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In Translation

Role of the Gut Microbiome in Uremia: A Potential Therapeutic Target

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Also known as the "second human genome," the gut microbiome plays important roles in both the maintenance of health and the pathogenesis of disease. The symbiotic relationship between host and microbiome is disturbed due to the proliferation of dysbiotic bacteria in patients with chronic kidney disease (CKD). Fermentation of protein and amino acids by gut bacteria generates excess amounts of potentially toxic compounds such as ammonia, amines, thiols, phenols, and indoles, but the generation of short-chain fatty acids is reduced. Impaired intestinal barrier function in patients with CKD permits translocation of gut-derived uremic toxins into the systemic circulation, contributing to the progression of CKD, cardiovascular disease, insulin resistance, and protein-energy wasting. The field of microbiome research is still nascent, but is evolving rapidly. Establishing symbiosis to treat uremic syndrome is a novel concept, but if proved effective, it will have a significant impact on the management of patients with CKD.
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INDEX WORDS: Gut microbiome; uremic toxin; microbial metabolite; metabolome; ammonia; urea; amine; thiol; phenol; indole; *p*-cresyl sulfate (PCS); uremic syndrome; chronic kidney disease (CKD); end-stage renal disease (ESRD); review.

BACKGROUND

Findings from the Human Microbiome Project (HMP) and the Metagenomics of the Human Intestinal Tract (Meta-HIT) project have shown that the human intestine is home to an extraordinarily complex and dynamic consortium of bacteria that play a pivotal role in human health and disease.[1,2] Bacteria have co-evolved with humans, and this symbiotic relationship has expanded our capabilities beyond what is coded in our own genome.[3] Genetically, we are vastly outnumbered by our own microbiome, the microbial genome. As the Nobel Laureate Joshua Lederberg has asserted, "We should think of each host and its parasites as a superorganism with the respective genomes yoked into a chimera of sorts."[4p9] The central role of the gut in human health has been long recognized, dating back to 400 BC, when Hippocrates stated, "death sits in the bowels."[5] This review provides an overview of the bidirectional relationship between chronic kidney disease (CKD) and the gut microbiome, discusses the consequences of gut dysbiosis in the pathogenesis of systemic inflammation and uremic toxicity, and highlights the recent advances in targeting the gut microbiome for therapeutic purposes.

CASE VIGNETTE

A 65-year-old man with CKD stage G4 presented with lethargy and chronic constipation to the emergency department. Clinical examination findings were unremarkable except for generalized muscle weakness and a distended abdomen with sluggish bowel sounds. Laboratory investigation showed the following values: sodium, 138 mEq/L; potassium, 6.3 mEq/L; chloride, 115 mEg/L; bicarbonate, 16 mEq/L; anion gap, 14; serum urea nitrogen, 60 mg/dL; serum creatinine, 3.8 mg/dL (corresponding to estimated glomerular filtration rate [eGFR] of 16 mL/min/1.73 m^2 using the IDMS-traceable 4-variable MDRD Study equation); glucose, 100 mg/dL; calcium, 8.1 mg/dL; phosphate, 7.1 mg/dL; albumin, 3.6 g/dL; white blood cell count, 8.1 $\times$ 10^9/L; and hemoglobin, 10.1 g/dL. Computed tomography of the brain was normal except for mild cortical atrophy. Computed tomography of the abdomen showed abundant fecal matter in a dilated rectum and sigmoid colon. After disimpaction with enemas and laxatives, the patient felt better. He was discharged with the recommendation to take laxatives on a regular basis.

The patient was seen in the outpatient clinic 2 months later. He appeared energetic and said that he was taking a prebiotic (*p*-inulin) and continuing the laxative when needed. Repeat

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laboratory evaluation showed the following values: sodium, 139 mEq/L; potassium, 4.0 mEq/L; chloride, 110 mEq/L; bicarbonate, 20 mEq/L; anion gap, 11; serum urea nitrogen, 51 mg/dL; serum creatinine, 3.4 mg/dL (corresponding to eGFR of 18 mL/min/1.73 m^2); glucose, 82 mg/dL; calcium, 8.3 mg/dL; phosphate, 6.2 mg/dL; and albumin, 3.8 g/dL.

In the case presented, the patient's clinical symptoms and biochemistry results improved with relief of constipation, possibly through the decreased generation and increased elimination of uremic toxins. This highlights the importance of colon health in patients with CKD.

PATHOGENESIS

Gut Microbiome in Health

The human gut harbors $\sim 10^{14}$ bacteria with an enormous metabolic potential.[6-8] Under physiologic conditions, the microbiota provide complementary functions by participating in metabolic activities that are not fully evolved in the human host, such as digestion of complex polysaccharides,[9] endogenous synthesis of certain vitamins and amino acids,[10] metabolism of bile acids,[11] degradation of dietary oxalates,[12] and maturation of the immune system.[13]

On average, an individual's gut microbiota is composed of 500 to 1,000 bacterial species.[14] Findings from the HMP suggest that each individual has a unique microbiome, each niche features one or a few signature taxa, and the gut microbiome is characterized by the greatest diversity with little variation over time.[15,16] The predominant bacterial groups in the human gastrointestinal tract are Bacteroidetes, Firmicutes, and Actinobacteria.[17,18] The phylogenetic composition of gut microbiota tends to be similar between individuals living in the same region, belonging to the same family, and having a similar diet.[19] Muegge et al[20] studied the gut microbiome profile in 33 mammalian species, including 18 humans, and reported that the difference in microbiome profiles stems from differing metabolic functions required to utilize the diet. Thus, the gut microbiome appears to change adaptively to the needs of the host organism.

Gut Microbiome in Kidney Disease

The term "dysbiosis" was first coined in the early 20th century by the Russian Nobel Laureate Elie Metchnikoff.[21] Dysbiosis is defined as an imbalanced intestinal microbial community with quantitative and qualitative alterations in the composition and metabolic activities of the gut microbiota. Preliminary evidence indicates that the microbiome profile might be altered in patients with chronic kidney failure and earlier stages of CKD[22] (Table 1). Vaziri et al[23] found that 190 microbial operational taxonomic units differed significantly in abundance between patients with end-stage renal disease and apparently healthy controls. Hida et al[24] reported that the number of aerobic bacteria, including *Enterobacteria* and *Enterococci* species, is higher in patients treated with maintenance hemodialysis than in controls. Among anaerobic bacteria, Hida et al[24] observed that hemodialysis patients have significantly lower numbers of *Bifidobacterium* species and higher organism counts for *Clostridium perfringens*.

The main contributing factors to gut microbiome dysbiosis in patients with kidney disease include slow intestinal transit time,[25] impaired protein assimilation,[26] decreased consumption of dietary fiber,[27] iron therapy,[28] and frequent use of antibiotics.[29,30] Antibiotic treatment decreases the diversity and alters the relative abundances of members of the bacterial community, with some patients exhibiting incomplete recovery post-treatment.[31]

Gut-Derived Uremic Toxins and Microbial Metabolites

In 1965, Einheber and Carter[32] showed that germ-free anephric mice survived longer than anephric mice with an intact gut microbiome. Aronov et al[33] showed that a number of uremic retention solutes are present only in hemodialysis patients with an intact colon. Recently, using untargeted metabolomic mass spectrometry, Wikoff et al[34] reported that the presence of several protein-bound uremic toxins, such as indoxyl sulfate (IS), hippuric acid, and phenylacetic acid, are dependent on the presence of gut microflora.

Impaired protein assimilation in uremia leads to large influx of undigested proteins into the distal intestine, which favors the proliferation of proteolytic bacteria[35] (Fig 1). Increased protein fermentation results in the generation of potentially toxic metabolites, such as ammonia, phenols, amines, indoles, and thiols.[36] Clinical manifestations of these uremic toxins are nonspecific and may include neurologic disorders, protein-energy wasting, cardiovascular disease (CVD), and progression of CKD. The potential pathways linking the accumulation of some of the major toxic metabolites to pathophysiologic consequences in patients with CKD are shown schematically in Fig 2. Increased levels of these toxins in patients with CKD may be related to increased generation from the dysbiotic microbiome or decreased elimination from reduced kidney function. In this review, we focus on the role of the gut microbiome in the generation of uremic toxins (Table 2).

Ammonia and Urea

Interdependency between humans and microbes in the metabolic process is exemplified by the urea nitrogen salvage pathway. The end product of mammalian protein catabolism is ammonia, which is toxic to cells in higher concentrations and thus is converted to urea through the ornithine-urea cycle. Mammals cannot break down urea, but gut bacteria expressing