Fatty liver and chronic kidney disease: Novel mechanistic insights and therapeutic opportunities

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Running title: NAFLD and CKD

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Abstract

Chronic kidney disease (CKD) is a risk factor for end-stage renal disease (ESRD) and cardiovascular disease (CVD). A substantial proportion of CKD patients receiving standard-of-care therapy develops ESRD or CVD and mortality in CKD remains unchanged. These data suggest key pathogenetic mechanisms underlying CKD progression go unaffected by current treatments. Growing evidence suggests non-alcoholic fatty liver disease (NAFLD) and CKD share common pathogenetic mechanisms and potential therapeutic targets, which will be discussed. Common nutritional conditions predisposing to both NAFLD and CKD include excessive fructose intake and vitamin D deficiency. Modulation of nuclear transcription factors regulating key pathways of lipid metabolism, inflammation and fibrosis, including Peroxisome Proliferator-activated Receptors (PPARs) and Farnesoid X Receptor (FXR) is advancing to stage III clinical development. The relevance of epigenetic regulation in the pathogenesis of NAFLD and CKD is also emerging, and modulation of miRNA21 is a promising therapeutic target.

While single antioxidant supplementation yielded variable results, modulation of key effectors of redox regulation and of molecular sensors of intracellular energy, nutrient or oxygen status gave promising preclinical results.

Other emerging therapeutic approaches target key mediators of inflammation, like chemokines, and of fibrogenesis, like galectin-3, or gut dysfunction through gut microbiota manipulation and incretin based therapies. Furthermore, NAFLD *per se* affects CKD through lipoprotein metabolism and hepatokine secretion and, conversely, targeting the renal tubule by Sodium-Glucose Linked Transporter-2 inhibitors can improve both CKD and NAFLD.

Implications for the treatment of NAFLD and CKD are discussed in light of this new therapeutic armamentarium.

Epidemiological evidence linking NAFLD and CKD

Chronic kidney disease (CKD) affects up to 8% of the world's adult population, with its prevalence increasing in a population that is ageing and beset by lifestyle-associated diseases such as obesity, metabolic syndrome, diabetes, hypertension, and smoking (1).

CKD may progress to end-stage renal disease (ESRD) and is an important cardiovascular disease (CVD) risk factor: importantly, most patients with CKD die from CVD before renal replacement therapy is initiated (1).

There is potential for improving recognition and treatment of CKD: in the Third National Health and Nutrition Survey (NHANES III), awareness among stage 3 CKD patients was lower than 8% (1). Furthermore, a substantial proportion of CKD patients receiving standard-of-care therapy develop ESRD or CVD and all-cause mortality remains unchanged in CKD population (2). These data suggest key pathogenetic mechanisms underlying renal disease progression go unaffected by current treatment and prompt search for easily identifiable risk factors and novel pharmacological targets.

The presence and severity of non-alcoholic fatty liver disease (NAFLD) has been recently related to the incidence and stage of CKD (3), independently of traditional CKD risk factors; conversely, the presence of CKD increased overall mortality in patients with NAFLD as compared to the general population (4). Further supporting a pathogenic link between NAFLD and CKD, NASH-related cirrhosis has a higher risk of renal failure than other aetiologies of cirrhosis, is an increasing indication for simultaneous liver-kidney transplantation and an independent risk factor for kidney graft loss and CVD (5,6).

Colelctively, these data suggest common pathogenic mechanisms subtend both liver and kidney injury and could be targeted to retard the progression of both NAFLD and CKD.

Potential pathogenic mechanisms contributing to NAFLD to CKD and therapeutic implications

Nutritional factors in NAFLD and CKD: fructose and vitamin D intake

Dietary intake of fructose, the main constituent of sugar-sweeteners, increased 2-fold over the last decade (7). Fructose may contribute to liver and kidney injury through several mechanisms, including uric acid overproduction and consistently, uric acid lowering agents improved fructose-induced experimental NAFLD and CKD (**Table 1**) (8,9). On this basis, the impact of xanthine oxidase inhibitors on CKD progression is being evaluated in the trials CKD-FIX (clinicaltrials.gov ID: NCT12611000791932) and FEATHER (FEbuxostat versus placebo rAndomized controlled Trial regarding reduced renal function in patients with Hyperuricemia complicated by chRonic kidney disease stage 3, UMIN ID: UMIN000008343) (**Table 2**).

Vitamin D attracted considerable interest because of its "pleiotropic" functions, with roles in regulation of cell proliferation, differentiation, immunity, inflammation, fibrogenesis and metabolism (**Table 1**), concurrent with au unsuspectectedly high prevalence of vitamin D deficiency, approaching 25% of the general adult population (10). Vitamin D deficiency has been linked to the pathogenesis and severity of NAFLD and CKD by observational and experimental data(**Table 1)** (11-13). However, the few trials with vitamin D supplementation yielded mixed results and the benefit of vitamin D supplementation remains uncertain (14). Concerning NAFLD and CKD, it should be noted that these conditions are characterized by vitamin D resistance, partly determined by impaired hepatic 25 hydroxylation and increased renal tubular 25(OH)D loss, and may require higher dose supplementation of vitamin D, calcitriol or vitamin D receptor agonists (e.g. paricalcitol) to be overcome(15, 16). The effects of vitamin D supplementation in CKD and in NASH are being evaluated in RCTs NCT00893451, NCT01623024 and NCT02098317.

Reversing ectopic fat deposition by targeting nuclear transcription factors in NAFLD and CKD

NAFLD and CKD are characterized by ectopic toxic lipid accumulation, which is determined by an extensive derangement in hepatic and renal lipid metabolism and triggers lipoperoxidative stress, cell apoptosis, inflammation and fibrosis (17,18,19)(**Figure 1**). These abnormalities are subtended by an

extensive deregulation of nuclear transcription factors regulating lipid metabolism, inflammation and fibrogenesis, including Peroxisome Proliferator-Activated Receptor (PPAR)-α, PPAR-δ and PPAR-γ, Sterol Regulatory Binding Protein (SREBP)-2 and Farnesoid X Receptor (FXR), which represent an attractive target for the treatment of NAFLD and CKD(20,21)**(Figure 1).**

Based on the finding that PPAR-α and PPAR-δ are down-regulated in NAFLD and CKD(20,22), potent, selective PPAR-α, PPAR-δ and dual PPAR-α/δ agonists (K-877, GW501516, MBX-8025 and GFT505, respectively) were evaluated in these two conditions, with encouraging results in preclinical models (22,23,24). Some of these compounds advanced to the clinical stage of development, and GFT505, a dual PPARα/δ agonist, improved steatohepatitis, fibrosis and glyco-lipid profile in the recently completed GOLDEN-505 trial (25)(**Table 2**).

The PPAR-γ agonists thiazolidinediones are another pharmacological class that significantly improved NASH and albuminuria in clinical trials (26, 27), but whose clinical use was limited by their side effects. These drawbacks prompted development of new compounds, including dual PPAR-α/γ agonsists, which mantained the therapeutic effectiveness of PPAR-γ agonists but were devoid of their unwanted effects **(Figure 1).** Saroglitazar, a potent PPAR-α/γ agonist , did not induce weight gain, peripheral edema or other adverse events after 1 year (28) and improved markers of NAFLD in diabetic patients (29), while aleglitazar slowed eGFR decline in diabetic nephropathy (30)(**Table 2**). A small, phase IIa trial enrolling biopsy-proven NASH patients has been completed but results are not available yet (CTRI registration no.: CTRI/2010/091/000108).

Larger and longer RCTs are needed to evaluate long-term clinical safety and effectiveness of these compounds in diabetic and non-diabetic patients.

Among different lipotoxic species accumulating in NAFLD and CKD, free cholesterol is believed to play a key pathogenic role in liver and renal injury (31,32). Ectopic cholesterol accumulation is driven by an inappropriate upregulation of transcription factor SREBP-2, with consequently increased cholesterol synthesis, influx and retention and reduced cholesterol excretion by liver and renal cells(31,32)(**Figure 1**). Such pervasive deregulation in all steps of cholesterol metabolism may diminish the effectiveness of available cholesterol-lowering drugs, that target single steps in

cholesterol metablism (33). Therefore, modulation of SREBP-2 activity represents an attractive therapeutic tool: while selective SREBP-2 antagonists are under development, several natural antioxidants, like myricetin, repressed SREBP-2 expression and ameliorated cholesterol-induced inflammation and fibrosis in experimental models(32)(**Table 2).**

Farnesoid X Receptor (FXR) is a nuclear transcription factor with prominent insulin sensitizing, anti-lipogenic, anti-inflammatory and antifibrotic properties; furthermore, FXR activation improves also endothelial function (34,35,36)(**Figure 1**).

FXR expression is down-regulated in the liver and kidney of NAFLD and CKD patients, respectively, and inversely related to disease severity. On this basis, potent semi-synthetic bile acid FXR agonists have been developed (**Table 2**). Obeticholic acid (OCA, or INT-747), a semi-synthetic chenodeoxycholic acid derivative, improved liver histology in the phase IIa multicenter, randomized "FXR Ligand NASH Treatment (FLINT)" trial and ameliorated renal histology and proteinuria in nutritional models of CKD (35,36). Several issues remain, however, including the effectiveness of FXR agonists in nondiabetic subjects and the impact of HDL-C reduction on long-term CVD risk of these patients.

Epigenetic regulation in NASH and CKD

MicroRNAs (miRNAs) are small (~22 base-pairs) endogenous non-coding RNAs

that regulate gene expression of at least 60% of protein-coding genes.

MicroRNA recognize mRNA targets through sequence complementarity between the miRNA and binding sites in the 3′ untranslated regions (3′ UTRs) of the target mRNAs or through interaction with

RNA-binding proteins.

MicroRNAs regulate gene expression in one of two ways, depending on the degree of complementarity between the miRNA and its target: miRNAs that bind to mRNA targets with

imperfect complementarity block target gene expression via translational silencing, while miRNAs binding their mRNA targets with perfect complementarity enhance target gene expression (38, 39).
To date, over 2500 miRNAs have been identified in the human genome and each miRNA can regulate several hundred target genes involved in diverse developmental and cellular processes, including cellular metabolism proliferation, differentiation and apoptosis.
Several miRNAs have been found to be dysregulated in NAFLD and CKD (38,39). On this basis, approaches inhibiting overexpressed miRNAs by antisense oligonucleotides (ASOs) or restoring the expression of down-regulated miRNAs by synthetic miRNA mimics have been attempted.
miRNA21, in particular, is an attractive target, as it regulates key metabolic, pro-inflammatory and profibrogenic pathways and its hepatic and renal overexpression in NASH and CKD leads to PPAR-α downregulation, SREBP-2 upregulation, mitochondrial dysfunction and pro-fibrogenic HSC activation and proximal tubular cell epithelial-to-mesenchimal transition(EMT)(40,41).
Consistently, in experimental models of NASH and CKD, anti-microRNA-21 ASOs induced weight loss, normalized metabolic dysregulation, and improved hepatic and renal inflammation and fibrosis, effects at least partly mediated by PPAR-α upregulation (40,41).
Despite these premises, several issues remain, including the stability and selective delivery of the pharmacological modulators to the target organs,and long-term safety of this approach, as miRNAs regulate also cell proliferation and cell cycle progression in diverse tissues and long-term consequences of its modulation on tumor onset and progression are unclear (39,40).

Role of cellular energy, oxygen and nutrient sensors

In mammals, cellular metabolism is finely orchestrated by molecular sensors of energy, nutrient and oxygen status to adapt to changing substrate availability. The dysregulation of some of these sensors, including 5-AMP-activated protein kinase (AMPK), hypoxia-inducible factor (HIF)-1α and mammalian target of rapamicin (mTOR), has been implicated in the pathogenesis of NAFLD and CKD, and could be targeted for the treatment of NAFLD and CKD.

AMPK is an ubiquitous kinase that preserves cell survival under calorie restriction or high energy demand (42). In response to cellular ATP depletion or to an increase in AMP/ATP ratio, AMPK activation enhances substrate oxidation and has anti-oxidant, anti-inflammatory and antifibrotic effects (43-45) (**Figure 2A**). On this basis, several natural AMPK activators were assessed in preclinical models of NASH and CKD, with encouraging results (43-45), and the synthetic AMPK-activator oltipraz is being evaluated in non-cirthotic NAFLD patients (clinicaltrials.gov ID: NCT01373554)(**Table 2**).

HIF-1α is a ubiquitous oxygen-sensitive protein that regulates transcription of genes involved in metabolic adaptation, energy conservation, angiogenesis, and cell survival in response to cellular hypoxia and to non-hypoxic stimuli like cholesterol overload (46). Inappropriate HIF-1α activation following stimuli like chronic intermittent hypoxia and cholesterol overload has been involved in NASH pathogenesis (47,48), while chronic hypoxia has been implicated in renal injury in early diabetic and obesity-related CKD (49, 50) (**Figure 2B).**

On this basis, small-molecule synthetic HIF-1α inhibitors, including YC-1, have been developed and showed potent anti-fibrotic properties in experimental models of NASH (51)(**Table 2**).

Several issues remain, however: in some studies HIF-1α activation protected against kidney injury (52), suggesting HIF-1α may not be the sole, or the best therapeutic target for reversing celluar effects of hypoxia in CKD.

mTOR is a serine/threonine kinase that, in response to changes in cellular nutrient levels, growth factors like insulin and IGF, and other stressors, associates with companion proteins to form two distinct signaling molecular complexes, mTOR complex 1 (mTORC1) and mTORC2 (53). mTORC1 has been more extensively studied and has been found to promote cellular anabolism by stimulating synthesis of protein, lipid, and nucleotides and blocking catabolic processes. Inappropriate mTORC1 activation in NAFLD and CKD inhibits autophagy (53,54) and promotes insulin resistance, ectopic lipid accumulation, lipotoxicity and proinflammatory monocyte recruitment in the liver and kidney (55,56)(**Figure 2B**). Consistently, mTORC1 inhibition reversed metabolic abnormalities and

attenuated lipid accumulation, inflammation, and fibrosis in diverse models of NASH and CKD (57,58) and may represent a therapeutic option for NAFLD and CKD (59)(**Table 2**).

Targeting redox regulation in the pathogenesis of NAFLD and CKD

Although increased oxidative stress is believed to play a central role in NASH and CKD progression, single antioxidant supplementation strategies yielded variable results (26), and other approaches targeting common effectors of redox regulation, like Apoptosis Signal-Regulating Kinase 1 (ASK1) and Nuclear Erythroid 2-related Factor 2 (Nrf2), are being investigated.

ASK1 is a serine/threonine kinase belonging to the mitogen-activated protein kinase kinase kinase .(MAP3Ks) family, which is activated in response to stresses like ROS, TNF-α, lipopolysaccharide (LPS) and ER stress (60). ASK1 activates downstream terminal MAPK kinases p38 and c-Jun N-terminal kinase (JNK), which promote insulin resistance, cell death, proinflammatory cytokine/chemokine production, and fibrogenesis(60)(**Figure 3**).

Recent data implicated ASK1 activation in oxidative stress-induced inflammation and fibrogenesis in NASH and CKD, and pharmacological ASK1 inhibition prevented diet-induced NASH and halted progression of diabetic and nondiabetic experimental CKD(61,62). On this basis, the highly selective oral ASK1 inhibitor GS-4997 is being evaluated in NASH patients with moderate-advanced fibrosis (ClinicalTrials.gov ID: NCT02466516) and in diabetic patients with stage 3/4 nephropathy (ClinicalTrials.gov ID: NCT02177786)(**Table 2**).

Several issues need to be clarified: ASK1 inhibition did not improve podocyte loss and albuminuria in experimental diabetic CKD, suggesting this kinase is not central for glomerular injury (62). Furthermore, the impact of ASK1 inhibitors in nondiabetic CKD is unknown.

Nrf2 is a transcription factor expressed ubiquitously in human tissues and most abundantly in the liver (63), where it regulates the expression of several antioxidant and detoxifying enzymes and has direct metabolic, anti-inflammatory and pro-autophagic actions (63)(**Figure 3**). Under basal conditions Nrf2 is kept transcriptionally inactive through binding to its inhibitor, Kelch-like ECH-associated protein 1

(KEAP1) (63). ROS and reactive nitrogen species (RNS) interact with KEAP1 and cause loosening of Nrf-2, which translocates to the nucleus and modulates transcription of its target genes **(Figure 3**). The relevance of Nrf2 in the pathogenesis of liver and kidney injury has emerged in diverse diet-induced models of NASH and CKD, where disease progression was accelerated by Nrf2 deletion and prevented by Nrf2 activators (64,65). On this basis, several natural and synthetic small-molecule Nrf2 activators are currently being evaluated in NASH and CKD patients (**Table 2**)**:** following early encouraging data, clinical development of bardoxolone methyl, a synthetic Nrf2 activator, was interrupted for safety concerns related to heart failure (66). A reanalysis of the potential risk/benefit ratio of the drug for the Japanese population and the observation that most of the severe adverse effects occurred in the first month of therapy prompted initiation of a dose-escalating RCT in Japan (clinicaltrials.gov ID: NCT02316821). Another potent synthetic Nrf2 activator, oltipraz, is being evaluated in NAFLD patients (clinicaltrials.gov ID: NCT01373554).

Targeting molecular effectors of inflammation and fibrosis

Chemokines are small proteins that regulate leukocyte migration into tissues and consequent inflammation, tissue remodeling and fibrosis (67). Among the over 40 chemokine ligands and 20 chemokine receptors currently identified, chemokine (C-C motif) ligand 2 (CCL2, or monocyte chemoattractant protein-1, MCP-1) and its receptor CCR2 have been implicated in the pathogenesis of NASH and CKD. In NAFLD, hepatic cells and adipocytes secrete CCL2, which attracts pro-inflammatory cells to the liver to promote NASH development (68,69), while genetic or pharmacological inhibition of CCL2/CCR2 axis reverses steatohepatitis and advanced hepatic fibrosis (69). In the kidney, tubule cells and podocytes secrete chemokines CCL2 and CCL5 in response to diverse pro-inflammatory stimuli to promote tubulo-interstitial inflammation and fibrosis, which are all reversed by chemokine antagonists (70). On this basis, chemokine antagonists are advancing to clinical stage of development: the small molecule CCR2 antagonist CCX140-B reduced albuminuria and slowed eGFR decline in diabetic nephropathy (71), while the dual chemokine receptor CCR2/CCR5 antagonists BMS-813160, PF-04634817 and cenicriviroc are being evaluated in

diabetic nephropathy(ClinicalTrials.gov ID**:** NCT01752985**,** NCT01712061) and in NASH (ClinicalTrials.gov ID**:** NCT02217475)(**Table 2**).

Galectin-3 is a lectin broadly expressed by immune and epithelial cells, where it localizes mainly in the cytoplasm, but also in the nucleus, in the cell surface and in the extracellular space (72). Galectin-3 regulates cell proliferation, apoptosis, cell adhesion and affinity for advanced glycation end-products(AGEs), exerting multiple and sometimes contrasting effects depending on its cellular location, cell type and mechanism(s) of injury(72)(**Figure 3**).

Galectin-3 is upregulated in the liver and kidney of patients with NASH and CKD and correlates with the severity of liver and renal disease (73,74). Furthermore, circulating galectin-3 levels predict renal function decline, cardiovascular and all-cause mortality in CKD patients (75). Consistent with epidemiological data, functional galectin-3 manipulation disclosed important pro-inflammatory and profibrotic effects of ths lectin (72-74)**.**

On this basis, pharmacological galectin-3 inhibition with small molecule competitive inhibitos, including GR-MD-02(galactoarabino-rhamnogalaturonan), GM-CT-01(galactomannan), and N-acetyllactosamine, prevented hypertensive nephropathy (76) and reversed diet-induced NASH and cirrhosis (77). GR-MD-02 was well tolerated and improved markers of hepatic fibrosis in a phase I RCT enrolling NASH patients with advanced fibrosis(ClinicalTrials.gov ID: NCT01899859), while a phase IIa RCT is exploring the effect of the galectin-3 antagonist GCS-100 on eGFR in CKD (ClinicalTrials.gov ID**:**: NCT01843790) (**Table 2).**

Data on galectin-3 inhibition are not univocal, however, and galectin-3 deletion exacerbated systemic inflammation, hyperglycemia, liver and kidney injury in diet-induced obese rodents (78). It has been suggested that inhibition of AGE uptake by the liver, which clears >90% of these end-products from the circulation, promotes their systemic accumulation and RAGE-mediated uptake by other tissues, thereby aggravating extrahepatic toxicity of these molecules. As both NASH and CKD are characterized by AGE accumulation, a better understanding of the impact of galectin-3 inhibitors on AGE-mediated tissue injury is warranted *in vivo.*

The gut connection: targeting incretins and gut microbiota for the treatment of NAFLD and CKD

Incretin based therapies, including glucagon-like peptide-1 (GLP-1) mimetics and dipeptidyl peptidase-4 (DPP-4) inhibitors, increase insulin release from the pancreas, reduce glucagon production and possess numerous extrapancreatic metabolic benefits which prompted evaluation for the treatment of NAFLD and CKD(79,80)(**Table 3**).

Preliminary human data suggest incretin mimetics may improve NAFLD: a meta-analysis of 6 RCTs from the "Liraglutide Effect and Action in Diabetes" (LEAD) program found a significant improvement in biochemical and radiological features of steatosis (81). Furthermore, in the " Liraglutide Efficacy and Action in NASH" (LEAN) trial, liraglutide 1.8 mg/day for 48 weeks induced NASH resolution and improved markers of lipotoxicity, inflammation and metabolic dysfunction as compared with placebo (82)(**Table 2**).

Incretin-based therapies have also the potential for nephroprotection independent of improved glycemic control, through several mechanisms(**Table 3).** GLP-1 administration induced natriuresis through inhibition of proximal tubular Na-H exchanger 3 (NHE3) and reduced activation of the AngII axis (83). These actions counteract the increase in proximal tubular Na reabsorption, which has been hypothesized to trigger glomerular hyperfiltration, the functional defect thought central to obesity-associated and diabetic CKD (83). Furthermore, GLP-1 mimetics demonstrated direct renal anti-inflammatory, anti-fibrotic and antioxidative actions(84,85) . Collectively, these properties may explain the attenuated progression of overt diabetic nephropathy in 2 preliminary small RCTs of treatment with liraglutide (84,85).

In addition to inactivating GLP-1, DPP-4 cleaves multiple other peptides, including brain-derived natriuretic peptide (BNP), neuropeptide Y (NPY), peptide YY (PYY) and stromal-derived factor (SDF)-1a(**Table 3**). Thus, the effect of DPP-4 inhibition depends on the actions of the different substrates inactivated in each tissue and organ. This may theoretically explain the slightly lower antti-hypertensive effect observed with DPP-4 inhibitors as compared to GLP-1 mimetics (86).

While the impact of DPP-4 inhibitors on NAFLD has to be assessed, results from the Saxagliptin Assessment of Vascular Outcomes Recorded in Patients with Diabetes Mellitus–Thrombolysis in Myocardial Infarction (SAVOR-TIMI) 53 trial (87) and from other 4 RCTs (88) suggest saxagliptin and linagliptin may reduce the development and progression of albuminuria. Renal effects of linagliptin are currently being investigated in the MARLINA-T2D(efficacy, safety and modifications os albuminuria in T2DM subjects with renal disease with LINAgliptin (clinicaltrials.gov ID: NCT01792518) and in the CARMELINA (Cardiovascular safety and renal microvascular outcome study with LINAgliptin(clinicaltrials.gov ID: NCT01897532)) trials.

In conclusion, incretin-based therapies address several of the pathophysiological mechanisms common to experimental NAFLD and CKD, but their impact on renal disease in non-diabetic CKD is unknown. Furthermore, incretin-based therapies did not affect the risk of CVD, a major cause of death in NAFLD and CKD (89).

The capacity for gut microbiota to interact with host metabolic and immune response and to contribute to the development of obesity-associated disorders is being increasingly recognized (90). Both NAFLD and CKD patients exhibit an altered gut microbiota composition, with a relative decrease in "healthy" Bacteroidetes, Lactobacillaceae and Prevotellaceae families and disruption of the normal gastrointestinal barrier (91,92). The resulting accumulation of gut-derived toxins induces inflammation (93), insulin resistance and ectopic fat deposition in liver and muscle through several mechanisms (**Table 3).** Some of these molecules, including endotoxin, indoxyl-sulphate, p-Cresyl sulphate (p-CS) and trimethylamine-N-oxide(TMAO), have documented clinical relevance for the development and progression of CKD (94-97).

CKD may also *per se* aggravate gut dysbiosis and systemic inflammation through accumulation of uremic toxic metabolites (URMs) normally eliminated by the kidneys, including urea and p-CS. The accumulation of urea may lead to influx into the gastrointestinal lumen, where it is hydrolysed to ammonia by microbial urease, then converted to ammonium hydroxide. Ammonia and ammonium hydroxide promote the growth of urea-metabolizing bacteria at the expense of carbohydrate-fermenting strains and disrupt intestinal epithelial tight junctions, enhancing passage of LPS and other

toxic luminal compounds into the circulation (98). Further highlighting the relevance of this mechanism to systemic inflammation, administration of oral activated charcoal absorbent AST-120 improved intestinal barrier function, and reduced systemic oxidative stress, inflammation and endotoxemia in rodent models of CKD (98). Gut microbiota manipulation with probiotics or prebiotics improved surrogate markers of NAFLD in small RCTs of short duration (99) and reduced URM levels in CKD patients (100). The impact of synbiotic administration on renal function in CKD is being investigated in the SYNbiotics Easing Renal failure by improving Gut microbiologY (SYNERGY) trial(Australian New Zealand Clinical Trials Registry Number: ACTRN12613000493741).

NAFLD as a determinant of CKD: targeting the liver to improve CKD

In NAFLD, liver disease *per se* contributes to kidney injury through several mechanisms: the liver contains up to 80% of all macrophages of the body and the steatotic liver may represent a more relevant source of proinflammatory cytokines than adipose tissue (101). Furthermore, the liver is a central regulator of lipoprotein metabolism and secretes hepatokines like FGF21, which can modulate whole body metabolism and inflammation.

1)hepatic secretion of VLDL, CETP and syndecan-1 in the pathogenesis of atherogenic dyslipidemia and kidney injury.

Atherogenic dyslipidemia is the commonest lipid abnormality in CKD and an independent predictor of the incidence and progression of CKD and of CVD in CKD patients (102,103). Atherogenic dyslipidemia promotes CKD through receptor-mediated uptake of qualitatively abnormal lipoproteins by glomerular and tubulo-interstitial cells (104). NAFLD may promote atherogenic dyslipidaemia through several mechanisms, which represent potential therapeutic targets**:** liver fat accumulation *per se* proportionally increases hepatic secretion rate of large VLDL1, which exchange Tg with cholesterol contained in circulating LDL and HDL particles, resulting in sLDL and HDL3 formation (31). Furthermore, recent studies demonstrated circulating cholesteryl ester transfer protein (CETP)

derives largely from hepatic Kupffer cells and its levels parallel the severity of histological necroinflammations in NASH (105). Intriguingly, CETP inhibitors alleviated high fat diet-induced steatohepatitis and fibrosis, possibly by reducing oxLDL uptake by hepatic Kupffer and stellate cells (106) and represent a potential therapeutic target for the treatment of both NAFLD and CKD(**Table 2**)..

Syndecan-1 is another key mediator of hepatic clearance of triglyceride-rich lipoproteins (107): syndecan-1 is a transmembrane heparan sulfate proteoglycan constitutively bound to hepatocyte membrane, where it binds LPL and apoE through its heparan sulfate chains and internalizes apoE-containing lipoproteins (108). Sulfation by hepatic sulfotransferases and binding to hepatocyte membrane are required for syndecan-1 biological activity. NAFLD is characterized by an increased shedding of syndecan-1 (109), as a result of increased hepatic metalloproteinase activity, and by a defective syndechan-1 sulfation, as a result of defective hepatic sulfotransferase activity (110, 111). These changes in hepatic syndecan-1 metabolism impair TRLP clearance and, accordingly, predicted atherogenic dyslipidemia in CKD (112). Beside mediating hepatic TRLP clearance, syndecan-1 is also a key constituent of endothelial glycocalyx layer and its shedding has been associated with loss of endothelial barrier integrity and endothelial dysfunction across progressive CKD stages (113). Inhibitors of syndecan-1 shedding, including the phospholipid sphingosine-1-phosphate, restored endothelial integrity experimentally (114) and may represent a potential therapeutic tool for the treatment of CKD.

2)Fibroblast growth factor(FGF)21

Fibroblast growth factors (FGFs) are a group of signalling proteins that regulate embryonic development, tissue regeneration, and diverse metabolic functions by binding extracellularly to four cell surface tyrosine kinase FGF receptors(FGFRs 1–4) (115). FGF21 is mainly secreted by the liver and exerts its multiple beneficial metabolic effects by binding to FGFRs in the presence of co-receptor β-Klotho(115): FGF21 administration ameliorates adipose and hepatic insulin sensitivity, suppresses hepatic gluconeogenesis and lipogenesis and enhances FFA oxidation and mitochondrial

function, at least in part by activating the AMPK-SIRT1-PGC-1α pathway (115, 116). Furthermore, FGF-21 has recently demonstrated direct anti-inflammatory and anti-fibrogenic activity by inhibiting the key NF-κB and TGF-β/smad2/3 signaling pathways (117). By virtue of these properties, FGF21 administration improved experimental NASH and CKD (115-117). The clinical development of FGF21 however, is hampered by its short half-life (0.5-5 hr) and by tissue FGF21 resistance, which is subtended by FGFR1 and β-Klotho downregulation (118) and is overcome by pharmacological doses of FGF21. To this aim, engineering of native molecule yielded FGF21 analogs with improved biophysical properties and one of these FGF21 analogs, LY2405319, ameliorated atherogenic dyslipidemia, insulin resistance and adiponectin in obese diabetic patients (119).

Targeting the /renal tubule to improve CKD and NAFLD

Sodium-Glucose Linked Transporter-2 (SGLT2) inhibitors block the activity of the SGLT2 protein, which is expressed in the S1 segment of the renal proximal tubule, leading to substantial glucosuria and a reduction in plasma glucose levels. Experimental evidence demonstrated SGLT2 inhibitors may confer neproprotection independently of their glucose-lowering or blood pressure-lowering properties. SGLT2 inhibitors attenuate glumerular hyperfiltration, which is thought to be the initial pathogenic alteration in diabetic and obesity-related CKD: in diabetes, hyperglycemia induces an increase in SGLT2-mediated proximal tubule NaCl reabsorption. The consequent reduction in NaCl distal delivery to the macula densa decreases tubulo-glomerular feedback-mediated afferent arteriolar vasoconstriction, thereby increasing glomerular afferent-to-efferent arteriolar tone, intraglomerular ultrafltration pressure and GFR (120). Proximal tubule SGLT2 upregulation and increased NaCl reabsorption have been documented in obesity-related CKD, as well, as a result of enhanced sympathetic activity (121,122) and TGFβ1/Smad3 axis activation (123). By virtue of these actions, SGLT2 inhibitors synergize with RAAS inhibitors and their combination may confer incremental renal benefits (124): simultaneous SGLT2–RAAS blockade induces afferent arteriole constriction (SGLT2 inhibition) and efferent arteriole vasodilatation (RAAS blockade), thereby more thoroughly

counteracting early intrarenal haemodynamic abnormalities underlying glomerular hyperfiltration in CKD. Additionally, SGLT2 inhibitors decreased inflammatory and fibrogenic responses, oxidative stress and cell apoptosis in diverse experimental models of CKD(125).

Available data on the impact of SGLT2 inhibitors on CKD derive from analysis of RCTs conducted with efficacy and safety end-points in diabetic population, where SGLT2 inhibitors slowed renal function decline and reduced albuminuria independently of glycemic control (126), while dedicated nephroprotection trials are underway (Evaluation of the effects of Canagliflozinon Renal and Cardiovascular Outcomes in Participants with Diabetic Nephropathy, CREDENCE trial-clinicaltrials.gov ID: NCT02065791).

Beside nephroprotection, SGLT2 inhibitors have also the potential for cardiovascular protection: in the EMPA-REG OUTCOME trial, empagliflozin added on top of standard care was associated with a lower rate of the primary composite outcome of cardiovascular and all-cause mortality (127).

SGLT2 inhibitors may also ameliorate NAFLD: in a post-hoc analysis of a RCT and a small RCT, remogliflozin etabonate and luseogliflozin improved liver fat accumulation and liver enzymes in diabetic patients (128,129). The conclusions of these RCTs are supported by preclinical data, whereby SGLT2 inhibitors prevented diet-induced hepatic steatosis, inflammation and fibrosis, independently of anti-hyperglycemic action (130,131). Potential mechanisms underlying liver-related benefits of these drugs include insulin sensitizing (128), and body fat loss-inducing properties mediated by enhanced lipolysis and fatty acid oxidation(130,131), attenuation of adipose tissue dysfunction and inflammation (132), increased ACE2 activation (133), and intrinsic drug-specific antioxidant and anti-RAGE axis activation properties (134). Furthermore, SGLT2 mRNA expression has also been documented in the liver, where its biological and clinical significance remain unknown (135).

Several issues with SGLT2 inhibitors warrant assessment, including the renoprotective effects in nondiabetic CKD patients, the impact on liver histology, long-term safety in patients with different degrees of renal and hepatic impairment, and the risk of ketoacidosis, which led the FDA to issue a safety warning in 2015.

Conclusions and future perspectives

Despite progress made in the last decade, CKD still remains a major health problem for 2 reasons: it often goes unrecognized and current therapeutic armamentarium has limited effectiveness in retarding disease progression. NAFLD is the most common chronic liver disease and, given the lack of an effective treatment, is becoming the leading indication for liver transplantation in the Western world (5). The shared unmet needs of NAFLD and CKD are therefore boosting research on novel therapeutic targets in each of these 2 conditions

Recent epidemiological data suggest a tight relationship between the presence and severity of NAFLD and the presence and stage of CKD and place NAFLD as an important contributor to the development and progression of CKD, independently of traditional risk factors. When analyzing the pathophysiological basis for this association, striking analogies can be found between fatty liver and CKD: like NAFLD, CKD is characterized by deranged cellular substrate metabolism, ectopic fat deposition, which trigger oxidative stress and inflammatory and profibrotic responses to drive the progression of both disease processes. Our review disclosed a wealth of cellular pathways and mechanisms that represent key contributors to liver and kidney injury and potential therapeutic targets (**Figure 4**). Most of these targets are being currently evaluated in phase II RCTs and some of them, like PPAR-α/δ agonists, FXR agonists and incretin analogues, gave promising results (136). Remarkably, few of them advanced to the same developmental stage in both NAFLD and CKD, reflecting a still low awareness of the similarities in the pathogenic mechanisms underlying these 2 conditions. Beside shared pathogenic mechanisms that promote both liver and kidney injury, the fatty liver may per se promote kidney injury and vice versa, with potentially relevant therapeutic implications: as an example, SGLT2 inhibitors target the renal tubule but may improve both CKD and NAFLD. Whether the relative merits of different therapeutic approaches will translate into a clinical benefit needs assessment in adequately powered, larger RCTs of longer duration with clinical end-points. A key challenge to therapeutic success of these variegated approaches will be the selection of the optimal therapeutic strategy for each patient: NAFLD and CKD progression is likely a

multi-factorial process, involving varied molecular pathways that may operate in different patient subsets and at different stages of disease.Within this context, recent developments in metabolic phenotyping with metabolomics and systems biology technologies will hopefully enable individualized treatment tailored to individual profile.

Given the increasing prevalence of CKD and NAFLD, their direct effects and their acceleration of CVD, strategies to reduce the incidence, progression and complications of these twin plagues are an important priority in healthcare.

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Author Contributions.

GM: conceived and designed the article, undertook literature search and acquired data, critically analyzed the results, drafted the article, gave final approval

GM is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

MC, SC, FDM, SP, FS, RG: undertook literature search and acquired data, critically analyzed the results, contributed to draft of the article, gave final approval

Conflicts of interest.

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FIGURE LEGENDS

Figure 1. Role of nuclear transcription factors in the pathogeness of NAFLD and CKD.

Each nuclear transcription factor is reported in the textbox at the centre of the scheme and is marked with a superscript number. The different molecular pathways affected by each nucelar transcription factors are written in the boxes surrounding the central box, with the superscript number referring to the corresponding nuclear transcription factor affecting the pathway.

Abbreviations:

HSC: hepatic stellate cell; JNK: c-Jun N-terminal kinase; MAPK: mitogen-activated protein kinase; NOX: NADPH oxidase; Smad: Mothers against decapentaplegic homolog ; VLDLR: VLDL receptor. ER: endoplasmic reticulum; FFA: free fatty acids; FXR: farnesoid X-receptor; GLUT: glucose transporter; MCP-1: monocyte chemotactic protein-1; NLRP3: NOD-like receptor family, pyrin domain containing 3; NOS: nitric oxide synthase; NOX: NADPH oxidase; PGC-1α: peroxisome proliferator-activated receptor-γ coactivator-1 α; RAGE: receptor for advanced glycation end-products; ROS: reactive oxygen species; SREBP: sterol-responsive element binding protein; TGF-β: transforming growth factor- β; VLDL: very low density lipoprotein.

Figure 2: Mechanisms connecting cellular sensors AMP-activated Kinase (AMPK)(panel A), hypoxia-induced factor (HIF)-1α and mammalian target of rapamicin complex 1 (mTORC1) (panel B) in the pathogenesis of NAFLD and CKD.

In panel B, HIF-1α and mTORC1 are reported in the textbox at the centre of the scheme and are marked with a superscript number. The different molecular pathways affected by each sensor are written in the boxes surrounding the central box, with the superscript number referring to the corresponding cellular sensor affecting the pathway.

Abbreviations: AMPK: adenosine-monophosphate kinase; EMT : epithelial-to-mesenchimal transition ; ER: endoplasmic reticulum; FAS: fatty acid synthase; FFA: free fatty acids; GLUT: glucose transporter; IRS-1: insulin receptor substrate-1; LOXL: Lysyl oxidase-like**;** MCP-1: monocyte chemotactic protein-1; NO: nitric oxide; NOX: NADPH oxidase; PGC-1α: peroxisome proliferator-activated receptor-γ coactivator-1 α; ROS: reactive oxygen species; SREBP: sterol-responsive element binding protein; TGF-β: transforming growth factor- β; VLDL: very low density lipoprotein;

Figure 3. Role of effectors of redox regulation Apoptosis Signal-Regulating Kinase 1 (ASK1) and Nuclear erythroid 2-related factor 2 (Nrf2) and of galectin-3 in the pathogenesis of NAFLD and CKD. ASK1, Nrf2 and galectin-3 are reported in the textbox at the centre of the scheme and are marked with a superscript number. The different molecular pathways affected by these 3 molecules are written in the boxes surrounding the central box, with the superscript number referring to the corresponding factor affecting the pathway.

Abbreviations: EMT: epithelial-to-mesenchimal transition; HSC: hepatic stellate cell; IRS: insulin receptor substrate; JNK: c-Jun N-terminal kinase; MAPK: mitogen-activated protein kinase; Smad: Mothers against decapentaplegic homolog ;Trx: thioredoxin

ABCA1: **A**TP-binding cassette transporters A1; ACC: acetyl-CoA carboxylase;

Cat: Catalase; Glt-Px: Glutathione peroxidase; Glt-R: Glutathione reductase ; *G6PD:* Glucose-6-phosphate 1-dehydrogenase; HPC: hepatic progenitor cells; *TXN-R: GST:* Glutathione *S*-transferase, Thioredoxin reductase; *SOD* Superoxide dismutase;; ER: endoplasmic reticulum; FAS: fatty acid synthase; FFA: free fatty acids; FXR: farnesoid X-receptor; GLUT: glucose transporter; MCP-1: monocyte chemotactic protein-1; NOS: NO synthase; NOX: NADPH oxidase; OCA: obeticholic acid; PGC-1α: peroxisome proliferator-activated receptor-γ coactivator-1 α; RAGE: receptor for advanced glycation end-products; ROS: reactive oxygen species; SREBP: sterol-responsive element binding protein; TGF-β: transforming growth factor- β.

Figure 4:Molecular pathways mediating the interplay between the liver, kidney, gut and adipose tissue in the pathogenesis of NAFLD and CKD and the effect of their modulation.

Panel 4A: in NAFLD-associated CKD, the gut promotes liver and kidney injury by reduced incretin Glucagon-like Peptide(GLP)-1 and by enhanced microbial production of lipopolysaccharide (LPS) and uremic toxins (URMs), whose excretion is reduced in CKD, further increasing their systemic levels.

Further injurious mechanisms contributing to NASH and CKD at a systemic level include inappropriate activation of Sterol Regulatory Element Binding Protein(SREBP)-2, which promotes intracellular cholesterol accumulation, of cellular sensors Hypoxia Inducible Factor(HIF)-1 and mammalian Target of Rapamicin Complex 1(mTORC1), of oxidative stress-activated Apoptosis Signal-Regulating Kinase 1(ASK1). Collectively, these factors induce insulin resistance(IR), ectopic lipid accumulation which trigger lipoperoxidative stress and enhance secretion of pro-ijflammatory cytokines, chemokines CCL2 and CCL5 and of profibrogenic factors like miRNA21 and Galectin-3. The liver, on its side, promotes CKD through enhanced secretion of uric acid and of pro-atherogenic factors including VLDL1 lipoproteins, Cholesterol-Ester Transfer Protein(CETP) and Syndecan-1, which promote atherogenic dyslipidemia, endothelial dysfunction and renal vascular disease. Additionally, in NAFLD there is also an impaired action of hepatokine Fibroblast Growth Factor(FGF)21, whose systemic levels are elevated because of tissue FGF21 resistance and of impaired renal FGF21 excretion in CKD.

Panel 4B: Beside directly antagonizing the action of noxious factors described in panel A, NAFLD and CKD can be ameliorated at intestinal levels by incretin mimetics and by modulating gut microbiota composition with prebiotics, probiotics or synbiotics and at systemic levels by activating cellular energy sensor AMP-activated Protein Kinae(AMPK) and several nuclear transcription factors, including Peroxisome Proliferator-Activated Receptor(PPAR)-α, PPAR-δ and PPAR-γ and Farnesoid X Receptor(FXR) and Nuclear Erythroid 2-related Factor(Nrf2). Please refer to Tables and text for a detailed description of molecular mechanisms underlying gut-liver-kidney connection,

REFERENCES

1McCullough K, Sharma P, Ali T, Khan I, Smith WC, MacLeod A, Black C. Measuring the population burden of chronic kidney disease: a systematic literature review of the estimated prevalence of impaired kidney function. Nephrol Dial Transplant. 2012;27:1812-21.

2 Palmer SC, Mavridis D, Navarese E, Craig JC, Tonelli M, Salanti G, Wiebe N, Ruospo M, Wheeler DC, Strippoli GF. Comparative efficacy and safety of blood pressure-lowering agents in adults with diabetes and kidney disease: a network meta-analysis. Lancet. 2015;385: 2047-56

3 Musso G, Gambino R, Tabibian JH, Ekstedt M, Kechagias S, Hamaguchi M, Hultcrantz R, Hagström H, Yoon SK, Charatcharoenwitthaya P, George J, Barrera F, Hafliðadóttir S, Björnsson ES, Armstrong MJ, Hopkins LJ, Gao X, Francque S, Verrijken A, Yilmaz Y, Lindor KD, Charlton M, Haring R, Lerch MM, Rettig R, Völzke H, Ryu S, Li G, Wong LL, Machado M, Cortez-Pinto H24, Yasui K, Cassader M Association of Non-alcoholic Fatty Liver Disease with Chronic Kidney Disease: A Systematic Review and Meta-analysis. PLoS Med. 2014 Jul 22;11: e1001680

4 Stepanova M, se Avila L, Birerdinc A, Noor B, Badoe N, Younossi Z. In female patients with NAFLD presence of type 2 diabetes and chronic kidney disease are independently associated with the risk of mortality. Hepatology 2015; 62: 1282°-83A

5 Singal AK, Salameh H, Kuo YF, Wiesner RH. Evolving frequency and outcomes of simultaneous liver kidney transplants based on liver disease etiology. Transplantation. 2014;98: 216-21

6 Mikolasevic I, Racki S, Zaputovic L, Lukenda V, Sladoje-Martinovic B, Orlic L. Nonalcoholic fatty liver disease (NAFLD) and cardiovascular risk in renal transplant recipients. Kidney Blood Press Res. 2014;39:308-14

7 Sugar and Sweeteners Team, Market and Trade Economics, Economic Research Service, US Department of Agriculture. US per capita caloric sweeteners estimated deliveries for domestic food

and beverage use, by calendar year. Available at: http://www.ers.usda.gov/briefing/Sugar/data/table50.xls. Accessed Jan 10th, 2014.

8Johnson RJ. Sugar, uric acid, and the etiology of diabetes and obesity. Diabetes. 2013;62: 3307-15.

9 Fan CY, Wang MX, Ge CX, Wang X, Li JM, Kong LD. Betaine supplementation protects against high-fructose-induced renal injury in rats. J Nutr Biochem. 2014;25:353-62.

10 Yetley EA. Assessing the vitamin D status of the US population. Am J Clin Nutr. 2008;88:558S-564S.

11 Barchetta I. Liver vitamin D receptor, CYP2R1, and CYP27A1 expression: relationship with liver histology and vitamin D3 levels in patients with nonalcoholic steatohepatitis or hepatitis C virus. Hepatology. 2012;56: 2180-7.

12 Beilfuss A. Vitamin D counteracts fibrogenic TGF-β signalling in human hepatic stellate cells both receptor-dependently and independently. Gut. 2014 Aug 18. pii: gutjnl-2014-307024. doi: 10.1136/gutjnl-2014-307024. [Epub ahead of print]

13 Wang XX, Jiang T, Shen Y, Santamaria H, Solis N, Arbeeny C, Levi M. Vitamin D receptor agonist doxercalciferol modulates dietary fat-induced renal disease and renal lipid metabolism. Am J Physiol Renal Physiol. 2011;300: F801-10.

14 Xu L. Impact of vitamin D on chronic kidney diseases in non-dialysis patients: a meta-analysis of randomized controlled trials. PLoS One. 2013; 8: e61387.

15 Dasarathy J, Varghese R, Kalinina I, Lopez R, McCullough AJ. Hypovitaminosis D in NAFLD requires high dose supplementation to reverse deficiency. Hepatology 2015; 62: 1273A

16 Parikh A, Chase HS, Vernocchi L, Stern L. Vitamin D resistance in chronic kidney disease (CKD). BMC Nephrol. 2014 19;15:47.

17 Xu Y, Huang J, Xin W, Chen L, Zhao X, Lv Z, Liu Y, Wan Q. Lipid accumulation is ahead of epithelial-to-mesenchymal transition and therapeutic intervention by acetyl-CoA carboxylase 2 silence in diabetic nephropathy. Metabolism. 2014;63:716-26.

18 Xin W, Zhao X, Liu L, Xu Y, Li Z, Chen L, Wang X, Yi F, Wan Q. Acetyl-CoA carboxylase 2 suppression rescues human proximal tubular cells from palmitic acid induced lipotoxicity via autophagy. Biochem Biophys Res Commun. 2015;463:364-9.

19 Herman-Edelstein M, Scherzer P, Tobar A, Levi M, Gafter U. Altered renal lipid metabolism and renal lipid accumulation in human diabetic nephropathy. J Lipid Res. 2014;55:561-72

20 Souza-Mello V. Peroxisome proliferator-activated receptors as targets to treat non-alcoholic fatty liver disease. World J Hepatol. 2015;7:1012-9

21 Pawlak M. The transrepressive activity of peroxisome proliferator-activated receptor alpha is necessary and sufficient to prevent liver fibrosis in mice. Hepatology. 2014;60:1593-606.

22 Zhou Y, Lin S, Zhang L, Li Y. Resveratrol prevents renal lipotoxicity in high-fat diet-treated mouse model through regulating PPAR-α pathway. Mol Cell Biochem. 2015 Sep 30. [Epub ahead of print] PMID: 26423427

23 Collino M, Benetti E, Rogazzo M, Mastrocola R, Yaqoob MM, Aragno M, Thiemermann C, Fantozzi R. Reversal of the deleterious effects of chronic dietary HFCS-55 intake by PPAR-δ agonism correlates with impaired NLRP3 inflammasome activation. Biochem Pharmacol. 2013;85:257-64

24 Lee EY, Kim GT, Hyun M, Kim S, Seok S, Choi R, Lee MY, Chung CH. Peroxisome proliferator-activated receptor-δ activation ameliorates albuminuria by preventing nephrin loss and restoring podocyte integrity in type 2 diabetes. Nephrol Dial Transplant. 2012;27:4069-79.

25 Sanyal AJ, Harrison SA, Francque SM, Bedossa P, Serfaty L, Romero-Gomez M, Cales P. The hepatic and extrahepatic profile of resolution of steatohepatitis induced by GFT-505. Hepatology 2015; 62: 1252A

26 Musso G, Cassader M, Rosina F, Gambino R. Impact of current treatments on liver disease, glucose metabolism and cardiovascular risk in non-alcoholic fatty liver disease (NAFLD): a systematic review and meta-analysis of randomised trials. Diabetologia. 2012; 55:885-904.

27 Sarafidis PA, Stafylas PC, Georgianos PI, Saratzis AN, Lasaridis AN. **Effect** of thiazolidinediones on albuminuria and proteinuria in diabetes: a meta-analysis. Am J Kidney Dis. 2010;55:835-47

28 Shashank R,Saroglitazar in Diabetic Dyslipidemia: 1-Year DataView session detail Abstract 126-LB; American Diabetes Association 75th Scientific Sessions, June 5 - 9, 2015, Boston, Massachusetts

29 Banshi D. To Assess the Effect of 4mg Saroglitazar on Patients of Diabetes Dyslipidemia with Nonalcoholic Fatty Liver Disease for 24 Weeks at Diabetes Care CentreView session detail
Abstract 712-P , American Diabetes Association 75th Scientific Sessions, June 5 - 9, 2015, Boston, Massachusetts

30 Ruilope L, Hanefeld M, Lincoff AM, Viberti G, Meyer-Reigner S, Mudie N, Wieczorek Kirk D, Malmberg K, Herz M. Effects of the dual peroxisome proliferator-activated receptor-α/γ agonist aleglitazar on renal function in patients with stage 3 chronic kidney disease and type 2 diabetes: a Phase IIb, randomized study. BMC Nephrol. 2014;15:180.

31 Musso G, Gambino R, Cassader M.Cholesterol metabolism and the pathogenesis of non-alcoholic steatohepatitis.Prog Lipid Res. 2013;52:175-91

32 Kandasamy N, Ashokkumar N. Renoprotective effect of myricetin restrains dyslipidemia and renal mesangial cell proliferation by the suppression of sterol regulatory element binding proteins in an experimental model of diabetic nephropathy. Eur J Pharmacol. 2014;743:53-62.

33 Chen Y, Zhao L, Li Q, Wheeler DC, Varghese Z, Moorhead JF, Powis SH, Ruan XZ . Inflammatory stress reduces the effectiveness of statins in the kidney by disrupting HMGCoA reductase feedback regulation.Nephrol Dial Transplant. 2014 ;29:1864-78.

34 Hu Z, Ren L, Wang C, Liu B, Song G. Effect of chenodeoxycholic acid on fibrosis, inflammation and oxidative stress in kidney in high-fructose-fed Wistar rats. Kidney Blood Press Res. 2012;36: 85-97

35 Wang XX. The farnesoid X receptor modulates renal lipid metabolism and diet-induced renal inflammation, fibrosis, and proteinuria. Am J Physiol Renal Physiol. 2009; 297:F1587-96.

36 Li J, Wilson A, Gao X, Kuruba R, Liu Y, Poloyac S, Pitt B, Xie W, Li S. Coordinated regulation of dimethylarginine dimethylaminohydrolase-1 and cationic amino acid transporter-1 by farnesoid X receptor in mouse liver and kidney and its implication in the control of blood levels of asymmetric dimethylarginine. J Pharmacol Exp Ther. 2009;331:234-43

37 Neuschwander-Tetri BA, Loomba R, Sanyal AJ, Lavine JE, Van Natta ML, Abdelmalek MF, Chalasani N, Dasarathy S, Diehl AM, Hameed B, Kowdley KV, McCullough A, Terrault N, Clark JM, Tonascia J, Brunt EM, Kleiner DE, Doo E. Farnesoid X nuclear receptor ligand obeticholic acid for non-cirrhotic, non-alcoholic steatohepatitis (FLINT): a multicentre, randomised, placebo-controlled trial. The Lancet 2015. 385: 956-65

38 Gerhard GS, DiStefano JK. Micro RNAs in the development of non-alcoholic fatty liver disease. World J Hepatol. 2015;7:226-34

39 Trionfini P, Benigni A, Remuzzi G. MicroRNAs in kidney physiology and disease. Nat Rev Nephrol. 2015;11:23-33

40 Dattaroy D, Pourhoseini S, Das S, Alhasson F, Seth RK, Nagarkatti M, Michelotti GA, Diehl AM, Chatterjee S.Micro-RNA 21 inhibition of SMAD7 enhances fibrogenesis via leptin-mediated NADPH oxidase in experimental and human nonalcoholic steatohepatitis. Am J Physiol Gastrointest Liver Physiol. 2015;308:G298-312.

41 Gomez IG, MacKenna DA, Johnson BG, Kaimal V, Roach AM, Ren S, Nakagawa N, Xin C, Newitt R, Pandya S, Xia TH, Liu X, Borza DB, Grafals M, Shankland SJ, Himmelfarb J, Portilla D, Liu S, Chau BN, Duffield JS. Anti-microRNA-21 oligonucleotides prevent Alport nephropathy progression by stimulating metabolic pathways. J Clin Invest. 2015;125:141-56.

42 Musso G, Gambino R, Cassader M. Emerging molecular targets for the treatment of nonalcoholic fatty liver disease. Annu Rev Med. 2010;61:375-92

43Li J. Hepatoprotective effects of berberine on liver fibrosis via activation of AMP-activated protein kinase. Life Sci. 2014;98:24-30

44 Hsu WH, Chen TH, Lee BH, Hsu YW, Pan I Monascin and ankaflavin act as natural AMPK activators with PPARα agonist activity to down-regulate nonalcoholic steatohepatitis in high-fat diet-fed C57BL/6 mice. Food Chem Toxicol. 2014; 64:94-103.

45 Soetikno V. Curcumin decreases renal triglyceride accumulation through AMPK-SREBP signaling pathway in streptozotocin-induced type 1 diabetic rats. J Nutr Biochem. 2013;24:796-802.

46 Anavi S, Hahn-Obercyger M, Madar Z, Tirosh O. Mechanism for HIF-1 activation by cholesterol under normoxia: a redox signaling pathway for liver damage. Free Radic Biol Med. 2014 ;71:61-9

47 Musso G, Cassader M, Olivetti C, Rosina F, Carbone G, Gambino R.Association of obstructive sleep apnoea with the presence and severity of non-alcoholic fatty liver disease. A systematic review and meta-analysis. Obes Rev. 2013;14:417-31

48 Musso G, Olivetti C, Cassader M, Gambino R.Obstructive sleep apnea-hypopnea syndrome and nonalcoholic fatty liver disease: emerging evidence and mechanisms. Semin Liver Dis. 2012 ;32:49-64

49 Shoji K, Tanaka T, Nangaku M. Role of hypoxia in progressive chronic kidney disease and implications for therapy. Curr Opin Nephrol Hypertens. 2014; 23:161-8.

50 Luo R, Zhang W, Zhao C, Zhang Y, Wu H, Jin J, Zhang W, Grenz A, Eltzschig HK, Tao L, Kellems RE, Xia Y. Elevated Endothelial Hypoxia-Inducible Factor-1α Contributes to Glomerular Injury and Promotes Hypertensive Chronic Kidney Disease. Hypertension. 2015;66:75-84.

51 Xiao J, Jin C, Liu Z, Guo S, Zhang X, Zhou X, Wu X. The design, synthesis, and biological evaluation of novel YC-1 derivatives as potent anti-hepatic fibrosis agents. Org Biomol Chem. 2015;13:7257-64.

52 Nordquist L, Friederich-Persson M, Fasching A, Liss P, Shoji K, Nangaku M, Hansell P, Palm F Activation of hypoxia-inducible factors prevents diabetic nephropathy. J Am Soc Nephrol. 2015;26:328-38.

53 Kim YC, Guan KL. mTOR: a pharmacologic target for autophagy regulation. J Clin Invest. 2015;125: 25-32

54 Xu Y, Liu L, Xin W, Zhao X, Chen L, Zhen J, Wan Q. The renoprotective role of autophagy activation in proximal tubular epithelial cells in diabetic nephropathy. J Diabetes Complications. 2015 Jul 22. pii: S1056-8727(15)00293-7. doi: 10.1016/j.jdiacomp.2015.07.021. [Epub ahead of print]

55Sapp V, Gaffney L, EauClaire SF, Matthews RP. Fructose leads to hepatic steatosis in zebrafish that is reversed by mechanistic target of rapamycin (mTOR) inhibition. Hepatology. 2014;60:1581-92

56 Chen H, Zhu J, Liu Y, Dong Z, Liu H, Liu Y, Zhou X, Liu F, Chen G. Lipopolysaccharide Induces Chronic Kidney Injury and Fibrosis through Activation of mTOR Signaling in Macrophages. Am J Nephrol. 2015;42:305-317.

57 Jiang H, Westerterp M, Wang C, Zhu Y, Ai D. Macrophage mTORC1 disruption reduces inflammation and insulin resistance in obese mice. Diabetologia. 2014;57: :2393-404.

58 Wang L. ALCAT1 controls mitochondrial etiology of fatty liver diseases, linking defective mitophagy to steatosis. Hepatology. 2015;61:486-96.

59Torricelli C. Phosphorylation-independent mTORC1 inhibition by the autophagy inducer Rottlerin. Cancer Lett. 2015 Feb 4. pii: S0304-3835(15)00074-9. doi: 10.1016/j.canlet.2015.01.040. [Epub ahead of print]

60 Kawarazaki Y, Ichijo H, Naguro I. Apoptosis signal-regulating kinase 1 as a therapeutic target. Expert Opin Ther Targets. 2014;18:651-64

61 Yamamoto E, Dong YF, Kataoka K, Yamashita T, Tokutomi Y, Matsuba S, Ichijo H, Ogawa H, Kim-Mitsuyama S. Olmesartan prevents cardiovascular injury and hepatic steatosis in obesity and diabetes, accompanied by apoptosis signal regulating kinase-1 inhibition. Hypertension. 2008;52:573-80

62 Tesch GH, Ma FY, Han Y, Liles JT, Breckenridge DG, Nikolic-Paterson DJ. **ASK1** Inhibitor Halts Progression of Diabetic Nephropathy in Nos3-Deficient Mice. Diabetes. 2015;64:3903-13

63Tebay LE, Robertson H, Durant ST, Vitale SR, Penning TM, Dinkova-Kostova AT, Hayes JD. Mechanisms of activation of the transcription factor Nrf2 by redox stressors, nutrient cues, and energy status and the pathways through which it attenuates degenerative disease. Free Radic Biol Med. 2015;88:108-46

64 Shimozono R. Nrf2 activators attenuate the progression of nonalcoholic steatohepatitis-related fibrosis in a dietary rat model. Mol Pharmacol. 2013;84:62-70

65 Choi BH, Kang KS, Kwak MK. Effect of redox modulating NRF2 activators on chronic kidney disease. Molecules. 2014;19:12727-59.

66 de Zeeuw D. Rationale and trial design of bardoxolone methyl evaluation in patients with chronic kidney disease and type 2 diabetes: the occurrence of renal events (BEACON). Am J Nephrol 2013; 37: 212–222

67 Kufareva I, Salanga CL, Handel TM. Chemokine and chemokine receptor structure and interactions: implications for therapeutic strategies. Immunol Cell Biol. 2015 Feb 24. doi: 10.1038/icb.2015.15.

68 Oh DY. Increased macrophage migration into adipose tissue in obese mice. Diabetes 2012;61:346–354.

69 Baeck C. Pharmacological inhibition of the chemokine C-C motif chemokine ligand 2 (monocyte chemoattractant protein 1) accelerates liver fibrosis regression by suppressing Ly-6C(+) macrophage infiltration in mice. Hepatology 2014; 59: 1060-72

70 Moreno JA, Moreno S, Rubio-Navarro A, Sastre C, Blanco-Colio LM, Gómez-Guerrero C, Ortiz A, Egido J. Targeting chemokines in proteinuria-induced renal disease. Expert Opin Ther Targets. 2012;16: 833-45.

71 de Zeeuw D, Bekker P, Henkel E, Hasslacher C, Gouni-Berthold I, Mehling H, Potarca A, Tesar V, Heerspink HJ, Schall TJ;The effect of CCR2 inhibitor CCX140-B on residual albuminuria in patients

with type 2 diabetes and nephropathy: a randomised trial. Lancet Diabetes Endocrinol. 2015 Aug 7. pii: S2213-8587(15)00261-2. doi: 10.1016/S2213-8587(15)00261-2. [Epub ahead of print]

72 Pugliese G, Iacobini C, Pesce CM, Menini S. **Galectin**-3: an emerging all-out player in metabolic disorders and their complications. Glycobiology. 2015;25:136-50.

73 Iacobini C. Galectin-3 ablation protects mice from diet-induced NASH: A major scavenging role for galectin-3 in liver. J Hep 2011; 54: 975–983

74 Calvier L, Martinez-Martinez E, Miana M, Cachofeiro V, Rousseau E, Sádaba JR, Zannad F, Rossignol P, López-Andrés N. The impact of galectin-3 inhibition on aldosterone-induced cardiac and renal injuries. JACC Heart Fail. 2015;3:59-67.

75 Drechsler C, Delgado G, Wanner C, Blouin K, Pilz S, Tomaschitz A, Kleber ME, Dressel A, Willmes C, Krane V, Krämer BK, März W, Ritz E, van Gilst WH, van der Harst P, de Boer RA.

Galectin-3, Renal Function, and Clinical Outcomes: Results from the LURIC and 4D Studies.

J Am Soc Nephrol. 2015;26:2213-21

76 Frenay AR, Yu L, van der Velde AR, Vreeswijk-Baudoin I, López-Andrés N, van Goor H, Silljé HH, Ruifrok WP, de Boer RA. Pharmacological inhibition of galectin-3 protects against hypertensive nephropathy. Am J Physiol Renal Physiol. 2015;308:F500-9

77 Traber PG et al. Regression of Fibrosis and Reversal of Cirrhosis in Rats by Galectin Inhibitors in Thioacetamide-Induced Liver Disease. PLoS ONE 2013; 8: e75361

78 Pang J, et al. Increased Adiposity, Dysregulated Glucose Metabolism and Systemic Inflammation in Galectin-3 KO Mice. PLoS One. 2013; 8:e57915.

79 Zhou SW, Zhang M, Zhu M. Liraglutide reduces lipid accumulation in steatotic L-02 cells by enhancing autophagy. Mol Med Rep 2014; 10:2351-7

80 Hwang HJ, Jung TW, Kim BH, Hong HC, Seo JA, Kim SG, Kim NH, Choi KM, Choi DS, Baik SH, Yoo HJ. A dipeptidyl peptidase-**IV inhibitor** improves hepatic steatosis and insulin resistance by

AMPK-dependent and JNK-dependent inhibition of LECT2 expression. Biochem Pharmacol. 2015 ;98:157-66

81 Armstrong MJ. Safety and efficacy of liraglutide in patients with type 2 diabetes and elevated liver enzymes: individual patient data meta-analysis of the LEAD program. Aliment Pharmacol Ther. 2013;37: 234-42.

82 Armstrong MJ, Hull D, Guo K, Barton D, Hazlehurst JM, Gathercole LL, Nasiri M, Yu J, Gough SC, Newsome PN, Tomlinson JW. Glucagon-Like Peptide 1 Decreases Lipotoxicity in Non-Alcoholic Steatophepatitis. J Hepatol. 2015 Sep 19. pii: S0168-8278(15)00624-8. doi: 10.1016/j.jhep.2015.08.038. [Epub ahead of print]

83 Skov J. Glucagon-like peptide-1 (GLP-1): effect on kidney hemodynamics and renin-angiotensin-aldosterone system in healthy men. J Clin Endocrinol Metab. 2013;98:E664-71.

84 Imamura S1, Hirai K, Hirai A.The glucagon-like peptide-1 receptor agonist, liraglutide, attenuates the progression of overt diabetic nephropathy in type 2 diabetic patients Tohoku J Exp Med. 2013;231:57-61..

85 Zhang H1, Zhang X, Hu C, Lu W.Exenatide reduces urinary transforming growth factor-β1 and type IV collagen excretion in patients with type 2 diabetes and microalbuminuria. Kidney Blood Press Res. 2012;35:483-8.

86 Bergenstal RM. Efficacy and safety of exenatide once weekly versus sitagliptin or pioglitazone as an adjunct to metformin for treatment of type 2 diabetes (DURATION-2): a randomised trial. Lancet. 2010;376:431-9.

87 Scirica BM, Bhatt DL, Braunwald E, Steg PG, Davidson J, Hirshberg B, Ohman P, Frederich R, Wiviott SD, Hoffman EB, Cavender MA, Udell JA, Desai NR, Mosenzon O, McGuire DK, Ray KK, Leiter LA, Raz I; SAVOR-TIMI 53 Steering Committee and Investigators. Saxagliptin and cardiovascular outcomes in patients with type 2 diabetes N Engl J Med. 2013;369:1317-26

88 Groop PH, Cooper ME, Perkovic V, Emser A, Woerle HJ, von Eynatten M. Linagliptin lowers albuminuria on top of recommended standard treatment in patients with type 2 diabetes and renal dysfunction. Diabetes Care. 2013;36: 3460-8.

89 Sanofi. ELIXA press release. Sanofi announces top-line results for cardiovascular outcomes study of Lyxumia_ (lixisenatide). 2015. Available from http://en.sanofi.com/NasdaQ_OMX/local/ press_releases/sanofi_announces_topline_resul_1904474_19-03-2015!07_00_00.aspx. Last accessed 14 September, 2015.

90 Musso G, Gambino R, Cassader M. Interactions between gut microbiota and host metabolism predisposing to obesity and diabetes. Annu Rev Med. 2011; 62:361-80.

91 Wong J, Piceno YM, Desantis TZ, Pahl M, Andersen GL, Vaziri ND. Expansion of urease- and uricase-containing, indole- and p-cresol-forming and contraction of short-chain fatty acid-producing intestinal microbiota in ESRD. Am J Nephrol. 2014;39:230-7.

92 Vaziri ND, Yuan J, Nazertehrani S, Ni Z, Liu S Chronic kidney disease causes disruption of gastric and small intestinal epithelial tight junction. Am J Nephrol. 2013;38(2):99-103.

93 Adesso S. The uremic toxin indoxyl sulphate enhances macrophage response to LPS. PLoS One. 2013;8:e76778.

94 Koppe L. p-Cresyl sulfate promotes insulin resistance associated with CKD.J Am Soc Nephrol. 2013;24:88-99.

95 Wu IW. p-Cresyl sulphate and indoxyl sulphate predict progression of chronic kidney disease. Nephrol. Dial. Transplant. 2011; 26: 938–47.

96 Nymark M1, Pussinen PJ, Tuomainen AM, Forsblom C, Groop PH, Lehto M Serum lipopolysaccharide activity is associated with the progression of kidney disease in finnish patients with type 1 diabetes. Diabetes Care. 2009;32:1689-93.

97 Tang W, Wang Z, Kennedy DJ, Wu Y, Buffa JA, Agatisa-Boyle B, Li XS, Levison BS, Hazen SL Gut Microbiota-Dependent Trimethylamine *N*-Oxide(TMAO) Pathway Contributes to Both Development of Renal Insufficiency and Mortality Risk in Chronic Kidney Disease. *Circ Res*. 2015;116:448-455.

98 Vaziri ND, Yuan J, Khazaeli M, Masuda Y, Ichii H, Liu S. Oral activated charcoal adsorbent (AST-120) ameliorates CKD-induced intestinal epithelial barrier disruption and systemic inflammation. Am. J. Nephrol 2013; 37; 518–25

99 Eslamparast T, Poustchi H, Zamani F, Sharafkhah M, Malekzadeh R, Hekmatdoost A. Synbiotic supplementation in nonalcoholic fatty liver disease: a randomized, double-blind, placebo-controlled pilot study. Am J Clin Nutr. 2014;99:535-42.

100 Rossi M, Klein K, Johnson DW, Campbell KL. Pre-, pro-, and synbiotics: do they have a role in reducing uremic toxins? A systematic review and meta-analysis**.** Int J Nephrol. 2012; 2012:673631.

101 **Musso G**, Cassader M, Cohney S, Pinach S, Saba F, Gambino R. Emerging Liver-Kidney Interactions in Nonalcoholic Fatty Liver Disease. Trends Mol Med. 2015;21: 645-62.

102 Penno G, Solini A, Zoppini G, Fondelli C, Trevisan R, Vedovato M, Gruden G, Lamacchia O, Pontiroli AE, Arosio M, Orsi E, Pugliese G.Hypertriglyceridemia Is Independently Associated with Renal, but Not Retinal Complications in Subjects with Type 2 Diabetes: A Cross-Sectional Analysis of the Renal Insufficiency And Cardiovascular Events (RIACE) Italian Multicenter Study. PLoS One. 2015 ;10:e0125512.

103 Sonmez A, Yilmaz MI, Saglam M, Unal HU, Gok M, Cetinkaya H, Karaman M, Haymana C, Eyileten T, Oguz Y, Vural A, Rizzo M, Toth PP. The role of plasma triglyceride/high-density lipoprotein cholesterol ratio to predict cardiovascular outcomes in chronic kidney disease. Lipids Health Dis. 2015; 14: 29.

104 Gyebi L, Soltani Z, Reisin E. Lipid nephrotoxicity: new concept for an old disease. Curr Hypertens Rep. 2012;14:177-81.

105 Wang Y, van der Tuin S, Tjeerdema N, van Dam AD, Rensen SS, Hendrikx T, Berbée JF, Atanasovska B, Fu J, Hoekstra M, Bekkering S, Riksen NP, Buurman WA, Greve JW, Hofker MH, Shiri-Sverdlov R, Meijer OC, Smit JW, Havekes LM, van Dijk KW, Rensen PC. Plasma cholesteryl ester transfer protein is predominantly derived from Kupffer cells. Hepatology. 2015 Jul 14. doi: 10.1002/hep.27985. [Epub ahead of print]

106 Liaw YW, Lin CY, Lai YS, Yang TC, Wang CJ, Whang-Peng J, Liu LF, Lin CP, Nieh S, Lu SC, Hwang J. A vaccine targeted at CETP alleviates high fat and high cholesterol diet-induced atherosclerosis and non-alcoholic steatohepatitis in rabbit. PLoS One. 2014;9:e111529.

107 Deng Y, Foley EM, Gonzales JC, Gordts PL, Li Y, Esko JD. Shedding of syndecan-1 from human hepatocytes alters very low density lipoprotein clearance. Hepatology. 2012 ;55: 277-86.

108 Stanford KI. Heparan sulfate 2-O-sulfotransferase is required for triglyceride-rich lipoprotein clearance. J Biol Chem 2010;285:286–94.

109 Yilmaz Y, Eren F, Colak Y, Senates E, Celikel CA, Imeryuz N. Hepatic expression and serum levels of syndecan 1 (CD138) in patients with **nonalcoholic** fatty liver disease. Scand J Gastroenterol. 2012;47:1488-93

110 Okazaki I. Fibrogenesis and Carcinogenesis in Nonalcoholic Steatohepatitis (NASH): Involvement of Matrix Metalloproteinases (MMPs) and Tissue Inhibitors of Metalloproteinase (TIMPs). Cancers. 2014;6:1220-55.

111 Hardwick RN. . Altered UDP-glucuronosyltransferase and **sulfotransferase** expression and function during progressive stages of human **nonalcoholic** fatty liver disease. Drug Metab Dispos. 2013;41:554-61.

112 Adepu S. Hepatic syndecan-1 changes associate with dyslipidemia after renal transplantation. Am J Transplant. 2014;14:2328-38.

113 Padberg JS. Damage of the endothelial glycocalyx in chronic kidney disease. Atherosclerosis. 2014;234:335-43.

114 Zeng Y, Adamson RH, Curry FR, Tarbell JM. Sphingosine-1-phosphate protects endothelial glycocalyx by inhibiting syndecan-1 shedding. Am J Physiol Heart Circ Physiol. 2014;306:H363-72.

115 Zhang J and Li Y (Fibroblast growth factor 21 analogs for treating metabolic disorders. Front. Endocrinol. 2015; 6:168.

116 Xu J. Fibroblast growth factor 21 reverses hepatic steatosis, increases energy expenditure, and improves insulin sensitivity in diet-induced obese mice. Diabetes 2009 58 250–259.

117Xu P, Zhang Y, Liu Y, Yuan Q, Song L, Liu M, Liu Z, Yang Y, Li J, Li D, Ren G.Fibroblast growth factor 21 attenuates hepatic fibrogenesis through TGF-β/smad2/3 and NF-κB signaling pathways. Toxicol Appl Pharmacol. 2015 Nov 21. pii: S0041-008X(15)30139-3. doi: 10.1016/j.taap.2015.11.012. [Epub ahead of print]

118 Fisher FM. Obesity is a fibroblast growth factor 21 (FGF21)-resistant state. Diabetes 2010; 11:2781–2789.

119 Gaich G, Chien JY, Fu H, Glass LC, Deeg MA, Holland WL, Kharitonenkov A, Bumol T, Schilske HK, Moller DE.. The effects of LY2405319, an FGF21 analog, in obese human subjects with type 2 diabetes. Cell Metab. 2013;18:333-40.

120 Skrtić M, Yang GK, Perkins BA, Soleymanlou N, Lytvyn Y, von Eynatten M, Woerle HJ, Johansen OE, Broedl UC, Hach T, Silverman M, Cherney DZ. Characterisation of glomerular haemodynamic responses to SGLT2 inhibition in patients with type 1 diabetes and renal hyperfiltration. Diabetologia. 2014;57:2599-602.

121 Zingerman B, Herman-Edelstein M, Erman A, Bar Sheshet Itach S, Ori Y, Rozen-Zvi B, Gafter U, Chagnac A. Effect of Acetazolamide on Obesity-Induced Glomerular Hyperfiltration: A Randomized Controlled Trial. PLoS One. 2015 ;10:e0137163.

112 Elliott RH, Matthews VB, Rudnicka C, Schlaich MP. Is it time to think about the sodium glucose co-transporter 2 sympathetically? Nephrology (Carlton). 2015 Sep 15. doi: 10.1111/nep.12620. [Epub ahead of print]

123 Panchapakesan U, Pegg K, Gross S, Komala MG, Mudaliar H, Forbes J, Pollock C, Mather A. Effects of SGLT2 inhibition in human kidney proximal tubular cells--renoprotection in diabetic nephropathy? PLoS One. 2013;8:e54442.

124 Lambers Heerspink HJ, Johnsson E, Gause-Nilsson I, Johannson K, Sjostrom CD. Dapagliflozin reduces albuminuria on top of renin-angiotensin system blockade in hypertensive diabetic patients. Diabetes. 2015;64:A303

125 Vallon V, Gerasimova M, Rose MA, Masuda T, Satriano J, Mayoux E, Koepsell H, Thomson SC, Rieg T. SGLT2 inhibitor empagliflozin reduces renal growth and albuminuria in proportion to hyperglycemia and prevents glomerular hyperfiltration in diabetic Akita mice. *Am J Physiol Renal Physiol* 2014; 306: F194–F204

126 Yale JF, Bakris G, Cariou B, Nieto J, David-Neto E, Yue D, Wajs E, Figueroa K, Jiang J, Law G, Usiskin K, Meininger G; DIA3004 Study Group. Efficacy and safety of canagliflozin over 52 weeks in patients with type 2 diabetes mellitus and chronic kidney disease. Diabetes Obes Metab. 2014;16:1016-27.

127 Zimman B, Wanner C, Lachin JM, Fitchett D, Bluhmki E, Hantel S, Mattheus M, Devins T, Johansen OR, Woerle HJ. Empagliflozin, cardiovascular outcomes, and mortality in type 2 diabetes. New Engl J Med 2015; doi:10.1056/NEJMoa1504720

128 Wilkison W, Cheatham B, Walker S. Remogliflozin etabonate reduces insulin resistance and liver function enzymes: Role for treatment of nashJ Hepatology 2015; 62(S2): S211-12

129 Fushimi N, Shibuya T, Takeishi S, Itou S, Kawai H, Mori A. Hepatic fat deposition is improved more with SGLT2 inhibitor luseogliflozin compared with sitagliptin: a randomised, crossover, controlled study using computed tomography. Diabetologia 2015; 58(S1): S392

130 Hayashizaki-Someya Y, Kurosaki E, Takasu T, Mitori H, Yamazaki S, Koide K, Takakura S.Ipragliflozin, an SGLT2 inhibitor, exhibits a prophylactic effect on hepatic **steatosis** and fibrosis induced by choline-deficient l-amino acid-defined diet in rats.
Eur J Pharmacol. 2015;754:19-24

131 Yokono M, Takasu T, Hayashizaki Y, Mitsuoka K, Kihara R, Muramatsu Y, Miyoshi S, Tahara A, Kurosaki E, Li Q, Tomiyama H, Sasamata M, Shibasaki M, Uchiyama Y. SGLT2 selective inhibitor ipragliflozin reduces body fat mass by increasing fatty acid oxidation in high-fat diet-induced obese rats. Eur J Pharmacol. 2014;727:66-74

132 Qiang S, Nakatsu Y, Seno Y, Fujishiro M, Sakoda H, Kushiyama A, Mori K, Matsunaga Y, Yamamotoya T, Kamata H, Asano T. Treatment with the SGLT2 inhibitor luseogliflozin improves nonalcoholic steatohepatitis in a rodent model with diabetes mellitus.
Diabetol Metab Syndr. 2015; 7:104.

133Cherney DZI, Perkins BA, Soleymanlou N, Xiao F, Zimpelmann J, Woerle HJ, Johansen OE, Broedl UC, von Eynatten M, Burns KD. Sodium glucose cotransport-2 inhibition and intrarenal RAS activity in people with type 1 diabetes.. Kidney Int 2014; 86: 1057–58.

134 Ojima A, Matsui T, Nishino Y, Nakamura N, Yamagishi S. Empagliflozin, an Inhibitor of Sodium-Glucose Cotransporter 2 Exerts Anti-Inflammatory and Antifibrotic Effects on Experimental Diabetic Nephropathy Partly by Suppressing AGEs-Receptor Axis. Horm Metab Res. 2015;47:686-92.

135 Dransfeld O, Gehrmann T, Köhrer K, Kircheis G, Holneicher C, Häussinger D, Wettstein M

Oligonucleotide microarray analysis of differential transporter regulation in the regenerating rat liver. Liver Int. 2005;25:1243-58.

136 Musso G, Cassader M, Gambino R. Non-alcoholic steatohepatitis: emerging molecular targets and therapeutic strategies. Nat Rev Drug Discov 2016; 15: 249-74

Table 1. Nutritional factors involved in liver and renal disease in NAFLD and CKD

Dietary fructose	
Cellular effects	**Biological effect**
Hypotalamus: ↑ dopaminergic tone→↑ appetite and calorie intake	Weight gain Ectopic fat deposition Insulin resistance inflammation fibrosis Albuminuria Hyperglycemia
Hepatocyte, skeletal miocyte: ↓ FFA oxidation and RE	
Adipocyte: Adipose tissue dysfunction→↓ adiponectin secretion	
Liver and kidney cells(uric acid-mediated): NLRP3 inflammasome activattion→↑ IL-1 secretion→↑macrophage accumulation ↓ AMPK activity→ ↓ FFA oxidation SREBP-1c activation→↑ *de novo* lipogenesis NOX activation→ ROS generation→podocyte loss, tubule cells EMT, endothelial ijury	
Pancreatic β-cell: ↓ postprandial insulin response	

vitamin D deficiency	
Cellular effects	**Biological effect**
Skeletal miocyte: ↓ IRS-1 activity→↓ insulin signaling	Insulin resistance, Ectopic fat
Hepatocyte, Kupffer cell: ↑ TLR-2/4/9 expression→↑ sensitivity to LPS and FFA-induced inflammatory cytokines ↑ ROS production→↑ hepatocyte apoptosis, ↑ inflammatory cell recruitment	

Hepatic stellate cell: VDR down-regulation→ ↑ TGF-β secretion and SMAD2 activation→↑ fibrogenesis	deposition Inflammation Hepatic fibrosis proteinuria ectopic fat deposition tubulo-interstitial inflammation and fibrosis
Glomerular podocyte: ↑p38- and ERK-mediated apoptosis→↑ podocyte injury→glomerular barrier disruption ↑ RAS activation→ glomerulosclerosis	
Renal tubule cell and interstitial macrophage: ↑ SREBP-1c/SREBP-2 →↑ toxic cholesterol and FFA lipid accumulation ↓ FXR/PPAR-α activation→↑ toxic cholesterol and FFA lipid accumulation ↑ NF-κB pathway activation→↑inflammation and macrophage accumulation ↑ TGF-β /Wnt pathway activation→ tubule cell EMT→↑ fibrogenesis	

Abbreviations: AMP: adenosine-monophosphate; AMPD: AMP deaminase; AMPK: adenosine-monophosphate kinase; ATP: adenosine triphosphate; EMT: epithelial-to-mesenchymal transition; CYP: cytochrome protein; ER: endoplasmic reticulum; ERK: extracellular signal-regulated kinase;; FFA: free fatty acids; FXR: farnesoid X-receptor; HSC: hepatic stellate cell; IL: interleukin; IRS-1: insulin receptor substrate-1; LPS: lipopolysaccharide; MCP-1: monocyte chemotactic protein-1; NF-κB: nuclear factor-κB; NLRP3: NOD-like receptor superfamily, pyrin domain containing 3; NO: nitric oxide; NOX: NADPH oxidase PPAR: peroxisome proliferators-activated receptor; RAS: rennin-angiotensin system; REE: resting energy expenditure; ROS: reactive oxygen species; SHP: small heterodimer partner; SMAD. small mother against decapentaplegic; SOCS-3: suppressor of cytokine signaling 3; SREBP: sterol-responsive element binding protein; TGF-β: transforming growth factor- β; TLR: toll-like receptor; TNF: tumor necrosis factor; VDR; vitamin D receptor;

Table 2. Mechanisms of action and developmental stages of drugs targeting both NAFLD and CKD

Mechanism of action	**Molecule**	**Developmental stage**	
		NAFLD	**CKD**
Xanthine oxidase inhibitors	Allopurinol, febuxostat	Preclinical	Iia CKD-FIX (clinicaltrials.gov ID: NCT12611000791932) FEATHER (UMIN ID: UMIN000008343)
Vitamin D supplementation	Natural (ergocalciferol, cholecalciferol, calcitriol), Synthetic VDR agonists(doxercalciferol, paricalcitol)	IIa Clinicaltrials.gov ID: NCT02098317.	IIa Clinicaltrials.gov ID: NCT00893451, NCT01623024
PPAR-δ agonists	GW0742	-	Preclinical (25)
	GW610742	-	Preclinical(26)
PPAR-α/δ agonists	GFT505	IIb GOLDEN-505(27)	-
PPAR- α/γ agonists	Saroglitazar	IIa (CTRI no.: CTRI/2010/091/000108	-
	Aleglitazar	-	IIb ((32)
SREBP-2 antagonists	Natural antioxidants(myricetin)	-	Preclinical (34)
FXR agonists	Obeticholic acid	IIa FLINT(39)	Preclinical (36,37)
miRNA-21	Antagomir-21	Preclinical(42)	Preclinical(43)

antagonists			
AMPK activators	**Natural**: curcumin berberine, monascin ankaflavin	Preclinical(45,46)	Preclinical (47)
	Synthetic: oltipraz	IIa(clinicaltrials.gov ID: NCT01373554).	-
HIF-1α inhibitors	YC-1, AC, POC	Preclinical (53)	-
Dual mTORC1/2 inhibitor	Rapamicin	Preclinical(55)	Preclinical(56)
ASK1 inhibitor	GS-4997	IIa (ClinicalTrials.gov ID: NCT02466516)	IIa(ClinicalTrials.gov ID: NCT02177786)
Nrf2 activators	Oltipraz , Bardoxolone methyl	IIa(clinicaltrials.gov ID: NCT01373554)	(clinicaltrials.gov ID: NCT02316821)
CCR2 receptor antagonist	CCX140-B	-	IIa(74)
CCR2/5 receptor antagonist	Cenicriviroc	Iia**:** CENTAUR (ClinicalTrials.gov ID: NCT02217475)	-
	BMS-813160 PF-04634817	-	IIa **(**ClinicalTrials.gov ID: NCT01752985, NCT01712061)
Galectin-3 inhibitors	GM-CT-01	Preclinical(84)	**-**
	GR-MD-02	Phase I (ClinicalTrials.gov ID: NCT01899859)	**-**
	GCS-100	**-**	Phase IIa: ClinicalTrials.gov ID:

			NCT01843790
Incretin-based therapies	**Incretin mimetics:** liraglutide, exenatide	Iia LEAN(84)	IIa(86,87)
	DPP-4 inhibitors: Saxagliptin, linagliptin	Preclinical(42)	IIa (MARLINA-T2D, CARMELINA)
Gut microbiota manipulation	Prebiotics, probiotics, synbiotics	IIa(101)	Iia(Australian New Zealand Clinical Trials Registry Number: ACTRN12613000493741)
CETP inhibitors	Fc-CETP6	Preclinical(108)	-
Inhibitors of syndecan-1 shedding	sphingosine-1-phosphate	-	Preclinical(116)
FGF-21 analogs	PEG-FGF21, recombinant FGF21, anti-FGFR1 mAb	Iia(121)	-
SGLT2 inhibitors	remogliflozin etabonate, luseogliflozin, ipragliflozin	Iia(130,131)	-
	dapagliflozin, canagliflozin, empagliflozin	-	Iia(134, CREDENCE trial)

Abbreviations: AC: 1-Adamantaneformoxyl-3-(5′-hydroxymethyl-2′-furyl)indazole; .FGFR. FGF-21 receptor; GM-CT-01: galactomannan; GR-MD-02: galactoarabino-rhamnogalaturonan; PEG: pegylated; mAb: monoclonal antibodies; YC-1: 1-Benzyl-3-(substituted aryl)-5-methylfuro[3,2-c]pyrazole

Table 3 Role of incretin-based therapies and gut microbiota in the pathogenesis of NAFLD and CKD.

Incretin analogues: GLP-1 receptor agonists	
Cellular mechanism	**Biological effect**
Hypothalamus: ↓ appetite	↓ calorie intake
Skeletal miocyte: ↑ Glucose uptake ↑ Glycogen synthesis	↑ insulin sensitivity
Hepatocyte: ↑ Glycogen synthesis and ↓ Gluconeogenesis ↑ Autophagy ↑ cAMP→↑AMPK and SIRT-1 activity ↓ FGF-21 secretion	↑ insulin sensitivity ↓ hepatic steatosis
Adipocyte: ↑ Lipolysis ↑ Glucose uptake ↑ Lipogenesis (↑ FFA synthesis, uptake and reesterification, ↑ LPL activity) ↑ Adiponectin secretion	↑ insulin sensitivity ↓ adipose tissue dysfunction
Proximal tubule cell: ↓ NHE3 activity→↑ Na-uresis →↓ Na hyperreabsorption ↑ PPAR-α→↑ FFA oxidation **Glomerular endothelial cell, mesangial cell, tubule cell:** ↓ apoptosis ↓ AGE/RAGE-axis activation ↓ IL-1/MCP-1/TGF-β secretion→↓ monocyte recruitment and fibrogenesis ↑ NO synthase activity ↑ cAMP→NOX down-regulation→↓ ROS generation ↓ AngII activity→↓ IRS-1 phosphorylation → ↑ IRS-1 signalling	↑ Na delivery to distal tubule→ ↓ tubulo-glomerular feed-back→ ↓ glomerular hyperfiltration ↓ blood pressure ↓ fat infiltration ↓ glomerular sclerosis ↓ inflammation and fibrosis ↓ endothelial dysfunction ↑ insulin sensitivity
Incretin analogues: **DPP-4 inhibitors**	
Liver, kidney: ↑ GLP-1 activity	Same effects of GLP-1R

Kidney: ↑ BNP→ ↑natriuresis and vasodilation, ↓ RAS and sympathetic activity ↑ NPY and PYY→↑ AngII-mediated vascocontriction ↑ SDF-1a→↑ mesenchimal stem cells recruitment	agonists Ultimate effect depends on local tissue activity of different substrates inactivated by DPP-4
Altered gut microbiota composition and intestinal barrier disruption	
Cellular mechanism	**Biological effect**
Hepatocyte, macrophage, HSC, renal vascular endothelial cell, podocytes : ↑LPS-TLR-4 axis activation→ ↑proinflammatory cytokines/TGF-β ↑LPS-TLR-4 axis activation→ ↑NOX type II activity→ROS production	 hepatic and renal inflammation hepatic and renal fibrogenesis endothelial dysfunction
Enterocyte: ↓ SCFA production →↑epithelial injury and ↓ GLP-1 secretion	gut barrier disruption
Hepatocyte: ↑ conversion of intestinal TMA to trimethylamine-N-oxide (TMAO) by flavin-containing monooxygenases	 Hepatic steatosis Hepatic necroinflammation
Tubule cells: ↑ TMAO-induced TGF-β/SMAD-3 axis activation	atherosclerosis tubule-interstitial fibrosis
Skeletal miocyte: ↑ URM-induced phosphorylation of IRS-1→↓ IRS-1 signalling	Insulin resistance
Hepatocyte: ↑ URM-induced *de novo* lipogenesis ↓ URM-induced VLDL secretion	Insulin resistance Hepatic steatosis
Adipocyte: ↑ URM-induced leptin secretion ↓ URM-induced *de novo* lipogenesis ↑ URM-induced Zinc-α2-glycoprotein (ZAG) and ↓ perlipin expression →↑ lipolysis **Macrophage:**	 Insulin resistance Adipose tissue dysfunction Adipose tissue and systemic inflammation

↑ LPS-induced NF-kB activation	

Abbreviations: AGE: advanced glycation end-products; AMP: adenosine-monophosphate; AMPK: adenosine-monophosphate kinase; AngII: angiotensin II; BNP: brain-derived natriuretic peptide; cAMP: cyclic adenosine-monophosphate; DPP-4: dipeptidyl peptidase protein-4; ERK: extracellular signal-regulated kinase;; FFA: free fatty acids; FGF: fibroblast growth factor; GLP-1: glucagon-like peptide-1; IL: interleukin; IRS-1: insulin receptor substrate-1; LPL: lipoptorein lipase; LPS: lipopolysaccharide; MCP-1: monocyte chemotactic protein-1; NHE3: Na+-H+ exchanger 3; NF-κB: nuclear factor-κB; NO: nitric oxide; NOX: NADPH oxidase; NPY: neuropeptide Y; PPAR: peroxisome proliferators-activated receptor; PYY: peptide YY; RAAS: rennin-angiotensin-aldosterone system; ROS: reactive oxygen species; SDF-1a: stromal-derived Factor; SIRT: sirtuin; TGF-β: transforming growth factor- β; TLR: toll-like receptor; TMA: trimethylamine; TNF: tumor necrosis factor; VLDL: very low density lipoprotein.

Figure 1

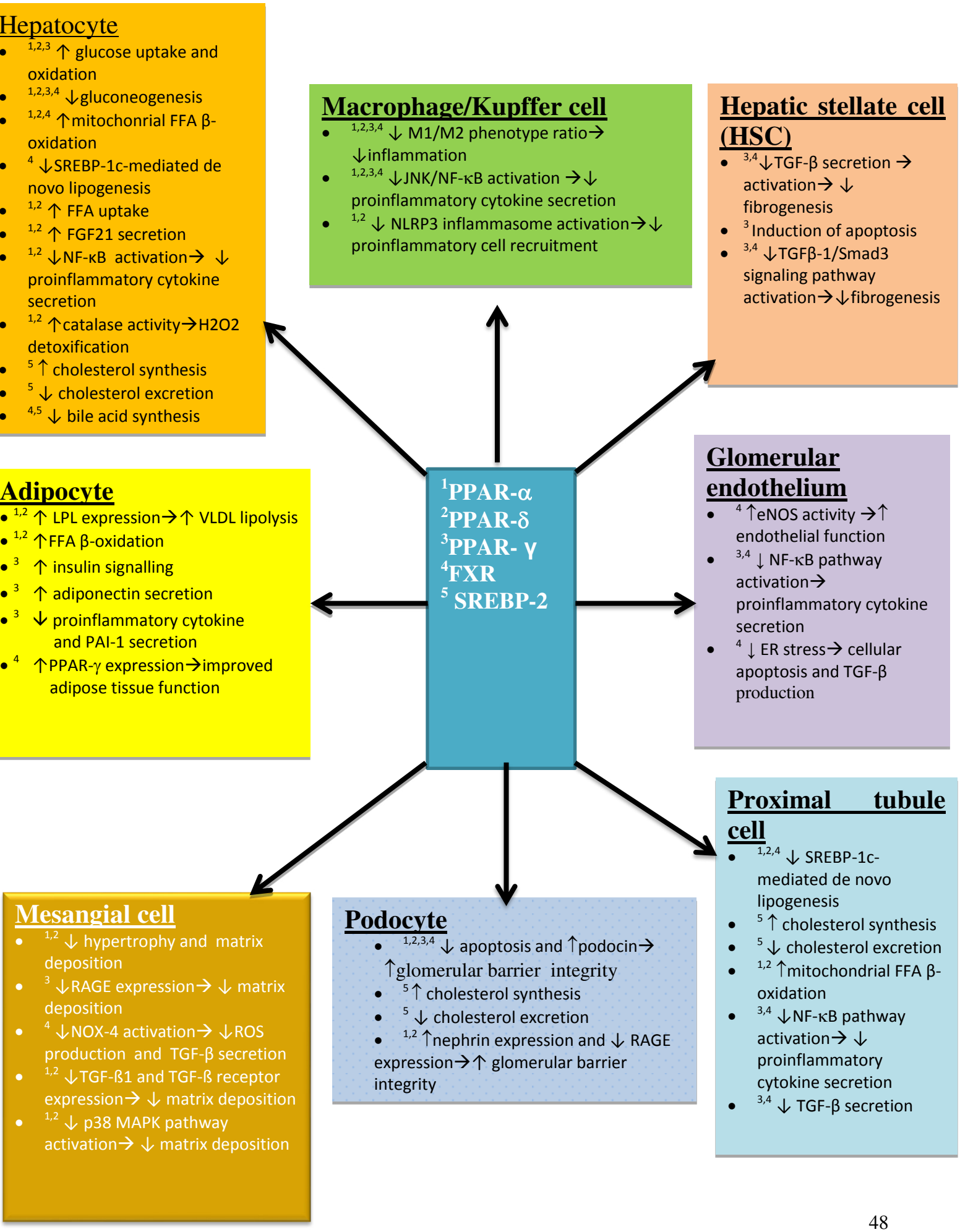

Hepatocyte
[1,2,3] ↑ glucose uptake and oxidation
[1,2,3,4] ↓gluconeogenesis
[1,2,4] ↑mitochonrial FFA β-oxidation
[4] ↓SREBP-1c-mediated de novo lipogenesis
[1,2] ↑ FFA uptake
[1,2] ↑ FGF21 secretion
[1,2] ↓NF-κB activation→ ↓ proinflammatory cytokine secretion
[1,2] ↑catalase activity→H2O2 detoxification
[5] ↑ cholesterol synthesis
[5] ↓ cholesterol excretion
[4,5] ↓ bile acid synthesis
Macrophage/Kupffer cell
[1,2,3,4] ↓ M1/M2 phenotype ratio→ ↓inflammation
[1,2,3,4] ↓JNK/NF-κB activation →↓ proinflammatory cytokine secretion
[1,2] ↓ NLRP3 inflammasome activation→↓ proinflammatory cell recruitment
Hepatic stellate cell (HSC)
[3,4]↓TGF-β secretion → activation→ ↓ fibrogenesis
[3] Induction of apoptosis
[3,4] ↓TGFβ-1/Smad3 signaling pathway activation→↓fibrogenesis
Adipocyte
[1,2] ↑ LPL expression→↑ VLDL lipolysis
[1,2] ↑FFA β-oxidation
[3] ↑ insulin signalling
[3] ↑ adiponectin secretion
[3] ↓ proinflammatory cytokine and PAI-1 secretion
[4] ↑PPAR-γ expression→improved adipose tissue function
[1]PPAR-α
[2]PPAR-δ
[3]PPAR- γ
[4]FXR
[5] SREBP-2
Glomerular endothelium
[4] ↑eNOS activity →↑ endothelial function
[3,4] ↓ NF-κB pathway activation→ proinflammatory cytokine secretion
[4] ↓ ER stress→ cellular apoptosis and TGF-β production
Mesangial cell
[1,2] ↓ hypertrophy and matrix deposition
[3] ↓RAGE expression→ ↓ matrix deposition
[4] ↓NOX-4 activation→ ↓ROS production and TGF-β secretion
[1,2] ↓TGF-ß1 and TGF-ß receptor expression→ ↓ matrix deposition
[1,2] ↓ p38 MAPK pathway activation→ ↓ matrix deposition
Podocyte
[1,2,3,4] ↓ apoptosis and ↑podocin→ ↑glomerular barrier integrity
[5] ↑ cholesterol synthesis
[5] ↓ cholesterol excretion
[1,2] ↑nephrin expression and ↓ RAGE expression→↑ glomerular barrier integrity
Proximal tubule cell
[1,2,4] ↓ SREBP-1c-mediated de novo lipogenesis
[5] ↑ cholesterol synthesis
[5] ↓ cholesterol excretion
[1,2] ↑mitochondrial FFA β-oxidation
[3,4] ↓NF-κB pathway activation→ ↓ proinflammatory cytokine secretion
[3,4] ↓ TGF-β secretion

Figure 2 Panel A

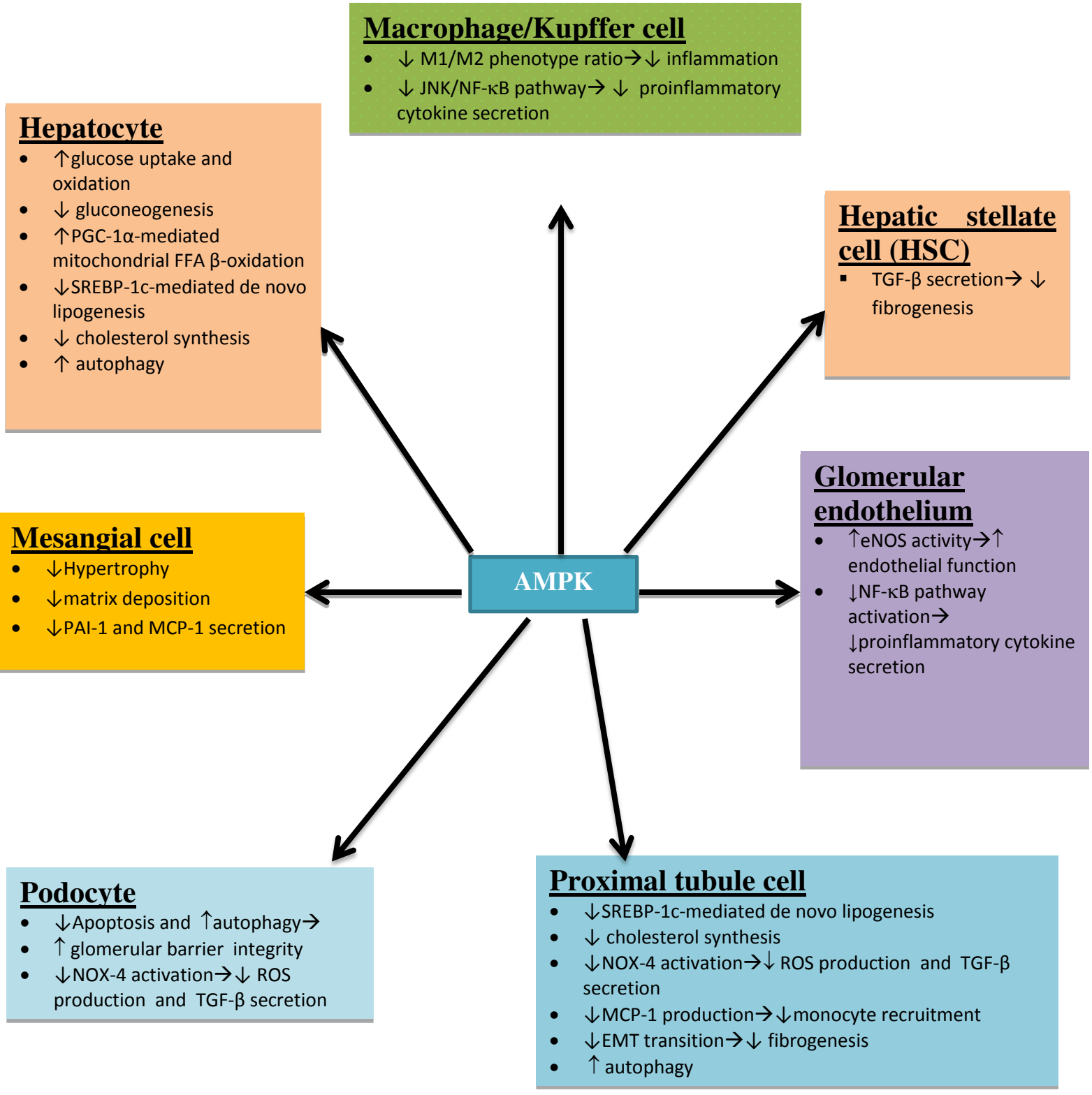


Figure 2 Panel B

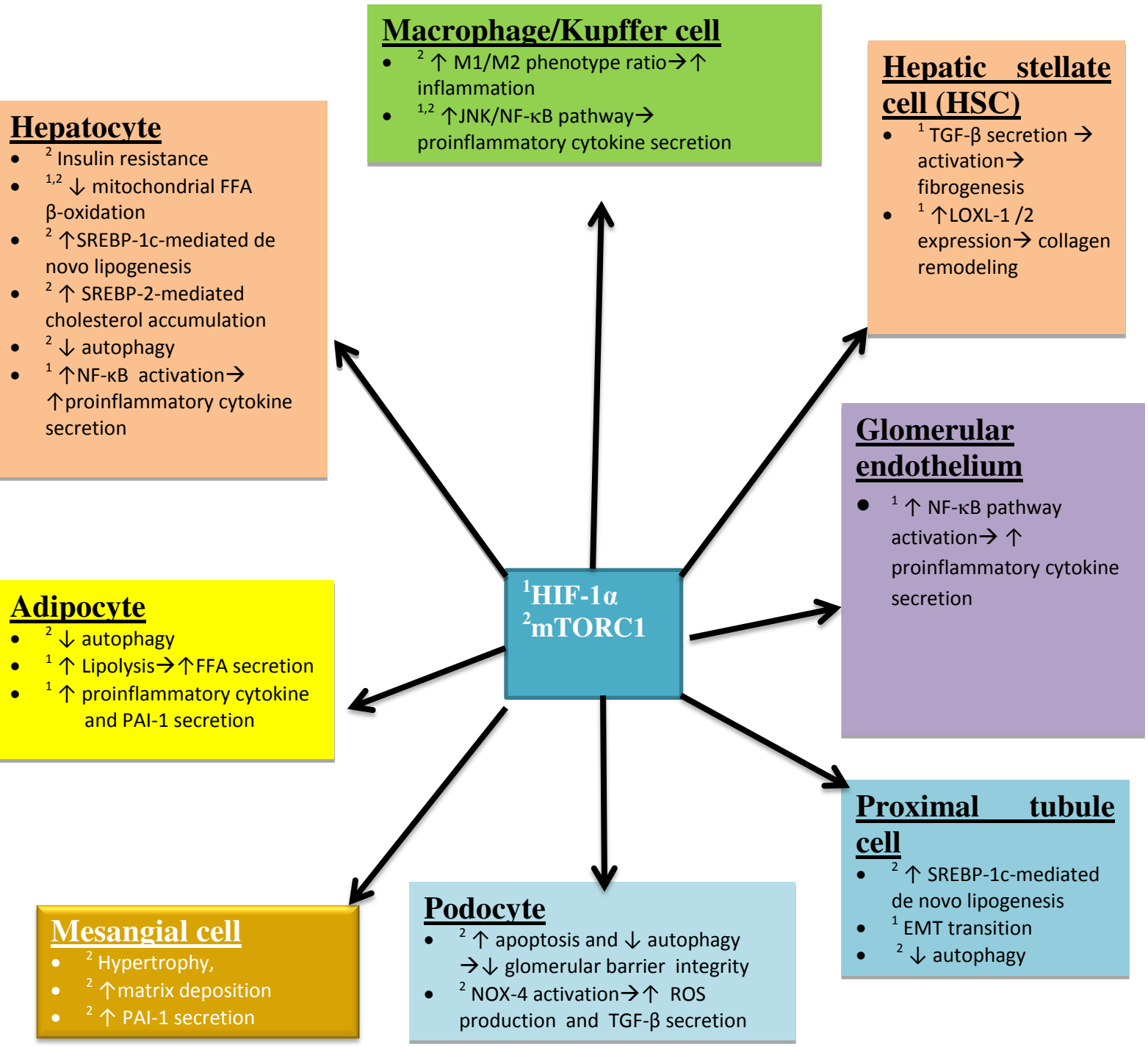


Figure 3

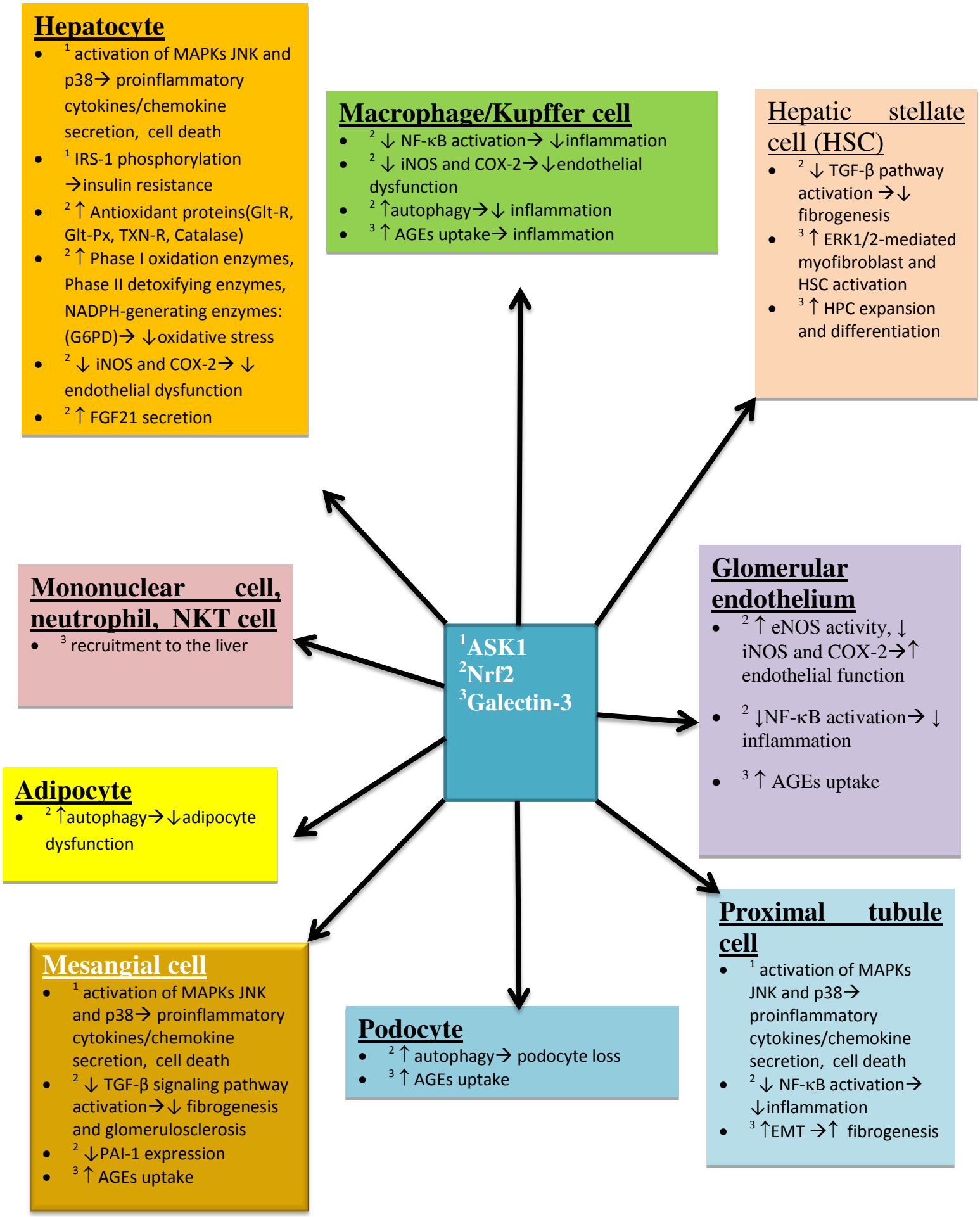

Hepatocyte
[1] activation of MAPKs JNK and p38→ proinflammatory cytokines/chemokine secretion, cell death
[1] IRS-1 phosphorylation →insulin resistance
[2] ↑ Antioxidant proteins(Glt-R, Glt-Px, TXN-R, Catalase)
[2] ↑ Phase I oxidation enzymes, Phase II detoxifying enzymes, NADPH-generating enzymes: (G6PD)→ ↓oxidative stress
[2] ↓ iNOS and COX-2→ ↓ endothelial dysfunction
[2] ↑ FGF21 secretion
Macrophage/Kupffer cell
[2] ↓ NF-κB activation→ ↓inflammation
[2] ↓ iNOS and COX-2→↓endothelial dysfunction
[2] ↑autophagy→↓ inflammation
[3] ↑ AGEs uptake→ inflammation
Hepatic stellate cell (HSC)
[2] ↓ TGF-β pathway activation →↓ fibrogenesis
[3] ↑ ERK1/2-mediated myofibroblast and HSC activation
[3] ↑ HPC expansion and differentiation
Mononuclear cell, neutrophil, NKT cell
[3] recruitment to the liver
[1]ASK1
[2]Nrf2
[3]Galectin-3
Glomerular endothelium
[2] ↑ eNOS activity, ↓ iNOS and COX-2→↑ endothelial function
[2] ↓NF-κB activation→ ↓ inflammation
[3] ↑ AGEs uptake
Adipocyte
[2] ↑autophagy→↓adipocyte dysfunction
Mesangial cell
[1] activation of MAPKs JNK and p38→ proinflammatory cytokines/chemokine secretion, cell death
[2] ↓ TGF-β signaling pathway activation→↓ fibrogenesis and glomerulosclerosis
[2] ↓PAI-1 expression
[3] ↑ AGEs uptake
Podocyte
[2] ↑ autophagy→ podocyte loss
[3] ↑ AGEs uptake
Proximal tubule cell
[1] activation of MAPKs JNK and p38→ proinflammatory cytokines/chemokine secretion, cell death
[2] ↓ NF-κB activation→ ↓inflammation
[3] ↑EMT →↑ fibrogenesis

Figure 4A

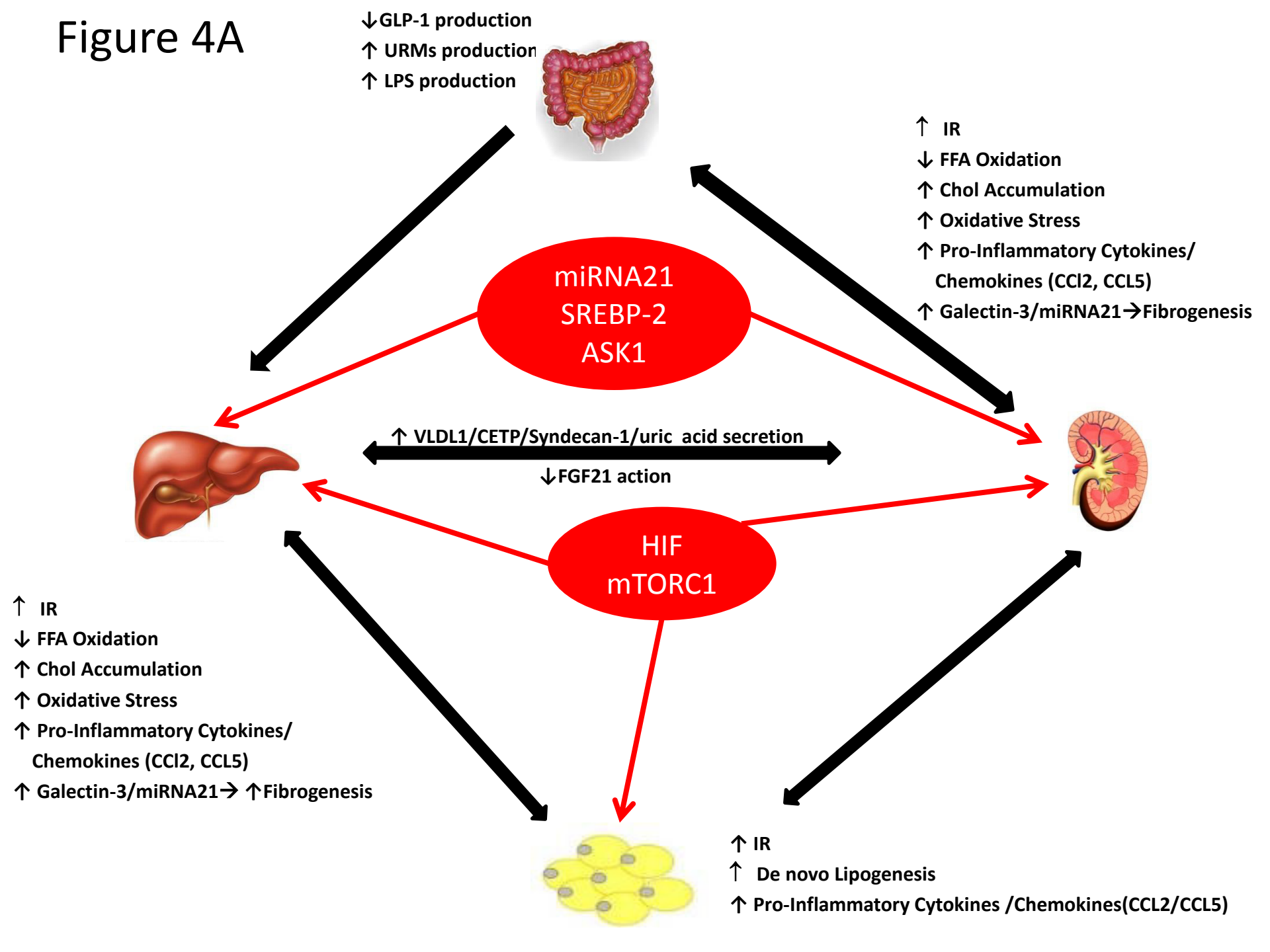
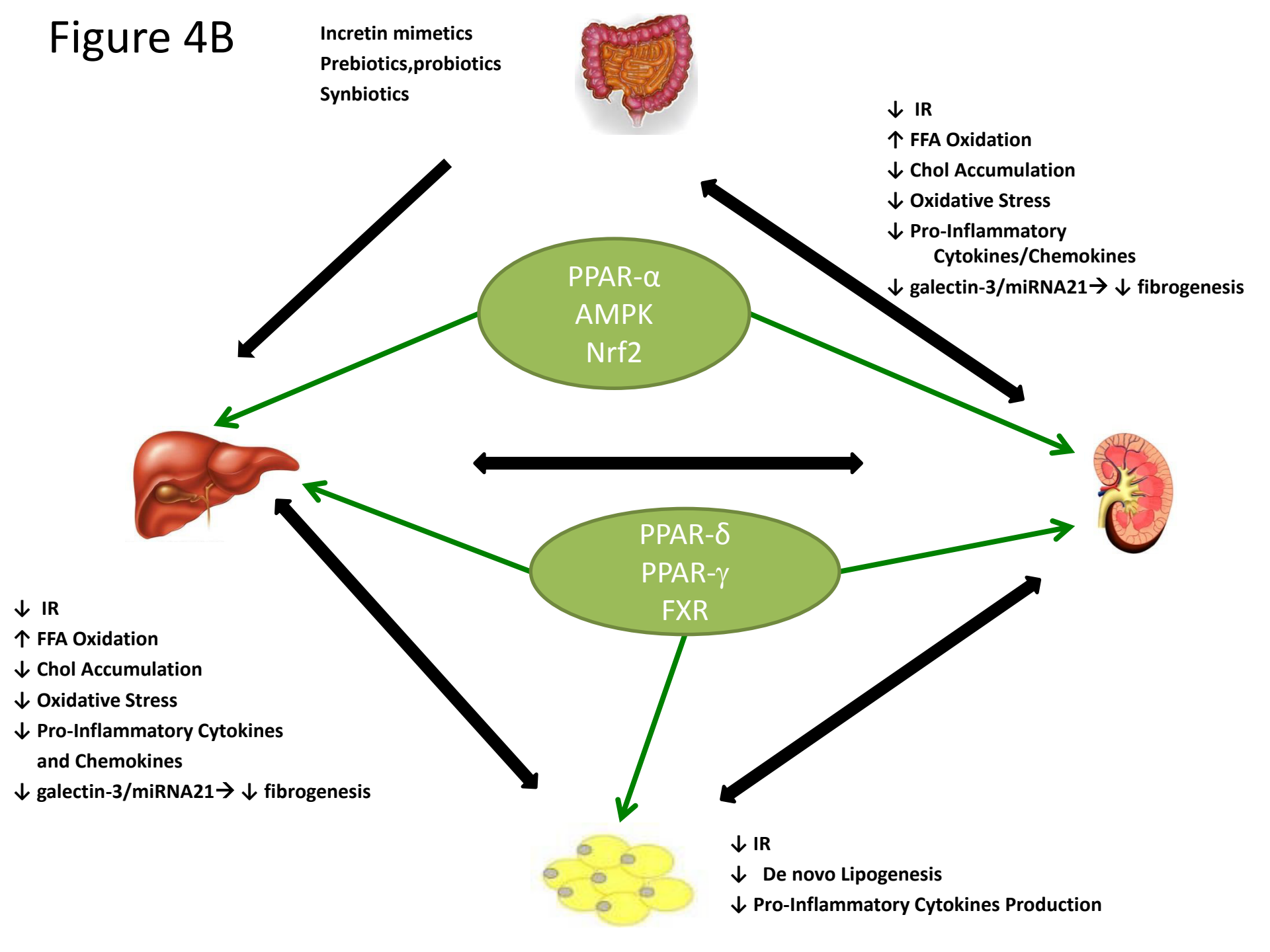

↓GLP-1 production
↑ URMs production
↑ LPS production
↑ IR
↓ FFA Oxidation
↑ Chol Accumulation
↑ Oxidative Stress
↑ Pro-Inflammatory Cytokines/
Chemokines (CCl2, CCL5)
↑ Galectin-3/miRNA21→Fibrogenesis
miRNA21
SREBP-2
ASK1
↑ VLDL1/CETP/Syndecan-1/uric acid secretion
↓FGF21 action
HIF
mTORC1
↑ IR
↓ FFA Oxidation
↑ Chol Accumulation
↑ Oxidative Stress
↑ Pro-Inflammatory Cytokines/
Chemokines (CCl2, CCL5)
↑ Galectin-3/miRNA21→ ↑Fibrogenesis
↑ IR
↑ De novo Lipogenesis
↑ Pro-Inflammatory Cytokines /Chemokines(CCL2/CCL5)

Figure 4B


Incretin mimetics
Prebiotics,probiotics
Synbiotics
↓ IR
↑ FFA Oxidation
↓ Chol Accumulation
↓ Oxidative Stress
↓ Pro-Inflammatory Cytokines/Chemokines
↓ galectin-3/miRNA21→ ↓ fibrogenesis
PPAR-α
AMPK
Nrf2
PPAR-δ
PPAR-γ
FXR
↓ IR
↑ FFA Oxidation
↓ Chol Accumulation
↓ Oxidative Stress
↓ Pro-Inflammatory Cytokines and Chemokines
↓ galectin-3/miRNA21→ ↓ fibrogenesis
↓ IR
↓ De novo Lipogenesis
↓ Pro-Inflammatory Cytokines Production