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BIOCARRIER COMPOSITION FOR AND METHOD OF DEGRADING POLLUTANTS

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246,261 BIOCARRIER COMPOSITION FOR AND METHOD OF DEGRADING POLLUTANTS Carl B. Fliermans DE-AC09-89SR18035

PATENT

BIOCARRIER COMPOSITION FOR AND METHOD OF DEGRADING POLLUTANTS

BACKGROUND OF THE INVENTION

1. Field of the Invention:

The present invention relates to biocarrier compositions. More particularly, the present invention relates to biocarrier compositions that attract and bond pollutant-degrading antigens that will degrade the pollutants. The United States Government has rights in this invention pursuant to Contract No. DE-AC09-89SR18035 between the U.S. Department of Energy and Westinghouse Savannah River Company.

2. Discussion of Background:

Biocarriers are known generally as a variety of inert or semi-inert compounds or structures having the ability to sequester (attract), hold and biomagnify (enhance) specific microorganisms within their structure. Glass or polystyrene beads are the most well known biocarriers. Biocarriers are frequently used in certain medical procedures, such as blood work, in which they are coated with antigens and dyes to assist in detecting the presence of antibodies in blood specimens. In a similar medical application, biocarriers are coated with antibodies to assist in detecting the presence of antigens in urine samples.

In U.S. Patent 5,045,201, Dubois, et al. use glass microbeads having a coating of at least one fixing agent and one binding agent for separating biological materials produced by animal cells. The fixing agent, similar to a biological adhesive, strongly attaches itself to the binding agent and the glass microbead to resist removal during processing. The binding agent may be an antibody or an antigen, although only a limited number are disclosed, including immunoglobulin of the type IgG or IgM (pentamer), protein A, and protein G.

Also, biocarriers have been used, to a certain extent, in detritiation processes. In their article "Application of Immobilized Hydrogenase for the Detritiation of Water", Alexander M. Klibanov and Jonathan Huber discuss the detritiation of contaminated water using whole bacteria cells of Alcaligenes eutrophus immobilized in calcium alginate or k-carrageenan gels.

However, biocarriers are not known to be used in pollutant degradation environments such as, for example, in-situ degradation of groundwater pollutants and other fluid streams and in fluid waste streams. There exists a need for new and effective techniques for this type of environmental remediation.

SUMMARY OF THE INVENTION

According to its major aspects and broadly stated, the present invention is a composition and method for using the composition for degrading pollutants. In particular, it is a biocarrier coated with an antigen-specific antibody for attracting pollutant-degrading microorganisms (antigens) thereto for subsequent use in degrading pollutants. The biocarrier, which is preferably in the form of glass microspheres, is coated with an antibody or group of antibodies that attract and react specifically with certain pollutant-degrading antigens.

The antibody, once bonded to the biocarrier, is used by the composition to attract and bond those pollutant-degrading antigens. Each antibody is specific for an antigen that is specific for a given pollutant. The resulting composition is subsequently exposed to an environment contaminated with pollutants for degradation. In the preferred use, the degrading composition is formed and then injected directly into or near a plume or source of contamination.

A major feature of the present invention is the combination of the biocarrier's inertness and its porosity. By being porous, it provides increased surface area for the antibodies and antigens; by being inert, it serves as a vehicle for their effective delivery directly of the antibodies and antigens into a plume of contaminants in the subsurface environment. It attacks and holds specific organisms (antigens) present in the groundwater where the pollutant is present.

Another feature of the present invention is the range of possible antigens available for use in bioremediation. The large number enables this technique to be applied to many types of pollutants.

Another feature of the present invention is the selection of specific antigens that degrade organic pollutants such as trichloroethylene, toluene, phenol and the like in groundwater, where the pollutants are most likely to be, since microorganisms are most active in liquids such as water.

Another feature is the use of microspheres of glass, polystyrene and the like as a biocarrier substrate for the degrading composition. The advantage of this feature is that the extremely small size of each microsphere (approximately 10^6 to 10^{12} microspheres per gram and smaller) allows them to be injected into contaminated subsurfaces and positioned in unusually small areas where pollutants might reside, for example, in fractured rocks.

Other features and advantages of the present invention will be apparent to those skilled in the art from a careful reading of the Detailed Description of a Preferred Embodiment presented below.

DETAILED DESCRIPTION OF A PREFERRED EMBODIMENT

The composition for degrading pollutants according to its preferred embodiment comprises a biocarrier having a preselected antibody or group of antibodies bound or chemically linked thereto. The antibody is chosen based on its ability to attract an antigen or group of antigens of interest that, in turn, are capable of degrading certain pollutants of interest. The method for pollutant degradation uses the disclosed composition in at least one of two embodiments. In the first embodiment, the biocarrier/antibody composition is used to attract at least one pollutant-degrading antigen, and the resulting composition is used to degrade pollutants. The second embodiment uses a biocarrier/antibody composition having at least one antigen fixed thereto for degrading pollutants.

As previously stated, biocarriers are inert or semi-inert compounds having the ability to sequester, hold and/or biomagnify microorganisms within their structure. "Sequester" means that the microorganism is kept isolated by the biocarrier from its environment. Also, "biomagnify" means that the sequestered microorganisms increase in number and possibly size within the biocarrier.

Biocarriers can be made of several substances, including glass, latex, polystyrene, rubber, wood and aragose. Also, biocarriers may include plastic materials of polypropylene, polyethylene, polyvinyl dichloride (PVDF), Halar, Tefzel, polyvinyl chloride (PVC), polytetrafluoroethylene (TEFLON®) and the like. Biocarriers may take the form of, but are not limited to microspheres and other beads, hollow membranes, injection foams, fluted filters, hollow fiber filters and the like. Other examples of biocarriers include glass beads, latex beads, and foams made of rubber. Biosphere's can be made to contain nutrients such as nitrogen and phosphorus that would stimulate the growth of the sequestered microorganisms and allow the organisms to grow optimally while degrading the pollutants, which serves as a carbon source for the antigen.

One of the important characteristics of biocarriers is that they have high surface areas for covalently bonding with other substances. Taking advantage of this characteristic, a biocarrier can be made to select and sequester a particular organism of choice. Such biocarriers are known as serobiocarriers, and here they are formed by binding a particular antibody or group of antibodies to the biocarrier so that the serobiocarrier will, in turn, attract and bond specific antigens.

An antibody is a protein characterizable in part by a specific reaction to a complementary antigen or small group of related antigens. Antibodies are often classified by the number of combining sites they have available, which is a factor in determining to which antigen they bond most effectively. "Monovalent" antibodies, obviously, have only one combining site; accordingly, "polyvalent" antibodies have many combining sites. Certain antibodies, referred to as "monoclonal" antibodies, react only with specific antigens. Similarly, "polyclonal" antibodies are less specific than monoclonal antibodies, that is, they bind with more than one type of antigen.

The antigens can be small organic chemicals, specific enzymes, a particular bacterium, or other substances that are attracted to a complementary antibody or group of antibodies. They may be naturally occurring, man-made or genetically engineered. Preferably, the antigens being attracted are those known to degrade one or more kinds of pollutants.

Pollutants are defined here as any harmful, organic or inorganic chemical or a radioactive particle or other toxic waste found in process streams, groundwater and similar areas. Such pollutants include but are not limited to chlorinated solvents, aliphatic compounds, hazardous and toxic waste chemicals, heavy metals or any other substance that is degradable or transformed by an antigen and presents a hazard to humans, animals and crops. Substances that are transformed by an antigen are inserted to allow chemical modification without degradation.

The choice of antigen to be attracted, and consequently the choice of antibody that is to be bound to the biocarrier, depends on the specific application, that is, the specific pollutants that are to be degraded and the particular environment in which they exist.

Thus, if desired, an antigen-specific serobiocarrier can be formed by binding a particular antibody to the surface and/or internal structure of a biocarrier. The choice of the particular antibody or group of antibodies used in this formation depends on the ability of that antibody to attract and hold the antigen or group of antigens of interest. The resulting serobiocarrier provides extensive surface area for the antigen of interest. Using the specificity of the particular antibody/antigen reaction in this manner, serobiocarriers can be formed that are adapted to degrade and transform selected pollutants from a variety of environments.

Preferably, the serobiocarrier composition is used for degrading pollutants. The preferred embodiment of the method comprises binding an antibody or group of antibodies to a biocarrier. The resulting composition is exposed to a pollutant-degrading antigen or group of antigens and then, in turn, exposed to an environment containing the pollutants of interest for degradation. An alternative embodiment of the method comprises exposing the biocarrier composition, having pollutant-degrading antigens bound thereto, to an environment containing the pollutants to be degraded.

The serobiocarrier is made by coating or otherwise binding the antibody or group of antibodies to the biocarrier of choice. Such binding involves chemically linking the antibody to the surface and internal structure of the biocarrier, and depends on the specific biocarrier being used. The particular biocarrier is chosen based on the expected application of the serobiocarrier, that is, the expected type of environment in which the composition will be exposed. This will be discussed in more detail later.

The antibody is bound to the biocarrier using any one of a number of conventional means. In preparing the composition, the reactions of the biocarrier with the antibody of interest is preferably conducted under conditions of standard temperature and pressure. Preferably, each biocarrier substrate is saturated with the antibody so that the binding sites present on the biocarrier are coated with the antibody. An incubation time of up to 24 hours usually follows, depending on the specific reactants used. The final step is to wash or rinse any unattached antibodies from the biocarrier using a buffered saline wash.

In use, the biocarrier/antibody composition, once formed into a serobiocarrier, is exposed, in a manner to be described more fully below to an environment containing pollutant-degrading antigens of interest. The known antibody/antigen reaction causes the antigens to be attracted and eventually bound to the specific antibody sites of the serobiocarrier. Exposure parameters such as time, serobiocarrier concentration, antigen concentration and the like, vary depending on the specific biocarrier, antibody, and antigen being used or sought, as well as the exposure environment. However, the determination of all parameters necessary for proper operation of the present invention can be determined reasonably by one skilled in the art after a modicum of experimentation.

Once the pollutant-degrading antigens of interest are attached to the antibodies bound in the biocarrier, the resulting serobiocarrier is exposed to the environment containing pollutants to be degraded. Again, the parameters for exposure to the contaminated environment depend on the particular biocarriers, antibodies, antigens and pollutants being used or sought and can be determined readily by one skilled in the art after a modicum of experimentation.

The pollutant-degrading method described above is adaptable to many different applications. Preferably, serobiocarriers are used in-situ for the remediation of groundwater contaminated with pollutants. In a preferred use, serobiocarriers are injected into a plume of the contaminated groundwater for direct degradation. In a more preferred embodiment, serobiocarriers are positioned into or near the plume via horizontal wells or a combination of horizontal and vertical wells.

When used in conjunction with horizontal wells, preferably in the form of injection wells positioned below the contaminated plume and extraction wells positioned above the contaminated plume, serobiocarriers are preferably suspended along the length of the shaft of the extraction wells positioned above the contaminated plume. The serocarriers can also be injected into the groundwater itself. Also, preferably, the serobiocarriers are prepared and packed in cylindrical filter beds suspended along the extraction well shaft. In this manner, during operation, the filters containing the serobiocarriers turn slowly to provide additional contact with the contaminated groundwater as the groundwater passes or is drawn from the plume through the extraction well. Thus, the degradation activity would take place along the extraction well. The extracted groundwater, which is no longer contaminated, is reinjected into the groundwater via the injection wells to provide gradient flow through the plume.

When the serobiocarriers are used in vertical wells, they are preferably suspended in filter beds in the screen zone of the vertical wells. Similar to their use in horizontal filters, the means for suspending the serobiocarriers are preferably configured to rotate so that, during operation, the filters turn slowly to provide additional contact between the serobiocarriers and the contaminated groundwater as the groundwater passes or is drawn through the well. Thus, the degradation activity occurs at the screen zone. This configuration is similar to a revolving disk function in a municipal water treatment facility.

In the manner described above, the serobiocarrier is preferably exposed initially to groundwater (not necessarily contaminated groundwater) to attract and attach antigens contained therein that are known for degrading the pollutants of interest. Then, the serobiocarrier is exposed to the contaminated groundwater so that the collected antigens can degrade the pollutants of interest. Alternatively, the serobiocarrier is equipped with specific antigens prior to exposure to the contaminated groundwater. In addition to degrading contaminated groundwater, both methods can be used for degrading pollutants contained in flowing streams, both natural and man-made.

In yet another application, serobiocarriers containing pollutant-degrading antigens can be used in conjunction with bioreactors. In this alternative application, the serobiocarriers are prepared and packed in the bed of a biological filter that can be contained within the bioreactor in one of many acceptable forms. The polluted environment, preferably contaminated liquids such as groundwater, is then brought to the bioreactor and pumped through the biological filter. Alternatively, the biological filter can be suspended on a rotational axis device within the bioreactor to provide movement to the filter during operation to allow greater interaction between the serobiocarriers in the filter bed and the contaminated liquid, thus increasing the effectiveness of the degradation.

The serobiocarrier applications as discussed above are used to remove various pollutants from contaminated environments through transformation and degradation.

Because of the ability of the biocarriers, antibodies and antigens to function physiologically in extreme environments, including those found in thermal hot springs and oligotrophic environments, serobiocarriers can also be used to degrade pollutants in the presence of waste containing both radioactive and chemical hazards, called "mixed" hazardous wastes.

Often, the nonradioactive portion of the mixed hazardous waste is readily degradable by one or more pollutant-degrading antigens.

Thus, in this application, mixed hazardous waste is fed continuously into bioreactors (see previous discussion). The level and rate of the continuous feed depend on the feed concentrate, the types of biocarriers, antibodies and antigens present, and the rate of reaction of the pollutant-degrading antigens with respect to the organic portion of the mixed hazardous waste.

Similar to the previously discussed bioreactor operation, the biological filters (with biocarrier beds) may be suspended in such a way that they rotate about an axis to provide movement of the mixed hazardous waste through the filter beds thus with greater surface area exposed to the waste and therefore greater interaction between the biocarriers and the mixed hazardous waste.

In this manner, the pollutant-degrading antigens contained in the biocarrier will effectively degrade the organic waste portion of the mixed hazardous waste while leaving behind radioisotopes, which can be vitrified or processed using conventional waste treatment technology.

It will be apparent to those skilled in the art that many changes and substitutions can be made to the preferred embodiment herein described without departing from the spirit and scope of the present invention as defined by the appended claims.

ABSTRACT OF THE DISCLOSURE

A composition and method for using the composition for degrading pollutants *in-situ*. The composition comprises a biocarrier coated with an antigen-specific antibody that attracts and binds pollution-degrading antigens. The biocarrier, which is preferably in the form of glass microspheres, is coated with one or more strains of antibody. The antibody may be placed into the ground in or near the source of pollutants where it may attract antigens present and bind them, or the antibodies may be first exposed to the antigens and then placed in the ground. Alternatively, the coated biocarriers may be used to degrade pollutants in ground water pumped to the surface and through a biofilter containing the biocarriers. The remediated groundwater can then be returned to the soil.