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(54) Title: THIOREDOXIN MIMETIC PRODRUGS AND USES THEREOF

(57) Abstract: The present invention provides thioredoxin (TRX) mimetic prodrugs, more particularly, derivatives or analogues of the amino acid sequence Cys-Pro-Cys or A_1 -Cys- A_2 -Pro-A_3 -Cys-A_4 wherein A_1 to A_4 each independently is either absent or an amino acid residue, as well as pharmaceutical compositions comprising them. These compounds, upon hydrolysis under physiological conditions, are converted into the native Cys-Pro-Cys or A_1-Cys-A_2 -Pro-A_3-Cys-A_4 sequence, and are thus useful for prevention, treatment or management of diseases, disorders or conditions mediated by redox stress.

THIOREDOXIN MIMETIC PRODRUGS AND USES THEREOF

TECHNICAL FIELD

[0001] The present invention relates to thioredoxin (TRX) mimetic prodrugs, pharmaceutical compositions comprising them, and uses thereof.

[0002] **Abbreviations**: **ALT**, alanine transaminase; **AST**, aspartate transaminase; **BAL**, bronchoalveolar lavage fluid; **BUN**, blood urea nitrogen; **CILI**, chlorine (Cl_2) inhalational lung injury; **CIN**, contrast media-induced nephropathy; **FiO_2**, fraction of inspired oxygen; **IMV**, intermittent mandatory ventilation; **IP**, intraperitoneally; **I/R**, ischemia-reperfusion; **IV**, intravenously; **LAD**, left anterior descending; **LCMS**, Liquid chromatography-mass spectrometry; **LPS**, lipopolysaccharide; **MDA**, malondialdehyde; **MIRI**, myocardial ischemia-reperfusion injury; **MPO**, myeloperoxidase; **NAC**, N-acetylcysteine; **PaO_2**, partial pressure of oxygen; **PBS**, phosphate-buffered saline; **PCO_2**, partial pressure of carbon dioxide; **PMN**, polymorphonuclear leukocytes; **PO_2**, oxygen partial pressure; **RIRI**, renal ischemia reperfusion injury; **ROS**, reactive oxygen species; **RT**, room temperature; **TV**, tidal volume; **TRX**, thioredoxin; **VILI**, ventilator-induced lung injury.

BACKGROUND ART

[0003] 1-(2-acetamido-3-mercaptopropanoyl)-N-(1-amino-3-mercapto-1-oxopropan-2-yl)pyrrolidine-2-carboxamide, 2-(1-(2-amino-3-mercaptopropanoyl) pyrrolidine-2-carboxamido)-3-mercaptopropanoic acid, and analogues thereof are TRX mimetics, thiol-rich tripeptide containing cysteine-proline-cysteine (Cys-Pro-Cys) or analogues, which are closely analogous to the native TRX motif. TRX is a multifunctional redox-active protein that scavenges ROS by itself or together with TRX-dependent peroxiredoxin, and is a critical element in the defense against redox stress. TRX also has chemotaxis-modulating functions and suppresses PMN infiltration into sites of inflammation (Hoshino *et al.*, 2003). The redox stress of acute lung injury is initially countered by endogenous reductants, especially TRX, but such thiol-rich reductant defenses are readily overwhelmed by massive oxidant insults. The subsequent depletion of TRX increases susceptibility to acute lung injury, as noted in models of hyperoxic lung injury (Tipple *et al.*, 2007). 1-(2-acetamido-3-mercaptopropanoyl)-N-(1-amino-3-mercapto-1-oxopropan-2-yl)pyrrolidine-2-carboxamide has been shown to be efficacious in a murine model of asthma induced by

sensitization and challenge with ovalbumin, but has not been recognized as having efficacy in halogen induced lung inhalation injury.

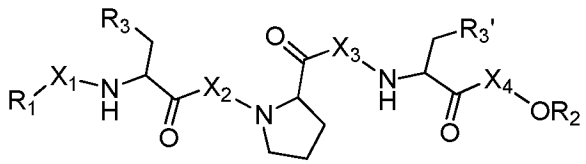
[0004] US 8,530,407 discloses antioxidant compounds of the general formula A-Y_1-Cys-Y_2-Cys-Y_3-B, wherein Cys is a cysteine residue, A and B are each individually a hydrophobic or non-charged moiety; and Y_1, Y_2 and Y_3 are each individually one or more amino acid residues in the range of 0-30 residues, with the provision that Y_1, Y_2 and Y_3 collectively provide for at least two amino acid residues in the peptide.

[0005] International Publication No. WO 2013/190497, herewith incorporated by reference in its entirety as if fully described herein, discloses *inter alia* particular TRX mimetics closely analogous to the native TRX motif, and shows that those compounds are useful in treatment of an inflammatory disease of the lung caused by inhalation of a toxic agent or an irritant, e.g., CILI.

SUMMARY OF INVENTION

[0006] In one aspect, the present invention provides a compound of the formula I:

I



or a pharmaceutically acceptable salt or solvate thereof,

wherein

X_1 is absent or an amino acid residue forming a peptide bond with the -NH- group adjacent thereto;

X_2 is absent or an amino acid residue forming peptide bonds with the carbonyl group and nitrogen atom adjacent thereto;

X_3 is absent or an amino acid residue forming peptide bonds with the carbonyl and -NH- groups adjacent thereto;

X_4 is absent or an amino acid residue forming a peptide bond with the carbonyl group adjacent thereto;

R_1 is H, -CO(C_1-C_8)alkyl, -CO(C_1-C_8)alkylene-NH((C_1-C_8)alkyl, -CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$, covalently linked to the N atom of the amino moiety of X_1, when present;

R_2 is H or (C_1-C_8)alkyl, covalently linked to the carbonyl moiety of X_4, when present; and

R_3 and R_3' each independently is -SH, -S-CO(C_1-C_8)alkyl, -S-CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -S-CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$; or R_3 and R_3' are both -S- and together form a disulfide bond; or one of R_3 and R_3' is -S- forming a disulfide bond with one of R_3 and R_3' of another identical or different compound of the formula I, and another of R_3 and R_3' is -SH, -S-CO(C_1-C_8)alkyl, -S-CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -S-CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$; or R_3 and R_3' are both -S-, wherein R_3 forms a disulfide bond with one of R_3 and R_3' of another identical or different compound of the formula I, and R_3' forms a disulfide bond with either another of R_3 and R_3' of said another compound of the formula I, or with one of R_3 and R_3' of a further identical or different compound of the formula I,

provided that at least one of R_1 and R_2 is not H, or at least one of R_3 and R_3' is not -SH, but excluding the compounds wherein R_3 and R_3' are each -SH, R_1 is -CO(C_1-C_8)alkyl, R_2 is H, and X_1, X_2, X_3 and X_4 are absent.

[0007] In another aspect, the present invention provides a pharmaceutical composition comprising a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, and a pharmaceutically acceptable carrier. The compounds and pharmaceutical compositions of the invention are useful for prevention, treatment or management of a disease, disorder or condition mediated by redox stress.

[0008] In a further aspect, the present invention thus relates to a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, for use in prevention, treatment or management of a disease, disorder or condition mediated by redox stress.

[0009] In yet another aspect, the present invention relates to use of a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, for the preparation of a pharmaceutical composition for prevention, treatment or management of a disease, disorder or condition mediated by redox stress.

[0010] In still another aspect, the present invention relates to a method for prevention, treatment or management of a disease, disorder or condition mediated by redox stress in an individual in need thereof, comprising administering to said individual an effective amount of a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof.

BRIEF DESCRIPTION OF DRAWINGS

[0011] **Fig. 1** shows the number of living mice at each time post-NaSH injection. Mice were injected IP with 30 mg/kg NaSH (LD_{70}). 1 hour post NaSH injection, the treated group was injected with R-911 (130 mg/kg), whereas the control group was injected with vehicle.

[0012] **Figs. 2A-2B** show the histological injury scoring of lungs of different groups post NaSH injection: Sham (n=3), Vehicle (n=6) and R-911 (n=8) (**2A**); and representative photos showing H&E stainings (×20) of histological samples that were used to generate the injury scoring data (**2B**). P value for difference between vehicle and R-911 groups was found to be highly significant ($p<10^{-7}$).

[0013] **Fig 3** shows the effect of R-911 in a lung radiation (RAD)-induced injury model. C57BL/6 male mice were exposed to 14 Gy radiation to the thorax. Five minutes before exposure, R-911 (125 mg/kg) or vehicle (RAD only group) was delivered IP. A second delivery of R-911 or vehicle was given 4 hours later on the day of radiation, and treatments continued twice daily for the next 6 days. Apoptosis was assessed by TUNEL staining on Day 7 post-radiation and the quantitative assessment of percent of apoptotic cells per lung section is shown. Un-irradiated mice (No RAD group) are shown as a negative control. * indicates significant difference by one-way ANOVA followed by Tukey's multiple comparison test comparing the indicated columns; overall ANOVA $p<0.0001$.

[0014] **Fig. 4** shows the prophylactic *vs.* therapeutic effect of R-911 in a lung radiation (RAD)-induced injury model. C57BL/6 male mice were exposed to 14 Gy radiation to the thorax. Either five minutes before exposure (pre) or 24 hours after exposure (post), R-911 (125 mg/kg) or vehicle (RAD only group) was delivered IP. A second delivery of R-911 or vehicle was given 4 hours later, and treatments continued twice daily for the next 5-6 days. Apoptosis was assessed by TUNEL staining on Day 7 post-radiation and the quantitative assessment of percent of apoptotic cells per lung section is shown. Un-irradiated mice (No RAD group) are shown as a negative control. * indicates significant difference by one-way ANOVA followed by Tukey's multiple comparison test comparing the indicated columns; overall ANOVA $p<0.0003$.

[0015] **Fig. 5** shows that the reduction in lung oxygenation is ameliorated in R-911 treated group. The significant elevation of PaO_2 in R-911 treated group indicates enhanced lung oxygenation.

[0016] **Figs. 6A-6B** show that R-911 resuscitated lung I/R-induced histological injury. Histlogical injury score (**6A**) and H&E stained histological images (**6B**) demonstrate the protective effect of R-911 in restoring lung tissue architecture.

[0017] **Figs. 7A-7B** show that R-911 significantly lessens inflammation induced by I/R injury. MPO (**7A**) and MDA (**7B**) were determined in wet lung tissue. Both markers were increased in treatment groups, and significantly reduced upon R-911 administration.

[0018] **Fig. 8** shows that R-911 significantly reduces protein exacerbation. BAL fluid content revealed an augmentation of proteins in treatment group, a sign of alveolar epithelial injury, in contrast to significant decrease of proteins in the R-911 group.

[0019] **Fig. 9** shows that R-911 reduces edema induced by lung I/R injury. Wet/dry ratio was determined in wet lung tissue, portraying a significant decrease in pulmonary edema when treated with R-911.

[0020] **Figs. 10A-10B** show that R-911 significantly attenuated CIN-induced BUN (**10A**) and creatinine (**10B**) elevations in CIN model in rats treated with R-911 or NAC (equimolar dose to 100 mg/kg/day of R-911). Sham rats are compared to CIN rats treated with a vehicle control or with 3 dose levels of R-911 (10, 30 and 100 mg/kg/day). P values: *<0.05, **<0.0002, ***<0.005, ****$\leq 10^{-5}$.

[0021] **Figs. 11A-11B** show that R-911 significantly restored tissue architecture in the kidney of contrast media-treated rats. **11A** shows representative H&E staining (×20) of histological samples that were used to generate the injury scoring data. R-911 treatment dramatically decreased the histological injury score and restored normal kidney tissue architecture. **11B** shows the histological injury scoring of kidneys of different groups post contrast media injection. P value for difference between vehicle and 100 mg/kg/day R-911 group, as well as the NAC group was found to be highly significant ($p<0.0008$).

[0022] **Figs. 12A-12B** show that R-911 significantly attenuated CIN-induced histological injury in a diabetic model. Representative histological kidney sections (**12A**, all slides are ×10) indicate that CIN caused severe damage to the kidney tissue. R-911 (300 mg/kg/day) restored the tissue architecture, which can be visualized by a reduction of 35% in the histological injury score (**12B**, $p<0.001$). An equimolar dose of NAC (94.5 mg/kg/day) was shown to be less effective when compared to contrast media vehicle groups ($p<0.05$).

[0023] **Figs. 13A-13B** show that R-911 significantly attenuated CIN-induced elevations in creatinine and BUN. CIN caused severe damage to the kidney tissue as BUN (**13A**) and creatinine (**13B**). Increasing volume did not cause any significant advantage when

administered with 300 mg/kg/day of R-911. R-911, given alone, decreased elevations of BUN by 19% and elevations of creatinine by 40% ($p<0.001$).

[0024] **Figs. 14A-14B** show that R-911 significantly restored tissue architecture in the lung in mouse lungs exposed to chlorine. **14A** shows representative H&E staining (×20) of histological samples that were used to generate the injury scoring data (**14B**). While Cl_2 injury elevated the histology score, R-911 treatment dramatically decreased this impacted histological injury score and restored normal lung tissue architecture.

[0025] **Fig. 15** shows that R-911 significantly restored VILI-induced decrease in PaO_2.

[0026] **Figs. 16A-16B** show that R-911 significantly restored VILI-induced histological damage, as seen in the representative images (**16A**). Remarkable injury is seen in the vehicle group, including edema, infiltration of neutrophils, and destruction of tissue architecture. The condition of all samples was scored (**16B**), and it was determined that R-911 reduced injury by 37.74% (right, *=$p<0.0001$).

[0027] **Fig. 17** shows that R-911 significantly reduced VILI-induced infiltrating neutrophils relative to VILI increased MPO levels, diminishing MPO levels by 29% (*=$p<10^{-6}$).

[0028] **Figs. 18A-18B** show that R-911 significantly attenuated RIRI-induced creatinine elevations. R-911 treatment groups dramatically reduced, in a dose dependent manner, creatinine elevations seen at 6 (**18A**) and 24 (**18B**) hours post reperfusion. P values in the highest dose of 300 mg/kg: ($p<0.003$, 6 hours; and $p<3.6\times10^{-5}$, 24 hours).

[0029] **Figs. 19A-19B** show that R-911 significantly attenuated RIRI-induced histological damage and restored histological tissue architecture in a dose dependent fashion. Representative slides are shown in **19A**, and the SEM values of histological injury scoring of all kidneys from the indicated treatment groups are shown in **19B**.

[0030] **Figs. 20A-20C** show that R-911 significantly decreases MIRI-induced augmentation of infarct size (**20A**), neutrophil infiltration (**20B**, $p<0.01$), and lipid peroxidation (**20C**, $p<0.0001$).

[0031] **Figs. 21A-21B** show that R-911 significantly reduces histological injury and restores cardiac tissue architecture as can be visualized in the representative slides (**21A**) and in **21B**, presenting the SEM histological injury scores indicating a dramatic attenuation of injury by R-911 (40%, $p<0.0003$ in a 2-tailed t-test).

DETAILED DESCRIPTION OF THE INVENTION

[0032] In one aspect, the present invention provides a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof. These compounds are, in fact, TRX mimetic prodrugs, i.e., derivatives or analogues of the amino acid sequence Cys-Pro-Cys or A_1-Cys-A_2-Pro-A_3-Cys-A_4 wherein A_1 to A_4 each independently is either absent or an amino acid residue, which upon hydrolysis under physiological conditions are converted into the native Cys-Pro-Cys or A_1-Cys-A_2-Pro-A_3-Cys-A_4 sequence, and are thus expected to be effective in all those clinical indications wherein administration of the native TRX motif is beneficial.

[0033] In one particular such aspect, the TRX mimetic prodrug of the present invention is a derivative or analogue of the amino acid sequence Cys-Pro-Cys or A_1-Cys-A_2-Pro-A_3-Cys-A_4 in which the thiol group of at least one of the Cys residues is protected by a group independently selected from -CO(C_1-C_8)alkyl, -CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$; and/or the terminal amino group is protected by a group selected from -CO(C_1-C_8)alkyl, -CO(C_1-C_8)alkylene-NH((C_1-C_8)alkyl, -CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$; and/or the terminal carboxyl group is protected by a group selected from (C_1-C_8)alkyl, but excluding the compound wherein both the thiol groups and the terminal carboxyl group are not protected; the terminal amino group is protected by a group selected from -CO(C_1-C_8)alkyl; and X_1 to X_4 are absent.

[0034] In another particular such aspect, the TRX mimetic prodrug of the invention is a derivative or analogue of the amino acid sequence Cys-Pro-Cys or A_1-Cys-A_2-Pro-A_3-Cys-A_4 in which at least one of the terminal amino group and terminal carboxyl group is protected as defined above, and/or the thiol group of each one of the Cys residues is deprotonated and the two sulfur atoms together form a disulfide bond.

[0035] In a further particular such aspect, the TRX mimetic prodrug of the invention is a derivative or analogue of the amino acid sequence Cys-Pro-Cys or A_1-Cys-A_2-Pro-A_3-Cys-A_4 in which at least one of the terminal amino group and terminal carboxyl group is protected as defined above, and/or the thiol group of at least one of the Cys residues is deprotonated wherein the sulfur atom forms a disulfide bond with the sulfur atom of a deprotonated thiol group of another identical or different compound of the formula I, and the thiol group of the other Cys residue is optionally protected as defined above. Conceptually, a compound according to this particular aspect is thus a dimer- or higher

multimer-like compound, in which two or more identical or different entities, each independently according to the formula I, are linked via disulfide bonds, wherein each disulfide bond links two entities.

[0036] In one particular embodiment, the dimer-like compound of the present invention results from a linkage between two compounds of the formula I, wherein only one of the thiol groups of each one of the compounds is deprotonated and the sulfur atom of the deprotonated thiol group of one of the compounds forms a disulfide bond with the sulfur atom of the deprotonated thiol group of the other compound. In another particular embodiment, the dimer-like compound of the present invention is a cyclic dimer resulting from a linkage between two compounds of the formula I, wherein the thiol group of each one of the Cys residues in each one of the compounds is deprotonated and each one of the sulfur atoms of the deprotonated thiol groups of one of the compounds forms a disulfide bond with a different one of the sulfur atoms of the deprotonated thiol groups of the other compound. In a further particular embodiment, the compound of the present invention is a higher multimer-like compound resulting from a linkage between more than two compounds of the formula I, wherein the thiol group of each one of the Cys residues in at least one of those compounds is deprotonated, and the sulfur atom of each one of the deprotonated thiol groups forms a disulfide bond with the sulfur atom of a deprotonated thiol group of a different compound.

[0037] Irrespective of the particular aspect, the cysteine and proline residues or residue derivatives constituting the TRX mimetic prodrug of the present invention may independently be of either L or D configuration, indicating the optical activity of the isomer of glyceraldehyde from which that amino acid can, in theory, be synthesized. In an alternative fashion, the (S) and (R) designators may be used to indicate the absolute stereochemistry of the compound, wherein proline naturally being (S) at the α carbon, and cysteine, having a sulfur atom at the second position in its side chain, naturally being (R).

[0038] The term "alkyl" as used herein typically means a linear or branched saturated hydrocarbon radical having 1-8 carbon atoms and includes, e.g., methyl, ethyl, n-propyl, isopropyl, n-butyl, sec-butyl, isobutyl, tert-butyl, n-pentyl, 2,2-dimethylpropyl, n-hexyl, n-heptyl, n-octyl, and the like. Preferred are (C_1-C_6)alkyl groups, more preferably (C_1-C_4)alkyl groups, most preferably methyl and ethyl.

[0039] The term "alkylene" typically means a divalent linear or branched hydrocarbon radical having 1-8 carbon atoms and includes, e.g., methylene, ethylene, propylene,

butylene, 2-methylpropylene, pentylene, 2-methylbutylene, hexylene, 2-methylpentylene, 3-methylpentylene, 2,3-dimethylbutylene, heptylene, octylene, and the like. Preferred are $(C_1$-$C_6)$alkylene, more preferably $(C_1$-$C_4)$alkylene, most preferably $(C_1$-$C_2)$alkylene.

[0040] The term "amino acid" as used herein refers to an organic compound comprising both amine and carboxylic acid functional groups, which may be either a natural or non-natural amino acid and may be of either L or D configuration. The twenty two natural amino acids are aspartic acid (Asp), tyrosine (Tyr), leucine (Leu), tryptophan (Trp), arginine (Arg), valine (Val), glutamic acid (Glu), methionine (Met), phenylalanine (Phe), serine (Ser), alanine (Ala), glutamine (Gln), glycine (Gly), proline (Pro), threonine (Thr), asparagine (Asn), lysine (Lys), histidine (His), isoleucine (Ile), cysteine (Cys), selenocysteine (Sec), and pyrrolysine (Pyl). Non-limiting examples of non-natural amino acids include diaminopropionic acid (Dap), diaminobutyric acid (Dab), ornithine (Orn), aminoadipic acid, β-alanine, 1-naphthylalanine, 3-(1-naphthyl)alanine, 3-(2-naphthyl)alanine, γ-aminobutiric acid (GABA), 3-(aminomethyl) benzoic acid, p-ethynyl-phenylalanine, p-propargly-oxy-phenylalanine, m-ethynyl-phenylalanine, p-bromophenylalanine, p-iodophenylalanine, p-azidophenylalanine, p-acetylphenylalanine, azidonorleucine, 6-ethynyl-tryptophan, 5-ethynyl-tryptophan, 3-(6-chloroindolyl)alanine, 3-(6-bromoindolyl)alanine, 3-(5-bromoindolyl)alanine, azidohomoalanine, p-chlorophenylalanine, α-aminocaprylic acid, O-methyl-L-tyrosine, N-acetylgalactosamine-α-threonine, and N-acetylgalactosamine-α-serine.

[0041] The term "amino acid residue" as used herein with respect to the groups X_1 to X_4 refers to a residue of an amino acid after removal of hydrogen atom from an amino group thereof, e.g., its α-amino group or side chain amino group if present, and -OH group from a carboxyl group thereof, e.g., its α-carboxyl group or side chain carboxyl group if present.

[0042] The term "peptide bond" or "amide bond" as used herein refers to the covalent bond -C(O)NH- formed between two molecules, e.g., two amino acids, when a carboxyl group of one of the molecules reacts with an amino group of the other molecule, causing the release of a molecule of water.

[0043] In certain embodiments, the compound of the present invention is a compound of the formula I, wherein R_1 is H, -CO$(C_1$-$C_4)$alkyl, preferably -CO$(C_1$-$C_2)$alkyl, -CO$(C_1$-$C_4)$alkylene-NH$((C_1$-$C_4)$alkyl, preferably -CO$(C_1$-$C_2)$alkylene-NH$((C_1$-$C_2)$alkyl, -CO$(C_1$-$C_4)$alkylene-N$((C_1$-$C_4)$alkyl$)_2$, preferably -CO$(C_1$-$C_2)$alkylene-N$((C_1$-$C_2)$alkyl$)_2$, or -

$CO(C_1\text{-}C_4)alkylene\text{-}N^+((C_1\text{-}C_4)alkyl)_3$, preferably $-CO(C_1\text{-}C_2)alkylene\text{-}N^+((C_1\text{-}C_2)alkyl)_3$. In particular such embodiments, R_1 is H, $-CO\text{-}CH_2\text{-}N(CH_3)_2$, or $-CO\text{-}CH_2\text{-}N^+(CH_3)_3$.

[0044] In certain embodiments, the compound of the present invention is a compound of the formula I, wherein R_2 is $(C_1\text{-}C_4)alkyl$, preferably $(C_1\text{-}C_2)alkyl$, or H. Particular such embodiments are those wherein R_2 is methyl, ethyl or H.

[0045] In certain embodiments, the compound of the present invention is a compound of the formula I, wherein R_3 and R_3' each independently is -SH, $-S\text{-}CO(C_1\text{-}C_4)alkyl$, preferably $-S\text{-}CO(C_1\text{-}C_2)alkyl$, $-S\text{-}CO(C_1\text{-}C_4)alkylene\text{-}N((C_1\text{-}C_4)alkyl)_2$, preferably $-S\text{-}CO(C_1\text{-}C_2)alkylene\text{-}N((C_1\text{-}C_2)alkyl)_2$, or $-S\text{-}CO(C_1\text{-}C_4)alkylene\text{-}N^+((C_1\text{-}C_4)alkyl)_3$, preferably $-S\text{-}CO(C_1\text{-}C_2)alkylene\text{-}N^+((C_1\text{-}C_2)alkyl)_3$; or R_3 and R_3' are both -S- and together form a disulfide bond. Particular such embodiments are those wherein R_3 and R_3' each independently is -SH, $-S\text{-}COCH_3$, $-S\text{-}CO\text{-}CH_2\text{-}N(CH_3)_2$, or $-S\text{-}CO\text{-}CH_2\text{-}N^+(CH_3)_3$; or R_3 and R_3' are both -S- and together form a disulfide bond.

[0046] In certain embodiments, the compound of the present invention is a dimer- or higher multimer-like compound as defined above, i.e., a compound of the formula I, wherein one of R_3 and R_3' is -S- forming a disulfide bond with one of R_3 and R_3' of another identical or different compound of the formula I, and another of R_3 and R_3' is -SH, $-S\text{-}CO(C_1\text{-}C_4)alkyl$, preferably $-S\text{-}CO(C_1\text{-}C_2)alkyl$, $-S\text{-}CO(C_1\text{-}C_4)alkylene\text{-}N((C_1\text{-}C_4)alkyl)_2$, preferably $-S\text{-}CO(C_1\text{-}C_2)alkylene\text{-}N((C_1\text{-}C_2)alkyl)_2$, or $-S\text{-}CO(C_1\text{-}C_4)alkylene\text{-}N^+((C_1\text{-}C_4)alkyl)_3$, preferably $-S\text{-}CO(C_1\text{-}C_2)alkylene\text{-}N^+((C_1\text{-}C_2)alkyl)_3$; or R_3 and R_3' are both -S-, wherein R_3 forms a disulfide bond with one of R_3 and R_3' of another identical or different compound of the formula I, and R_3' forms a disulfide bond with either another of R_3 and R_3' of said another compound of the formula I, or with one of R_3 and R_3' of a further identical or different compound of the formula I. In particular such embodiments, one of R_3 and R_3' is -S-, and another of R_3 and R_3' is -SH, $-S\text{-}COCH_3$, $-S\text{-}CO\text{-}CH_2\text{-}N(CH_3)_2$, or $-S\text{-}CO\text{-}CH_2\text{-}N^+(CH_3)_3$; or R_3 and R_3' are both -S-.

[0047] In certain embodiments, the compound of the present invention is a compound of the formula I, wherein X_1, X_2, X_3 and X_4 are absent, or at least one of X_1, X_2, X_3 and X_4 is present. Particular such embodiments are those wherein (i) one of X_1, X_2, X_3 and X_4 is an amino acid residue as defined above, and the other three of X_1, X_2, X_3 and X_4 are absent, e.g., wherein X_1, X_2 and X_3 are absent, and X_4 is a glycine residue; X_1, X_2 and X_4 are absent, and X_3 is a glycine residue; X_1, X_3 and X_4 are absent, and X_2 is a glycine residue; or X_2, X_3 and X_4 are absent, and X_1 is a glycine residue; (ii) two of X_1, X_2, X_3 and X_4 are

amino acid residues each independently as defined above, and the other two of X_1, X_2, X_3 and X_4 are absent, e.g., wherein X_1 and X_2 are absent, and X_3 and X_4 each is a glycine residue; X_1 and X_3 are absent, and X_2 and X_4 each is a glycine residue; X_1 and X_4 are absent, and X_2 and X_3 each is a glycine residue; X_2 and X_3 are absent, and X_1 and X_4 each is a glycine residue; X_2 and X_4 are absent, and X_1 and X_3 each is a glycine residue; X_3 and X_4 are absent, and X_1 and X_2 each is a glycine residue; (iii) three of X_1, X_2, X_3 and X_4 are amino acid residues each independently as defined above, and the other one of X_1, X_2, X_3 and X_4 is absent, e.g., wherein X_1 is absent, and X_2, X_3 and X_4 each is a glycine residue; X_2 is absent, and X_1, X_3 and X_4 each is a glycine residue; X_3 is absent, and X_1, X_2 and X_4 each is a glycine residue; X_4 is absent, and X_1, X_2 and X_3 each is a glycine residue; or (iv) the four of X_1, X_2, X_3 and X_4 are amino acid residues each independently as defined above, e.g., wherein X_1, X_2, X_3 and X_4 each is a glycine residue.

[0048] In certain particular embodiments, the compound of the present invention is a compound of the formula I according to some of the embodiments defined above, wherein R_1 is H, -CO(C_1-C_4)alkyl, preferably -CO(C_1-C_2)alkyl, -CO(C_1-C_4)alkylene-NH((C_1-C_4)alkyl, preferably -CO(C_1-C_2)alkylene-NH((C_1-C_2)alkyl, -CO(C_1-C_4)alkylene-N((C_1-C_4)alkyl)$_2$, preferably -CO(C_1-C_2)alkylene-N((C_1-C_2)alkyl)$_2$, or -CO(C_1-C_4)alkylene-N^+((C_1-C_4)alkyl)$_3$, preferably -CO(C_1-C_2)alkylene-N^+((C_1-C_2)alkyl)$_3$; R_2 is (C_1-C_4)alkyl, preferably (C_1-C_2)alkyl, or H; R_3 and R_3' each independently is -SH, -S-CO(C_1-C_4)alkyl, preferably -S-CO(C_1-C_2)alkyl, -S-CO(C_1-C_4)alkylene-N((C_1-C_4)alkyl)$_2$, preferably -S-CO(C_1-C_2)alkylene-N((C_1-C_2)alkyl)$_2$, or -S-CO(C_1-C_4)alkylene-N^+((C_1-C_4)alkyl)$_3$, preferably -S-CO(C_1-C_2)alkylene-N^+((C_1-C_2)alkyl)$_3$, or R_3 and R_3' are both -S- and together form a disulfide bond; and X_1, X_2, X_3 and X_4 are absent, or at least one of X_1, X_2, X_3 and X_4 is present. In more particular such embodiments, R_1 is H, -CO-CH_2-N(CH_3)$_2$, or -CO-CH_2-N^+(CH_3)$_3$; R_2 is H, methyl, or ethyl; and R_3 and R_3' each independently is -SH, -S-$COCH_3$, -S-CO-CH_2-N(CH_3)$_2$, or -S-CO-CH_2-N^+(CH_3)$_3$, or R_3 and R_3' are both -S- and together form a disulfide bond, wherein each one of the combinations of R_1, R_2, R_3 and R_3' constitutes a separate embodiment.

[0049] Specific compounds of the formula I described herein, wherein X_1, X_2, X_3 and X_4 are absent, are herein identified by the Arabic numbers **1-9** in bold, corresponding to PEPTIDE ID No. 1-9, respectively (**Table 1**), and their full chemical structures are shown in **Table 2**. Compound **1** is also identified R-951; compound **2** is also identified R-956; compound **3** is also identified R-952; compound **4** is also identified R-908; compound **5** is

also identified R-911; compound **6** is also identified R-958; compound **7** is also identified R-909; compound **8** is also identified R-910; and compound **9** is also identified R-953. Other specific compounds of the formula I, wherein X_2 is a glycine residue and X_1, X_3 and X_4 are absent, are herein identified by the Arabic numbers **10-18** in bold, corresponding to PEPTIDE ID No. 10-18 (also referred to as SEQ ID No. 1-9), respectively (**Table 1**), and their full chemical structures are depicted in **Table 3**.

Table 1: Sequence description of compounds/PEPTIDE ID No. **1-18**

PEPTIDE ID No.	Amino acid sequence
1	Cys-Pro-(Cys-ethyl ester)
2	(dimethylaminomethylcarbonyl-Cys)-Pro-Cys
3	(dimethylaminomethylcarbonyl-Cys)-Pro-(Cys-ethyl ester)
4	(S-acetyl Cys)-Pro-(S-acetyl Cys)
5	(dimethylaminomethylcarbonyl-S-acetyl Cys)-Pro-(Cys-ethyl ester, S-acetyl)
6	(dimethylaminomethylcarbonyl-S-acetyl Cys)-Pro-(S-acetyl Cys)
7	Dimethylaminomethylcarbonyl-Cys, S-dimethylaminomethylcarbonyl)-Pro-(Cys-ethyl ester, S-dimethylaminomethylcarbonyl)
8	(S-trimethylammoniomethylcarbonyl-Cys, trimethylammoniomethylcarbonyl)-Pro-(Cys ethyl ester, S-trimethylammoniomethylcarbonyl)
9	(dimethylaminomethylcarbonyl-Cys)-Pro-(Cys-ethyl ester)
10	Cys-Gly-Pro-(Cys ethyl ester)
11	(dimethylaminomethylcarbonyl-Cys)-Gly-Pro-Cys
12	(dimethylaminomethylcarbonyl-Cys)-Gly-Pro-(Cys-ethyl ester)
13	(S-acetyl Cys)-Gly-Pro-(S-acetyl Cys)
14	(dimethylaminomethylcarbonyl-S-acetyl Cys)-Gly-Pro-(Cys-ethyl ester, S-acetyl)
15	(dimethylaminomethylcarbonyl-S-acetyl Cys)-Gly-Pro-(S-acetyl Cys)
16	dimethylaminomethylcarbonyl-Cys, S-dimethylaminomethylcarbonyl)-Gly-Pro-(Cys-ethyl ester, S-dimethylaminomethylcarbonyl)
17	(S-trimethylammoniomethylcarbonyl-Cys, trimethylammoniomethylcarbonyl)-Gly-Pro-(Cys ethyl ester, S-trimethylammoniomethylcarbonyl)
18	(dimethylaminomethylcarbonyl-Cys)-Gly-Pro-(Cys-ethyl ester)

[0050] In certain specific embodiments, the compound of the present invention is a compound of the formula I, wherein X_1, X_2, X_3 and X_4 are absent, i.e., a derivative or analogue of the amino acid sequence Cys-Pro-Cys, and: (i) R_1 is H, R_2 is ethyl, R_3 is -SH, and R_3' is -SH (compound **1**; R-951); (ii) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is H, R_3 is -SH, and R_3' is -SH (compound **2**; R-956); (iii) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, R_3 is -SH, and R_3' is -SH (compound **3**; R-952); (iv) R_1 is H, R_2 is H, R_3 is $-S-COCH_3$, and R_3' is -S-

$COCH_3$ (compound **4**; R-908); (v) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, R_3 is $-S-COCH_3$, and R_3' is $-S-COCH_3$ (compound **5**; R-911); (vi) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is H, R_3 is $-S-COCH_3$, and R_3' is $-S-COCH_3$ (compound **6**; R-958); (vii) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, R_3 is $-S-CO(CH_2)-N(CH_3)_2$, and R_3' is $-S-CO(CH_2)-N(CH_3)_2$ (compound **7**; R-909); (viii) R_1 is $-CO(CH_2)-N^+(CH_3)_3$, R_2 is ethyl, R_3 is $-S-CO(CH_2)-N^+(CH_3)_3$, and R_3' is $-S-CO(CH_2)-N^+(CH_3)_3$ (compound **8**; R-910); or (ix) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, and R_3 and R_3' are both -S- and together form a disulfide bond (compound **9**; R-953).

[0051] In other specific embodiments, the compound of the present invention is a compound of the formula I, wherein X_2 is a glycine residue; and X_1, X_3 and X_4 are absent, i.e., a derivative or analogue of the amino acid sequence Cys-Gly-Pro-Cys, and wherein: (i) R_1 is H, R_2 is ethyl, R_3 is -SH, and R_3' is -SH (compound **10**); (ii) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is H, R_3 is -SH, and R_3' is -SH (compound **11**); (iii) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, R_3 is -SH, and R_3' is -SH (compound **12**); (iv) R_1 is H, R_2 is H, R_3 is $-S-COCH_3$, and R_3' is $-S-COCH_3$ (compound **13**); (v) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, R_3 is $-S-COCH_3$, and R_3' is $-S-COCH_3$ (compound **14**); (vi) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is H, R_3 is $-S-COCH_3$, and R_3' is $-S-COCH_3$ (compound **15**); (vii) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, R_3 is $-S-CO(CH_2)-N(CH_3)_2$, and R_3' is $-S-CO(CH_2)-N(CH_3)_2$ (compound **16**); (viii) R_1 is $-CO(CH_2)-N^+(CH_3)_3$, R_2 is ethyl, R_3 is $-S-CO(CH_2)-N^+(CH_3)_3$, and R_3' is $-S-CO(CH_2)-N^+(CH_3)_3$ (compound **17**); or (ix) R_1 is $-CO(CH_2)-N(CH_3)_2$, R_2 is ethyl, and R_3 and R_3' are both -S- and together form a disulfide bond, (compound **18**).

[0052] In certain particular embodiments, the compound of the present invention is a dimer- or higher multimer-like compound as defined above, i.e., a compound of the formula I, wherein R_1 is H, $-CO(C_1-C_4)$alkyl, preferably $-CO(C_1-C_2)$alkyl, $-CO(C_1-C_4)$alkylene-NH$((C_1-C_4)$alkyl, preferably $-CO(C_1-C_2)$alkylene-NH$((C_1-C_2)$alkyl, $-CO(C_1-C_4)$alkylene-N$((C_1-C_4)$alkyl$)_2$, preferably $-CO(C_1-C_2)$alkylene-N$((C_1-C_2)$alkyl$)_2$, or $-CO(C_1-C_4)$alkylene-$N^+((C_1-C_4)$alkyl$)_3$, preferably $-CO(C_1-C_2)$alkylene-$N^+((C_1-C_2)$alkyl$)_3$; R_2 is (C_1-C_4)alkyl, preferably (C_1-C_2)alkyl, or H; one of R_3 and R_3' is -S- forming a disulfide bond with one of R_3 and R_3' of another compound of the formula I, and another of R_3 and R_3' is -SH, $-S-CO(C_1-C_4)$alkyl, preferably $-S-CO(C_1-C_2)$alkyl, $-S-CO(C_1-C_4)$alkylene-N$((C_1-C_4)$alkyl$)_2$, preferably $-S-CO(C_1-C_2)$alkylene-N$((C_1-C_2)$alkyl$)_2$, or $-S-CO(C_1-C_4)$alkylene-$N^+((C_1-C_4)$alkyl$)_3$, preferably $-S-CO(C_1-C_2)$alkylene-$N^+((C_1-C_2)$alkyl$)_3$, or R_3 and R_3' are both -S-, wherein R_3 forms a disulfide bond with one of R_3 and R_3' of another compound of the formula I, and R_3' forms a disulfide bond with either

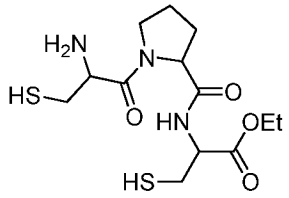
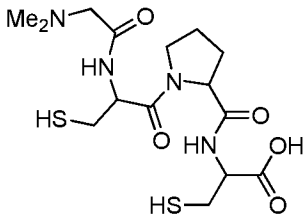
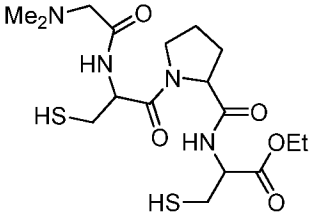
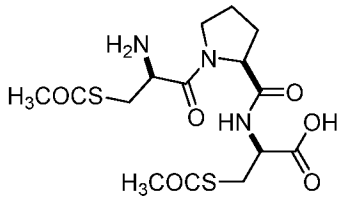
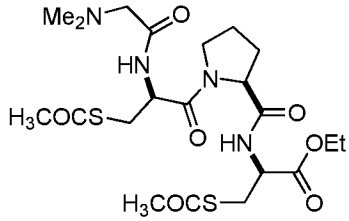
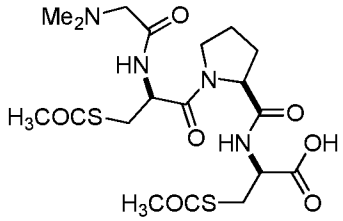
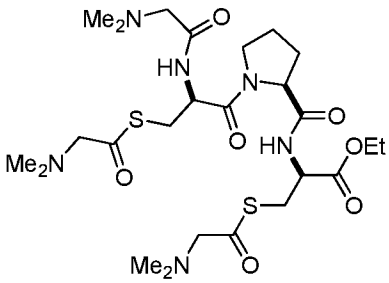
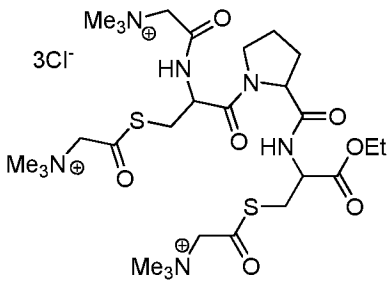
another of R_3 and R_3' of said another compound of the formula I (i.e., a cyclic dimer-like compound), or with one of R_3 and R_3' of a further compound of the formula I (i.e., a multimer-like compound); and X_1, X_2, X_3 and X_4 are absent, or at least one of X_1, X_2, X_3 and X_4 is present. In more particular such embodiments, R_1 is H, -CO-CH_2-N(CH_3)$_2$, or -CO-CH_2-N^+(CH_3)$_3$; R_2 is H, methyl, or ethyl; and either one of R_3 and R_3' is -S-, and another of R_3 and R_3' is -SH, -S-$COCH_3$, -S-CO-CH_2-N(CH_3)$_2$, or -S-CO-CH_2-N^+(CH_3)$_3$, or R_3 and R_3' are both -S-, wherein each one of the combinations of R_1, R_2, R_3 and R_3' constitutes a separate embodiment. In certain particular such dimer- or higher multimer-like compounds, wherein at least one of X_1, X_2, X_3 and X_4 is a glycine residue, and the other of X_1, X_2, X_3 and X_4 are absent.

[0053] The compounds of the formula I may be synthesized according to any technology or procedure known in the art, e.g., as described in Example 1 hereinafter.

[0054] The compounds of the formula I may have one or more asymmetric centers, and may accordingly exist both as enantiomers, i.e., optical isomers (R, S, or racemate, wherein a certain enantiomer may have an optical purity of 90%, 95%, 99% or more) and as diastereoisomers. Specifically, those chiral centers may be, e.g., in each one of the carbon atoms located at position alpha to any one of the carbonyl groups in the formula I. It should be understood that the present invention encompasses all such enantiomers, isomers and mixtures thereof, as well as pharmaceutically acceptable salts and solvates thereof.

[0055] Optically active forms of the compounds of the formula I may be prepared using any method known in the art, e.g., by resolution of the racemic form by recrystallization techniques; by chiral synthesis; by extraction with chiral solvents; or by chromatographic separation using a chiral stationary phase. A non-limiting example of a method for obtaining optically active materials is transport across chiral membranes, i.e., a technique whereby a racemate is placed in contact with a thin membrane barrier, the concentration or pressure differential causes preferential transport across the membrane barrier, and separation occurs as a result of the non-racemic chiral nature of the membrane that allows only one enantiomer of the racemate to pass through. Chiral chromatography, including simulated moving bed chromatography, can also be used. A wide variety of chiral stationary phases are commercially available.

Table 2: Compounds of the formula I, herein identified **1-9**

1	**2**
	
3	**4**
	
5	**6**
	
7	**8**
	
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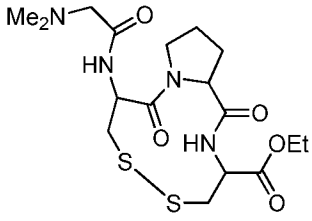
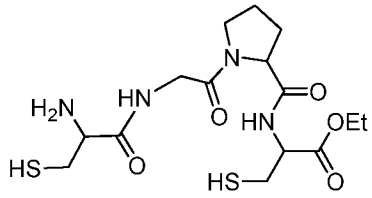
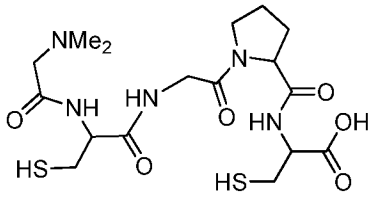
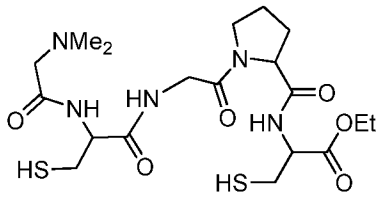
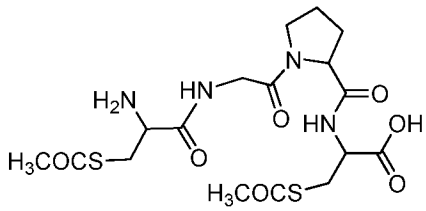
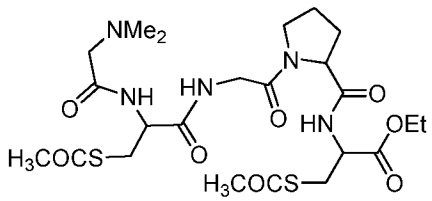
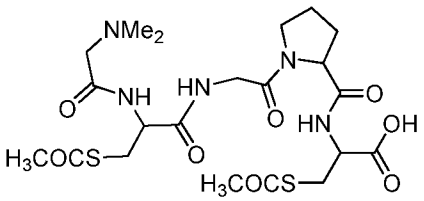
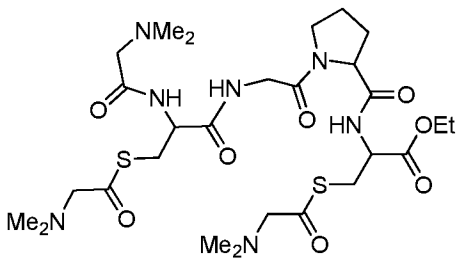
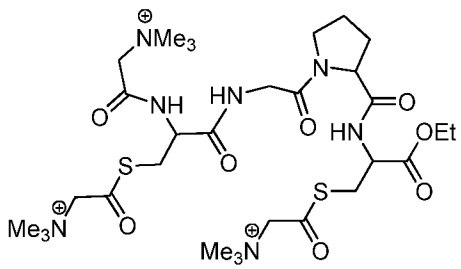
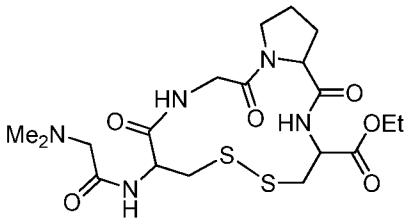
	

Table 3: Compounds of the formula I, herein identified **10-18**

10	**11**
	
12	**13**
	
14	**15**
	
16	**17**
	
18	
	

[0056] Reduction of the disulfide bond of TRX superfamily proteins, a critical step in providing intracellular redox defense, results from the donation of NADPH-derived electrons to TRX, followed by their transfer to recipient TRX superfamily proteins via a dithiolate exchange mechanism (Røhr *et al.*, 2013). The compounds of the present invention, e.g., compound **2**, are TRX mimetics with vicinal thiol groups that may

substitute for deficient TRX in mediating dithiolate exchange. These compounds have been rendered suitable for long-term shelf storage by overcoming the instability of their free thiols through formation of acetyl thioesters to yield inert prodrugs, e.g., compound **5**, wherein upon contact with biological fluids, the prodrugs regenerate the active di-thiol compounds. In contrast to existing antioxidants, that merely scavenge or catalyze degradation of ROS, the compounds of the present invention mimic the enzymatic function of TRX, serving as both a powerful reducing agent *per se* and a master intracellular switch regulating the activity of the TRX superfamily of proteins that govern cytoprotection, inflammation, and apoptosis (Kim *et al.*, 2010).

[0057] TRX mimetics and compounds providing reducing thiol equivalents are useful in the treatment of indications where oxidative stress occurs and ROS are generated. These compounds may scavenge ROS directly or via TRX-mimetic activity. However, compounds containing thiols and especially those with multiple thiols in close proximity are subject to facile oxidation to various disulfides and other products. This can occur on standing, as a solid, in solution and in biological matrixes. Prodrugs that mask the thiols prevent their instability, but allow the thiol equivalents to be released when administered to subjects.

[0058] The compounds of the present invention, which are TRX mimetic prodrugs, are thus expected to be effective in all those clinical indications wherein administration of the native TRX motif is beneficial, i.e., in the prevention, treatment or management of any disease, disorder, or condition mediated by redox stress. Such diseases, disorders or conditions include, without being limited to, chlorine inhalational lung injury, phosgene inhalational lung injury, hydrogen sulfide inhalational lung injury, ventilator-induced lung injury, lung ischemia reperfusion injury, chronic obstructive pulmonary disease (COPD), bronchopulmonary dysplasia, adult respiratory distress syndrome (ARDS), radiation induced lung fibrosis, congestive heart failure (CHF), myocardial infarction, myocardial ischemia-reperfusion injury, stroke, cardiopulmonary bypass surgery, doxorubicin-induced cardiomyopathy, limb ischemia reperfusion injury, renal ischemia repercussion injury, contrast media-induced nephropathy, hemorrhagic shock, endotoxic shock, septic shock, hepatic reperfusion injury, primary lung graft dysfunction, lung transplantation, renal transplantation, burn injury, angioplasty, traumatic brain injury, Parkinson's disease, ischemic bowel disease, mesenteric ischemia, retinopathy of prematurity, retinal degenerative disease, glaucoma, acute macular degeneration, cataracts, Crohn's disease,

and vitiligo. As shown herein, these compounds allow stable formulation and sustained delivery of the active thiol compounds that would otherwise be prone to rapid oxidation during storage and rapid oxidative metabolism *in vivo*.

[0059] In another aspect, the present invention thus provides a pharmaceutical composition comprising a pharmaceutically acceptable carrier and an active agent, i.e., a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof. In certain embodiments, the active agent is compound selected from compounds **1**-**18**, or a pharmaceutically acceptable salt or solvate thereof.

[0060] The pharmaceutical compositions of the present invention can be provided in a variety of formulations, e.g., in a pharmaceutically acceptable form and/or in a salt form, as well as in a variety of dosages.

[0061] In one embodiment, the pharmaceutical composition of the present invention comprises a non-toxic pharmaceutically acceptable salt of the active agent as defined above. Suitable pharmaceutically acceptable salts include acid addition salts such as, without being limited to, the mesylate salt, the maleate salt, the fumarate salt, the tartrate salt, the hydrochloride salt, the hydrobromide salt, the esylate salt, the *p*-toluenesulfonate salt, the benzenesulfonate salt, the benzoate salt, the acetate salt, the phosphate salt, the sulfate salt, the citrate salt, the carbonate salt, and the succinate salt. Additional pharmaceutically acceptable salts include salts of ammonium (NH_4^+) or an organic cation derived from an amine of the formula R_4N^+, wherein each one of the Rs independently is selected from H, C_1-C_{22}, preferably C_1-C_6 alkyl, such as methyl, ethyl, propyl, isopropyl, n-butyl, sec-butyl, isobutyl, tert-butyl, n-pentyl, 2,2-dimethylpropyl, n-hexyl, and the like, phenyl, or heteroaryl such as pyridyl, imidazolyl, pyrimidinyl, and the like, or two of the Rs together with the nitrogen atom to which they are attached form a 3-7 membered ring optionally containing a further heteroatom selected from N, S and O, such as pyrrolydine, piperidine and morpholine. Furthermore, where the compounds of the formula I carry an acidic moiety, suitable pharmaceutically acceptable salts thereof may include metal salts such as alkali metal salts, e.g., lithium, sodium or potassium salts, and alkaline earth metal salts, e.g., calcium or magnesium salts.

[0062] Further pharmaceutically acceptable salts include salts of a cationic lipid or a mixture of cationic lipids. Cationic lipids are often mixed with neutral lipids prior to use as delivery agents. Neutral lipids include, but are not limited to, lecithins; phosphatidylethanolamine; diacyl phosphatidylethanolamines such as dioleoyl

phosphatidylethanolamine, dipalmitoyl phosphatidylethanolamine, palmitoyloleoyl phosphatidylethanolamine and distearoyl phosphatidylethanolamine; phosphatidylcholine; diacyl phosphatidylcholines such as dioleoyl phosphatidylcholine, dipalmitoyl phosphatidylcholine, palmitoyloleoyl phosphatidylcholine and distearoyl phosphatidylcholine; phosphatidylglycerol; diacyl phosphatidylglycerols such as dioleoyl phosphatidylglycerol, dipalmitoyl phosphatidylglycerol and distearoyl phosphatidylglycerol; phosphatidylserine; diacyl phosphatidylserines such as dioleoyl- or dipalmitoyl phosphatidylserine; and diphosphatidylglycerols; fatty acid esters; glycerol esters; sphingolipids; cardiolipin; cerebrosides; ceramides; and mixtures thereof. Neutral lipids also include cholesterol and other 3β hydroxy-sterols.

[0063] Examples of cationic lipid compounds include, without being limited to, Lipofectin® (Life Technologies, Burlington, Ontario) (1:1 (*w/w*) formulation of the cationic lipid N-[1-(2,3-dioleyloxy)propyl]-N,N,N-trimethylammonium chloride and dioleoylphosphatidyl-ethanolamine); Lipofectamine™ (Life Technologies, Burlington, Ontario) (3:1 (*w/w*) formulation of polycationic lipid 2,3-dioleyloxy-N-[2(spermine-carboxamido)ethyl]-N,N-dimethyl-1-propanamin-iumtrifluoroacetate and dioleoylphosphatidyl-ethanolamine), Lipofectamine Plus (Life Technologies, Burlington, Ontario) (Lipofectamine and Plus reagent), Lipofectamine 2000 (Life Technologies, Burlington, Ontario) (Cationic lipid), Effectene (Qiagen, Mississauga, Ontario) (Non liposomal lipid formulation), Metafectene (Biontex, Munich, Germany) (Polycationic lipid), Eu-fectins (Promega Biosciences, San Luis Obispo, Calif.) (ethanolic cationic lipids numbers 1 through 12: $C_{52}H_{106}N_6O_4 \cdot 4CF_3CO_2H$, $C_{88}H_{178}N_8O_4S_2 \cdot 4CF_3CO_2H$, $C_{40}H_{84}NO_3P \cdot CF_3CO_2H$, $C_{50}H_{103}N_7O_3 \cdot 4CF_3CO_2H$, $C_{55}H_{116}N_8O_2 \cdot 6CF_3CO_2H$, $C_{49}H_{102}N_6O_3 \cdot 4CF_3CO_2H$, $C_{44}H_{89}N_5O_3 \cdot 2CF_3CO_2H$, $C_{100}H_{206}N_{12}O_4S_2 \cdot 8CF_3CO_2H$, $C_{162}H_{330}N_{22}O_9 \cdot 13CF_3CO_2H$, $C_{43}H_{88}N_4O_2 \cdot 2CF_3CO_2H$, $C_{43}H_{88}N_4O_3 \cdot 2CF_3CO_2H$, $C_{41}H_{78}NO_8P$); Cytofectene (Bio-Rad, Hercules, Calif.) (mixture of a cationic lipid and a neutral lipid), GenePORTER® (Gene Therapy Systems, San Diego, Calif.) (formulation of a neutral lipid (Dope) and a cationic lipid) and FuGENE 6 (Roche Molecular Biochemicals, Indianapolis, Ind.) (Multi-component lipid based non-liposomal reagent).

[0064] The pharmaceutically acceptable salts of the present invention may be formed by conventional means, e.g., by reacting the free base form of the active agent with one or more equivalents of the appropriate acid in a solvent or medium in which the salt is insoluble, or in a solvent such as water which is removed *in vacuo* or by freeze drying, or

by exchanging the anions of an existing salt for another anion on a suitable ion exchange resin.

[0065] The present invention encompasses solvates of the various active agents defined above as well as salts thereof, e.g., hydrates.

[0066] The pharmaceutical compositions provided by the present invention may be prepared by conventional techniques, e.g., as described in Remington: The Science and Practice of Pharmacy, 19th Ed., 1995. The compositions can be prepared, e.g., by uniformly and intimately bringing the active agent, i.e., the compound of the formula I, into association with a liquid carrier, a finely divided solid carrier, or both, and then, if necessary, shaping the product into the desired formulation. The compositions may be in liquid, solid or semisolid form and may further include pharmaceutically acceptable fillers, carriers, diluents or adjuvants, and other inert ingredients and excipients. In one embodiment, the pharmaceutical composition of the present invention is formulated as nanoparticles.

[0067] The compositions can be formulated for any suitable route of administration, but they are preferably formulated for parenteral administration, e.g., intravenous, intraarterial, intramuscular, intraperitoneal, intrathecal, intrapleural, intratracheal or subcutaneous administration, as well as for inhalation. The dosage will depend on the state of the patient, and will be determined as deemed appropriate by the practitioner.

[0068] The pharmaceutical composition of the invention may be in the form of a sterile injectable aqueous or oleagenous suspension, which may be formulated according to the known art using suitable dispersing, wetting or suspending agents. The sterile injectable preparation may also be a sterile injectable solution or suspension in a non-toxic parenterally acceptable diluent or solvent. Acceptable vehicles and solvents that may be employed include, without limiting, water, Ringer's solution and isotonic sodium chloride solution.

[0069] Pharmaceutical compositions according to the present invention, when formulated for inhalation, may be administered utilizing any suitable device known in the art, such as metered dose inhalers, liquid nebulizers, dry powder inhalers, sprayers, thermal vaporizers, electrohydrodynamic aerosolizers, and the like.

[0070] Pharmaceutical compositions according to the present invention, when formulated for administration route other than parenteral administration, may be in a form suitable for oral use, e.g., as tablets, troches, lozenges, aqueous, or oily suspensions, dispersible

powders or granules, emulsions, hard or soft capsules, or syrups or elixirs. Compositions intended for oral use may be prepared according to any method known to the art for the manufacture of pharmaceutical compositions and may further comprise one or more agents selected from sweetening agents, flavoring agents, coloring agents and preserving agents in order to provide pharmaceutically elegant and palatable preparations. Tablets contain the active agent(s) in admixture with non-toxic pharmaceutically acceptable excipients, which are suitable for the manufacture of tablets. These excipients may be, e.g., inert diluents such as calcium carbonate, sodium carbonate, lactose, calcium phosphate, or sodium phosphate; granulating and disintegrating agents, e.g., corn starch or alginic acid; binding agents, e.g., starch, gelatin or acacia; and lubricating agents, e.g., magnesium stearate, stearic acid, or talc. The tablets may be either uncoated or coated utilizing known techniques to delay disintegration and absorption in the gastrointestinal tract and thereby provide a sustained action over a longer period. For example, a time delay material such as glyceryl monostearate or glyceryl distearate may be employed. They may also be coated using the techniques described in the US Patent Nos. 4,256,108, 4,166,452 and 4,265,874 to form osmotic therapeutic tablets for control release. The pharmaceutical composition of the invention may also be in the form of oil-in-water emulsion.

[0071] The pharmaceutical compositions of the invention may be formulated for controlled release of the active agent. Such compositions may be formulated as controlled-release matrix, e.g., as controlled-release matrix tablets in which the release of a soluble active agent is controlled by having the active diffuse through a gel formed after the swelling of a hydrophilic polymer brought into contact with dissolving liquid (*in vitro*) or gastro-intestinal fluid (*in vivo*). Many polymers have been described as capable of forming such gel, e.g., derivatives of cellulose, in particular the cellulose ethers such as hydroxypropyl cellulose, hydroxymethyl cellulose, methylcellulose or methyl hydroxypropyl cellulose, and among the different commercial grades of these ethers are those showing fairly high viscosity. In other configurations, the compositions comprise the active agent formulated for controlled release in microencapsulated dosage form, in which small droplets of the active agent are surrounded by a coating or a membrane to form particles in the range of a few micrometers to a few millimeters.

[0072] Another contemplated formulation is depot systems, based on biodegradable polymers, wherein as the polymer degrades, the active agent is slowly released. The most common class of biodegradable polymers is the hydrolytically labile polyesters prepared

from lactic acid, glycolic acid, or combinations of these two molecules. Polymers prepared from these individual monomers include poly (D,L-lactide) (PLA), poly (glycolide) (PGA), and the copolymer poly (D,L-lactide-co-glycolide) (PLG).

[0073] The pharmaceutical compositions of the present invention are useful for prevention, treatment or management of a disease, disorder or condition mediated by redox stress as defined above. The term "treatment" as used herein with respect to a disease, disorder or condition mediated by redox stress refers to administration of a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, after the onset of symptoms of said disease, disorder or condition, regardless of the cause for that medical condition. The term "prevention" as used herein refers to administration of a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, prior to the onset of symptoms of said disease, disorder or condition, particularly to patients at risk for such disorder or condition; and the term "management" as used herein refers to prevention of recurrence of said disease, disorder or condition in a patient previously suffered from that medical condition.

[0074] The therapeutic and/or prophylactic efficacy of compounds of the formula I, in particular of R-911 exemplified herein, in preventing mortality caused by NaSH injection; preventing and/or reducing apoptosis in a lung radiation-induced injury model; protecting the lung from ischemia/reperfusion injury, chlorine inhalational lung injury, or ventilator-induced lung injury; protecting against LPS-induced injury; protecting the kidney from contrast media-induced nephropathy; treatment of renal ischemia-reperfusion injury; and reducing infarct size and myocardial damage resulting from myocardial ischemia-reperfusion injury, is shown in Examples 3-11 hereinafter.

[0075] In a further aspect, the present invention thus relates to a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, for use in prevention, treatment or management of a disease, disorder or condition mediated by redox stress. Particular such compounds are those herein identified as compounds **1-18**, e.g., R-911, or pharmaceutically acceptable salts or solvates thereof.

[0076] In yet another aspect, the present invention relates to use of a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, for the preparation of a pharmaceutical composition for prevention, treatment or management of a disease, disorder or condition mediated by redox stress. Particular such compounds are

those herein identified as compounds **1-18**, e.g., R-911, or pharmaceutically acceptable salts or solvates thereof.

[0077] In still another aspect, the present invention relates to a method for prevention, treatment or management of a disease, disorder or condition mediated by redox stress in a subject, e.g., an individual, in need thereof, comprising administering to said subject an effective amount of a compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof. Particular such compounds are those herein identified compounds **1-18**, e.g., R-911, or pharmaceutically acceptable salts or solvates thereof.

[0078] The term "effective amount" as used herein refers to the quantity of the compound of the formula I as defined above, or a pharmaceutically acceptable salt or solvate thereof, that is useful to prevent, treat, or manage the particular disease, disorder or condition that is to be prevented, treated or managed in said individual, and may thus, in fact, be a "prophylactically effective amount" or "therapeutically effective amount".

[0079] The invention will now be illustrated by the following non-limiting Examples.

EXAMPLES

Example 1. Synthesis of R-911 and R-952

[0080] The tetra-peptide prodrug R-911 was assembled as described below, following the linear synthesis shown in **Scheme 1** (see Appendix) performed from the commercially available staring materials Boc-L-Cys(trt)-OH, Boc-L-Pro-OH, Fmoc-L-Cys(trt)-OH and Fmoc-Pro-OH.

[0081] In step 1, N-Boc-(S-trityl)-Cys(OH) (**a**; 7.6 gm, 0.016 mol) was suspended in dry acetonitrile (50 ml). Diisopropylethylamine (4 ml, 0.025 mol) and iodoethane (2.6 ml, 0.032 mol) was added. After the addition, the reaction mixture became clear. It was stirred at RT for 2 days. Additional, 2.6 ml of iodoethane was added and stirred further for 6 hours. The reaction mixture was concentrated. Water (200 ml) and EtOAC (200 ml) was added. N-Boc-(S-trityl)-Cys(OEt) (**b**) was collected, dried on Na_2SO_4 and concentrated (7.7 gm).

[0082] In step 2, N-Boc-(S-trityl)-Cys(OEt) (7.5 gm) was dissolved in dry EtOAc (40 ml). Solution of HCl in dioxane (4M, 5 eq) was added, and the reaction was stirred at RT until the starting material disappeared. The reaction mixture was concentrated on rotary evaporator at 40°C. Residue was washed with hexane (50 ml) to remove the trityl chloride

impurity, and was then dried under vacuum (~100% yield of crude residue); and the material was used as such for the next reaction.

[0083] In step 3, N-Boc-Pro(OH) (3.5 gm, 0.016 mol) was suspended in dry CH_2Cl_2 (50 ml). N,N'-dicyclohexylcarbodiimide (DCC; 3.350 gm) and N-methyl morpholine (NMM, 3 eq.) was added. The mixture was stirred at RT for 30 min. To the above mixture, a solution of NH_2-(S-trityl)-Cys(OEt) HCl salt (0.016 mol) in dichloromethane (25 ml) was added, and the reaction was stirred at RT until the starting material disappeared (~4 hours). The reaction mixture was filtered to remove the DCC urea; the filtrate was concentrated; and EtOAC (100 ml) was added and urea separated out was filtered again. The filtrate was washed with sat $NaHCO_3$, water and brine. It was dried on Na_2SO_4 and concentrated. The residue was purified on silica gel column on combi-flash 25% (5 min); 25-40% (5 min) and 40% (5 min). Product (**d**; N-Boc-Pro-Cys-(OEt) (S-trityl)) eluted ~40% EA-hexane (8.1 gm).

[0084] In step 4, **d** (7.3 gm) was dissolved in dry EtOAc (50 ml) and 4M solution of HCl in dioxane (15.5 ml, 0.062 mol) was added. The reaction was stirred at RT until the starting material disappeared. The reaction mixture was concentrated on rotary evaporator at 50°C. Residue was washed with hexane (50 ml) to remove the trivial amount of trityl chloride impurity and HCl, and was then dried under vacuum to remove the excess HCl from product (**e**).

[0085] In step 5, a suspension of Fmoc-Cys(OH)-S-Trt (730 mg, 1 eq), DCC (260 mg), NMM (3 eq) in CH_2Cl_2 (20 ml) was stirred at RT for 30 min. Intermediate **e** HCl salt (650 mg, 0.0012 mol) was added and stirred at RT for overnight. The reaction mixture was concentrated. EtOAC (20 ml) was added and the DCC urea was filtered. The filtrate was washed with water, $NaHCO_3$ and brine (to reduce the emulsion), and was then dried on sodium sulfate and concentrated. The residue was purified on silica gel column using 50% EA-hexane (820 mg).

[0086] In step 6, intermediate **f** (820 mg) was dissolved in acetonitrile (15 ml) and diethylamine (5 ml) was added. The mixture was stirred at RT for 2 hours, and concentrated on rotary evaporator. The residue was purified on silica gel column 5% MeOH-CH_2Cl_2, which provided the amino intermediate **g** (425 mg).

[0087] In step 7, intermediate **g** HCl salt (27 gm mixture of two isomers) was dissolved in EtOAC (200 ml) and $NaHCO_3$ (200 ml), and the solution was stirred for 5-10 min. Chloroacetyl chloride (3 eq) was slowly added. The reaction mixture was stirred for 1

hour. EtOAC layer was separated, and washed with water. It was dried on Na_2SO_4 and concentrated. The residue was purified on silica gel column in 3-4% MeOH-CH_2Cl_2 to provide **h** as a mixture of isomer 1 (18 gm) and isomer 2 (4 gm).

[0088] In step 8, intermediate **h** (mixture of isomers, 8 gm) was dissolved in tetrahydrofuran (THF), and a 2M solution of dimethylamine in THF (70 ml) was added. The mixture was stirred at RT for overnight, and was then concentrated. The residue was purified on silica gel column using 3-5% methanol-CH_2Cl_2 to provide intermediate **i** (6 gm).

[0089] In step 9, intermediate **i** (2.2 gm) was dissolved in methylene chloride (25 ml) and triethyl silane (4 eq) was added. The mixture was treated with trifluoroacetic acid (TFA; 4 eq) at RT, and was then stirred for 6 hours. It was concentrated on rotary evaporator. The residue was suspended in hexane (20 ml). Hexane was removed. The residue was washed again with hexane (20 ml) and dried under vacuum to give the TFA salt of R-952 (compound **3**), which was used as such for the next reaction.

[0090] In step 10, triethylamine (4 eq) and acetic anhydride (4 eq) was added to a solution of R-952 (1.2 gm) in methylene chloride (25 ml). The reaction mixture was stirred at RT for 16 hours. Excess triethylamine and acetic anhydride was removed on rotary evaporator. The corresponding crude acetate salt was dissolved in water and purified on C-18 column using 0.1% solution of acetic acid in water and acetonitrile. The fractions collected at 20% to 40% gradient were re-purified on C-18 column to produce R-911 isomer 1 (30 mg) MS (ES+): 519.1423 [M+1], 541.1207 [M+Na]; and isomer 2 (380 mg). MS (ES+): 519.1422 [M+1], 541.1212 [M+Na].

[0091] A convergent synthesis of R-911 is shown in **Scheme 2** (see Appendix).

Example 2. R-911 is converted under physiological conditions to its corresponding active form

[0092] R-911 was incubated with diluted rat plasma (1 vol in 10 vol water) at 37°C, in a final concentration of 1 mg/ml. Samples of the incubation mixture were extracted after 15 min and 18 hours with 2 volumes of cold ethanol. After mixing and incubation for 10 min on ice, the solutions were centrifuged for 15 min and the supernatant taken diluted in 0.1% TFA in water. LCMS analysis of the diluted supernatants indicated the absence of the parent, R-911 (expected retention time (Rt)=4.56 min, [M+H]+ 519.31+ ions). The following new peaks were observed: (i) at t=3.62 min with an intense [M+H]+ ion at

491.27+ Da., corresponding to de-esterification of the ethyl ester functionality in R-911, giving the "free acid" derivative; (ii) at t=3.22 min with an [M+H]+ ion at 449.25 + Da., corresponding to de-esterification of one of the thio esters of the free acid; (iii) at t=2.68 min with an [M+H]+ ion at 407.23+ Da., corresponding to de-esterification of both the thio esters of the free acid R-956 (compound **2**). In addition, a series disulphide compounds corresponding to dimers of the mono and di-thiols were observed.

[0093] These results show that not only is the active R-956 generated from the prodrug (R-911), but the active is still being generated after 18 hours. Thus, the prodrug serves as a sustained source of the active under conditions where the active would otherwise be rapidly oxidized to disulphides.

Example 3. R-911 increases survival following NaSH injection

[0094] In order to evaluate the therapeutic efficacy of R-911 in preventing mortality caused by NaSH, a survival study was conducted. 28 adult male mice were IP injected with 30 mg/kg NaSH (LD_{70}) and were then divided into two equal sized groups. 1 hour post NaSH injection, the treated group was injected with 130 mg/kg of R-911, whereas the control group was injected with vehicle. Mortality rate was observed among groups for a week. Whereas only 3 of the control group mice were alive at 156 hours post NaSH injection (21% survival), 10 of the treatment group mice were alive at the same time point (**Table 4**; **Fig. 1**), pointing at the remarkable therapeutic potential of R-911 in scenarios of NaSH toxicity.

Table 4: Number of living mice at each time post-NaSH injection for each group

	Time after NaSH administration (h)											
	0	1	3	6	12	24	48	72	96	108	132	156
Vehicle	14	12	9	6	6	4	3	3	3	3	3	3
R-911	14	12	11	10	10	10	10	10	10	10	10	10

[0095] Considering that the lung is the prominent site of injury upon inhalation of NaSH, the next experiment was aimed at verifying whether R-911 can protect lung from injury in our model. We repeated the experimental protocol described above, but this time sacrificed the mice 5 hours post NaSH injection. Out of 14 mice in each group, 6 mice remained alive at 5 hours post NaSH injection in the control group, whereas 8 remained alive at the R-911 treated group. These remaining mice were sacrificed and their lungs were histologically

examined. **Fig. 2** shows that R-911 had a dramatic protective effect on lungs of mice injected with NaSH and treated with R-911 as evidenced by histological injury scoring.

Example 4. R-911 has prophylactic and therapeutic anti-apoptotic effects in a lung radiation-induced injury model

[0096] In these studies, both the prophylactic and therapeutic anti-apoptotic effects of R-911 were evaluated in a lung radiation-induced injury model.

[0097] In the first study, R-911 was delivered IP at a dose of 250 mg/kg/day for 7 days. C57BL/6 male mice were exposed to 14 Gy radiation to the thorax. Five minutes before exposure, R-911 (125 mg/kg) or vehicle (radiation only group) was delivered. A second delivery of R-911 or vehicle was given 4 hours later on the day of radiation and treatments continued twice daily for the next 6 days. Mice were necropsied on Day 7 post-radiation and their lungs examined by terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL) assay to assess the percent apoptotic cells. As shown in **Fig. 3**, R-911 treatment significantly decreased radiation-induced apoptosis.

[0098] In the second study, R-911 was delivered IP at a dose of 250 mg/kg/day for 6-7 days. C57BL/6 male mice were exposed to 14 Gy radiation to the thorax. In group A (pre-radiation), treatment was initiated 5 minutes before radiation was delivered, and continued 4 hours later and twice daily for the next 6 consecutive days. In group B (post-radiation), treatment was initiated 24 hours after radiation was delivered, and continued 4 hours later and twice daily for the next 5 consecutive days. In both groups, mice were necropsied on Day 7 post-radiation and their lungs examined by TUNEL assay to assess the percent apoptotic cells. As shown in **Fig. 4**, R-911 treatment significantly decreased radiation-induced apoptosis when delivered either pre- or post-radiation.

Example 5. R-911 protects against LPS-induced injury of multiple organs

[0099] In this study, 15 male Balb/C mice were randomly divided to three groups: Sham (n=3), Vehicle (n=6) and R-911 (n=6). LPS (0.25 mg/25 g mouse) was IP injected to mice in Vehicle and R-911 groups. At 1, 6 and 11 hours post LPS injection, R-911 (100 mg/kg per each dose) was IP administered. Mice in all groups were sacrificed 18 hours post LPS injection. Serum was collected as well as liver, kidney, lung and ileum tissues for further analysis.

[00100] R-911 protected mice from LPS-induced injury as indicated by multiple biomarkers of LPS pathology. As shown in **Table 5**, creatinine and BUN levels, important markers of renal health, were reduced, determining a protective effect of R-911 on kidney function. ALT and AST levels, which increase during acute liver failure, were also markedly reduced, demonstrating efficacy of R-911 in preventing liver damage. Histological injury was evaluated in lung, kidney and liver, and the histological score was based on a scale between 0 (no damage) and 4 (severe injury). All three yielded scores portraying significant reduction of histological destruction and restoration of tissue architecture. The *p*-values in the chart are the difference between the vehicle control group and R-911 treatment group, all of which suggest data are exceedingly significant (± value is Standard Error).

Table 5: R-911 protects mice from LPS-induced injury as indicated by multiple biomarkers of LPS pathology

	Sham	LPS+Vehicle	LPS+R-911	Correction (%)	*P* value
Creatinine (mg/dl)	0.140±0.006	0.381±0.009	0.258±0.012	51%	$>10^{-5}$
BUN (mg/dl)	30.67±1.453	101.83±2.271	65.50±1.707	51%	$>10^{-7}$
ALT (U/l)	25.33±0.88	59.17±3.67	32.33±0.99	79%	$>10^{-4}$
AST (U/l)	152.33±1.76	662.00±10.53	381.67±14.59	55%	$>10^{-7}$
Histological injury score					
Lung	0.00±0.00	3.17±0.16	1.17±0.16	63%	$>10^{-5}$
Kidney	0.00±0.00	2.33±0.21	1.00±0.00	57%	$>10^{-4}$
Liver	0.00±0.00	1.67±0.21	0.67±0.21	60%	$>10^{-2}$

Example 6. R-911 is effective in lung ischemia-reperfusion injury model

[00101] Lungs of adult male Sprague-Dawley rats were rendered ischemic for 1 hour and then re-perfused for 4 hours. 80 mg/kg of R-911 were administered twice to treatment group (n=6): 3 minutes infusion right before reperfusion was followed one hour later by 3 hours long infusion. Vehicle was similarly given to a control group (n=6). Rats of a third sham group underwent thoracotomy but neither experienced ischemia nor drug therapy (n=6). Rats were sacrificed after reperfusion and lungs were harvested for examination.

[00102] R-911 protected the lung from I/R injury as indicated by multiple biomarkers of the disease. Evidence of improved lung oxygenation was demonstrated by elevation of PaO_2 (222 mmHg in control group *vs.* 351 mmHg in R-911 group, $p<0.001$, **Fig. 5**). A histological examination of lung tissue revealed a dramatic reduction in injury to the

treatment group (2.33 in the treatment group *vs.* 4.17 in the control group, $p<0.001$, **Fig. 6**). MPO concentration, an index of neutrophil infiltration in lung tissue, and MDA concentration, an index of lipid peroxidation, are commonly utilized as markers of inflammation in I/R injury model. Both significantly displayed the protective effect of R-911. MPO was 0.83 U/gr in the treatment group, compared to 1.22 U/gr in the control group ($p<0.001$, **Fig. 7A**). MDA was 283.83 μM/100 mg of wet tissue in treatment group, compared to 450.67 μM/100 mg of wet tissue in the control ($p<0.001$, **Fig. 7B**). Protein concentration was quantified in BAL, a method used to determine the integrity of the lung epithelium. A significant reduction was found in the protein concentration in the BAL of R-911 treated group (691.33 mg/ml in control group to 409.5 mg/ml in R-911 treated group, $p<0.001$, **Fig. 8**). Wet/dry ratio, a method that measures the increase of water in the lungs, is utilized to determine the presence of edema induced by pulmonary injury. Here, a 50% reduction in R-911 treated group when compared with treatment, indicating a decrease in edema ($p<0.001$, **Fig. 9**). Taken together, the data collected establish R-911 as a potent therapy in improving the symptoms of lung I/R injury and modifying the course of injury by restoring lung function.

Example 7. R-911 prevents kidney damage in models of contrast media-induced nephropathy

[00103] In these studies, the therapeutic effects of R-911 were assessed in uni-nephrectomy and diabetic models of CIN.

Non-diabetic model

[00104] Three weeks prior to contrast media (Omnipaque) application, Sprague-Dawley rats ($n=5$ in each group) underwent right-sided nephrectomy. The rats were divided into the following treatment groups: (i) Vehicle (saline); (ii) R-911 10 mg/kg/day; (iii) R-911 30 mg/kg/day; (iv) R-911 100 mg/kg/day; (v) NAC (equimolar to 100 mg/kg/day of R-911); and (vi) Sham operation. A full treatment was divided into three equal portions, which were administered by IP injection during the course of the study.

[00105] Twenty-four hours prior to Omnipaque application, the animals were deprived of drinking water; 30 minutes prior to Omnipaque application, they received the first portion of the treatment by IP injection; and 15 minutes prior to Omnipaque application, they were injected with indomethacin, a non-steroidal anti-inflammatory drug, to increase ischemic

susceptibility of the renal medulla. At 8 and 16 hours following Omnipaque application, the animals received the second and third portions of the treatment.

[00106] At 24 hours following Omnipaque application, the animals were sacrificed, blood was collected for plasma preparation, and a kidney was removed from each animal. One half of each kidney was snap-frozen and the other half was fixed in formalin. Creatinine and BUN concentrations in the isolated plasma samples were determined using standard methods.

[00107] Kidney tissue samples were fixed for 1 week in 10% (w/v) PBS-buffered formaldehyde solution at RT, dehydrated using graded ethanol, and embedded in Paraplast (Sherwood Medicab l, Mahwah, NJ, USA). Sections were then deparaffinized with xylene, and stained with hematoxylin and eosin. All sections were studied using Axiovision Zeiss (Milan, Italy) microscope. The following morphological criteria were used for scoring: 0, normal kidney; grade 1, minimal edema or infiltration; grade 3, moderate edema and inflammatory cell infiltration without obvious damage to kidney architecture; grade 4, severe inflammatory cell infiltration with obvious damage to kidney architecture. All the histological studies were performed in a blinded fashion.

[00108] All values expressed as mean ± standard error of the mean (SEM) of N (number of animals studied) observations. The results were analyzed by Student's paired test. A *p*-value of less than 0.05 was considered significant.

[00109] As found, contrast media application severely damaged the kidneys, as was obvious from the dramatically elevated plasma BUN and creatinine levels (**Fig. 10A**) indicating that the kidneys are poorly functioning. R-911 treatment attenuated these changes in a dose-dependent manner. The 100 mg/ kg dose was found to improve kidney function better than the comparable NAC treatment.

[00110] Contrast media-induced severe kidney damage was also clear from the dramatic changes in kidney architecture shown in **Fig. 11**. After scoring the condition of each section and averaging the results for each group, it was clear that R-911 treatment attenuated these changes in a dose-dependent manner. The 100 mg/kg dose was found to improve kidney structure better than the comparable NAC treatment.

Diabetic model

[00111] The protocol in this case was similar to that in the non-diabetic model, except that 2 weeks prior to contrast media (Omnipaque) application, Sprague-Dawley rats (*n*=5 in

each group) were rendered diabetic via streptozotocin (STZ; 60 mg/kg) injection. Additionally, the added effects of aggressive volume resuscitation during treatment were assessed. The study included the following treatment groups: (i) Sham (n=3) no contrast media, saline injections 0.5 ml; (ii) Vehicle (saline) injections 0.5 ml; (iii) R-911 300 mg/kg/day (100 mg/kg/dose ×3 doses) - each injection in 0.5 ml saline; (iv) NAC 94.5 mg/kg/day (31.5 mg/kg/dose ×3 doses) in 0.5 ml saline; (v) R-911 300 mg/kg/day (100 mg/kg/dose ×3 doses) in increased volume 10 ml/kg - each injection 2.5 ml saline for a 250g rat; and (vi) increased volume alone 10 ml/kg - each injection 2.5 ml saline for a 250g rat.

[00112] In this model, kidney damage was somewhat greater than in the non-diabetic model according to serum BUN levels (**Fig. 13A**). However, the histological assessment (**Fig. 12**) and serum creatinine levels (**Fig 13B**) did not significantly differ in the models. As in the non-diabetic model, R-911 was found to attenuate the changes of the model disease somewhat better than NAC. Hydration, alone or in combination with R-911, had no apparent effect on any of the assessed parameters.

[00113] The data obtained in both non-diabetic and diabetic models support the hypothesis that R-911 treatment can improve both functional and structural kidney damage caused by contrast media even when diabetes is present and may be a potent therapy for individuals at high-risk for CIN.

Example 8. R-911 is effective as a rescue therapy in a murine Cl_2 exposure model

[00114] In this study, the therapeutic effect of R-911 in treatment of CILI was tested.

[00115] In a chemical fume hood, Balb/c mice (n=6 in each group) were exposed in a cylindrical glass chamber (4 mice per exposure) that is flushed continuously for 30 minutes at a rate of 2 liters/minute with humidified gas obtained from a calibrated cylinder containing air and 400 ppm Cl_2. After the end of the 30-minute exposure, the chamber was opened and mice were removed and placed immediately in cages in room air. Thirty minutes after the conclusion of Cl_2 exposure, mice were administered IP with 300 mg/kg/dose of R-911 in 200 µl. At 24 hours post-exposure to the Cl_2-containing air, a midline incision from the neck to the pubis was created for access to the chest and abdominal cavities. The pulmonary circulation was flushed through the main pulmonary artery with 20 ml of normal saline. The lungs were separated from the mediastinal tissues

and were taken for histological examination (H&E staining). The following morphological criteria were used for scoring: 0, normal lung; grade 1, minimal edema or infiltration of alveolar or bronchiolar walls; grade 3, moderate edema and inflammatory cell infiltration without obvious damage to lung architecture; grade 4, severe inflammatory cell infiltration with obvious damage to lung architecture. All the histological studies were performed in a blinded fashion.

[00116] As shown in **Fig. 14**, CILI was evident in the vehicle control group animals according to dramatically worsened histological scoring. By comparison, when administered 30 minutes post a 30-minute exposure to Cl_2-containing air, R-911 significantly attenuated CILI in mice 24 hours post exposure as demonstrated by the improved histology scores, reducing injury by 42% ($p<0.00005$).

Example 9. R-911 is effective as a rescue therapy in a ventilator-induced lung injury model

[00117] In this study, the therapeutic effect of R-911 in treatment of VILI was tested.

[00118] Male Sprague-Dawley rats ($n=5$ in each group) were subjected to high TV mechanical ventilation with positive end expiratory pressure of 3 cm H_2O. TV was initially set at 30 ml/kg and then reduced to limit peak inspiratory pressure to 30 cm H_2O. Animals were dosed with 40 mg/kg R-911 or an equal volume of saline (0.2 ml) via endotracheal tube 1 minute before the onset of high volume mechanical ventilation. Both groups were ventilated at a FiO_2 of 100% and the rate adjusted to maintain PCO_2=40 torr. Arterial PO_2 was checked at 1, 2, 3, and 4 hours. Rats were extubated after 4 hours and allowed to recover. Rats were sacrificed at 24 hours. MPO was measured and lung histology was assessed. In order to assess histology, lung tissue samples were fixed for 1 week in 10% (w/v) PBS-buffered formaldehyde solution at room temperature, dehydrated using graded ethanol, and embedded in Paraplast (Sherwood Medicab l, Mahwah, NJ, USA). Sections were then deparaffinized with xylene, and stained with hematoxylin and eosin. All sections were studied using Axiovision Zeiss (Milan, Italy) microscope. The following morphological criteria were used for scoring: 0, normal lung; grade 1, minimal edema or infiltration of alveolar or bronchiolar walls; grade 3, moderate edema and inflammatory cell infiltration without obvious damage to lung architecture; grade 4, severe inflammatory cell infiltration with obvious damage to lung architecture. All the histological studies were performed in a blinded fashion.

[00119] As shown in **Fig. 15**, VILI was evident in the vehicle control group animals according to dramatically worsened PaO_2 over the 4-hour period. R-911 administration attenuated PaO_2 decrease in rats during this period.

[00120] Prominent VILI-induced damage was also observed in histology samples (**Fig. 16**). This included edema, infiltration of neutrophils, and destruction of tissue architecture. R-911 significantly improved the lung tissue condition after VILI. This improvement was also apparent according to the levels of infiltrating neutrophils according to changes in MPO levels. While VILI caused extreme elevations in MPO levels (**Fig. 17**), R-911 diminished these levels by 29% ($*=p<10^{-6}$). These results indicate that R-911 can be a useful therapy for treating VILI.

Example 10. R-911 is effective as a therapy in a renal ischemia reperfusion injury model

[00121] In this study, the therapeutic effect of R-911 in prevention of RIRI was tested.

[00122] Anesthetized male Sprague-Dawley rats (n=4 in each group) underwent bilateral clamping of the renal pedicles for 30 minutes, followed by 6 hours of reperfusion. The rats were divided into the following treatment groups: (i) Vehicle (saline); (ii) R-911 30 mg/kg loading dose, 10 mg/kg/hr infusion rate over; (iii) R-911 100 mg/kg loading dose, 33 mg/kg/hr infusion rate; (iv) R-911 300 mg/kg loading dose, 100 mg/kg/hr infusion rate; and (v) Sham operation.

[00123] The loading R-911 (or vehicle) dose was administered 5 minutes prior to reperfusion, and subsequent infusions immediately followed reperfusion. Blood samples were drawn at 6 and at 24 hours, when the animals were terminated, to determine plasma creatinine levels. At termination, kidneys were removed and samples were fixed for histological examination. As shown in **Fig. 18**, administration of R-911 before reperfusion profoundly attenuated elevations in plasma creatinine in a dose dependent manner (**Fig. 18**). Histologic studies (**Fig. 19**) revealed that R-911 provided near total protection against severe renal tubular necrosis. These results indicate that R-911 can be a useful therapy for preventing RIRI.

Example 11. R-911 reduces infarct size and myocardial damage in a rodent model of myocardial ischemia-reperfusion injury

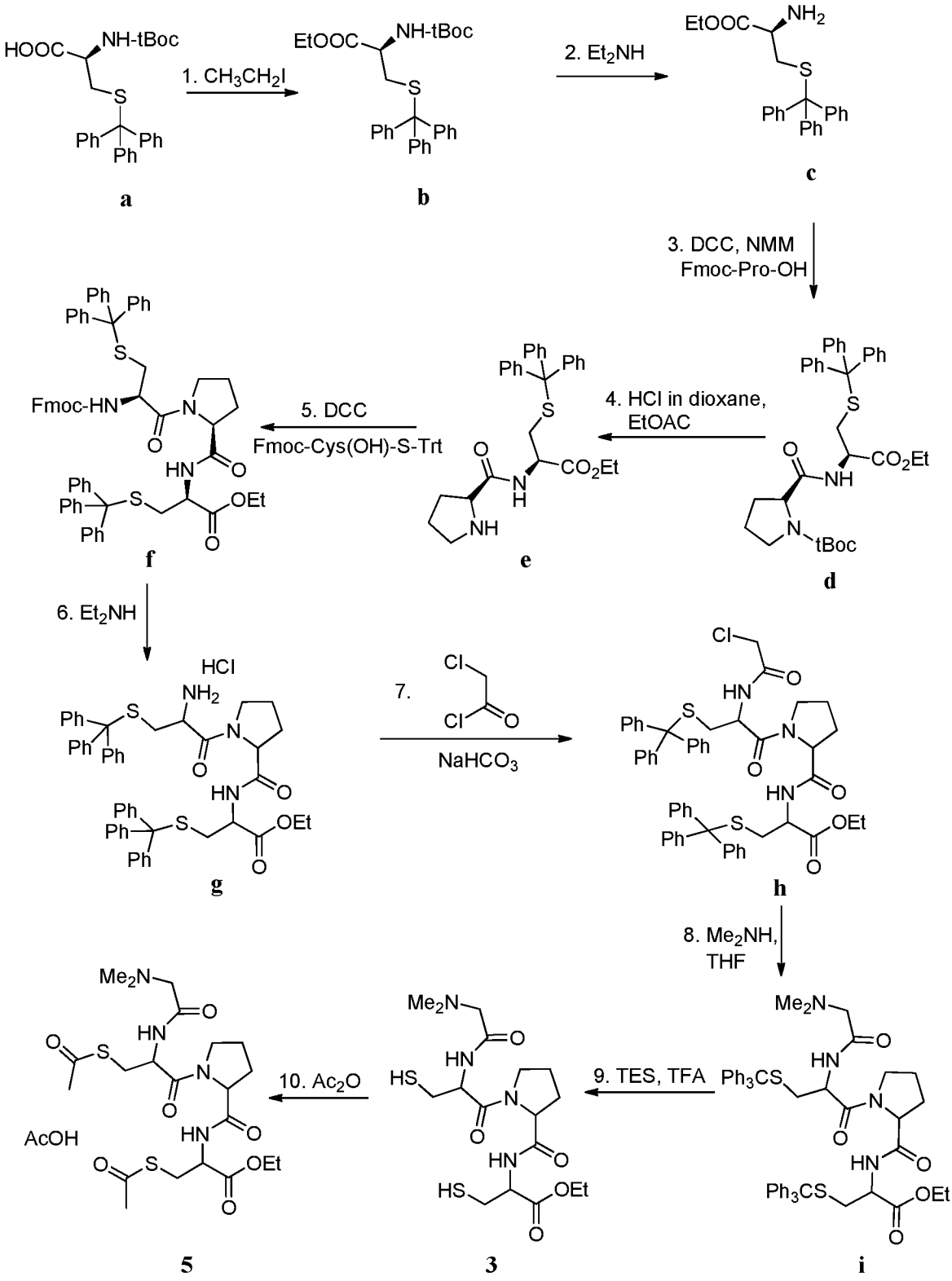
[00124] In this study, the therapeutic effect of R-911 in prevention of MIRI was tested.

[00125] Male adult Wistar rats (250-300 g), anesthetized with thiopentone sodium and mechanically ventilated (FiO_2=30%, IMV=70 bpm, TV=8-10 ml/kg), underwent placement of a ligature around the LAD coronary artery approximately 1-2 mm below its origin. Ischemia was induced by tightening the threads of the coronary suture and was maintained for 20 min. Reperfusion for 2 hours was obtained by reopening the chest and cutting the ligature around the coronary artery. Rats were randomly allocated to the following groups (n=5 per each group): (i) MIRI+vehicle group: rats were subjected to coronary artery occlusion (20 min) followed by reperfusion (2 hours); (ii) MIRI+R-911: rats were subjected to the surgical procedures described above and were treated with an IV bolus of 200 mg/kg R-911, followed by a continuous IV infusion of 50 mg/kg/h; and (iii) Sham+vehicle group: rats were subjected to identical surgical procedures, except for coronary artery occlusion, and were kept under anaesthesia for the duration of the experiment. At the end of the 2-hour reperfusion period, the LAD was re-occluded, and 1 ml of Evans blue dye (2% wt/vol) was injected to the animal via the jugular vein. The area at risk, i.e., the non-perfused and thus non-stained myocardium, was separated from the non-ischemic tissue and expressed as a percentage of the left ventricle. The tissue from the are at risk staining with p-nitroblue tetrazolium was separated from the infarcted tissue and weighed, and the infarct size was expressed as a percentage of the are at risk.

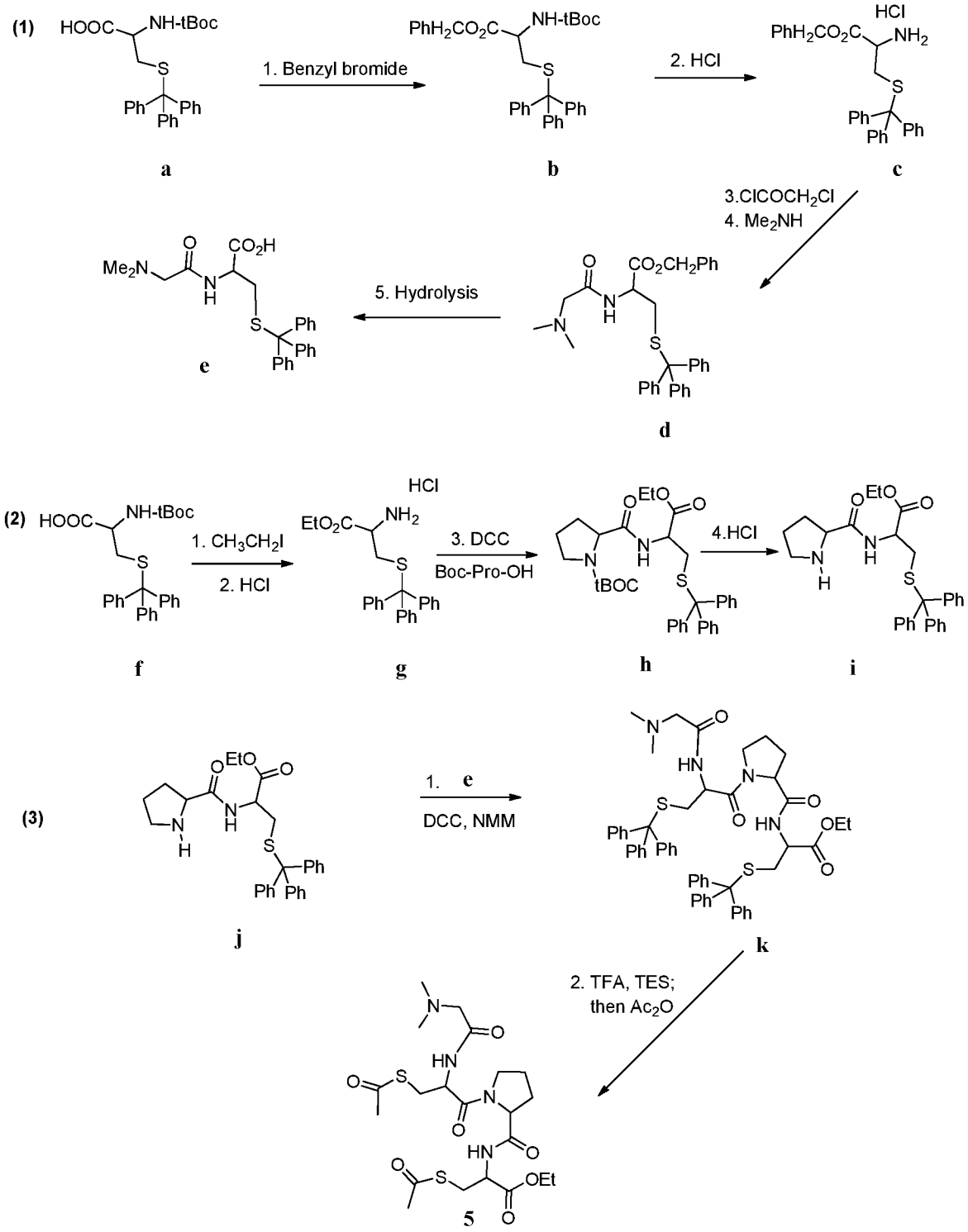
[00126] **Fig. 20** shows that R-911 significantly reduced tissue infarction, myocardial MPO, and lipid peroxidation. **Fig. 21A** shows that MIRI induced severe myocyte necrosis, edema and neutrophil infiltration, compared to a sham control, and administration of R-911 prior to reperfusion profoundly attenuated virtually all histologic features of injury. **Fig. 21B**, presenting histological injury scores, indicates a dramatic attenuation of injury by R-911 (40%, p<0.0003 in a 2-tailed t-test). These results indicate that R-911 can be a useful therapy for preventing MIRI.

APPENDIX

Scheme 1: Linear process for the synthesis of R-952 and R-911 (compounds **3** and **5**)



Scheme 2: Convergent process for the synthesis of R-911 (compound **5**)



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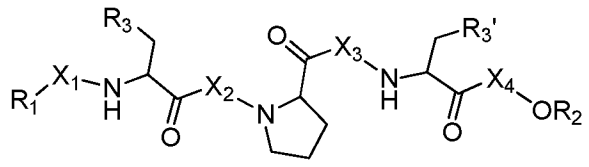
CLAIMS

1. A compound of the formula I:

I 

or a pharmaceutically acceptable salt or solvate thereof,

wherein

X_1 is absent or an amino acid residue forming a peptide bond with the -NH- group adjacent thereto;

X_2 is absent or an amino acid residue forming peptide bonds with the carbonyl group and nitrogen atom adjacent thereto;

X_3 is absent or an amino acid residue forming peptide bonds with the carbonyl and -NH- groups adjacent thereto;

X_4 is absent or an amino acid residue forming a peptide bond with the carbonyl group adjacent thereto;

R_1 is H, -CO(C_1-C_8)alkyl, -CO(C_1-C_8)alkylene-NH((C_1-C_8)alkyl, -CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$, covalently linked to the N atom of the amino moiety of X_1, when present;

R_2 is H or (C_1-C_8)alkyl, covalently linked to the carbonyl moiety of X_4, when present; and

R_3 and R_3' each independently is -SH, -S-CO(C_1-C_8)alkyl, -S-CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -S-CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$; or R_3 and R_3' are both -S- and together form a disulfide bond; or one of R_3 and R_3' is -S- forming a disulfide bond with one of R_3 and R_3' of another identical or different compound of the formula I, and another of R_3 and R_3' is -SH, -S-CO(C_1-C_8)alkyl, -S-CO(C_1-C_8)alkylene-N((C_1-C_8)alkyl)$_2$, or -S-CO(C_1-C_8)alkylene-N^+((C_1-C_8)alkyl)$_3$; or R_3 and R_3' are both -S-, wherein R_3 forms a disulfide bond with one of R_3 and R_3' of another identical or different compound of the formula I, and R_3' forms a disulfide bond with either another of R_3 and R_3' of said another compound of the formula I, or with one of R_3 and R_3' of a further identical or different compound of the formula I,

provided that at least one of R_1 and R_2 is not H, or at least one of R_3 and R_3' is not -SH, but excluding the compounds wherein R_3 and R_3' are each -SH, R_1 is -CO(C_1-C_8)alkyl, R_2 is H, and X_1, X_2, X_3 and X_4 are absent.

2. The compound of claim 1, wherein R_1 is H, -CO(C_1-C_4)alkyl, preferably -CO(C_1-C_2)alkyl, -CO(C_1-C_4)alkylene-NH((C_1-C_4)alkyl, preferably -CO(C_1-C_2)alkylene-NH((C_1-C_2)alkyl, -CO(C_1-C_4)alkylene-N((C_1-C_4)alkyl)$_2$, preferably -CO(C_1-C_2)alkylene-N((C_1-C_2)alkyl)$_2$, or -CO(C_1-C_4)alkylene-N^+((C_1-C_4)alkyl)$_3$, preferably -CO(C_1-C_2)alkylene-N^+((C_1-C_2)alkyl)$_3$.

3. The compound of claim 2, wherein R_1 is H, -CO-CH_2-N(CH_3)$_2$, or -CO-CH_2-N^+(CH_3)$_3$.

4. The compound of claim 1, wherein R_2 is (C_1-C_4)alkyl, preferably (C_1-C_2)alkyl, or H.

5. The compound of claim 4, wherein R_2 is ethyl or H.

6. The compound of claim 1, wherein R_3 and R_3' each independently is -SH, -S-CO(C_1-C_4)alkyl, preferably -S-CO(C_1-C_2)alkyl, -S-CO(C_1-C_4)alkylene-N((C_1-C_4)alkyl)$_2$, preferably -S-CO(C_1-C_2)alkylene-N((C_1-C_2)alkyl)$_2$, or -S-CO(C_1-C_4)alkylene-N^+((C_1-C_4)alkyl)$_3$, preferably -S-CO(C_1-C_2)alkylene-N^+((C_1-C_2)alkyl)$_3$; or R_3 and R_3' are both -S- and together form a disulfide bond.

7. The compound of claim 6, wherein R_3 and R_3' each independently is -SH, -S-$COCH_3$, -S-CO-CH_2-N(CH_3)$_2$, or -S-CO-CH_2-N^+(CH_3)$_3$; or R_3 and R_3' are both -S- and together form a disulfide bond.

8. The compound of claim 1, wherein one of R_3 and R_3' is -S- forming a disulfide bond with one of R_3 and R_3' of another compound of the formula I, and another of R_3 and R_3' is -SH, -S-CO(C_1-C_4)alkyl, preferably -S-CO(C_1-C_2)alkyl, -S-CO(C_1-C_4)alkylene-N((C_1-C_4)alkyl)$_2$, preferably -S-CO(C_1-C_2)alkylene-N((C_1-C_2)alkyl)$_2$, or -S-CO(C_1-C_4)alkylene-N^+((C_1-C_4)alkyl)$_3$, preferably -S-CO(C_1-C_2)alkylene-N^+((C_1-C_2)alkyl)$_3$; or R_3 and R_3' are both -S-, wherein R_3 forms a disulfide bond with one of R_3 and R_3' of another compound of the formula I, and R_3' forms a disulfide bond with either another of R_3 and

R_3' of said another compound of the formula I, or with one of R_3 and R_3' of a further compound of the formula I.

9. The compound of claim 8, wherein one of R_3 and R_3' is -S-, and another of R_3 and R_3' is -SH, -S-$COCH_3$, -S-CO-CH_2-$N(CH_3)_2$, or -S-CO-CH_2-$N^+(CH_3)_3$; or R_3 and R_3' are both -S-.

10. The compound of claim 1, wherein X_1, X_2, X_3 and X_4 are absent, or at least one of X_1, X_2, X_3 and X_4 is present.

11. The compound of claim 10, wherein X_1, X_2 and X_3 are absent, and X_4 is a glycine residue; X_1, X_2 and X_4 are absent, and X_3 is a glycine residue; X_1, X_3 and X_4 are absent, and X_2 is a glycine residue; or X_2, X_3 and X_4 are absent, and X_1 is a glycine residue; X_1 and X_2 are absent, and X_3 and X_4 each is a glycine residue; X_1 and X_3 are absent, and X_2 and X_4 each is a glycine residue; X_1 and X_4 are absent, and X_2 and X_3 each is a glycine residue; X_2 and X_3 are absent, and X_1 and X_4 each is a glycine residue; X_2 and X_4 are absent, and X_1 and X_3 each is a glycine residue; X_3 and X_4 are absent, and X_1 and X_2 each is a glycine residue; X_1 is absent, and X_2, X_3 and X_4 each is a glycine residue; X_2 is absent, and X_1, X_3 and X_4 each is a glycine residue; X_3 is absent, and X_1, X_2 and X_4 each is a glycine residue; X_4 is absent, and X_1, X_2 and X_3 each is a glycine residue; or X_1, X_2, X_3 and X_4 each is a glycine residue.

12. The compound of any one of claims 1 to 7, 10 or 11, wherein R_1 is H, -CO(C_1-C_4)alkyl, preferably -CO(C_1-C_2)alkyl, -CO(C_1-C_4)alkylene-NH((C_1-C_4)alkyl, preferably -CO(C_1-C_2)alkylene-NH((C_1-C_2)alkyl, -CO(C_1-C_4)alkylene-N((C_1-C_4)alkyl)$_2$, preferably -CO(C_1-C_2)alkylene-N((C_1-C_2)alkyl)$_2$, or -CO(C_1-C_4)alkylene-N^+((C_1-C_4)alkyl)$_3$, preferably -CO(C_1-C_2)alkylene-N^+((C_1-C_2)alkyl)$_3$; R_2 is (C_1-C_4)alkyl, preferably (C_1-C_2)alkyl, or H; R_3 and R_3' each independently is -SH, -S-CO(C_1-C_4)alkyl, preferably -S-CO(C_1-C_2)alkyl, -S-CO(C_1-C_4)alkylene-N((C_1-C_4)alkyl)$_2$, preferably -S-CO(C_1-C_2)alkylene-N((C_1-C_2)alkyl)$_2$, or -S-CO(C_1-C_4)alkylene-N^+((C_1-C_4)alkyl)$_3$, preferably -S-CO(C_1-C_2)alkylene-N^+((C_1-C_2)alkyl)$_3$, or R_3 and R_3' are both -S- and together form a disulfide bond; and X_1, X_2, X_3 and X_4 are absent, or at least one of X_1, X_2, X_3 and X_4 is present.

13. The compound of claim 12, wherein R_1 is H, -CO-CH_2-$N(CH_3)_2$, or -CO-CH_2-$N^+(CH_3)_3$; R_2 is H or ethyl; and R_3 and R_3' each independently is -SH, -S-$COCH_3$, -S-CO-

CH_2-$N(CH_3)_2$, or -S-CO-CH_2-$N^+(CH_3)_3$, or R_3 and R_3' are both -S- and together form a disulfide bond.

14. The compound of claim 13, wherein at least one of X_1, X_2, X_3 and X_4 is a glycine residue, and the other of X_1, X_2, X_3 and X_4 are absent.

15. The compound of claim 13, wherein X_1, X_2, X_3 and X_4 are absent and: (i) R_1 is H, R_2 is ethyl, R_3 is -SH, and R_3' is -SH, herein identified compound **1**; (ii) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is H, R_3 is -SH, and R_3' is -SH, herein identified compound **2**; (iii) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, R_3 is -SH, and R_3' is -SH, herein identified compound **3**; (iv) R_1 is H, R_2 is H, R_3 is -S-$COCH_3$, and R_3' is -S-$COCH_3$, herein identified compound **4**; (v) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, R_3 is -S-$COCH_3$, and R_3' is -S-$COCH_3$, herein identified compound **5**; (vi) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is H, R_3 is -S-$COCH_3$, and R_3' is -S-$COCH_3$, herein identified compound **6**; (vii) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, R_3 is -S-$CO(CH_2)$-$N(CH_3)_2$, and R_3' is -S-$CO(CH_2)$-$N(CH_3)_2$, herein identified compound **7**; (viii) R_1 is -$CO(CH_2)$-$N^+(CH_3)_3$, R_2 is ethyl, R_3 is -S-$CO(CH_2)$-$N^+(CH_3)_3$, and R_3' is -S-$CO(CH_2)$- $N^+(CH_3)_3$, herein identified compound **8**; or (ix) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, and R_3 and R_3' are both -S- and together form a disulfide bond, herein identified compound **9**.

16. The compound of claim 15, wherein R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, R_3 is -S-$COCH_3$, and R_3' is -S-$COCH_3$, herein identified compound **5**.

17. The compound of claim 14, wherein X_2 is a glycine residue; X_1, X_3 and X_4 are absent; and: (i) R_1 is H, R_2 is ethyl, R_3 is -SH, and R_3' is -SH, herein identified compound **10**; (ii) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is H, R_3 is -SH, and R_3' is -SH, herein identified compound **11**; (iii) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, R_3 is -SH, and R_3' is -SH, herein identified compound **12**; (iv) R_1 is H, R_2 is H, R_3 is -S-$COCH_3$, and R_3' is -S-$COCH_3$, herein identified compound **13**; (v) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, R_3 is -S-$COCH_3$, and R_3' is -S-$COCH_3$, herein identified compound **14**; (vi) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is H, R_3 is -S-$COCH_3$, and R_3' is -S-$COCH_3$, herein identified compound **15**; (vii) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, R_3 is -S-$CO(CH_2)$-$N(CH_3)_2$, and R_3' is -S-$CO(CH_2)$-$N(CH_3)_2$, herein identified compound **16**; (viii) R_1 is -$CO(CH_2)$-$N^+(CH_3)_3$, R_2 is ethyl, R_3 is -S-$CO(CH_2)$-$N^+(CH_3)_3$, and R_3' is -S-$CO(CH_2)$- $N^+(CH_3)_3$, herein identified compound **17**;

or (ix) R_1 is -$CO(CH_2)$-$N(CH_3)_2$, R_2 is ethyl, and R_3 and R_3' are both -S- and together form a disulfide bond, herein identified compound **18**.

18. The compound of any one of claims 1 to 5, or 8 to 11, wherein R_1 is H, -$CO(C_1$-$C_4)$alkyl, preferably -$CO(C_1$-$C_2)$alkyl, -$CO(C_1$-$C_4)$alkylene-$NH((C_1$-$C_4)$alkyl, preferably -$CO(C_1$-$C_2)$alkylene-$NH((C_1$-$C_2)$alkyl, -$CO(C_1$-$C_4)$alkylene-$N((C_1$-$C_4)$alkyl$)_2$, preferably -$CO(C_1$-$C_2)$alkylene-$N((C_1$-$C_2)$alkyl$)_2$, or -$CO(C_1$-$C_4)$alkylene-$N^+((C_1$-$C_4)$alkyl$)_3$, preferably -$CO(C_1$-$C_2)$alkylene-$N^+((C_1$-$C_2)$alkyl$)_3$; R_2 is $(C_1$-$C_4)$alkyl, preferably $(C_1$-$C_2)$alkyl, or H; one of R_3 and R_3' is -S- forming a disulfide bond with one of R_3 and R_3' of another compound of the formula I, and another of R_3 and R_3' is -SH, -S-$CO(C_1$-$C_4)$alkyl, preferably -S-$CO(C_1$-$C_2)$alkyl, -S-$CO(C_1$-$C_4)$alkylene-$N((C_1$-$C_4)$alkyl$)_2$, preferably -S-$CO(C_1$-$C_2)$alkylene-$N((C_1$-$C_2)$alkyl$)_2$, or -S-$CO(C_1$-$C_4)$alkylene-$N^+((C_1$-$C_4)$alkyl$)_3$, preferably -S-$CO(C_1$-$C_2)$alkylene-$N^+((C_1$-$C_2)$alkyl$)_3$, or R_3 and R_3' are both -S-, wherein R_3 forms a disulfide bond with one of R_3 and R_3' of another compound of the formula I, and R_3' forms a disulfide bond with either another of R_3 and R_3' of said another compound of the formula I, or with one of R_3 and R_3' of a further compound of the formula I; and X_1, X_2, X_3 and X_4 are absent, or at least one of X_1, X_2, X_3 and X_4 is present.

19. The compound of claim 18, wherein R_1 is H, -CO-CH_2-$N(CH_3)_2$, or -CO-CH_2-$N^+(CH_3)_3$; R_2 is H or ethyl; and either one of R_3 and R_3' is -S-, and another of R_3 and R_3' is -SH, -S-$COCH_3$, -S-CO-CH_2-$N(CH_3)_2$, or -S-CO-CH_2-$N^+(CH_3)_3$, or R_3 and R_3' are both -S-.

20. The compound of claim 19, wherein at least one of X_1, X_2, X_3 and X_4 is a glycine residue, and the other of X_1, X_2, X_3 and X_4 are absent.

21. A pharmaceutical composition comprising a compound of any one of claims 1 to 20, preferably selected from the group consisting of the herein identified compounds **1-18**, or a pharmaceutically acceptable salt or solvate thereof, and a pharmaceutically acceptable carrier.

22. The pharmaceutical composition of claim 21, for intravenous, intraarterial, intramuscular, intraperitoneal, intrathecal, intrapleural, intratracheal, subcutaneous, topical, inhalational, or oral administration.

23. The pharmaceutical composition of claim 21 or 22, for prevention, treatment or management of a disease, disorder or condition mediated by redox stress.

24. The pharmaceutical composition of claim 23, wherein said disease, disorder or condition mediated by redox stress is chlorine inhalational lung injury, phosgene inhalational lung injury, hydrogen sulfide inhalational lung injury, ventilator-induced lung injury, lung ischemia reperfusion injury, chronic obstructive pulmonary disease, bronchopulmonary dysplasia, adult respiratory distress syndrome, radiation induced lung fibrosis, congestive heart failure, myocardial infarction, myocardial ischemia-reperfusion injury, stroke, cardiopulmonary bypass surgery, doxorubicin-induced cardiomyopathy, limb ischemia reperfusion injury, renal ischemia repercussion injury, contrast media-induced nephropathy, hemorrhagic shock, endotoxic shock, septic shock, hepatic reperfusion injury, primary lung graft dysfunction, lung transplantation, renal transplantation, burn injury, angioplasty, traumatic brain injury, Parkinson's disease, ischemic bowel disease, mesenteric ischemia, retinopathy of prematurity, retinal degenerative disease, glaucoma, acute macular degeneration, cataracts, Crohn's disease, or vitiligo.

25. A compound of any one of claims 1 to 20, preferably selected from the group consisting of the herein identified compounds **1-18**, or a pharmaceutically acceptable salt or solvate thereof, for use in prevention, treatment or management of a disease, disorder or condition mediated by redox stress.

26. Use of a compound of any one of claims 1 to 20, preferably selected from the group consisting of the herein identified compounds **1-18**, or a pharmaceutically acceptable salt or solvate thereof, for the preparation of a pharmaceutical composition for prevention, treatment or management of a disease, disorder or condition mediated by redox stress.

27. A method for prevention, treatment or management of a disease, disorder or condition mediated by redox stress in an individual in need thereof, said method comprising administering to said individual an effective amount of a compound of any one of claims 1 to 20, preferably selected from the group consisting of the herein identified compounds **1-18**, or a pharmaceutically acceptable salt or solvate thereof.

Fig. 1

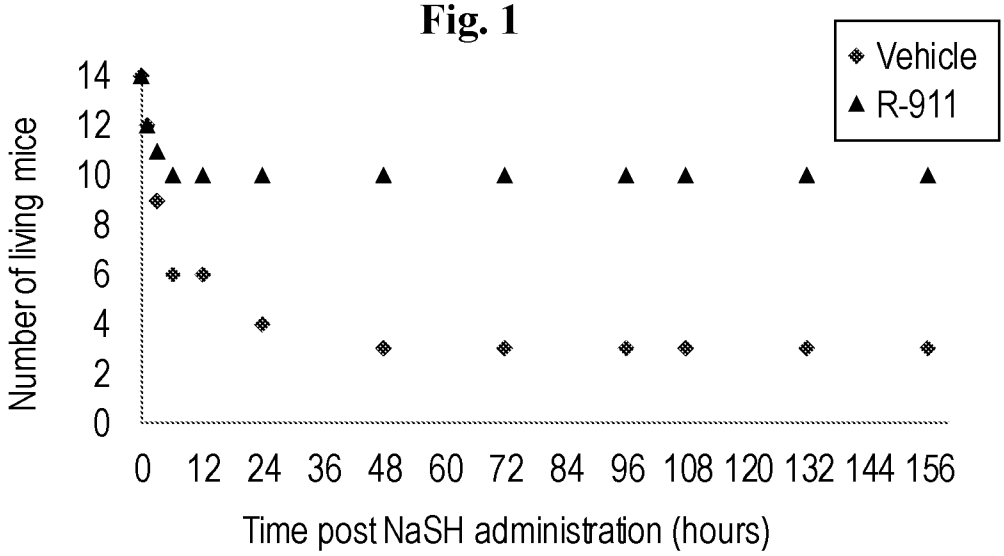


Fig. 2A

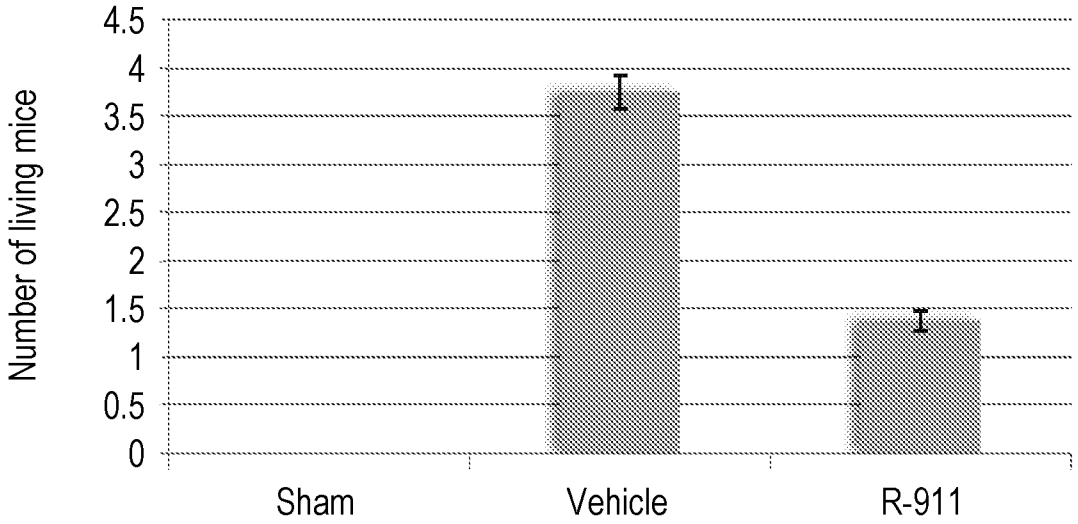


Fig. 2B

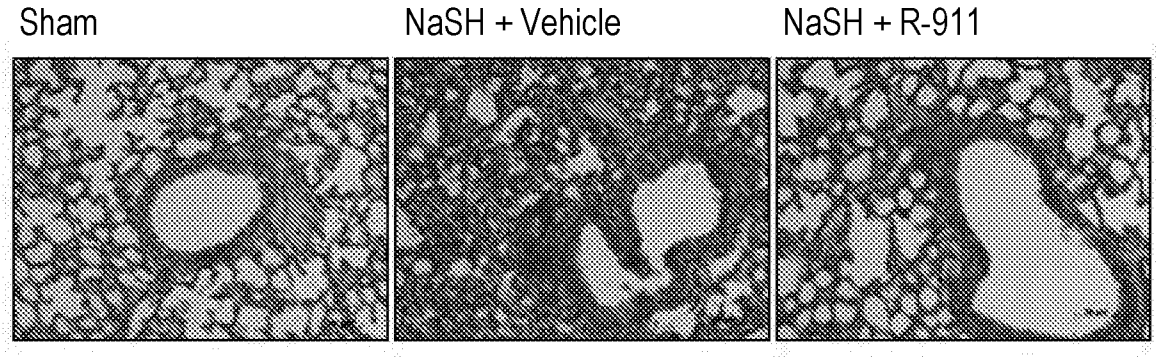


Fig. 3

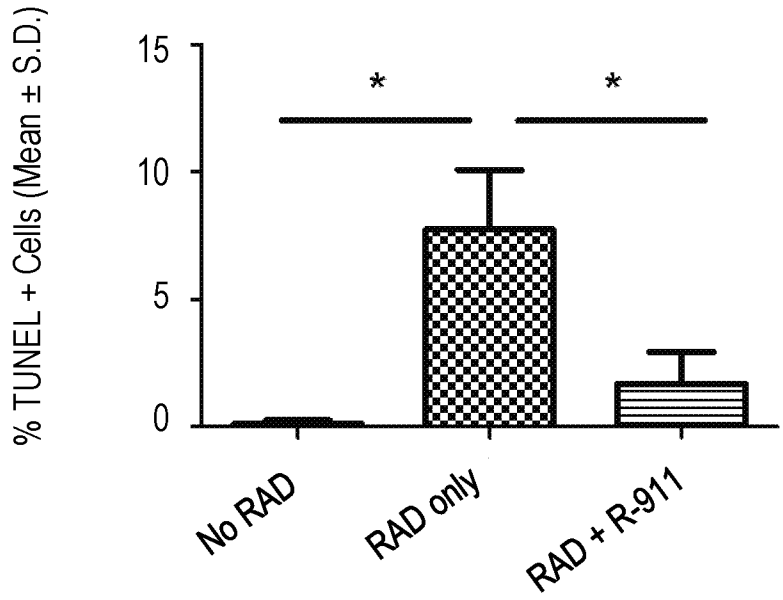

% TUNEL + Cells (Mean ± S.D.)
15
10
5
0
*
*
No RAD
RAD only
RAD + R-911

Fig. 4

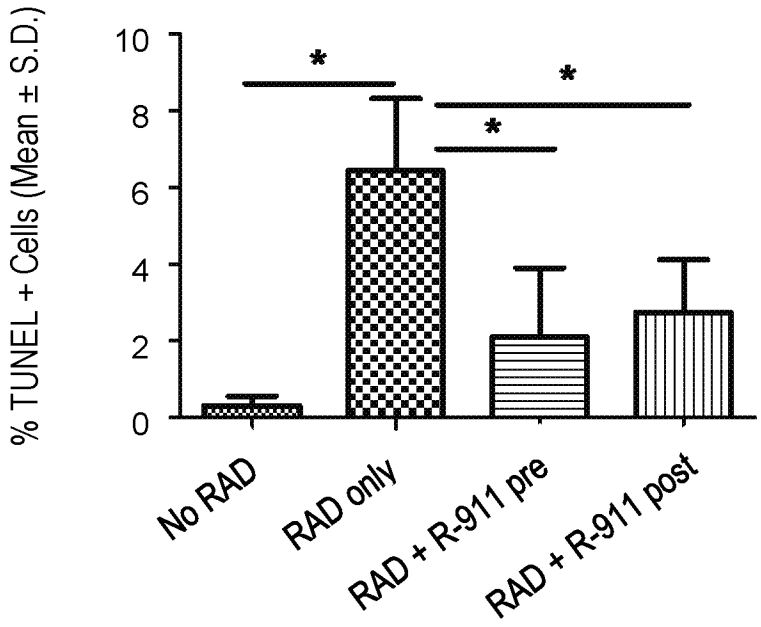

% TUNEL + Cells (Mean ± S.D.)
10
8
6
4
2
0
*
*
*
No RAD
RAD only
RAD + R-911 pre
RAD + R-911 post

Fig. 5

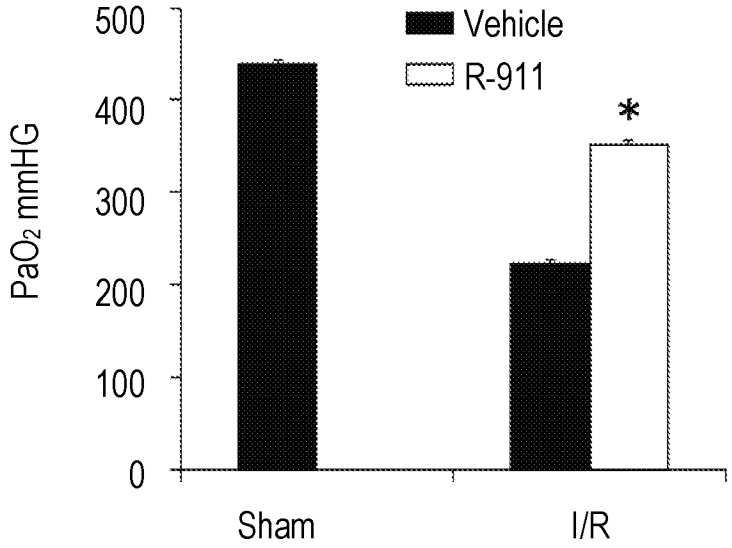


Fig. 6A

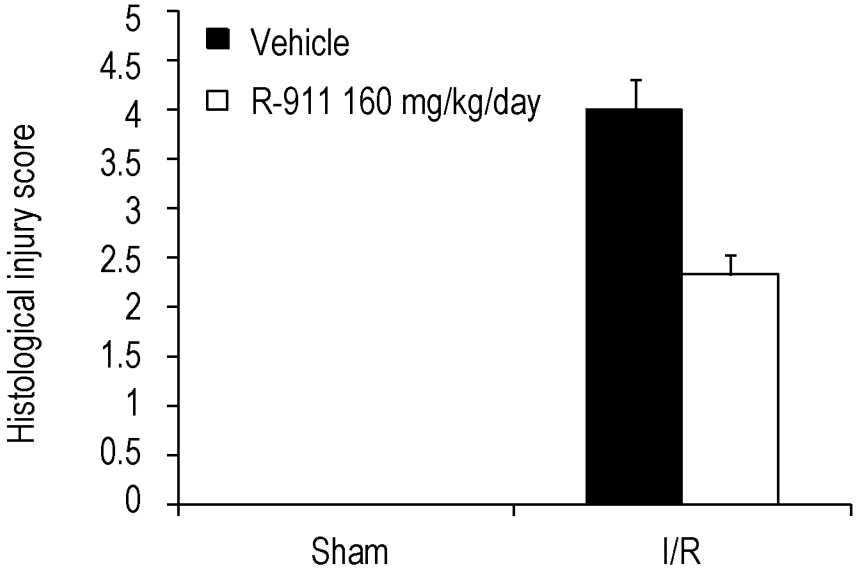


Fig. 6B

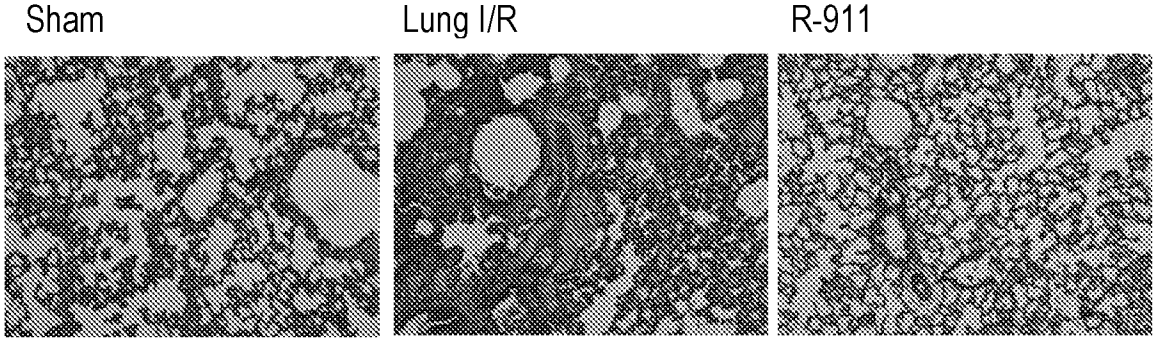


Fig. 7A

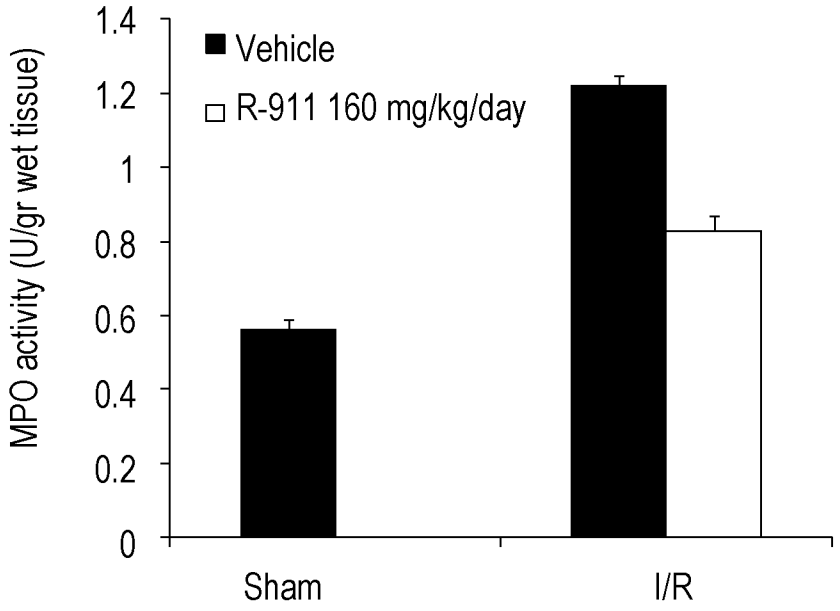

MPO activity (U/gr wet tissue)
1.4
1.2
1
0.8
0.6
0.4
0.2
0
Vehicle
R-911 160 mg/kg/day
Sham
I/R

Fig. 7B

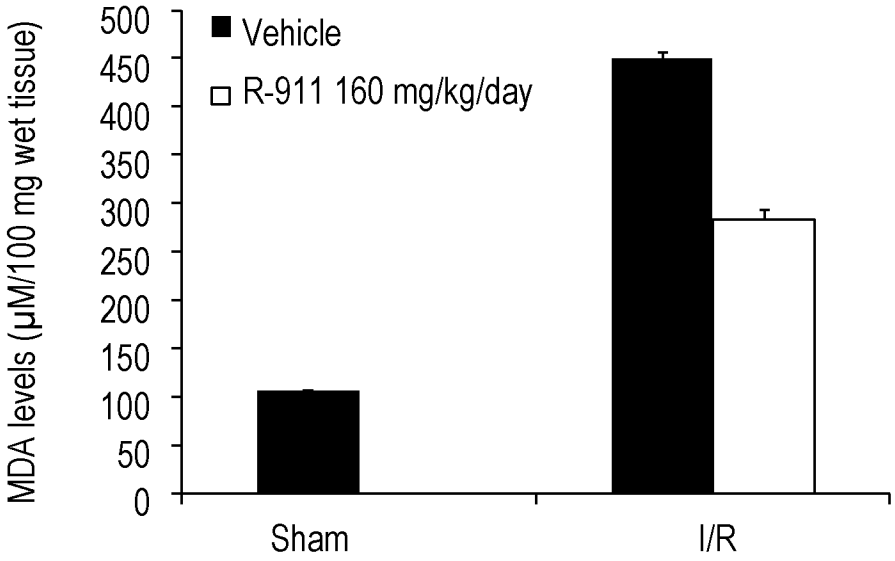

MDA levels (µM/100 mg wet tissue)
500
450
400
350
300
250
200
150
100
50
0
Vehicle
R-911 160 mg/kg/day
Sham
I/R

Fig. 8

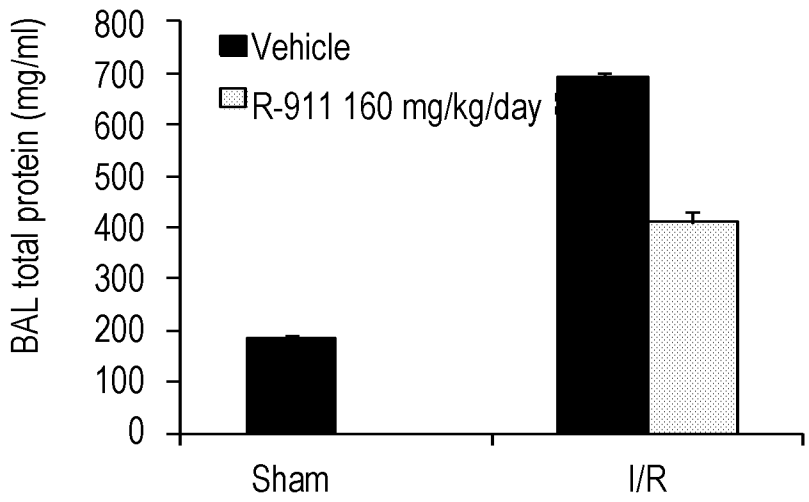

BAL total protein (mg/ml)
800
700
600
500
400
300
200
100
0
Vehicle
R-911 160 mg/kg/day
Sham
I/R

Fig. 9

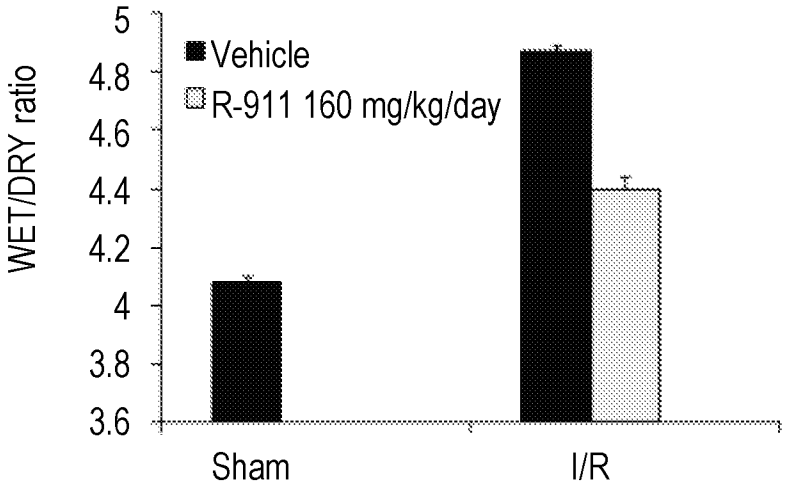

WET/DRY ratio
5
4.8
4.6
4.4
4.2
4
3.8
3.6
Vehicle
R-911 160 mg/kg/day
Sham
I/R

Fig. 10A

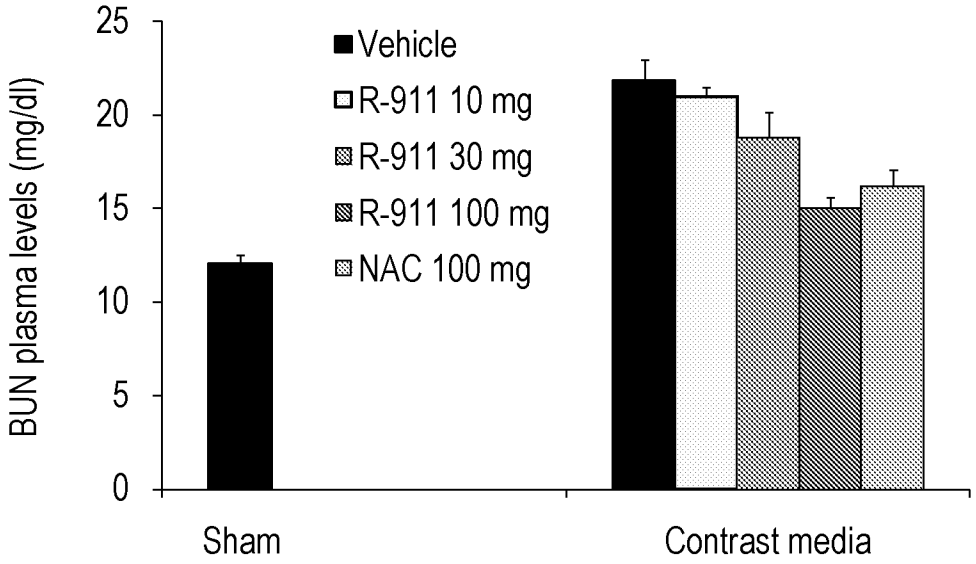


Fig. 10B

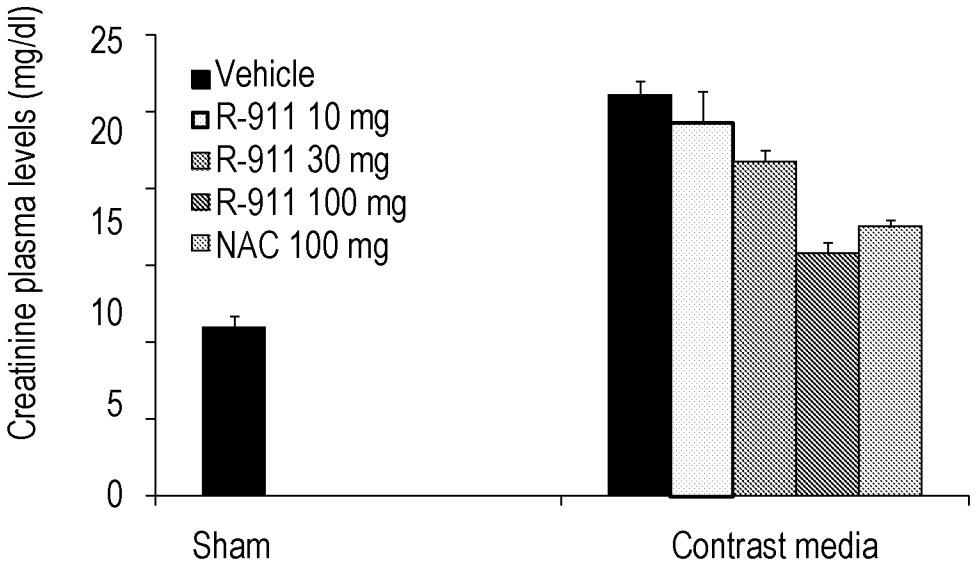


Fig. 11A

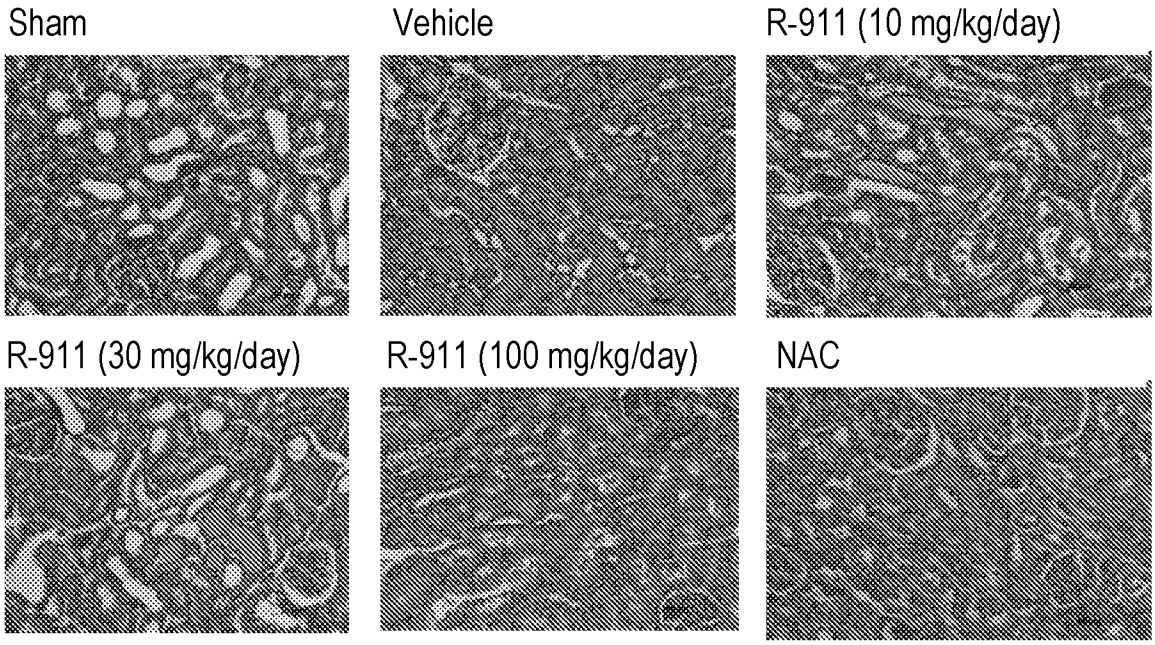


Fig. 11B

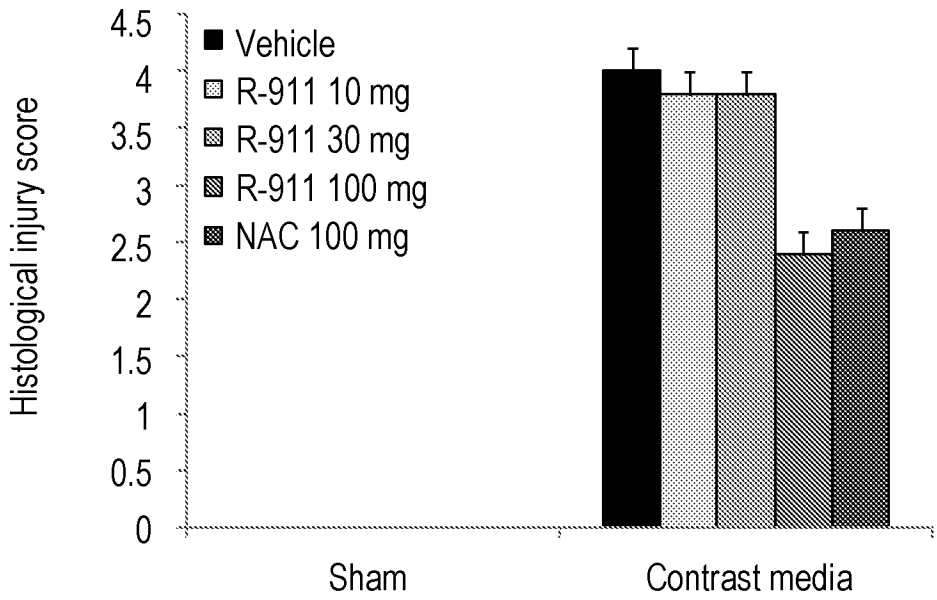


Fig. 12A

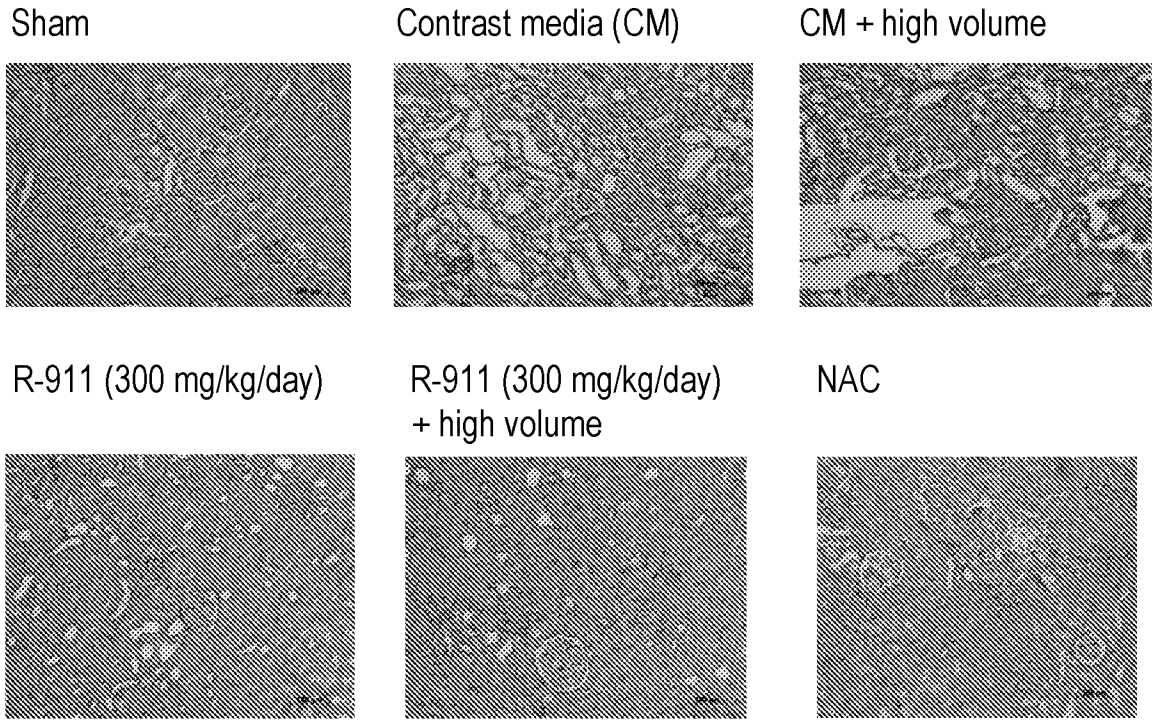


Fig. 12B

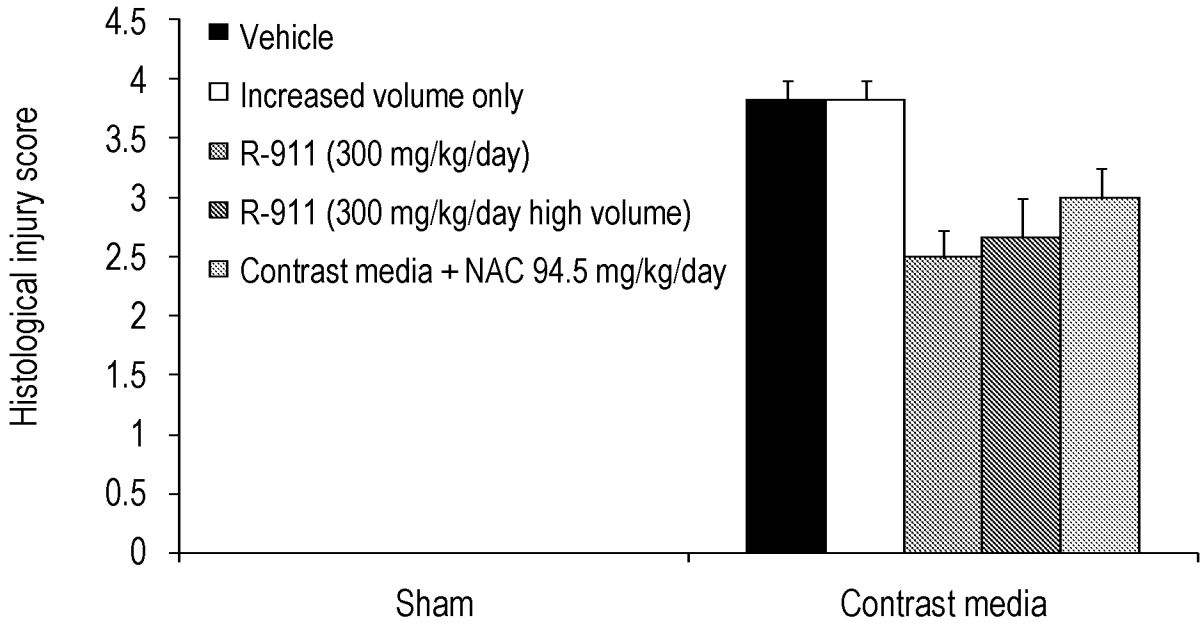


Fig. 13A

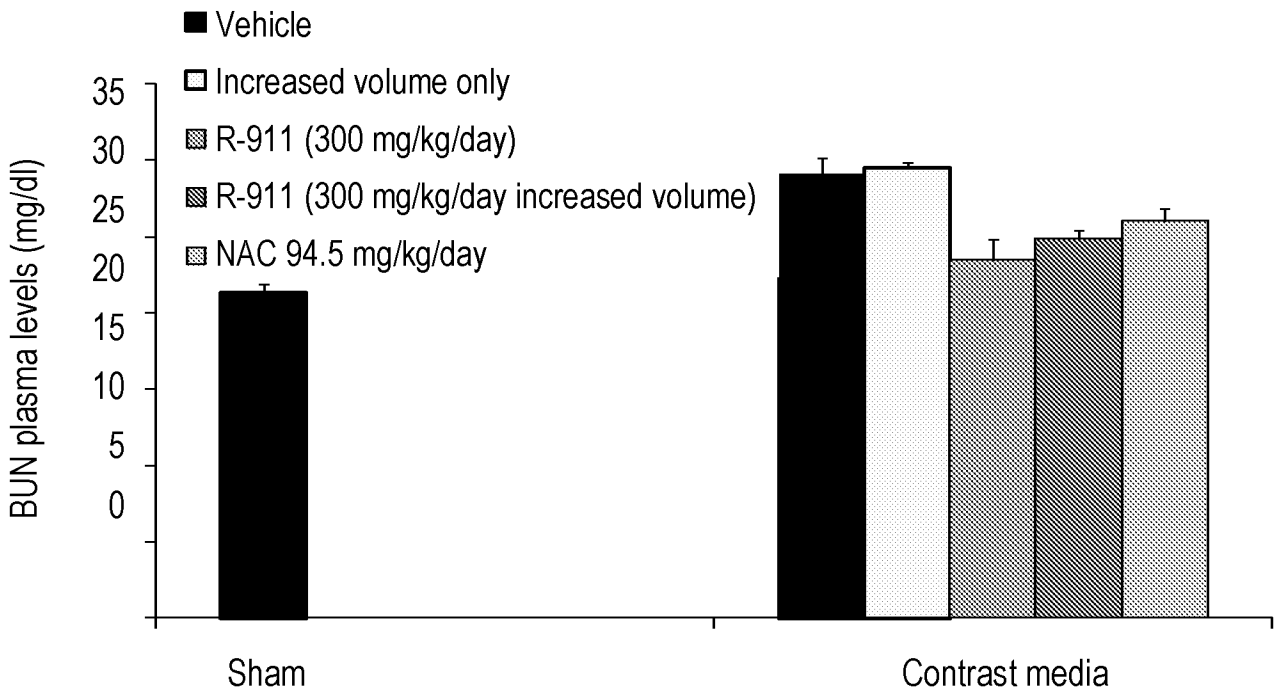


Fig. 13B

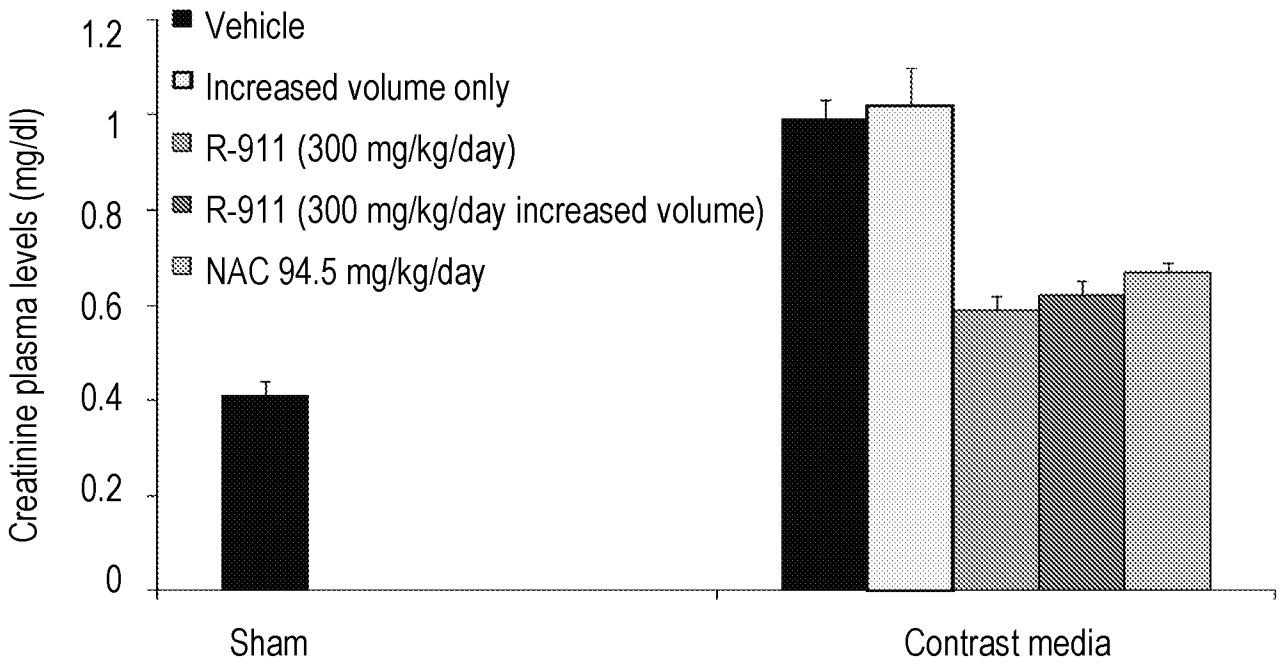


Fig. 14A

Fig. 14B

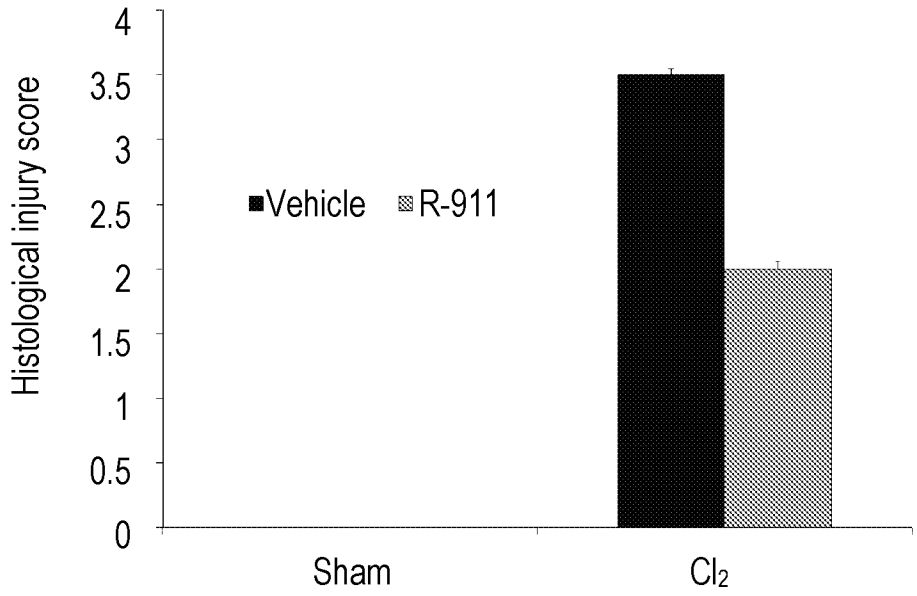


Fig. 15

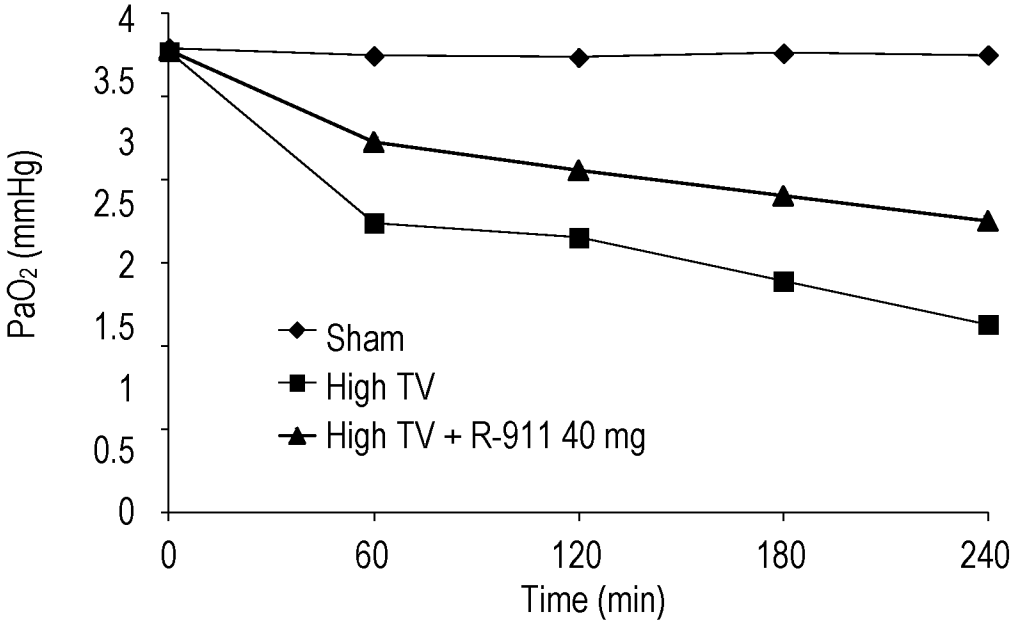


Fig. 16A

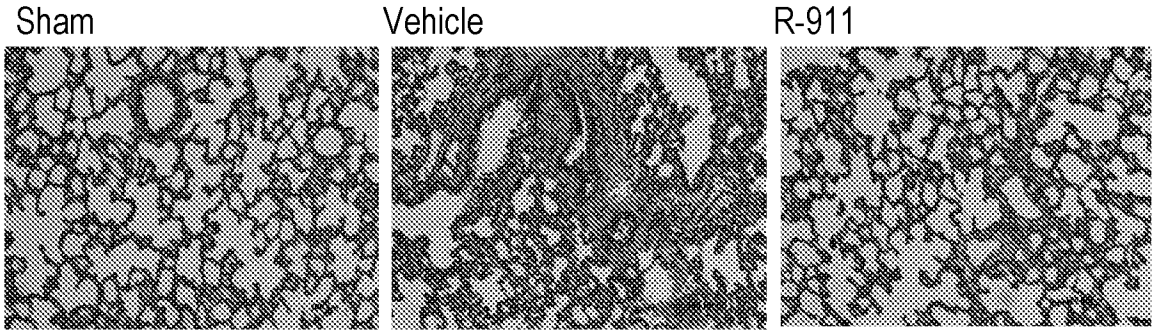


Fig. 16B

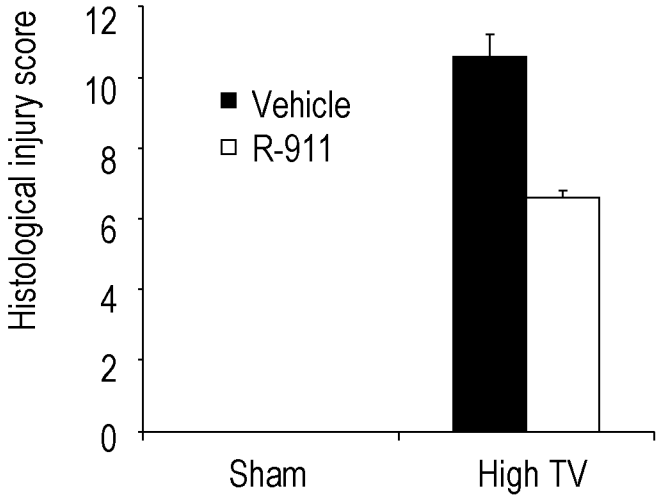


Fig. 17

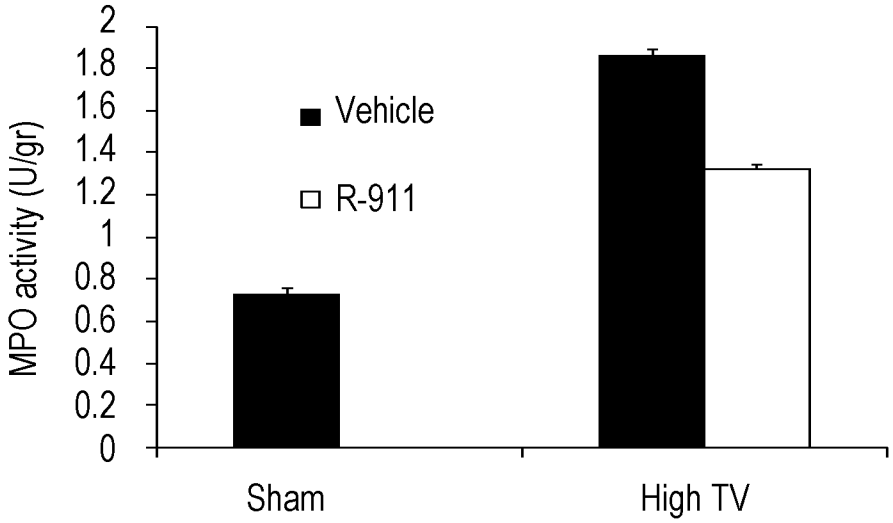


Fig. 18A

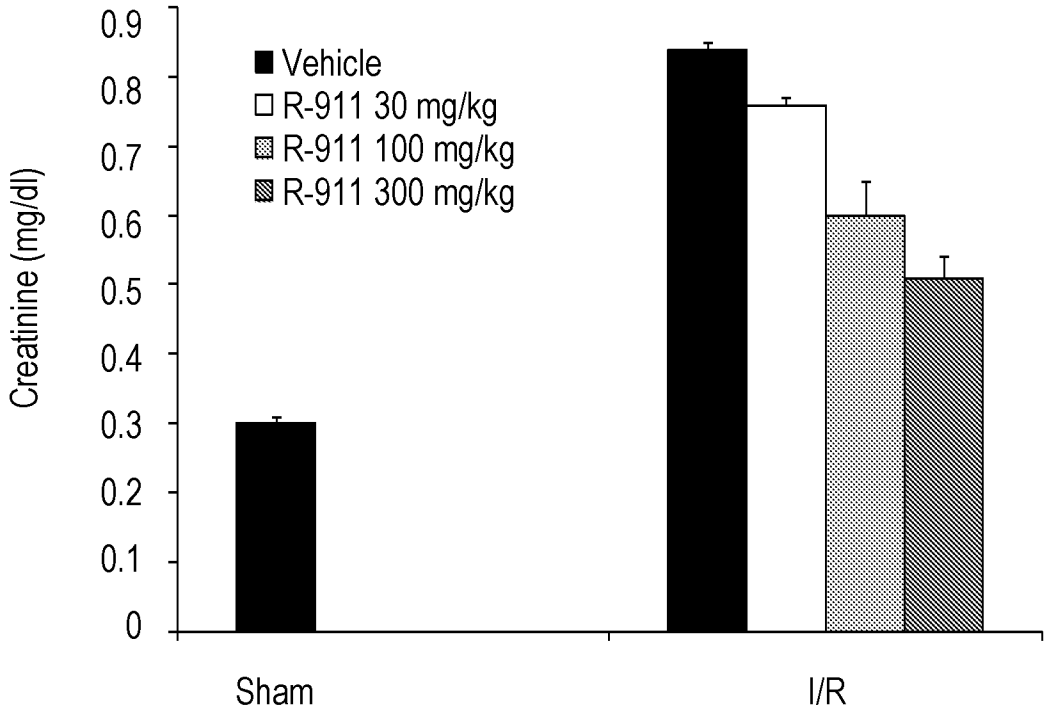


Fig. 18B

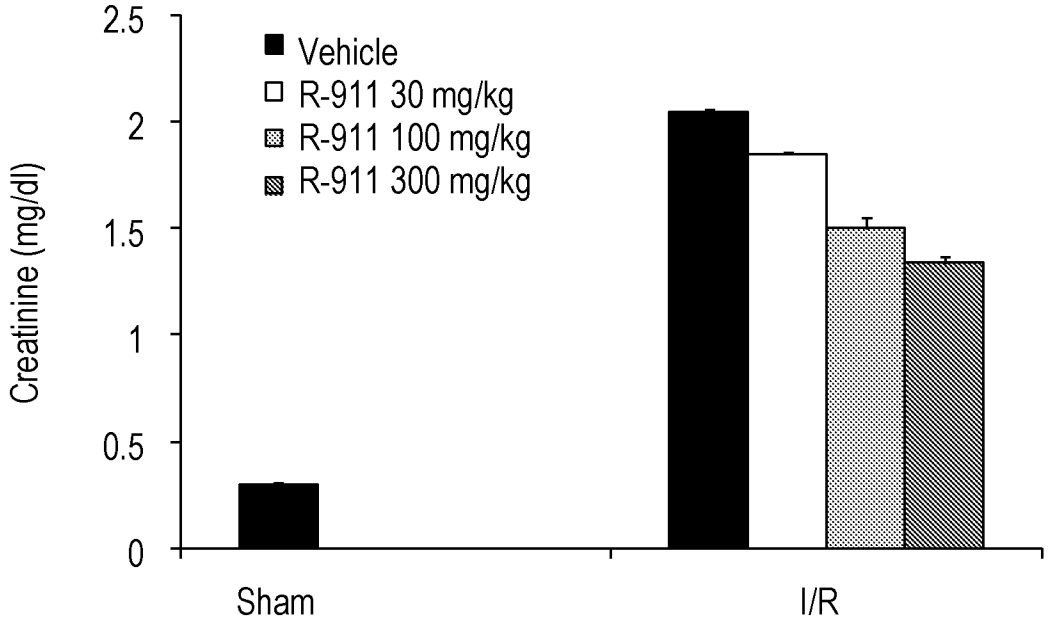


Fig. 19A

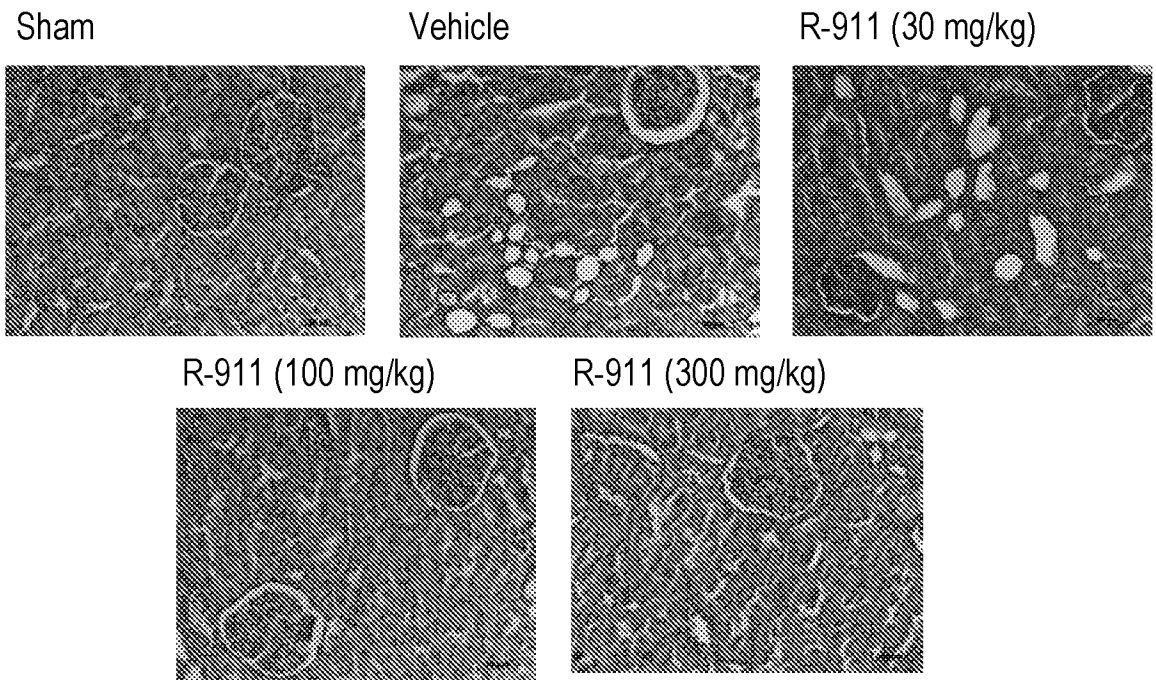


Fig. 19B

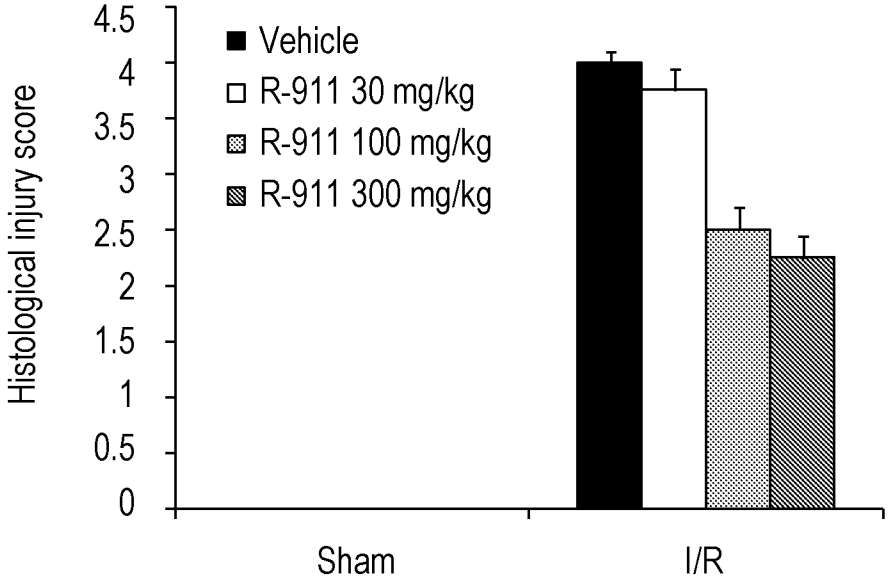


Fig. 20A

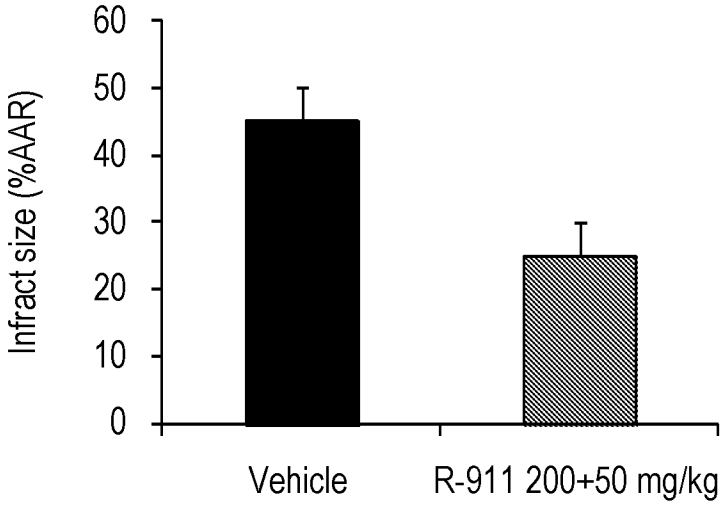


Fig. 20B

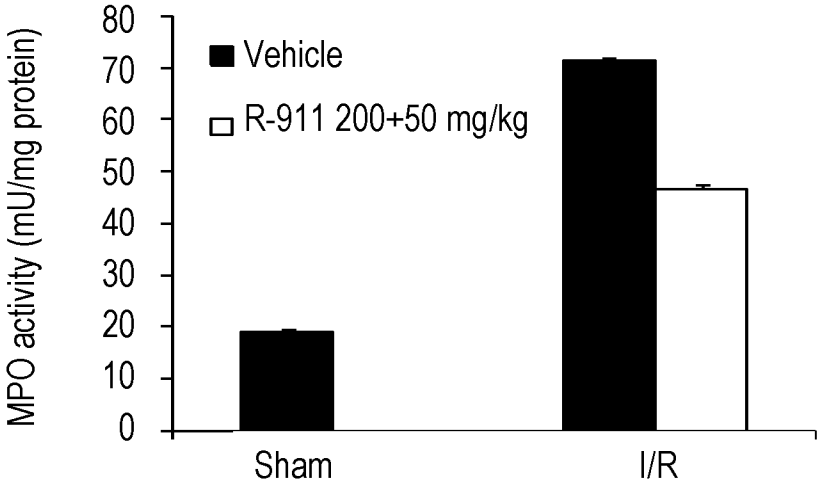


Fig. 20C

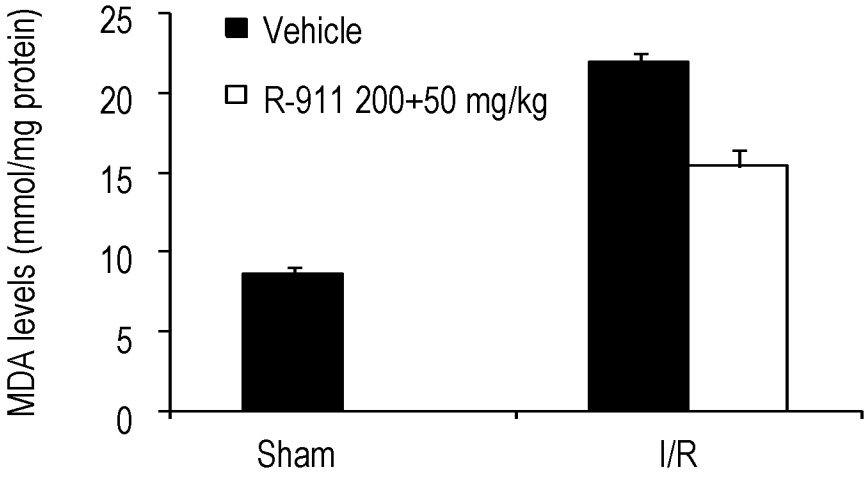


Fig. 21A

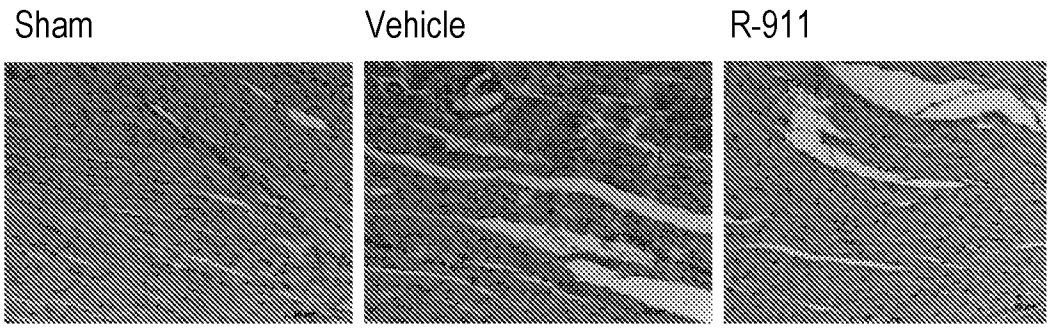
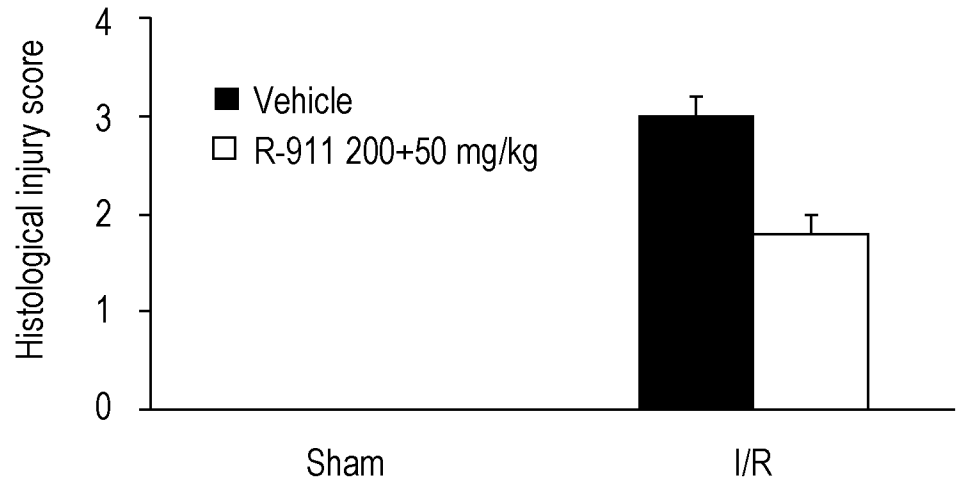


Fig. 21B



INTERNATIONAL SEARCH REPORT

International application No
PCT/IB2015/055307

A. CLASSIFICATION OF SUBJECT MATTER
INV. C07K5/083 C07K7/06 C07K5/103 A61P11/00 A61P25/16
A61P9/10 A61P27/00 A61P1/00 A61P13/12
ADD.
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, WPI Data, BIOSIS, CHEM ABS Data, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	CASTANEDO G M ET AL: "Solid-Phase synthesis of dual alpha4beta1/alpha4beta7 Integrin antagonists: TWO SCAFFOLDS WITH OVERLAPPING PHARMACOPHORES", BIOORGANIC & MEDICINAL CHEMISTRY LETTERS, PERGAMON, AMSTERDAM, NL, vol. 12, 1 October 2002 (2002-10-01), pages 2913-2917, XP002250476, ISSN: 0960-894X, DOI: 10.1016/S0960-894X(02)00597-8 compound 1 ----- -/--	1,2,4-7, 10,12, 21,22

[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

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Date of the actual completion of the international search	Date of mailing of the international search report
24 November 2015	07/12/2015

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Jetter, Sonya

INTERNATIONAL SEARCH REPORT

International application No
PCT/IB2015/055307

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
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X	JACKSON D Y ET AL: "Potent alpha4beta1 peptide antagonists as potential anti-inflammatory agents", JOURNAL OF MEDICINAL CHEMISTRY, AMERICAN CHEMICAL SOCIETY, US, vol. 40, no. 21, 1 January 1997 (1997-01-01), pages 3359-3368, XP002148351, ISSN: 0022-2623, DOI: 10.1021/JM970175S compounds 1, 2, 3, 11, 16-34	1-7,10, 12,13, 21,22

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A	figure 1	8,9, 15-20

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INTERNATIONAL SEARCH REPORT

International application No
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C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2013/190497 A2 (RADIKAL THERAPEUTICS INC [US]) 27 December 2013 (2013-12-27)	1-7,10, 12,13, 21-27
A	claims 1-3, 11-13, 41; compounds 25, 26	8,9, 15-20

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/IB2015/055307

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2013190497 A2	27-12-2013	NONE	