Guben Xiezhuo Recipe alleviated Colon Dialysis on Urotoxin-induced Intestinal Barrier Injury and Systemic Inflammation

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Studies have shown that, dh404 produces anti-in ammatory and anti-oxidative stress e ects by stimulating the Nrf2 signaling pathway. It has a protective e ect on chronic kidney disease in rats [6].

Pyrrolidine dithiocarbamate(PDTC), is a speci c inhibitor of NF-κB, which can inhibit the activation of NF-κB and reduce in ammation. It is also an antioxidant, which can directly eliminate free radicals and inhibit the generation of free radicals [7].

Claudin is mainly located on the side of epithelial cells near the lumen. Its function is to close the intercellular space and prevent the free entry and exit of materials inside and outside the epithelial layer of the lumen. It is the material basis for the selection of permeabilization of epithelial cells [8]. Tight junctions are a composite structure formed under the interaction of multiple proteins, which are mainly composed of two components, which are transmembrane protein and cytoplasmic protein. e main transmembrane proteins are occludin and claudin-1. Both occludin and claudin-1 are four-pass transmembrane proteins, and the amino and carboxyl ends are located in the cytoplasm [9]. Tight junctions can e ectively prevent the paracellular transport of bacteria, toxins and other substances in the intestinal cavity through regulation under physiological conditions, and maintain the integrity of the intestinal mucosal epithelial barrier function.

Zonulaocludens-1(ZO-1) is a protein that is related to tightly connect. In recent years, it has been discovered that it is related to the maintenance and regulation of epithelial fence and barrier function, and also participates in the regulation of cell material transport, maintenance of epithelial polarity, cell proliferation and di erentiation, tumor cell metastasis and other important processes [10]. Since the structure and function of ZO-1 are closely related to other members of tight junctions, in most cases, as long as ZO-1 is damaged, the functions of tight junctions will change accordingly, so ZO-1 is o en used to observe the tight junctions of various tissues. Indicators of barrier function and permeability function, such as blood-brain barrier, intestinal epithelial cells, retinal pigment epithelial cells, etc [11].

Nrf2 is a key factor in the cellular oxidative stress response. It is regulated by kelch-like ECH-associated protein 1(Keap-1) and interacts with the antioxidant response element to regulate antioxidant proteins and phase II detoxi cation enzymes expression [12].

In this work, immuno uorescence was used to analyze the expression characteristics of intestinal epithelial tight junction protein occludin in the intestinal barrier injury model. is work uses Western Blot technology to analyze the expression of proteins ZO-1 and Cluadin-1 in intestinal epithelial cells of the intestinal barrier injury model, and analyzes the mRNA of Keap-1, Nrf2, ROS, and NF-κB in intestinal epithelial cells by RT-PCR technology. e above test is used to evaluate the destruction and repair e ect of the intestinal epithelial barrier function of CKD patients. In this work, Elisa was used to analyze the in ammatory signal hypersensitive C-reactive protein and interleukin-6 (IL-6) content in the cell culture medium to evaluate the in ammatory response caused by CKD and the reduction of in ammatory response by colon dialysis with Guben Dialysis e ect.

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Cultivate T84 cells at 37°C and 5% CO_2 with DMEM/F12 medium containing 10% fetal calf serum (FCS) and 50 U/L penicillin-streptomycin in cell culture asks, and replace the medium every 1-2 days. en induce and establish an intestinal epithelial injury model with low and high doses of urea.e by c20mor c3(fu9 72.64is a k culturof et d)nt yOnto j0.028 Tgh epithelof the intelture0% n a g.06p. Coms iatetioxida

standard samples were added to the wells according to the concentration requirements and serially diluted. The sample volume in each well was 50μl. Then a blank control well and the sample well to be tested were set. The blank control well would not add sample and enzyme-labeled reagent; the other steps were the same with the sample well. Add 40μl of sample diluent to the sample well, and then add 10μl of the sample. The final dilution of the sample is 5 times. Seal the plate with a sealing film and incubate at 37 for 30 minutes. After incubation, wash the plate with washing solution and dry it. After drying, add 50μl of enzyme-labeled reagent to each well except the blank well. Repeat the incubation and washing steps. Then add 50μl of developer A and 50μl of developer B in each well sequentially, and develop color at 37 for 15 minutes in the dark, and lastly add 50μl of stop solution to stop the reaction (the blue turns to yellow immediately). Set the blank control well as zero, and measure the absorbance (OD value) of each well in sequence at 450nm wavelength.