O_3 Effect on Kidney Damage

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Ozone (O_3) is a reactive oxygen species (ROS) that can interact with cellular components and cause oxidative stress. Following said logic, if O_3 induces such a stressful milieu, how does it exert antioxidant functions? This is mediated by controlled toxicity produced by low concentrations of O_3, which enhance the cell's suppliance of antioxidant properties without causing any further damage. O_3 therapy has been shown to be effective when applied before or after traumatic renal procedures, whether caused by ischemia, xenobiotics, chronic damage, or other models.

ozone ozone therapy kidney disease oxidative stress

1. O_3 Therapy Protects the Kidney against Ischemic Damage

Ischemic damage in renal tissue occurs when kidneys experiment periods of diminished or restricted blood supply. In contrast, oxidative damage occurs when tissue is re-oxygenated, which might happen during experimental procedures in rats, such as clamping and unclamping renal pedicle, or during renal transplantation [1]. This kind of damage is proposedly produced through xanthine oxidase (XO). This enzyme degrades nucleotides upon cell ischemia. However, after O_2 reperfusion, XO forms uric acid and high quantities of superoxide radical, which further produces oxidative stress [2]. This explains why treatment with XO inhibitors, such as tungsten [2], allopurinol [3], or even XO knockout models [4], ameliorates ischemia-reperfusion injury (IRI) and oxidative stress after short periods of ischemia. Finding auxiliary treatments for oxidative damage is clinically important since ischemic-producing scenarios are highly prevalent. Just in 2010, for instance, more than 2 million patients received renal transplants [5].

O_3 therapy has previously been used before IRI (preconditioning) [6][7][8][9][10][11] or after IRI (postconditioning) [12][13][14][15][16] and has been described as a potential treatment (**Table 1**). O_3 therapy is demonstrated to act with similar efficacy, but not synergic, to that achieved when IRI preconditioning is made with other protective strategies, such as inducing short, repeated periods of ischemia before the main IRI. This prepares the renal tissue against the IRI via similar controlled mechanisms as that of the O_3 and is called ischemic (pre)conditioning [17]. Interestingly, when administered after the main IRI, ischemic postconditioning in conjunction with O_3 therapy upregulate beneficial effects and even diminishes cell death [18]. After transplantation, rats also show a protective effect against the oxidative state when treated with O_3 [19][20]. Antioxidant enzymes are also upregulated in cultured kidney cells after they were submitted to hypoxia and reoxygenation [21].

Table 1. Ozone (O_3) effects on ischemic damage models.

Damage Model	Induced Procedure	O_3 Administration	Effects in O_3 Treated Rats	Ref.
O_3 oxidative preconditioning Therapy				
Kidney transplantation	Right Nephrectomy and left transplant	15 (1 daily) preconditioning rectal insufflations 1 mg/kg at [50 µg/mL] to the donor rat	↑ SOD, GSH Px ↓ SCr, BUN, MDA ↓ Morphologic damage ↓ IL-6, IL-18, COX2 ↓ NF-κB, HMGB1	[19]
Kidney transplantation	Right nephrectomy and left transplant	15 (1 daily) preconditioning rectal insufflations 1 mg/kg at [50 µg/mL] to the donor rat	↑ SOD, GSH, CAT ↑ Nrf2, HO-1 ↓ SCr, BUN, MDA ↓ Morphologic damage	[20]
Right nephrectomy and left pedicle clamping	45 min ischemia 24 h reperfusion	Preconditioning therapy 15 previous rectal insufflations, 1 mg/kg at [50 µg/mL]	↓ BUN, SCr ↓ Medullar Hemorrhage ↓ TNF-α, IL-1β, IL-6, ICAM-1, ↓ MCP-1, TLR4, NF-kB	[6]
Right nephrectomy and left pedicle clamping	60 min ischemia 60 min reperfusion	Preconditioning therapy OA, 1 mL of blood added with 5 mL of O_3 [50 µg/mL] before and after IR	↑ iNOS ↑ β NADPH diaphorase ↓ BUN, SCr ↓ Medullar damage	[7]
Right nephrectomy and left pedicle clamping	45 min ischemia 24, 48, 72 h reperfusion	Preconditioning therapy 15 previous rectal insufflations, 1 mg/kg at [50 µg/mL]	↑ GSH, GSH-Px, SOD ↑ NO, iNOS, eNOS ↓ BUN, SCr ↓ Morphologic damage ↓ MDA ↓ ET-1	[8]
Right nephrectomy and left pedicle clamping	45 min ischemia 8-week reperfusion	Preconditioning therapy rectal pathway, 1 mg/kg at [50 µg/dL]	↑ SMAD-7 ↓ α- SMA, TGF-β BUN, SCr not significant	[9]
Right nephrectomy and left pedicle clamping	45 min ischemia and reperfusion	Preconditioning 15 (1 daily) doses by rectal insufflation, 1 mg/kg at [50 µg/mL]	↓ SCr, BUN, MDA ↓ Morphologic damage ↓ICAM-1, IL-1β, TNF-α, Caspase 3	[10]

Damage Model	Induced Procedure	O_3 Administration	Effects in O_3 Treated Rats	Ref.
Bilateral pedicle clamping	30 min ischemia and 3 h reperfusion	Preconditioning 15 (1 daily) 2.5–2.6 mL at [50 mg/mL] at a dose of 0.5 mg/kg by rectal insufflation	↑ RPF, GFR (inulin) ↑ SOD ↓ Morphologic damage	[11]
O_3 oxidative postconditioning therapy				
Bilateral Renal Artery Occlusion	60 min ischemia 6 h reperfusion	Postconditioning therapy single 0.7 µg/kg i.p. immediately after reperfusion	↑ SOD, GSH-Px, ↓ SCr, BUN ↓ AST, Neopterin ↓ MDA, PCC, NOx ↓ Morphologic damage	[14]
Left nephrectomy and right pedicle clamping	45 min ischemia 24 h reperfusion	Postconditioning therapy 1 and 2 mg/kg; 15 (1 daily) doses after IRI at [50 µg/mL] by rectal insufflation	↑ SOD ↓ SCr, BUN, MDA ↓ Morphologic damage ↓ BAX, PARP, CREB, c-Fos	[13]
Right Nephrectomy and Left pedicle clamping	45 min ischemia 10 days reperfusion	Postconditioning therapy 10 daily rectal insufflations after IRI, a 2.5 mL volume at 0.5 mg/kg/min [50 µg/mL]	↑ SOD ↓ SCr, BUN ↓ MDA, MPO ↓ Morphologic damage ↓ α-SMA, TGF-β, p-SMAD-2	[12]
Renal vascular bundles clamping	60 min ischemia 10 days reperfusion	Postconditioning therapy Daily 10 days after IRI At 0.5 mg/kg/min via rectal insufflation	↓ Proteinuria ↑ RPF, Glomerular Filtration Rate ↓ Morphologic Damage	[15]
Bilateral Renal Artery Occlusion	60 min ischemia and 10-day reperfusion	10 (1 daily) 2.5–2.6 mL at [50 mg/mL], representing a dose of 0.5 mg/kg weight rectal insufflations	↑ CAT, SOD ↓ SCr, Fructosamine ↓ Phospholipase A2	[16]
Right nephrectomy and left pedicle clamping	45 min ischemia and 24 h reperfusion	Ischemic Preconditioning vs. O_3 Preconditioning, 15 rectal insufflations at [50 µg/mL])	↑ NO ↑ GSH, GSP-Px, SOD ↓ BUN, SCr, MDA	[17]
Right nephrectomy and left pedicle clamping	45 min ischemia and 24 h reperfusion	Comparison Ischemic Post conditioning vs. O_3 post conditioning, 2 mg/kg	↓ IL 1, IL 18, Caspase 1 ↓ SCr, BUN, MDA	[18]

dants that protective mechanism favored by O_3 therapy against IRI inflammation and vasoconstriction caused by Endothelin-1 [7][8]. In fact, nitrate-derived NO, when applied topically, is an effective therapy against IRI damage [23].

Damage Model	Induced Procedure	O_3 Administration	Effects in O_3 Treated Rats	Ref.
			↓ Morphologic Damage	

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clinical conditions that reduce renal blood flow, such as those that produce AKI.

Abbreviatures: ↑: significant increase, ↓: significant decrease, α-SMA: α-smooth muscle actin, AST: aspartate aminotransferase, BAX: bcl-2-associated X, BUN: blood urea nitrogen, CAT: catalase, COX2: cyclooxygenase 2, CREB: cAMP response element-binding, eNOS: endothelial nitric oxide synthase, ET-1: endothelin-1, FF: filtration fraction, GFR: glomerular filtration rate, GSH-Px: glutathione peroxidase, GSH: glutathione, HMGB1: high mobility group Box 1, HO-1: heme oxygenase 1, ICAM-1: intercellular adhesion molecule 1, IL-1β: interleukin 1β, IL-6: interleukin 6, iNOS: inducible nitric oxide synthase, IRI: ischemia/reperfusion injury, MCP-1: monocyte chemoattractant protein 1, MDA: malondialdehyde, NF-kB: nuclear factor kappa B, NO: nitric oxide, O_3: ozone, OA: ozonated autohemotherapy, PARP: polymerase 1, PCC: protein carbonyl content, RPF: renal plasma fraction, SCr: serum creatinine, SMAD 7 and 2: suppressor of mothers against decapentaplegic family members 7 and 2, SOD: superoxide dismutase, β NADPH diaphorase: β-nicotinamide adenine dinucleotide phosphate diaphorase, TGF-β: transforming growth factor β, TLR 4: Toll-Like receptor 4, TNF-α: tumor necrosis factor α.

2. O_3 Therapy Protects the Kidney against Xenobiotic-Induced Damage

Xenobiotics are exogenous chemicals not synthesized by a certain organism, therefore, they are not essential for its physiological functions and processes. That way, synthetical drugs, metals, and environmental factors, amongst others, are considered as such [24]. In this section, the mechanisms through which some of these xenobiotics cause nephrotoxicity will be discussed, along with the described protective effects of O_3 therapy against it, looking forward to discovering the usage of new therapeutic alternatives against damaging products we are constantly in contact with (**Table 2**).

Acetaminophen (APAP), a common anti-inflammatory drug, has been demonstrated to produce severe nephrotoxicity [25]. Proposed mechanisms include APAP's hepatic degradation and further enzymatic formation of a highly toxic and reactive metabolite, N-acetyl-p-benzoquinone (NAPQI), which glutathione (GSH) normally neutralizes. However, in APAP overdose, NAPQI is formed in major quantities, proving uncontainable by antioxidant enzymes, and therefore producing oxidative damage, especially in proximal tubules [26]. O_3 therapy has proven to be an effective antioxidant therapy by enhancing antioxidant enzymes and diminishing oxidation [25]. Interestingly, the administration of O_3 therapy in APAP induced nephrotoxicity, when combined with another antioxidant therapy, N-acetylcysteine (NAC), produced no significant changes in the kidney's function (creatinine, urea) and inflammation (IL-6, IL-10) but did produce significant changes against oxidative stress, showing lower levels of MDA, as well as a reduction of histopathologic glomerular, tubular, and interstitial damage [27].

Cadmium (Cd) is a heavy non-essential metal that is accumulated in body tissues progressively [28] and to which humans are exposed through air particles [29], occupational exposure [30] and seafood such as mollusks, crustaceans, or fish [31]. Cd can produce nephrotoxicity by many mechanisms, including DNA damage, altered gene expression, and, most importantly, oxidative damage by depleting cells' antioxidant defenses, such as selenium, which binds to Cd to neutralize it [32]. Other proteins, e.g., metallothionein (MT), bind Cd in others to diminish its toxicity in organs such as kidneys and testis [33][34]. O_3 therapy can diminish Cd accumulation, augment MT levels, and reduce morphologic damage, serving as an effective protective mechanism against Cd^{2+} renal damage [33]. It also reduces N-acetyl-β-D-glucosaminidase (NAG) [35], a lysosomal enzyme found mainly in proximal convoluted tubules, its function is the digestion of cell's glycoconjugates [36]. The NAG increase is mediated by loss of the tubular brush border, thus liberating the enzyme into the urine [37]; such an increase is associated with pathologic processes such as Cd intoxication and malignancies of the kidney, liver, pancreas, lung, and breast, amongst many others [35][38], as well as an increased risk of requiring dialysis treatment and lethality in hospitalized patients [37]. Even when stimulating lipid peroxidation, as a result, O_3 was also demonstrated to induce antioxidant enzymes in Cd-treated rats [39].

Some antineoplastics are proven to cause nephrotoxicity. For instance, doxorubicin, often known as Adriamycin, binds to cell membranes and inhibits nucleotide replication. However, it can be oxidized into forming reactive species like hydroxyl radicals [40]. It is demonstrated to cause severe progressive damage, fibrosis, and proteinuria [41]. O_3 therapy, in certain doses, has proven to mediate protective effects against this morphologic damage, and arterial pressure, as well as proteinuria, have been ameliorated in rats receiving this treatment [42].

Another example is cisplatin (CDDP), an FDA (American Food and Drug Administration) approved treatment for advanced solid cancers such as that of the testis, ovary, and bladder [43]. CDDP is a molecule composed of a single platinum atom bound to chloride and ammonium; due to its small size, it filtrates freely into the glomerular barrier without tubular reabsorption [44]. It then enters tubular cells and dissociates into its toxic components, which damage DNA, membrane transporters, and mitochondrial function, thus producing oxidative stress, inflammation, and apoptosis [44][45]. O_3 has been used as a therapy against CDDP induced damage, improving function and augmenting antioxidant defenses. Thiobarbituric acid reactive substances (TBARS, an assay used to measure lipid peroxidation; [46]), as well as NAG and morphologic damage, displayed decreased values when treated with O_3 [47][48][49]. Protective effects, however, varied according to the administered O_3 concentration, given that the administration of 0.36 mg/kg might be therapeutic [34] or might not [49]. On the other hand, 1.1 mg/kg always shows protective tendencies in CDDP-induced damage [47][48][49]. Higher concentrations, e.g., 1.8 mg/kg, might be protective [36]. However, due to the high formation rate of hydrogen peroxide and oxidative stress mediated by O_3, toxic effects might be produced [47]. Very similar protective morphologic, anti-inflammatory, and antioxidant effects have been found against the damage induced by methotrexate, another cancer drug, in the kidneys, as well as the intestines and liver [50].

Radiographic contrast media (CM) is constantly used in clinical procedures which require the observation of vascular compartments. Mechanisms through which CM might cause renal dysfunction include direct oxygen-free radical damage, modified hemodynamics, and hypoxic renal medullary injury mediated by shortness of blood flow and an increase in tubular O_2 supply. Therefore, the employment of CM produces high toxicity [51], which can be treated with O_3. Neutrophil gelatinase-associated lipocalin (NGAL) is a damage marker observed in contrast-induced nephropathy (CIN) which augmented its expression when treated with O_3; no further discussion was provided, although the initial oxidation by O_3 might have produced it [52][53].

In the medical field, the use of xenobiotics as drugs to treat and diagnose diseases is an irreplaceable factor. However, during their metabolism and excretion, some might become nephrotoxic by accumulation, directing damage, the formation of free radicals, and depletion of antioxidant substances. This represents a risk for patients with neoplasia or other conditions which require constant chemical induction or those in contact with environmental components such as Cd, which is also demonstrated to cause similar renal damage. However, O_3 is an effective treatment against this damage, at least experimentally, and thus the importance of further research in clinical environments.

Table 2. Ozone (O_3) effects on chemical-induced damage models.

Damage Model	Induced Procedure	O_3 Administration	Effects in O_3 Treated Rats	Ref.
APAP toxicity	A 1.0 g/kg dose suspended in H_2O, 3 mL: orally	Single i.p. 0.7 mg/kg dose at [60 mg/mL] Immediately after APAP induction	↑ SOD, GSH-Px ↓ SCr, BUN ↓ MDA ↓ Morphologic damage	[25]
APAP toxicity	A 1.0 g/kg dose suspended in H_2O, 3 mL: gastric tube	5 daily 0.7 mg/kg doses i.p. at [60 mg/mL] Immediately after APAP induction	↑ GSH-Px, IL-10 ↓ Morphologic damage ↓ MDA ↓TNF-α	[27]
Experimental toxic adriamycin-induced glomerulonephritis	Adriamycin single 7.5 mg/kg dose through a jugular vein; 10-week evolution	After 10 weeks, daily for 15 days at 0.3 mg/kg or 0.5 mg/kg or 0.7 mg/kg, or 1.1 mg/kg	(0.3 mg/kg) ↓ Arterial pressure ↓ Proteinuria (0.5 mg/kg) ↓ Morphologic damage (0.7 and 1.1 mg/kg) No significant changes	[42]
Cd intoxication	Drinking water with Cd^{2+} (50 mg/L) in the form of Cadmium Acetate for 12 weeks	10 (1 daily) 1 mL i.p. doses at [40 µg/mL]	↓ Morphologic damage ↓ Glomerulonephritis ↓ NAG	[35]
Cd Intoxication	Drinking water with Cd^{2+} (50 mg/L) in the form of Cadmium Acetate for 12 weeks	10 (1 daily) 1 mL i.p. doses at [40 µg/mL]	↑ MT ↓ Morphologic damage	[33]
CDDP induced nephrotoxicity	Single 6 mg/kg CDDP injection	Preconditioning 15 (1 daily) doses by rectal insufflation, 9 mL at concentrations of [0.36, 0.72, 1.1, 1.8, 2.5 mg/kg]	↑ GSH, SOD, CAT, GSH-Px ↓ SCr ↓ TBARS	[47]
CDDP induced nephrotoxicity	Single 6 mg/kg CDDP injection	Postconditioning 6 (1 daily) rectal insufflations, 9 mL volume with concentrations of: 10 mg at [0.36 mg/kg] or 30 mg at [1.10 mg/kg] or 50 mg at [1.80 mg/kg]	↑ GSH, SOD, CAT, GSH-Px ↓ SCr ↓ TBARS	[49]
CDDP induced nephrotoxicity	Single 6 mg/kg CDDP injection	Daily; 5 days before and 5 days after CDDP injection. i.p.at 1.1 mg/kg	↑ CAT, SOD ↑ NAG, TGF-β1, IL-6 ↓ Morphologic damage ↓Urea, creatinine, uric	[48]

Damage Model	Induced Procedure	O_3 Administration	Effects in O_3 Treated Rats	Ref.
			acid, phosphorus, calcium, sNGAL, albumin ↓ NF-a, IL-1B,	
CIN	10 mg/kg injected through the tail vein	1. 6 h before and 6 h after OR 2. For 5 days after; contrast agent introduction. O_3 at 1 mg/kg, 95% i.p.	1. ↑ NGAL ↓Hemorrhage 2. ↑TAC, similar SCr ↓Renal tubular injury	[53]
CIN	6 mL/kg of meglumine/sodium diatrizoate through the tail vein	Five 0.7 mg/kg/d doses i.p. [70 µg/mL] For 5 days before CIN	↑ NO ↑ TAS ↓ SCr, BUN ↓ MDA ↓ Tubular necrosis	[52]

Abbreviatures: ↑: significant increase, ↓: significant decrease. APAP: acetaminophen, BUN: blood urea nitrogen, CAT: catalase, Cd: cadmium, CDDP: cisplatin, CIN: contrast-induced nephropathy, GSH: glutathione, GSH-Px: glutathione peroxidase, IL-10: interleukin 10, i.p.: intraperitoneal route, MDA: malondialdehyde, MT: metallothionine, NAG: N-acetyl-β-D-glucosaminidase, NGAL: neutrophil gelatinase-associated lipocalin, NO: nitric oxide, O_3: ozone, SCr: serum creatinine, SOD: superoxide dismutase, TAC: total antioxidant capacity, TAS: total antioxidant system, TBARS: thiobarbituric acid reactive substances, TGF-β1: transforming growth factor β1, TNF-α: tumor necrosis factor-alpha.

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other abnormalities detected by imaging, for at least three months [55]. Many factors are involved in its development, such as hypertension, pollution, glomerulonephritis, and, most importantly, type 2 diabetes mellitus [56]. In this section, the effects of O_3 therapy against CKD will be discussed, hoping to decipher the use of new therapeutic alternatives to delay or prevent this pathology (**Table 3**).

Several procedures are induced in rats to simulate CKD, such as subtotal (5/6) nephrectomy, which exposes remaining renal tissue to high pressure and perfusion, eventually diminishing renal function and hence great inflammation. O_3 can ameliorate this condition, enhancing kidney function and antioxidant status. TBARS showed higher levels, possibly due to O_3 mediated oxidative stress [57][58]. Adenine administration also simulates CKD through its enzymatic degradation by xanthine dehydrogenase and further accumulation of the product 2,8-dihydroxyadenine (DHA) in the renal tubules, leading to inflammation and oxidative stress [59]. O_3 ameliorated this damaging condition mainly by stimulating the expression of antioxidant enzymes and reducing inflammation [60][61].

Diabetic kidney disease (DKD) is the main cause of CKD. It is a chronic condition caused by diabetes (whether type 1 or 2) via apoptosis, formation of free radicals, advanced glycation end-products (AGES), inflammatory cytokines, and other growth molecules. [62]. Diagnosis is made essentially through diminished GFR and proteinuria in humans. Risk factors include smoking habits and high arterial pressure. The discussion of this disease becomes important since its prevalence, and therefore that of CKD, is augmenting [63]. In experimental DKD studies that use streptozotocin (STZ) as a toxic component to β-cells, O_3 has shown beneficial anti-apoptotic and antioxidative effects in response [64][65].

Table 3. Ozone (O_3) effects on chronic kidney damage models.

Damage Model	Induced Procedure	O_3 Administration	Effects in O_3 Treated Rats	Ref.
Adenine Induced CKD	0.75% adenine diet for 4 weeks	1.1 mg/kg at [50 µg/mL] Via rectal insufflation	↓ SCr, BUN, K, Ca ↓ Morphologic damage ↓ MCP-1, TNFα, IL-1b,	[60]

Damage Model	Induced Procedure	O_3 Administration	Effects in O_3 Treated Rats	Ref.
			IL-6 ↓TLR 4, NFkB, p65	
Subtotal Nephrectomy CKD	Right nephrectomy and left subtotal ablation. 10-week evolution	1.1 mg/kg at [50 μg/mL] Via rectal insufflation Once a day for 2 weeks	↓ TNFα, IL-1β, IL-6, ↓ SCr, BUN, K, Ca ↓ Morphologic damage ↓NLRP3, NFkB, ASC, Caspase 1	[57]
Subtotal Nephrectomy CKD	Right nephrectomy and left subtotal ablation. 10-week evolution	2.5 mL at [50 μg/mL] Dose of 0.5 mg/kg Once a day for 15 days	↑ RPF, GFR ↑ SOD, CAT, GSH, TBARS ↓ Systolic arterial pressure ↓ SCr, BUN ↓ Morphologic damage	[58]
Diabetic Nephropathy	Streptozotocin induced Diabetes 6-week evolution	1.1 mg/kg [50 μg/mL] i.p.	↑ SOD, GPx, CAT ↓ BP, Hb A_{1c} % ↓ BUN, SCr, AR, MDA	[64]
Diabetic Nephropathy	Streptozotocin induced Diabetes 6-week evolution	1.1 mg/kg [50 μg/mL] once a day for 6 weeks	↓ Caspases 1, 3, 9; HIF-1α, TNF-α, Glc, morphologic damage	[65]

Abbreviations: ↑: significant increase, ↓: significant decrease, AR: aldose reductase, ASC: apoptosis-associated speck-like protein containing a CARD, BP: blood pressure, BUN: blood urea nitrogen, Ca: calcium, CAT: catalase, CKD: chronic kidney disease, GFR: glomerular filtration rate, Glc: glucose, GPx: glutathione peroxidase, GSH: glutathione, Hb A_{1c} %: glycosylated hemoglobin, HIF-1α: hypoxia inducible factor 1α, IL: Interleukins, i.p.: Intraperitoneal route, K: potassium, MDA: malondialdehyde, MCP-1: monocyte chemoattractant protein-1, NFkB: nuclear factor kappa B, NLRP3: (NLR) family pyrin domain containing 3, O_3: ozone, p65: 5 kDa polypeptide, RPF: renal plasma flow, SCr: serum creatinine, SOD: superoxide dismutase, TBARS: thiobarbituric acid reactive substances, TLR 4: Toll-like receptor 4, TNF-α: tumor necrosis factor alpha.

ialysis, as the indicated treatment [66]. O_3 has been shown as a coadjutant therapy to dialysis, as demonstrated by case reports in which conventional treatment did not work. For example, Biedunkiewicz and collaborators [67] described the case of a dialyzed patient with calciphylaxis-induced ulcerations who did not respond to antibiotics and surgical treatment. Ozonated autohemotherapy in concentrations of 50 μg/mL, as well as O_3 topic administration, allowed a successful skin transplant. Authors propose that effects are mediated through O_3 induced the synthesis of platelet-derived growth factor (PDGF), TGF-β1, and IL-8. Paolo and collaborators [68] described the case of a hemodialyzed patient who presented necrotizing fasciitis and a fatal prognosis. However, after extracorporeal blood oxygenation and ozonization (EBOO) and O_3 topic administration, drastic wellness, including diminished hyperpyrexia and restoration of skin lesions, was reported. EBOO might be a more comfortable and practical alternative to O_3 administration to patients over i.p. or rectal insufflation pathways, and its safety has also been proven experimentally [69]. A clinical trial in hemodialyzed patients conducted by Tylicki and collaborators [57] showed diminished GSH levels after nine weeks of O_3 treatment, possibly caused by an augmented antioxidant system that consumes GSH. The same authors found no difference in NK cell activity after O_3 therapy, indicating it as a safe treatment in hemodialyzed patients [70]. Interestingly, another case report concluded that this therapy might cause heart failure in complex patients, such as those with CKD, diabetes, and hypertension. This association resulted from the speculation that O_3 therapy augmented K^+ serum levels, which, along the diminished excretion, produced sinus arrest [71]. Contrasting effects were found by Gu and collaborators [72], who treated patients suffering from chronic hepatitis with O_3 and, while measuring kidney function, found diminished renal damage, augmented renal blood flow, and even a significant association with lesser fatalities.

To sum up, CKD is usually caused by diabetes. Both are highly prevalent, and dialysis is the standard treatment in advanced stages. O_3 treatment is useful against these chronic diseases by reducing inflammation and oxidative stress. On top of that, O_3 works as a coadjutant therapy for dialyzed patients to ameliorate not only kidney function, but aggravated topical microbial infections, which are common. **Figure 1** shows the effects of ozone on ischemia/reperfusion, renal damage by xenobiotics, and chronic kidney disease.

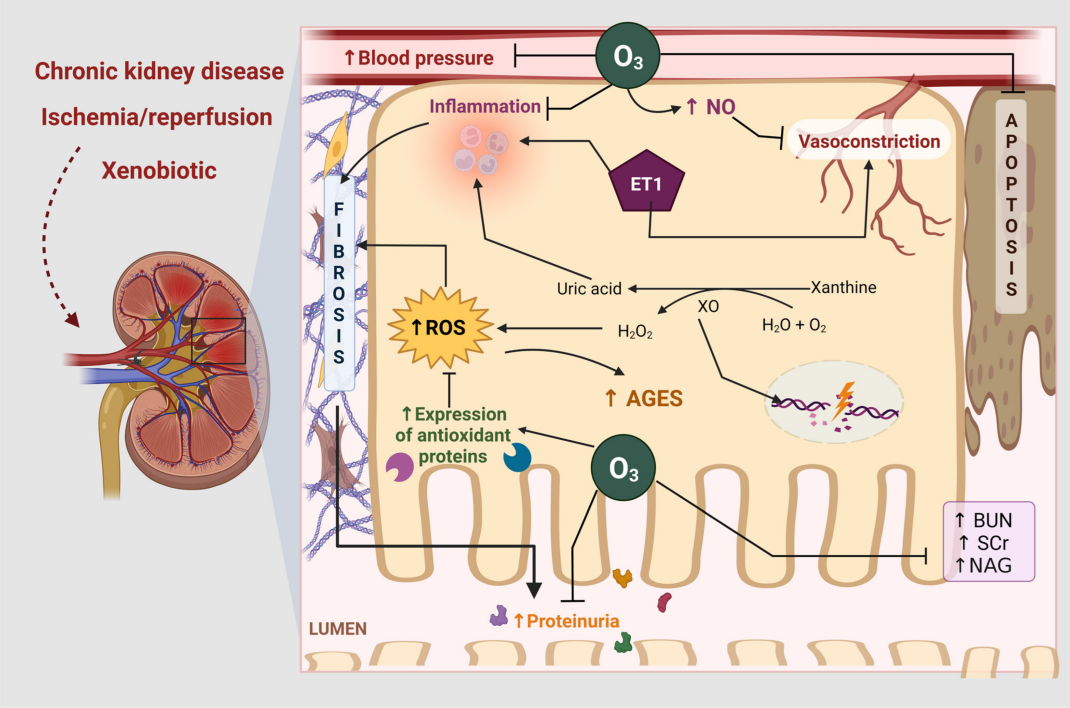

Figure 1. Effects of ozone therapy (O_3) against xenobiotics, ischemia-reperfusion (IRI) and chronic kidney disease (CKD). O_3 inhibits inflammation and ROS production by increasing the expression of antioxidant enzymes in all models. Additionally, during IRI, xanthine oxidase (XO) degrades nucleotides and forms uric acid, generating large amounts of reactive oxygen species (ROS) and inflammation. Endothelin-1 (ET-1) causes vasoconstriction and exacerbates inflammation leading to fibrosis. O_3 therapy increases nitric oxide (NO), which inhibits vasoconstriction. While O_3, by inhibiting ROS, causes a decrease in advanced glycation end products (AGES) and apoptosis, preventing CKD. H_2O: water, H_2O_2: hydrogen peroxide, O_2: oxygen molecule, NAG: N-acetyl-β-D-glucosaminidase. Created with Biorender.com, accessed on 10 February 2023.

4. Otherapeutic Uses of O_3 in Kidney

Extracorporeal shock wave lithotripsy is the first-line treatment for patients with renal calculi of under 2.0 cm; therapy fragments such stones and is highly efficient. Nevertheless, adverse effects such as hematuria might be present after the procedure [73]. Experimentally, O_3 treatment has been proven as effective against the morphological and oxidative damage caused by shock wave therapy [74]. The novel therapy, due to its antimicrobial

capacity, has also ameliorated oxidative damage caused by microorganisms in kidney infection (pyelonephritis) [63] and septic shock in kidneys [75], as well as in other organs [76].

References

1. Pérez Fernandez, R.; Martín Mateo, M.C.; De Vega, L.; Bustamante Bustamante, J.; Herrero, M.; Bustamante Munguira, E. Antioxidant enzyme determination and a study of lipid peroxidation in renal transplantation. Ren. Fail. 2002, 24, 353–359.
2. Linas, S.L.; Whittenburg, D.; Repine, J.E. Role of xanthine oxidase in ischemia/reperfusion injury. Am. J. Physiol. Physiol. 1990, 258, F711–F716.
3. Zhou, J.-Q.; Qiu, T.; Zhang, L.; Chen, Z.-B.; Wang, Z.-S.; Ma, X.-X.; Li, D. Allopurinol preconditioning attenuates renal ischemia/reperfusion injury by inhibiting HMGB1 expression in a rat model. Acta Cir. Bras. 2016, 31, 176–182.
4. Heim, C.; Glas, K. Ozone I: Characteristics/Generation/Possible Applications. Brew. Sci. 2011, 64, 8–12.
5. Liyanage, T.; Ninomiya, T.; Jha, V.; Neal, B.; Patrice, H.M.; Okpechi, I.; Zhao, M.-h.; Lv, J.; Garg, A.X.; Knight, J.; et al. World-wide access to treatment for end-stage kidney disease: A systematic review. Lancet 2015, 385, 1975–1982.
6. Xing, B.; Chen, H.; Wang, L.; Weng, X.; Chen, Z.; Li, X. Ozone oxidative preconditioning protects the rat kidney from reperfusion injury via modulation of the TLR4-NF-κB pathway. Acta Cir. Bras. 2015, 30, 60–66.
7. Foglieni, C.; Fulgenzi, A.; Belloni, D.; Sciorati, C.; Ferrero, E.; Ferrero, M.E. Ozonated autohemotherapy: Protection of kidneys from ischemia in rats subjected to unilateral nephrectomy. BMC Nephrol. 2011, 12, 61.
8. Chen, H.; Xing, B.; Liu, X.; Zhan, B.; Zhou, J.; Zhu, H.; Chen, Z. Ozone Oxidative Preconditioning Protects the Rat Kidney from Reperfusion Injury: The Role of Nitric Oxide. J. Surg. Res. 2008, 149, 287–295.
9. Wang, L.; Chen, H.; Liu, X.-H.; Chen, Z.-Y.; Weng, X.-D.; Qiu, T.; Liu, L.; Zhu, H.-C. Ozone oxidative preconditioning inhibits renal fibrosis induced by ischemia and reperfusion injury in rats. Exp. Ther. Med. 2014, 8, 1764–1768.
10. Chen, H.; Xing, B.; Liu, X.; Zhan, B.; Zhou, J.; Zhu, H.; Chen, Z. Ozone oxidative preconditioning inhibits inflammation and apoptosis in a rat model of renal ischemia/reperfusion injury. Eur. J. Pharmacol. 2008, 581, 306–314.

11. Barber, E.; Menéndez, S.; León, O.S.; Barber, M.O.; Merino, N.; Calunga, J.L.; Cruz, E.; Bocci, V. Prevention of renal injury after induction of ozone tolerance in rats submitted to warm ischaemia. Mediat. Inflamm. 1999, 8, 37–41.
12. Jiang, B.; Su, Y.; Chen, Q.; Dong, L.; Zhou, W.; Li, H.; Wang, Y. Protective Effects of Ozone Oxidative Postconditioning on Long-term Injury after Renal Ischemia/Reperfusion in Rat. Transplant. Proc. 2020, 52, 365–372.
13. Wang, L.; Chen, Z.; Liu, Y.; Du, Y.; Liu, X. Ozone oxidative postconditioning inhibits oxidative stress and apoptosis in renal ischemia and reperfusion injury through inhibition of MAPK signaling pathway. Drug Des. Dev. Ther. 2018, 12, 1293–1301.
14. Oztosun, M.; Akgul, E.O.; Cakir, E.; Cayci, T.; Uysal, B.; Ogur, R.; Ozcan, A.; Ozgurtas, T.; Guven, A.; Korkmaz, A. The Effects of Medical Ozone Therapy on Renal Ischemia/Reperfusion Injury. Ren. Fail. 2012, 34, 921–925.
15. Fernández, A.; González, L.; Calunga, J.L.; Rodríguez, S.; Santos, E. Ozone Postconditioning in Renal Ischaemia-Reperfusion Model. Functional and Morphological Evidences. Soc. Española Nefrol. 2011, 31, 379–504.
16. Calunga, J.L.; Trujillo, Y.; Zamora, Z.; Alonso, Y.; Merino, N.; Montero, T.; Menéndez, S. Ozone oxidative post-conditioning in acute renal failure. J. Pharm. Pharmacol. 2009, 61, 221–227.
17. Chen, H.; Xing, B.; Liu, X.; Zhan, B.; Zhou, J.; Zhu, H.; Chen, Z. Similarities Between Ozone Oxidative Preconditioning and Ischemic Preconditioning in Renal Ischemia/Reperfusion Injury. Arch. Med. Res. 2008, 39, 169–178.
18. Wang, L.; Chen, Z.; Weng, X.; Wang, M.; Du, Y.; Liu, X. Combined Ischemic Postconditioning and Ozone Postconditioning Provides Synergistic Protection Against Renal Ischemia and Reperfusion Injury Through Inhibiting Pyroptosis. Urology 2019, 123, 296.e1–296.e8.
19. Wang, Z.; Han, Q.; Guo, Y.-L.; Liu, X.-H.; Qiu, T. Effect of ozone oxidative preconditioning on inflammation and oxidative stress injury in rat model of renal transplantation. Acta Cir. Bras. 2018, 33, 238–249.
20. Qiu, T.; Wang, Z.-S.; Liu, X.-H.; Chen, H.; Zhou, J.-Q.; Chen, Z.-Y.; Wang, M.; Jiang, G.-J.; Wang, L.; Yu, G.; et al. Effect of ozone oxidative preconditioning on oxidative stress injury in a rat model of kidney transplantation. Exp. Ther. Med. 2017, 13, 1948–1955.
21. Wang, L.; Chen, H.; Liu, X.-H.; Chen, Z.-Y.; Weng, X.-D.; Qiu, T.; Liu, L. The protective effect of ozone oxidative preconditioning against hypoxia/reoxygenation injury in rat kidney cells. Ren. Fail. 2014, 36, 1449–1454.
22. Yu, L.; Gengaro, P.; Niederberger, M.; Burke, T.J.; Schrier, R.W. Nitric oxide: A mediator in rat tubularhypoxia/reoxygenation injury. Proc. Natl. Acad. Sci. USA 1994, 91, 1691–1695.

23. Tripatara, P.; Patel, N.S.; Webb, A.; Rathod, K.; Lecomte, F.M.; Mazzon, E.; Cuzzocrea, S.; Yaqoob, M.M.; Ahluwalia, A.; Thiemermann, C. Nitrite-Derived Nitric Oxide Protects the Rat Kidney against Ischemia/Reperfusion Injury In Vivo: Role for Xanthine Oxidoreductase. J. Am. Soc. Nephrol. 2007, 18, 570–580.
24. Juchau, M.R.; Chen, H. Developmental Enzymology. In Handbook of Developmental Neurotoxicology; Elsevier: Humana Totowa, NJ, USA, 1998; pp. 321–337.
25. Demirbag, S.; Uysal, B.; Guven, A.; Cayci, T.; Ozler, M.; Ozcan, A.; Kaldirim, U.; Surer, I.; Korkmaz, A. Effects of medical ozone therapy on acetaminophen-induced nephrotoxicity in rats. Ren. Fail. 2010, 32, 493–497.
26. Reshi, M.S.; Yadav, D.; Uthra, C.; Shrivastava, S.; Shukla, S. Acetaminophen-induced renal toxicity: Preventive effect of silver nanoparticles. Toxicol. Res. 2020, 9, 406–412.
27. Ucar, F.; Taslipinar, M.Y.; Alp, B.F.; Aydin, I.; Aydin, F.N.; Agilli, M.; Toygar, M.; Ozkan, E.; Macit, E.; Oztosun, M.; et al. The Effects of N-Acetylcysteine and Ozone Therapy on Oxidative Stress and Inflammation in Acetaminophen-Induced Nephrotoxicity Model. Ren. Fail. 2013, 35, 640–647.
28. Lewis, G.; Coughlin, L.; Jusko, W.; Hartz, S. Contribution of cigarette smoking to cadmium accumulation in man. Lancet 1972, 299, 291–292.
29. Vijayakumar, V.; Abern, M.; Jagai, J.; Kajdacsy-Balla, A. Observational Study of the Association between Air Cadmium Exposure and Prostate Cancer Aggressiveness at Diagnosis among a Nationwide Retrospective Cohort of 230,540 Patients in the United States. Int. J. Environ. Res. Public Health 2021, 18, 8333.
30. Singh, P.; Mitra, P.; Goyal, T.; Sharma, S.; Sharma, P. Blood lead and cadmium levels in occupationally exposed workers and their effect on markers of DNA damage and repair. Environ. Geochem. Health 2020, 43, 185–193.
31. Ramon, D.; Morick, D.; Croot, P.; Berzak, R.; Scheinin, A.; Tchernov, D.; Davidovich, N.; Britzi, M. A survey of arsenic, mercury, cadmium, and lead residues in seafood (fish, crustaceans, and cephalopods) from the south-eastern Mediterranean Sea. J. Food Sci. 2021, 86, 1153–1161.
32. Rani, A.; Kumar, A.; Lal, A.; Pant, M. Cellular mechanisms of cadmium-induced toxicity: A review. Int. J. Environ. Health Res. 2013, 24, 378–399.
33. Milnerowicz, H.; Śliwińska-Mossoń, M.; Sobiech, K.A. The effect of ozone on the expression of metallothionein in tissues of rats chronically exposed to cadmium. Environ. Toxicol. Pharmacol. 2017, 52, 27–37.
34. Ohta, H.; Qi, Y.; Ohba, K.; Toyooka, T.; Wang, R.-S. Role of metallothionein-like cadmium-binding protein (MTLCdBP) in the protective mechanism against cadmium toxicity in the testis. Ind. Health 2019, 57, 570–579.

35. Śliwińska-Mossoń, M.; Sobiech, K.; Dolezych, B.; Madej, P.; Milnerowicz, H. N-acetyl-beta-D-Glucosaminidase in Tissues of Rats Chronically Exposed to Cadmium and Treated with Ozone. Ann. Clin. Lab. Sci. 2019, 49, 193–203.
36. Le Hir, M.; Dubach, U.C.; Schmidt, U. Quantitative distribution of lysosomal hydrolases in the rat nephron. Histochem. 1979, 63, 245–251.
37. Liangos, O.; Perianayagam, M.C.; Vaidya, V.S.; Han, W.K.; Wald, R.; Tighiouart, H.; MacKinnon, R.W.; Li, L.; Balakrishnan, V.S.; Pereira, B.J.; et al. Urinary N-Acetyl-β-(D)-Glucosaminidase Activity and Kidney Injury Molecule-1 Level Are Associated with Adverse Outcomes in Acute Renal Failure. J. Am. Soc. Nephrol. 2007, 18, 904–912.
38. Boyer, M.J.; Tannock, I.F. Lysosomes, Lysosomal Enzymes, and Cancer. Adv. Cancer Res. 1992, 60, 269–291.
39. Łaszczyca, P.; Kawka-Serwecińska, E.; Witas, I.; Dolezych, B.; Falkus, B.; Mekail, A.; Ziółkowska, B.; Madej, P.; Migula, P. Lipid peroxidation and activity of antioxidative enzymes in the rat model of ozone therapy. Mater. Med. Pol. 1996, 28, 155–160.
40. U.S. Food and Drug Administration (FDA). ADRIAMYCIN (DOXOrubicin HCl) for Injection; FDA: Silver Spring, MD, USA, 2020.
41. Okuda, S.; Oh, Y.; Tsuruda, H.; Onoyama, K.; Fujimi, S.; Fujishima, M. Adriamycin-induced nephropathy as a model of chronic progressive glomerular disease. Kidney Int. 1986, 29, 502–510.
42. Calunga, J.; Bello, M.; Chaple, M.; Barber, E.; Menéndez, S.; Merino, N. Ozonoterapia en la glomerulonefritis tóxica experimental por adriamicina. Rev. Cuba. Invest. Biomed. 2004, 23, 139–143.
43. U.S. Food and Drug Administration (FDA). Cisplatin Injection. Available online: https://www.accessdata.fda.gov/drugsatfda_docs/label/2019/018057s089lbl.pdf (accessed on 19 October 2022).
44. Sánchez-González, P.D.; López-Hernández, F.J.; Lopez-Novoa, J.M.; Morales, A.I. An integrative view of the pathophysiological events leading to cisplatin nephrotoxicity. Crit. Rev. Toxicol. 2011, 41, 803–821.
45. Jo, S.-K.; Cho, W.Y.; Sung, S.A.; Kim, H.K.; Won, N.H. MEK inhibitor, U0126, attenuates cisplatin-induced renal injury by decreasing inflammation and apoptosis. Kidney Int. 2005, 67, 458–466.
46. Ghani, M.A.; Barril, C.; Bedgood, D.R., Jr.; Prenzler, P.D. Measurement of antioxidant activity with the thiobarbituric acid reactive substances assay. Food Chem. 2017, 230, 195–207.
47. Borrego, A.; Zamora, Z.B.; González, R.; Romay, C.; Menéndez, S.; Hernández, F.; Montero, T.; Rojas, E. Protection by ozone preconditioning is mediated by the antioxidant system in cisplatin-

induced nephrotoxicity in rats. Mediat. Inflamm. 2004, 13, 13–19.

48. Mohamed, A.A.-R.; Khater, S.I.; Metwally, M.M.; Bin Emran, T.; Nassan, M.A.; El-Emam, M.M.A.; Mostafa-Hedeab, G.; El-Shetry, E.S. TGF-β1, NAG-1, and antioxidant enzymes expression alterations in Cisplatin-induced nephrotoxicity in a rat model: Comparative modulating role of Melatonin, Vit. E and Ozone. Gene 2022, 820, 146293.
49. González, R.; Borrego, A.; Zamora, Z.; Romay, C.; Hernández, F.; Menéndez, S.; Montero, T.; Rojas, E. Reversion by ozone treatment of acute nephrotoxicity induced by cisplatin in rats. Mediat. Inflamm. 2004, 13, 307–312.
50. Kesik, V.; Uysal, B.; Kurt, B.; Kismet, E.; Koseoglu, V. Ozone ameliorates methotrexate-induced intestinal injury in rats. Cancer Biol. Ther. 2009, 8, 1623–1628.
51. Heyman, S.N.; Rosenberger, C.; Rosen, S. Regional alterations in renal haemodynamics and oxygenation: A role in contrast medium-induced nephropathy. Nephrol. Dial. Transplant. 2005, 20, i6–i11.
52. Kurtoglu, T.; Durmaz, S.; Akgullu, C.; Gungor, H.; Eryilmaz, U.; Meteoglu, I.; Karul, A.; Boga, M. Ozone preconditioning attenuates contrast-induced nephropathy in rats. J. Surg. Res. 2015, 195, 604–611.
53. Ozturk, O.; Eroglu, H.A.; Ustebay, S.; Kuzucu, M.; Adali, Y. An experimental study on the preventive effects of N-acetyl cysteine and ozone treatment against contrast-induced nephropathy. Acta Cir. Bras. 2018, 33, 508–517.
54. Mills, K.T.; Xu, Y.; Zhang, W.; Bundy, J.D.; Chen, C.-S.; Kelly, T.N.; Chen, J.; He, J. A systematic analysis of worldwide population-based data on the global burden of chronic kidney disease in 2010. Kidney Int. 2015, 88, 950–957.
55. Webster, A.C.; Nagler, E.V.; Morton, R.L.; Masson, P. Chronic Kidney Disease. Lancet 2017, 389, 1238–1252.
56. Kalantar-Zadeh, K.; Jafar, T.H.; Nitsch, D.; Neuen, B.L.; Perkovic, V. Chronic kidney disease. Lancet 2021, 398, 786–802.
57. Yu, G.; Bai, Z.; Chen, Z.; Chen, H.; Wang, G.; Wang, G.; Liu, Z. The NLRP3 inflammasome is a potential target of ozone therapy aiming to ease chronic renal inflammation in chronic kidney disease. Int. Immunopharmacol. 2017, 43, 203–209.
58. Calunga, J.L.; Zamora, Z.B.; Borrego, A.; Del Río, S.; Barber, E.; Menéndez, S.; Hernández, F.; Montero, T.; Taboada, D. Ozone Therapy on Rats Submitted to Subtotal Nephrectomy: Role of Antioxidant System. Mediat. Inflamm. 2005, 2005, 221–227.
59. Zhao, Y.-Y.; Feng, Y.-L.; Bai, X.; Tan, X.-J.; Lin, R.-C.; Mei, Q. Ultra Performance Liquid Chromatography-Based Metabonomic Study of Therapeutic Effect of the Surface Layer of Poria

cocos on Adenine-Induced Chronic Kidney Disease Provides New Insight into Anti-Fibrosis Mechanism. PLoS ONE 2013, 8, e59617.

60. Chen, Z.; Liu, X.; Yu, G.; Chen, H.; Wang, L.; Wang, Z.; Qiu, T.; Weng, X. Ozone therapy ameliorates tubulointerstitial inflammation by regulating TLR4 in adenine-induced CKD rats. Ren. Fail. 2016, 38, 822–830.
61. Yu, G.; Liu, X.; Chen, Z.; Chen, H.; Wang, L.; Wang, Z.; Qiu, T.; Weng, X. Ozone Therapy Could Attenuate Tubulointerstitial Injury in Adenine-Induced CKD Rats by Mediating Nrf2 and NF-KB. Iran J. Basic Med. Sci. 2016, 19, 1136–1143.
62. Umanath, K.; Lewis, J.B. Update on Diabetic Nephropathy: Core Curriculum 2018. Am. J. Kidney Dis. 2018, 71, 884–895.
63. Tuttle, K.R.; Bakris, G.L.; Bilous, R.W.; Chiang, J.L.; de Boer, I.H.; Goldstein-Fuchs, J.; Hirsch, I.B.; Kalantar-Zadeh, K.; Narva, A.S.; Navaneethan, S.D.; et al. Diabetic Kidney Disease: A Report from an ADA Consensus Conference. Diabetes Care 2014, 37, 2864–2883.
64. Morsy, M.D.; Hassan, W.N.; Zalat, S. Improvement of renal oxidative stress markers after ozone administration in diabetic nephropathy in rats. Diabetol. Metab. Syndr. 2010, 2, 29.
65. Güçlü, A.; Erken, H.A.; Erken, G.; Dodurga, Y.; Yay, A.; Özçoban, Ö.; Şimşek, H.; Akçılar, A.; Kocak, F.E. The effects of ozone therapy on caspase pathways, TNF-α, and HIF-1α in diabetic nephropathy. Int. Urol. Nephrol. 2015, 48, 441–450.
66. Varghese, R.T.; Jialal, I. Diabetic Nephropathy. Available online: https://www.ncbi.nlm.nih.gov/books/NBK534200/ (accessed on 9 November 2022).
67. Biedunkiewicz, B.; Tylicki, L.; Lichodziejewska-Niemierko, M.; Liberek, T.; Rutkowski, B. Ozonetherapy in a dialyzed patient with calcific uremic arteriolopathy. Kidney Int. 2003, 64, 367–368.
68. Di Paolo, N.; Bocci, V.; Cappelletti, F.; Petrini, G.; Gaggiotti, E. Necrotizing Fasciitis Successfully Treated with Extracorporeal Blood Oxygenation and Ozonization (EBOO). Int. J. Artif. Organs 2002, 25, 1194–1198.
69. Sancak, E.B.; Turkön, H.; Çukur, S.; Erimsah, S.; Akbas, A.; Gulpinar, M.T.; Toman, H.; Sahin, H.; Uzun, M. Major Ozonated Autohemotherapy Preconditioning Ameliorates Kidney Ischemia-Reperfusion Injury. Inflammation 2016, 39, 209–217.
70. Biedunkiewicz, B.; Tylicki, L.; Rachon, D.; Hak, L.; Nieweglowski, T.; Chamienia, A.; Debska-Slizien, A.; Mysliwska, J.; Rutkowski, B. Natural Killer Cell Activity Unaffected by Ozonated Autohemotherapy in Patients with End-Stage Renal Disease on Maintenance Renal Replacement Therapy. Int. J. Artif. Organs 2004, 27, 766–771.

71. Tang, W.-J.; Jiang, L.; Wang, Y.; Kuang, Z.-M. Ozone therapy induced sinus arrest in a hypertensive patient with chronic kidney disease. Medicine 2017, 96, e9265.
72. Gu, X.-B.; Yang, X.-J.; Zhu, H.-Y.; Xu, Y.-Q.; Liu, X.-Y. Effect of medical ozone therapy on renal blood flow and renal function of patients with chronic severe hepatitis. Chin. Med. J. 2010, 123, 2510–2513.
73. Torricelli, F.C.M.; Danilovic, A.; Vicentini, F.; Marchini, G.S.; Srougi, M.; Mazzucchi, E. Extracorporeal shock wave lithotripsy in the treatment of renal and ureteral stones. Rev. Assoc. Med. Bras. 2015, 61, 65–71.
74. Uğuz, S.; Demirer, Z.; Uysal, B.; Alp, B.F.; Malkoc, E.; Guragac, A.; Turker, T.; Ateş, F.; Karademir, K.; Ozcan, A.; et al. Medical ozone therapy reduces shock wave therapy-induced renal injury. Ren. Fail. 2016, 38, 974–981.
75. Madej, P.; Plewka, A.; Madej, J.A.; Nowak, M.; Plewka, D.; Franik, G.; Golka, D. Ozonotherapy in an Induced Septic Shock. I. Effect of Ozonotherapy on Rat Organs in Evaluation of Free Radical Reactions and Selected Enzymatic Systems. Inflammation 2007, 30, 52–58.
76. Madej, P.; Plewka, A.; Madej, J.A.; Plewka, D.; Mroczka, W.; Wilk, K.; Dobrosz, Z. Ozone Therapy in Induced Endotoxemic Shock. II. The Effect of Ozone Therapy Upon Selected Histochemical Reactions in Organs of Rats in Endotoxemic Shock. Inflammation 2007, 30, 69–86.