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**DHA-enriched phosphatidylcholine alleviate lupus nephritis by regulating
inflammation and the gut–kidney axis**

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Abstract: Lupus nephritis is a severe autoimmune kidney disease characterized by chronic inflammation and immune dysregulation. Nutritional modulation of inflammatory responses and gut microbiota homeostasis has emerged as a promising adjunct strategy for managing lupus nephritis. Docosahexaenoic acid (DHA), a functional lipid derived from marine foods, exhibits well-documented anti-inflammatory and immunomodulatory activities. Notably, the biological effects of DHA are strongly related to its chemical structure. Compared to conventional forms, phospholipid-bound DHA shows improved bioavailability and metabolic advantages, highlighting its potential in regulating inflammation and the gut–kidney axis. This study aimed to explore the protective effects of DHA-enriched phosphatidylcholine (DHA-PC) against lupus nephritis and compare its efficacy with conventional DHA supplementation in an animal model of lupus nephritis. The results demonstrated that DHA-PC markedly improved renal function indices by decreasing the levels of serum urea nitrogen and creatinine, alleviating renal pathological injury, and reducing proteinuria in lupus-prone mice, accompanied by a significant suppression of pro-inflammatory cytokines. Mechanistically, DHA-PC inhibited activation of the NF- κ B-NLRP3 inflammasome pathway, thereby attenuating renal inflammatory responses. Moreover, DHA-PC reshaped the composition of the gut microbiota by enriching anti-inflammatory genera, such as *Lactobacillus* and *Odoribacter*, while suppressing pro-inflammatory bacteria, including *Lachnoclostridium*, *Corynebacterium*, *Enterorhabdus*, and *Desulfovibrio*, accompanied by functional shifts in microbial metabolic pathways. Collectively, these findings indicate that DHA-PC confers robust protection against lupus nephritis through the coordinated regulation of inflammatory signaling and the gut–kidney axis. This study provides novel evidence that DHA-PC is a functional lipid involved in the prevention and management of autoimmunity-mediated renal injury.

Keywords: DHA-enriched phosphatidylcholine; inflammation; microbiota; lupus nephritis

1. Introduction

Lupus nephritis (LN) is one of the most severe clinical manifestations of systemic lupus erythematosus, affecting approximately 40–70% of patients during the disease course and serving as a major determinant of morbidity and mortality. LN is driven by the excessive production of autoantibodies, immune complex deposition in renal tissues, complement activation, and the subsequent amplification of inflammatory cascades, ultimately leading to glomerular and tubular injuries.^{1 2} Emerging evidence has suggested that inflammation plays a critical role in sustaining renal damage. More recently, the gut–kidney axis has been proposed as a key contributor to LN pathogenesis. Alterations in the gut microbiota composition and intestinal barrier integrity may facilitate the translocation of microbial-derived metabolites and endotoxins, thereby exacerbating systemic inflammation and autoimmune responses.^{3 4 5} Given the limitations and adverse effects of long-term immunosuppressive therapy, nutritional interventions, particularly functional foods that modulate inflammatory signaling and gut microbiota homeostasis, are increasingly recognized as promising and sustainable adjunct strategies for the management of lupus nephritis.⁶

Docosahexaenoic acid (DHA) is a member of the omega-3 polyunsaturated fatty acid family. It is not only a structural component for maintaining normal physiological functions but also plays a role in regulating lipid metabolism, controlling inflammation, and balancing immune function.⁷ ⁸The main sources of DHA in our daily diet are seafood, such as salmon (*Atlantic salmon*), tuna, sardines, Antarctic krill, and sea cucumbers. ⁹DHA supplementation has been shown to attenuate renal inflammation and improve renal function in various kidney disease models. ^{10 11 12}In autoimmune disorders, ω -3 polyunsaturated fatty acids modulate immune responses and alleviate disease severity, suggesting their therapeutic relevance in LN. Clinical and experimental studies have reported that DHA-rich fish oil supplementation reduces proteinuria and circulating inflammatory markers in LN, supporting the feasibility of DHA-based nutritional interventions. ¹³The biological efficacy of DHA was strongly dependent on its molecular form and bioavailability. Among the dietary sources, DHA predominantly exists as triglycerides, ethyl esters, and phospholipid-bound species. Among these, phospholipid-bound DHA has attracted increasing attention owing to its superior intestinal absorption, enhanced tissue distribution, and preferential incorporation into cellular membranes. Compared with the triglyceride or free fatty acid forms,

DHA-phospholipids are more efficiently integrated into membrane phospholipid bilayers, thereby exerting stronger regulatory effects on membrane-associated inflammatory and immune signaling pathways.⁹ In addition to these physicochemical advantages, DHA-esterified to phosphatidylcholine (DHA-PC) has emerged as a bioactive lipid with distinct functional properties.¹⁴ Recent studies have demonstrated that DHA-PL supplementation modulates the gut microbiota composition in depression-like mouse models, characterized by the enrichment of beneficial bacteria and attenuation of inflammation-associated dysbiosis, highlighting its capacity to regulate the microbiota-immune axis.¹⁵ In addition, DHA-PC exhibits pronounced anti-inflammatory effects in metabolic disorders, such as metabolic syndrome and atherosclerosis, by suppressing pro-inflammatory signaling and improving lipid metabolic homeostasis.^{16 17} Notably, DHA-PC has been shown to increase renal DHA incorporation and alleviate renal injury in models of acute kidney injury.¹⁸ Recent advances in renal lipidomics have further underscored lipid metabolism as a critical determinant of kidney disease progression. Profound alterations in phospholipid profiles have been identified in chronic kidney disease, especially in phosphatidylcholine and phosphatidylethanolamine, suggesting that targeted modulation of phospholipid composition may represent a promising strategy for renal protection.¹⁹ Taken together, DHA-PC integrates the anti-inflammatory properties of DHA with the structural and functional advantages of PC, and emerging evidence supports its role in gut microbiota modulation, inflammation suppression, and renal protection. However, despite these advances, the effects of DHA-PC on LN, particularly in comparison with conventional DHA and in the context of the gut-kidney axis, remain largely unexplored.

Therefore, the present study systematically investigated the differential effects of DHA-PC and DHA on LN progression using a lupus nephritis animal model. We systematically evaluated the effects of DHA-phospholipid supplementation on renal function, histopathological alterations, and systemic inflammatory status. In parallel, the gut microbiota composition and diversity were analyzed to determine whether DHA-phospholipids could restore microbial homeostasis and modulate the gut-kidney axis. This study provides novel evidence that DHA-enriched phospholipids are functional lipids with potential therapeutic value in the nutritional management of lupus nephritis.

2. Results

2.1 Effects of DHA-PC on immune organs and kidney indices

At 14 week of age, MRL/lpr mice begin to exhibit obvious pathological manifestations, including cervical skin lesions and scab formation on the ears, which are typical clinical features of systemic lupus and indicate progressive organ injury. From week 10 onward, a marked increase in daily urine output was observed, suggesting the onset of renal dysfunction. Compared to normal mice, the body weight was significantly increased in the model group. Although the DHA and DHA-PC interventions tended to reduce body weight, no statistically significant differences were observed. The thymus and spleen are key immune organs enriched in various immune cell populations. Compared to the normal group, both the spleen and thymus indices were markedly elevated in the model group ($P < 0.05$), with a particularly pronounced increase in the spleen index(Fig 1A-D). Dietary supplementation with DHA or DHA-PC significantly reduced both spleen and thymus indices ($P < 0.05$). Consistently, spleen length was significantly increased in model mice compared to normal controls, accompanied by obvious edema, whereas DHA and DHA-PC intervention alleviated splenomegaly ($P < 0.05$). Furthermore, renal weight and kidney index were significantly higher in MRL/lpr mice than in the normal group (Fig 1E-F). Both DHA and DHA-PC supplementation reduced renal weight and kidney index, indicating the attenuation of renal hypertrophy.

2.2 Effects on serum inflammatory cytokines and renal function

BUN and Cr are commonly used clinical indicators of renal function.²⁰ As shown in Figures 2A-B, BUN and Cr levels were significantly elevated in the model group compared to those in the normal group($P < 0.05$). Compared with the model group, DHA and DHA-PC supplementation effectively reduced serum BUN and Cr levels, with DHA-PC showing a more pronounced effect on lowering Cr ($P < 0.05$). In addition, urinary protein and urinary creatinine levels were markedly altered in the model group relative to normal controls, whereas DHA-PC supplementation significantly ameliorated these abnormalities (Fig 2C-D).

Serum levels of inflammatory cytokines were further assessed. As shown in the figure 2E-H, serum lipopolysaccharide (LPS) and tumor necrosis factor- α (TNF- α) levels were significantly

increased in the model group compared with the normal group ($P < 0.05$). DHA-PC supplementation markedly reduced the elevated LPS and TNF- α levels ($P < 0.05$). Although serum interleukin-6 (IL-6) and interleukin-1 β (IL-1 β) showed an increasing trend in model mice, the differences were not statistically significant ($P > 0.05$).

2.3 Renal histopathological analysis

Renal histopathological alterations were evaluated using H&E and PAS staining. As shown in the Fig 3A-B, kidneys from model mice exhibited severe pathological damage compared to normal controls, including glomerular atrophy, interstitial inflammatory cell infiltration, and proteinaceous casts within the renal tubules. Dietary supplementation with DHA-PC was markedly alleviated by these pathological lesions, showing an improved glomerular structure and reduced inflammatory infiltration. Notably, the protective effect of DHA-PC on renal histopathology was superior to that observed with DHA treatment (Fig 3C).

2.4 Changes in inflammatory signaling-related proteins

Given the critical role of the NF- κ B signaling pathway in the progression of nephritis, the expression of NF- κ B pathway-related proteins in renal tissues was assessed by western blotting. As shown in Figure 4A-D, the expression of NF- κ B p65 was significantly increased in the kidneys of model mice compared with normal controls ($P < 0.05$). DHA-PC supplementation significantly reduced NF- κ B p65 expression ($P < 0.05$), while DHA exhibited a comparable inhibitory effect. Compared to the normal group, renal caspase-1 expression was markedly elevated in the model mice ($P < 0.05$). Dietary supplementation with DHA-PC or DHA reduced caspase-1 expression, although the difference was not statistically significant ($P > 0.05$). Additionally, the protein expression levels of ASC and NLRP3 were significantly increased in the model group. Compared to the model group, DHA-PC supplementation significantly reduced the expression of caspase-1 and NLRP3 ($P < 0.05$).

2.5 Renal fatty acid composition

DHA and arachidonic acid (AA) are important polyunsaturated fatty acids in renal lipids that are closely associated with renal physiological function.¹²⁻²¹ Compared to the normal group, the proportion of AA in renal tissues was significantly increased in model mice ($P < 0.05$), whereas DHA-PC supplementation markedly reduced renal AA levels (Fig 4D). In contrast, the renal DHA content was significantly decreased in the model group ($P < 0.05$). Dietary supplementation with

either DHA-PC or DHA significantly increased the proportion of DHA in the kidneys of mice with renal injury.

2.6 Gut microbiota composition and diversity

Changes in the gut microbiota composition following dietary intervention were further analyzed (Figure 5A-D). Alpha diversity analysis revealed that the Ace, Chao, and Sobs indices were significantly increased ($P < 0.05$), whereas the Simpson index was significantly decreased in nephritic mice compared to that in normal controls, which may be related to the mouse strain. Supplementation with DHA and DHA-PC significantly increased the Simpson index, with DHA-PC showing a more pronounced effect ($P < 0.01$). Beta diversity analysis based on principal coordinate analysis for the PcoA results of OTU level demonstrated a clear separation between nephritic and normal mice, indicating substantial differences in gut microbial composition. The intervention groups were also separated from the model group, suggesting that DHA and DHA-PC altered the structure of the gut microbiota (Figure 5E). Venn diagram analysis further revealed distinct OTU distributions among the groups, with 193 OTUs commonly involved in lupus nephritis progression (Figure 5F).

At the phylum level, gut microbiota was dominated by *Firmicutes*, *Bacteroidota*, *Actinobacteriota*, *Desulfobacterota*, and *Proteobacteria* (Figure 6A). In normal mice, *Bacteroidota* and *Firmicutes* together accounted for over 93% of the total abundance. Compared with the normal group, the relative abundance of *Firmicutes*, *Actinobacteriota*, *Desulfobacterota*, and *Proteobacteria*, as well as the *Firmicutes/Bacteroidota* (F/B) ratio, was increased in the model group, whereas *Bacteroidota* abundance was significantly reduced (Figure 6B). DHA-PC supplementation significantly decreased the relative abundances of *Actinobacteriota* and *Desulfobacterota* and reduced the F/B ratio. At the genus level, significant changes were observed for *Lactobacillus*, *Norank_f_Muribaculaceae*, *Dubosiella*, and *Turicibacter* (Figure 6C). Compared with normal controls, the relative abundances of *Lactobacillus*, *Odoribacter*, *Lachnoclostridium*, *Corynebacterium*, *Enterorhabdus*, and *Desulfovibrio* were significantly higher in the model mice (Figure 6D-I). DHA-PC supplementation significantly increased the abundance of *Lactobacillus* and *Odoribacter*, while reducing that of *Lachnoclostridium*, *Corynebacterium*, *Enterorhabdus*, and *Desulfovibrio*, with effects superior to those of DHA.

Heatmap analysis revealed distinct microbial community compositions on OUT, phylum and genus of level between nephritic and normal mice, with similar overall modulation patterns for DHA and DHA-PC (Fig 6J-L). Differential OTU analysis indicated that OTU251, OTU312, OTU134/261, and OTU227 are closely involved in lupus nephritis progression. DHA-PC primarily ameliorated nephritis by modulating OTU251, OTU134, and OTU234, whereas DHA mainly exerted its effects through OTU227 and OTU234. Phylogenetic analysis indicated that *Lactobacillus*, *norank_f_Muribaculaceae*, *Dubosiella*, and *Enterorhabdus* were closely associated with disease progression. DHA-PC predominantly exerts its effects by regulating the *Lactobacillus* and *Odoribacter* species.

Circos analysis showed that *Lactobacillus*, *norank_f_Muribaculaceae*, *Dubosiella*, *unclassified_f_Lachnospiraceae*, and *Enterorhabdus* were closely associated with lupus nephritis. DHA-PC alleviated nephritis mainly by modulating *Lactobacillus* and *Odoribacter*, whereas DHA exerted its effects primarily through *Norank_f_Muribaculaceae* and *Enterorhabdus* (Figure 7A). Functional prediction using PICRUSt2 revealed that alterations in the gut microbiota of nephritic mice were mainly associated with secondary metabolite biosynthesis, inorganic ion transport and metabolism, lipid transport and metabolism, coenzyme transport and metabolism, nucleotide transport and metabolism, amino acid transport and metabolism, carbohydrate transport and metabolism, and energy production and conversion (Figure 7B). KEGG pathway analysis further indicated that gut microbiota functions were primarily enriched in carbohydrate metabolism, amino acid metabolism, energy metabolism, membrane transport, translation, lipid metabolism, folding, sorting and degradation, and glycan biosynthesis and metabolism at the second level, and in pathways such as biosynthesis of secondary metabolites, microbial metabolism in diverse environments, biosynthesis of amino acids, carbon metabolism, ribosome, ABC transporters, purine metabolism, starch and sucrose metabolism, glycolysis/gluconeogenesis, and amino sugar and nucleotide sugar metabolism at the third level (Figure 7D-E).

3. Discussion

LN is one of the most severe complications of systemic lupus erythematosus, and remains a leading cause of morbidity and mortality. Its progression is driven by persistent immune dysregulation,

chronic inflammation, and progressive structural renal damage.² In recent years, nutritional interventions targeting the immune and inflammatory pathways have attracted increasing attention as an adjunct strategy for LN management.²² In the present study, we demonstrated that dietary supplementation with DHA-PC markedly ameliorated lupus nephritis in MRL/lpr mice, as evidenced by improvements in immune organ indices, renal function, renal histopathology, inflammatory signaling pathways, renal lipid composition, and gut microbiota homeostasis (Figure 8). Importantly, DHA-PC consistently exhibited superior protective effects compared with free DHA.

LN is one of the most common secondary glomerulonephritis.²³ In this experiment, mice exhibited skin damage, ear crusting, and polyuria during feeding. Compared with the normal group, the model group mice showed significantly increased body weight, enlarged immune organs, renal edema, increased serum urea nitrogen and creatinine levels, upregulated inflammatory factor levels, and renal interstitial inflammatory infiltration. These results indicated the successful establishment of an animal model of glomerulonephritis. While current treatments for glomerulonephritis primarily utilize glucocorticoids and immunosuppressants, dietary intervention is also an effective strategy for improving kidney damage.⁶ Numerous studies have shown that certain naturally active substances possess anti-inflammatory activities and can inhibit the development of nephritis. DHA and EPA, which are marine-derived polyunsaturated fatty acids, have good anti-inflammatory activity and can improve various inflammation-related diseases such as enteritis and neuroinflammation.^{8,10} Recent studies have found that phospholipid-based DHA/EPA exhibits several advantages over triglycerides (TG) or ethyl ester DHA/EPA. For example, DHA-PC is almost completely absorbed by the intestine and readily binds to cell membrane molecular structures, owing to its unique properties.²⁵ Previous studies have shown that DHA-PC is more effective than TG-DHA in inhibiting MPTP-induced oxidative damage in the brain.²⁶ Early life supplementation with DHA-PL can rapidly increase DHA levels in the brains of young mice, with higher supplementation efficiency than that of TG-DHA.²⁷ Furthermore, DHA-PC was more effective than the ethyl ester DHA in improving vancomycin-induced acute kidney injury.²⁸ The results of this experiment showed that the renoprotective effects of DHA-PC were consistently more pronounced than those of DHA, and DHA-PC supplementation effectively improved these renal functional

parameters, with a particularly pronounced reduction in serum creatinine, attenuated glomerular atrophy, and inflammatory cell infiltration.

Chronic activation of inflammatory signaling pathways is a central driver of lupus nephritis progression, and increasing evidence suggests that functional dietary components can ameliorate renal injury by targeting the inflammatory pathway.^{29 30} In particular, the suppression of the NF- κ B signaling cascade and downstream inflammasome activation have emerged as a common mechanistic axis underlying the renoprotective effects of various functional foods in lupus nephritis models. Several natural bioactive compounds alleviate lupus nephritis by inhibiting NF- κ B-mediated inflammatory responses. Resveratrol and curcumin have been shown to reduce renal inflammation in lupus-prone mice by suppressing NF- κ B activation and decreasing the expression of pro-inflammatory cytokines, including TNF- α and IL-6.^{31 32} Similarly, quercetin and epigallocatechin gallate (EGCG) have demonstrated protective effects against renal injury through attenuation of NF- κ B signaling and oxidative stress, thereby improving renal histopathology and function.^{33 34} Inflammasome activation, particularly of the NLRP3 inflammasome, is increasingly recognized as a key contributor to lupus-associated renal inflammation.³⁵ Omega-3 polyunsaturated fatty acids has been reported to inhibit NLRP3 inflammasome assembly and caspase-1 activation, thereby reducing IL-1 β production and renal inflammatory damage.³⁶ The present findings extend previous observations by demonstrating that DHA-PC effectively suppresses both upstream NF- κ B activation and downstream NLRP3 inflammasome signaling in renal tissues. This suggests that the phospholipid-bound form of DHA may enhance the anti-inflammatory efficacy of omega-3 fatty acids in lupus nephritis.

Increasing evidence has established that gut microbiota is a pivotal regulator of host immune homeostasis and systemic inflammation.³⁷ The gut microbiome interacts with the host through multiple pathways, including the production of microbial metabolites such as short-chain fatty acids (SCFAs), modulation of intestinal barrier integrity, and regulation of endotoxin (lipopolysaccharide, LPS) translocation.³⁸ Dysbiosis, defined as an imbalance in the microbial composition, can disrupt these interactions, contributing to elevated systemic inflammation, immune dysregulation, and exacerbation of chronic inflammatory diseases.³⁹ Inflammatory mediators activated by microbial disturbances can further propagate local and systemic tissue damage through pathways such as NF-

κB and the NLRP3 inflammasome. Gut-derived microbial metabolites and endotoxins can access systemic circulation and modulate immune responses, with direct implications for kidney inflammation and function. In autoimmune and inflammatory kidney diseases, including lupus nephritis, gut dysbiosis has been associated with increased intestinal permeability, elevated circulating LPS, and exacerbated renal inflammation. Preclinical studies have consistently shown that restoring the gut microbial balance can attenuate renal injury, reduce inflammatory cytokine levels, and improve renal histopathology.⁴⁰ Functional foods and dietary interventions that modulate the gut microbiota have shown promise in improving the outcomes of lupus nephritis. Probiotic supplementation with *Lactobacillus* strains has been demonstrated to ameliorate renal dysfunction and reduce systemic inflammation in lupus-prone models. *Lactobacillus* species contribute to SCFA production (acetate, butyrate), which are known to enhance epithelial barrier integrity,⁴¹ suppress pro-inflammatory signaling pathways such as NF-κB, and promote regulatory T cell differentiation. An enhanced abundance of *Lactobacillus* has been correlated with reduced levels of circulating inflammatory mediators and improved renal function, indicating its potential as an anti-inflammatory microbial marker in LN. Similarly, genera such as *Odoribacter*, recognized for their SCFA-producing capacity, have been linked to anti-inflammatory effects and improved barrier function in inflammatory diseases. Increased *Odoribacter* abundance has been associated with the modulation of host immune responses, reduced pro-inflammatory cytokine production, and reinforcement of gut epithelial integrity, thereby mitigating systemic inflammation.⁴² In contrast, *Lachnospirillum*, *Corynebacterium*, *Enterorhabdus*, and *Desulfovibrio* have been associated with pro-inflammatory states.^{43 44 45} Members of the *Lachnospirillum* genus are associated with mucin degradation and impaired gut-barrier function, potentially facilitating endotoxin translocation and systemic immune activation.⁴⁶ An elevated abundance of *Corynebacterium* species has been linked to Th17-driven inflammatory responses in autoimmune settings, whereas *Enterorhabdus* enrichment correlates with dysregulated host metabolism and inflammatory phenotypes.⁴⁷ ⁴⁸*Desulfovibrio*, a sulfate-reducing bacterium, produces hydrogen sulfide, which can compromise epithelial barriers and exacerbate inflammatory cascades at high concentrations.^{45 49} Functional food interventions that reduce the relative abundance of pro-inflammatory taxa have been reported to attenuate systemic inflammation and improve renal pathology in experimental autoimmune

models.^{50 51} In the present study, DHA-PC supplementation restored microbial diversity and shifted the gut microbial profile toward a composition associated with anti-inflammatory potential. Specifically, DHA-PC increased *Lactobacillus* and *Odoribacter* abundance while reducing *Lachnoclostridium*, *Corynebacterium*, *Enterorhabdus*, and *Desulfovibrio*, which corresponded to decreased serum LPS levels and lowered pro-inflammatory signaling in renal tissues. These findings align with research on other functional food interventions, where the modulation of key microbial taxa was linked to the suppression of systemic inflammation and attenuation of lupus nephritis progression. Functional prediction analyses further suggested that DHA-PC-modulated microbial communities were enriched in pathways related to lipid and amino acid metabolism, energy production, and secondary metabolite biosynthesis, which interface with host immune regulation and inflammatory responses. These results indicate that targeting gut dysbiosis and enhancing microbial metabolic functions may therefore represent a promising nutritional strategy for mitigating autoimmune renal injury through the gut–kidney axis.

In summary, this study demonstrated that DHA-PC exerts a pronounced protective effect against lupus nephritis through the coordinated regulation of inflammation and gut microbiota homeostasis. DHA-PC supplementation significantly ameliorated renal dysfunction and pathological injury, reduced systemic and renal inflammatory responses, and suppressed activation of the NF- κ B–NLRP3–ASC–caspase-1 signaling pathway. Importantly, DHA-PC reshaped the gut microbial community by enriching anti-inflammatory bacteria such as *Lactobacillus* and *Odoribacter* while reducing pro-inflammatory taxa, including *Lachnoclostridium*, *Corynebacterium*, *Enterorhabdus*, and *Desulfovibrio*, thereby alleviating endotoxemia and disrupting the inflammatory amplification loop along the gut–kidney axis. Compared to free DHA, DHA-PC exhibited superior protective effects, highlighting its potential as a functional phospholipid ingredient for nutritional intervention in lupus nephritis.

4. Materials and methods

4.1 Preparation of DHA-PC

DHA-PC was prepared by enriching DHA in the phosphatidylcholine (PC) fraction derived from *Sthenoteuthis oualaniensis* refer to the Qu et al.⁵² Briefly, crude lipids were extracted from *S.*

oualaniensis using a chloroform–methanol method and then subjected to fractionation to obtain a PC-rich extract, followed by purification to enrich the DHA-containing PC species. The purity and composition of DHA-PC were confirmed using thin-layer chromatography and high-performance liquid chromatography. Fatty acid profiles were determined using gas chromatography. All lipid samples were protected from oxidation, flushed with nitrogen, and stored at –80 °C until use.

4.2 Animal experimental design

Four-week-old female MRL/lpr mice were purchased from Nanjing Junke Bioengineering Co., Ltd. (Nanjing, China). After one week of acclimatization under standard laboratory conditions, the mice underwent a two-week gut microbiota homogenization period to minimize baseline inter-individual microbial variation. Subsequently, the MRL/lpr mice were randomly assigned to three groups according to their body weight: model group (M), DHA–phosphatidylcholine group (DPC), and DHA group (DHA). Age-matched female C57BL/6J mice served as normal controls (N). Each group consisted of eight mice. The mice were administered the corresponding test substances once daily via oral gavage in the afternoon. The DHA gavage dose was set at 300 mg/kg body weight (BW), calculated based on the recommended human dietary reference intake and adjusted using body surface area conversion. The intervention period lasted 10 wk. The model and normal control groups received an equivalent volume of the vehicle. At the end of the experimental period, the mice were anesthetized with chloral hydrate and placed in metabolic cages for 24 h urine collection. Serum was separated by centrifugation, and the kidney tissue and fecal samples were harvested immediately. All samples were weighed, rapidly frozen in liquid nitrogen, and stored at –80 °C until further analysis. All experimental procedures were approved by the Institutional Animal Care and Use Committee and were conducted with accordance with ethical guidelines (Approval number: HNUAUCC-2023-00137).

4.3 Determination of serum biochemical and inflammatory parameters

Serum was obtained by centrifugation at $3000 \times g$ for 15 min at 4 °C. Serum blood urea nitrogen (BUN) and creatinine (Cr) levels were measured using commercial diagnostic kits to assess renal function. Urinary protein and creatinine concentrations were determined, and the urinary protein-to-creatinine ratio was calculated as an indicator of renal injury. The levels of lipopolysaccharide (LPS), tumor necrosis factor- α (TNF- α), interleukin-6 (IL-6), and interleukin-1 β (IL-1 β) were

quantified using enzyme-linked immunosorbent assay (ELISA) kits according to the manufacturers' instructions.

4.4 Renal histopathological examination

The kidney tissue was fixed with 4% paraformaldehyde for 24 hours, then embedded in paraffin and sliced to a thickness of 4 μ m. Finally, the overall morphology of the kidney, including glomerular and tubular structures as well as inflammatory cell infiltration, was evaluated using hematoxylin and eosin (H&E) staining. In addition, periodic acid-Schiff (PAS) staining was used to evaluate glomerular basement membrane thickening, mesangial matrix dilation, and other subtle structural changes associated with renal injury. Histopathological changes were examined under a light microscope by a professional researcher. Renal injury was semi-quantitatively scored based on glomerular and tubular pathological features.

4.5 Analysis of renal fatty acid composition

Total lipids were extracted from kidney tissues using the chloroform-methanol method, according to the method described by Liu.⁵³ The lipid extracts were transmethylated to generate fatty acid methyl esters (FAMES). The FAMES were analyzed using a gas chromatograph equipped with a flame ionization detector and a capillary column suitable for fatty acid separation. Individual fatty acids were identified using authentic standards and expressed as relative percentages of the total fatty acids.

4.6 Western blot analysis

Approximately 100 mg of renal tissue was collected from each mouse and homogenized in ice-cold RIPA lysis buffer supplemented with protease inhibitors, following a previously described protocol with minor modifications (Shi et al.).⁵⁴ The homogenates were centrifuged at $12,000 \times g$ for 15 min at 4 °C, and the supernatants were collected for protein analysis. Protein concentrations were determined by the bicinchoninic acid assay. Twenty micrograms of total protein from each sample was loaded and separated by sodium dodecyl sulfate-polyacrylamide gel electrophoresis according to molecular weight. The proteins were then transferred onto polyvinylidene difluoride membranes using a semi-dry transfer system. After transfer, the membranes were blocked with 5% nonfat milk in Tris-buffered saline containing 0.1% Tween-20 (TBST) for 2 h at room temperature with gentle shaking. The membranes were incubated overnight at 4 °C with primary antibodies against NF- κ B

p65, caspase-1, apoptosis-associated speck-like protein containing a CARD (ASC), NOD-like receptor protein 3 (NLRP3), and β -actin at the dilutions recommended by the manufacturers. The following day, the membranes were equilibrated at room temperature for 30 min and washed thoroughly with TBST for 40 min. Subsequently, the membranes were incubated with the appropriate horseradish peroxidase-conjugated secondary antibodies for 2 h at room temperature. After extensive washing with TBST, the protein bands were visualized using enhanced chemiluminescence reagents and captured using a Tanon 5200 chemiluminescence imaging system (Tanon Science & Technology, Shanghai, China). Band intensities were quantified by densitometric analysis using ImageJ software, and target protein expression levels were normalized to β -actin.

4.7 Gut microbiota analysis based on 16s rrna sequencing

Cecal and colonic contents were collected under sterile conditions, immediately placed into sterile microcentrifuge tubes, and stored at -80°C until further analysis. The gut microbiota composition in the cecal contents of the normal (N), model (M), DHA-PC, and DHA groups ($n = 4$ per group) was analyzed using 16S rRNA gene sequencing. Total microbial genomic DNA was extracted using a FastPure Stool DNA Isolation Kit (MJYH, Shanghai, China) according to the manufacturer's instructions. The integrity of the extracted DNA was assessed by 1% agarose gel electrophoresis, and the DNA concentration and purity were determined using a NanoDrop 2000 spectrophotometer (Thermo Scientific, USA). The V3–V4 hypervariable regions of the bacterial 16S rRNA gene were amplified by polymerase chain reaction using barcoded universal primers. The resulting amplicons were purified, quantified, and used for library construction using a NEXTFLEX Rapid DNA-Seq Kit. High-throughput sequencing was performed using an Illumina NextSeq 2000 platform (Shanghai Majorbio Bio-Pharm Technology Co., Ltd., Shanghai, China). Raw sequencing data were quality-filtered and denoised to generate amplicon sequence variants (ASVs). Taxonomic assignment was conducted using the naïve Bayes classifier implemented in QIIME 2 based on the SILVA 16S rRNA gene reference database (version 138). Functional profiles of the gut microbiota were predicted using the PICRUST2 software (version 2.2.0).

4.8 Statistical analysis

Data are presented as mean $\pm$ standard deviation. Statistical analyses were conducted using corresponding software. Intergroup differences were analyzed using one-way analysis of variance

(ANOVA), and post-hoc multiple comparison tests were conducted. Spearman's rank correlation analysis was used. Statistical significance was set at $P < 0.05$. significant.

Data Availability

Data will be made available upon request.

Notes

The authors declare no competing financial interest.

Authorship contribution statement

Haohao Shi: Writing-original draft, Writing-review & editing, Visualization, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. Lipin Chen: Software, Methodology, Formal analysis. Tiantian Zhang: Methodology, Investigation. Yuming Wang: Investigation, Writing-review & editing, Software, Resource.

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REFERENCES

- 1 Parodis, I. *et al.* Lupus nephritis. *Nat Rev Dis Primers* **11**, 69, doi:10.1038/s41572-025-00653-y (2025).
- 2 Lichtnekert, J. & Anders, H. J. Lupus nephritis-related chronic kidney disease. *Nat Rev Rheumatol* **20**, 699-711, doi:10.1038/s41584-024-01158-w (2024).
- 3 Nozaki, Y. The Network of Inflammatory Mechanisms in Lupus Nephritis. *Front Med (Lausanne)* **7**, 591724, doi:10.3389/fmed.2020.591724 (2020).
- 4 Mohd, R., Chin, S. F., Shaharir, S. S. & Cham, Q. S. Involvement of Gut Microbiota in SLE and Lupus Nephritis. *Biomedicines* **11**, doi:10.3390/biomedicines11030653 (2023).
- 5 Zhang, L. *et al.* Gut Microbiome and Metabolites in Systemic Lupus Erythematosus: Link, Mechanisms and Intervention. *Front Immunol* **12**, 686501, doi:10.3389/fimmu.2021.686501 (2021).
- 6 Mok, C. C., Teng, Y. K. O., Saxena, R. & Tanaka, Y. Treatment of lupus nephritis: consensus,

evidence and perspectives. *Nat Rev Rheumatol* **19**, 227-238, doi:10.1038/s41584-023-00925-5 (2023).

7 Li, J., Pora, B. L. R., Dong, K. & Hasjim, J. Health benefits of docosahexaenoic acid and its bioavailability: A review. *Food Sci Nutr* **9**, 5229-5243, doi:10.1002/fsn3.2299 (2021).

8 Siriwardhana, N., Kalupahana, N. S. & Moustaid-Moussa, N. Health benefits of n-3 polyunsaturated fatty acids: eicosapentaenoic acid and docosahexaenoic acid. *Adv Food Nutr Res* **65**, 211-222, doi:10.1016/b978-0-12-416003-3.00013-5 (2012).

9 Zhang, T. T., Xu, J., Wang, Y. M. & Xue, C. H. Health benefits of dietary marine DHA/EPA-enriched glycerophospholipids. *Prog Lipid Res* **75**, 100997, doi:10.1016/j.plipres.2019.100997 (2019).

10 Hong, S. & Lu, Y. Omega-3 fatty acid-derived resolvins and protectins in inflammation resolution and leukocyte functions: targeting novel lipid mediator pathways in mitigation of acute kidney injury. *Front Immunol* **4**, 13, doi:10.3389/fimmu.2013.00013 (2013).

11 Fassett, R. G., Gobe, G. C., Peake, J. M. & Coombes, J. S. Omega-3 polyunsaturated fatty acids in the treatment of kidney disease. *Am J Kidney Dis* **56**, 728-742, doi:10.1053/j.ajkd.2010.03.009 (2010).

12 Syren, M. L. *et al.* The polyunsaturated fatty acid balance in kidney health and disease: A review. *Clin Nutr* **37**, 1829-1839, doi:10.1016/j.clnu.2017.11.019 (2018).

13 Pestka, J. J. n-3 polyunsaturated fatty acids and autoimmune-mediated glomerulonephritis. *Prostaglandins Leukot Essent Fatty Acids* **82**, 251-258, doi:10.1016/j.plefa.2010.02.013 (2010).

14 Cao, B. *et al.* Interaction between dietary omega-3 polyunsaturated fatty acids, obesity and gut microbiota in preclinical models: A systematic review of randomized controlled trials. *Diabetes Obes Metab* **27**, 4643-4661, doi:10.1111/dom.16535 (2025).

15 Cao, W. *et al.* DHA-phospholipids (DHA-PL) and EPA-phospholipids (EPA-PL) prevent intestinal dysfunction induced by chronic stress. *Food Funct* **10**, 277-288, doi:10.1039/c8fo01404c (2019).

16 Ding, L. *et al.* DHA-Enriched Phosphatidylcholine and DHA-Enriched Phosphatidylserine Improve Age-Related Lipid Metabolic Disorder through Different Metabolism in the Senescence-Accelerated Mouse. *European Journal of Lipid Science and Technology* **120**, doi:10.1002/ejlt.201700490 (2018).

17 Wang, Q. *et al.* Comparison of the Effect of Phospholipid Extracts from Salmon and Silver Carp Heads on High-Fat-Diet-Induced Metabolic Syndrome in C57BL/6J Mice. *Mar Drugs* **21**, doi:10.3390/md21070409 (2023).

18 Shi, H. H. *et al.* Comparative evaluation of phosphatidylcholine and phosphatidylserine with different fatty acids on nephrotoxicity in vancomycin-induced mice. *Biosci Biotechnol Biochem* **85**, 1873-1884, doi:10.1093/bbb/zbab105 (2021).

19 Fan, X. *et al.* Combined UPLC-QqQ-MS/MS and AP-MALDI Mass Spectrometry Imaging Method for Phospholipidomics in Obese Mouse Kidneys: Alleviation by Feeding Sea Cucumber Phospholipids. *J Agric Food Chem* **72**, 16312-16322, doi:10.1021/acs.jafc.4c02692 (2024).

20 Krstic, D. *et al.* Biochemical Markers of Renal Function. *Curr Med Chem* **23**, 2018-2040, doi:10.2174/0929867323666160115130241 (2016).

21 Wang, T. *et al.* Arachidonic Acid Metabolism and Kidney Inflammation. *Int J Mol Sci* **20**, doi:10.3390/ijms20153683 (2019).

22 Ahn, S. S. *et al.* Comparison of the Clinical Implications among Five Different Nutritional

Indices in Patients with Lupus Nephritis. *Nutrients* **11**, doi:10.3390/nu11071456 (2019).

23 Satoskar, A. A., Parikh, S. V. & Nadasdy, T. Epidemiology, pathogenesis, treatment and outcomes of infection-associated glomerulonephritis. *Nat Rev Nephrol* **16**, 32-50, doi:10.1038/s41581-019-0178-8 (2020).

24 Komprda, T. Eicosapentaenoic and docosahexaenoic acids as inflammation-modulating and lipid homeostasis influencing nutraceuticals: A review. *Journal of Functional Foods* **4**, 25-38, doi:10.1016/j.jff.2011.10.008 (2012).

25 Ju, S. N. *et al.* Characterization, stability, digestion and absorption of a nobiletin nanoemulsion using DHA-enriched phosphatidylcholine as an emulsifier in vivo and in vitro. *Food Chem* **397**, 133787, doi:10.1016/j.foodchem.2022.133787 (2022).

26 Wang, C. *et al.* DHA enriched phospholipids with different polar groups (PC and PS) had different improvements on MPTP-induced mice with Parkinson's disease. *Journal of Functional Foods* **45**, 417-426, doi:10.1016/j.jff.2018.04.017 (2018).

27 Wen, M. *et al.* Short-term supplementation of DHA as phospholipids rather than triglycerides improve cognitive deficits induced by maternal omega-3 PUFA deficiency during the late postnatal stage. *Food Funct* **12**, 564-572, doi:10.1039/d0fo02552f (2021).

28 Shi, H. H. *et al.* DHA-PC protects kidneys against cisplatin-induced toxicity and its underlying mechanisms in mice. *Food Funct* **10**, 1571-1581, doi:10.1039/c8fo02386g (2019).

29 Braunstein, M. Taming renal inflammation: signaling pathways and therapeutic advances in lupus nephritis. *BMC Nephrol* **26**, 507, doi:10.1186/s12882-025-04434-3 (2025).

30 Islam, M. A., Khandker, S. S., Kotyla, P. J. & Hassan, R. Immunomodulatory Effects of Diet and Nutrients in Systemic Lupus Erythematosus (SLE): A Systematic Review. *Front Immunol* **11**, 1477, doi:10.3389/fimmu.2020.01477 (2020).

31 Yang, H. *et al.* Curcumin attenuates lupus nephritis by inhibiting neutrophil migration via PI3K/AKT/NF- κ B signalling pathway. *Lupus Sci Med* **11**, doi:10.1136/lupus-2024-001220 (2024).

32 Huo, R. *et al.* Potential of resveratrol in the treatment of systemic lupus erythematosus (Review). *Mol Med Rep* **30**, doi:10.3892/mmr.2024.13306 (2024).

33 Chen, H. Y. *et al.* Quercetin Ameliorates Renal Injury and Pyroptosis in Lupus Nephritis through Inhibiting IL-33/ST2 Pathway In Vitro and In Vivo. *Antioxidants (Basel)* **11**, doi:10.3390/antiox11112238 (2022).

34 Tsai, P. Y. *et al.* Epigallocatechin-3-gallate prevents lupus nephritis development in mice via enhancing the Nrf2 antioxidant pathway and inhibiting NLRP3 inflammasome activation. *Free Radic Biol Med* **51**, 744-754, doi:10.1016/j.freeradbiomed.2011.05.016 (2011).

35 Oliveira, C. B., Lima, C. A. D., Vajgel, G. & Sandrin-Garcia, P. The Role of NLRP3 Inflammasome in Lupus Nephritis. *Int J Mol Sci* **22**, doi:10.3390/ijms222212476 (2021).

36 Favor, O. K. *et al.* Lipidome modulation by dietary omega-3 polyunsaturated fatty acid supplementation or selective soluble epoxide hydrolase inhibition suppresses rough LPS-accelerated glomerulonephritis in lupus-prone mice. *Front Immunol* **14**, 1124910, doi:10.3389/fimmu.2023.1124910 (2023).

37 Pickard, J. M., Zeng, M. Y., Caruso, R. & Núñez, G. Gut microbiota: Role in pathogen colonization, immune responses, and inflammatory disease. *Immunol Rev* **279**, 70-89, doi:10.1111/imr.12567 (2017).

38 Hakansson, A. & Molin, G. Gut microbiota and inflammation. *Nutrients* **3**, 637-682,

doi:10.3390/nu3060637 (2011).

39 Al Bander, Z., Nitert, M. D., Mousa, A. & Naderpoor, N. The Gut Microbiota and Inflammation: An Overview. *Int J Environ Res Public Health* **17**, doi:10.3390/ijerph17207618 (2020).

40 Glorieux, G., Gryp, T. & Perna, A. Gut-Derived Metabolites and Their Role in Immune Dysfunction in Chronic Kidney Disease. *Toxins (Basel)* **12**, doi:10.3390/toxins12040245 (2020).

41 Mirfeizi, Z., Mahmoudi, M. & Faridzadeh, A. Probiotics as a complementary treatment in systemic lupus erythematosus: A systematic review. *Health Sci Rep* **6**, e1640, doi:10.1002/hsr2.1640 (2023).

42 Huber-Ruano, I. *et al.* Orally administered *Odoribacter laneus* improves glucose control and inflammatory profile in obese mice by depleting circulating succinate. *Microbiome* **10**, 135, doi:10.1186/s40168-022-01306-y (2022).

43 Fang, J. *et al.* Mechanistic study of *Dendrobium huoshanense* polysaccharides improving ulcerative colitis by promoting *Lachnoclostridium edouardi* metabolism of short-chain fatty acids. *J Ethnopharmacol* **362**, 121321, doi:10.1016/j.jep.2026.121321 (2026).

44 Kushkevych, I. *et al.* Comparison of microbial communities and the profile of sulfate-reducing bacteria in patients with ulcerative colitis and their association with bowel diseases: a pilot study. *Microb Cell* **11**, 79-89, doi:10.15698/mic2024.03.817 (2024).

45 Huang, G. *et al.* *Desulfovibrio vulgaris* caused gut inflammation and aggravated DSS-induced colitis in C57BL/6 mice model. *Gut Pathog* **16**, 39, doi:10.1186/s13099-024-00632-w (2024).

46 Husain, N., Kumar, A., Anbazhagan, A. N., Gill, R. K. & Dudeja, P. K. Intestinal luminal anion transporters and their interplay with gut microbiome and inflammation. *Am J Physiol Cell Physiol* **328**, C1455-c1472, doi:10.1152/ajpcell.00026.2025 (2025).

47 Shamsuzzaman, M., Dahal, R. H., Kim, S. & Kim, J. Genome insight and probiotic potential of three novel species of the genus *Corynebacterium*. *Front Microbiol* **14**, 1225282, doi:10.3389/fmicb.2023.1225282 (2023).

48 Zhou, Z. *et al.* Critical roles of NLRP3 inflammasome in IL-1 β secretion induced by *Corynebacterium pseudotuberculosis* in vitro. *Mol Immunol* **116**, 11-17, doi:10.1016/j.molimm.2019.09.016 (2019).

49 Zhou, H., Huang, D., Sun, Z. & Chen, X. Effects of intestinal *Desulfovibrio* bacteria on host health and its potential regulatory strategies: A review. *Microbiol Res* **284**, 127725, doi:10.1016/j.micres.2024.127725 (2024).

50 Wagenaar, C. A. *et al.* The Effect of Dietary Interventions on Chronic Inflammatory Diseases in Relation to the Microbiome: A Systematic Review. *Nutrients* **13**, doi:10.3390/nu13093208 (2021).

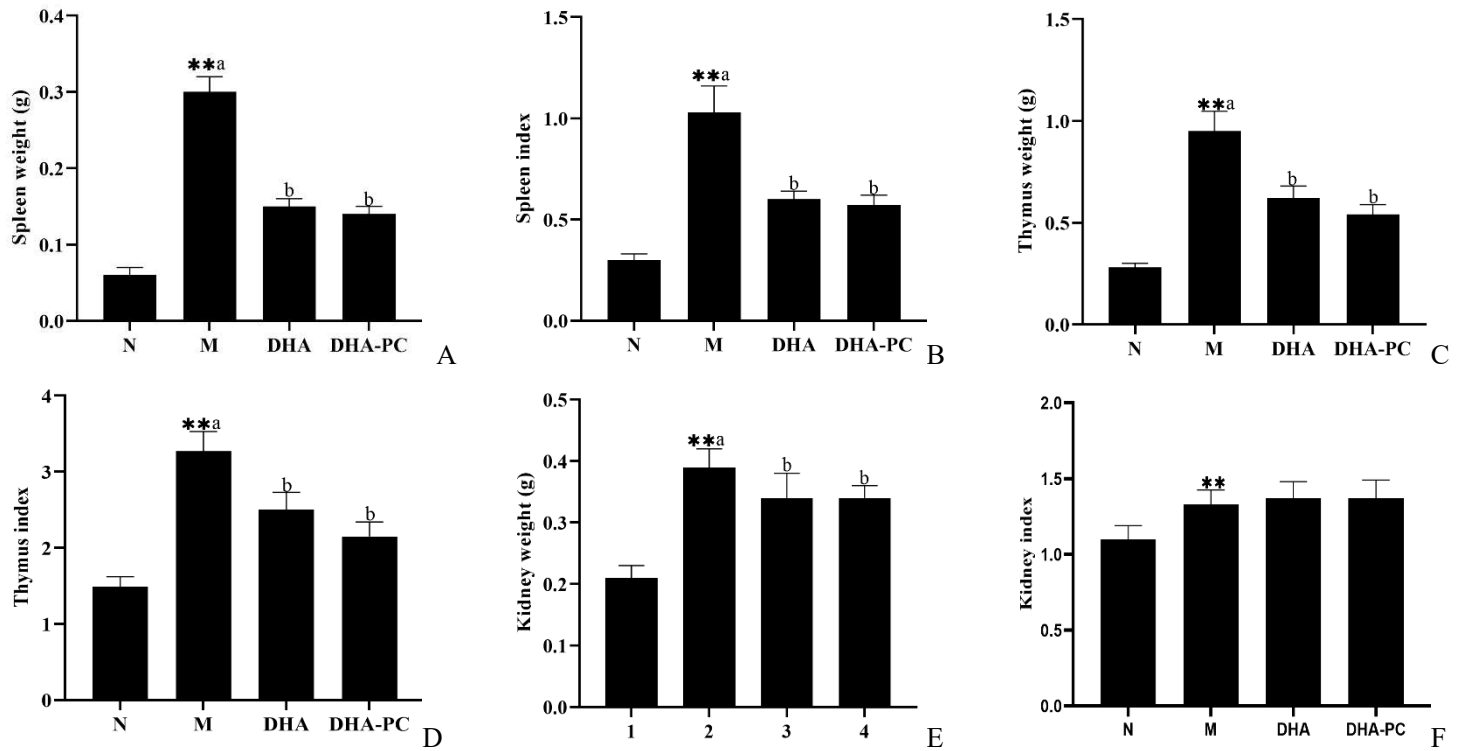
51 Mafra, D. *et al.* Food as medicine: targeting the uraemic phenotype in chronic kidney disease. *Nat Rev Nephrol* **17**, 153-171, doi:10.1038/s41581-020-00345-8 (2021).

52 Qu, M. H. *et al.* Docosahexaenoic Acid-Phosphatidylcholine Improves Cognitive Deficits in an A β 23-35-Induced Alzheimer's Disease Rat Model. *Curr Top Med Chem* **16**, 558-564, doi:10.2174/1568026615666150813144437 (2016).

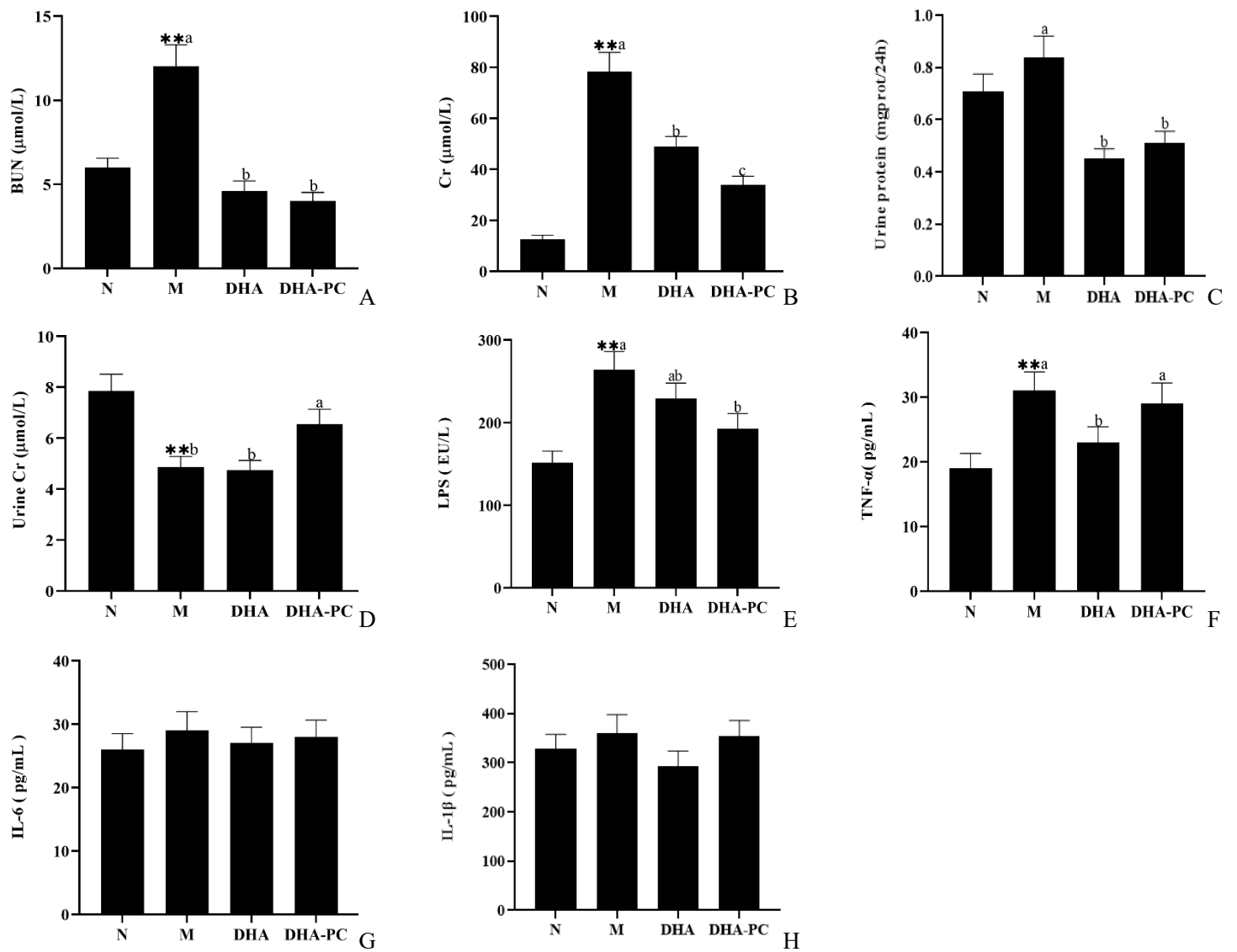
53 Liu, X. *et al.* Eicosapentaenoic acid-enriched phospholipid ameliorates insulin resistance and lipid metabolism in diet-induced-obese mice. *Lipids Health Dis* **12**, 109, doi:10.1186/1476-511x-12-109 (2013).

54 Shi, H. *et al.* Protective Effects of DHA-PC against Vancomycin-Induced Nephrotoxicity through the Inhibition of Oxidative Stress and Apoptosis in BALB/c Mice. *J Agric Food Chem*

579 **66**, 475-484, doi:10.1021/acs.jafc.7b04565 (2018).
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581



582 Figure 1. Effects of different samples on the immune organs and kidneys of mice. A, spleen weight;
 583 B, spleen index; C, thymus weight; D, thymus index; E, kidney weight; F, kidney index. All data
 584 were expressed as mean $\pm$ S.E.M. for 8 mice. **Significantly different compared to normal group
 585 ($p < 0.01$). Different letters represented significant differences between experimental groups, and
 586 means with the same letter were not significantly different ($P < 0.05$).



587 Figure 2. Effects of different samples on the levels of BUN(A), Cr(B), Urine protein(C), Urine
588 Cr(D), LPS(E), TNF-α(F), IL-6(G), and IL-1β(H) in serum of mice. All data were expressed as mean
589 ± S.E.M. for 8 mice. **Significantly different compared to normal group ($p < 0.01$). Different letters
590 represented significant differences between experimental groups, and means with the same letter
591 were not significantly different ($P < 0.05$).

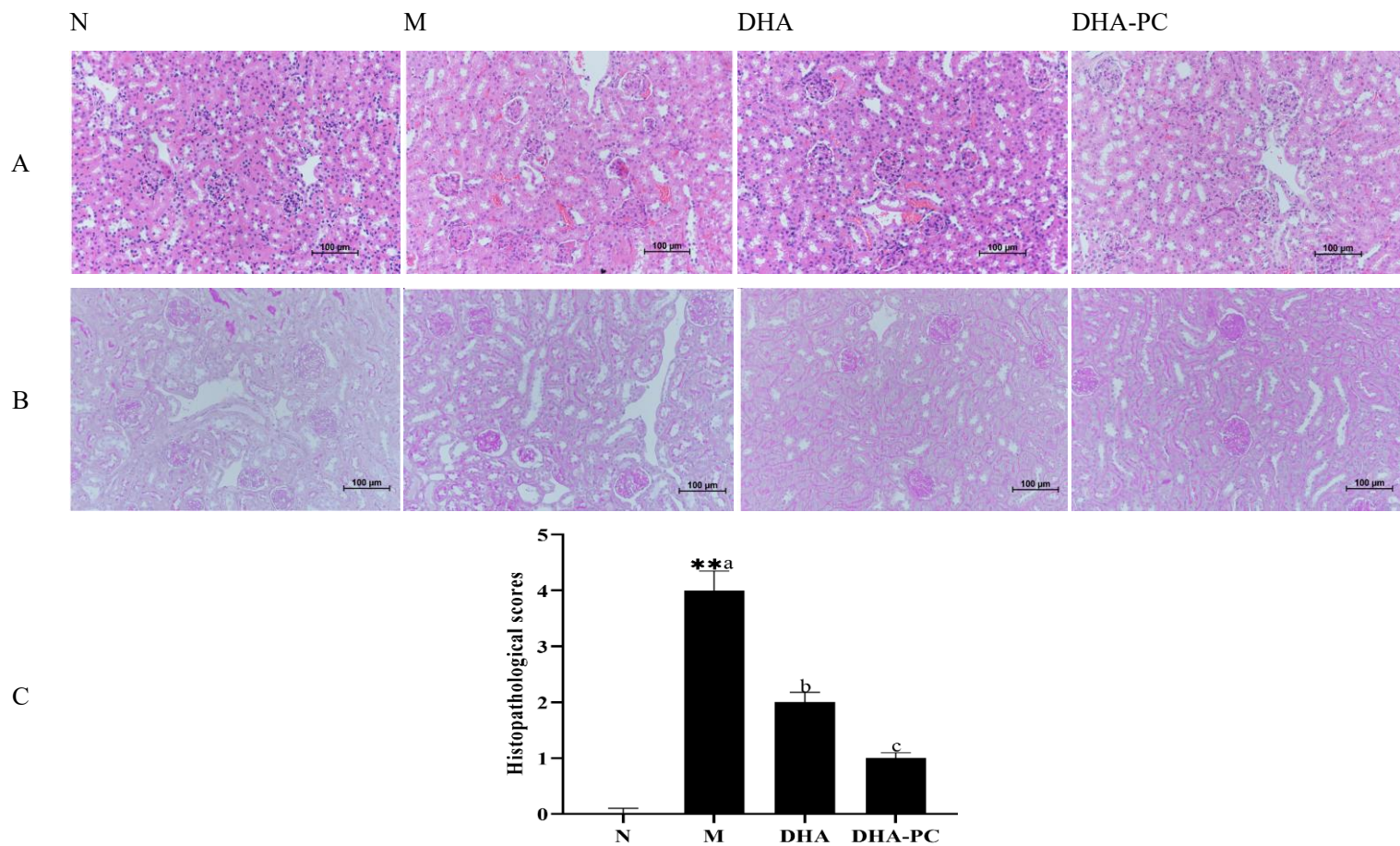


Figure 3. Histopathological results of different samples against lupus nephritis mice as assessed by H&E(A) and PAS(B) staining of kidney tissues (100× magnification); the results of total histopathological scores reflecting tubular damage in each group (C). All data were expressed as mean ± S.E.M. for 8 mice. **Significantly different compared to normal group ($p < 0.01$). Different letters represented significant differences between experimental groups, and means with the same letter were not significantly different ($P < 0.05$).

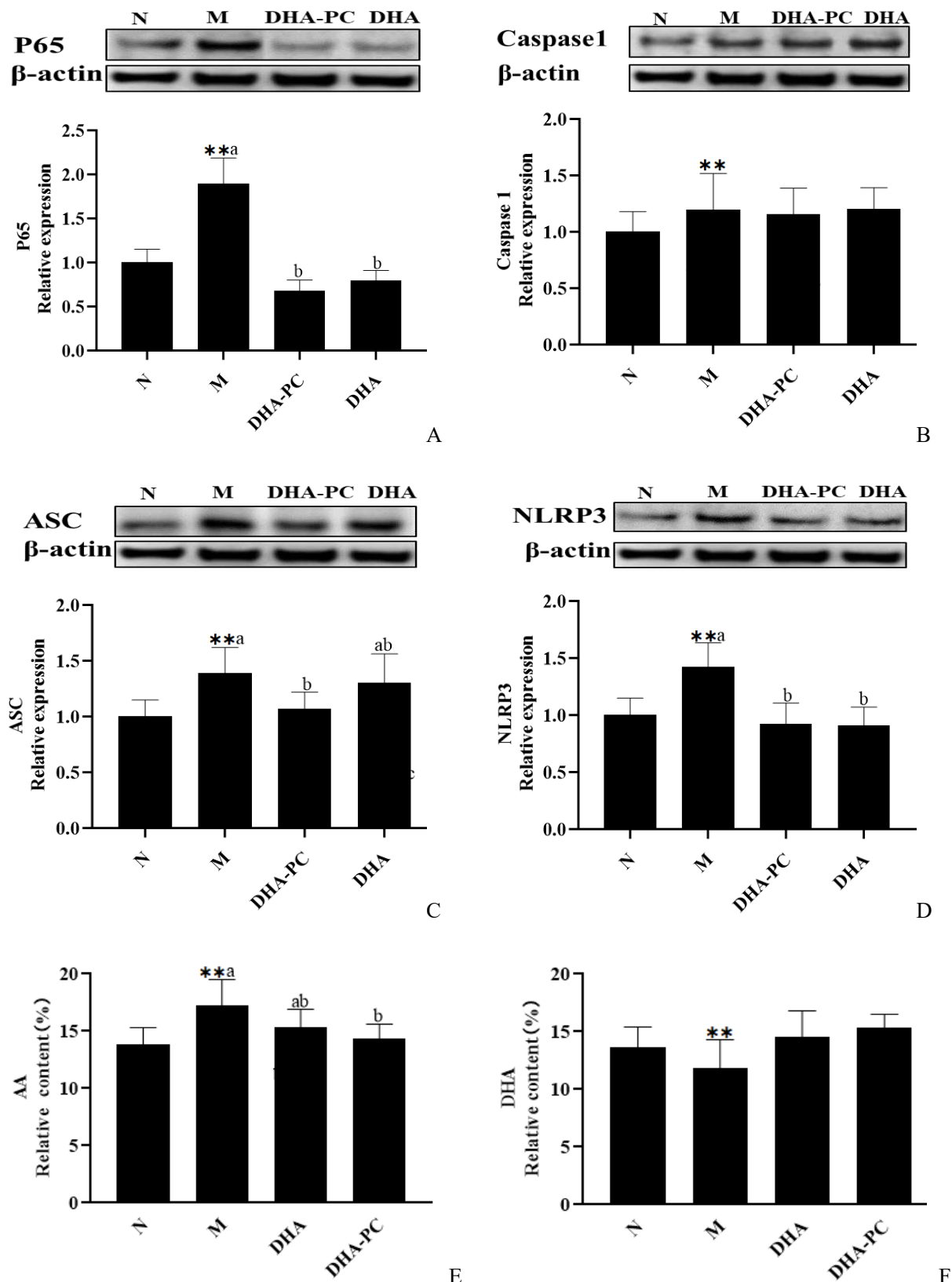


Figure 4. Expressions of P65, Caspase1, ASC, and NLRP3 in the kidney were analyzed by Western blotting with β -actin as the loading control of total proteins (A-D); effects of different samples on the relative levels of AA(E), and DHA(F) in the kidney of mice. The band density was quantified

601 by scanning densitometry. All data were expressed as mean $\pm$ S.E.M. for 8 mice. * **Significantly
602 different compared to normal group ($P < 0.01$). Different letters represented significant differences
603 between experimental groups, and means with the same letter were not significantly different ($P <$
604 0.05).

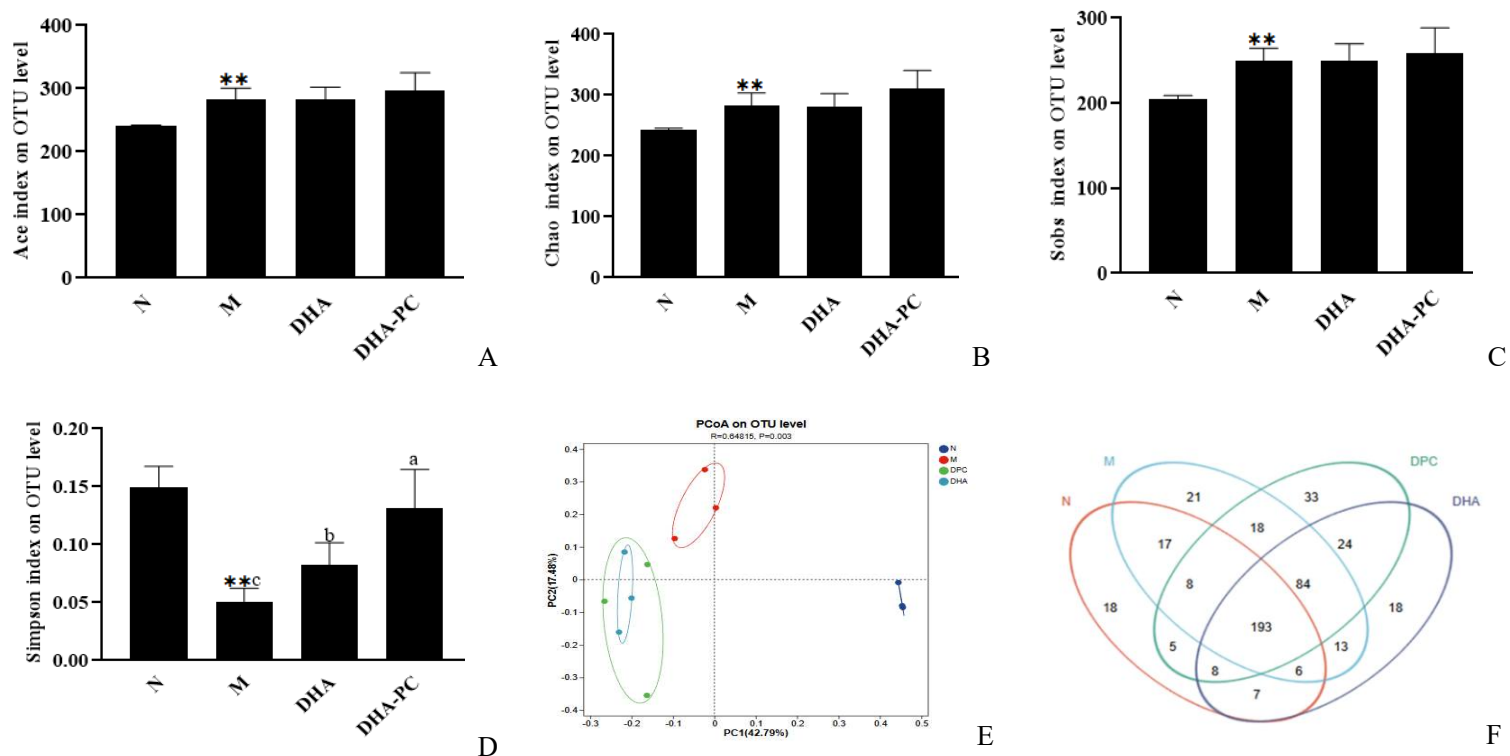
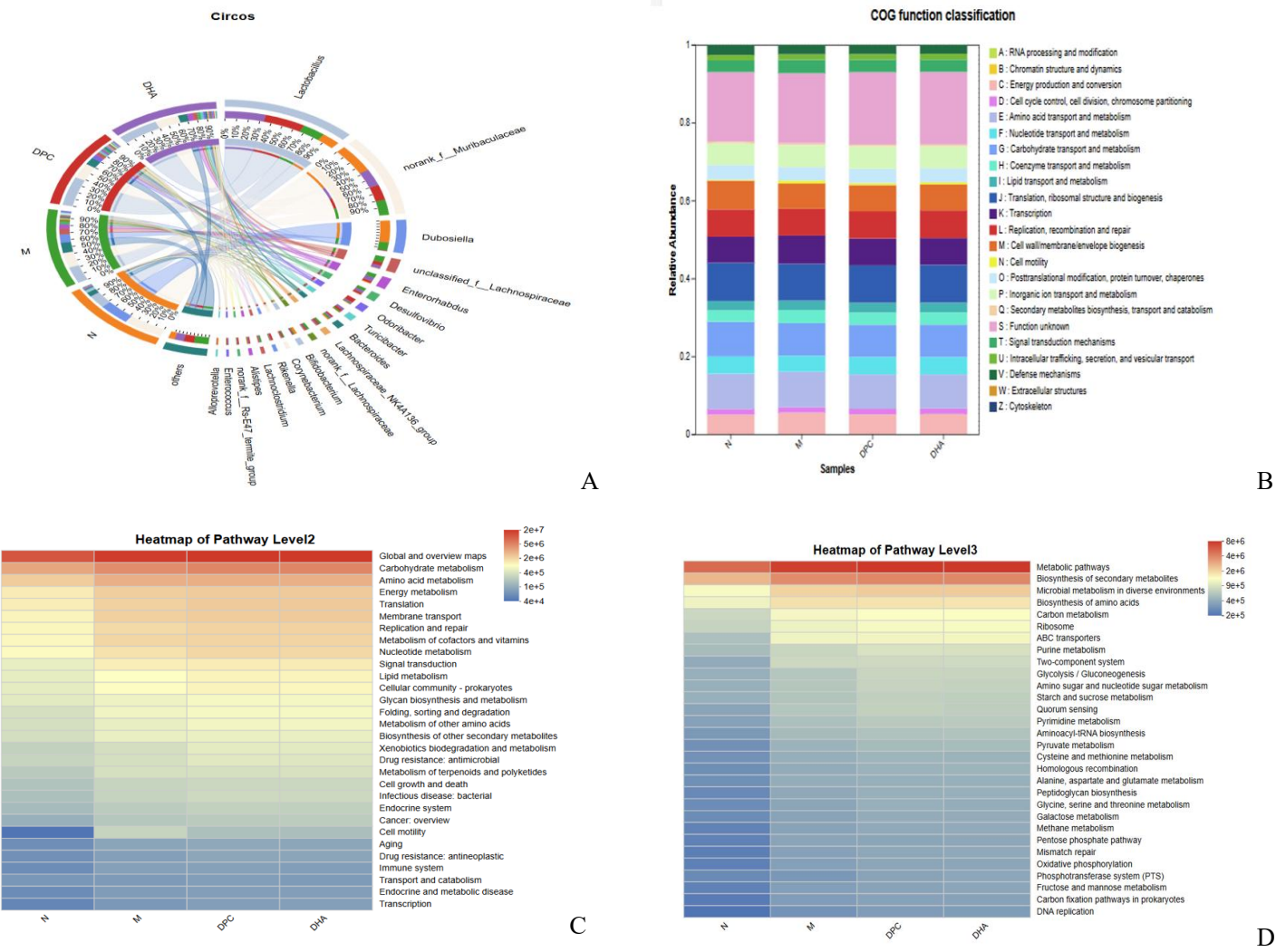
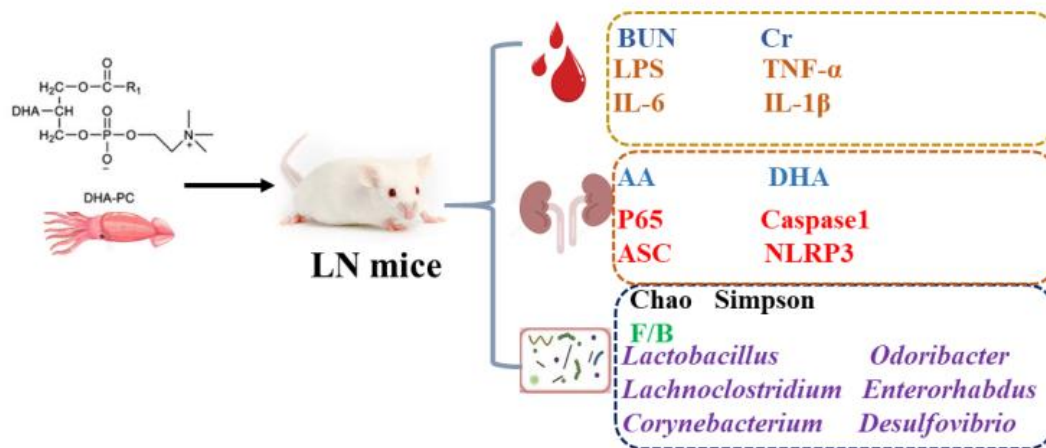


Figure 5. Effects of different samples on the change of alpha diversity index (A-D), PcoA(E), and Venn diagram(F) in mice. PCR was performed on the V3-V4 hypervariable region of the 16S rRNA gene on an Ion S5TM XL platform and 600 bp single-end reads were generated. For each representative sequence, the Silva Database was used based on Mothur algorithm to annotate taxonomic information. **Significantly different compared to normal group ($P < 0.01$). Different letters represented significant differences between experimental groups, and means with the same letter were not significantly different ($P < 0.05$).



618 Figure 7. Effects of different samples on the change of Circos results on the genus in mice(A);
619 Functional prediction using PICRUST2 of COG function classification(B), KEGG functional
620 heatmap pathway (C, D) of different pathway levels in mice.



621

622 Figure 8. The potential molecular mechanisms by which DHA-PC improves lupus nephritis

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