The Intestinal Microflora, the Immune System and Probiotics.

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Today there is great scientific interest in the functions of the intestine and the action of intestinal microflora. Research on these issues is ongoing all over the world and one of the main issues is how the intestinal microflora influences the immune system and what effect supplementation of probiotics might have on the immune system.

The intestinal microflora and the immune system

There are an enormous number of bacteria colonising our intestines. These bacteria are referred to as " the normal intestinal microflora". The total amount of bacteria colonising our bodies and above all our intestines is ten times higher than the amount of the cells of the body. It follows that 90% of our body cells are microorganisms!
The majority of the bacteria are to be found in the large intestine (approximately 10^{11-12} bacteria/g). The bacterial counts of the small intestine are considerably lower (approximately 10^{4-7} bacteria/ml). The dominating species in the large intestine are bifidobacteria and bacteroides whereas lactobacilli and streptococci dominate the small intestinal flora.
The intestinal microflora have numerous functions the majority of which have yet to be identified. It is a well-established fact that the intestinal microflora influence the digestion and absorption of food, the function of the immune system, peristalsis, production of vitamins such as B-vitamins and influence the turnover of the intestinal epithelial cells. In addition the metabolism of the microflora influences the secretion of hormones.

One of the most interesting issues regarding the intestinal microflora is its interaction with the immune system. The mucosal membrane of the intestines, with an area of approximately 200 m^2, is constantly challenged by the enormous amount of antigens from food and from the intestinal microflora and from inhaled particles that also reach the intestines. It is not surprising therefore that approximately 80 % of the immune system is found in the area of the intestinal tract and are particularly prevalent in the small intestine.

The immune system of the intestine is referred to as GALT (gut-associated-lymphoid tissue). It consists of the Peyer's patches, which are units of lymphoid cells; single lymphocytes scattered in the lamina propria and intra-epithelial lymphocytes spread in the intestinal epithelia.

The immune system of infants is not fully developed. Bacterial colonisation of the intestine is important for the development of the immune system. The immune system has two equally important main functions: (i) to react to antigens that are harmful or (ii) *not* to react to harmless antigens like nutrients and the tissues of the body. Research shows that these functions are interconnected. Microbial stimulation of the immune system decreases the reactivity against harmless antigens (1-3), which is one of the reasons why scientists are interested in the relationship between the composition and activity of the intestinal microflora and the development of allergies. Research shows that the composition of the intestinal microflora in populations of the western world has changed over the decades and differs from the flora of people in developing countries both in adults (4) and in children (5-6). The intestinal flora of Swedish children consists of fewer strains and has a slower turnover than that of the children from Pakistan. This may well cause a decreased bacterial stimulation of the immune system.

The intestine has an important function in working as a barrier against the surrounding world. This barrier is maintained by tight-junctions between the epithelial cells, by production of IgA antibodies and by influencing the normal microbial flora. It is extremely important that only harmless substances are absorbed while the harmful substances are secreted via the faeces.
Studies show that individuals allergic to cow´s milk have defective IgA production (7) and an increased permeability of the intestinal mucosa (8). This results in an increased absorption of macromolecules by the intestinal mucosa (9). The increased permeability is most probably caused by local inflammations due to immunological reactions against the allergen. This damages the intestinal mucosa (10).

What may cause changes in the composition of the intestinal microflora?

During the past century our lifestyle has changed dramatically regarding hygienic measures, diet, standards of living and usage of medical drugs. Today our diet largely comprises industrially produced sterilised food and the use of different kinds of preservatives. Refrigerators and freezers have replaced the natural processes of the fermentation of the food as was used in past times. This has led to a decreased intake of bacteria, particularly lactic acid producing bacteria (11).

The widespread use of antibiotics in healthcare and agriculture, antibacterial substances in toothpaste, deodorants, food etc. is also something new for human kind. We have in so many ways sterilised our environment, which is detrimental to the microflora.

Probiotics

Probiotics can be defined as microorganisms that positively influence health by improving the intestinal microbial balance. The most commonly used probiotics are *Lactobacillus spp, Bifidobacterium spp.* and *Streptococcus spp.* At the beginning of the 20th century the scientist Elie Metchnikoff postulated his hypothesis on the influence of the intestinal microflora on human ageing. He claimed that the putrefactive processes in the intestine led to the formation of toxins that contributed to the degeneration of the body. He proposed that supplementation with lactic acid producing bacteria i.e. probiotic bacteria, in the form of yoghurt decreased the negative effects of putrefactive bacteria and thus reduced the degenerative processes in the body (12).

Today a great deal of research has been undertaken to reveal the effects of probiotics and the results of that research demonstrate that intake of probiotic bacteria appears to have many positive effects on the body. In an article in the September -99 issue of Trends of Immunology Today, regarding the effects of probiotics on the immune system (13) it was suggested that the beneficial effects of probiotic supplementation could contribute to improved digestion and absorption, prevention of cancer, prevention of infections in the gastrointestinal channel, regulation of peristalsis, prevention of degenerative processes such as osteoporosis and arteriosclerosis etc. It seems that Metchnicoff might have been right!

Research shows that the intake of probiotics has a number of effects on the immune system such as increased production of IgA antibodies (14), increased macrophage activity (15)(16) and increased phagocytosis (17). Supplementation of certain bacteria also decreases the number of inflammatory mediators like TNF-α and α -1-antitrypsin, which may indicate that probiotic bacteria have a healing effect on the intestinal mucosal membrane. Lactic acid producing bacteria inhibit growth of putrefactive bacteria due to the production of organic acids, which lowers the intestinal pH. The activity of putrefactive bacteria decreases with a lower pH.

Prebiotics

Prebiotics are complex carbohydrates such as oligo- and polysaccharides, which are not digested by the enzymes of the small intestine, so thus reach the large intestine in a viable form. They are the nutrients for certain bacteria like bifidobacteria. Intake of different kinds of prebiotics reduces the count of putrefactive bacteria and influences the body beneficially in many different ways and it may decrease the risk of cancer (18). Prebiotics improve the stability of probiotic cultures both in the food and the intestinal tract, which in turn improves the effect of the probiotic bacteria. Products with a combination of probiotics and prebiotics appear to be more favourable and are referred to as symbiotics.

Choice of probiotics and bacterial logistics.

The means of transport of probiotic cultures from the mouth to the regions of the stomach or the intestines and the stability of the cultures during storage can best be defined as “bacterial logistics”. There are today various kinds of probiotic formulations. They may be in the form of liquids like yoghurt, with or without the addition of bacterial cultures of intestinal origin. To improve stability, the bacterial cultures should be freeze-dried. The process of freeze-drying makes it possible to introduce them in powder, capsule or tablet form. The use of tablets and capsules in turn improves the chances for the bacteria to survive the passage through the gastric and bile acids where in other formulations the greater part of the bacteria dies.

Most of the pharmaceutically distributed probiotic products are in the form of gelatine capsules. The problem with capsules of this kind is that they usually consist of 10-15% water causing decreased stability and viability in the cultures (19). To ensure stability and proper distribution the best way to pack probiotic bacteria is in tablet form where the tablet is produced with low compression forces. A tablet with regulated disintegration in the gastrointestinal tract will continuously expose the intestinal system to viable bacteria. Traditional methods of manufacturing tablets cannot be used when the tablet contains living bacterial cultures. Most of the disintegration of a tablet/capsule should take place in the small intestine. Depending on the choice of probiotic strain the optimal time of disintegration may vary. The advantage with probiotics produced in tablet form is that the disintegration time can be regulated.
It is easier to influence the microflora of the small intestine with supplementation of probiotics since the bacterial counts in the small intestine are much lower than in the large intestine. Prebiotics like inulin (fructooligo- or polysaccharides) have a greater impact on the large intestine.

Which bacterial culture is the most effective?
There are different ideas as to which bacterial strains are the most effective. The original theory regarding probiotics according to Metchnicoff referred to the use of cultures with food origin like yoghurt and sauerkraut (12).
Today the general scientific belief is that bacteria with intestinal origins are preferable. This follows from the proposition that supplemented probiotic bacteria must be able to colonise the intestinal mucosa if they are to be effective. The composition of the intestinal microflora of most individuals is quite stable, especially in the large intestine and it is hard to find studies proving the "colonisation theory".

It is easier, however, to influence the metabolic activity of the microflora. According to several studies probiotics have various effects on the microflora regarding enzyme activity, influence on the endocrine system (20) and immunological activity (14-16). These effects have been demonstrated when using both "food derived bacteria" and " intestinal derived bacteria" although the definition of these groups is complex since the same bacterial species can be found in both groups!

From a historical/ecological point of view, supplementation with probiotics replaces the loss of lactic acid producing bacteria in the food caused by the change of preservation methods (11). Since the loss of probiotic bacteria in food does not refer to a particular strain but rather to many different kinds of strains, the probiotic supplements with a mixture of strains are preferable. Studies also show that different bacteria have different niches in relation to the immune system. A complex microflora is important for the immune system. However it is important to note that supplementation with single strain products also have various beneficial effects and this has been documented and patented by different producers of probiotic products.

Difficult to compare different studies.
The effect of probiotic supplementation is probably influenced by the choice of bacteria, the counts of bacteria, the quality, the viability and the stability of the bacteria and how the bacteria is distributed from the mouth to the intestine. The method of distributing bacteria certainly influences the dose required to obtain particular effects. Due to this the variation in dose may be as great as E4-5= 10.000-100.000 per day or per time of dosage. It probably requires higher dosages to influence the microflora of the large intestine than that of the small intestine regarding the bacterial counts in these areas are different.
As most studies on the effects of probiotic supplementation have not considered bacterial logistics, it is very difficult to compare them and this probably explains the different results presented. ***The role of bacterial logistics is one of the biggest challenges for future studies.*** Whilst waiting for the results of more clinical studies, we should perhaps be cautious in our claims that probiotics are effective against specific diseases but rather emphasise that probiotics have a beneficial effect on health as a whole.

Abstract: There is a need for probiotics. Modern life style implies a reduced intake of beneficial bacteria. There is a relationship between the health problems of today and microbial ecology. In this relation the microflora interacts with the immune system. The concern of bacterial logistics is important when choosing probiotic products.

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