

increased residence time greater than noncross-linked oxyhemoglobin, and cross-linked with a covalent cross-linking agent selected from the group consisting of heterocyclic triazines, bis(diazobenzidines), halogenated aromatics, cycloalkanes having at least two cross-linking moieties, dialdehydes, glutaredehyde, divinyl sulfone, diisocyanates and diepoxides, and wherein the quantity transfused is an effective amount for increasing the volume of the system.

19. A method for the treatment of shock which method comprises administering into the circulatory system of an animal in shock, a blood plasma substitute comprising intramolecularly cross-linked, stromal-free oxyhemoglobin, having a molecular weight of about 64,500, cross-linked with a covalent cross-linking agent selected from the group consisting of heterocyclic triazines, bis(diazobenzidines), halogenated aromatic, cycloalkanes having at least two cross-linking moieties, dialdehydes, glutaraldehyde, divinyl sulfone, diisocyanates, and diepoxides, and wherein the hemoglobin is mixed with an isotonic physiologically acceptable carrier and is administered in an effective amount for alleviating said shock.

20. A method for supplying oxygen (a) to animal tissues and organs, and (b) for maintaining isolated animal organs in a viable state, which method comprises, (c) transfusing an effective amount of intramolecularly cross-linked stromal-free oxyhemoglobin into the circulatory system of an animal for supplying oxygen to said tissues and organs, (d) perfusing the isolated organ with an effective amount of a perfusate comprising intramolecularly cross-linked stromal-free oxyhemoglobin, and wherein the oxyhemoglobin has a molecular weight of

about 64,500 and is mixed with a pharmaceutically acceptable, liquid carrier.

21. A method for increasing the shelf-life and in vivo life of oxyhemoglobin which method comprises the steps of separating erythrocytes from whole blood, lysing the erythrocytes to disrupt the cellular wall of the erythrocytes, isolating oxyhemoglobin from the lysed erythrocytes substantially free of cellular wall material and stroma, cross-linking the stromal-free oxyhemoglobin with a polyfunctional, covalent, cross-linking agent in an oxygen containing environment to form intramolecularly cross-linked, stromal-free oxyhemoglobin, soluble in aqueous and physiological fluids, capable of binding oxygen and releasing it in vivo, having a molecular weight of 64,500, and wherein said intramolecularly cross-linked, stromal-free oxyhemoglobin has an increased shelf-life an increased in vivo persistence in an animal at least twice that of noncross-linked oxyhemoglobin, and has an osmolarity substantially equal to whole blood.

22. Intramolecularly cross-linked, stromal-free oxyhemoglobin formed by cross-linking oxyhemoglobin with a covalent cross-linking agent selected from the group consisting of heterocyclic triazines, halogenated aromatic cycloalkanes having at least two cross-linking sites, dialdehydes, glutaraldehyde, divinyl sulfone, diisocyanates and diepoxides, said oxyhemoglobin having an increased intravascular peristance, a molecular weight of about 64,500, soluble in aqueous and physiological fluids, capable of reversibly binding a gaseous ligand, and having a partial oxygen pressure at half-saturation, P<sub>50</sub>, of at least 2.5 mm Hg at physiological temperature and physiological pH.

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