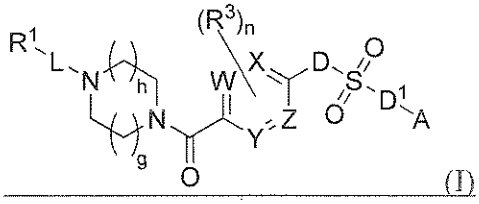
chronic transfusions and/or splenectomy at early development or during physiological stress or serious infections. Most affected individuals who are asymptomatic, paradoxically due to enhanced oxygen-transfer capacity, do not require any treatment. However, for some of the most severe cases, while extremely rare population-wise with estimated prevalence of 51 per million (Beutler, E. *Blood* **2000,** *95* (11), 3585-8), there is no disease-modifying treatment available for these patients other than palliative care (Tavazzi, D. et al., *Pediatr Ann* **2008,** *37* (5), 303-10). These hereditary non-spherocytic hemolytic anemia (HNSHA) patients present a clear unmet medical need.

Heterogenous genetic mutations in PKR lead to dysregulation of its catalytic activity. Since the initial cloning of PKR and report of a single point mutation Thr^{384}>Met associated with a HNSHA patient (Kanno, H. et al., *Proc Natl Acad Sci U S A* **1991,** *88* (18), 8218-21), there are now nearly 200 different reported mutations associated with this disease reported worldwide (Zanella, A. et al., *Br J Haematol* **2005,** *130* (1), 11-25; Kedar, P., et al., *Clin Genet* **2009,** *75* (2), 157-62; Fermo, E. et al., *Br J Haematol* **2005,** *129* (6), 839-46; Pissard, S. et al., *Br J Haematol* **2006,** *133* (6), 683-9). Although these mutations represent wide range genetic lesions that include deletional and transcriptional or translational abnormalities, by far the most common type is missense mutation in the coding region that one way or another affects conserved residues within domains that are structurally important for optimal catalytic function of PKR. The pattern of mutation prevalence seems to be unevenly distributed toward specific ethnic backgrounds. For instance, the most frequent codon substitutions reported for North American and European patients appear to be Arg^{486}>Trp and Arg^{510}>Gln, while mutations Arg^{479}>His, Arg^{490}>Trp and Asp^{331}>Gly were more frequently found in Asian patients (Kedar, P., et al., *Clin Genet* **2009,** *75* (2), 157-62).

SUMMARY

In one aspect, there is provided use of a compound in the manufacture of a medicament for increasing the lifetime of red blood cells (RBCs) comprising an

effective amount of (1) a compound of formula I or a pharmaceutically acceptable salt thereof; (2) a pharmaceutically acceptable composition comprising a compound of formula I or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier,



(I)

wherein:

W, X, Y and Z are each independently selected from CH or N;

D and D^1 are independently selected from a bond or NR^b;

A is optionally substituted aryl or optionally substituted heteroaryl;

L is a bond, -C(O)-, $-(CR^cR^c)_m$-, -OC(O)-, $-(CR^cR^c)_m$-OC(O)-, $-(CR^cR^c)_m$-C(O)-, $-NR^bC(S)$-, or $-NR^bC(O)$- (wherein the point of the attachment to R^1 is on the left-hand side);

R^1 is selected from alkyl, cycloalkyl, aryl, heteroaryl, and heterocyclyl; each of which is substituted with 0-5 occurrences of R^d;

each R^3 is independently selected from halo, haloalkyl, alkyl, hydroxyl and $-OR^a$, or two adjacent R^3 taken together with the carbon atoms to which they are attached form an optionally substituted cyclyl;

each R^a is independently selected from alkyl, acyl, hydroxyalkyl and haloalkyl;

each R^b is independently selected from hydrogen and alkyl;

each R^c is independently selected from hydrogen, halo, alkyl, alkoxy and halo alkoxy or two R^c taken together with the carbon atoms to which they are attached form an optionally substituted cycloalkyl;

each R^d is independently selected from halo, haloalkyl, haloalkoxy, alkyl, alkynyl, nitro, cyano, hydroxyl, $-C(O)R^a$, $-OC(O)R^a$, $-C(O)OR^a$, $-SR^a$, $-NR^aR^b$ and $-OR^a$, or two R^d taken together with the carbon atoms to which they are attached form an optionally substituted heterocyclyl;

n is 0, 1, or 2;

m is 1, 2 or 3;

h is 0, 1, 2; and

g is 0, 1 or 2, wherein the compound is to be contacted with blood; and wherein the medicament is formulated for administration to a subject having pyruvate kinase deficiency (PKD).

In another aspect, there is provided use of a compound in the manufacture of a medicament for reducing 2,3-diphosphoglycerate (2,3-DPG) levels or preventing an increase in 2,3-DPG levels in blood comprising an effective amount of (1) a compound of formula I or a pharmaceutically acceptable salt thereof; (2) a pharmaceutically acceptable composition comprising a compound of formula I or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier; wherein the compound is to be contacted with blood; and wherein formula I is as defined herein; and wherein the medicament is formulated for administration to a subject having pyruvate kinase deficiency (PKD).

In another aspect, there is provided use of a compound in the manufacture of a medicament for treating hemolytic anemia comprising (1) a compound of formula I or a pharmaceutically acceptable salt thereof; or (2) a pharmaceutically acceptable composition comprising a compound of formula I or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier; wherein formula I is as defined herein, and wherein the medicament is formulated for administration to a subject in need thereof.

In another aspect, there is provided use of a compound in the manufacture of a medicament for treating pyruvate kinase deficiency (PKD) in a subject comprising (1) a compound of formula I or a pharmaceutically acceptable salt thereof; or (2) a pharmaceutically acceptable composition comprising a compound of formula I or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier; wherein formula I is as defined herein, and wherein the medicament is formulated for administration to a subject in need thereof.

In another aspect, there is provided use of a compound in the manufacture of a medicament for treating sickle cell anemia comprising (1) a compound of formula I or a pharmaceutically acceptable salt thereof; or (2) a pharmaceutically acceptable composition comprising a compound of formula I or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier; wherein formula I is as defined herein, and wherein the medicament is formulated for administration to a subject in need thereof.

In another aspect, there is provided use of a compound in the manufacture of a medicament for treating thalassemia, e.g., beta-thalassemia; hereditary spherocytosis; hereditary elliptocytosis; abetalipoproteinemia or Bassen-Kornzweig syndrome; paroxysmal nocturnal hemoglobinuria; acquired hemolytic anemia, e.g., congenital anemias, e.g., enzymopathies; or anemia of chronic diseases comprising (1) a compound of formula I or a pharmaceutically acceptable salt thereof; or (2) a pharmaceutical composition comprising a compound of formula I or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier; wherein formula I is as defined herein, and wherein the medicament is formulated for administration to a subject in need thereof.

In another aspect, there is provided use of a compound in the manufacture of a medicament for treating beta-thalassemia comprising (1) a compound of formula I or a pharmaceutically acceptable salt thereof; or (2) a pharmaceutical composition comprising a compound of formula I or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier; wherein formula I is as defined herein, and wherein the medicament is formulated for administration to a subject in need thereof.

Disclosed is~~The present invention provides~~ a method for increasing lifetime of the red blood cells (RBCs) in need thereof comprising contacting blood with an effective amount of (1) a compound disclosed herein or a pharmaceutically acceptable salt thereof; (2) a composition comprising a compound disclosed herein or a salt thereof and a carrier; or (3) a pharmaceutical composition comprising a compound disclosed

herein or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier.

Disclosed is~~The present invention further provides~~ a method for regulating 2,3-diphosphoglycerate levels in blood in need thereof comprising contacting blood with an effective amount of (1) a compound disclosed herein or a pharmaceutically acceptable salt thereof; (2) a composition comprising a compound disclosed herein or a salt thereof and a carrier; or (3) a pharmaceutical composition comprising a compound disclosed herein or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier.

Disclosed is~~The present invention also provides~~ a method for treating hereditary non-spherocytic hemolytic anemia comprising administering to a subject in need thereof a therapeutically effective amount of (1) a compound disclosed herein or a pharmaceutically acceptable salt thereof; (2) a pharmaceutical composition comprising a compound disclosed herein or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier.

Disclosed is~~The present invention further provides~~ a method for treating sickle cell anemia comprising administering to a subject in need thereof a therapeutically effective amount of (1) a compound disclosed herein or a pharmaceutically acceptable salt thereof; (2) a pharmaceutical composition comprising a compound disclosed herein or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier.

Disclosed is~~The present invention further provides~~ a method for treating hemolytic anemia (e.g., chronic hemolytic anemia caused by phosphoglycerate kinase deficiency, Blood Cells Mol Dis, 2011; 46(3):206) comprising administering to a subject in need thereof a therapeutically effective amount of (1) a compound disclosed herein or a pharmaceutically acceptable salt thereof; (2) a pharmaceutical composition comprising a compound disclosed herein or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier.

Disclosed is~~The present invention further provides~~ a method for treating thalassemia (e.g., beta-thalassemia), hereditary spherocytosis, hereditary elliptocytosis, abetalipoproteinemia (or Bassen-Kornzweig syndrome), paroxysmal nocturnal hemoglobinuria, acquired hemolytic anemia (e.g., congenital anemias (e.g.,

enzymopathies)), or anemia of chronic diseases comprising administering to a subject in need thereof a therapeutically effective amount of (1) a compound disclosed herein or a pharmaceutically acceptable salt thereof; (2) a pharmaceutical composition comprising a compound disclosed herein or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier.

Disclosed is~~The present invention further provides~~ a method for treating diseases or conditions that are associated with increased 2,3-diphosphoglycerate levels (e.g., liver diseases (Am J Gastroenterol, 1987;82(12):1283) and Parkinson's (J. Neurol, Neurosurg, and Psychiatry 1976,39:952) comprising administering to a subject in need thereof a therapeutically effective amount of (1) a compound disclosed herein or a pharmaceutically acceptable salt thereof; (2) a pharmaceutical composition comprising a compound disclosed herein or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier.

Compounds and compositions described herein are activators of PKR mutants having lower activities compared to the wild type, thus are useful for methods of the present ~~invention~~disclosure. Such mutations in PKR can affect enzyme activity (catalytic efficiency), regulatory properties (modulation by fructose bisphosphate (FBP)/ATP), and/or thermostability of the emzyme. Examples of such mutations are described in Valentini et al, JBC 2002. Some examples of the mutants that are activated by the compounds described herein include G332S, G364D, T384M, G37E, R479H, R479K, R486W, R532W, R510Q, and R490W. Without being bound by theory, compounds described herein affect the activities of PKR mutants by activating FBP non-responsive PKR mutants, restoring thermostability to mutants with decreased stability, or restoring catalytic efficiency to impaired mutants. The activating activity of the present compounds against PKR mutants may be tested following a method described in Example 1. Compounds described herein are also activators of wild type PKR.

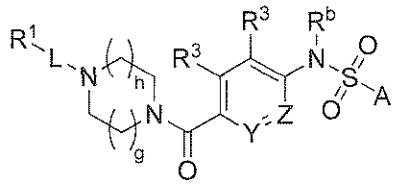
In an embodiment, to increase the lifetime of the red blood cells, a compound, composition or pharmaceutical composition described herein is added directly to whole blood or packed cells extracorporeally or be provided to the subject (e.g., the patient) directly (e.g., by i.p., i.v., i.m., oral, inhalation (aerosolized delivery),

22. The use of claim 15, wherein L is $-(CR^cR^c)_m$-OC(O)- and R^1 is heterocyclyl or cycloalkyl substituted with 0-5 occurrences of R^d.

23. The use of claim 10, wherein n is 0.

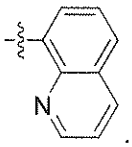
24. The use of claim 10, wherein n is 1 and R^3 is CH_3, CH_2CH_3, OCH_3, OCH_2CH_3, OH, F, Cl, or CF_3.

25. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is represented by formula (Id):

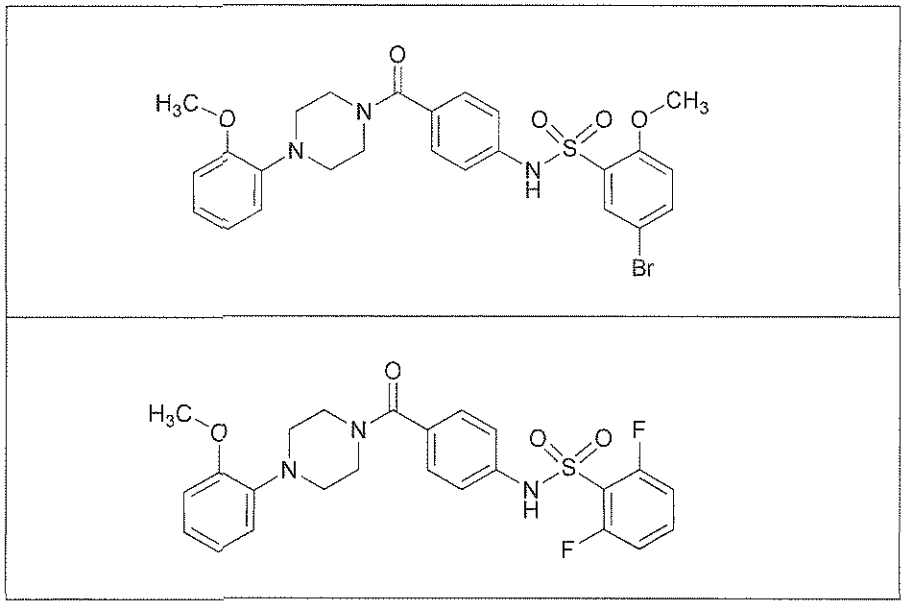


(Id).

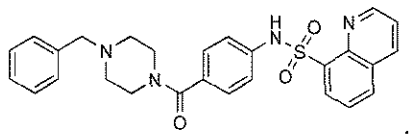
26. The use of claim 25, wherein A is optionally substituted quinolinyl.

27. The use of claim 25, wherein A is .

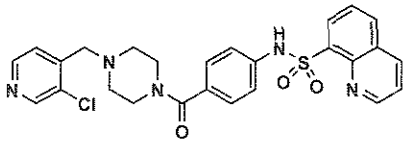
28. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



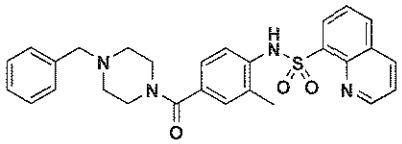
30. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



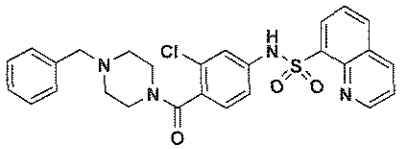
31. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



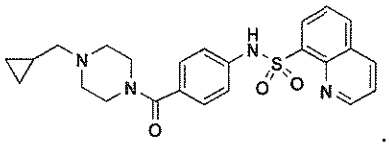
32. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



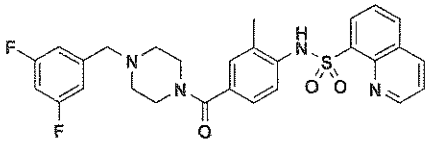
33. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



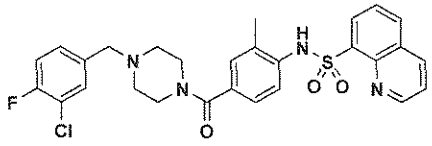
34. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



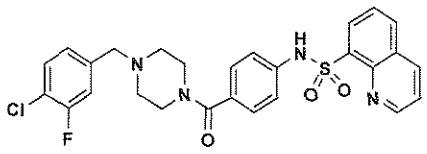
35. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



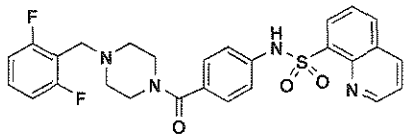
36. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



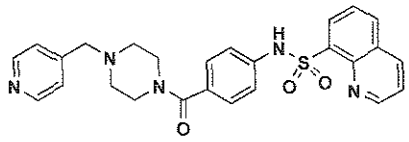
37. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



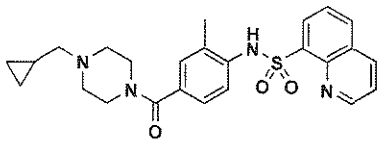
38. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



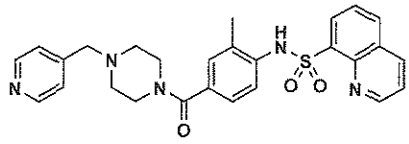
39. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



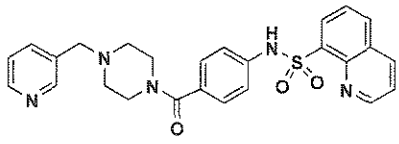
40. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



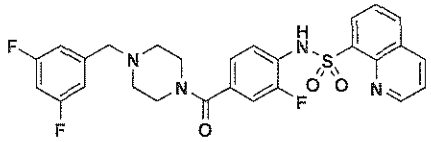
41. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



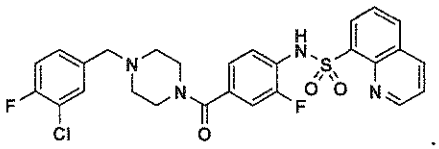
42. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



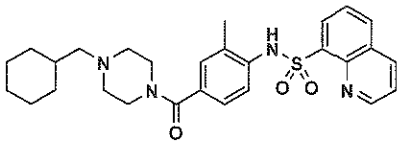
43. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



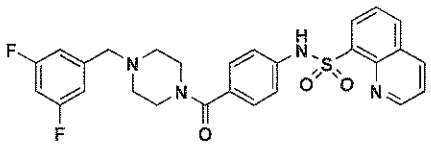
44. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



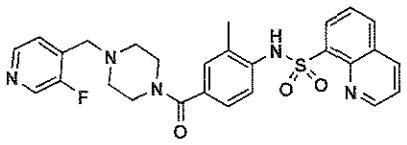
45. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



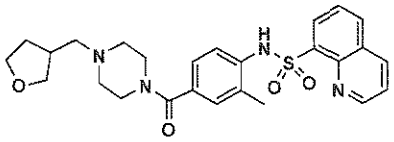
46. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



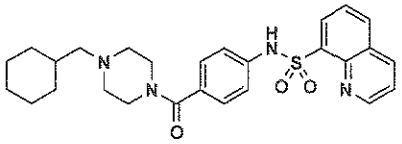
47. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



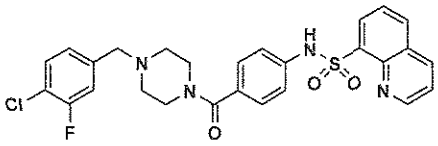
48. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



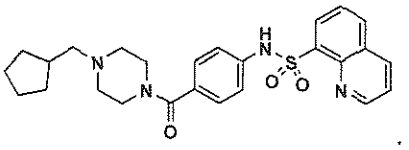
49. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



50. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



51. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:



52. The use of any one of claims 1 to 9 or a pharmaceutically acceptable salt thereof, wherein the compound is selected from:

