sIgA ELISA kit (Immunodagnostik AG, Bensheim, Germany).

**Illness symptoms**

Subjects recorded symptoms of GI, URTI and lower respiratory illness at the end of the synbiotic consumption respect to their initial status using a Wisconsin Upper Respiratory Symptom Survey (0-no symptoms, 3-mild, 5-moderate, 7-severe). Briefly, symptoms of GI illness included nausea, vomiting, diarrhoea, abdominal pain bloating, flatulence, stomach “rumbles” and loss of appetite. URTI symptoms included throat soreness, sneeze, a blocked or runny nose and cough. Lower respiratory illness symptoms including coughing with chest congestion and/or wheezing.

**Bowel habits**

The overall assessment of bowel well-being was also recorded. Primary outcome measures were intestinal regularity and stool volume, while as secondary outcome measures were also investigated: ease at defecation, bloating, constipation, abdominal pain, intestinal cramps, feeling of incomplete defecation, incontinence and halitosis. The procedures applied by Silvi et al. (2014) were followed and stool consistency was defined by the Bristol Stool Form Scale (Lewis and Heaton, 1997).

**Health-related quality of life**

HRQoL of subjects was assessed by self-administration of Psychological General Well-Being Index (PGWBI) (Dupuy, 1984) that is a general questionnaire measuring psychological well-being and distress and is composed of twenty-two items which constitute six dimensions (anxiety, depression, self-control, positive well-being, general health and vitality). The scores of all dimensions can be summarized to provide a global score. The score range from 0 to 100 (best).

**Statistical analysis**

The results are expressed as mean ± standard deviation (SD) or error (SE). Statistical significance of the differences between the synbiotic group and the control group was analyzed using Student t-test. Significant differences between mean values were determined by Tukey’s test after one-way analysis of variance ANOVA (1-way ANOVA) using GraphPad PRISM 5.1 program. A P-value less than 0.05 was considered statistically significant. For the bowel habits, where P<0.05 reflects significant difference, a Four Multiple Comparison test was used.

**RESULTS**

**Survey on functional foods**

**Questionnaire**

Taking into account consumers’ general attitudes towards FFs, our research tested the level of knowledge of such products displayed by young athletes. The data analysis shows that they were not greatly informed about the concept of FFs. The 80.4% of the subjects answered that they never heard the term functional foods and the 83% didn’t know what a functional food is. Regarding the analysis of available information flow to consumers, the results show that the main sources from which respondents obtain information on FFs are product labels (76.2%) and friends and family (56.7%), followed by farmer (49.3%) and brand/industry (48.7%). The information flow by the radio was the lowest respect to all the other sources (11.5%) (Figure 1A).

**FIGURE 1. Survey on Functional foods with two possible answers, yes (■) and no (■). Question A: “Is this your main information sources on FFs?”; Question B: “Do you know this kind of FFs?”**

Within a list of FFs shown to the subjects respect, the most popular product known by the young athletes was the sport drinks (95.1%), followed by probiotic yogurts (86.6%), while the less known product was the spread added with calcium (67.1%) (Figure 1B).

Taking in consideration the consumption of these products, the 67.1% of respondents stated that they had never consumed Omega-3 milk, while 29.9% were occasional consumers of the sport drinks, followed by those consuming probiotics occasionally (26.6%). The 9.1% of participants consumed
A high percentage of athletes consume daily probiotic yogurts compared to the other percentages of the listed functional foods (Figure 2).

Regarding the question about beneficial characteristics of functional foods (Figure 3), participants answered that if the claim was scientifically proved, the 74.3% would like having a protection against tumors followed by cardiovascular prevention, instead skin improvement was considered by being less important (1.21%).

Finally, we asked the subject’s parents to indicate the degree of agreement or disagreement on some statements related to functional and conventional foods. The 50% of the subjects agreed with the statement about the benefits of FFs and the capacity to prevent some diseases, 50% agreed to pay more for a product that is good for the health, 78% totally disagreed that FFs are mean just for elderly and sick people, 32.2% doesn’t know if FFs taste worse than a conventional food (data not shown).

**Intervention study**

**Microbial analysis**

Table 1 shows the faecal microbiota composition in synbiotic and control group before and at the end of the intervention period. Lactobacilli and Bifidobacteria had a significantly higher value in the synbiotic group after intervention, while no differences were registered in the control group.

**Lipid oxidation in buccal cells**

In order to evaluate the effect on lipid peroxidation, the probe DPPP was employed since it reacts with hydroperoxides in the cell membrane yielding a measurable fluorescent product. The lipid peroxidation value was measured before and after the intervention period and outcome was expressed as percentage difference. As shown in Table 2, the control group shows a significant increase of the percentage of lipid hydroperoxide (22.2%) with respect to the synbiotic group that stayed stable (1.72%).

**Determination of sIgA in saliva**

The results indicate a significant increase of secretory IgA in daily probiotic yogurts respect to the other percentages of the listed functional foods (Figure 2).

Regarding the question about beneficial characteristics of functional foods (Figure 3), participants answered that if the claim was scientifically proved, the 74.3% would like having a protection against tumors follow by cardiovascular prevention, instead skin improvement was considered by being less important (1.21%).

Finally, we asked the subject’s parents to indicate the degree of agreement or disagreement on some statements related to synbiotic group after the dietary intervention (Table 2), this relate with the increase of mucosal immunity.

**Illness symptoms, Bowel habits and Health-related quality of life**

Table 3 summarizes the change in frequency of gastrointestinal, upper respiratory and general symptoms at the end of intervention period. A significant improvement in intestinal regularity, ease of defecation and stool consistency has been registered in synbiotic group respect to control group. Also a lower incidence of upper respiratory symptoms

**TABLE 1. Faecal microbiota composition of synbiotic and control group before and after intervention period.** *Significantly different (*P*<0.05) from the initial value and # respect to the control group after the intervention by One-way ANOVA test.

<table>
<thead>
<tr>
<th>Bacterial group</th>
<th>Log CFU/g of faeces</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Synbiotic group</td>
<td>Control group</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Before intervention</td>
<td>After intervention</td>
<td>Before intervention</td>
<td>After intervention</td>
</tr>
<tr>
<td><strong>Bacteroides-Prev.-Porphyrib. spp.</strong></td>
<td>7.63±0.08</td>
<td>7.45±0.00</td>
<td>7.23±0.04</td>
<td>7.44±0.22</td>
</tr>
<tr>
<td><strong>Staphylococcus spp.</strong></td>
<td>4.92±0.05</td>
<td>4.84±0.02</td>
<td>5.11±0.05</td>
<td>5.27±0.00</td>
</tr>
<tr>
<td><strong>Cl. coccoides-Eubact. rectale group</strong></td>
<td>10.52±0.06</td>
<td>10.19±0.14</td>
<td>10.27±0.04</td>
<td>10.26±0.03</td>
</tr>
<tr>
<td><strong>Lactobacillus spp.</strong></td>
<td>4.09±0.02</td>
<td>5.53±0.09*</td>
<td>4.22±0.18</td>
<td>3.79±0.06</td>
</tr>
<tr>
<td><strong>Bifidobacterium spp.</strong></td>
<td>6.23±0.09</td>
<td>7.75±0.01*</td>
<td>6.16±0.01</td>
<td>6.32±0.09</td>
</tr>
<tr>
<td><strong>Enterobacteriaceae</strong></td>
<td>4.23±0.02</td>
<td>4.09±0.29</td>
<td>3.77±0.06</td>
<td>4.12±0.02</td>
</tr>
</tbody>
</table>
was noticed in synbiotic group respect to control group. Stool consistency was generally evaluated of type 3 or 4 and presented significant difference between the synbiotic and control group. The PGWBI score of synbiotic group didn’t differ from control group. The synbiotic consumption was very well tolerated, and no side effects were registered.

**DISCUSSION**

The concern regarding the human health became crucial in the last years and the consumers are more careful about their diet knowing food having a positive impact on improvement and maintenance of health status. With the emphasis being on cost-effective health care, the significance of dietary changes for improved health and disease prevention is widely recognized and it is also important to better understand the consumer knowledge, attitudes and perceptions on functional foods. Data from this study indicated that only 17% of the young Italian athletes interviewed were familiar with the term "functional food" despite the fact that the majority of the subjects who participated were aware of the link between diet and health. So, attention to diet - health connection is accepted as the initial phase in stimulating enthusiasm for obtaining knowledge concerning good dieting decisions.

Results of the explorative analysis reveal that young Italian consumers are confused due to the ambiguity of what FF products are, despite having a marked awareness of the link between diet and health and a high level of interest in the nutritional and health aspects of their food choices. Similar findings were found in other studies conducted in other European countries, such as UK, Germany, France, Spain, Netherlands and Sweden (Del Giudice and Pascucci, 2010). The second factor found in the present analysis is the consumer confidence in FFs; it describes whether individuals consider these products as safe and to what extent their belief in the scientific basis underlying the information about FFs health effects. Moreover, some respondents were suspicious of possible harmful effects of FFs; these perceived risks of FFs can be a strong barrier to the consumption of such products.

Additionally, consumers from this study felt that the most reliable source of information regarding FFs could be provided by product labels and friends and family, followed by farmer and brand/industry and the scientists. As part of the continuing effort to promote public health, health professional could thus help in the endeavour of promoting FFs in view of their potential health benefits.

In Italy, there is the availability of a wide variety of FFs on the local market and these products were thus included as options in the questionnaire. Subjects were familiar with the products “probiotic yogurt”, “sport drinks” and “enriched breakfast cereal” and they were also aware of their potential health benefits. However, level of awareness regarding “Omega-3 milk”, “vitamin-enriched juices”, “spread added with calcium” and “low cholesterol butter” was relatively lower among the subjects. Similarly, respondents were confident
about the health benefits of “probiotic yogurt” especially because of the wide variety of products on the shelves as well as because of the massive promotion of these products for their beneficial effects. The same behaviour could be ascribed to “enriched breakfast cereals” that are massively present on the local market. Moreover, the high level of awareness to “sport drinks” could be attributed not only to the wide range of products available on the local market, but also to the young population involved in the survey very attracted to this type of product. However, the beneficial effects of “Omega-3 milk”, “vitamin-enriched juices”, “spread added with calcium” and “low cholesterol butter” were not familiar to them, that could be ascribed to a low promotion of food containing these bioactive components.

Previous studies have suggested that the acceptance of FFs depends on the basic product that serves as a carrier for the functional ingredient (Siro et al., 2008). Our results indicate that Italian consumers are more familiar with fortified cereals, probiotic yogurt, and vitamin juice as suitable and credible carriers of FFs, showing less interest in “spreads added with calcium” and “low-cholesterol butter”.

As for the satisfaction factor, the present findings show that consumers do not perceive FFs less tasty than conventional products, since the taste is the main organoleptic characteristic used in the food consuming and purchasing. The results of this study also show that consumers are available to accept high price for food able to improve their health status. Anyway difficult availability, lacking in labelling information easy to understand and the difficulty to distinguish FFs from conventional foods can be considered the main obstacles to purchase these products. Finally, in terms of marketing strategies, FFs need to be promoted with the aim of making them much more visible and recognizable to final consumers in order to avoid confusion with other generic health foods (such as light or diet products). Since the present analysis highlighted that the perception of healthiness is the main factor affecting consumer attitudes toward FFs, food companies should focus their marketing strategies on reinforcing FF properties and trying to communicate them clearly and less scientifically. In this regard, and in line with findings elsewhere (Annunziata and Vecchio, 2011), the role of labelling should be strengthened.

The present study allowed to come to a better understanding of young Italian athletes knowledge on ingredient functionality that can be used for successful product positioning of new FF products. Consumer acceptance is a complex matter, more researches are needed to scrutinize the different influence factors related to different product categories within the FF sector.

The results of the survey on FFs consumer awareness were the basis for a human trial test showed the beneficial effects of a synbiotic fermented milk on gut health, incidence of URTIs and immune markers in athletes. Consumption of dairy products, including yogurt, by European consumers is considerable and dairy products currently lead the probiotic market.

The consumption of the synbiotic fermented milk tested in this study, containing the two probiotic strains L. rhamnosus IMC 501° and L. paracasei IMC 502° and the prebiotic substrate oat bran, determined a significant increase in lactobacilli and bifidobacteria in the intestine of the synbiotic group subjects at the end of the consumption period. A mechanism of action for this effect was not investigated as part of this study, but we would speculate that the increase in bifidobacteria observed may be attributed to the intestinal pH decrease induced by lactic acid or other fermented products produced by L. rhamnosus IMC 501° and L. paracasei IMC 502° (Verdenelli et al., 2011). The probiotics may metabolize luminal components to generate a substrate that may be a preferred fuel source or create a preferred physical environment, such as localized pH, advantageous for the growth of Bifidobacterium. Even the presence of oat bran could determine the increase of bifidobacteria due to is known bifidogenic effect (Abnous et al., 2009).

A significant improvement in intestinal regularity, stool consistency and ease of defecation, has been registered in synbiotic group after the intervention respect to control group. Also a tendency to improvement in upper respiratory symptoms was registered in synbiotic group respect to control group. These results are very interesting because the recruited subjects had a normal bowel function before the intervention study; thus, the synbiotic diet improves bowel habits without any side effects. Taken together, consumption of synbiotic fermented milk is likely to improve bowel habits through stimulating the growth and activity of bifidobacteria and lactobacilli.

Recently, the biological roles of lipid peroxidation products have received a great deal of attention not only for elucidating pathological mechanisms but also for practical clinical applications as biomarkers. Lipids are susceptible to oxidation and lipid peroxidation products are potential biomarkers of oxidative stress status in vivo and its related diseases. In the present study the percentage of lipid hydroperoxide increased in the control group, while in the synbiotic group remain stable, indicating a protective effect of the synbiotic consumption in athletes. In the light of this, the ability of synbiotic product to reduce the oxidative stress in active people was investigated. Normally, muscular cells have an efficacious defence system to control the overproduction of reactive species. Indeed, endogenous or dietary antioxidants play a protective role, being capable of scavenging free radicals, and therefore they may prevent muscle damage. Moreover, several studies have shown that training results in increased activity of antioxidant defence and also that dietary supplementation with antioxidants has favourable effects on peroxidation processes after exercise. However, if reactive oxygen species (ROS) production is exaggerated and/or muscle cell ability to inactivate such reactive species is reduced, cell undergoes to free radicals damage, despite antioxidant system. Indeed, ROS can attack any organic substrates thus producing hydroperoxides. Because their relative stability and their good oxidant capacity,
hydroperoxides are detectable in biological fluids. In the present study a new approach to measure lipid hydroperoxides using the mucosal buccal cells was used. This is a rapid and no invasive method that could be used in different situation and group of people. The data from the study showed a significant increase of lipid hydroperoxides in control group at the end of dietary intervention, while no differences were registered for synbiotic group before and after the intervention. The authors can speculate that the synbiotic-fermented milk could neutralize the effects of reactive oxygen species; even a greater subject’s number is needed. Anyway, the results obtained in this study are in line with those of Martarelli et al. (2011) in which the intake of L. rhamnosus IMC 501 and L. paracasei IMC 502 as a lyophilized powder by athletes exerted strong antioxidant activity.

It is well known that a decreased salivary secretory immunoglobulin A, found in saliva, intestinal secretions, urine and other mucosal fluids, may increase the URTIs incidence in the athletes. The intake of synbiotic-fermented milk has led to significant increase of sIgA in synbiotic group after the dietary intake. This is an important finding because the augmentation of mucosal immunity by probiotics can protect against infection with pathogens that penetrate the mucosa, results in accordance with those of Neville et al. (2008), demonstrating an association between low levels of total secretory IgA and clinical endpoints (increased susceptibility to URTIs) in athletes. The results also showed that the athletes who take probiotics stay healthier overall.

In conclusion the data support the hypothesis that an adequate synbiotic supplementation can improve gut health, oxidative status and mucosal immunity with beneficial effects on health status of active people. Future studies are required to elucidate the mechanisms of action and also the different dose-response in various exercise settings, by designing a study enrolling a higher number of subjects.

REFERENCES


