

APPENDIX H - SAMPLE PROVISIONAL APPLICATION

METHODS OF USING APOPTOSIS COMMITTED CELLS

Field of The Invention

[0001] The field of the invention is the use of dendritic cells.

Background of The Invention

[0002] Cells can become diseased under a number of different conditions, including nutritional or metabolic insults, viral, bacterial and mycotic infections, and various infestations. In some instances, the cells can also become neoplastic, resulting in cancer. In many instances the diseased cells can be removed from the organism in some manner, inactivated in some manner, or even returned to a healthy state. Often, however, the treatments are quite invasive, and may not even be particularly effective. Thus, there continues to be a need for less invasive and more effective treatments.

[0003] Diseased cells can be treated *ex vivo*, i.e., outside of system in which they became diseased. Typically, the system is a body such as a human or animal body, and diseased cells are removed from the body, treated in some manner, and then reintroduced into the body. There are many methods for treating cells *ex vivo*, including irradiation, interaction with various chemicals, and stimulation by antigens.

[0004] US 5,788,963 to Murphy et al. (Aug. 1998) teaches that prostate cancer can be treated by removing immune system cells from a patient, activating the removed cells via presentation with prostate cancer antigen, expanding the activated cells, and then introducing the expanded cells into the patient. Although Murphy reports significant results using this method, there are significant drawbacks.

[0005] One problem with the Murphy method is selection of antigen to be presented to the immune system cells. Library or "stock" antigen is unavailable for each different type of cancer, let alone other diseases, and even if library antigen were available for every type of disease, such antigens would likely correlate only poorly with the target antigens for the system being treated. Murphy's solution is to remove a population of diseased cells from the system being treated, lyse the cells, and then present the lysate to the immune system cells. That solution undoubtedly does provide some target antigen, but the target antigen is mixed among a large amount of non-target antigen.

[0006] Another problem with the Murphy method is presentation of antigen to the immune system cells. It was already known that free antigen is extremely inefficient at activating T cells and other lymphocytes *ex vivo*, and Murphy attempted to improve the activation efficiency by presenting the antigen to dendritic cells. The dendritic cells become activated in this manner, and are then used to activate T-cells against the antigen. Unfortunately, antigen in lysate is relatively inefficient at activating dendritic cells. Worse still, it is entirely possible that the dendrocytes will become activated against non-target antigen, and that immune system components activated in this manner will provoke an auto-immune reaction in the system upon reintroduction.

[0007] Thus, there is still a need to improve antigen selection and antigen presentation in the *ex vivo* stimulation of immune system components.

Summary of the Invention

[0008] According to the present invention diseased cells are removed from a body or other system, and committed to apoptosis *ex vivo*. During this process the apoptosis committed cells present surface antigen, and that antigen is employed to activate dendrocytes or other APCs. Such antigen is then employed against diseased cell remaining *in vivo*.

[0009] In preferred methods both the selection of antigen and the presentation of antigen is performed by the cells undergoing apoptosis, and in especially preferred methods the apoptosis committed cells present antigen to dendrocytes or other APCs, which then activate NK or other lytic lymphocytes. Such methods are thought to mimic antigen presentation processes normally taking place with a body, and are contemplated to be particularly efficient. In alternative methods, however, apoptosis selected antigen can be reintroduced directly back into the system. In still other methods, apoptosis selected antigen can be employed to directly stimulate immune system components other than APCs, and those components can be reintroduced into the system.

[0010] Various objects, features, aspects and advantages of the present invention will become more apparent from the following detailed description of preferred embodiments of the invention.

Brief Description of the Drawing

[0011] Figure 1 is a flowchart of a preferred method for treating a patient according to the present invention.

Detailed Description

[0012] In **Figure 1** a method for treating a patient generally comprises the following steps: removing a sample of diseased cells from a patient (10); committing the removed cells to apoptosis (20); and introducing antigen presented by the committed cells to cells of the patient's immune system (30).

[0013] The patient is contemplated to be any higher organism having diseased cells present in its body at the time of the treatment. Contemplated patients include vertebrates, especially mammals, and most especially humans. Treatment of livestock and pets, such as cats and dogs, are also of particular interest. Diseased cells are contemplated to be any cells of which the patient wants to eliminate. Contemplated diseased cells include those invaded by cancer, viruses, fungus, toxins, or bacteria. A sample of diseased cells from the patient may be collected by any suitable harvesting procedure, including, for example, scraping, resection, or aspiration. The number of removed cells may therefore vary greatly, and it is contemplated that the number may vary from 10^6 cells or less, to 10^7 cells or more. It is contemplated that diseased cells can be removed from anywhere on the patient's body. Contemplated areas of the patient's body which are available for cell harvesting include the brain, skin, bone marrow, reproductive organs, or other major organs.

[0014] Cells can be committed to apoptosis using any suitable methods and apparatus. A highly preferred method comprises incubating the removed cells in interferon, especially type 1 interferon, for a suitable period of time. Experimentation shows that such time spans are commonly in excess of 24 hours, and possibly up to two weeks or longer. Other methods are also contemplated, including nutrient, toxic, and physical stressors, as well as genetic inhibitors or inducers. For example, it is contemplated that factors which down regulate any of the stress accommodation genes may be used to commit cells to apoptosis.

[0015] Committing the diseased cells to apoptosis caused the committed cells to exhibit surface antigens corresponding to the related disease. In this manner the committed cells more or less selectively present the target antigens. It is contemplated that such antigen can be presented to cells of the patient's immune system either *in vivo* or *ex vivo*.

[0016] Thus, specific embodiments and applications of methods of using apoptosis committed cells have been disclosed. It should be apparent, however, to those skilled in the art that many more modifications besides those already described are possible without departing from the inventive concepts herein. The inventive subject matter, therefore, is not to be restricted except in the spirit of the appended claims.

CLAIMS

What is claimed is:

1. A method of treating diseased cells in a system, comprising:
removing a sample of the diseased cells from the system;
committing the removed cells to apoptosis *ex vivo*, whereby the
committed cells selectively produce surface antigen; and
employing the surface antigen to treat diseased cells remaining in the
system.
2. The method of claim 1 further comprising:
harvesting the surface antigen; and
depositing the harvested antigen back into the system.
3. The method of claim 1 further comprising presenting the surface antigen
to a collection of cells of the patient's immune system *ex vivo*.

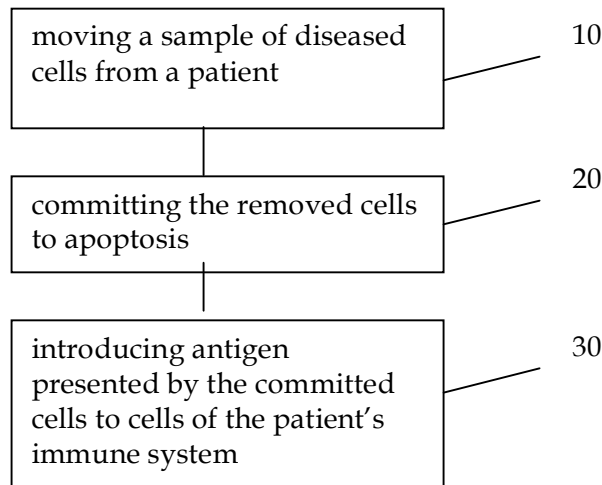


Figure 1