EDITORIAL

Nanoparticles, the Microbiome, and Probiotics

During the last couple of decades, we have learned much about the microbiome, and studies continue to illuminate and expound on its many functions and effects on the human body.[1] There is increasing interest in nanoparticles (NPs) and there have been more studies lately on their effect on the microbiome.

The first description of NP in scientific terms was by Michael Faraday in his classic work in 1857.[2] NPs are defined by the US National Nanotechnology Initiative as materials having at least 1 diameter measuring 100 nm or less, with 1 nm being equal to 10^{-9} m, smaller than cells and many viruses. They are used in food packaging materials; as flame retardants; to help enhance imaging in magnetic resonance imaging (MRI) and positron emission tomography (PET) scans; in common consumer products; in genetic and tissue engineering; and in research for possible use in delivery of drugs, including chemotherapeutic agents in cancer treatment.

We have known of effects of NPs on the lungs from smoking tobacco, from fires indoors used for cooking in developing countries leading to early mortality, and from diesel NPs damaging the cardiovascular system in animal studies.[3,4] Published reports have shown that NPs can affect the gut and microbiome. There are other facets for NPs that are emerging as well, and for this editorial we shall focus on the consequences on the microbiome and how in the future probiotics may modify some of these outcomes for the better.

There are potential benefits from the use of NPs but also concerns over the potential for adverse human health effects. NPs may initiate medical problems, mostly due to the high surface-to-volume ratios, making the particles very reactive. They are able to pass through cell membranes, and their effects in humans are still being studied.

The gastrointestinal tract (GIT) includes a selective mucosal barrier that is considerable in surface area, estimated at 200 m^2 or the size of a tennis court. The average American eats approximately 1 ton of food per year, providing ample opportunity for NPs from foods to affect the microbiome. The surface area of the GIT provides a large zone for interaction with ingested NPs. In the normal gut, the majority of bacteria are commensal and do not leave the lumen. Most of them belong to the phyla *Firmicutes* and *Bacteroides*. These bacteria have very important and critical roles in digestion and in the immunological function of the GIT, including the regulation of colonic enterocyte health by the production of short-chain fatty acid butyrate, making vitamin B_{12} and vitamin K, the conjugation of bile acids, the maturation of the immune system, and the detoxification of toxins from drugs or plant sources, among others.[5]

Particles move through the intestinal barrier in a multistep route involving diffusion through the mucus layer, contact with M cells or enterocytes, and via paracellular transport or cellular entry. NPs move into epithelial cells via endocytosis according to studies by Fröhlich et al.[6] However, ingested NPs may have toxic effects if they cause changes of the normal microbiome, even though they may not be absorbed through the intestinal barrier. In the case of a pre-existing alteration of the microbiome, as in a Gram-negative bacterial overgrowth, the absorption of NPs may be affected by the adherence of NPs to lipopolysaccharides, which may result in increased delivery of the lipopolysaccharides and the NPs. NPs may also affect microbial metabolism, which could result in changes of nutrient absorption.[6]

NPs of titanium dioxide, silicon dioxide, and zinc oxide are the most common NPs found in what we usually ingest. The larger versions of these ingredients have been used in food and medicines for decades. The US Food and Drug Administration (FDA) requires that any substance added to food be evaluated for safety and these are considered "generally recognized as safe" (GRAS).[7] NPs can be used as anticaking agents, as gelatin forming agents, to purify water, and in packaging to protect against ultraviolent (UV) light.

Titanium dioxide nanoparticles (NTiOs) are currently used in many products. They can be found in personal-care products, paints, plastics, and coatings. These include toothpaste, a number of sunscreens, shampoos, deodorants, and shaving creams. Studies show that approximately one-third of the titanium dioxide in foods is in the form of NPs. NTiOs are frequently added to foods to whiten or brighten color. The highest amount of NTiOs in common food products are found in candies, powdered doughnuts and icing, sweets, and chewing gums. Titanium oxide (TiO) is also found in cheese, cereal, and Greek yogurt. Pharmaceutical drugs that have high prescription rates can also contain NTiO. Human exposure analysis identified children as having the highest exposures to NTiO because it is higher in sweets than most other foods. The typical exposure of an adult in the United States is approximately 1 mg of titanium per kilogram of body weight per day.[8] A study evaluating the acute toxicity of zinc NPs in mice showed that symptoms of lethargy, vomiting, and diarrhea occurred, as well as elevated levels of alanine aminotransferase (ALT), alkaline phosphatase (ALP), and lactate dehydrogenase (LDH). The authors found severe renal damage and a significant decrease in hemoglobin (HGB) and hematocrit (HCT), resulting in severe anemia.[9] Another study published in 2015 looked at the effects of NPs of silicon dioxide, titanium dioxide, and zinc oxide used by the food industry on cells taken from the human intestinal lining. All 3 caused inflammation, with zinc oxide (ZnO) being the most damaging and inducing the highest production of intracellular reactive oxygen species (ROS). These in turn caused significant damage to the DNA of cells, resulting in cell death.[10] There also may be exposures from the consumption of fish or shellfish that have accumulated NPs due to ingestion or absorption of contaminated effluent.[10] Silver NPs (AgNPs) are increasingly used by the food and food packaging industry. A recent study examined the effects of AgNPs on intestinal epithelial cells. Exposure for 24 hours of AgNPs caused 15% necrotic cell death and an 80% decrease in metabolic activity, as well markers indicating oxidative stress.[11] This finding is significant in that AgNPs are shed by nonedible products, such as AgNP-coated toothbrushes, containers of food and beverages, and even baby bottles and pacifiers.[12]

Probiotics are live active lactobacilli and *Bifidobacterium*; sometimes other microorganisms are added as well into commercially available products. Prebiotics develop specific beneficial changes in the microbiome; synbiotics combine probiotics with probiotics. In order to be effective, probiotics have to survive gastric acids and bile before they reach the colon where they exert their effects. The FDA does not regulate probiotics under drug regulations, as they are considered a dietary supplement; they are also not covered by health insurance.

Élie Metchnikoff, winner of the Nobel Prize for Medicine in 1908 along with Paul Ehrlich, was one of the first to realize the benefits of probiotics. In Greek, the word *pro* means "to promote" and *biotic* means "life." Metchnikoff's discoveries brought about the foundations of immunology, especially innate immunity. He observed that Bulgarians living in rural areas in poverty and in harsh climate conditions died at a very old age, attributing that to their consumption of yoghurt. Metchnikoff is credited with having coined the word *gerontology* and theorized that aging is caused by toxic bacteria in the gut. He concluded that health could be improved and senility delayed by enhancing the microbiome with the bacteria found in sour milk. In the last couple of decades, research has backed this up and has found even more benefits to probiotics.[13]

The effects of probiotics are beneficial immunomodulation, improved lactose digestion, and protection against colon cancer. They have also been found helpful in cases of diarrhea and inflammatory bowel disease. Probiotics aid in the maintenance of intestinal homeostasis and in the promotion of the function of the gut barrier.[14,15] Probiotics and their use are considered safe and complications are rare. A review panel report by the US government has supported the findings of epidemiological studies that there is no increase in population risk.[16,17]

Advances in genomics, investigations of the human microbiome, and the functional abilities of probiotics continue to show promise in bringing about changes of our understanding of how the microbiome affects our health and how probiotics may be directed to either fight or resist diseases and improve life. In the future, we may well see probiotic products addressing specific diseases in disorders for patients.[18]

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