Furthermore, the present inventors have shown that in humans, anti-3rd party CTLs, although depleted of alloreactivity, exhibit potent in-vitro killing of B-CLL and different types of primary lymphoma cells [Lask A et al. (submitted 2010); Arditti FD et al., Blood (2005) 105:3365-3371]. This unique form of GVL was shown to be independent of TCR recognition and it was found to be mediated both by autologous and by allogeneic anti-3rd party CTLs. Furthermore, this TCR independent killing of B cell malignancies by anti-3rd party CTLs was shown to be mediated via a rapid adhesion through ICAM1-LFA1 binding, followed by slow induction of apoptosis upon a critical interaction between CD8 on the CTL and MHC class I on the tumor cell. Moreover, the killing was shown to be independent of the classical CTLs death molecules: FASL, perforin, TNF, and Trail [Lask A et al., supra].

Additional background art includes WO/2010/049935.

SUMMARY ~~OF THE INVENTION~~

According to ~~an aspect of~~ some embodiments of the present ~~invention~~disclosure there is provided a method of treating a disease in a subject in need thereof, the method comprising: (a) transplanting a non-syngeneic cell or tissue graft to the subject; and (b) administering to the subject a therapeutically effective amount of an isolated population of cells comprising non-graft versus host (GVHD) inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, the cells being tolerance-inducing cells and capable of homing to the lymph nodes following transplantation, and further wherein the cells are either: (i) non-syngeneic with both the subject and the graft; or (ii) non-syngeneic with the graft and syngeneic with the subject, thereby treating the subject.

In one aspect, there is provided a method of generating an isolated population of cells comprising non-graft versus host (GVHD) inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said central memory T-lymphocyte (Tcm) phenotype comprising a CD8+/CD62L+/CD45RO+ signature and wherein at least 50 % of the isolated population of cells have said signature, said cells being tolerance-inducing cells and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) contacting peripheral blood mononuclear cells (PBMC) with a third party antigen or antigens in the presence of IL-21 under conditions which allow elimination of GVH reactive cells; and

(b) culturing said cells resulting from step (a) in the presence of IL-15 in an antigen free environment under conditions which allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

According to ~~an aspect of~~ some embodiments of the present ~~invention~~disclosure there is provided a use of an isolated population of cells comprising non-graft versus host (GVHD) inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, the cells being tolerance-inducing cells and capable of homing to the lymph nodes following transplantation, as an adjuvant treatment for eradication of a disease in a subject who has been transplanted with a non-syngeneic cell or tissue graft, wherein

the cells are either: (i) non-syngeneic with both the subject and the graft; or (ii) non-syngeneic with the graft and syngeneic with the subject.

In another aspect, there is provided the use of an isolated population of cells generated according to the method as defined above for the manufacture of a medicament identified for eradication of a disease in a subject who has been transplanted with a non-syngeneic cell or tissue graft, wherein said cells are either:

(i) non-syngeneic with both the subject and said graft; or

(ii) non-syngeneic with said graft and syngeneic with the subject.

According to ~~an aspect of~~ some embodiments of the present ~~invention~~disclosure there is provided an isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, the cells being tolerance-inducing cells and capable of homing to the lymph nodes following transplantation, and further wherein the cells are either: (i) non-syngeneic with both a host subject and a graft, wherein the subject and the graft are non-syngeneic; or (ii) non-syngeneic with a graft and syngeneic with a host subject, wherein the subject and the graft are non-syngeneic.

According to ~~an aspect of~~ some embodiments of the present ~~invention~~disclosure there is provided a method of treating a disease in a subject in need thereof, the method comprising: (a) transplanting immature hematopoietic cells to the subject; and (b) administering to the subject a therapeutically effective amount of an isolated population of cells comprising non-graft versus host (GVHD) inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, the cells being tolerance-inducing cells and capable of homing to the lymph nodes following transplantation, wherein when the immature hematopoietic cells are syngeneic with the subject, the isolated population of cells are selected syngeneic with the subject or non-syngeneic with the subject.

According to ~~an aspect of~~ some embodiments of the present ~~invention~~disclosure there is provided a method of treating a disease in a subject in need thereof, the method comprising: (a) transplanting immature hematopoietic cells to the subject; and (b) administering to the subject a therapeutically effective amount of an isolated population of cells comprising non-graft versus host (GVHD) inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, the cells being tolerance-inducing

cells and capable of homing to the lymph nodes following transplantation, wherein when the immature hematopoietic cells are non-syngeneic with the subject, the isolated population of cells are selected syngeneic with the subject or non-syngeneic with both the subject and the immature hematopoietic cells.

In another aspect, there is provided the use of a therapeutically effective amount of an isolated population of cells generated according to the method as defined above for the manufacture of a medicament identified for treating a disease in a subject who has been transplanted with immature hematopoietic cells, and further wherein when said immature hematopoietic cells are syngeneic with the subject, said isolated population of cells are selected syngeneic with the subject or non-syngeneic with the subject.

In another aspect, there is provided the use of a therapeutically effective amount of an isolated population of cells generated according to the method as defined above for the manufacture of a medicament identified for treating a disease in a subject who has been transplanted with immature hematopoietic cells, wherein when said immature hematopoietic cells are non-syngeneic with the subject, said isolated population of cells are selected syngeneic with the subject or non-syngeneic with both the subject and said immature hematopoietic cells.

According to some embodiments of the ~~invention~~disclosure the non-syngeneic cell or tissue graft is derived from a donor selected from the group consisting of an HLA identical donor and an HLA non-identical donor.

According to some embodiments of the ~~invention~~disclosure the subject and the donor are both humans.

According to some embodiments of the ~~invention~~disclosure the subject is a human subject.

According to some embodiments of the ~~invention~~disclosure the cell or tissue graft is non-autologous and the isolated population of cells are autologous.

According to some embodiments of the ~~invention~~disclosure the cell or tissue graft and the isolated population of cells are from different donors.

According to some embodiments of the ~~invention~~disclosure the disease comprises a malignancy.

According to some embodiments of the ~~invention~~disclosure the malignancy comprises a B cell malignancy.

According to some embodiments of the ~~invention~~disclosure the malignancy comprises a leukemia.

According to some embodiments of the ~~invention~~disclosure the malignancy comprises a lymphoma.

According to some embodiments of the ~~invention~~disclosure the graft comprises bone marrow cells.

According to some embodiments of the ~~invention~~disclosure the bone marrow cells comprise immature hematopoietic cells.

According to some embodiments of the ~~invention~~disclosure the immature hematopoietic cells are non-syngeneic with the subject, the isolated population of cells are syngeneic with the subject.

According to some embodiments of the ~~invention~~disclosure the immature hematopoietic cells are non-autologous and the isolated population of cells are autologous.

According to some embodiments of the ~~invention~~disclosure when the immature hematopoietic cells are non-syngeneic with the subject, the isolated population of cells are non-syngeneic with both the subject and with the graft.

According to some embodiments of the ~~invention~~disclosure the immature hematopoietic cells and the isolated population of cells are from different donors.

According to some embodiments of the ~~invention~~disclosure the method further comprises administering at least one immunosuppressant drug to the subject.

According to some embodiments of the ~~invention~~disclosure the subject is further conditioned under sublethal, lethal or supralethal conditioning prior to step (a).

According to some embodiments of the ~~invention~~disclosure step (a) and step (b) are effected concomitantly.

According to some embodiments of the ~~invention~~disclosure step (b) is effected prior to step (a).

According to some embodiments of the ~~invention~~disclosure step (b) is effected one day after step (a).

According to some embodiments of the ~~invention~~disclosure the central memory T-lymphocyte (Tcm) phenotype comprises a $CD8^+/CD62L^+$ signature.

According to some embodiments of the ~~invention~~disclosure at least 50 % of the isolated population of cells have the signature.

According to some embodiments of the ~~invention~~disclosure the lymph nodes comprise peripheral lymph nodes.

According to some embodiments of the ~~invention~~disclosure the lymph nodes comprise mesenteric lymph nodes.

According to some embodiments of the ~~invention~~disclosure the cells non-syngeneic with the graft and syngeneic with the subject comprise autologous cells.

According to some embodiments of the ~~invention~~disclosure the cells non-syngeneic with the graft and syngeneic with the host subject are autologous.

According to some embodiments of the ~~invention~~disclosure the immature hematopoietic cells and the isolated population of cells are autologous.

According to some embodiments of the ~~invention~~disclosure the immature hematopoietic cells are autologous and the isolated population of cells are non-autologous.

According to some embodiments of the ~~invention~~disclosure the immature hematopoietic cells are non-autologous and the isolated population of cells are autologous.

According to some embodiments of the ~~invention~~disclosure the immature hematopoietic cells and the isolated population of cells are from different donors.

According to some embodiments of the ~~invention~~disclosure the anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, the cells being tolerance-

inducing cells and capable of homing to the lymph nodes following transplantation are generated by: (a) contacting peripheral blood mononuclear cells (PBMC) with a third party antigen or antigens in the presence or absence of IL-21 under conditions which allow elimination of GVH reactive cells; and (b) culturing the cells resulting from step (a) in the presence of IL-15 in an antigen free environment under conditions which allow proliferation of cells comprising the central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

According to some embodiments of the ~~invention~~disclosure the conditions which allow elimination of GVH reactive cells comprise culturing for 1-5 days.

According to some embodiments of the ~~invention~~disclosure the conditions which allow elimination of GVH reactive cells comprise 1-5 days in a culture deprived of cytokines.

According to some embodiments of the ~~invention~~disclosure the third party antigen or antigens comprise dendritic cells.

According to some embodiments of the ~~invention~~disclosure the third party antigen or antigens is selected from the group consisting of third party cells, a cell antigen, a viral antigen, a bacterial antigen, a protein extract, a purified protein and a synthetic peptide presented by autologous presenting cells, non-autologous presenting cells or on an artificial vehicle or artificial antigen presenting cell.

According to some embodiments of the ~~invention~~disclosure the conditions which allow proliferation of cells further comprise IL-7.

According to some embodiments of the ~~invention~~disclosure the culturing in the presence of IL-15 is effected for 3-30 days.

According to some embodiments of the ~~invention~~disclosure the culturing in the presence of IL-15 is effected for 6-30 days.

According to some embodiments of the ~~invention~~disclosure the culturing in the presence of IL-15 is effected for 3-20 days.

According to some embodiments of the ~~invention~~disclosure the conditions which allow proliferation of cells comprise culturing for 3-10 days.

According to some embodiments of the ~~invention~~disclosure the conditions which allow proliferation of cells comprise culturing for 3-7 days.

Unless otherwise defined, all technical and/or scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which

the ~~invention~~disclosure pertains. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of embodiments of the ~~invention~~disclosure, exemplary methods and/or materials are described below. In case of conflict, the patent specification, including definitions, will control. In addition, the materials, methods, and examples are illustrative only and are not intended to be necessarily limiting.

BRIEF DESCRIPTION OF THE DRAWINGS

Some embodiments of the invention are herein described, by way of example only, with reference to the accompanying drawings. With specific reference now to the drawings in detail, it is stressed that the particulars shown are by way of example and for purposes of illustrative discussion of embodiments of the invention. In this regard, the description taken with the drawings makes apparent to those skilled in the art how embodiments of the invention may be practiced.

In the drawings:

FIGs. 1A-I depict inhibition of tumor relapse by syngeneic derived anti-3rd party Tcm cells after syngeneic bone marrow transplantation. Lethally irradiated (8 Gy) BALB/c mice (H-2^{d}) received intravenously a transplant of 3 x 10^{6} syngeneic T cell depleted bone marrow (BALB/c-NUDE (H-2^{d}) in the presence or absence of 5 x 10^{3} A20-luc lymphoma cells (day 0). Mice were then intravenously injected with or without the indicated numbers of BALB/c derived anti-3rd party Tcm cells (indicated as leukemia cells + Tcm, n = 7; or with only leukemia cells, n =7, respectively) on day +1. Figures 1A-H are pictures depicting tumor growth monitored by bioluminescence imaging (BLI) from day 14 in weekly intervals; Figure 1I is a graph depicting the survival rate of the animals from the different treatment groups. X-axis indicates days after tumor cell injection and the y-axis indicates the proportion of recipient mice surviving.

FIGs. 2A-G depict inhibition of tumor relapse by F1 derived anti-3rd party CD8 T cells after syngeneic bone marrow transplantation. Lethally irradiated (8 Gy) BALB/c mice (H-2^{d}) received intravenously a transplant of 3 x 10^{6} syngeneic T cell depleted bone marrow cells (BALB/c-NUDE (H-2^{d}) in the presence or absence of 5 x 10^{3} A20-luc lymphoma cells (day 0). Mice were then intravenously injected with or without 2 x 10^{7} F1 derived anti-3rd party Tcm cells (Leukemia cells + Tcm, n = 7) or without (Leukemia cells alone, n=5) on day +1; Figures 2A-F are pictures depicting tumor