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(54) **METHODS AND COMPOSITIONS FOR
VACCINATION COMPRISING NUCLEIC
ACID AND/OR POLYPEPTIDE SEQUENCES
OF CHLAMYDIA**

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(57) **ABSTRACT**

The instant invention relates to antigens and nucleic acids encoding such antigens obtainable by screening a *Chlamydia* genome. In more specific aspects, the invention relates to methods of isolating such antigens and nucleic acids and to methods of using such isolated antigens for producing immune responses. The ability of an antigen to produce an immune response may be employed in vaccination or antibody preparation techniques.

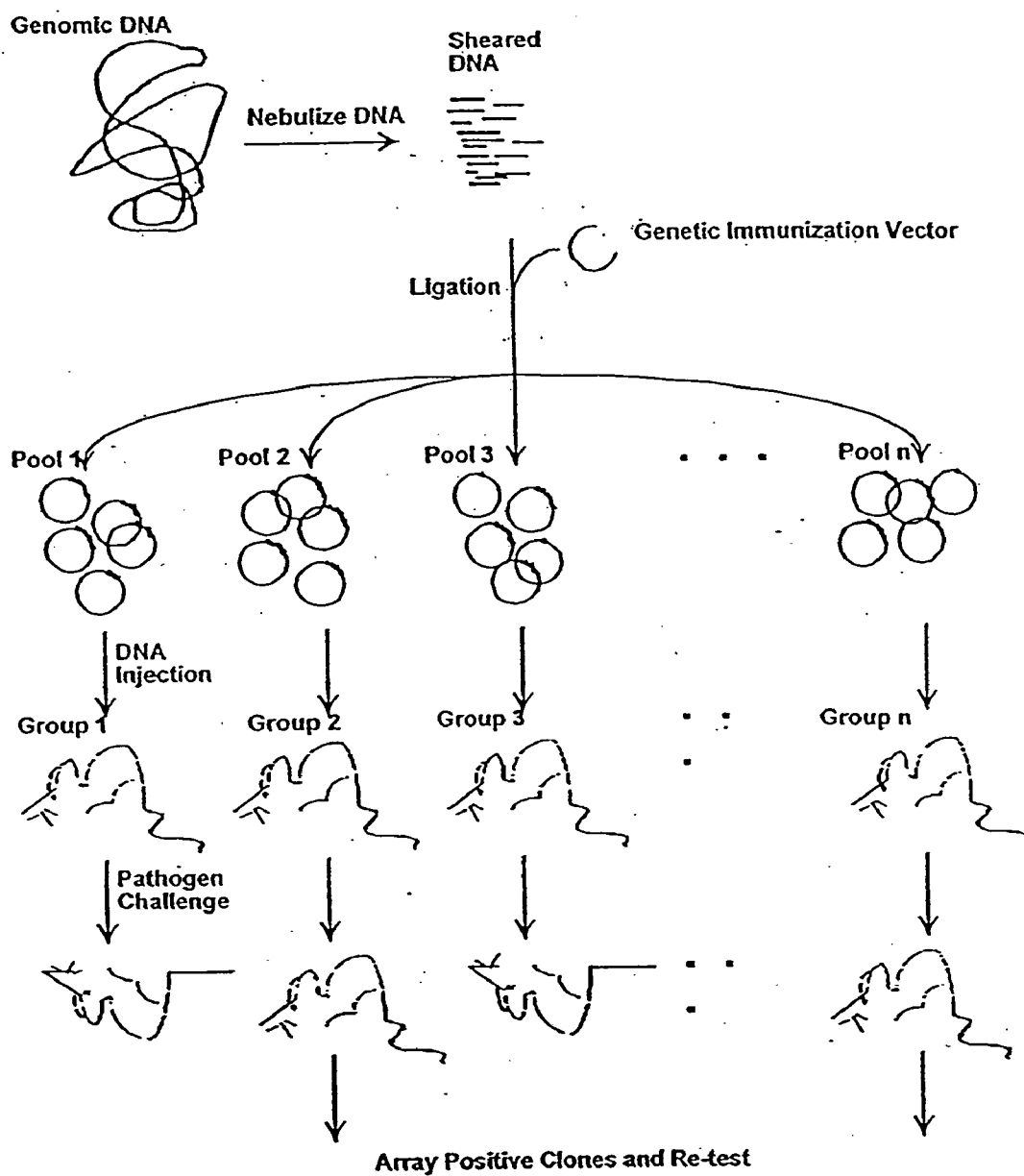


FIG. 1

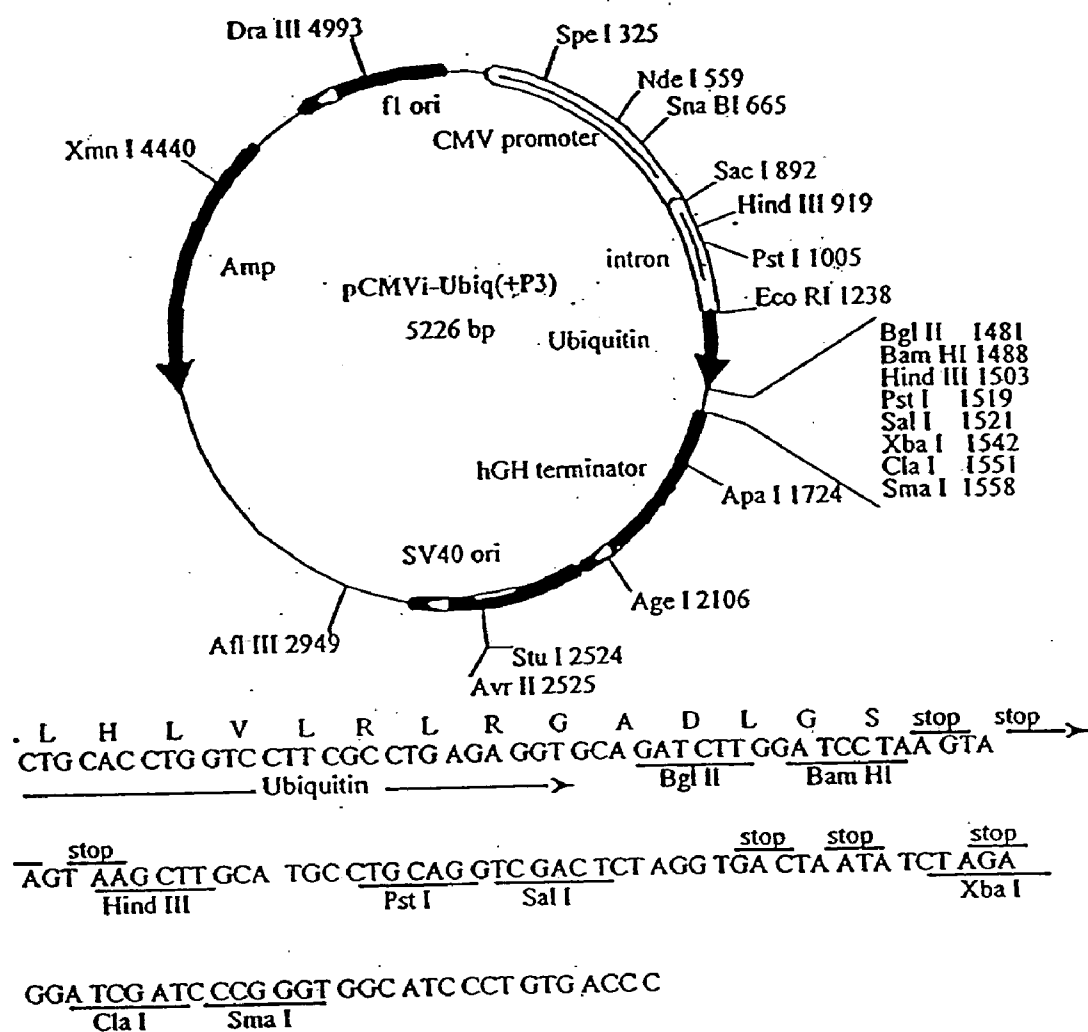


FIG. 2

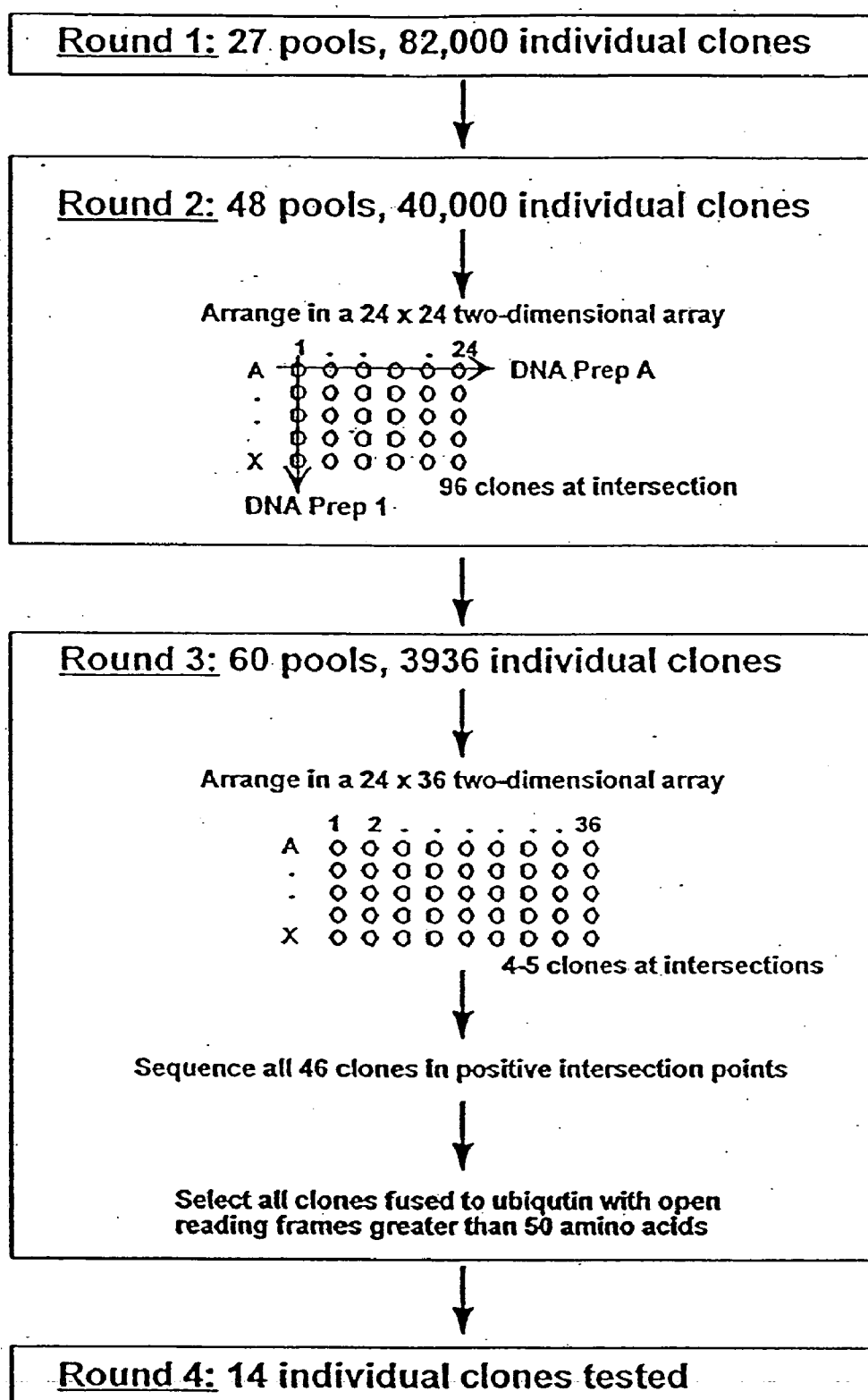


FIG. 3

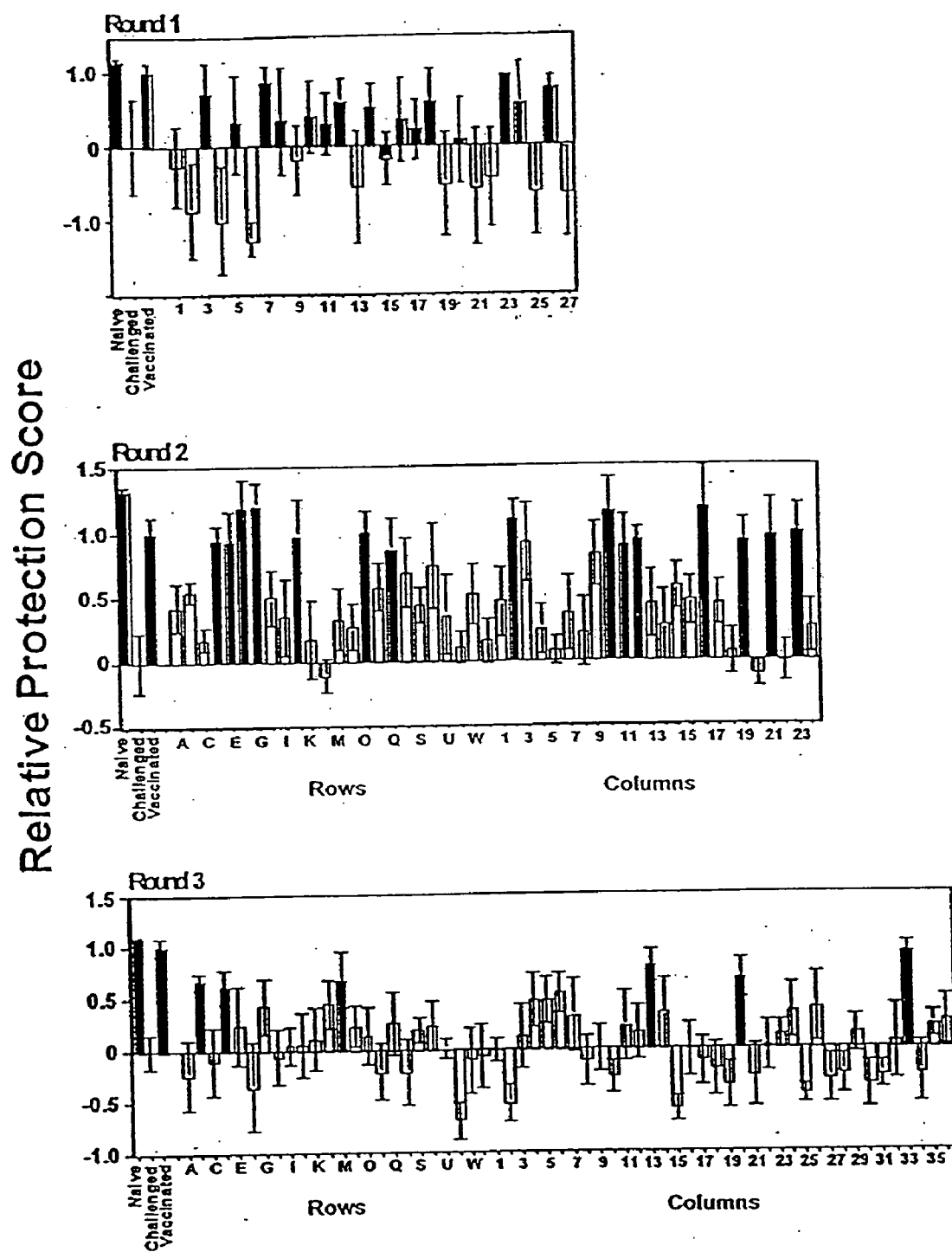


FIG. 4

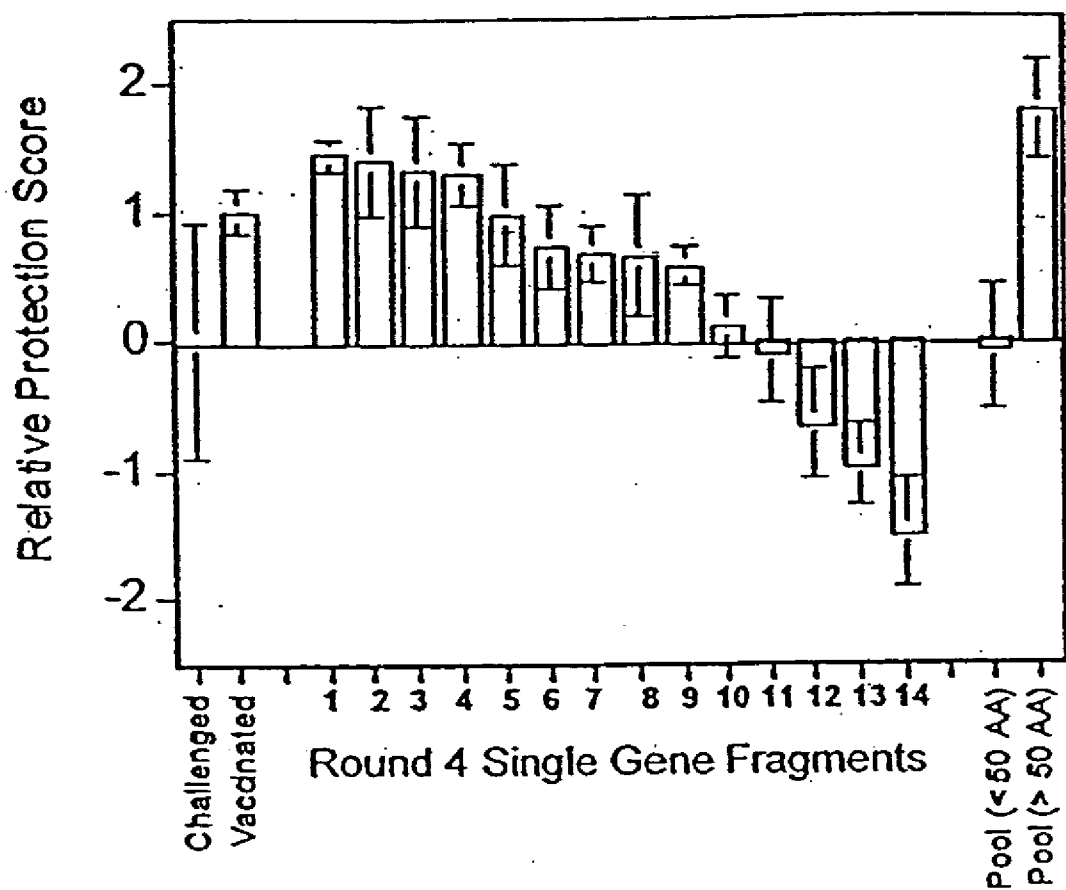


FIG. 5

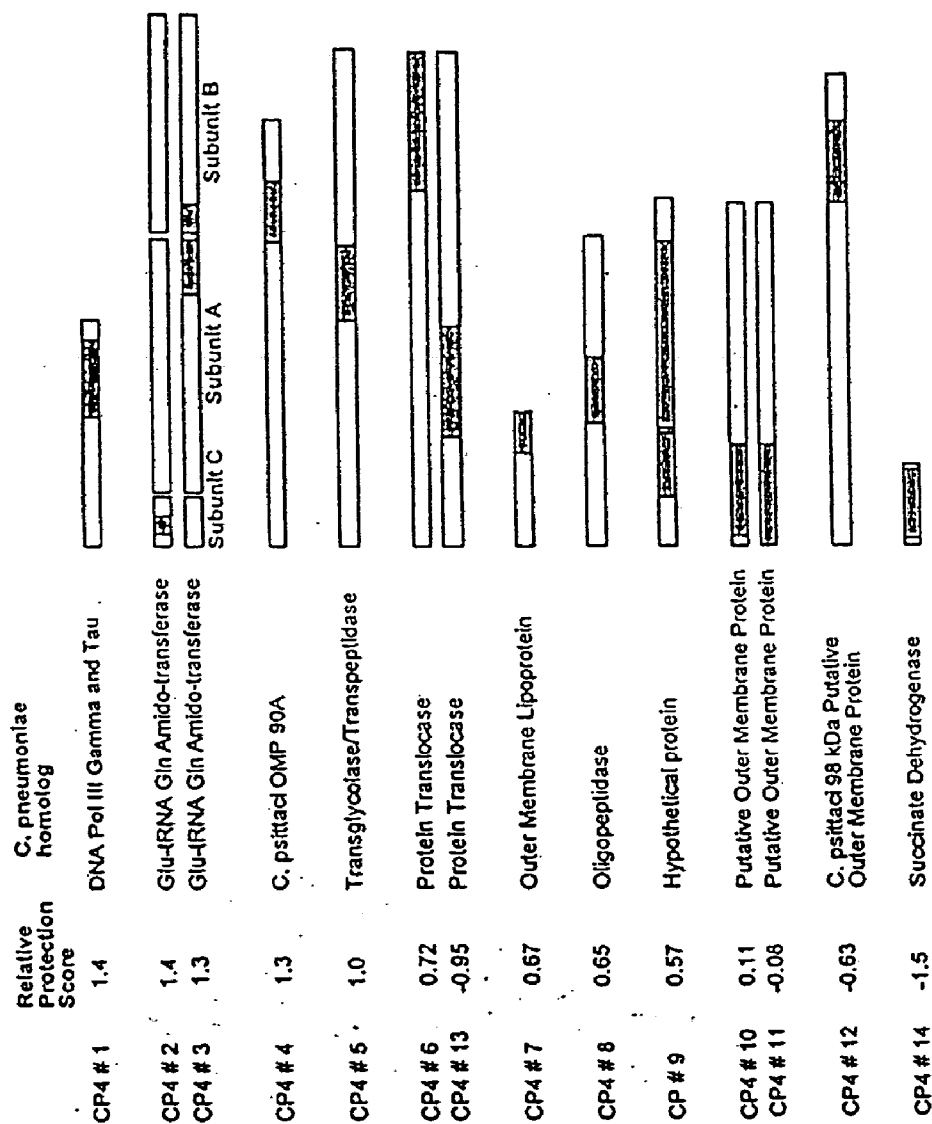


FIG. 6

Chlamydia psittaci Addition Experiments

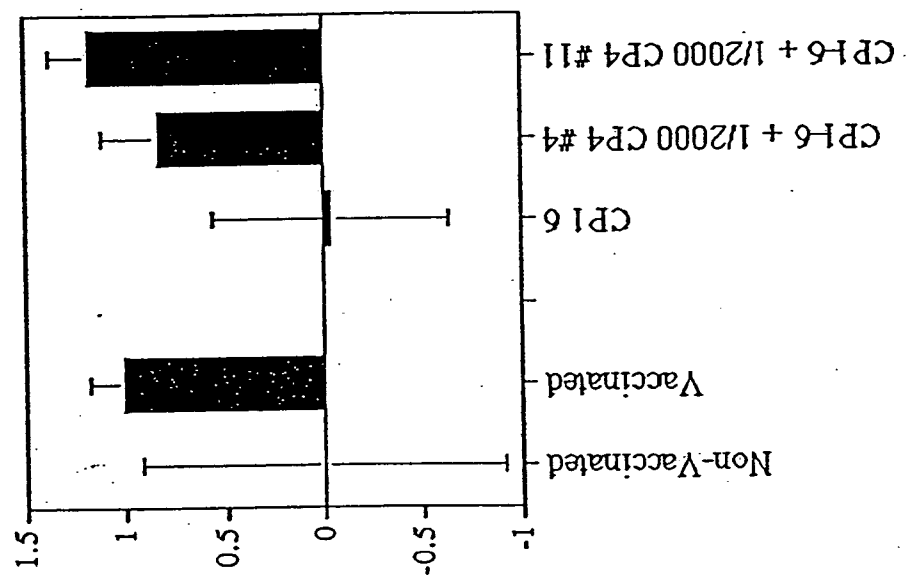
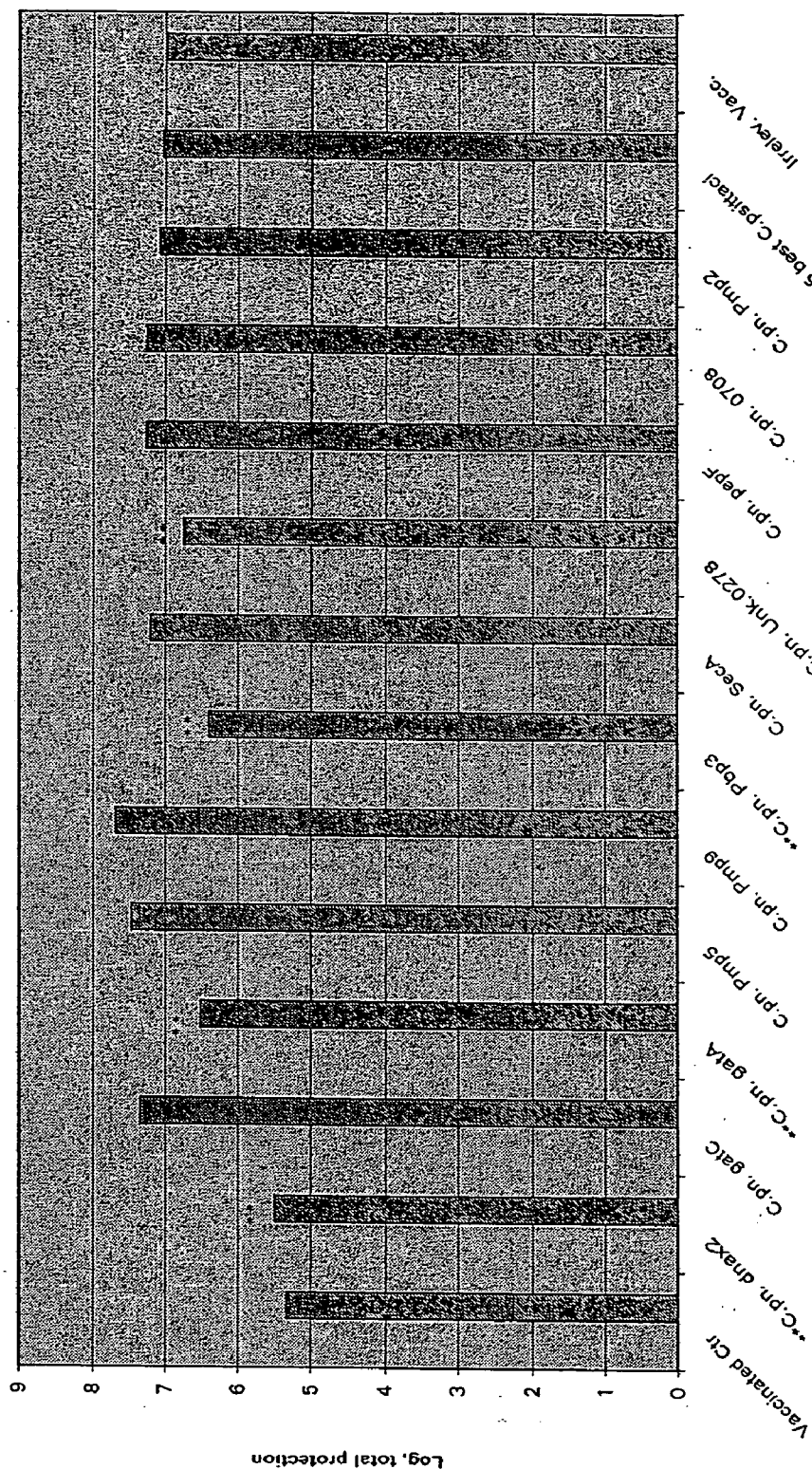


FIG. 7

Protection against *C. Pneumoniae*



Gene's Name

FIG. 8

METHODS AND COMPOSITIONS FOR VACCINATION COMPRISING NUCLEIC ACID AND/OR POLYPEPTIDE SEQUENCES OF CHLAMYDIA

BACKGROUND OF THE INVENTION

[0001] The present application claims priority to co-pending U.S. Provisional Patent Application Serial No: 60/225,839 filed on Dec. 15, 2000. The entire text of the above-referenced disclosure is specifically incorporated herein by reference without disclaimer. The government owns rights in the present invention pursuant to DARPA grant number MDA 972-97-1-0013.

[0002] 1. Field of the Invention

[0003] The present invention relates generally to the fields of immunology, bacteriology and molecular biology. More particularly, the invention relates to methods for screening and obtaining vaccines generated from the administration of expression libraries constructed from a *Chlamydia psittaci* genome or corresponding homologs from other *Chlamydia* species. In particular embodiments, it concerns methods and compositions for the vaccination of vertebrate animals against *Chlamydia* bacterial infections, wherein vaccination of the animal is via a protein or gene derived from part or all of the genes validated as vaccines.

[0004] 2. Description of Related Art

[0005] Intracellular bacteria of the genus *Chlamydia* are important pathogens in both man and vertebrate animals, causing blindness in man, sexually transmitted disease, and community-acquired pneumonia, and most likely act as co-factors in atherosclerotic plaque formation in human coronary heart disease.

[0006] Ubiquitous *Chlamydia* (*C. psittaci*) infections in cattle cause mastitis, infertility and abortion. A primary economic impact of *Chlamydia* in dairy cattle is the loss of milk production and quality. Serological evidence for infection with ruminant *Chlamydia psittaci* is found in virtually all cattle (Kaltenbock et al., 1997). These infections typically do not cause overt signs of disease, but under stress of the host animal may elicit transient inflammation of the mammary gland and uterus. These stress-related herd health problems, while not clinically pronounced, result in major losses for animal agriculture due to reduced output and quality of animal products like milk.

[0007] Most existing vaccines for the treatment of bacterial infections are composed of live/attenuated or killed pathogens (Babiuk, 1999). Live/attenuated vaccines present the risk of residual, or reacquisition of, pathogenicity, and are associated with a high cost of production. In addition, efficacious live/attenuated vaccines cannot be developed against many pathogens, or are impractical to produce. Killed pathogens typically have less utility than live/attenuated vaccines, as they are not usually effective in eliciting cellular immune responses. An alternative is subunit vaccines that consist of one or a few proteins of the pathogen (Babiuk, 1999; Ellis, 1999). The proteins being developed for these vaccines are typically based on a dominant immune response in infected hosts, and/or on surmised importance in the disease process. Due to the high genetic complexity of bacteria or protozoa, the empirical approach to identify these proteins often requires extensive research on the pathogen's

biology and produces a small, biased set of potential vaccine candidates. However, this is currently the only practical method when proteins are the commodity for testing a vaccine.

[0008] The development of genetic (DNA) immunization (Tang et al., 1992) not only offers a new method of vaccine delivery, but also enables a new, unbiased, approach to vaccine discovery. The inventors have proposed that the whole genome of a pathogen could be searched for protein vaccine candidates by directly assessing protection from challenge, termed expression library immunization (ELI) (U.S. Pat. No. 5,703,057, specifically incorporated herein by reference). It involves making an expression library representing the whole genome of the pathogen in a genetic immunization vector. The library is subdivided into smaller groups, and DNA from each library is used to vaccinate animals that are subsequently challenged. The advantage of this approach is that all of the potentially protective genes could be discovered and used in any useful combination to reconstitute a vaccine devoid of non-protective, immunopathological, or immunosuppressive antigens. The potential of ELI was demonstrated in a murine *Mycoplasma pulmonis* infection, against which random *M. pulmonis* libraries were protective (Barry et al., 1995). Since then, others have reported on protective libraries (Brayton et al., 1998; Piedrafita et al., 1999), but the reduction of these libraries to individual genes has not been demonstrated.

[0009] As described above, the widespread human and animal infections by the genus *Chlamydia* represents a particular challenge for vaccinology. *Chlamydia psittaci* infections in cattle cause mastitis, infertility and abortion. A primary economic impact of *Chlamydia* in dairy cattle is the loss of milk production and quality. Thus, an effective vaccine against *Chlamydia* bacterial infections in cattle would be of great economic importance. However, there presently have been no effective vaccines developed against any *Chlamydia*.

SUMMARY OF THE INVENTION

[0010] The present invention overcomes these difficulties and problems in the art and provides for methods of immunization using *Chlamydia* antigens and polynucleotides. The instant invention relates to antigens and nucleic acids encoding such antigens obtainable by screening a *Chlamydia* genome. In more specific aspects, the invention relates to methods of isolating such antigens and nucleic acids and to methods of using such isolated antigens for producing immune responses. The ability of an antigen to produce an immune response may be employed in vaccination or antibody preparation techniques.

[0011] In some embodiments, the invention relates to isolated polynucleotides having a region that comprises a sequence of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:34, SEQ ID NO:36, SEQ ID NO:38, SEQ ID NO:40, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:50, SEQ ID NO:52, SEQ ID NO:58, SEQ ID NO:60, SEQ ID NO:62, SEQ ID NO:64, SEQ ID NO:66, or SEQ ID NO:68 a complement of any of these sequences, or fragments thereof, or sequences closely related to these

sequences. In some more specific embodiments, the invention relates to such polynucleotides comprising a region having a sequence comprising at least 17, 20, 25, 30, 35, 40, 45, 50, 60, 70, 80, 90, 100, 125, 150, 200, or more contiguous nucleotides in common with at least one of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:34, SEQ ID NO:36, SEQ ID NO:38, SEQ ID NO:40, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:46, SEQ ID NO:58, SEQ ID NO:60, SEQ ID NO:62, SEQ ID NO:64, SEQ ID NO:66, or SEQ ID NO:68 or its complement. Of course, such polynucleotides may comprise a region having all nucleotides in common with at least one of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:34, SEQ ID NO:36, SEQ ID NO:38, SEQ ID NO:40, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:50, SEQ ID NO:52, SEQ ID NO:58, SEQ ID NO:60, SEQ ID NO:62, SEQ ID NO:64, SEQ ID NO:66, or SEQ ID NO:68 or its complement.

[0012] In another aspect, the invention relates to polypeptides having sequences of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:1, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31, SEQ ID NO:33, SEQ ID NO:35, SEQ ID NO:37, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:43, SEQ ID NO:45, SEQ ID NO:47, SEQ ID NO:59, SEQ ID NO:61, SEQ ID NO:63, SEQ ID NO:65, SEQ ID NO:67, or SEQ ID NO:69 or fragments thereof, or sequences closely related to these sequences. The invention also relates to methods of producing such polypeptides using recombinant methods, for example, using the polynucleotides described above.

[0013] The invention relates to antibodies against *Chlamydia psittaci* antigens, including those directed against an antigen having sequences of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31, SEQ ID NO:33, SEQ ID NO:35, SEQ ID NO:37, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:43, SEQ ID NO:45, SEQ ID NO:47, SEQ ID NO:49, SEQ ID NO:51, SEQ ID NO:53, SEQ ID NO:55, SEQ ID NO:5, SEQ ID NO:59, SEQ ID NO:61, SEQ ID NO:63, SEQ ID NO:65, SEQ ID NO:67, or SEQ ID NO:69 or an antigenic fragment thereof, or sequences closely related to these sequences. The antibodies may be polyclonal or monoclonal and produced by methods known in the art.

[0014] The invention contemplates vaccines comprising: (a) a pharmaceutically acceptable carrier, and (b) at least one polynucleotide having a *Chlamydia* sequence. In presently preferred embodiments, the at least one polynucleotide has a *Chlamydia psittaci*, *Chlamydia pneumoniae*, *Chlamydia trachomatis*, or *Chlamydia pecorum* sequence. In some specific embodiments at least one polynucleotide has a *Chlamydia psittaci* or *Chlamydia pneumoniae* sequence.

[0015] The at least one polynucleotide may be isolated from a *Chlamydia* genomic DNA expression library but it need not be. For example, the polynucleotide may also be a

sequence from one species that is determined to be protective based on the protective ability of a homologous sequence in another species. For example, the polynucleotide could be a *Chlamydia pneumoniae* sequence that was determined to be protective after analysis of the *Chlamydia pneumoniae* genomic sequence for homologues of *Chlamydia psittaci* antigens that had previously been shown to be protective. As discussed below, the polynucleotides need not be of natural origin, or to encode an antigen that is precisely a naturally occurring *Chlamydia* antigen. It is anticipated that polynucleotides and antigens within the scope of this invention may be synthetic and/or engineered to mimic, or improve upon, naturally occurring polynucleotides and still be useful in the invention.

[0016] In some embodiments, the at least one polynucleotide has a sequence isolated from *Chlamydia psittaci*, for example, a sequence of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:34, SEQ ID NO:36, SEQ ID NO:38, SEQ ID NO:40, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:50, SEQ ID NO:52, SEQ ID NO:54, SEQ ID NO:56, SEQ ID NO:58, or SEQ ID NO:60, or fragment thereof, or sequences closely related to these sequences. In more specific such embodiments, the at least one polynucleotide has a sequence of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, or SEQ ID NO:26, or fragment thereof, or sequences closely related to these sequences. In even more specific embodiments, the at least one polynucleotide has a sequence of SEQ ID NO:6, SEQ ID NO:10, SEQ ID NO:14, SEQ ID NO:20, or SEQ ID NO:24.

[0017] In some embodiments, the polynucleotide encodes an antigen having a sequence of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31, SEQ ID NO:33, SEQ ID NO:35, SEQ ID NO:37, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:43, SEQ ID NO:45, SEQ ID NO:47, SEQ ID NO:49, SEQ ID NO:51, SEQ ID NO:53, SEQ ID NO:55, SEQ ID NO:57, SEQ ID NO:59, SEQ ID NO:61, SEQ ID NO:63, SEQ ID NO:65, SEQ ID NO:67, or SEQ ID NO:69; or antigenic fragment thereof, or sequences closely related to these sequences.

[0018] In many embodiments, the polynucleotide is comprised in a genetic immunization vector. Such a vector may, but need not, comprise a gene encoding a mouse ubiquitin fusion polypeptide. The vector, in some preferred embodiments, will comprise a promoter operable in eukaryotic cells, for example, but not limited to a CMV promoter. Such promoters are well known to those of skill in the art. In some embodiments, the polynucleotide is comprised in a viral expression vector, for example, but not limited to, a vector selected from the group consisting of adenovirus, adeno-associated virus, retrovirus and herpes-simplex virus.

[0019] The vaccines of the invention may comprise multiple polynucleotide sequences from one or more *Chlamydia* species. In some embodiments, the vaccine will comprise at

least a first polynucleotide having a *Chlamydia psittaci* sequence and a second polynucleotide having a sequence, wherein the first polynucleotide and the second polynucleotide have different sequences. In some more specific embodiments, the first polynucleotide may have a sequence of SEQ ID NO:50.

[0020] The present invention also involves vaccines comprising: (a) a pharmaceutically acceptable carrier; and (b) at least one *Chlamydia* antigen. The at least one *Chlamydia* antigen can be a *Chlamydia psittaci*, *Chlamydia pneumoniae*, *Chlamydia trachomatis*, or *Chlamydia pecorum* antigen or an antigen of any other *Chlamydia* species. In some cases, the at least one *Chlamydia* antigen is a *Chlamydia psittaci* antigen. In some embodiments, the at least one *Chlamydia* antigen has a sequence of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31, SEQ ID NO:33, SEQ ID NO:35, SEQ ID NO:37, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:43, SEQ ID NO:45, SEQ ID NO:47, SEQ ID NO:49, SEQ ID NO:51, SEQ ID NO:53, SEQ ID NO:55, SEQ ID NO:57, SEQ ID NO:59, SEQ ID NO:61, SEQ ID NO:63, SEQ ID NO:65, SEQ ID NO:67, or SEQ ID NO:69 or antigenic fragment thereof, or sequences closely related to these sequences. In some specific embodiments, the at least one *Chlamydia* antigen has a sequence of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, or SEQ ID NO:27, or an antigenic fragment thereof, or sequences closely related to these sequences. In even more specific embodiments, the at least one *Chlamydia* antigen has a sequence of SEQ ID NO:7, SEQ ID NO:11, SEQ ID NO:15, SEQ ID NO:21, or SEQ ID NO:25.

[0021] The invention also relates to methods of immunizing an animal comprising providing to the animal at least one *Chlamydia* antigen, or antigenic fragment thereof, in an amount effective to induce an immune response. Again, the at least one *Chlamydia* antigen can be of *Chlamydia psittaci*, *Chlamydia pneumoniae*, *Chlamydia trachomatis*, *Chlamydia pecorum* or any other *Chlamydia* species. In some cases, the at least one *Chlamydia* antigen is a *Chlamydia psittaci* antigen, while in others it will not be. In further examples the *Chlamydia pneumoniae* antigens are comprised of SEQ ID NO: 63; SEQ ID NO: 65; SEQ ID NO: 675; SEQ ID NO: 69: As discussed above, and described in detail below, the *Chlamydia* antigens useful in the invention need not be native antigens. Rather, these antigens may have sequences that have been modified in any number of ways known to those of skill in the art, -so long as they result in or aid in an antigenic response.

[0022] In some embodiments of the invention, the provision of the at least one *Chlamydia* antigen comprises: (a) preparing a cloned expression library from fragmented genomic DNA, cDNA or sequenced genes of *Chlamydia*; (b) administering at least one clone of the library in a pharmaceutically acceptable carrier into the animal; and (c) expressing at least one *Chlamydia* antigen in the animal. The expression library may comprise at least one or more polynucleotides having a sequence of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28,

SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:34, SEQ ID NO:36, SEQ ID NO:38, SEQ ID NO:40, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:50, SEQ ID NO:52, SEQ ID NO:54, SEQ ID NO:56, SEQ ID NO:58, or SEQ ID NO:60, SEQ ID NO:62; SEQ ID NO:64; SEQ ID NO:66; or SEQ ID NO:68; or fragment thereof, or sequences closely related to these sequences. The expression library may be cloned in a genetic immunization vector, such as a vector of SEQ ID NO:1, or any other suitable vector. The vector may comprise a gene encoding a mouse ubiquitin fusion polypeptide designed to link the expression library polynucleotides to the ubiquitin gene. The vector may comprise a promoter operable in eukaryotic cells, for example a CMV promoter, or any other suitable promoter. In such methods, the polynucleotide may be administered by a intramuscular injection or epidermal injection. The polynucleotide may likewise be administered by intravenous, subcutaneous, intralesional, intraperitoneal, oral or inhaled routes of administration. In some specific, exemplary embodiments, the administration may be via intramuscular injection of at least 1.0 µg to 200 µg of the polynucleotide. In other exemplary embodiments, administration may be epidermal injection of at least 0.01 µg to 5.0 µg of the polynucleotide. In some cases, a second administration, for example, an intramuscular injection and/or epidermal injection, may administered at least about three weeks after the first administration. In these methods, the polynucleotide may be, but need not be, cloned into a viral expression vector, for example, a viral expression vector selected from the group consisting of adenovirus, herpes-simple virus, retrovirus and adeno-associated virus. The polynucleotide may also be administered in any other method disclosed herein or known to those of skill in the art.

[0023] In some embodiments, the provision of the *Chlamydia* antigen(s) may comprise: (a) preparing a pharmaceutical composition comprising at least one polynucleotide * encoding a *Chlamydia* antigen or fragment thereof, (b) administering one or more clones of the library in a pharmaceutically acceptable carrier into the animal; and (c) expressing one or more *Chlamydia* antigens in the animal. The one or more polynucleotides can be comprised in one or more expression vectors, as described above and elsewhere in this specification.

[0024] Alternatively, the provision of the *Chlamydia* antigen(s) may comprise: (a) preparing a pharmaceutical composition of at least one *Chlamydia* antigen or an antigenic fragment thereof; and (b) administering the at least one antigen or fragment into the animal. The antigen(s) may be administered by a first intramuscular injection, intravenous injection, parenteral injection, epidermal injection, inhalation or oral route.

[0025] In preferred embodiments of the invention, the animal is a mammal. In some cases the mammal is a bovine, in others, the mammal is a human.

[0026] In some embodiments, these methods may induce an immune response against *Chlamydia psittaci*. Alternatively; these methods may be practiced in order to induce an immune response against a *Chlamydia* species other than *Chlamydia psittaci*, for example, but not limited to, *Chlamydia pneumoniae*, *Chlamydia trachomatis*, and/or *Chlamydia pecorum*. In some embodiments, these methods may be employed to induce an immune response against a non-*Chlamydia* infection or other disease.

[0027] These methods may comprise administering to the animal an antigen or antigenic fragment from a *Chlamydia* species other than *Chlamydia psittaci*. Also, these methods may comprise administering to the animal an antigen or antigenic fragment from a non-*Chlamydia* species.

[0028] This specification discusses methods of obtaining polynucleotide sequences effective for generating an immune response against the genus *Chlamydia* in a non-human animal comprising: (a) preparing a cloned expression library from fragmented genomic DNA of the genus *Chlamydia*; (b) administering one or more clones of the library in a pharmaceutically acceptable carrier into the animal in an amount effective to induce an immune response; and (c) selecting from the library the polynucleotide sequences that induce an immune-response, wherein the immune response in the animal is protective against *Chlamydia* infection. Such methods may further comprise testing the animal for immune resistance against a *Chlamydia* bacterial infection by challenging the animal with *Chlamydia*. In some cases, the genomic DNA has been fragmented physically or by restriction enzymes, for example, but not limited to, fragments that average, about 200-1000 base pairs in length. In some cases, each clone in the library may comprise a gene encoding a mouse ubiquitin fusion polypeptide designed to link the expression library polynucleotides to the ubiquitin gene, but this is not required in all cases. In some cases, the library may comprise about 1×10^3 to about 1×10^6 clones; in more specific cases, the library could have 1×10^5 clones. In some preferred methods, about 0.01 μg to about 200 μg of DNA, from the clones is administered into the animal. In some situations the genomic DNA, cDNA or sequenced gene is introduced by intramuscular injection or epidermal injection. In some versions of these protocols, the cloned expression library further comprises a promoter operably linked to the DNA that permits expression in a vertebrate animal cell.

[0029] The application also discloses methods of preparing antigens that confer protection against infection in a vertebrate animal comprising the steps of: (a) preparing a cloned expression library from fragmented genomic DNA of the genus *Chlamydia*; (b) administering one or more clones of the library in a pharmaceutically acceptable carrier into the animal in an amount effective to induce an immune response; (c) selecting from the library the polynucleotide sequences that induce an immune response and expressing the polynucleotide sequences in cell culture; and (d) purifying the polypeptide(s) expressed in the cell culture. Often, these methods further comprise testing the animal for immune resistance against infection by challenging the animal with one or more *Chlamydia* or other pathogens.

[0030] The invention relates to methods of preparing antibodies against a *Chlamydia* antigen comprising the steps of: (a) identifying a *Chlamydia* antigen that confers immune resistance against *Chlamydia* bacterial infection when challenged with the *Chlamydia* species in which the antigen was prepared; (b) generating an immune response in a vertebrate animal with the antigen identified in step (a); and (c) obtaining antibodies produced in the animal.

[0031] The invention also relates to methods of assaying for the presence of *Chlamydia* infection in a vertebrate animal comprising: (a) obtaining an antibody directed against a *Chlamydia* antigen; (b) obtaining a sample from

the animal; (c) admixing the antibody with the sample; and (d) assaying the sample for antigen-antibody binding, wherein the antigen-antibody binding indicates *Chlamydia* infection in the animal. In some cases, the antibody directed against the antigen is further defined as a polyclonal antibody. In others, the antibody directed against the antigen is further defined as a monoclonal antibody. In some embodiments, the *Chlamydia* antigen has a sequence of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31, SEQ ID NO:33, SEQ ID NO:35, SEQ ID NO:37, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:43, SEQ ID NO:45, SEQ ID NO:47, SEQ ID NO:49, SEQ ID NO:51, SEQ ID NO:53, SEQ ID NO:55, SEQ ID NO:57, SEQ ID NO:59, SEQ ID NO:61, SEQ ID NO:63, SEQ ID NO:65, SEQ ID NO:67, or SEQ ID NO:69; or fragment thereof, or sequences closely related to these sequences. The assaying the sample for antigen-antibody binding may be by precipitation reaction, radioimmunoassay, ELISA, Western blot, immunofluorescence, or any other method known to those of skill in the art.

[0032] The invention also relates to kits for assaying a *Chlamydia* infection comprising, in a suitable container: (a) a pharmaceutically acceptable carrier; and (b) an antibody directed against a *Chlamydia* antigen.

[0033] The invention further relates to methods of assaying for the presence of a *Chlamydia* infection in an animal comprising: (a) obtaining an oligonucleotide probe comprising a sequence comprised within one of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:34, SEQ ID NO:36, SEQ ID NO:38, SEQ ID NO:40, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:50, SEQ ID NO:52, SEQ ID NO:54, SEQ ID NO:56, SEQ ID NO:58, or SEQ ID NO:60, SEQ ID NO:62, SEQ ID NO:64, SEQ ID NO:66, or SEQ ID NO:68; or a complement thereof; and (b) employing the probe in a PCR or other detection protocol.

[0034] As used herein in the specification, "a" or "an" may mean one or more. As used herein, when used in conjunction with the word "comprising", the words "a" or "an" may mean one or more than one. As used herein "another" may mean at least a second or more.

[0035] As used herein, "plurality" means more than one. In certain specific aspects, a plurality may mean 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 125, 150, 175, 200, 250, 300, 400, 500, 750, 1,000, 2,000, 3,000, 4,000, 5,000, 7,500, 10,000, 15,000, 20,000, 30,000, 40,000, 50,000, 60,000, 70,000, 80,000, 90,000, 100,000, 125,000, 150,000, 200,000 or more, and any integer derivable therein, and any range derivable therein.

[0036] As used herein, "any integer derivable therein" means a integer between the numbers described in the specification, and "any range derivable therein" means any range selected from such numbers or integers.

[0037] As used herein, a “fragment” refers to a sequence having or having at least 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 105, 110, 115, 120, 125, 130, 135, 140, 145, 150, 155, 160, 165, 170, 175, 180, 185, 190, 195, 200, 210, 220, 230, 240, 250, 260, 270, 280, 290, 300, 310, 320, 330, 340, 350, 360, 370, 380, 390, 400, 410, 420, 430, 440, 450, 460, 470, 480, 490, 500, 510, 520, 530, 540, 550, 560, 570, 580, 590, 600, 610, 620, 630, 640, 650, 660, 670, 680, 690, 700, 710, 720, 730, 740, 750, 760, 770, 780, 790, 800, 810, 820, 830, 840, 850, 860, 870, 890, 900, 910, 920, 930, 940, 950, 960, 970, 980, 990, 1000 or more contiguous residues of the recited SEQ ID NOS, but less than the full-length of the SEQ. ID. NOS.. It is contemplated that the definition of “fragment” can be applied to amino acid and nucleic acid fragments.

[0038] As used herein, an “antigenic fragment” refers to a fragment, as defined above, that can elicit an immune response in an animal.

[0039] Reference to a sequence in an organism, such as a “*Chlamydia* sequence” refers to a segment of contiguous residues that is unique to that organism or that constitutes a fragment (or full-length region(s)) found in that organism (either amino acid or nucleic acid).

BRIEF DESCRIPTION OF THE DRAWINGS

[0040] The following drawings form part of the present specification and are included to further demonstrate certain aspects of the present invention. The invention may be better understood by reference to one or more of these drawings in combination with the detailed description of specific embodiments presented herein.

[0041] FIG. 1. Scheme for Expression Library Immunization.

[0042] FIG. 2. Production of the *Chlamydia psittaci* Library. The *Chlamydia psittaci* library was produced by first physically shearing the genomic DNA, strain BGM/B577, and size selecting fragments of 300-800 base pairs. The fragments were ligated into the Bgl II site of pCMV-Ubs(+P3); see Sykes and Johnston, 1999 for details. The nucleotide sequence shown in this figure is given as SEQ ID NO:1.

[0043] FIG. 3. Flowchart depicting the process for deconvolution of the libraries. Each round consists of preparation of DNA samples, vaccination of mice, challenge and determination of the relative protection in each group.

[0044] FIG. 4. Results of protection assays in Rounds 1, 2 and 3. Protection was scored as lung weight relative to average of the vaccinated, maximum protection, positive control and the non-vaccinated, challenged, maximum disease, negative control. The relative protection score was calculated by assigning the score 1 to animals with lung weight equal to the vaccinated control and the score 0 to animals with lung weights equal to the challenged, non-vaccinated control. These points define a line; animals with lower lung weight, hence better protection, have a higher relative protection score. Animals that have worse disease than challenged, non-vaccinated controls, i.e. heavier lungs, will have a negative relative protection score. The unchallenged Naive group consistently had lung weights slightly lower than the maximum protection, positive controls (Vaccinated) due to the peribronchiolar accumulation of lymphatic cells.

In Rounds 2 and 3 the pools of plasmids from columns in the two-dimensional arrays are assigned numbers and the rows assigned letters. The solid bars indicate pools that were designated as protective and entered into the subsequent round. The error bars represent one standard deviation on either side of the mean.

[0045] FIG. 5. Results of protection assays of testing individual gene fragments in Round 4. Protection was scored as lung weight relative to the average of the vaccinated, maximum protection, positive control (Vaccinated=1) and the non-vaccinated, challenged, maximum disease, negative control (Challenged=0). The Pool<50AA is the DNA consisting of the pool of the 32 plasmids from Round 3 having predicted open-reading frames less than 50 amino acids long. Pool>50AA is the DNA consisting of all the 14 plasmids containing *Chlamydia psittaci* inserts encoding in-frame proteins more than 50 amino acids long. The numbers of each individual gene fragment tested correspond to the numbers in FIG. 4. The error bars represent one standard deviation of the mean.

[0046] FIG. 6. Summary of characterization of the single gene fragments of Round 4. The Relative Protection score of each *Chlamydia psittaci* (CP) gene fragment is provided along with the designation of the gene in *Chlamydia pneumonia* that has the highest similarity (*Chlamydia pneumonia* homologue). In two cases, gene fragment CP #4 and CP #12, the *Chlamydia psittaci* gene could also be identified. On the right is a linear map showing the location in each gene of the fragment that conferred protection (shaded).

[0047] FIG. 7. Protection data from DNA pools. CP1-6 is a negative pool from round 1. To test whether a single protective gene could be detected in a negative pool, 25 ng of either CP4 #4 or CP4 #11 was added to 50 µg of CP1-6.

[0048] FIG. 8. Protection against *Chlamydia pneumoniae* challenge by various homologs of *Chlamydia pneumoniae* from ELI-selected *Chlamydia psittaci* (CP) gene.

DESCRIPTION OF ILLUSTRATIVE EMBODIMENTS

[0049] The widespread human and animal infections by the genus *Chlamydia* represents a particular challenge for vaccinology. For example, *Chlamydia psittaci* infections in cattle cause mastitis, infertility and abortion. A primary economic impact of *Chlamydia* in dairy cattle is the loss of milk production and quality. Thus, an effective treatment for *Chlamydia* bacterial infections in human and other vertebrate animals would be of clinical and economic importance.

[0050] The present invention provides compositions and methods for the immunization of vertebrate animals, including humans, against infections using nucleic acid sequences and polypeptides elucidated by screening *Chlamydia psittaci*. These compositions and methods will be useful for immunization against *Chlamydia psittaci* bacterial infections and other infections and disease states. In particular embodiments, a vaccine composition directed against *Chlamydia* infections is provided. The vaccine according to the present invention comprises *Chlamydia* genes and polynucleotides identified by the inventors, that confer protective resistance in vertebrate animals to *Chlamydia* bacterial infections, and other infections. In other embodiments, the

invention provides methods for immunizing an animal against *Chlamydia* infections, methods for preparing a cloned library via expression library immunization and methods for screening and identifying *Chlamydia* genes that confer protection against infection.

A. EXPRESSION LIBRARY IMMUNIZATION

[0051] In particular embodiments, the immunization of vertebrate animals according to the present invention includes a cloned library of *Chlamydia* expression constructs. In specific embodiments, a cloned expression library of *Chlamydia psittaci* is provided. Expression library immunization, ELI herein, is well known in the art (U.S. Pat. No. 5,703,057, specifically incorporated herein by reference). In certain embodiments, the invention provides an ELI method applicable to virtually any pathogen and requires no knowledge of the biological properties of the pathogen. The method operates on the assumption, generally accepted by those skilled in the art, that all the potential antigenic determinants of any pathogen are encoded in its genome. The inventors have previously devised methods of identifying vaccines using a genomic expression library representing all of the antigenic determinants of a pathogen (U.S. Pat. No. 5,703,057). The method uses to its advantage the simplicity of genetic immunization to sort through a genome for immunological reagents in an unbiased, systematic fashion.

[0052] The preparation of an expression library is performed using the techniques and methods familiar one of skill in the art. The pathogen's genome, may or may not be known or possibly may even have been cloned. Thus one obtains DNA (or cDNA), representing substantially the entire genome of the pathogen (e.g., *Chlamydia psittaci*). The DNA is broken up, by physical fragmentation or restriction endonuclease, into segments of some length so as to provide a library of about 10^5 (approximately $18\times$ the genome size) members. The library is then tested by inoculating a subject with purified DNA of the library or sub-library and the subject challenged with a pathogen, wherein immune protection of the subject from pathogen challenge indicates a clone that confers a protective immune response against infection.

B. NUCLEIC ACIDS

[0053] The present invention provides *Chlamydia* polynucleotide compositions and methods that induce a protective immune response in vertebrate animals challenged with a *Chlamydia* bacterial infection. The preparation and purification of antigenic *Chlamydia* polypeptides, or fragments thereof (Section C) and antibody preparations directed against *Chlamydia* antigens, or fragments thereof (Section E) are described below.

[0054] Thus, in certain embodiments of the present invention, genes or polynucleotides encoding *Chlamydia* polypeptides or fragments thereof are provided. It is contemplated in other embodiments, that a polynucleotide encoding a *Chlamydia* polypeptide or polypeptide fragment will be expressed in prokaryotic or eukaryotic cells and the polypeptides purified for use as anti-*Chlamydia* antigens in the vaccination of vertebrate animals or in generating antibodies immunoreactive with *Chlamydia* polypeptides (i.e., antigens). The genomes of *Chlamydia pneumoniae* and

Chlamydia trachomatis have been completely sequenced. The *Chlamydia* genes are quite similar, with the four most protective genes identified being 30-71% identical and 45-85% similar in amino acid sequence.

[0055] Genes for various species of the genus *Chlamydia* have been cloned, identified and compared (Kalman et al., 1999; Meijer et al., 1999). For example, the genomes of *Chlamydia pneumoniae*, *Chlamydia trachomatis*, *Chlamydia psittaci* and *Chlamydia pecorum* have been studied. The present invention is not limited in scope to the genes of *Chlamydia psittaci*, however, as one of ordinary skill in the art could, using these nucleic acids, readily identify related homologues in various other species. In addition, it should be clear that the present invention is not limited to the specific nucleic acids disclosed herein. As discussed below, a specific "*Chlamydia*" gene or polynucleotide fragment may contain a variety of different bases and yet still produce a corresponding polypeptide that is functionally indistinguishable, and in some cases structurally indistinguishable, from the polynucleotide sequences disclosed herein by reference in U.S. patent application Ser. No: 09/738,269 filed on Dec. 15, 2000.

[0056] 1. Nucleic Acids Encoding *Chlamydia* Polypeptides

[0057] The present invention provides polynucleotides encoding antigenic *Chlamydia psittaci* polypeptides capable of inducing a protective immune response in vertebrate animals and for use as an antigen to generate anti-*Chlamydia psittaci* or other pathogen antibodies. In certain instances, it may be desirable to express *Chlamydia psittaci* polynucleotides encoding a particular antigenic *Chlamydia psittaci* polypeptide domain or sequence to be used as a vaccine or in generating anti-*Chlamydia psittaci* or other pathogen antibodies. Nucleic acids according to the present invention may encode an entire *Chlamydia psittaci* gene, or any other fragment of the *Chlamydia psittaci* sequences set forth herein. The nucleic acid may be derived from genomic DNA, i.e., cloned directly from the genome of a particular organism. In other embodiments, however, the nucleic acid may comprise complementary DNA (cDNA). A protein may be derived from the designated sequences for use in a vaccine or to isolate useful antibodies.

[0058] The term "cDNA" is intended to refer to DNA prepared using messenger RNA (mRNA) as template. The advantage of using a cDNA, as opposed to genomic DNA or DNA polymerized from a genomic, non- or partially-processed RNA template, is that the cDNA primarily contains coding sequences of the corresponding protein. There may be times when the full or partial genomic sequence is preferred, such as where the non-coding regions are required for optimal expression.

[0059] It also is contemplated that a given *Chlamydia* polynucleotide from a given species may be represented by natural variants that have slightly different nucleic acid sequences but, nonetheless, encode the same polypeptide (see Table 2 below). In addition, it is contemplated that a given *Chlamydia* polypeptide from a species may be generated using alternate codons that result in a different nucleic acid sequence but encodes the same polypeptide.

[0060] As used in this application, the term "a nucleic acid encoding a *Chlamydia* polynucleotide" refers to a nucleic

acid molecule that has been isolated tree of total cellular nucleic acid. The term “functionally equivalent codon” is used herein to refer to codons that encode the same amino acid, such as the six codons for arginine or serine (Table 2, below), and also refers to codons that encode biologically equivalent amino acids, as discussed in the following pages.

TABLE 1

Amino Acids		Codons			
Alanine	Ala	A	GCA	GCC	GCG GCU
Cysteine	Cys	C	UGC	UGU	
Aspartic acid	Asp	D	GAC	GAU	
Glutamic acid	Glu	E	GAA	GAG	
Phenylalanine	Phe	F	UUC	UUU	
Glycine	Gly	G	GGA	GGC	GGG GGU
Histidine	His	H	CAC	CAU	
Isoleucine	Ile	I	AUA	AUC	AUU
Lysine	Lys	K	AAA	AAG	
Leucine	Leu	L	UUA	UUG	CUA CUC CUG CUU
Methionine	Met	M	AUG		
Asparagine	Asn	N	AAC	AAU	
Proline	Pro	P	CCA	CCC	CCG CCU
Glutamine	Gln	Q	CAA	CAG	
Arginine	Arg	R	AGA	AGG	CGA CGC CGG CGU
Serine	Ser	S	AGC	AGU	UCA UCC UCG UCU
Threonine	Thr	T	ACA	ACC	ACG ACU
Valine	Val	V	GUA	GUC	GUG GUU
Tryptophan	Trp	W	UGG		
Tyrosine	Tyr	Y	UAC	UAU	

[0061] Allowing for the degeneracy of the genetic code, sequences that have at least about 50%, usually at least about 60%, more usually about 10%, most usually about 80%, preferably at least about 90% and most preferably about 95% of nucleotides that are identical to the nucleotides of given *Chlamydia* gene or polynucleotide. Sequences that are essentially the same as those set forth in a *Chlamydia* gene or polynucleotide may also be functionally defined as sequences that are capable of hybridizing to a nucleic acid segment containing the complement of a *Chlamydia* polynucleotide under standard conditions.

[0062] The DNA segments of the present invention include those encoding biologically functional equivalent *Chlamydia* proteins and peptides, as described above. Such sequences may arise as a consequence of codon redundancy and amino acid functional equivalency that are known to occur naturally within nucleic acid sequences and the proteins thus encoded. Alternatively, functionally equivalent proteins or peptides may be created via the application of recombinant DNA technology, in which changes in the protein structure may be engineered, based on consider-

ations of the properties of the amino acids being exchanged. Changes designed by man may be introduced through the application of site-directed mutagenesis techniques or may be introduced randomly and screened later for the desired function, as described below.

[0063] 2. Oligonucleotide Sequences

[0064] Naturally, the present invention also encompasses DNA segments that are complementary, or essentially complementary to the sequences of a *Chlamydia* polynucleotide. Nucleic acid sequences that are “complementary” are those that are capable of base-pairing according to the standard Watson-Crick complementary rules. As used herein, the term “complementary sequences means nucleic acid sequences that are substantially complementary, as may be assessed by the same nucleotide comparison set forth above, or as defined as being capable of hybridizing to the nucleic acid segment of a *Chlamydia* polynucleotide under relatively stringent conditions such as those described herein. Such sequences may encode the entire *Chlamydia* polypeptide or functional or non-functional fragments thereof.

[0065] Alternatively, the hybridizing segments may be shorter oligonucleotides. Sequences of 17 bases long should occur only once in the human genome and, therefore, suffice to specify a unique target sequence. Although shorter oligomers are easier to make and increase in vivo accessibility, numerous other factors are involved in determining the specificity of hybridization. Both binding affinity and sequence specificity of an oligonucleotide to its complementary target increases with increasing length. It is contemplated that exemplary oligonucleotides of 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100 or more base pairs will be used, although others are contemplated. Longer polynucleotides encoding 250, 500, 1000, 1212, 1500, 2000, 2500, 3000 or 3500 bases and longer are contemplated as well. Such oligonucleotides will find use, for example, as probes in Southern and Northern blots and as primers in amplification reactions, or for vaccines.

[0066] Suitable hybridization conditions will be well known to those of skill in the art. In certain applications, for example, substitution of amino acids by site-directed mutagenesis, it is appreciated that lower stringency conditions are required. Under these conditions, hybridization may occur even though the sequences of probe and target strand are not perfectly complementary, but are mismatched at one or more positions. Conditions may be rendered less stringent by increasing salt concentration and decreasing temperature. For example, a medium stringency condition could be provided by about 0.1 to 0.25 M NaCl at temperatures of about 37° C. to about 55° C., while a low stringency condition could be provided by about 0.15 M to about 0.9 M salt, at temperatures ranging from about 20° C. to about 55° C. Thus, hybridization conditions can be readily manipulated, and thus will generally be a method of choice depending on the desired results.

[0067] In other embodiments, hybridization may be achieved under conditions of, for example, 50 mM Tris-HCl (pH 8.3), 75 mM KCl, 3 mM MgCl₂, 10 mM dithiothreitol, at temperatures between approximately 20° C. to about 37° C. Other hybridization conditions utilized could include approximately 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 1.5

mM MgCl₂, at temperatures ranging from approximately 40° C. to about 72° C. Formamide and SDS also may be used to alter the hybridization conditions.

[0068] One method of using probes and primers of the present invention is in the search for genes related to *Chlamydia* or, more particularly, homologues of *Chlamydia* from other species. Normally, the target DNA will be a genomic or cDNA library, although screening may involve analysis of RNA molecules. By varying the stringency of hybridization, and the region of the probe, different degrees of homology may be discovered.

[0069] Another way of exploiting probes and primers of the present invention is in site-directed, or site-specific mutagenesis. Site-specific mutagenesis is a technique useful in the preparation of individual peptides, or biologically functional equivalent proteins or peptides, through specific mutagenesis of the underlying DNA. The technique further provides a ready ability to prepare and test sequence variants, incorporating one or more of the foregoing considerations, by -introducing one or more nucleotide sequence changes into the DNA. Site-specific mutagenesis allows the production of mutants through the use of specific oligo-nucleotide sequences which encode the DNA sequence of the desired mutation, as well as a sufficient number of adjacent nucleotides, to provide a primer sequence of sufficient size and sequence complexity to form a stable duplex on both sides of the deletion junction being traversed. Typically, a primer of about 17 to 25 nucleotides in length is preferred, with about 5 to 10 residues on both sides of the junction of the sequence being altered.

[0070] The technique typically employs a bacteriophage vector that exists in both a single stranded and double stranded form. Typical vectors useful in site-directed mutagenesis include vectors such as the M13 phage. These phage vectors are commercially available and their use is generally well known to those skilled in the art. Double stranded plasmids are also routinely employed in site directed mutagenesis, which eliminates the step of transferring the gene of interest from a phage to a plasmid.

[0071] In general, site-directed mutagenesis is performed by first obtaining a single-stranded vector, or melting of two strands of a double stranded vector which includes within its sequence a DNA sequence encoding the desired protein. An oligonucleotide primer bearing the desired mutated sequence is synthetically prepared. This primer is then annealed with the single-stranded DNA preparation, taking into account the degree of mismatch when selecting hybridization conditions, and subjected to DNA polymerizing enzymes such as *E. coli* polymerase I Klenow fragment, in order to complete the synthesis of the mutation-bearing strand. Thus, a heteroduplex is formed wherein one strand encodes the original non-mutated sequence and the second strand bears the desired mutation. This heteroduplex vector is then used to transform appropriate cells, such as *E. coli* cells, and clones are selected that include recombinant vectors bearing the mutated sequence arrangement.

[0072] The preparation of sequence variants of the selected gene using site-directed mutagenesis is provided as a means of producing potentially useful species and is not meant to be limiting, as there are other ways in which sequence variants of genes may be obtained. For example,

recombinant vectors encoding the desired gene may be treated with mutagenic agents, such as hydroxylamine, to obtain sequence variants.

C. POLYPEPTIDES AND ANTIGENS

[0073] For the purposes of the present invention a *Chlamydia* polypeptide used as an antigen may be a naturally-occurring *Chlamydia* polypeptide that has been extracted using protein extraction techniques well known to those of skill in the art. In particular embodiments, a *Chlamydia* antigen is identified by ELI and prepared in a pharmaceutically acceptable carrier for the vaccination of an animal against *Chlamydia* infection.

[0074] In alternative embodiments, the *Chlamydia* polypeptide or antigen may be a synthetic peptide. In still other embodiments, the peptide may be a recombinant peptide produced through molecular engineering techniques. The present section describes the methods and compositions involved in producing a composition of *Chlamydia* polypeptides for use as antigens in the present invention.

[0075] 1. *Chlamydia* Polypeptides as Antigens

[0076] Section A describes methods for preparing a cloned *Chlamydia* library via ELI. Described also are methods for screening and identifying *Chlamydia* genes that confer protection against *Chlamydia* infection. Thus, *Chlamydia* polypeptide encoding genes or their corresponding cDNA identified in the present invention can be inserted into an appropriate cloning vehicle for the production of *Chlamydia* polypeptides as antigens for the present invention. In addition, sequence variants of the polypeptide can be prepared. These may, for instance, be minor sequence variants of the polypeptide that arise due to natural variation within the population or they may be homologues found in other species. They also may be sequences that do not occur naturally, but that are sufficiently similar that they function similarly and/or elicit an immune response that cross-reacts with natural forms of the polypeptide. Sequence variants can be prepared by standard methods of site-directed mutagenesis such as those described below in the following section.

[0077] Another synthetic or recombinant variation of a *Chlamydia*-antigen is a polyepitopic moiety comprising repeats of epitopic determinants found naturally on *Chlamydia* proteins. Such synthetic polyepitopic proteins can be made up of several homomeric repeats of any one *Chlamydia* protein epitope; or can comprise of two or more heteromeric epitopes expressed on one or several *Chlamydia* protein epitopes.

[0078] Amino acid sequence variants of the polypeptide can be substitutional, insertional or deletion variants. Deletion variants lack one or more residues of the native protein which are not essential for function or immunogenic activity, and are exemplified by the variants lacking a transmembrane sequence described above. Another common type of deletion variant is one lacking secretory signal sequences or signal sequences directing a protein to bind to a particular part of a cell.

[0079] Substitutional variants typically contain the exchange of one amino acid for another at one or more sites within the protein, and may be designed to modulate one or more properties of the polypeptide such as stability against

proteolytic cleavage. Substitutions preferably are conservative, that is, one amino acid is replaced with one of similar shape and charge. Conservative substitutions are well known in the art and include, for example, the changes of alanine to serine; arginine to lysine; asparagine to glutamine or histidine; aspartate to glutamate; cysteine to serine; glutamine to asparagine; glutamate to aspartate; glycine to proline; histidine to asparagine or glutamine; isoleucine to leucine or valine; leucine to valine or isoleucine; lysine to arginine; methionine to leucine or isoleucine; phenylalanine to tyrosine, leucine or methionine; serine to threonine; threonine to serine; tryptophan to tyrosine; tyrosine to tryptophan or phenylalanine; and valine to isoleucine or leucine.

[0080] Insertional variants include fusion proteins such as those used to allow rapid purification of the polypeptide and also can include hybrid proteins containing sequences from other proteins and polypeptides which are homologues of the polypeptide. For example, an insertional variant could include portions of the amino acid sequence of the polypeptide from one species, together with portions of the homologous polypeptide from another species. Other insertional variants can include those in which additional amino acids are introduced within the coding sequence of the polypeptide. These typically are smaller insertions than the fusion proteins described above and are introduced, for example, into a protease cleavage site.

[0081] In one embodiment, major antigenic determinants of the polypeptide may be identified by an empirical approach in which portions of the gene encoding the polypeptide are expressed in a recombinant host, and the resulting proteins tested for their ability to elicit an immune response. For example, the polymerase chain reaction (PCR) can be used to prepare a range of cDNAs encoding peptides lacking successively longer fragments of the C-terminus of the protein. The immunogenic activity of each of these peptides then identifies those fragments or domains of the polypeptide that are essential for this activity. Further experiments in which only a small number of amino acids are removed or added at each iteration then allows the location of other antigenic determinants of the polypeptide. Thus, the polymerase chain reaction, a technique for amplifying a specific segment of DNA via multiple cycles of denaturation-renaturation, using a thermostable DNA polymerase, deoxyribonucleotides and primer sequences is contemplated in the present invention. (Mullis, 1990; Mullis et al., 1992).

[0082] Another embodiment for the preparation of the polypeptides according to the invention is the use of peptide mimetics. Mimetics are peptide-containing molecules that mimic elements of protein secondary structure. Because many proteins exert their biological activity via relatively small regions of their folded surfaces, their actions can be reproduced by much smaller designer (mimetic) molecules that retain the bioactive surfaces and have potentially improved pharmacokinetic/dynamic properties (Fairlie et al., 1998).

[0083] The underlying rationale behind the use of peptide mimetics is that the peptide backbone of proteins exists chiefly to orient amino acid side chains in such a way as to facilitate molecular interactions, such as those of antibody and antigen. However, unlike proteins, peptides often lack

well defined three dimensional structure in aqueous solution and tend to be conformationally mobile. Progress has been made with the use of molecular constraints to stabilize the bioactive conformations. By affixing or incorporating templates that fix secondary and tertiary structures of small peptides, synthetic molecules (protein surface mimetics) can be devised to mimic the localized elements of protein structure that constitute bioactive surfaces. Methods for mimicking individual elements of secondary structure (helices, turns, strands, sheets) and for assembling their combinations into tertiary structures (helix bundles, multiple loops, helix-loop-helix motifs) have been reviewed (Fairlie et al., 1998; Moore, 1994).

[0084] Methods for predicting, preparing, modifying, and screening mimetic peptides are described in U.S. Pat. No. 5,933,819 and U.S. Pat. No. 5,869,451 (each specifically incorporated herein by reference). It is contemplated in the present invention, that peptide mimetics will be useful in screening modulators of an immune response.

[0085] Modifications and changes may be made in the structure of a gene and still obtain a functional molecule that encodes a protein or polypeptide with desirable characteristics. The following is a discussion based upon changing the amino acids of a protein to create an equivalent, or even an improved, second-generation molecule. The amino acid changes may be achieved by changing the codons of the DNA sequence, according to the following data.

[0086] For example, certain amino acids may be substituted for other amino acids in a protein structure without appreciable loss of interactive binding capacity with structures such as, for example, antigen-binding regions of antibodies or binding sites on substrate molecules. Since it is the interactive capacity and nature of a protein that defines that protein's biological functional activity, certain amino acid substitutions can be made in a protein sequence, and its underlying DNA coding sequence, and nevertheless obtain a protein with like properties. It is thus contemplated by the inventor that various changes may be made in the DNA sequences of genes without appreciable loss of their biological utility or activity. Table 1 shows the codons that encode particular amino acids.

[0087] In making such changes, the hydropathic index of amino acids may be considered. The importance of the hydropathic amino acid index in conferring interactive biological function on a protein is generally understood in the art by Doolittle, 1982).

[0088] It is accepted that the relative hydropathic character of the amino acid contributes to the secondary structure of the resultant protein, which in turn defines the interaction of the protein with other molecules, for example, enzymes, substrates, receptors, DNA, antibodies, antigens, and the like.

[0089] Each amino acid has been assigned a hydropathic index on the basis of their hydrophobicity and charge characteristics (Kyte and Doolittle, 1982), these are: Isoleucine (+4.5); valine (+4.2); leucine (+3.8); phenylalanine (+2.8); cysteine/cystine (+2.5); methionine (+1.9); alanine (+1.8); glycine (-0.4); threonine (-0.7); serine (-0.8); tryptophan (-0.9); tyrosine (-1.3); proline (-1.6); histidine (-3.2); glutamate (-3.5); glutamine (-3.5); aspartate (-3.5); asparagine (-3.5); lysine (-3.9); and arginine (-4.5).

[0090] It is known in the art that certain amino acids may be substituted by other amino acids having a similar hydrophobic index or score and still result in a protein with similar biological activity, i.e., still obtain a biological functionally equivalent protein. In making such changes, the substitution of amino acids whose hydrophobic indices are within ± 2 is preferred, those which are within ± 1 are particularly preferred, and those within ± 0.5 are even more particularly preferred.

[0091] It also is understood in the art that the substitution of like amino acids can be made effectively on the basis of hydrophilicity. U.S. Pat. No. 4,554,101, incorporated herein by reference, states that the greatest local average hydrophilicity of a protein, as governed by the hydrophilicity of its adjacent amino acids, correlates with a biological property of the protein.

[0092] As detailed in U.S. Pat. No. 4,554,101, the following hydrophilicity values have been assigned to amino acid residues: arginine (+3.0); lysine (+3.0); aspartate (+3.0 $\pm$ 1); glutamate (+3.0 $\pm$ 1); serine (+0.3); asparagine (+0.2); glutamine (+0.2); glycine (0); threonine (-0.4); proline (-0.5 $\pm$ 1); alanine (-0.5); histidine. *-0.5); cysteine (-1.0); methionine (-1.3); valine (-1.5); leucine (-1.8); isoleucine (-1.8); tyrosine (-2.3); phenylalanine (-2.5); tryptophan (-3.4).

[0093] It is understood that an amino acid can be substituted for another having a similar hydrophilicity value and still obtain a biologically equivalent and immunologically equivalent protein. In such changes, the substitution of amino acids whose hydrophilicity values are within ± 2 is preferred, those that are within ± 1 are particularly preferred, and those within ± 0.5 are even more particularly preferred.

[0094] As outlined above, amino acid substitutions generally are based on the relative similarity of the amino acid side-chain substituents, for example, their hydrophobicity, hydrophilicity, charge, size, and the like. Exemplary substitutions that take various of the foregoing characteristics into consideration are well known to those of skill in the art and include: arginine and lysine; glutamate and aspartate; serine and threonine; glutamine and asparagine; and valine, leucine and isoleucine.

[0095] 2. Synthetic Polypeptides

[0096] Contemplated in the present invention are *Chlamydia* Psittaci proteins and related peptides for use as antigens. In certain embodiments, the synthesis of a *Chlamydia* peptide fragment is considered. The peptides of the invention can be synthesized in solution or on a solid support in accordance with conventional techniques. Various automatic synthesizers are commercially available and can be used in accordance with known protocols. See, for example, Stewart and Young, (1984); Tam et al., (1983); Merrifield, (1986); and Barany and Merrifield (1979), each incorporated herein by reference. Alternatively, recombinant DNA technology may be employed wherein a nucleotide sequence which encodes a peptide of the invention is inserted into an expression vector, transformed or transfected into an appropriate host cell and cultivated under conditions suitable for expression.

[0097] 3. *Chlamydia* Polypeptide/Antigen Purification

[0098] *Chlamydia* polypeptides, including *Chlamydia psittaci* polypeptides, of the present invention are used as

antigens for inducing a protective immune response in an animal and for the preparation of anti-*Chlamydia* antibodies. Thus, certain aspects of the present invention concern the purification, and in particular embodiments, the substantial purification, of a *Chlamydia* polypeptide that is described herein above. The term "purified protein or peptide" as used herein, is intended to refer to a composition, isolatable from other components, wherein the protein or peptide is purified to any degree relative to its naturally-obtainable state. A purified protein or peptide therefore also refers to a protein or peptide, free from the environment in which it may naturally occur.

[0099] Generally, "purified" will refer to a protein or peptide composition that has been subjected to fractionation to remove various other components, and which composition substantially retains its expressed biological activity. Where the term "substantially purified" is used, this designation will refer to a composition in which the protein or peptide forms the major component of the composition, such as constituting about 50% or more of the proteins in the composition.

[0100] Various methods for quantifying the degree of purification of the protein or peptide will be known to those of skill in the art in light of the present disclosure. These include, for example, determining the specific activity of an active fraction, or assessing the number of polypeptides within a fraction by SDS/PAGE analysis. A preferred method for assessing the purity of a fraction is to calculate the specific activity of the fraction, to compare it to the specific activity of the initial extract, and to thus calculate the degree of purity, herein assessed by a "-fold purification number." The actual units used to represent the amount of activity will, of course, be dependent upon the particular assay technique chosen to follow the purification and whether or not the expressed protein or peptide exhibits a detectable activity.

[0101] Various techniques suitable for use in protein purification will be well known to those of skill in the art. These include, for example, precipitation with ammonium sulphate, PEG, antibodies and the like or by heat denaturation, followed by centrifugation; chromatography steps such as ion exchange, gel filtration, reverse phase, hydroxylapatite and affinity chromatography; isoelectric focusing; gel electrophoresis; and combinations of such and other techniques. As is generally known in the art, it is believed that the order of conducting the various purification steps may be changed, or that certain steps may be omitted, and still result in a suitable method for the preparation of a substantially purified protein or peptide.

[0102] There is no general requirement that the protein or peptide always be provided in their most purified state. Indeed, it is contemplated that less substantially purified products will have utility in certain embodiments. Partial purification may be accomplished by using fewer purification steps in combination, or by utilizing different forms of the same general purification scheme. For example, it is appreciated that a cation-exchange column chromatography performed utilizing an HPLC apparatus will generally result in a greater -fold purification than the same technique utilizing a low pressure chromatography system. Methods exhibiting a lower degree of relative purification may have advantages in total recovery of protein product, or in maintaining the activity of an expressed protein.

[0103] It is known that the migration of a polypeptide can vary, sometimes significantly, with different conditions of SDS/PAGE (Capaldi et al., 1977). It will therefore be appreciated that under differing electrophoresis conditions, the apparent molecular weights of purified or partially purified expression products may vary.

[0104] High Performance Liquid Chromatography (HPLC) is characterized by a very rapid separation with extraordinary resolution of peaks. This is achieved by the use of very fine particles and high pressure to maintain an adequate flow rate. Separation can be accomplished in a matter of minutes, or at most an hour. Moreover, only a very small volume of the sample is needed because the particles are so small and close-packed that the void volume is a very small fraction of the bed volume. Also, the concentration of the sample need not be very great because the bands are so narrow that there is very little dilution of the sample.

[0105] Gel chromatography, or molecular sieve chromatography, is a special type of partition chromatography that is based on molecular size. The theory behind gel chromatography is that the column, which is prepared with tiny particles of an inert substance that contain small pores, separates larger molecules from smaller molecules as they pass through or around the pores, depending on their size. As long as the material of which the particles are made does not adsorb the molecules, the sole factor determining rate of flow is the size. Hence, molecules are eluted from the column in decreasing size, so long as the shape is relatively constant. Gel chromatography is unsurpassed for separating molecules of different size because separation is independent of all other factors such as pH, ionic strength, temperature, etc. There also is virtually no adsorption, less zone spreading and the elution volume is related in a simple manner to molecular weight.

[0106] Affinity Chromatography is a chromatographic procedure that relies on the specific affinity between a substance to be isolated and a molecule that it can specifically bind to. This is a receptor-ligand type interaction. The column material is synthesized by covalently coupling one of the binding partners to an insoluble matrix. The column material is then able to specifically adsorb the substance from the solution. Elution occurs by changing the conditions to those in which binding will not occur (alter pH, ionic strength, temperature, etc.).

D. GENE DELIVERY

[0107] In certain embodiments of the invention, an expression construct comprising a *Chlamydia* gene or other polynucleotide segment under the control of a heterologous promoter operable in eukaryotic cells is provided. For example, the delivery of *Chlamydia psittaci*, antigen-encoding expression constructs can be provided in this manner. The general approach in certain aspects of the present invention is to provide a cell with an expression construct encoding a specific protein, polypeptide or peptide fragment, thereby permitting the antigenic expression of the protein, polypeptide or peptide fragment to take effect in the cell. Following delivery of the expression construct, the protein, polypeptide or peptide fragment encoded by the expression construct is synthesized by the transcriptional and translational machinery of the cell, as well as any that may be provided by the expression construct.

[0108] Viral and non-viral vector systems are the two predominant categories for the delivery of an expression construct encoding a therapeutic protein, polypeptide, polypeptide fragment. Both vector systems are described in the following sections. There also are two primary approaches utilized in the delivery of an expression construct for the purposes of gene therapy; either indirect, ex vivo methods or direct, in vivo methods. Ex vivo gene transfer comprises vector modification of (host) cells in culture and the administration or transplantation of the vector modified cells to a gene therapy recipient. In vivo gene transfer comprises direct introduction of the vector (e.g., injection, inhalation) into the target source or therapeutic gene recipient.

[0109] In certain embodiments of the invention, the nucleic acid encoding the gene or polynucleotide may be stably integrated into the genome of the cell. In yet further embodiments, the nucleic acid may be stably or transiently maintained in the cell as a separate, episomal segment of DNA. Such nucleic acid segments or "episomes" encode sequences sufficient to permit maintenance and replication independent of or in synchronization with the host cell cycle. How the expression construct is delivered to a cell and/or where in the cell the nucleic acid remains is dependent on the type of vector employed. The following gene delivery methods provide the framework for choosing and developing the most appropriate gene delivery system for a preferred application.

[0110] 1. Non-Viral Polynucleotide Delivery

[0111] In one embodiment of the invention, a polynucleotide expression construct consists of naked recombinant DNA or plasmids. In preferred embodiments of the invention, an expression construct comprising, for example, a *Chlamydia psittaci* polynucleotide is administered to a subject via injection and/or particle bombardment (e.g., a gene gun). Thus, in one preferred embodiment, polynucleotide expression constructs are transferred into cells by accelerating DNA-coated microprojectiles to a high velocity, allowing the DNA-coated microprojectiles to pierce cell membranes and enter cells. In another preferred embodiment, polynucleotides are administered to a subject by injection. Injection of a polynucleotide expression construct may be given by intramuscular, intravenous, subcutaneous, or intraperitoneal injection, as long as the polynucleotide expression construct can effectively be delivered to a desired target.

[0112] a. Particle Bombardment

[0113] Particle Bombardment depends on the ability to accelerate DNA-coated microprojectiles to a high velocity allowing them to pierce cell membranes and enter cells without killing them (Klein et al., 1987). Several devices for accelerating small particles have been developed. The most commonly used forms rely on high-pressure helium gas (Sanford et al., 1991), of which one of the present inventors is a co-inventor. The microprojectiles used have consisted of biologically inert substances such as tungsten or gold beads.

[0114] For microprojectile bombardment transformation using the constructs of the instant invention, both physical and biological parameters may be optimized. Physical factors are those that involve manipulating the DNA/microprojectile precipitate or those that affect the flight and

velocity of either the macro- or microprojectiles. Biological factors include all steps involved in manipulation of cells before and immediately after bombardment, such as the osmotic adjustment of target cells to help alleviate the trauma associated with bombardment, the orientation of an immature embryo or other target tissue relative to the particle trajectory, and also the nature of the transforming DNA, such as linearized DNA or intact supercoiled plasmids.

[0115] Accordingly, it is contemplated that one may wish to adjust various bombardment parameters in small scale studies to fully optimize the conditions. One may particularly wish to adjust physical parameters such as DNA concentration, gap distance, flight distance, tissue distance, and helium pressure. It is further contemplated that the grade of helium may effect transformation efficiency. One may also optimize the trauma reduction factors (TRFs) by modifying conditions which influence the physiological state of the recipient cells and which may therefore influence transformation and integration efficiencies. For example, the osmotic state, tissue hydration and the subculture stage or cell cycle of the recipient cells may be adjusted for optimum transformation.

[0116] Other physical factors include those that involve manipulating the DNA/microprojectile precipitate or those that affect the flight and velocity of either the macro- or microprojectiles. Biological factors include all steps involved in manipulation of cells immediately before and after bombardment. The pre-bombardment culturing conditions, such as osmotic environment, the bombardment parameters, and the plasmid configuration have been adjusted to yield the maximum numbers of stable transformants.

[0117] For microprojectile bombardment, one will attach (i.e., "coat") DNA to the microprojectiles such that it is delivered to recipient cells in a form suitable for transformation thereof. In this respect, at least some of the transforming DNA must be available to the target cell for transformation to occur, while at the same time during delivery the DNA must be attached to the microprojectile. Therefore, availability of the transforming DNA from the microprojectile may comprise the physical reversal of interactions between transforming DNA and the microprojectile following delivery of the microprojectile to the target cell. This need not be the case, however, as availability to a target cell may occur as a result of breakage of unbound segments of DNA or of other molecules which comprise the physical attachment to the microprojectile. Availability may further occur as a result of breakage of bonds between the transforming DNA and other molecules, which are either directly or indirectly attached to the microprojectile. It is further contemplated that transformation of a target cell may occur by way of direct illegitimate or homology-dependent recombination between the transforming DNA and the genomic DNA of the recipient cell. Therefore, as used herein, a "coated" microprojectile will be one which is capable of being used to transform a target cell, in that the transforming DNA will be delivered to the target cell, yet will be accessible to the target cell such that transformation may occur.

[0118] Any technique for coating microprojectiles which allows for delivery of transforming DNA to the target cells may be used. Methods for coating microprojectiles which

have been demonstrated to work well with the current invention have been specifically disclosed herein. DNA may be bound to microprojectile particles using alternative techniques, however. For example, particles may be coated with streptavidin and DNA end labeled with long chain thiol cleavable biotinylated nucleotide chains. The DNA adheres to the particles due to the streptavidin-biotin interaction, but is released in the cell by reduction of the thiol linkage through reducing agents present in the cell.

[0119] Alternatively, particles may be prepared by functionalizing the surface of a gold oxide particle, providing free amine groups. DNA, having a strong negative charge, binds to the functionalized particles. Furthermore, charged particles may be deposited in controlled arrays on the surface of mylar flyer disks used in the PDS-1000 Biolistics device, thereby facilitating controlled distribution of particles delivered to target tissue.

[0120] b. Other Non-Viral Methods of Polynucleotide Delivery

[0121] Transfer of a cloned expression construct in the present invention also may be performed by any of the methods which physically or chemically permeabilize the cell membrane (e.g., calcium phosphate precipitation, DEAE-dextran, electroporation, direct microinjection, DNA-loaded liposomes and lipofectamine-DNA complexes, cell sonication, gene bombardment using high velocity microprojectiles and receptor-mediated transfection).

[0122] In certain embodiments, the use of lipid formulations and/or nanocapsules is contemplated for the introduction of a *Chlamydia psittaci* polynucleotide or polypeptide, or a gene therapy vector into host cells.

[0123] Nanocapsules can generally entrap compounds in a stable and/or reproducible way. To avoid side effects due to intracellular polymeric overloading, such ultrafine particles (sized around 0.1 μm) should be designed using polymers able to be degraded in vivo. Biodegradable polyalkyl-cyanoacrylate nanoparticles that meet these requirements are contemplated for use in the present invention, and/or such particles may be easily made.

[0124] In a preferred embodiment of the invention, the polynucleotide or polypeptide may be associated with a lipid. The polynucleotide or polypeptide associated with a lipid may be encapsulated in the aqueous interior of a liposome, interspersed within the lipid bilayer of a liposome, attached to a liposome via a linking molecule that is associated with both the liposome and the oligonucleotide, entrapped in a liposome, complexed with a liposome, dispersed in a solution containing a lipid, mixed with a lipid, combined with a lipid, contained as a suspension in a lipid, contained or complexed with a micelle, or otherwise associated with a lipid. The lipid or lipid/polynucleotide or polypeptide associated compositions of the present invention are not limited to any particular structure in solution. For example, they may be present in a bilayer structure, as micelles, or with a "collapsed" structure. They may also simply be interspersed in a solution, possibly forming aggregates which are not uniform in either size or shape.

[0125] Lipids suitable for use according to the present invention can be obtained from commercial sources. For example, dimyristyl phosphatidylcholine ("DMPC") can be obtained from Sigma Chemical Co., dicetyl phosphate

("DCP") is obtained from K & K Laboratories (Plainview, N.Y.); cholesterol ("Chol") is obtained from Calbiochem-Behring; dimyristyl phosphatidylglycerol ("DMPG") and other lipids may be obtained from Avanti Polar Lipids, Inc. (Birmingham, Ala.). Stock solutions of lipids in chloroform or chloroform/methanol can be stored at about -20°C . Preferably, chloroform is used as the only solvent since it is more readily evaporated than methanol.

[0126] "Liposome" is a generic term encompassing a variety of single and multilamellar lipid vehicles formed by the generation of enclosed lipid bilayers or aggregates. Liposomes may be characterized as having vesicular structures with a phospholipid bilayer membrane and an inner aqueous medium. Multilamellar liposomes have multiple lipid layers separated by aqueous medium. They form spontaneously when phospholipids are suspended in an excess of aqueous solution. The lipid components undergo self-rearrangement before the formation of closed structures and entrap water and dissolved solutes between the lipid bilayers (Ghosh and Bachhawat, 1991). However, the present invention also encompasses compositions that have different structures in solution than the normal vesicular structure. For example, the lipids may assume a micellar structure or merely exist as non-uniform aggregates of lipid molecules. Also contemplated are lipofectamine-nucleic acid complexes.

[0127] Liposomes within the scope of the present invention can be prepared in accordance with known laboratory procedures, for example: the method of Bangham et al. (1965), the contents of which are incorporated herein by reference; the method of Gregoriadis, as described in *DRUG CARRIERS IN BIOLOGY AND MEDICINE*, G. Gregoriadis ed. (1979) pp. 287-341, the contents of which are incorporated herein by reference; the method of Deamer and Uster (1983), the contents of which are incorporated by reference; and the reverse-phase evaporation method as described by Szoka and Papahadjopoulos (1978).

[0128] Other vector delivery systems which can be employed to deliver a nucleic acid encoding a therapeutic gene into cells are receptor-mediated delivery vehicles. These take advantage of the selective uptake of macromolecules by receptor-mediated endocytosis in almost all eukaryotic cells. Because of the cell type-specific distribution of various receptors, the delivery can be highly specific (Wu and Wu, 1993).

[0129] Receptor-mediated gene targeting vehicles generally consist of two components: a cell receptor-specific ligand and a DNA-binding agent. Several ligands have been used for receptor-mediated gene transfer. The most extensively characterized ligands are asialoorosomucoid (ASOR) (Wu and Wu, 1987) and transferring (Wagner et al., 1990). Recently, a synthetic neoglycoprotein, which recognizes the same receptor as ASOR, has been used as a gene delivery vehicle (Ferkol et al, 1993; Perales et al, 1994) and epidermal growth factor (EGF) has also been used to deliver genes to squamous carcinoma cells (Myers, EPO 0273085).

[0130] In other embodiments, the delivery vehicle may comprise a ligand and a liposome. For example, Nicolau et al (1987) employed lactosyl-ceramide, a galactose-terminal asialganglioside, incorporated into liposomes and observed an increase in the uptake of the insulin gene by hepatocytes. Thus, it is feasible that a nucleic acid encoding a therapeutic

gene also may be specifically delivered into a cell type such as prostate, epithelial or tumor endothelial cells, by any number of receptor-ligand systems with or without liposomes. For example, the human prostate-specific antigen (Watt et al., 1986) may be used as the receptor for mediated delivery of a nucleic acid in prostate tissue.

[0131] In another embodiment of the invention, the expression construct may simply consist of naked recombinant DNA or plasmids. Transfer of the construct may be performed by any of the methods mentioned above which physically or chemically permeabilize the cell membrane. This is applicable particularly for transfer in vitro, however, it may be applied for in vivo use as well. Dubensky et al. (1984) successfully injected polyomavirus DNA in the form of CaPO_4 precipitates into liver and spleen of adult and newborn mice demonstrating active viral replication and acute infection. Benvenisty and Neshif (1986) also demonstrated that direct intraperitoneal injection of CaPO_4 precipitated plasmids results in expression of the transfected genes. It is envisioned that DNA encoding a *Chlamydia psittaci* gene or polynucleotide of interest may also be transferred in a similar manner in vivo and express the gene or polynucleotide product.

[0132] 2. Viral Vectors

[0133] In certain embodiments, it is contemplated that a *Chlamydia psittaci* gene or other polynucleotide that confers immune resistance to infection pursuant to the invention may be delivered by a viral vector. The capacity of certain viral vectors to efficiently infect or enter cells, to integrate into a host cell genome and stably express viral genes, have led to the development and application of a number of different viral vector systems (Robbins et al., 1998). Viral systems are currently-being developed for use as vectors for ex vivo and in vivo gene transfer. For example, adenovirus, herpes-simple virus, retrovirus and adeno-associated virus vectors are being evaluated currently for treatment of diseases such as cancer, cystic fibrosis, Gaucher disease, renal disease and arthritis (Robbins and Ghivizzani, 1998; Imai et al., 1998; U.S. Pat. No. 5,670,488). The various viral vectors described below, present specific advantages and disadvantages, depending on the particular gene-therapeutic application.

[0134] a. Adenoviral Vectors

[0135] In particular embodiments, an adenoviral expression vector is contemplated for the delivery of expression constructs. "Adenovirus expression vector" is meant to include those constructs containing adenovirus sequences sufficient to (a) support packaging of the construct and (b) to ultimately express a tissue or cell-specific construct that has been cloned therein.

[0136] Adenoviruses comprise linear double stranded DNA, with a genome ranging from 30 to 35 kb in size (Reddy et al., 1998; Morrison et al., 1997; Chillon et al., 1999). An adenovirus expression vector according to the present invention comprises a genetically engineered form of the adenovirus. Advantages of adenoviral gene transfer include the ability to infect a wide variety of cell types, including non-dividing cells, a mid-sized genome, ease of manipulation, high infectivity and they can be grown to high titers (Wilson, 1996). Further, adenoviral infection of host cells does not result in chromosomal integration because

adenoviral DNA can replicate in an episomal manner, without potential genotoxicity associated with other viral vectors. Adenoviruses also are structurally stable (Marienfeld et al., 1999) and no genome rearrangement has been detected after extensive amplification (Parks et al., 1997; Bett et al., 1993).

[0137] Salient features of the adenovirus genome are an early region (E1, E2, E3 and E4 genes), an intermediate region (pIX gene, Iva2 gene), a late region (L1, L2, L3, L4 and L5 genes), a major late promoter (MLP), inverted-terminal-repeats (ITRs) and a ψ sequence (Zheng, et al., 1999; Robbins et al., 1998; Graham and Prevec, 1995). The early genes E1, E2, E3 and E4 are expressed from the virus after infection and encode polypeptides that regulate viral gene expression, cellular gene expression, viral replication, and inhibition of cellular apoptosis. Further on during viral infection, the MLP is activated, resulting in the expression of the late (L) genes, encoding polypeptides required for adenovirus encapsidation. The intermediate region encodes components of the adenoviral capsid. Adenoviral inverted terminal repeats (ITRs; 100-200 bp in length), are cis elements, function as origins of replication and are necessary for viral DNA replication. The ψ sequence is required for the packaging of the adenoviral genome.

[0138] A common approach for generating an adenovirus for use as a gene transfer vector is the deletion of the E1 gene (E1⁻), which is involved in the induction of the E2, E3 and E4 promoters (Graham and Prevec, 1995). Subsequently, a therapeutic gene or genes can be inserted recombinantly in place of the E1 gene, wherein expression of the therapeutic gene(s) is driven by the E1 promoter or a heterologous promoter. The E1⁻, replication-deficient virus is then proliferated in a "helper" cell line that provides the E1 polypeptides in trans (e.g., the human embryonic kidney cell line 293). Thus, in the present invention it may be convenient to introduce the transforming construct at the position from which the E1-coding sequences have been removed. However, the position of insertion of the construct within the adenovirus sequences is not critical to the invention. Alternatively, the E3 region, portions of the E4 region or both may be deleted, wherein a heterologous nucleic acid sequence under the control of a promoter operable in eukaryotic cells is inserted into the adenovirus genome for use in gene transfer (U.S. Pat. No. 5,670,488; U.S. Pat. No. 5,932,210, each specifically incorporated herein by reference).

[0139] Although adenovirus based vectors offer several unique advantages over other vector systems, they often are limited by vector immunogenicity, size constraints for insertion of recombinant genes and low levels of replication. The preparation of a recombinant adenovirus vector deleted of all open reading frames, comprising a full length dystrophin gene and the terminal repeats required for replication (Haecker et al., 1997) offers some potentially promising advantages to the above mentioned adenoviral shortcomings. The vector was grown to high titer with a helper virus in 293 cells and was capable of efficiently transducing dystrophin in mdx mice, in myotubes in vitro and muscle fibers in vivo. Helper-dependent viral vectors are discussed below.

[0140] A major concern in using adenoviral vectors is the generation of a replication-competent virus during vector

production in a packaging cell line or during gene therapy treatment of an individual. The generation of a replication-competent virus could pose serious threat of an unintended viral infection and pathological consequences for the patient. Armentano et al., describe the preparation of a replication-defective adenovirus vector, claimed to eliminate the potential for the inadvertent generation of a replication-competent adenovirus (U.S. Pat. No. 5,824,544, specifically incorporated herein by reference). The replication-defective adenovirus method comprises a deleted E1 region and a relocated protein IX gene, wherein the vector expresses a heterologous, mammalian gene.

[0141] Other than the requirement that the adenovirus vector be replication defective, or at least conditionally defective, the nature of the adenovirus vector is not believed to be crucial to the successful practice of the invention. The adenovirus may be of any of the 42 different known serotypes and/or subgroups A-F. Adenovirus type 5 of subgroup C is the preferred starting material in order to obtain the conditional replication-defective adenovirus vector for use in the present invention. This is because adenovirus type 5 is a human adenovirus about which a great deal of biochemical and genetic information is known, and it has historically been used for most constructions employing adenovirus as a vector.

[0142] As stated above, the typical vector according to the present invention is replication defective and will not have an adenovirus E1 region. Adenovirus growth and manipulation is known to those of skill in the art, and exhibits broad host range in vitro and in vivo (U.S. Pat. No. 5,670,488; U.S. Pat. No. 5,932,210; U.S. Pat. No. 5,824,544). This group of viruses can be obtained in high titers, e.g., 10^9 to 10^{11} plaque-forming units per ml, and they are highly infective. The life cycle of adenovirus does not require integration into the host cell genome. The foreign genes delivered by adenovirus vectors are episomal and, therefore, have low genotoxicity to host cells. Many experiments, innovations, pre-clinical studies and clinical trials are currently under investigation for the use of adenoviruses as gene delivery vectors. For example, adenoviral gene delivery-based gene therapies are being developed for liver diseases (Han et al., 1999), psychiatric diseases (Lesch, 1999), neurological diseases (Smith, 1998; Hermens and Verhaagen, 1998), coronary diseases (Feldman et al., 1996), muscular diseases (Petrof, 1998), gastrointestinal diseases (Wu, 1998) and various cancers such as colorectal (Fujiwara and Tanaka, 1998; Dorai et al., 1999), pancreatic (Carrion et al., 1999), bladder (Irie et al., 1999), head and neck (Blackwell et al., 1999), breast (Stewart et al., 1999), lung (Batra et al., 1999) and ovarian (Vanderkwaak et al., 1999).

[0143] b. Retroviral Vectors

[0144] In certain embodiments of the invention, the use of retroviruses for gene delivery are contemplated. Retroviruses are RNA viruses comprising an RNA genome. When a host cell is infected by a retrovirus, the genomic RNA is reverse transcribed into a DNA intermediate which is integrated into the chromosomal DNA of infected cells. This integrated DNA intermediate is referred to as a provirus. A particular advantage of retroviruses is that they can stably infect dividing cells with a gene of interest (e.g., a therapeutic gene) by integrating into the host DNA, without expressing immunogenic viral proteins. Theoretically, the

integrated retroviral vector will be maintained for the life of the infected host cell, expressing the gene of interest.

[0145] The retroviral genome and the proviral DNA have three genes: gag, pol, and env, which are flanked by two long terminal repeat (LTR) sequences. The gag gene encodes the internal structural (matrix, capsid, and nucleocapsid) proteins; the pol gene encodes the RNA-directed DNA polymerase (reverse transcriptase) and the env gene encodes viral envelope glycoproteins. The 5' and 3' LTRs serve to promote transcription and polyadenylation of the virion RNAs. The LTR contains all other cis-acting sequences necessary for viral replication.

[0146] A recombinant retrovirus of the present invention may be genetically modified in such a way that some of the structural, infectious genes of the native virus have been removed and replaced instead with a nucleic acid sequence to be delivered to a target cell (U.S. Pat. No. 5,858,744; U.S. Pat. No. 5,739,018, each incorporated herein by reference). After infection of a cell by the virus, the virus injects its nucleic acid into the cell and the retrovirus genetic material can integrate into the host cell genome. The transferred retrovirus genetic material is then transcribed and translated into proteins within the host cell. As with other viral vector systems, the generation of a replication-competent retrovirus during vector production or during therapy is a major concern. Retroviral vectors suitable for use in the present invention are generally defective retroviral vectors that are capable of infecting the target cell, reverse transcribing their RNA genomes, and integrating the reverse transcribed DNA into the target cell genome, but are incapable of replicating within the target cell to produce infectious retroviral particles (e.g. the retroviral genome transferred into the target cell is defective in gag, the gene encoding virion structural proteins, and/or in pol, the gene encoding reverse transcriptase). Thus, transcription of the provirus and assembly into infectious virus occurs in the presence of an appropriate helper virus or in a cell line containing appropriate sequences enabling encapsidation without coincident production of a contaminating helper virus.

[0147] The growth and maintenance of retroviruses is known in the art (U.S. Pat. No. 5,955,331; U.S. Pat. No. 5,888,502, each specifically incorporated herein by reference). Nolan et al. describe the production of stable high titre, helper-free retrovirus comprising a heterologous gene (U.S. Pat. No. 5,830,725, specifically incorporated herein by reference). Methods for constructing packaging cell lines useful for the generation of helper-free recombinant retroviruses with amphoteric or ecotrophic host ranges, as well as methods of using the recombinant retroviruses to introduce a gene of interest into eukaryotic cells in vivo and in vitro are contemplated in the present invention (U.S. Pat. No. 5,955,331).

[0148] Currently, the majority of all clinical trials for vector mediated gene delivery use murine leukemia virus (MLV)-based retroviral vector gene delivery (Robbins et al., 1998; Miller et al., 1993). Disadvantages of retroviral gene delivery includes a requirement for ongoing cell division for stable infection and a coding capacity that, prevents the delivery of large genes. However, recent development of vectors such as lentivirus (e.g., HIV), simian immunodeficiency virus (SIV) and equine infectious-anemia virus (EIAV), which can infect certain non-dividing cells, poten-

tially allow the in vivo use of retroviral vectors for gene therapy applications (Amado and Chen, 1999; Klimatcheva et al., 1999; White et al., 1999; Case et al., 1999). For example, HIV-based vectors have been used to infect non-dividing cells such as neurons (Takashi et al., 1999; Miyake et al., 1999), islets (Leibowitz et al., 1999) and muscle cells (Johnston et al., 1999). The therapeutic delivery of genes via retroviruses are currently being assessed for the treatment of various disorders such as inflammatory disease (Moldawer et al., 1999), AIDS (Amado et al., 1999; Engel and Kohn, 1999), cancer (Clay et al., 1999), cerebrovascular disease (Weihl et al., 1999) and hemophilia (Kay, 1998).

[0149] c. Herpes-Simplex Viral Vectors

[0150] Herpes simplex virus (HSV) type I and type II contain a double-stranded, linear DNA genome of approximately 150 kb, encoding 70-80 genes. Wild type HSV are able to infect cells lytically and to establish latency in certain cell types (e.g., neurons). Similar to adenovirus, HSV also can infect a variety of cell types including muscle (Yeung et al., 1999), ear (Derby et al., 1999), eye (Kaufman et al., 1999), tumors (Yoon et al., 1999; Howard et al., 1999), lung (Kohut et al., 1998), neuronal (Garrido et al., 1999; Lachmann and Efstathiou, 1999), liver (Miyake et al., 1999; Kooby et al., 1999) and pancreatic islets (Rabinovitch et al., 1999).

[0151] HSV viral genes are transcribed by cellular RNA polymerase II and are temporally regulated, resulting in the transcription and subsequent synthesis of gene products in roughly three discernable phases or kinetic classes. These phases of genes are referred to as the Immediate Early (IE) or alpha genes, Early (E) or beta genes and Late (L) or gamma genes. Immediately following the arrival of the genome of a virus in the nucleus of a newly infected cell, the IE genes are transcribed. The efficient expression of these genes does not require prior viral protein synthesis. The products of IE genes are required to activate transcription and regulate the remainder of the viral genome.

[0152] For use in therapeutic gene delivery, HSV must be rendered replication-defective. Protocols for generating replication-defective HSV helper virus-free cell lines have been described (U.S. Pat. No. 5,879,934; U.S. Pat. No. 5,851,826, each specifically incorporated herein by reference in its entirety). One IE protein, Infected Cell Polypeptide 4 (ICP4), also known as alpha 4 or Vmw175, is absolutely required for both virus infectivity and the transition from IE to later transcription. Thus, due to its complex, multifunctional nature and central role in the regulation of HSV gene expression, ICP4 has typically been the target of HSV genetic studies.

[0153] Phenotypic studies of HSV viruses deleted of ICP4 indicate that such viruses will be potentially useful for gene transfer purposes (Krisky et al., 1998a). One property of viruses deleted for ICP4 that makes them desirable for gene transfer is that they only express the five other IE genes: ICP0, ICP6, ICP27, ICP22 and ICP47 (DeLuca et al., 1985), without the expression of viral genes encoding proteins that direct viral DNA synthesis, as well as the structural proteins of the virus. This property is desirable for minimizing possible deleterious effects on host cell metabolism or an immune response following gene transfer. Further deletion of IE genes ICP22 and ICP27, in addition to ICP4, substantially improve reduction of HSV cytotoxicity and prevented early and late viral gene expression (Krisky et al., 1998b).

[0154] The therapeutic potential of HSV in gene transfer has been demonstrated in various in vitro model systems and in vivo for diseases such as Parkinson's (Yamada et al., 1999), retinoblastoma (Hayashi et al., 1999), intracerebral and intradermal tumors. (Moriuchi et al., 1998), B cell malignancies (Suzuki et al., 1998), ovarian cancer (Wang et al., 1998) and Duchenne muscular dystrophy (Huard et al., 1997).

[0155] d. Adeno-Associated Viral Vectors

[0156] Adeno-associated virus (AAV), a member of the parvovirus family, is a human virus that is increasingly being used for gene delivery therapeutics. AAV has several advantageous features not found in other viral systems. First, AAV can infect a wide range of host cells, including non-dividing cells. Second, AAV can infect cells from different species. Third, AAV has not been associated with any human or animal disease and does not appear to alter the biological properties of the host cell upon integration. For example, it is estimated that 80-85% of the human population has been exposed to AAV. Finally, AAV is stable at a wide range of physical and chemical conditions which lends itself to production, storage and transportation requirements.

[0157] The AAV genome is a linear, single-stranded DNA molecule containing 4681 nucleotides. The AAV genome generally comprises an internal non-repeating genome flanked on each end by inverted terminal repeats (ITRs) of approximately 145 bp in length. The ITRs have multiple functions, including origins of DNA replication, and as packaging signals for the viral genome. The internal non-repeated portion of the genome includes two large open reading frames, known as the AAV replication (rep) and capsid (cap) genes. The rep and cap genes code for viral proteins that allow the virus to replicate and package the viral genome into a virion. A family of at least four viral proteins are expressed from the AAV rep region, Rep 78, Rep 68, Rep 52, and Rep 40, named according to their apparent molecular weight. The AAV cap region encodes at least three proteins, VP1, VP2, and VP3.

[0158] AAV is a helper-dependent virus requiring co-infection with a helper virus (e.g., adenovirus, herpesvirus or vaccinia) in order to form AAV virions. In the absence of co-infection with a helper virus, AAV establishes a latent state in which the viral genome inserts into a host cell chromosome, but infectious virions are not produced. Subsequent infection by a helper virus "rescues" the integrated genome, allowing it to replicate and package its genome into infectious AAV virions. Although AAV can infect cells from different species, the helper virus must be of the same species as the host cell (e.g., human AAV will replicate in canine cells co-infected with a canine adenovirus).

[0159] AAV has been engineered to deliver genes of interest by deleting the internal non-repeating portion of the AAV genome and inserting a heterologous gene between the ITRs. The heterologous gene may be functionally linked to a heterologous promoter (constitutive, cell-specific, or inducible) capable of driving gene expression in target cells. To produce infectious recombinant AAV (rAAV) containing a heterologous gene, a suitable producer cell line is transfected with a rAAV vector containing a heterologous gene. The producer cell is concurrently transfected with a second plasmid harboring the AAV rep and cap genes under the

control of their respective endogenous promoters or heterologous promoters. Finally, the producer cell is infected with a helper virus.

[0160] Once these factors come together, the heterologous gene is replicated and packaged as though it were a wild-type AAV genome. When target cells are infected with the resulting rAAV virions, the heterologous gene enters and is expressed in the target cells. Because the target cells lack the rep and cap genes and the adenovirus helper genes, the rAAV cannot further replicate, package or form wild-type AAV.

[0161] The use of helper virus, however, presents a number of problems. First, the use of adenovirus in a rAAV production system causes the host cells to produce both rAAV and infectious adenovirus. The contaminating infectious adenovirus can be inactivated by heat treatment (56 degree. C. for 1 hour). Heat treatment, however, results in approximately a 50% drop in the titer of functional rAAV virions. Second, varying amounts of adenovirus proteins are present in these preparations. For example, approximately 50% or greater of the total protein obtained in such rAAV virion preparations is free adenovirus fiber protein. If not completely removed, these adenovirus proteins have the potential of eliciting an immune response from the patient. Third, AAV vector production methods which employ a helper virus require the use and manipulation of large amounts of high titer infectious helper virus, which presents a number of health and safety concerns, particularly in regard to the use of a herpesvirus. Fourth, concomitant production of helper virus particles in rAAV virion producing cells diverts large amounts of host cellular resources away from rAAV virion production, potentially resulting in lower rAAV virion yields.

[0162] e. Other Viral Vectors

[0163] The development and utility of viral vectors for gene delivery is constantly improving and evolving. Other viral vectors such as poxvirus; e.g., vaccinia virus (Gnant et al., 1999; Gnant et al., 1999), alpha virus; e.g., sindbis virus, Semliki forest virus (Lundstrom, 1999), reovirus (Coffey et al., 1998) and influenza A virus (Neumann et al., 1999) are contemplated for use in the present invention and may be selected according to the requisite properties of the target system.

[0164] In certain embodiments, vaccinia viral vectors are contemplated for use in the present invention. Vaccinia virus is a particularly useful eukaryotic viral vector system for expressing heterologous genes. For example, when recombinant vaccinia virus is properly engineered, the proteins are synthesized, processed and transported to the plasma membrane. Vaccinia viruses as gene delivery vectors have recently been demonstrated to transfer genes to human tumor cells, e.g., EMAP-II (Gnant, et al., 1999), inner ear (Derby et al., 1999), glioma cells, e.g., p 53 (Timiryasova et al., 1999) and various mammalian cells, e.g., P-450 (U.S. Pat. No. 5,506,138). The preparation, growth and manipulation of vaccinia viruses are described in U.S. Pat. No. 5,849,304 and U.S. Pat. No. 5,506,138 (each specifically incorporated herein by reference).

[0165] In other embodiments, sindbis viral vectors are contemplated for use in gene delivery. Sindbis virus is a species of the alphavirus genus (Garoff and Li, 1998) which

includes such important pathogens as Venezuelan, Western and Eastern equine encephalitis viruses (Sawai et al., 1999; Mastrangelo et al., 1999). In vitro, sindbis virus infects a variety of avian, mammalian, reptilian, and amphibian cells. The genome of sindbis virus consists of a single molecule of single-stranded RNA, 11,703 nucleotides in length. The genomic RNA is infectious, is capped at the 5' terminus and polyadenylated at the 3' terminus, and serves as mRNA. Translation of a vaccinia virus 26S mRNA produces a polyprotein that is cleaved co- and post-translationally by a combination of viral and presumably host-encoded proteases to give the three virus structural proteins, a capsid protein (C) and the two envelope glycoproteins (E1 and PE2, precursors of the virion E2).

[0166] Three features of sindbis virus suggest that it would be a useful vector for the expression of heterologous genes. First, its wide host range, both in nature and in the laboratory. Second, gene expression occurs in the cytoplasm of the host cell and is rapid and efficient. Third, temperature-sensitive mutations in RNA synthesis are available that may be used to modulate the expression of heterologous coding sequences by simply shifting cultures to the non-permissive temperature at various time after infection. The growth and maintenance of sindbis virus is known in the art (U.S. Pat. No. 5,217,879, specifically incorporated herein by reference).

[0167] f. Chimeric Viral Vectors

[0168] Chimeric or hybrid viral vectors are being developed for use in therapeutic gene delivery and are contemplated for use in the present invention. Chimeric poxviral/retroviral vectors (Holzer et al., 1999), adenoviral/retroviral vectors (Feng et al., 1997; Bilbao et al., 1997; Caplen et al., 1999) and adenoviral/adenovirus-associated viral vectors (Fisher et al., 1996; U.S. Pat. No. 5,871,982) have been described.

[0169] These "chimeric" viral gene transfer systems can exploit the favorable features of two or more parent viral species. For example, Wilson et al., provide a chimeric vector construct which comprises a portion of an adenovirus, AAV 5' and 3' ITR sequences and a selected transgene, described below (U.S. Pat. No. 5,871,983, specifically incorporate herein by reference).

[0170] The adenovirus/AAV chimeric virus uses adenovirus nucleic acid sequences as a shuttle to deliver a recombinant AAV/transgene genome to a target cell. The adenovirus nucleic acid sequences employed in the hybrid vector can range from a minimum sequence amount, which requires the use of a helper virus to produce the hybrid virus particle, to only selected deletions of adenovirus genes, which deleted gene products can be supplied in the hybrid viral production process by a selected packaging cell. At a minimum, the adenovirus nucleic acid sequences employed in the pAdA shuttle vector are adenovirus genomic sequences from which all viral genes are deleted and which contain only those adenovirus sequences required for packaging adenoviral genomic DNA into a preformed capsid head. More specifically, the adenovirus sequences employed are the cis-acting 5' and 3' inverted terminal repeat (ITR) sequences of an adenovirus (which function as origins of replication) and the native 5' packaging/enhancer domain, that contains sequences necessary for packaging linear Ad genomes and enhancer elements for the E1 promoter. The adenovirus sequences may be modified to contain desired

deletions, substitutions, or mutations, provided that the desired function is not eliminated.

[0171] The AAV sequences useful in the above chimeric vector are the viral sequences from which the rep and cap polypeptide encoding sequences are deleted. More specifically, the AAV sequences employed are the cis-acting 5' and 3' inverted terminal repeat (ITR) sequences. These chimeras are characterized by high titer transgene delivery to a host cell and the ability to stably integrate the transgene into the host cell chromosome (U.S. Pat. No. 5,871,983, specifically incorporate herein by reference). In the hybrid vector construct, the AAV sequences are flanked by the selected adenovirus sequences discussed above. The 5' and 3' AAV ITR sequences themselves flank a selected transgene sequence and associated regulatory elements, described below. Thus, the sequence formed by the transgene and flanking 5' and 3' AAV sequences may be inserted at any deletion site in the adenovirus sequences of the vector. For example, the AAV sequences are desirably inserted at the site of the deleted E1a/E1b genes of the adenovirus. Alternatively, the AAV sequences may be inserted at an E3 deletion, E2a deletion, and so on. If only the adenovirus 5' ITR/packaging sequences and 3' ITR sequences are used in the hybrid virus, the AAV sequences are inserted between them.

[0172] The transgene sequence of the vector and recombinant virus can be a gene, a nucleic acid sequence or reverse transcript thereof, heterologous to the adenovirus sequence, which encodes a protein, polypeptide or peptide fragment of interest. The transgene is operatively linked to regulatory components in a manner which permits 5L transgene transcription. The composition of the transgene sequence will depend upon the use to which the resulting hybrid vector will be put. For example, one type of transgene sequence includes a therapeutic gene which expresses a desired gene product in a host cell. These therapeutic genes or nucleic acid sequences typically encode products for administration and expression in a patient in vivo or ex vivo to replace or correct an inherited or non-inherited genetic defect or treat an epigenetic disorder or disease.

E. *CHLAMYDIA* ANTIBODIES

[0173] In another aspect, the present invention provides antibody compositions that are immunoreactive with a *Chlamydia* polypeptide of the present invention, or any portion thereof.

[0174] An antibody can be a polyclonal or a monoclonal antibody. An antibody may also be monovalent or bivalent. A prototype antibody is an immunoglobulin composed by four polypeptide chains, two heavy and two light chains, held together by disulfide bonds. Each pair of heavy and light chains forms an antigen binding site, also defined as complementarity-determining region (CDR). Therefore, the prototype antibody has two CDRs, can bind two antigens, and because of this feature is defined bivalent. The prototype antibody can be split by a variety of biological or chemical means. Each half of the antibody can only bind one antigen and, therefore, is defined monovalent. Means for preparing and characterizing antibodies are well known in the art (see, e.g., Howell and Lane, 1988).

[0175] Peptides corresponding to one or more antigenic determinants of a *Chlamydia* polypeptide of the present

invention also can be prepared. Such peptides should generally be at least five or six amino acid residues in length, will preferably be about 10, 15, 20, 25 or about 30 amino acid residues in length, and may contain up to about 35-50 residues or so. Synthetic peptides will generally be about 35 residues long, which is the approximate upper length limit of automated peptide synthesis machines, such as those available from Applied Biosystems (Foster City, Calif.). Longer peptides also may be prepared, e.g., by recombinant means.

[0176] The identification and preparation of epitopes from primary amino acid sequences on the basis of hydrophilicity is taught in U.S. Pat. No. 4,554,101 (Hopp), incorporated herein by reference. Through the methods disclosed in Hopp, one of skill in the art would be able to identify epitopes from within an amino acid sequence such as a *Chlamydia* polypeptide sequence.

[0177] Numerous scientific publications have also been devoted to the prediction of secondary structure, and to the identification of epitopes, from analyses of amino acid sequences (Chou & Fasman, 1974a; Chou & Fasman, 1974b; Chou & Fasman, 1978a; Chou & Fasman, 1978b; Chou & Fasman, 1979). Any of these may be used, if desired, to supplement the teachings of Hopp in U.S. Pat. No. 4,554,101.

[0178] Moreover, computer programs are currently available to assist with predicting antigenic portions and epitopic core regions of proteins. Examples include those programs based upon the Jameson-Wolf analysis (Jameson & Wolf, 1988; Wolf et al., 1988), the program PEPLOT® (Brutlag et al., 1990; Weinberger et al., 1985), and other new programs for protein tertiary structure prediction (Fetrow & Bryant, 1993). Another commercially available software program capable of carrying out such analyses is MACVECTOR (IBI, New Haven, Conn.).

[0179] In further embodiments, major antigenic determinants of a *Chlamydia* polypeptide may be identified by an empirical approach in which portions of the gene encoding the polypeptide are expressed in a recombinant host, and the resulting proteins tested for their ability to elicit an immune response. For example, PCR can be used to prepare a range of peptides lacking successively longer fragments of the C-terminus of the protein. The immunoactivity of each of these peptides is determined to identify those fragments or domains of the polypeptide that are immunodominant. Further studies in which only a small number of amino acids are removed at each iteration then allows the location of the antigenic determinants of the polypeptide to be more precisely determined.

[0180] Another method for determining the major antigenic determinants of a polypeptide is the SPOTS system (Genosys Biotechnologies, Inc., The Woodlands, Tex.). In this method, overlapping peptides are synthesized on a cellulose membrane, which following synthesis and deprotection, is screened using a polyclonal or monoclonal antibody. The antigenic determinants of the peptides which are initially identified can be further localized by performing subsequent syntheses of smaller peptides with larger overlaps, and by eventually replacing individual amino acids at each position along the immunoreactive peptide.

[0181] Once one or more such analyses are completed, polypeptides are prepared that contain at least the essential

features of one or more antigenic determinants. The peptides are then employed in the generation of antisera against the polypeptide. Minigenes or gene fusions encoding these determinants also can be constructed and inserted into expression vectors by standard methods, for example, using PCR cloning methodology.

[0182] The use of such small peptides for antibody generation or vaccination typically requires conjugation of the peptide to an immunogenic carrier protein, such as hepatitis B surface antigen, keyhole limpet hemocyanin or bovine serum albumin. Methods for performing this conjugation are well known in the art.

[0183] 1. Anti-*Chlamydia* Antibody Generation

[0184] The present invention provides monoclonal antibody compositions that are immunoreactive with a *Chlamydia* polypeptide. As detailed above, in addition to antibodies generated against a full length *Chlamydia* polypeptide, antibodies also may be generated in response to smaller constructs comprising epitopic core regions, including wild-type and mutant epitopes. In other embodiments of the invention, the use of anti-*Chlamydia* single chain antibodies, chimeric antibodies, diabodies and the like are contemplated.

[0185] As used herein, the term "antibody" is intended to refer broadly to any immunologic binding agent such as IgG, IgM, IgA, IgD and IgE. Generally, IgG and/or IgM are preferred because they are the most common antibodies in the physiological situation and because they are most easily made in a laboratory setting.

[0186] Monoclonal antibodies (mAbs) are recognized to have certain advantages, e.g., reproducibility and large-scale production, and their use is generally preferred.

[0187] However, "humanized" *Chlamydia* antibodies also are contemplated, as are chimeric antibodies from mouse, rat, goat or other species, fusion proteins, single chain antibodies, diabodies, bispecific antibodies, and other engineered antibodies and fragments thereof. As defined herein, a "humanized" antibody comprises constant regions from a human antibody gene and variable regions from a non-human antibody gene. A "chimeric antibody" comprises constant and variable regions from two genetically distinct individuals. An anti-*Chlamydia* humanized or chimeric antibody can be genetically engineered to comprise a *Chlamydia* antigen binding site of a given molecular weight and biological lifetime, as long as the antibody retains its *Chlamydia* antigen binding site.

[0188] The term "antibody" is used to refer to any antibody-like molecule that has an antigen binding region, and includes antibody fragments such as Fab', Fab; F(ab')₂, single domain antibodies (DABs), Fv, scFv (single chain Fv), chimeras and the like. Methods and techniques of producing the above antibody-based constructs and fragments are well known in the art (U.S. Pat. No. 5,889,157; U.S. Pat. No. 5,821,333, U.S. Pat. No. 5,888,773, each specifically incorporated herein by reference).

[0189] U.S. Pat. No. 5,889,157 describes a humanized B3 scFv antibody preparation. The B3 scFv is encoded from a recombinant, fused DNA molecule, that comprises a DNA sequence encoding humanized Fv heavy and light chain regions of a B3 antibody and a DNA sequence that encodes

an effector molecule. The effector molecule can be any agent having a particular biological activity which is to be directed to a particular target cell or molecule. Described in U.S. Pat. No. 5,888,773, is the preparation of scFv antibodies produced in eukaryotic cells, wherein the scFv antibodies are secreted from the eukaryotic cells into the cell culture medium and retain their biological activity: It is contemplated that similar methods for preparing multi-functional anti-*Chlamydia* fusion proteins, as described above, may be utilized in the present invention.

[0190] Means for preparing and characterizing antibodies also are well known in the art (See, e.g., *Antibodies: A Laboratory Manual*, Cold Spring Harbor Laboratory, 1988; incorporated herein by reference). The methods for generating monoclonal antibodies (mAbs) generally begin along the same lines as those for preparing polyclonal antibodies. Briefly, a polyclonal antibody is prepared by immunizing an animal with an immunogenic *Chlamydia* polypeptide composition in accordance with the present invention and collecting antisera from that immunized animal.

[0191] A wide range of animal species can be used for the production of antisera. Typically the animal used for production of antisera is a rabbit, a mouse, a rat, a hamster, a guinea pig or a goat. Because of the relatively large blood volume of rabbits, a rabbit is a preferred choice for production of polyclonal antibodies.

[0192] As is well known in the art, a given composition may vary in its immunogenicity. It is often necessary therefore to boost the host immune system, as may be achieved by coupling a peptide or polypeptide immunogen to a carrier. Exemplary and preferred carriers are keyhole limpet hemocyanin (KLH) and bovine serum albumin (BSA). Other albumins such as ovalbumin, mouse serum albumin or rabbit serum albumin also can be used as carriers. Means for conjugating a polypeptide to a carrier protein are well known in the art and include glutaraldehyde, m-maleimido-benzoyl-N-hydroxysuccinimide ester, carbodiimide and bis-biazotized benzidine.

[0193] As also well known in the art, the immunogenicity of a particular immunogen composition can be enhanced by the use of non-specific stimulators of the immune response, known as adjuvants. Suitable molecule adjuvants include all acceptable immunostimulatory compounds, such as cytokines, toxins or synthetic compositions.

[0194] Adjuvants that may be used include IL-1, IL-2, IL-4, IL-7, IL-12, γ -interferon, GM-CSF, BCG, aluminum hydroxide, MDP compounds, such as thur-MDP and nor-MDP, CGP (MTP-PE), lipid A, and monophosphoryl lipid A (MPL). RIBI, which contains three components extracted from bacteria, MPL, trehalose dimycolate (TDM) and cell wall skeleton (CWS) in a 2% squalene/Tween 80 emulsion also is contemplated. MHC antigens may even be used. Exemplary, often preferred adjuvants include complete Freund's adjuvant (a non-specific stimulator of the immune response containing killed *Mycobacterium tuberculosis*), incomplete Freund's adjuvants and aluminum hydroxide adjuvant.

[0195] In addition to adjuvants, it may be desirable to coadminister biologic response modifiers (BRM), which have been shown to upregulate T cell immunity or down-regulate suppressor cell activity. Such BRMs include, but

are not limited to, Cimetidine (CIM; 1200 mg/d) (Smith-Kline Beecham, Pa.); low-dose Cyclophosphamide (CYP; 300 mg/m²) (Johnson/Mead, N.J.), cytokines such as γ -interferon, IL-2, or IL-12 or genes encoding proteins involved in immune helper functions, such as B-7.

[0196] The amount of immunogen composition used in the production of polyclonal antibodies varies upon the nature of the immunogen as well as the animal used for immunization. A variety of routes can be used to administer the immunogen (subcutaneous, intramuscular, intradermal, intravenous and intraperitoneal). The production of polyclonal antibodies may be monitored by sampling blood of the immunized animal at various points following immunization.

[0197] A second, booster injection, also may be given. The process of boosting and titering is repeated until a suitable titer is achieved. When a desired level of immunogenicity is obtained, the immunized animal can be bled and the serum isolated and stored, and/or the animal can be used to generate mAbs.

[0198] For production of rabbit polyclonal antibodies, the animal can be bled through an ear vein or alternatively by cardiac puncture. The removed blood is allowed to coagulate and then centrifuged to separate serum components from whole cells and blood clots. The serum may be used as is for various applications or else the desired antibody fraction may be purified by well-known methods, such as affinity chromatography using another antibody, a peptide bound to a solid matrix, or by using, e.g., protein A or protein G chromatography.

[0199] mAbs may be readily prepared through use of well-known techniques, such as those exemplified in U.S. Pat. No. 4,196,265, incorporated herein by reference. Typically, this technique involves immunizing a suitable animal with a selected immunogen composition, e.g., a purified or partially purified *Chlamydia* polypeptide, peptide or domain, be it a wild-type or mutant composition. The immunizing-composition is administered in a manner effective to stimulate antibody producing cells.

[0200] The methods for generating monoclonal antibodies (mAbs) generally begin along the same lines as those for preparing polyclonal antibodies. Rodents such as mice and rats are preferred animals, however, the use of rabbit, sheep or frog cells also is possible. The use of rats may provide certain advantages (Goding, 1986, pp. 60-61), but mice are preferred, with the BALB/c mouse being most preferred as this is most routinely used and generally gives a higher percentage of stable fusions.

[0201] The animals are injected with antigen, generally as described above. The antigen may be coupled to carrier molecules such as keyhole limpet hemocyanin if necessary. The antigen would typically be mixed with adjuvant, such as Freund's complete or incomplete adjuvant. Booster injections with the same antigen would occur at approximately two-week intervals, or the gene encoding the protein of interest can be directly injected.

[0202] Following immunization, somatic cells with the potential for producing antibodies, specifically B lymphocytes (B cells), are selected for use in the mAb: generating protocol. These cells may be obtained from biopsied spleens, tonsils or lymph nodes, or from a peripheral blood

sample. Spleen cells and peripheral blood cells are preferred, the former because they are a rich source of antibody-producing cells that are in the dividing plasmablast stage, and the latter because peripheral blood is easily accessible.

[0203] Often, a panel of animals will have been immunized and the spleen of an animal with the highest antibody titer will be removed and the spleen lymphocytes obtained by homogenizing the spleen with a syringe. Typically, a spleen from an immunized mouse contains approximately 5×10^8 to 2×10^8 lymphocytes.

[0204] The antibody-producing B lymphocytes from the immunized animal are then fused with cells of an immortal myeloma cell, generally one of the same species as the animal that was immunized. Myeloma cell lines suited for use in hybridoma-producing fusion procedures preferably are non-antibody-producing, have high fusion efficiency, and enzyme deficiencies that render them incapable of growing in certain selective media which support the growth of only the desired fused cells (hybridomas).

[0205] Any one of a number of myeloma cells may be used, as are known to those of skill in the art (Goding, pp. 65-66, 1986; Campbell, pp. 75-83, 1984). For example, where the immunized animal is a mouse, one may use P3-X63/Ag8, X63-Ag8.653, NS1/1.Ag 4 1, Sp210-Ag14, FO, NSO/U, MPC-11, MPC11-X45-GTG 1.7 and S194/5XX0 Bul; for rats, one may use R210.RCY3, Y3-Ag 1.2.3, IR983F and 4B210; and U-266, GM1500-GRG2, LICR-LON-HMy2 and UC729-6 are all useful in connection with human cell fusions.

[0206] One preferred murine myeloma cell is the NS-1 myeloma cell line (also termed P3-NS-1-Ag4-1), which is readily available from the NIGMS Human Genetic Mutant Cell Repository by requesting cell line repository number GM3573. Another mouse myeloma cell line that may be used is the 8-azaguanine-resistant mouse murine myeloma SP2/0 non-producer cell line.

[0207] Methods for generating hybrids of antibody-producing spleen or lymph node cells and myeloma cells usually comprise mixing somatic cells with myeloma cells in a 2:1 proportion, though the proportion may vary from about 20:1 to about 1:1, respectively, in the presence of an agent or agents (chemical or electrical) that promote the fusion of cell membranes. Fusion methods using Sendai virus have been described by Kohler and Milstein (1975; 1976), and those using polyethylene glycol (PEG), such as 37% (v/v) PEG, by Geffer et al. (1977). The use of electrically induced fusion methods also is appropriate (Goding pp. 71-74, 1986).

[0208] Fusion procedures usually produce viable hybrids at low frequencies, about 1×10^{-6} to 1×10^{-8} . However, this does not pose a problem, as the viable, fused hybrids are differentiated from the parental, unfused cells (particularly the unfused myeloma cells that would normally continue to divide indefinitely) by culturing in a selective medium. The selective medium is generally one that contains an agent that blocks the de novo synthesis of nucleotides in the tissue culture media. Exemplary and preferred agents are aminopterin, methotrexate, and azaserine. HAT medium, as growth medium containing hypoxanthine, aminopterin and thymidine, is well known in the art as a medium for selection of hybrid cells. Aminopterin and methotrexate block de novo

synthesis of both purines and pyrimidines, whereas azaserine blocks only purine synthesis. Where aminopterin or methotrexate is used, the media is supplemented with hypoxanthine and thymidine as a source of nucleotides (HAT medium). Where azaserine is used, the media is supplemented with hypoxanthine.

[0209] The preferred selection medium is HAT. Only cells capable of operating nucleotide salvage pathways are able to survive in HAT medium. The myeloma cells are defective in key enzymes of the salvage pathway, e.g., hypoxanthine phosphoribosyl transferase (HPRT), and they cannot survive. The B cells can operate this pathway, but they have a limited life span in culture and generally die within about two weeks. Therefore, the only cells that can survive in the selective media are those hybrids formed from myeloma and B cells.

[0210] This culturing provides a population of hybridomas from which specific hybridomas are selected. Typically, selection of hybridomas is performed by culturing the cells by single-clone dilution in microtiter plates, followed by testing the individual clonal supernatants (after about two to three weeks) for the desired reactivity. The assay should be sensitive, simple and rapid, such as radioimmunoassays, enzyme immunoassays, cytotoxicity assays, plaque assays, dot immunobinding assays, and the like.

[0211] The selected hybridomas then would be serially diluted and cloned into individual antibody-producing cell lines, which clones can then be propagated indefinitely to provide mAbs. The cell lines may be exploited for mAb production in two basic ways. First, a sample of the hybridoma can be injected (often into the peritoneal cavity) into a histocompatible animal of the type that was used to provide the somatic and myeloma cells for the original fusion (e.g., a syngeneic mouse). Optionally, the animals are primed with a hydrocarbon, especially oils such as pristane (tetramethylpentadecane) prior to injection. The injected animal develops tumors secreting the specific monoclonal antibody produced by the fused cell hybrid. The body fluids of the animal, such as serum or ascites fluid, can then be tapped to provide mAbs in high concentration. Second, the individual cell lines could be cultured in vitro, where the mAbs are naturally secreted into the culture medium from which they can be readily obtained in high concentrations. mAbs produced by either means may be further purified, if desired, using filtration, centrifugation and various chromatographic methods such as HPLC or affinity chromatography. Fragments of the monoclonal antibodies of the invention can be obtained from the monoclonal antibodies so produced by methods which include digestion with enzymes, such as pepsin or papain, and/or by cleavage of disulfide bonds by chemical reduction. Alternatively, monoclonal antibody fragments encompassed by the present invention can be synthesized using an automated peptide synthesizer.

[0212] It also is contemplated that a molecular cloning approach may be used to generate monoclonals. For this, combinatorial immunoglobulin phagemid libraries are prepared from RNA isolated from the spleen of the immunized animal, and phagemids expressing appropriate antibodies are selected by panning using cells expressing the antigen and control cells. The advantages of this approach over conventional hybridoma techniques are that approximately

10^4 times as many antibodies can be produced and screened in a single round, and that new specificities are generated by H and L chain combination which further increases the chance of finding appropriate antibodies.

[0213] Alternatively, monoclonal antibody fragments encompassed by the present invention can be synthesized using an automated peptide synthesizer, or by expression of full-length gene or of gene fragments in, for example, *E. coli*.

F. PHARMACEUTICAL COMPOSITIONS

[0214] Aqueous compositions of the present invention comprise an effective amount of a purified *Chlamydia* polynucleotide and/or a purified *Chlamydia* a protein, polypeptide, peptide, epitopic core region, and the like, dissolved and/or dispersed in a pharmaceutically acceptable carrier and/or aqueous medium. Aqueous compositions of gene therapy vectors expressing any of the foregoing are also contemplated.

[0215] The phrases “pharmaceutically and/or pharmacologically acceptable” refer to molecular entities and/or compositions that do not produce an adverse, allergic and/or other untoward reaction when administered to an animal.

[0216] As used herein, “pharmaceutically acceptable carrier” includes any and/or all solvents, dispersion media, coatings, antibacterial and/or antifungal agents, isotonic and/or absorption delaying agents and the like. The use of such media and/or agents for pharmaceutical active substances is well known in the art. Except insofar as any conventional media and/or agent is incompatible with the active ingredient, its use in the therapeutic compositions is contemplated. Supplementary active ingredients can also be incorporated into the compositions. For animal and more particularly human administration, preparations should meet sterility, pyrogenicity, general safety and/or purity standards as required by FDA Office of Biologics standards.

[0217] The biological material should be extensively dialyzed to remove undesired small molecular weight molecules and/or lyophilized for more ready formulation into a desired vehicle, where appropriate. The active compounds may generally be formulated for parenteral administration, e.g., formulated for injection via the intravenous, intramuscular, sub-cutaneous, intralesional, and/or even intraperitoneal routes, or formulated for oral or inhaled delivery. The preparation of an aqueous compositions that contain an effective amount of purified *Chlamydia* polynucleotide or polypeptide agent as an active component and/or ingredient will be known to those of skill in the art in light of the present disclosure. Typically, such compositions can be prepared as injectables, either as liquid solutions and/or suspensions; solid forms suitable for using to prepare solutions and/or suspensions upon the addition of a liquid prior to injection can also be prepared; and/or the preparations can also be emulsified.

[0218] The pharmaceutical forms suitable for injectable use include sterile aqueous solutions and/or dispersions; formulations including sesame oil, peanut oil and/or aqueous propylene glycol; and/or sterile powders for the extemporaneous preparation of sterile injectable solutions and/or dispersions. In all cases the form must be sterile and/or must be fluid to the extent that easy syringability exists. It must be

stable under the conditions of manufacture and/or storage and/or must be preserved against the contaminating action of microorganisms, such as bacteria and/or fungi.

[0219] Solutions of the active compounds as free base and/or pharmacologically acceptable salts can be prepared in water suitably mixed with a surfactant, such as hydroxypropylcellulose. Dispersions can also be prepared in glycerol, liquid polyethylene glycols, and/or mixtures thereof and/or in oils. Under ordinary conditions of storage and/or use, these preparations contain a preservative to prevent the growth of microorganisms.

[0220] A *Chlamydia* polynucleotide or polypeptide of the present invention can be formulated into a composition in a neutral and/or salt form. Pharmaceutically acceptable salts, include the acid addition salts (formed with the free amino groups of the protein) and/or which are formed with inorganic acids such as, for example, hydrochloric and/or phosphoric acids, and/or such organic acids as acetic, oxalic, tartaric, mandelic, and/or the like. Salts formed with the free carboxyl groups can also be derived from inorganic bases such as, for example, sodium, potassium, ammonium, calcium, and/or ferric hydroxides, and/or such organic bases as isopropylamine, trimethylamine, histidine, procaine and/or the like. In terms of using peptide therapeutics as active ingredients, the technology of U.S. Pat. Nos. 4,608,251; 4,601,903; 4,599,231; 4,599,230; 4,596,792; and/or 4,578,770, each incorporated herein by reference, may be used.

[0221] The carrier can also be a solvent and/or dispersion medium containing, for example, water, ethanol, polyol (for example, glycerol, propylene glycol, and/or liquid polyethylene glycol, and/or the like), suitable mixtures thereof, and/or vegetable oils. The proper fluidity can be maintained, for example, by the use of a coating, such as lecithin, by the maintenance of the required particle size in the case of dispersion and/or by the use of surfactants. The prevention of the action of microorganisms can be brought about by various antibacterial and/or antifungal agents, for example, parabens, chlorobutanol, phenol, sorbic acid, thimerosal, and/or the like. In many cases, it will be preferable to include isotonic agents, for example, sugars and/or sodium chloride. Prolonged absorption of the injectable compositions can be brought about by the use in the compositions of agents delaying absorption, for example, aluminum monostearate and/or gelatin.

[0222] Sterile injectable solutions are prepared by incorporating the active compounds in the required amount in the appropriate solvent with various of the other ingredients enumerated above, as required, followed by filtered sterilization. Generally, dispersions are prepared by incorporating the various sterilized active ingredients into a sterile vehicle which contains the basic dispersion medium and/or the required other ingredients from those enumerated above. In the case of sterile powders for the preparation of sterile injectable solutions, the preferred methods of preparation are vacuum-drying and/or freeze-drying techniques which yield a powder of the active ingredient plus any additional desired ingredient from a previously sterile-filtered solution thereof. The preparation of more, and/or highly, concentrated solutions for direct injection is also contemplated, where the use of DMSO as solvent is envisioned to result in extremely rapid penetration, delivering high concentrations of the active agents to a small tumor area.

[0223] Upon formulation, solutions will be administered in a manner compatible with the dosage formulation and/or in such amount as is therapeutically effective. The formulations are easily administered in a variety of dosage forms, such as the type of injectable solutions described above, but drug release capsules and/or the like can also be employed.

[0224] For parenteral administration in an aqueous solution, for example, the solution should be suitably buffered if necessary and/or the liquid diluent first rendered isotonic with sufficient saline and/or glucose. These particular aqueous solutions are especially suitable for intravenous, intramuscular, subcutaneous and/or intraperitoneal administration. In this connection, sterile aqueous media which can be employed will be known to those of skill in the art in light of the present disclosure. For example, one dosage could be dissolved in 1 ml of isotonic NaCl solution and/or either added to 1000 ml of hypodermoclysis fluid and/or injected at the proposed site of infusion, (see for example, "Remington's Pharmaceutical Sciences" 15th Edition, pages 1035-1038 and/or 1570-1580). Some variation in dosage will necessarily occur depending on the condition of the subject being treated. The person responsible for administration will, in any event, determine the appropriate dose for the individual subject.

[0225] A *Chlamydia* polynucleotide or protein-derived peptides and/or agents may be formulated within a therapeutic mixture to comprise about 0.0001 to 1.0 milligrams, and/or about 0.001 to 0.1 milligrams, and/or about 0.1 to 1.0 and/or even about 10 milligrams per dose and/or so. Multiple doses can also be administered.

[0226] In addition to the compounds formulated for parenteral administration, such as intravenous and/or intramuscular injection, other pharmaceutically acceptable forms include, e.g., tablets and/or other solids for oral administration; liposomal formulations; time release capsules; and/or any other form currently used, including cremes.

[0227] One may also use nasal solutions and/or sprays, aerosols and/or inhalants in the present invention. Nasal solutions are usually aqueous solutions designed to be administered to the nasal passages in drops and/or sprays. Nasal solutions are prepared so that they are similar in many respects to nasal secretions, so that normal ciliary action is maintained. Thus, the aqueous nasal solutions usually are isotonic and/or slightly buffered to maintain a pH of 5.5 to 6.5. In addition, antimicrobial preservatives, similar to those used in ophthalmic preparations, and/or appropriate drug stabilizers, if required, may be included in the formulation. Various commercial nasal preparations are known and/or include, for example, antibiotics and/or antihistamines and/or are used for asthma prophylaxis.

[0228] Additional formulations which are suitable for other modes of administration include vaginal suppositories and/or pessaries. A rectal pessary and/or suppository may also be used. Suppositories are solid dosage forms of various weights and/or shapes, usually medicated, for insertion into the rectum, vagina and/or the urethra. After insertion, suppositories soften, melt and/or dissolve in the cavity fluids. In general, for suppositories, traditional binders and/or carriers may include, for example, polyalkylene glycols and/or triglycerides; such suppositories may be formed from mixtures containing the active ingredient in the range of 0.5% to 10%, preferably 1%-2%.

[0229] Oral formulations include such normally employed excipients as, for example, pharmaceutical grades of mannitol, lactose, starch, magnesium stearate, sodium saccharine, cellulose, magnesium carbonate and/or the like. These compositions take the form of solutions, suspensions, tablets, pills, capsules, sustained release formulations and/or powders. In certain defined embodiments, oral pharmaceutical compositions will comprise an inert diluent and/or assimilable edible carrier, and/or they may be enclosed in hard and/or soft shell gelatin capsule, and/or they may be compressed into tablets, and/or they may be incorporated directly with the food of the diet. For oral therapeutic administration, the active compounds may be incorporated with excipients and/or used in the form of ingestible tablets, buccal tablets, troches, capsules, elixirs, suspensions, syrups, wafers, and/or the like. Such compositions and/or preparations should contain at least 0.1% of active compound. The percentage of the compositions and/or preparations may, of course, be varied and/or may conveniently be between about 2 to about 75% of the weight of the unit, and/or preferably between 25-60%. The amount of active compounds in such therapeutically useful compositions is such that a suitable dosage will be obtained.

[0230] The tablets, troches, pills, capsules and/or the like may also contain the following: a binder, as gum tragacanth, acacia, cornstarch, and/or gelatin; excipients, such as dicalcium phosphate; a disintegrating agent, such as corn starch, potato starch, alginic acid and/or the like; a lubricant, such as magnesium stearate; and/or a sweetening agent, such as sucrose, lactose and/or saccharin may be added and/or a flavoring agent, such as peppermint, oil of wintergreen, and/or cherry flavoring. When the dosage unit form is a capsule, it may contain, in addition to materials of the above type, a liquid carrier. Various other materials may be present as coatings and/or to otherwise modify the physical form of the dosage unit. For instance, tablets, pills, and/or capsules may be coated with shellac, sugar and/or both. A syrup of elixir may contain the active compounds sucrose as a sweetening agent methyl and/or propylparabens as preservatives, a dye and/or flavoring, such as cherry and/or orange flavor.

G. KITS

[0231] Therapeutic kits of the present invention are kits comprising a *Chlamydia* polynucleotide or polypeptide or an antibody to the polypeptide. Such kits will generally contain, in a suitable container, a pharmaceutically acceptable formulation of a *Chlamydia* polynucleotide or polypeptide, or an antibody to the polypeptide, or vector expressing any of the foregoing in a pharmaceutically acceptable formulation. The kit may have a single container, and/or it may have a distinct container for each compound.

[0232] When the components of the kit are provided in one and/or more liquid solutions, the liquid solution is an aqueous solution, with a sterile aqueous solution being particularly preferred. The *Chlamydia* polynucleotide or polypeptide, or antibody compositions may also be formulated into a syringeable composition. In which case, the container may itself be a syringe, pipette, and/or other such like apparatus, from which the formulation may be applied to an infected area of the body, injected into an animal, and/or even applied to and/or mixed with the other components of the kit.

[0233] However, the components of the kit may be provided as dried powder(s). When reagents and/or components are provided as a dry powder, the powder can be reconstituted by the addition of a suitable solvent. It is envisioned that the solvent may also be provided in another container.

[0234] The container will generally include at least one vial, test tube, flask, bottle, syringe and/or other container, into which the *Chlamydia* polynucleotide or polypeptide, or antibody formulation are placed, preferably, suitably allocated. The kits may also comprise a second container for containing a sterile, pharmaceutically acceptable buffer and/or other diluent.

[0235] The kits of the present invention will also typically include a means for containing the vials in close confinement for commercial sale, such as, e.g., injection and/or blow-molded plastic containers into which the desired vials are retained.

[0236] Irrespective of the number and/or type of containers, the kits of the invention may also comprise, and/or be packaged with, an instrument for assisting with the injection/administration and/or placement of the ultimate *Chlamydia* polynucleotide or polypeptide, or an antibody to the polypeptide within the body of an animal. Such an instrument may be a syringe, pipette, forceps, and/or any such medically approved delivery vehicle.

EXAMPLES

[0237] The following examples are included to demonstrate preferred embodiments of the invention. It should be appreciated by those of skill in the art that the techniques disclosed in the examples which follow represent techniques discovered by the inventor to function well in the practice of the invention, and thus can be considered to constitute preferred modes for its practice. However, those of skill in the art should, in light of the present disclosure, appreciate that many changes can be made in the specific embodiments which are disclosed and still obtain a like or similar result without departing from the spirit and scope of the invention.

Example 1

Exemplary ELI Protocol

[0238] The following sections outline general methodology that one might use to prepare, screen and utilize ELI according to the present invention. Of course the following methods are merely general guidelines and should not limit one of skill in the art from modifying the present invention to accomplish a desired goal using ELI.

[0239] 1. Library-Construction

[0240] The present invention provides expression library constructs of genus *Chlamydia psittaci*. An expression library of *Chlamydia psittaci* can be produced by first physically shearing the genomic DNA of *Chlamydia psittaci* (e.g., *Chlamydia psittaci* strain B577) and size-selecting fragments of 300-800 base pairs. The protocol used by the present inventors to produce a *Chlamydia psittaci* library is similar to that described in Sykes and Johnston (1999). Adaptors were added and the DNA fragments ligated into a genetic immunization vector (FIG. 2) designed to link fragments to the mouse ubiquitin gene. However, the fragments can be blunt-end cloned.

[0241] This vector is known to enhance MHC class I-restricted immune responses (Sykes and Johnston, 1999), while sterilizing immunity against *Chlamydia* is thought to be MHC class II-dependent (Morrison et al., 1995). However, any genetic immunization procedure, by the mechanism of intracellular expression of the inserted genes, will target towards class I antigen presentation. Nevertheless, both MHC class I- and class II-restricted immune responses to the expressed antigens are well documented (Barry et al., 1995; Sykes and Johnston, 1999). The inventors observed, for instance, pronounced delayed-type hypersensitivity responses, mediated by MHC II-restricted CD4⁺Th1 cells, against protective *Chlamydia psittaci* B577 antigens, which were expressed from the ubiquitin fusion vector. In addition to the fact that MHC M-restricted immunity is generated by the ubiquitin fusion vector, MHC I-restricted immunity appears to mediate protection in the early phase of chlamydial infection (Morrison et al., 1995; Rottenberg et al., 1999). This duality of the cellular immune response generated by the ubiquitin fusion vector might explain the efficacy of this vector for genetic immunization against intracellular bacteria.

[0242] A library of approximately 82,000 individual members was created and tested as 27 sub-libraries each with 2,400-3,400 plasmid clones. The average insert frequency was approximately 67% and the average insert size was 660 base pairs. Nitrocellulose replica filters were made of each original colony plating of a sub-library pool for subsequent retrieval of positive clones. This generated a library with approximately six-fold expression-equivalent redundancy. One expression equivalent is defined as the number of in-frame fragments necessary to completely represent all authentic open reading frames. Since the genome size of *Chlamydia psittaci* is approximately 1×10^6 base pairs and only one-sixth of the actual open-reading frames will be cloned in the right orientation and frame, it requires at least six genomic equivalents to encode one expression equivalent. Each sub-library was propagated on plates and harvested to prepare DNA. DNA representing each sub-library was used for genetic immunization of mice in the following section.

[0243] 2. Vaccination and Challenge

[0244] For the first round of testing, outbred, 6-week old, female NIH-Swiss Webster mice were inoculated with the purified DNA of each sub-library using both intra-muscular (i.m.) and epidermal injection. The epidermal injection was effected with a gene gun (Sanford et al., 1991). Each mouse was given 50 μ g DNA i.m. and 5 μ g DNA by gene gun. It has been argued that the gene gun immunization favors a Th2 and the i.m. injection a Th1 type response (Feltquate et al., 1997), therefore both types of injection were given to each group. In the first round of testing, the prime inoculation was followed by a boost 9 weeks later, before intranasal challenge with 3×10^6 inclusion forming units (IFU) of *Chlamydia psittaci* strain B577 13 weeks after prime inoculation. The animals were sacrificed 12 days after the challenge, and lungs were weighed.

[0245] 3. Library Deconvolution

[0246] The basic scheme for handling the reduction of the libraries is depicted in FIG. 3. Fourteen groups out of the first round looked promising, so the individual clones from these groups were picked and grown in 96 well microtiter

plates. This gave approximately 40,000 wells in microtiter plates, therefore about 40,000 clones. The second round was reduced using a two dimensional array format. As depicted in FIG. 3, the DNA was prepared from colonies pooled from rows and columns of the array. The rationale was that if a row and column conferred protection, the colonies at the intersection would be responsible. This scheme is premised on largely additive effects of the protective clones. This 24x24 array yielded pools of ~1,700 clones with each intercession having ~96 clones. Currently the inventors deconvolute the second round with a 3-dimensional array.

[0247] Since the lung weight was highly variable in the outbred NIH-Swiss mice with variable MHC background, the inventors decided to use inbred BALB/c mice in subsequent rounds. The 48 DNA pools for round two were i.m. injected into BALB/c mice at 50 µg DNA/animal, and the animals were boosted at seven weeks by both gene gun inoculation and i.m. injection. The mice were given a higher *Chlamydia psittaci* challenge, 1.6×10^6 IFU *Chlamydia psittaci* B577, at approximately 12 weeks, again to further differentiate the groups. Animals were sacrificed and results evaluated as in round one.

[0248] In the fourth round, the animals received two boosts rather than one, and the challenge inoculum was increased to 3×10^6 IFU *Chlamydia psittaci* B577 to increase the selectivity of protection scoring. Furthermore, because too much DNA may lead to a decrease in cellular immune response, the amount of each individual clone was reduced by half, with the difference made up with pUC118 DNA, so each mouse received a total 50 µg DNA for i.m. immunization, but only 25 µg of the specific clone. The inventors also decreased the gene gun DNA in the same manner: 1.25 µg/ear of the specific clone and 1.25 µg pUC 18. Mice were boosted i.m. at both four and nine weeks after prime inoculation, and were challenged. The results of this final round are depicted in FIG. 5.

[0249] 4. Analysis of Sequences

[0250] The clones conferring protection were re-sequenced and then compared by BLAST search to Genbank and particularly to the recently completed *Chlamydia pneumoniae* (Kalman et al., 1999) genome sequences (FIG. 6). Of the 14 single genes identified in this study, ten are internal fragments and three contain the C-terminus of the protein. Of the five most protective clones, one was from a putative outer membrane protein and one was from a cell surface protein. The other three were from cytosolic proteins.

[0251] Four of the 14 clones have sequence similarity to a class of proteins known as putative outer membrane proteins (POMPs) in *Chlamydia psittaci* and *Chlamydia pneumoniae*. Many of the "putative" outer membrane proteins are known to be localized to the outer membrane and to be highly immunogenic (Longbottom et al., 1996; Tan et al., 1990).

[0252] 5. Mixing Experiment

[0253] The two dimensional approach used to find protective gene fragments assumes that the protection is due to a single highly protective gene within a pool. To verify that such genes would be found, 25 ng (i.e. $1/2000$) of either of the two most protective genes was added to a pool that scored negative (pool 6 round 1). As depicted in FIG. 7, spiking with either clone converted the negative library to a positive.

Example 2

Materials and Methods

[0254] Library construction. *Chlamydia psittaci* strain B577 (ATCC VR-656) was grown in BGMK cells and elementary bodies (EB) were purified by renograff gradient centrifugation as described (Huang et al., 1999). Genomic DNA was isolated from EB by proteinase K and RNase digestion followed by cetyl-trimethyl ammonium bromide (Kaltenbock et al., 1997).

[0255] Genomic DNA was physically sheared using a nebulizer (Glas Col, Terra Haute, IN), then size fractionated on a 1.5% TBE agarose gel. Agarose with fragments between 300-700 base pairs was excised and the DNA was electroeluted. Adaptors (top strand 5': GATCTGGATCCGAT (SEQ ID NO.2) ATCGGGCTCCA (SEQ ID NO.3) onto the fragments, then the fragments were cloned into pCMVi-UBs at the Bgl II site (See FIG. 6 and Sykes and Johnston, 1999 for more details). The ligations were transformed into DH 5 alpha electrocompetent cells and plated onto 150 mm diameter YT-Ampicillin (75 µg/mL final concentration) plates. The resulting plates had between 2400-3400 individual clones per plate. After the plates were incubated overnight at 37° C., the colonies from were lifted using nitrocellulose filters soaked in L-Broth with 8% DMSO, and these filters were stored at -80° C. The original agar plates were then incubated at 37° C. for an additional six hours. Ten mL of L Broth was added to each plate, the *E. coli* was scraped into 150 mL of L-Broth and grown at 37° C. for 30 minutes. Ampicillin was then added to a final concentration of 50 µg/mL, and the cultures were grown overnight at 37° C. Cells were pelleted and the DNA was purified using Qiagen tip 500 columns.

[0256] Inoculation of DNA. Round One: DNA from the pools was injected into 6-week old female NIH-Swiss mice. All mice received 50 µg total DNA by i.m. injections; evenly distributed between the quadriceps and tibialis anterior muscles. Eighteen of the groups also received gene gun inoculations (wand), with 2.5 µg DNA inoculated into each ear. The animals were boosted once at nine weeks in the same manner as the primary inoculation—all mice received i.m. injections, but only the same 18 groups received gene gun injections—then intranasally challenged with 5.5×10^5 IFU of *Chlamydia psittaci* strain B577 at 13 weeks. The mice were sacrificed 11 days after the challenge, and lungs were weighed.

[0257] Round Two: Nitrocellulose filters from the positive pools were placed on L-Broth Bio-Assay plates supplemented with 75 µg/mL ampicillin and 2% agar. The filters were incubated on the plates for approximately 15 minutes, then the nitrocellulose was discarded. The colonies were grown at 30° C. for 12 hours. The majority of the colonies were picked into 96 well microtiter plates containing HYT

media (1.6% Bacto-tryptone, 1.0% Bacto-yeast extract, 85.5 mM NaCl, 36 mM K_2HPO_4 , 13.2 mM KH_2PO_4 , 1.7 mM Sodium citrate, 0.4 mM $MgSO_4$, 6.8 mM ammonium sulfate, 4.4% wt/vol glycerol) supplemented with 75 μ g/mL ampicillin, using a Hybaid colony picker; the plates were then visually inspected and the remainder of the colonies were hand-picked. The microtiter plates were designated by their original pool-number and by the order in which they were picked. Hence, plate 5.10 was from original pool 5 and was the tenth plate picked. The colonies were subdivided into groups as is indicated in FIG. 2. All of the microtiter plates comprising a pool were stamped onto on L-Broth Bio-Assay plates supplemented with 75 μ g/mL ampicillin and were grown overnight at 37° C. The cells from these plates were harvested by adding L-Broth to the plates and scraping off the cells. The cells were pelleted by centrifugation then resuspended in Qiagen buffer P1. The remainder of the DNA prep proceeded according to manufacture's instructions.

[0258] These 48 DNA pools were i.m. injected into 6-week old BALB/c mice at 50 μ g DNA/animal. For the initial inoculation, the mice did not receive gene gun inoculations. At seven weeks, the mice were boosted with 50 μ g DNA/animal. In addition to the i.m. injections, the first 31 groups received gene gun (Rumsey-Loomis) inoculations at 2.5 μ g DNA/ear; however, the gene gun failed at group 32, and the last 17 groups received only i.m. injections. The mice were given a higher challenge, 1.6×10^6 IFU *Chlamydia psittaci* B577, at 12 weeks. Animals were sacrificed as in round one.

[0259] Round Three: Colonies from the microtiter plates that were judged to be positive were arrayed as in FIG. 2. For each pool, new microtiter plates with HYT media supplemented with 75 μ g/mL ampicillin were constructed from all of the colonies-which comprise the. Colonies were grown and DNA prepared as in round two.

[0260] The mice received both gene gun (wand) and i.m. inoculations at the dosage indicated above. At six weeks, the mice were boosted with 50 μ g DNA/animal, but only by i.m. injections. The challenge schedule was the same as in Round Two.

[0261] Round Four: *E coli* from wells at either full by full protection or full by partial protection was streaked out onto YT-plates supplemented with 75 μ g/mL ampicillin. Six colonies from each of the plates were tested by PCR colony screening, using the primers FS-UB 5': CCGCAC-CCTCTCTGATTAC, (SEQ ID No: 4) CTGGAGTG-GCAACTTCC. (SEQ ID NO. 5) Colonies with different sizes, hence different inserts, were sequenced using ABI Big Dye terminator and the FS-UB primer. Samples were purified on G-50 spin columns, and run on an ABI 377

Sequencer. The generated sequences were analyzed for open reading frames using a program designed by Simon Raynor, Ph.D.

Example 3

Vaccination and Challenge

[0262] It was established that the weight increase of the infected lung over the lung weight of naïve, uninfected controls (~120. mg) correlated strongly with disease intensity. Maximum disease in this model resulted in approximately 250% lung weight increase, while further lung weight increases were lethal. The lung disease on day 12 after inoculation was characterized by areas of gross lung tissue consolidation and the presence of mononuclear interstitial infiltrates in consolidated tissue. *Chlamydia* 1 inclusions were observed by immunohistochemistry in many macrophages, but rarely in other cells. Controls for complete protection were established by low level intranasal infection of naïve mice with 3×10 IFU of *Chlamydia psittaci* strain B577 4 weeks prior to challenge. These mice were completely protected from disease after challenge infection and had lung weight increases of 10-30% compared to naïve animals. Lungs of completely protected mice did not show gross lung lesions, and pathohistological examination revealed no interstitial infiltrates, but prominent peribronchiolar lymphocytic cuffs, interpreted as sign of protective immune stimulation. The chlamydial lung burden on day 11 after challenge was typically $1-3 \times 10^6$ IFU per 100 mg lung tissue in protected, and $2-6 \times 10^6$ IFU per 100 mg lung in diseased animals. Since the lowest chlamydial burden was, however, not consistently associated with lowest disease, the inventors used the disease-dependent parameter lung weight rather than chlamydial burden as readout for evaluation of protection. The lung weights were transformed to relative protection scores in a linear equation that assumed the high average lung weight I of the severely ill, naïve, challenged mice as 0 and that of fully protected controls as 1 (FIG. 4).

Example 4

Deconvolution of the Libraries

[0263] Since the lung weight was highly variable in the outbred NIH-Swiss mice with variable MEC background, the inventors decided to use inbred BALB/c mice in subsequent rounds. The 48 DNA pools for round two were i.m. injected into BALB/c mice at 50 μ g DNA/animal, and the animals were boosted at seven weeks by both gene gun inoculation and i.m. injection. The mice were given a higher *Chlamydia psittaci* challenge, 1.6×10^6 IFU *Chlamydia psittaci* B577, at approximately 12 weeks, again to further differentiate the groups. Animals were sacrificed and results evaluated as in round one.

[0264] The results of the Round two challenge are depicted in FIG. 4. Of the 48 groups from round two, 15

were judged to be positive, giving a total of 3936 wells. These wells were again arrayed as in round two, but the array had 112 colonies per column and 156 per row with 4-5 colonies per intersection (See FIG. 3). The mice received both gene -gun and i.m. injections at the dosage indicated above. At six weeks, the mice were boosted. Both the challenge and the sacrifice were performed as in Round two.

[0265] The positive 46 colonies from the intersection wells from Round three were sequenced, and those clones with open reading frames greater than 50 amino acids long were prepared individually and shot into mice as single genes and as a pool. Fourteen clones met these criteria. The disease scoring on each pool in rounds 1-3 are depicted in FIG. 4.

[0266] In the fourth round, the animals received two boosts rather than one, and the challenge inoculum was increased to 3×10^6 IFU *Chlamydia psittaci* B577 to increase the selectivity of protection scoring. Furthermore, because too much DNA may lead to a decrease in cellular immune response, the amount of each individual clone was reduced by half but made up the difference with pUC118 DNA, and each mouse received a total 50 μ g DNA for i.m. immunization, but only 25 μ g of the specific clone. The inventors also decreased the gene gun DNA in the same manner: 1.25 μ g/ear of the specific clone and 1.25 μ g pUC118. Mice were boosted i.m. at both four and nine weeks after prime inoculation, and were challenged. The results of this final round are depicted in FIG. 5.

Example 5

Comparison of Clones

[0267] Based on the hypothesis that sequences from genes conferring a high level of protection might be selected more

than once in the ELI process, the clones were compared against each other for overlaps. Interestingly, one of the clones, CP4 #10, did overlap with another gene, CP4 #11. The gene from which these two clones arise had been partially sequenced (Longbottom et al., 1998).

[0268] Two of the genes, CP4 #5 and CP4 #9, had an overlapping region, but they were fused to ubiquitin in opposite orientations. CP4 #5, is composed of two different *Chlamydia psittaci* DNA fragments, fused in opposite orientations. The first gene is fused to ubiquitin in the correct orientation and the correct reading frame. Interestingly, the second gene, which is in the opposite orientation to the ubiquitin gene, has an overlapping sequence to CP4 #5. It is doubtful that the protein from the second gene is produced in the mouse.

Example 6

Analysis of Sequences

[0269] The clones conferring protection were re-sequenced and then compared by BLAST search to Genbank and particularly to the recently completed *Chlamydia pneumoniae* (Kalman et al., 1999) genome sequences (FIG. 6). The full-length *Chlamydia psittaci* genes were next isolated and sequenced. Upon analysis, all nucleic acid sequences, except #4, #10, #11, and #12, were previously undisclosed in any context. Further, only portions of the sequences encoding #10 and #11 were previously disclosed. Since most protective genes would not have been predicted by any bioinformatics or information-based approach, it is likely that one will need to apply an unbiased, global approach, such as ELI to define vaccine candidates.

[0270] Table 2, lists a comparison of the *Chlamydia psittaci* genes with homologues from *Chlamydia trachomatis* and *Chlamydia pneumoniae*.

TABLE 2

	<i>Chlamydia ps</i>	<i>Chlamydia trachomatis</i>	Identity/ Similarity	<i>Chlamydia pneumoniae</i>	Identity/ Similarity
CP4 #1		DNA Pol III Gamma and Tau	62/73	DNA Pol III Gamma and Tau	66/76
CP4 #2		Glu-tRNA Gln Amidotransferase (C subunit)	49/70	Glu-tRNA Gln Amidotransferase (C subunit)	48/63
CP4 #3		Glu-tRNA Gln Amidotransferase (A subunit)	71/85	Glu-tRNA Gln Amidotransferase (A subunit)	71/84
CP4 #4	OMP 90A	Outer Membrane Protein 5	30/45	Outer Membrane Protein G Family	40/54
				Outer Membrane Protein G/I Family	28/46
CP4 #5		Transglycolase/transpeptidase	67/80	Transglycolase/transpeptidase	67/77
CP4 #6		Protein Translocase	80/89	Protein Translocase	84/92
CP4 #7				Outer Membrane Lipoprotein	60/79
CP4 #8		Oligopeptidase	60/75	Oligopeptidase	61/74
CP4 #9		Hypothetical protein	62/76	Hypothetical protein	62/77
CP4 #10		Outer Membrane Protein 4	27/42	Outer Membrane Protein G family	33/51
CP4 #11		Outer Membrane Protein 4	27/42	Outer Membrane Protein G family	33/51
CP4 #12	OMP 98 kDa	Outer Membrane Protein 5	30/43	Outer membrane Protein G family	44/58
CP4 #13		Protein Translocase	80/89	Protein Translocase	84/92
CP4 #14		Succinate Dehydrogenase	60/76	Succinate Dehydrogenase	61/77

[0271] Table 3 lists all of the cloned fragments, their corresponding full length nucleotide sequences, and the amino acid sequences encoded by both the fragments and the full length sequences. Table 2 further describes the fragments.

TABLE 3

SEQUENCE LISTING INDEX		
SEQ ID NO	CP4 NO	Description
SEQ ID NO: 6	CP4 #1	(fragment) homolog to <i>Chlamydia pneumoniae</i> DNA Pol III Gamma and Tau subunits (dnaX2 gene)
SEQ ID NO: 7	CP4 #1	Polypeptide translation corresponding to SEQ ID NO. 6, homolog to <i>Chlamydia pneumoniae</i> DNA Pol III Gamma and Tau subunits (dnaX2 gene)
SEQ ID NO: 8	CP4 #1	(full length) homolog to <i>Chlamydia pneumoniae</i> DNA Pol III Gamma and Tau subunits (dnaX2 gene)
SEQ ID NO: 9	CP4 #1	Polypeptide translation corresponding to SEQ ID NO. 8, homolog to <i>Chlamydia pneumoniae</i> DNA Pol III Gamma and Tau subunits (dnaX2 gene)
SEQ ID NO: 10	CP4 #2	(fragment) homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (C subunit) (gatC gene)
SEQ ID NO: 11	CP4 #2	Polypeptide translation corresponding to SEQ ID NO. 10, homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (C subunit) (gatC gene)
SEQ ID NO: 12	CP4 #2	(full length) homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (C subunit) (gatC gene)
SEQ ID NO: 13	CP4 #2	Polypeptide translation corresponding to SEQ ID NO. 12, homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (C subunit) (gatC gene)
SEQ ID NO: 14	CP4 #3	(fragment) homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (A subunit) (gatA gene)
SEQ ID NO: 15	CP4 #3	Polypeptide translation corresponding to SEQ ID NO. 14, homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (A subunit) (gatA gene)
SEQ ID NO: 16	CP4 #3	(full length) homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (A subunit) (gatA gene)
SEQ ID NO: 17	CP4 #3	Polypeptide translation corresponding to SEQ ID NO. 16, homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (A subunit) (gatA gene)
SEQ ID NO: 18	CP4 #3	(full length) homolog to <i>Chlamydia pneumoniae</i> Glu-tRNA Gln Amido-transferase (B subunit) (gatB gene)
SEQ ID NO: 19	CP4 #3	Polypeptide translation corresponding to SEQ ID NO. 18, homolog to <i>Chlamydia pneumoniae</i> Glu-Trna Gln Amido-transferase (B subunit) (gatB gene)
SEQ ID NO: 20	CP4 #4	(fragment) <i>Chlamydia psittaci</i> 90 kDa outer membrane protein (OMP90A gene) (Previously sequenced by Longbottom, et al); homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Protein G/I (pmp 9) and Outer Membrane Protein G (pmp 5)
SEQ ID NO: 21	CP4 #4	Polypeptide translation corresponding to SEQ ID NO. 20, <i>Chlamydia psittaci</i> 90 kDa outer membrane protein (OMP90A gene); homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Protein G/I (pmp 9) and Outer Membrane Protein G (pmp 5)
SEQ ID NO: 22	CP4 #4	(full length) <i>Chlamydia psittaci</i> 90 kDa outer membrane protein (OMP90A gene) (Previously sequenced by Longbottom, et al); homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Protein G/I (pmp 9) and Outer Membrane Protein G (pmp 5)
SEQ ID NO: 23	CP4 #4	Polypeptide translation corresponding to SEQ ID NO. 22, <i>Chlamydia psittaci</i> 90 kDa outer membrane protein (OMP90A gene); homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Protein G/I (pmp 9) and Outer Membrane Protein G (pmp 5)
SEQ ID NO: 24	CP4 #5	(fragment) homolog to <i>Chlamydia pneumoniae</i> transglycolase/transpeptidase (pbp3 gene)
SEQ ID NO: 25	CP4 #5	Polypeptide translation corresponding to SEQ ID NO. 24, homolog to <i>Chlamydia pneumoniae</i> transglycolase/transpeptidase (pbp3 gene)
SEQ ID NO: 26	CP4 #5	(full length) homolog to <i>Chlamydia pneumoniae</i> transglycolase/transpeptidase (pbp3 gene)
SEQ ID NO: 27	CP4 #5	Polypeptide translation corresponding to SEQ ID NO. 26, homolog to <i>Chlamydia pneumoniae</i> transglycolase/transpeptidase (pbp3 gene)

TABLE 3-continued

SEQUENCE LISTING INDEX		
SEQ ID NO	CP4 NO	Description
SEQ ID NO: 28	CP4 #6	(fragment) homolog to <i>Chlamydia pneumoniae</i> Protein Translocase (secA2 gene)
SEQ ID NO: 29	CP4 #6	Polypeptide translation corresponding to SEQ ID NO. 28, homolog to <i>Chlamydia pneumoniae</i> Protein Translocase (secA2 gene)
SEQ ID NO: 30	CP4 #13	(fragment) homolog to <i>Chlamydia pneumoniae</i> Protein Translocase (secA2 gene)
SEQ ID NO: 31	CP4 #13	Polypeptide translation corresponding to SEQ ID NO. 30, homolog to <i>Chlamydia pneumoniae</i> Protein Translocase (secA2 gene)
SEQ ID NO: 32	CP4 #6 & 13	(full length) homolog to <i>Chlamydia pneumoniae</i> Protein Translocase (secA2 gene)
SEQ ID NO: 33	CP4 #6 & 13	Polypeptide translation corresponding to SEQ ID NO. 32, homolog to <i>Chlamydia pneumoniae</i> Protein Translocase (secA2 gene)
SEQ ID NO: 34	CP4 #7	(fragment) homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Lipoprotein (Cpn 0278)
SEQ ID NO: 35	CP4 #7	Polypeptide translation corresponding to SEQ ID NO. 34, homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Lipoprotein (Cpn 0278 gene)
SEQ ID NO: 36	CP4 #7	(full length) homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Lipoprotein (Cpn 0278)
SEQ ID NO: 37	CP4 #7	Polypeptide translation corresponding to SEQ ID NO. 36, homolog to <i>Chlamydia pneumoniae</i> Outer Membrane Lipoprotein (Cpn 0278 gene)
SEQ ID NO: 38	CP4 #8	(fragment) homolog to <i>Chlamydia pneumoniae</i> Oligopeptidase (pepF gene)
SEQ ID NO: 39	CP4 #8	Polypeptide translation corresponding to SEQ ID NO. 38, homolog to <i>Chlamydia pneumoniae</i> Oligopeptidase (pepF gene)
SEQ ID NO: 40	CP4 #8	(full length) homolog to <i>Chlamydia pneumoniae</i> Oligopeptidase (pepF gene)
SEQ ID NO: 41	CP4 #8	Polypeptide translation corresponding to SEQ ID NO. 40, homolog to <i>Chlamydia pneumoniae</i> Oligopeptidase (pepF gene)
SEQ ID NO: 42	CP4 #9	(fragment) homolog to <i>Chlamydia pneumoniae</i> gene of unknown function, co-translationally coupled to Yop N Flagellar-Type ATPase (Cpn 0708 gene)
SEQ ID NO: 43	CP4 #9	Polypeptide translation corresponding to SEQ ID NO. 42, homolog to <i>Chlamydia pneumoniae</i> gene of unknown function, co-translationally coupled to Yop N Flagellar-Type ATPase (Cpn 0708 gene)
SEQ ID NO: 44	CP4 #9	(full length) homolog to <i>Chlamydia pneumoniae</i> gene of unknown function, co-translationally coupled to Yop N Flagellar-Type ATPase (Cpn 0708 gene)
SEQ ID NO: 45	CP4 #9	Polypeptide translation corresponding to SEQ ID NO. 44, homolog to <i>Chlamydia pneumoniae</i> gene of unknown function, co-translationally coupled to Yop N Flagellar-Type ATPase (Cpn 0708 gene)
SEQ ID NO: 46	CP4 #9	(full length) homolog to <i>Chlamydia pneumoniae</i> Yop N Flagellar-Type ATPase (yscN gene)
SEQ ID NO: 47	CP4 #9	Polypeptide translation corresponding to SEQ ID NO. 46, homolog to <i>Chlamydia pneumoniae</i> Yop N Flagellar-Type ATPase (yscN gene)
SEQ ID NO: 48	CP4 #10	(fragment) homolog to <i>Chlamydia pneumoniae</i> outer membrane protein G (pmp 2 gene) (Nucleotides 1-423 were previously sequenced by Longbottom et al.)
SEQ ID NO: 49	CP4 #10	Polypeptide translation corresponding to SEQ ID NO. 48, homolog to <i>Chlamydia pneumoniae</i> outer membrane protein G (pmp 2 gene)
SEQ ID NO: 50	CP4 #11	(fragment) homolog to <i>Chlamydia pneumoniae</i> outer membrane protein G (pmp 2 gene) (Nucleotides 1-301 were previously sequenced by Longbottom et al.)
SEQ ID NO: 51	CP4 #11	Polypeptide translation corresponding to SEQ ID NO. 50, homolog to <i>Chlamydia pneumoniae</i> outer membrane protein G (pmp 2 gene)
SEQ ID NO: 52	CP4 #10 & 11	(full length) homolog to <i>Chlamydia pneumoniae</i> outer membrane protein G (pmp 2 gene). This gene immediately follows the OMP90A gene on <i>Chlamydia psittaci</i> , and nucleotides 1-502 were published by Longbottom et al., although they did not report this as a gene.

TABLE 3-continued

SEQUENCE LISTING INDEX		
SEQ ID NO	CP4 NO	Description
SEQ ID NO: 53	CP4 #10 & 11	Polypeptide translation corresponding to SEQ ID NO. 52, homolog to <i>Chlamydia pneumoniae</i> outer membrane protein G (pmp 2 gene).
SEQ ID NO: 54	CP4 #12	(fragment) <i>Chlamydia psittaci</i> 98 kDa outer membrane protein (POMP gene) (Previously sequenced by Longbottom, et al)
SEQ ID NO: 55	CP4 #12	Polypeptide translation corresponding to SEQ ID NO. 54, <i>Chlamydia psittaci</i> 98 kDa outer membrane protein (POMP gene)
SEQ ID NO: 56	CP4 #12	(full length) <i>Chlamydia psittaci</i> 98 kDa outer membrane protein (POMP gene) (Previously sequenced by Longbottom et al.)
SEQ ID NO: 57	CP4 #12	Polypeptide translation corresponding to SEQ ID NO. 56, <i>Chlamydia psittaci</i> 98 kDa outer membrane protein (POMP gene)
SEQ ID NO: 58	CP4 #14	(fragment) homolog to <i>Chlamydia pneumoniae</i> Succinate Dehydrogenase (sdhC)
SEQ ID NO: 59	CP4 #14	Polypeptide translation corresponding to SEQ ID NO. 58, homolog to <i>Chlamydia pneumoniae</i> Succinate Dehydrogenase (sdhC gene)
SEQ ID NO: 60	CP4 #14	(full length) homolog to <i>Chlamydia pneumoniae</i> Succinate Dehydrogenase (sdhC)
SEQ ID NO: 61	CP4 #14	Polypeptide translation corresponding to SEQ ID NO. 60, homolog to <i>Chlamydia pneumoniae</i> Succinate Dehydrogenase (sdhC gene)

[0272] Of the 14 single genes identified in this study, ten are internal fragments and three contain the C-terminus of the protein. Of the five most protective clones (CP4 #1-5), one was from a putative outer membrane protein (CP4 #4) and one was from a cell surface protein-(CP4 #5). The other three were from cytosolic proteins, with CP4 #2 and CP4 #3 deriving independently from genes encoding a particular amidotransferase complex.

[0273] Four of the 14 clones have sequence similarity to a class of proteins known as putative outer membrane proteins (POMPs) in *Chlamydia psittaci* and *Chlamydia pneumoniae* (CP4 #4, CP4 #10, CP4 #11 and CP4 #12). Many of the "putative" outer membrane proteins are known to be localized to the outer membrane and to be highly immunogenic (Longbottom et al., 1996; Tan et al., 1990). The clone designated CP4 #4, is an in-frame fragment of POMP90A (Longbottom et al., 1998) and CP4 #12 is an in-frame fragment of a 98 kDa POMP which has been completely sequenced (Accession. U72499). The clones CP4 #10 and CP4 #11 immediately follow CP4 #4 in the genome and have sequence similarity to POMPs in *Chlamydia psittaci*, *Chlamydia trachomatis* and *Chlamydia pneumoniae*. As stated earlier, the clone CP4 #10 overlaps the CP4 #11 clone. Of these clones only CP4 #4 confers significant protection in isolation so clearly the criteria of being an outer membrane protein is not sufficient to predict a protective vaccine.

Example 7

Mixing Experiment

[0274] The two dimensional approach used to find protective gene fragments assumes that the protection is due to a single highly protective gene within a pool. To verify that such genes would be found, 25 ng (i.e. $\frac{1}{2000}$) of either CP4 #4 or CP4 #11 was added to a pool that scored negative (pool

6 round 1). As depicted in FIG. 7, spiking with either clone converted the negative library to a positive. Of note is that CP4 #11 did not confer protection when tested individually, however, it does protect in combination.

[0275] The fact that a CP4 #4 positive library confers protection validates the sensitivity of the system. The fact that a CP4 #11 positive library protects implies that CP4 #11 can be a useful component of a vaccine, but that it may depend upon having other antigens. present. A likely explanation is that CP4 #11 is a good vaccine antigen, but requires immunological help.

Example 8

Vaccination in Cattle

[0276] An important question is whether the genes identified in this manner in a mouse model are clinically relevant. Of course, this concern is not peculiar to genetic vaccines or ELI, but any system that uses models to identify vaccine candidates. In this case the clinically relevant situation is protection of cattle. In a preliminary experiment, the inventors evaluated the pool of 14 individual clones in the original host in a fertility challenge model. All fourteen clones were used as the individual test data on each clone in mice was not available by the time it was necessary to initiate the cow trial.

TABLE 4

<i>Chlamydia psittaci</i> Vaccine in Cows			
	Percent Pregnant	Pregnant	Not Pregnant
Not Challenged	75	3	1
Challenged,	0	0	4
Not Vaccinated			

TABLE 4-continued

<i>Chlamydia psittaci</i> Vaccine in Cows			
	Percent Pregnant	Pregnant	Not Pregnant
EB Vaccine	25	1	3
Genetic Vaccine (14 gene pool)	33	2	4

[0277] *Chlamydia psittaci* is normally introduced by the fecal-oral and respiratory routes in cattle, and disseminates to other tissues including reproductive organs. *Chlamydia psittaci* infection of the uterine mucosa reduces fertility, the basis of the economic interest in a *Chlamydia psittaci* vaccine. Four groups of heifers were used. One group was the naïve unchallenged control, another the naïve, challenged control, a third received the same pool of fourteen gene fragments that were tested in mice, and the fourth group was vaccinated with an experimental, inactivated vaccine of elementary bodies (EB) and also challenged. This EB vaccine had shown great promise in field trials but is too expensive to produce. After a prime and one boost, the heifers were estrus synchronized by prostaglandin injection, were in heat 2-3 days later, and were artificially inseminated, simultaneously receiving an intracervical chlamydial challenge of 3×10^7 inclusion forming units. The heifers were palpated for pregnancy at six weeks after insemination. This challenge was very high in order to maximize the difference between positive and negative control animals. This was necessary because only a small number of cows could be justified for this high-risk experiment.

[0278] Although the animal numbers are small, the results are quite encouraging. As is seen in Table 4, three out of four animals became pregnant in the positive control (non-challenged) group, 0/4 in the negative control (non-vaccinated, challenged) group, 2/6 in the genetic immunization group, and 1/4 in the elementary body vaccine group. The genetic vaccine of the pooled genes performed at least as well as the EB vaccine. Also relative to the inventor's interest in therapeutic vaccines, these cows were not sterile with respect to *Chlamydia psittaci* at the time of the prime inoculation. The vaccination was in the face of previous exposure and low level *Chlamydia psittaci* infection, as determined by the high titers of preinoculation antichlamydial antibodies, and occasional positivity of *Chlamydia omp1* PCRs from vaginal scrapings.

[0279] The next phase in developing a cow vaccine will be to experimentally verify the effectiveness of particular groups of the protective genes and then convert the codon usage of the *Chlamydia psittaci* genes to that of a mammal. This should increase the expression of the antigen in cows and increase the effectiveness of the vaccine. The inventors will test different combinations of those genes which have been found to be individually protective, as well as combinations with CP4 #11. Both the original fragments and their full-length versions can be tested, both as nucleic acid segments and proteins. Once the combinations have been verified in mice or other small mammals, those combinations showing the most promise will be tested in cows. After immunization, the cows will be challenged with *Chlamydia psittaci*, either by direct challenge at insemination or infec-

tion by herd-mates. Direct challenge at insemination is a very severe and unnatural form of challenge. Therefore, even if protection is not demonstrated in the wake of such challenge, this does not necessarily mean that no protection has been conferred upon the cows.

Example 9

Fertility at 42 Days Post Breeding in Heifers Vaccinated with the Pool of the 5 Best Mouse-Protective Genes of *Chlamydia psittaci*

[0280] Because it is known that bacterial genes are not expressed efficiently in mammalian cells, the five most protective genes were chemically resynthesized to give an optimal mammalian codon bias. In addition, the full-length genes corresponding to the fragments isolated in the screen were used.

[0281] One group of five heifers was vaccinated with this pool. Another group of six heifers was vaccinated with an Alum-Quil A based vaccine containing per dose 100 µg each of the affinity-purified protein fragments expressed in *E. coli* from these genes. The control group of twelve heifers was vaccinated with a plasmid expressing an unrelated bacterial gene. Six weeks after the initial immunization all groups received booster vaccinations. Eight weeks later all heifers, including a cohort of 27 non-vaccinated heifers, were estrus-synchronized by prostaglandin injection. After coming into heat two to three days later, the non-vaccinated cohort heifers were infected with an intrauterine chlamydial inoculum of 108 IFUs *C. psittaci* B577. The function of this group was to shed chlamydiae, and thus to challenge through natural infection routes the vaccinated animals at the time of breeding. Eleven days later, the vaccinated animals were re-synchronized, and inseminated at estrus. The heifers were rectally palpated for pregnancy determination at six weeks after insemination.

[0282] The Genetic Vaccine group was vaccinated with DNA comprised of the pool of 5 full length, mammalianized genes, the Protein Vaccine group with the 5 full-length proteins, and the control group with DNA of an unrelated gene from *Salmonella typhimurium*. During the 3-week period prior to *C. psittaci* infection, heifers of all groups, including the non-vaccinated challenge cohort, shed low levels of *C. psittaci* (0.5 ± 0.2 genomes/swab) as determined by qPCR of weekly collected vaginal cytobrush swabs. To challenge the vaccinated animals via natural transmission at the time of breeding, a cohort of 27 non-vaccinated animals was intracervically infected with *C. psittaci*. Eleven days later, all vaccinated groups were estrus-synchronized and inseminated. During the 4 weeks following the infection, the infected cohort animals shed high levels of chlamydiae (3826 ± 2052 genomes per swab), and then returned to low baseline shedding (24.2 ± 10.9 genomes per swab) for the remaining 5-week observation period. All vaccinated heifers were exposed to the natural challenge infection, as evident in their 7-fold increased post-breeding shedding of chlamydiae (3.6 ± 1.2 genomes/swab; $p < 0.05$) compared to pre-breeding shedding of all heifers. No difference in chlamydial shedding before or after breeding was found between the *C. psittaci* vaccinated and the control vaccinated groups.

TABLE 5

Fertility in cows vaccinated with a pool of the 5 best mouse-protective *Chlamydia psittaci* genes.

Group	Percent Pregnant	Pregnant	Not Pregnant
Control Group	50	6	6
Genetic Vaccine	80	4	1
Protein Vaccine	83	5	1

[0283] As is seen in Table 5, six out of twelve animals (50% fertility) became pregnant in the control group, 4/5 or (80% fertility in the genetic vaccine group, and five out of six (83% fertility) in the protein vaccine group. Thus, 9/11 animals in both vaccine groups were pregnant. The genetic vaccine of the pooled genes performed as well as the protein vaccine. These fertility data correspond very well with typical data of bovine herds with and without fertility problems. When both vaccine groups combined are compared to the controls, the 1-tailed Fisher's exact test indicates with a $p=0.122$ that vaccination is effective to improve *Chlamydia*-induced reduction of fertility. The odds ratio for improvement of fertility by vaccination is 4.5 (0.67-30.23, 95% confidence interval). These data are important in view of the fact that all heifers in the experiment had been previously exposed to chlamydiae and experienced low-level herd infection with *C. psittaci*, as determined by positive *C. psittaci* B577 MOMP-peptide ELISA and sporadic detection by quantitative PCR of low levels of *C. psittaci* in pre-challenge vaginal cytobrush swabs.

Example 10

Creation and Testing of Vaccines Using *Chlamydia psittaci* Nucleic Acid and Amino Acid Sequences To Protect Non-Bovine Species

[0284] The *Chlamydia psittaci* sequences and antigens disclosed in this application are envisioned to be used in vaccines for *Chlamydia psittaci* in commercially important animals such as dairy cattle: Field trials in cattle are being conducted, as described above. However, these *Chlamydia psittaci* sequences may be used to create vaccines for other species as well, including other species of *Chlamydia* and other bacterial pathogens.

[0285] For example, one may use the information gained concerning *Chlamydia psittaci* to identify a sequence in another bacterial pathogen that had substantial homology to the *Chlamydia psittaci* sequences. In many cases, this homology would be expected to be more than 30% amino acid sequence identity or similarity and could be for only part of a protein, e.g. 30 amino acids, in the other species. The gene encoding such identity/similarity may be isolated and tested as a vaccine candidate in the appropriate model system either as a protein or nucleic acid. Alternatively, the *Chlamydia psittaci* homologs may be tested directly in an animal species of interest since having so few genes to screen (10 or less) and given that the genes had been demonstrated to be protective in another species the probability of success would be high. Alternatively, proteins or peptides corresponding to the homologs to the *Chlamydia psittaci* genes may be used to assay in animals or humans for immune responses in people or animals infected with the

relevant pathogen. If such immune responses are detected, particularly if they correlated with protection, then the genes, proteins or peptides corresponding to the homologs may be tested directly in animals or humans as vaccines.

Example 11

Creation and Testing of Commercial Vaccines Using *Chlamydia psittaci* Nucleic Acid and Amino Acid Sequences

[0286] The genes identified and claimed as vaccine: candidates can be developed into commercial vaccines in the following manner. The genes identified can be converted to optimized mammalian expression sequences by changing the codons. This is a straightforward procedure, which can be easily do by one of skill in the art, and has been done for the *Chlamydia psittaci* sequences. The genes can then be tested in the relevant host, for example, cattle, for the relevant protection, for example, fertility. Genetic immunization affords a simple method to test vaccine candidate for efficacy and this form of delivery has been used in a wide variety of animals including humans. Alternatively, the genes may be transferred to another vector, for example, a vaccinia vector, to be tested in the relevant host in this form. Alternatively, the corresponding protein, with or without adjuvants may be tested. These tests may be done on a relatively small number of animals. Once conducted, a decision can be made as to how many of the protective antigens to include in a larger test. Only a subset may be chosen based on the economics of production. A large field trial may be conducted using the formulation arrived at. Based on the results of the field trial, possibly done more than once at different locations, a commercial vaccine would go into production.

Example 12

Creation and Testing of Vaccines Against Other Pathogens Using *Chlamydia* Nucleic Acid and Amino Acid Sequences

[0287] Since *Chlamydia pneumonia* has a similar pathobiology as *Chlamydia psittaci*, the inventors take advantage of the screening already accomplished on the *Chlamydia psittaci* genome to test *Chlamydia pneumoniae* for homologs corresponding to the ones from *Chlamydia psittaci* as vaccine candidates. Those of ordinary skill may expect that, as one moved evolutionarily away from *Chlamydia psittaci*, the likelihood that the homologs would protect would presumably decline. However, researchers would be likely to test the homologs identified from even disparate species for protective ability in regard to relevant diseases, as this could reduce the search of a genome for vaccine candidates ~200-1,000 fold. Once the homologs have been identified and isolated, they may be tested in the appropriate animal model system for efficacy as a vaccine. For example, the *Chlamydia pneumonia* homologs as genes or proteins can be tested in a mouse pneumonia model or in a mouse or rabbit atherosclerosis model.

[0288] In an example, showing the applicability of the use of homology to determine protective antigens in differing genera, it has been shown that hsp65, the *Mycobacterium tuberculosis* homolog of the *Chlamydia pneumonia* hsp60 gene, is protective against *Mycobacterium tuberculosis*, just

as hsp60 is protective against *Chlamydia pneumoniae*. This validates that homologous genes from two different pathogens can result in protective genetic vaccines against those pathogens. Therefore, there is a strong impetus to use the *Chlamydia* gene sequences that have been disclosed as protective herein, and other such sequences that may be determined by the methods disclosed herein, to search for protective sequences of other species.

[0289] To prove this concept, full length gene of *Chlamydia pneumoniae* homolog of *Chlamydia psittaci* underwent PCR and the animals were challenged with *Chlamydia pneumoniae*. The gene that conferred protection: against *Chlamydia psittaci* gave the best protection against *Chlamydia pneumoniae*. As demonstrated in FIG. 8 and Table 6, the genes of *Chlamydia pneumoniae* dnaX2 (SEQ. ID NO 62), gatA (SEQ. ID NO 64); pbp3 (SEQ. ID NO 66); and the unknown gene 0278 (SEQ. ID NO 68), and their respective amino acid sequences (SEQ. ID NO 63, SEQ. ID NO 65, SEQ. ID NO 67, and SEQ. ID NO 69), conferred protection against *Chlamydia pneumoniae*.

TABLE 6

Protection (log of colonies in lung) against <i>Chlamydia pneumoniae</i>	
Name of gene	Protection
Vaccinated Ctr	5.3
**C.pn. dnax2	5.4
C.pn. gatC	7.3
**C.pn. gatA	6.5
C.pn. Pmp5	7.4
C.pn. Pmp9	7.6
**C.pn. Pbp3	6.4
C.pn. SecA	7.2
**C.pn. Unk.0278	6.7
C.pn.pepF	7.2
C.pn. 0708	7.2
C.pn. Pmp2	7.0
pool, 5 best <i>C. psittaci</i>	7.0
Irrelev. Vacc.	6.9

**genes conferred protection

[0290] The above study indicates that, once one of ordinary skill has access to the *Chlamydia* sequences disclosed in this specification, or to additional sequences determined to be protective using any of the methods disclosed in this specification, it is easy to run a computer-based search of relevant genetic databases in order to determine homologous sequences in other pathogens. For example, these searches can be run in the BLAST database in GenBank.

[0291] Once a sequence which is homologous to a protective sequence is determined, it is possible to obtain the homologous sequence using any of a number of methods known to those of skill. For example, it is easy to PCR amplify the pathogen homolog genes from genomic DNA and clone the genes into an appropriate genetic immunization vector, such as those used for ELI. These homolog genes can then be tested in an animal model appropriate for the pathogen for which protection is sought, to determine whether homologs of the *Chlamydia* genes will protect a host from challenge with that pathogen.

[0292] For example, the dnaX2 gene from *Chlamydia psittaci* is homologous to the dnaX2 gene from *Helicobacter pylori*. Therefore, one can will amplify the dnaX2 gene from *Helicobacter pylori* genomic DNA and clone it into a genetic

immunization vector. The clone could then be tested for protection by inoculating animals with the *Helicobacter pylori* dnaX2 clone, then challenging the inoculated animals with *Helicobacter pylori* bacteria.

[0293] Of course, it is possible for one of ordinary skill to use the *Chlamydia* genes that are disclosed as protective herein, or determined to be protective using the methods disclosed herein, to obtain protective sequences from a first non-*Chlamydia* organism, then to use the protective sequences from the non-*Chlamydia* organism to search for homologous sequences in a second non-*Chlamydia* or *Chlamydia* organism. So long as a protective *Chlamydia* sequence is used as the starting point for determining at least one homology in such a chain of searches and testing, such methods are within the scope of this invention.

[0294] All of the compositions and methods disclosed and claimed herein can be made and executed without undue experimentation in light of the present disclosure. While the compositions and methods of this invention have been described in terms of preferred embodiments, it will be apparent to those of skill in the art that variations may be applied to the compositions and methods and in the steps or in the sequence of steps of the method described herein without departing from the concept, spirit and scope of the invention. More specifically, it will be apparent that certain agents which are both chemically and physiologically related may be substituted for the agents described herein while the same or similar results would be achieved. All such similar substitutes and modifications apparent to those skilled in the art are deemed to be within the spirit, scope and concept of the invention as defined by the appended claims.

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- [0295] The following references, to the extent that they provide exemplary procedural or other details supplementary to those set forth herein, are specifically incorporated herein by reference.
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- [0297] U.S. Pat. No. 5,770,414
- [0298] U.S. Pat. No. 5,824,544
- [0299] U.S. Pat. No. 5,830,725
- [0300] U.S. Pat. No. 5,851,826
- [0301] U.S. Pat. No. 5,858,744
- [0302] U.S. Pat. No. 5,879,934
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gctgaatctc tttatgatta tgtcataggg ttattcccta catctttatc cccagagttg 720
gttgcacagc cattaggttt attatctcaa gacaccttag ctacattatc agaatgtatt 780
cgacgcaaa aatacgctga agctttgctt cctgtcacga cagcgatcaa ttctggagtc 840
gcgcctatca ccttcctcca tgacctcact gttttttatc gcgatgtact gctaaacaaa 900
gatcagggaa attctcctct atcggccatc gccatgcact attccagtga atgtttatta 960
gaaatcattg atttccttgg tgaagcggcc aaacatctac aacaaactat tttgaaaaa 1020
acatttttag aaacagtcac catccatctt attcggatat gccaacgtcc ctctttagaa 1080
actctgtttt ctcaactgaa aacatccacg tttgatacag tgagaaacgt accccagcag 1140
caagaaccct cgaaaccgag tatacaacct gaaaaaact atcaagatca gagtttctta 1200
acttcacctt ctcccacgcc aaaagttcag catcaaaaag aagcttcccc ttcttttagt 1260
ggatcagcta ctatagatac gcttttacia tttgctgttg ttgagttttc cggaatttta 1320
accaaggagt aa 1332

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<210> SEQ ID NO 9

<211> LENGTH: 443

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 9

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Met Thr Ser Ala Thr Tyr Gln Val Ser Ser Arg Lys Tyr Arg Pro Gln
1           5           10           15
Thr Phe Ala Glu Met Leu Gly Gln Asp Ala Val Val Thr Val Leu Lys
20          25          30
Asn Ala Leu Gln Phe Gln Arg Val Ala His Ala Tyr Leu Phe Ser Gly
35          40          45
Ile Arg Gly Thr Gly Lys Thr Thr Leu Ala Arg Ile Phe Ala Lys Ala
50          55          60
Leu Asn Cys Lys Glu Leu Thr Pro Glu His Glu Pro Cys Asn Gln Cys
65          70          75          80
Cys Val Cys Lys Glu Ile Ser Ser Gly Thr Ser Leu Asp Val Ile Glu
85          90          95
Ile Asp Gly Ala Ser His Arg Gly Ile Glu Asp Ile Arg Gln Ile Asn
100         105         110
Glu Thr Val Leu Phe Thr Pro Ala Lys Ser Gln Tyr Lys Ile Tyr Ile
115         120         125

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Ile 130	Asp	Glu	Val	His	Met	Leu 135	Thr	Lys	Glu	Ala	Phe 140	Asn	Ser	Leu	Leu
Lys 145	Thr	Leu	Glu	Glu	Pro 150	Pro	Ser	His	Val	Lys 155	Phe	Phe	Leu	Ala	Thr 160
Thr	Glu	Asn	Tyr	Lys 165	Ile	Pro	Ser	Thr	Ile 170	Leu	Ser	Arg	Cys	Gln	Lys 175
Met	His	Leu	Lys 180	Arg	Ile	Pro	Glu	Thr	Met 185	Ile	Val	Asp	Lys 190	Leu	Ala
Ser	Ile	Ser	Gln	Ala	Gly	Gly	Ile 200	Glu	Thr	Ser	Arg	Glu 205	Ala	Leu	Leu
Pro	Ile 210	Ala	Arg	Ala	Ala	Gln 215	Gly	Ser	Leu	Arg	Asp 220	Ala	Glu	Ser	Leu
Tyr 225	Asp	Tyr	Val	Ile	Gly 230	Leu	Phe	Pro	Thr	Ser 235	Leu	Ser	Pro	Glu	Leu 240
Val	Ala	Asp	Ala	Leu 245	Gly	Leu	Leu	Ser	Gln 250	Asp	Thr	Leu	Ala	Thr 255	Leu
Ser	Glu	Cys	Ile 260	Arg	Thr	Gln	Lys	Tyr 265	Ala	Glu	Ala	Leu	Leu	Pro	Val
Thr	Thr 275	Ala	Ile	Asn	Ser	Gly 280	Val	Ala	Pro	Ile	Thr	Phe 285	Leu	His	Asp
Leu	Thr 290	Val	Phe	Tyr	Arg	Asp 295	Val	Leu	Leu	Asn	Lys 300	Asp	Gln	Gly	Asn
Ser 305	Pro	Leu	Ser	Ala	Ile 310	Ala	Met	His	Tyr	Ser 315	Ser	Glu	Cys	Leu	Leu 320
Glu	Ile	Ile	Asp	Phe 325	Leu	Gly	Glu	Ala	Ala 330	Lys	His	Leu	Gln	Gln	Thr 335
Ile	Phe	Glu	Lys 340	Thr	Phe	Leu	Glu	Thr 345	Val	Ile	Ile	His	Leu 350	Ile	Arg
Ile	Cys	Gln	Arg	Pro	Ser	Leu 360	Glu	Thr	Leu	Phe	Ser	Gln 365	Leu	Lys	Thr
Ser 370	Thr	Phe	Asp	Thr	Val	Arg 375	Asn	Val	Pro	Gln	Gln 380	Gln	Glu	Pro	Ser
Lys 385	Pro	Ser	Ile	Gln	Pro 390	Glu	Lys	His	Tyr	Gln 395	Asp	Gln	Ser	Phe	Leu 400
Thr	Ser	Pro	Ser	Pro 405	Thr	Pro	Lys	Val	Gln 410	His	Gln	Lys	Glu	Ala	Ser 415
Pro	Ser	Leu	Val 420	Gly	Ser	Ala	Thr	Ile 425	Asp	Thr	Leu	Leu	Gln 430	Phe	Ala
Val	Val 435	Glu	Phe	Ser	Gly	Ile	Leu 440	Thr	Lys	Glu					

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<210> SEQ ID NO 10
<211> LENGTH: 123
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci
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<400> SEQUENCE: 10

1999, 2000, 2001, 2002, 2003, 2004, 2005, 2006, 2007, 2008, 2009, 2010, 2011, 2012, 2013, 2014, 2015, 2016, 2017, 2018, 2019, 2020, 2021, 2022, 2023, 2024, 2025, 2026, 2027, 2028, 2029, 2030, 2031, 2032, 2033, 2034, 2035, 2036, 2037, 2038, 2039, 2040, 2041, 2042, 2043, 2044, 2045, 2046, 2047, 2048, 2049, 2050, 2051, 2052, 2053, 2054, 2055, 2056, 2057, 2058, 2059, 2060, 2061, 2062, 2063, 2064, 2065, 2066, 2067, 2068, 2069, 2070, 2071, 2072, 2073, 2074, 2075, 2076, 2077, 2078, 2079, 2080, 2081, 2082, 2083, 2084, 2085, 2086, 2087, 2088, 2089, 2090, 2091, 2092, 2093, 2094, 2095, 2096, 2097, 2098, 2099, 2100, 2101, 2102, 2103, 2104, 2105, 2106, 2107, 2108, 2109, 2110, 2111, 2112, 2113, 2114, 2115, 2116, 2117, 2118, 2119, 2120, 2121, 2122, 2123, 2124, 2125, 2126, 2127, 2128, 2129, 2130, 2131, 2132, 2133, 2134, 2135, 2136, 2137, 2138, 2139, 2140, 2141, 2142, 2143, 2144, 2145, 2146, 2147, 2148, 2149, 2150, 2151, 2152, 2153, 2154, 2155, 2156, 2157, 2158, 2159, 2160, 2161, 2162, 2163, 2164, 2165, 2166, 2167, 2168, 2169, 2170, 2171, 2172, 2173, 2174, 2175, 2176, 2177, 2178, 2179, 2180, 2181, 2182, 2183, 2184, 2185, 2186, 2187, 2188, 2189, 2190, 2191, 2192, 2193, 2194, 2195, 2196, 2197, 2198, 2199, 2200, 2201, 2202, 2203, 2204, 2205, 2206, 2207, 2208, 2209, 2210, 2211, 2212, 2213, 2214, 2215, 2216, 2217, 2218, 2219, 2220, 2221, 2222, 2223, 2224, 2225, 2226, 2227, 2228, 2229, 2230, 2231, 2232, 2233, 2234, 2235, 2236, 2237, 2238, 2239, 2240, 2241, 2242, 2243, 2244, 2245, 2246, 2247, 2248, 2249, 2250, 2251, 2252, 2253, 2254, 2255, 2256, 2257, 2258, 2259, 2260, 2261, 2262, 2263, 2264, 2265, 2266, 2267, 2268, 2269, 2270, 2271, 2272, 2273, 2274, 2275, 2276, 2277, 2278, 2279, 2280, 2281, 2282, 2283, 2284, 2285, 2286, 2287, 2288, 2289, 2290, 2291, 2292, 2293, 2294, 2295, 2296, 2297, 2298, 2299, 2300, 2301, 2302, 2303, 2304, 2305, 2306, 2307, 2308, 2309, 2310, 2311, 2312, 2313, 2314, 2315, 2316, 2317, 2318, 2319, 2320, 2321, 2322, 2323, 2324, 2325, 2326, 2327, 2328, 2329, 2330, 2331, 2332, 2333, 2334, 2335, 2336, 2337, 2338, 2339, 2340, 2341, 2342, 2343, 2344, 2345, 2346, 2347, 2348, 2349, 2350, 2351, 2352, 2353, 2354, 2355, 2356, 2357, 2358, 2359, 2360, 2361, 2362, 2363, 2364, 2365, 2366, 2367, 2368, 2369, 2370, 2371, 2372, 2373, 2374, 2375, 2376, 2377, 2378, 2379, 2380, 2381, 2382, 2383, 2384, 2385, 2386, 2387, 2388, 2389, 2390, 2391, 2392, 2393, 2394, 2395, 2396, 2397, 2398, 2399, 2400, 2401, 2402, 2403, 2404, 2405, 2406, 2407, 2408, 2409, 2410, 2411, 2412, 2413, 2414, 2415, 2416, 2417, 2418, 2419, 2420, 2421, 2422, 2423, 2424, 2425, 2426, 2427, 2428, 2429, 2430, 2431, 2432, 2433, 2434, 2435, 2436, 2437, 2438, 2439, 2440, 2441, 2442, 2443, 2444, 2445, 2446, 2447, 2448, 2449, 2450, 2451, 2452, 2453, 2454, 2455, 2456, 2457, 2458, 2459, 2460, 2461, 2462, 2463, 2464, 2465, 2466, 2467, 2468, 2469, 2470, 2471, 2472, 2473, 2474, 2475, 2476, 2477, 2478, 2479, 2480, 2481, 2482, 2483, 2484, 2485, 2486, 2487, 2488, 2489, 2490, 2491, 2492, 2493, 2494, 2495, 2496, 2497, 2498, 2499, 2500, 2501, 2502, 2503, 2504, 2505, 2506, 2507, 2508, 2509, 2510, 2511, 2512, 2513, 2514, 2515, 2516, 2517, 2518, 2519, 2520, 2521, 2522, 2523, 2524, 2525, 2526, 2527, 2528, 2529, 2530, 2531, 2532, 2533, 2534, 2535, 2536, 2537, 2538, 2539, 2540, 2541, 2542, 2543, 2544, 2545, 2546, 2547, 2548, 2549, 2550, 2551, 2552, 2553, 2554, 2555, 2556, 2557, 2558, 2559, 2560, 2561, 2562, 2563, 2564, 2565, 2566, 2567, 2568, 2569, 2570, 2571, 2572, 2573, 2574, 2575, 2576, 2577, 2578, 2579, 2580, 2581, 2582, 2583, 2584, 2585, 2586, 2587, 2588, 2589, 2590, 2591, 2592, 2593, 2594, 2595, 2596, 2597, 2598, 2599, 2600, 2601, 2602, 2603, 2604, 2605, 2606, 2607, 2608, 2609, 2610, 2611, 2612, 2613, 2614, 2615, 2616, 2617, 2618, 2619, 2620, 2621, 2622, 2623, 2624, 2625, 2626, 2627, 2628, 2629, 2630, 2631, 2632, 2633, 2634, 2635, 2636, 2637, 2638, 2639, 2640, 2641, 2642, 2643, 2644, 2645, 2646, 2647, 2648, 2649, 2650, 2651, 2652, 2653, 2654, 2655, 2656, 2657, 2658, 2659, 2660, 2661, 2662, 2663, 2664, 2665, 2666, 2667, 2668, 2669, 2670, 2671, 2672, 2673, 2674, 2675, 2676, 2677, 2678, 2679, 2680, 26

gagtttattc aaga

9a9cccccccc 229a9c299a 229cccccccc 2

atcgctatcg atgctac

qaa	123
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944

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<210> SEQ ID NO 11
 <211> LENGTH: 41
 <212> TYPE: PRT
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 11

Glu	Phe	Ile	Gln	Glu	Tyr	Glu	Ser	Ser	Leu	Asn	Glu	Val	Ile	Lys	Thr
1				5					10					15	
Met	Ala	Ala	Ser	Ile	Ala	Met	Asp	Val	Thr	Asp	Val	Val	Ile	Glu	Val
			20				25						30		
Gly	Leu	Ser	His	Val	Ile	Ser	Pro	Glu							
		35				40									

<210> SEQ ID NO 12
 <211> LENGTH: 303
 <212> TYPE: DNA
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 12

atgacacaac cctatgtaac tagagaagac attatacttc tggcgaagag ttcagctctg	60
gaattaagcg aagagtttat tcaagagtat gaaagttcct taaatgaagt cattaanaact	120
atggcagcat ccatcgctat ggatgtaacc gacgtgggta ttgaggttg tttatcccat	180
gtgatcagtc ccgaagattt acgagaagat atcgttgcct caagtttctc tctgaggag	240
tttctaacta atgtccctga atccttaggg ggattagtaa aagtaccac agtcattaag	300
tag	303

<210> SEQ ID NO 13
 <211> LENGTH: 100
 <212> TYPE: PRT
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 13

Met	Thr	Gln	Pro	Tyr	Val	Thr	Arg	Glu	Asp	Ile	Ile	Leu	Leu	Ala	Lys
1				5					10					15	
Ser	Ser	Ala	Leu	Glu	Leu	Ser	Glu	Glu	Phe	Ile	Gln	Glu	Tyr	Glu	Ser
			20				25						30		
Ser	Leu	Asn	Glu	Val	Ile	Lys	Thr	Met	Ala	Ala	Ser	Ile	Ala	Met	Asp
		35				40					45				
Val	Thr	Asp	Val	Val	Ile	Glu	Val	Gly	Leu	Ser	His	Val	Ile	Ser	Pro
	50				55						60				
Glu	Asp	Leu	Arg	Glu	Asp	Ile	Val	Ala	Ser	Ser	Phe	Ser	Arg	Glu	Glu
65				70					75					80	
Phe	Leu	Thr	Asn	Val	Pro	Glu	Ser	Leu	Gly	Gly	Leu	Val	Lys	Val	Pro
			85					90					95		
Thr	Val	Ile	Lys												
			100												

<210> SEQ ID NO 14
 <211> LENGTH: 514
 <212> TYPE: DNA
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 14

gaaaagtgtg atgtgattgc gatgcctgta tgctcatgcc cagcattcgc cgatggcgaa	60
atccttgatc ctacctctct atatctccag gatattctata ccgtggctat gaatttagcc	120

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tacctcccag ctatcgccgt tccttcaggg tttctcgcag aagggtgcc tctaggattc 180
caggtgattg gacaaaaggg taaagatcaa caggtgtgcc aggtaggcta tagcttccaa 240
gaacattcag gaattaagaa tttataccct aaaggatgta acaaacttgt tgatggagag 300
gtgaaataat gagcgacgtt tatgctgatt gggaatccgt cataggtcctt gaagtccacg 360
tagaattaaa cacaaaaatct aaattgttca gttgtgcacg caaccgtttt ggagacgaac 420
ctaatacaaa catctctcct gtatgcaccg gcatgccggg gtcactgcca gtactgaata 480
aagaagcagt gagaaaggct gttttatttg gttg 514

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<210> SEQ ID NO 15
<211> LENGTH: 102
<212> TYPE: PRT
<213> ORGANISM: Chlamydia psittaci

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<400> SEQUENCE: 15

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Glu Lys Cys Asp Val Ile Ala Met Pro Val Cys Ser Cys Pro Ala Phe
1           5           10          15
Ala Asp Gly Glu Ile Leu Asp Pro Thr Ser Leu Tyr Leu Gln Asp Ile
20          25          30
Tyr Thr Val Ala Met Asn Leu Ala Tyr Leu Pro Ala Ile Ala Val Pro
35          40          45
Ser Gly Phe Ser Arg Glu Gly Leu Pro Leu Gly Phe Gln Val Ile Gly
50          55          60
Gln Lys Gly Lys Asp Gln Gln Val Cys Gln Val Gly Tyr Ser Phe Gln
65          70          75          80
Glu His Ser Gly Ile Lys Asn Leu Tyr Pro Lys Gly Cys Asn Lys Leu
85          90          95
Val Asp Gly Glu Val Lys
100

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<210> SEQ ID NO 16
<211> LENGTH: 1476
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci

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<400> SEQUENCE: 16

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atgtatcaga agagtgcctt agagttaaga aatgccgtag tgagtggaga gtcttcagct 60
acagcaatag caaagtattt ttataataga ataaaaacag aagacaatca gataggagct 120
ttcttttctc tttgtgaaga aagagcttat gagaaagcag ctatcataga tgcgaaagtg 180
gcgcgaggag aacctttggg gaaactcgca ggtgtcccca tcgggataaa agataatatt 240
catattcggg gtttgcgcac cacttgtgct tctaaaatgt tagaaaatta tatagcgctt 300
tttgatgcta cagtcgtcga acggatagaa gctgaagatg gggtcatttt aggcaaactc 360
aatatggatg agtttgctat gggatcgaca acgcagtatt ctgctttcca tcctacgaaa 420
aatccttggg gttttacctg tgtgccagga ggatcttcag ggggatccgc cgccgcagtt 480
tctgcaagat tttgtcctat agcgttaggt tcggataccg gtggatctat acgtcagcca 540
gcagcatttt gtggagtgtt ggggtttaag cctcctatg gagccgtctc ccgttacggt 600
ttagtcgctt ttgggtcttc attagatcag ataggccctt taacaacagt tgtogaagat 660
gtcgccttag ctatggatgt attcgcaggt aaggatgata gagatgcaac ttctcagaag 720

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ttttttacag gatctttcca agaggccttg tcttttagacg ttccgagttt gatcggcgtg   780
cctatgggat ttttagacgg ttacgtgat gatgttaaag agaatttctt tgcctcttta   840
agtattttgg aacgtcaggg tagccgcatt gttgaagtgg atcttaacat cttagatcac   900
gctgtctctg tttactacat tgtcgcttct gcagaagccg caacaaatct tgcaagattt   960
gatggtattc gttacggcta tcgttctcca gaagcgcata gtatagaaga tattttatcg  1020
atctcccgcg tacaaggctt cggtaaggaa gtcatgcgta ggattctttt aggtaactat  1080
gtgttatcca ctgagcgcca aaatgtctat tataagaaag gctccgcaat tcgagcaaaa  1140
atcattcaag cttttcaaaa agcttatgaa aagtgtgatg tgattgcgat gcctgtatgc  1200
tcatgcccg cattcgccga tggcgaaatc cttgatccta cctctctata tctccaggat  1260
atctataccg tggtatgaa tttagcctac ctcccagcta tcgccgttcc ttcagggttt  1320
tctcgagaag ggctgcctct aggattccag gtgattggac aaaagggtaa agatcaacag  1380
gtgtgccagg taggctatag cttccaagaa cattcaggaa ttaagaattt ataccctaaa  1440
ggatgtaaca aacttgttga tggagaggtg aaataa                               1476

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<210> SEQ ID NO 17
<211> LENGTH: 491
<212> TYPE: PRT
<213> ORGANISM: Chlamydia psittaci

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<400> SEQUENCE: 17

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Met Tyr Gln Lys Ser Ala Leu Glu Leu Arg Asn Ala Val Val Ser Gly
1          5          10          15
Glu Ser Ser Ala Thr Ala Ile Ala Lys Tyr Phe Tyr Asn Arg Ile Lys
20        25        30
Thr Glu Asp Asn Gln Ile Gly Ala Phe Leu Ser Leu Cys Glu Glu Arg
35        40        45
Ala Tyr Glu Lys Ala Ala Ile Ile Asp Ala Lys Val Ala Arg Gly Glu
50        55        60
Pro Leu Gly Lys Leu Ala Gly Val Pro Ile Gly Ile Lys Asp Asn Ile
65        70        75        80
His Ile Arg Gly Leu Arg Thr Thr Cys Ala Ser Lys Met Leu Glu Asn
85        90        95
Tyr Ile Ala Pro Phe Asp Ala Thr Val Val Glu Arg Ile Glu Ala Glu
100       105       110
Asp Gly Val Ile Leu Gly Lys Leu Asn Met Asp Glu Phe Ala Met Gly
115       120       125
Ser Thr Thr Gln Tyr Ser Ala Phe His Pro Thr Lys Asn Pro Trp Gly
130       135       140
Leu Ser Cys Val Pro Gly Gly Ser Ser Gly Gly Ser Ala Ala Ala Val
145       150       155       160
Ser Ala Arg Phe Cys Pro Ile Ala Leu Gly Ser Asp Thr Gly Gly Ser
165       170       175
Ile Arg Gln Pro Ala Ala Phe Cys Gly Val Val Gly Phe Lys Pro Ser
180       185       190
Tyr Gly Ala Val Ser Arg Tyr Gly Leu Val Ala Phe Gly Ser Ser Leu
195       200       205
Asp Gln Ile Gly Pro Leu Thr Thr Val Val Glu Asp Val Ala Leu Ala
210       215       220

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Met	Asp	Val	Phe	Ala	Gly	Lys	Asp	Asp	Arg	Asp	Ala	Thr	Ser	Gln	Lys	
225					230					235					240	
Phe	Phe	Thr	Gly	Ser	Phe	Gln	Glu	Ala	Leu	Ser	Leu	Asp	Val	Pro	Ser	
				245					250					255		
Leu	Ile	Gly	Val	Pro	Met	Gly	Phe	Leu	Asp	Gly	Leu	Arg	Asp	Asp	Val	
			260					265					270			
Lys	Glu	Asn	Phe	Phe	Ala	Ser	Leu	Ser	Ile	Leu	Glu	Arg	Gln	Gly	Ser	
		275					280					285				
Arg	Ile	Val	Glu	Val	Asp	Leu	Asn	Ile	Leu	Asp	His	Ala	Val	Ser	Val	
	290					295					300					
Tyr	Tyr	Ile	Val	Ala	Ser	Ala	Glu	Ala	Ala	Thr	Asn	Leu	Ala	Arg	Phe	
305				310						315					320	
Asp	Gly	Ile	Arg	Tyr	Gly	Tyr	Arg	Ser	Pro	Glu	Ala	His	Ser	Ile	Glu	
			325					330					335			
Asp	Ile	Tyr	Thr	Ile	Ser	Arg	Val	Gln	Gly	Phe	Gly	Lys	Glu	Val	Met	
		340					345					350				
Arg	Arg	Ile	Leu	Leu	Gly	Asn	Tyr	Val	Leu	Ser	Thr	Glu	Arg	Gln	Asn	
		355				360						365				
Val	Tyr	Tyr	Lys	Lys	Gly	Ser	Ala	Ile	Arg	Ala	Lys	Ile	Ile	Gln	Ala	
	370				375						380					
Phe	Gln	Lys	Ala	Tyr	Glu	Lys	Cys	Asp	Val	Ile	Ala	Met	Pro	Val	Cys	
385				390					395						400	
Ser	Cys	Pro	Ala	Phe	Ala	Asp	Gly	Glu	Ile	Leu	Asp	Pro	Thr	Ser	Leu	
			405					410					415			
Tyr	Leu	Gln	Asp	Ile	Tyr	Thr	Val	Ala	Met	Asn	Leu	Ala	Tyr	Leu	Pro	
	420						425					430				
Ala	Ile	Ala	Val	Pro	Ser	Gly	Phe	Ser	Arg	Glu	Gly	Leu	Pro	Leu	Gly	
	435					440					445					
Phe	Gln	Val	Ile	Gly	Gln	Lys	Gly	Lys	Asp	Gln	Gln	Val	Cys	Gln	Val	
	450				455					460						
Gly	Tyr	Ser	Phe	Gln	Glu	His	Ser	Gly	Ile	Lys	Asn	Leu	Tyr	Pro	Lys	
465				470				475						480		
Gly	Cys	Asn	Lys	Leu	Val	Asp	Gly	Glu	Val	Lys						
		485					490									

<210> SEQ ID NO 18

<211> LENGTH: 1464

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 18

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aacacaaaat ctaaattgtt cagttgtgca cgcaaccgtt ttggagacga acctaataca	120
aacatctctc ctgtatgcac cggcacgccg gggtcactgc cagtactgaa taaagaagca	180
gtgagaaagg ctgttttatt tggttgtgct gttgaaggcg aagtagcttt gtcagccgt	240
tttgatagaa agtcctattt ttatcccgat agcccaagga attttcaa at taccgaattc	300
gaacatccta ttgtgcgagg aggacatata aaagctatcg ttcacggtga ggaacgtcat	360
tttgaactgg ctcaagcgca tatcgaagat gatgccggta tgctaaaaca ttctggagaa	420
tttgctggag tagattataa ccgcgctggt gtacotttaa tagagattgt gtctaagccg	480
tgcatgtttt gtgctgatga tgctgttgct tatgccacag ctttggtatc cttattagac	540

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tacataggca tttctgactg taatatggaa gaaggctcgg tacgctttga tgtaaacata    600
tccgtacgtc ctaaaggtag cgaagaacta cgcaataaag tagaaattaa aaatatgaac    660
tcctttgctt ttatggccca agctctagaa gccgagcgtt gtcgtcagat cgatgcatat    720
ttagacaatc caaatgcaga ccccaaaact gttattccag gagcgacata ccgttgggat    780
cctgaaaaga aaaaaacagt gttgatgcgt cttaaggaac gagctgaaga ttacaagtat    840
ttcatagagc ctgatctccc agtattgcaa ttaacagaag catatatgta tgaaattcgt    900
catacgcttc ccgagctccc tttcaacaaa taccaaaggt atttgcacga atatgctctt    960
gccgaagaca tcgctgccat tttaattagc gataagcata gtgcgcactt ctttgaatta   1020
gccgctcagg aatgtaaaaa ctacagagcc ctttctaatt ggttaactgt tgagtttgcc   1080
ggacgttgta aactcaaggg taagaatctc gctttctcag gtatcctgcc cagtagtgta   1140
gtcagcgttg tgaattttat tgatcaaggg gtgattaccg gaaagatcgc taaggatatc   1200
gcagacatga tgatggaatc tcctgaaaag agtcctgaga ctatcctcaa agaaaatcct   1260
gaaatgttgc ccatgcaga tgaaagtgcg ttggtggcga tcatttccga ggtgattacc   1320
gcaaatccgc agtctgtcgt agactacaaa agtggtgaaga ccaaggcgtt aggattttta   1380
gttggggcaa ttatgaaacg taccagggc aaggcccctc caaatagggt aaatgaactt   1440
ttgcttgtagg aattaagtaa ataa                                     1464

```

<210> SEQ ID NO 19

<211> LENGTH: 487

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 19

```

Met Ser Asp Val Tyr Ala Asp Trp Glu Ser Val Ile Gly Leu Glu Val
 1             5             10             15

His Val Glu Leu Asn Thr Lys Ser Lys Leu Phe Ser Cys Ala Arg Asn
      20             25             30

Arg Phe Gly Asp Glu Pro Asn Thr Asn Ile Ser Pro Val Cys Thr Gly
      35             40             45

Met Pro Gly Ser Leu Pro Val Leu Asn Lys Glu Ala Val Arg Lys Ala
      50             55             60

Val Leu Phe Gly Cys Ala Val Glu Gly Glu Val Ala Leu Leu Ser Arg
      65             70             75             80

Phe Asp Arg Lys Ser Tyr Phe Tyr Pro Asp Ser Pro Arg Asn Phe Gln
      85             90             95

Ile Thr Gln Phe Glu His Pro Ile Val Arg Gly Gly His Ile Lys Ala
      100            105            110

Ile Val His Gly Glu Glu Arg His Phe Glu Leu Ala Gln Ala His Ile
      115            120            125

Glu Asp Asp Ala Gly Met Leu Lys His Phe Gly Glu Phe Ala Gly Val
      130            135            140

Asp Tyr Asn Arg Ala Gly Val Pro Leu Ile Glu Ile Val Ser Lys Pro
      145            150            155            160

Cys Met Phe Cys Ala Asp Asp Ala Val Ala Tyr Ala Thr Ala Leu Val
      165            170            175

Ser Leu Leu Asp Tyr Ile Gly Ile Ser Asp Cys Asn Met Glu Glu Gly
      180            185            190

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Ser Val Arg Phe Asp Val Asn Ile Ser Val Arg Pro Lys Gly Ser Glu
 195 200 205
 Glu Leu Arg Asn Lys Val Glu Ile Lys Asn Met Asn Ser Phe Ala Phe
 210 215 220
 Met Ala Gln Ala Leu Glu Ala Glu Arg Cys Arg Gln Ile Asp Ala Tyr
 225 230 235 240
 Leu Asp Asn Pro Asn Ala Asp Pro Lys Thr Val Ile Pro Gly Ala Thr
 245 250 255
 Tyr Arg Trp Asp Pro Glu Lys Lys Lys Thr Val Leu Met Arg Leu Lys
 260 265 270
 Glu Arg Ala Glu Asp Tyr Lys Tyr Phe Ile Glu Pro Asp Leu Pro Val
 275 280 285
 Leu Gln Leu Thr Glu Ala Tyr Ile Asp Glu Ile Arg His Thr Leu Pro
 290 295 300
 Glu Leu Pro Phe Asn Lys Tyr Gln Arg Tyr Leu His Glu Tyr Ala Leu
 305 310 315 320
 Ala Glu Asp Ile Ala Ala Ile Leu Ile Ser Asp Lys His Ser Ala His
 325 330 335
 Phe Phe Glu Leu Ala Ala Gln Glu Cys Lys Asn Tyr Arg Ala Leu Ser
 340 345 350
 Asn Trp Leu Thr Val Glu Phe Ala Gly Arg Cys Lys Leu Lys Gly Lys
 355 360 365
 Asn Leu Ala Phe Ser Gly Ile Leu Pro Ser Ser Val Ala Gln Leu Val
 370 375 380
 Asn Phe Ile Asp Gln Gly Val Ile Thr Gly Lys Ile Ala Lys Asp Ile
 385 390 395 400
 Ala Asp Met Met Met Glu Ser Pro Glu Lys Ser Pro Glu Thr Ile Leu
 405 410 415
 Lys Glu Asn Pro Glu Met Leu Pro Met Thr Asp Glu Ser Ala Leu Val
 420 425 430
 Ala Ile Ile Ser Glu Val Ile Thr Ala Asn Pro Gln Ser Val Val Asp
 435 440 445
 Tyr Lys Ser Gly Lys Thr Lys Ala Leu Gly Phe Leu Val Gly Gln Ile
 450 455 460
 Met Lys Arg Thr Gln Gly Lys Ala Pro Pro Asn Arg Val Asn Glu Leu
 465 470 475 480
 Leu Leu Val Glu Leu Ser Lys
 485

<210> SEQ ID NO 20

<211> LENGTH: 379

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 20

```

tatttagtgt cgaaaacaa cgccaacatt tacgcagggt ctctctatta tcagcatatc      60
tcctattgga gcgcttggca gaatctgcta caaaacacta tcggtgcaga agctccgtta      120
gtccttaacg cacagttaac ttattgtcat gcttcaaacg acatgaaaac caacatgacg      180
actacttacg ctctctgtaa aacaacgtat gcagaaatca aggggtgattg gggtaacgat      240
tgtttcggag tcgagcttgg tgcaactgtg cctatccaaa cagaatcttc tctoctattc      300

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gatatgtact caccttttct gaagtttcaa cttgtgcata cgcaccaaga tgactttaag 360

gaaaacaata gcgatcagg 379

<210> SEQ ID NO 21

<211> LENGTH: 126

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 21

Tyr Leu Val Ser Lys Asn Asn Ala Asn Ile Tyr Ala Gly Ser Leu Tyr
1 5 10 15

Tyr Gln His Ile Ser Tyr Trp Ser Ala Trp Gln Asn Leu Leu Gln Asn
20 25 30

Thr Ile Gly Ala Glu Ala Pro Leu Val Leu Asn Ala Gln Leu Thr Tyr
35 40 45

Cys His Ala Ser Asn Asp Met Lys Thr Asn Met Thr Thr Thr Tyr Ala
50 55 60

Pro Arg Lys Thr Thr Tyr Ala Glu Ile Lys Gly Asp Trp Gly Asn Asp
65 70 75 80

Cys Phe Gly Val Glu Leu Gly Ala Thr Val Pro Ile Gln Thr Glu Ser
85 90 95

Ser Leu Leu Phe Asp Met Tyr Ser Pro Phe Leu Lys Phe Gln Leu Val
100 105 110

His Thr His Gln Asp Asp Phe Lys Glu Asn Asn Ser Asp Gln
115 120 125

<210> SEQ ID NO 22

<211> LENGTH: 2520

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 22

atgaaacatc cagtctactg gttcttaata tcctcgagcc tatttgctc gaattctttg 60

agcttcgcta acgacgetca aacagcctta actccctccg atagctataa tggaaatgtg 120

acctctgagg agttccaggt aaaagaaact tcatcaggaa caacgtatac ttgtgaaggc 180

aatgtgtgta tctcctttgc agggaaagat tcaggtctaa agaaaagttg tttctcagct 240

actgataacc ttaccttctt aggaaacggg tatactcttt gctttgataa tattactact 300

acagctagta accccggagc cattaatgtt caaggtcaag gaaaaacctt aggcactctca 360

ggattttctt tattttcatg tgcttattgt cctccaggca caactgggta cggagctata 420

cagactaaag gcaacacaac tttaaaagat aactctagtc ttgtcttcca taaaaactgc 480

tcaacagcag aaggtggggc tatccagtgt aaaggaagca gtgatgctga attaaaaata 540

gaaaataatc agaatctggt tttctcagaa aactcctcca cttcaaaagg cggggctatt 600

tatgctgata aactcaccat tgtctcaggt gggcctacat tattttctaa caactctgta 660

tccaacgggt catcccttaa aggcggagct attagcataa aagattcaag tggatgaatgt 720

agcctaaccg ctgatctcgg agatattacc ttcgatggga acaaaatcat caaaactagt 780

ggtggaagtt ctacagtaac aagaaattcc atagatctcg gcacagggaa atttaciaaag 840

ctacgtgcta aagacggcct cggaattttc ttctatgacc ctattactgg gggaggatct 900

gatgaactaa acattaataa aaaagaaact gttgattata caggaaagat cgtcttctca 960

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ggtgaaaaat tatccgatga agaaaaagca cgagcggaaa acctagcttc tactttcaac 1020
caacccatca cattatcagc aggatctctt gtacttaaag atgggtgtatc tgtaaccgca 1080
aaacaagtaa cgcaggaagc gggatctacc gttgtcatgg atctagggac cacattacag 1140
acgccttctt caggtggaga aaccatcacc ctaactaatc tagatattaa catcgccctcg 1200
ttgggggggg ggggggggtac ctctcctgct aaactcgcaa caaatacagc aagtcaagct 1260
ataactatta acgctgtcaa tctagtcgat gctgatggca atgcttatga agatcctatt 1320
cttgctactg ctaaaccctt cacagcaata gtactacaa ctaacgctag tacagtcaca 1380
cagcctacag ataactctaa aaattatgtc cctcctactc attacgggta ccaaggaaat 1440
tggacagtaa ctgggacac cgaaacagct acaaaaacag ccactctaac ttgggaacaa 1500
actggctact cccctaacc agaacgtcaa ggaccttag tccgaatac tctttgggg 1560
gcattctctg acctcagagc tataaaaaac ttaatggata ttagcgtcaa tggcgctgac 1620
taccatagag gtttttgggt atccgggtcta gctaacttct tacacaaaag tggctctgat 1680
actaaacgca agttccgtca caatagcgcc ggatacgctt taggcgtcta cgcaaaaact 1740
ccttctgatg atattttcag tgcggcttct tgccaactct tcggaaagga caaagactat 1800
ttagtgtcga aaaacaacgc caacatttac gcagggttct tctattatca gcatatctcc 1860
tattggagcg ctgggcagaa tctgtctaaa aacactatcg gtgcagaagc tccgttagtc 1920
cttaacgcac agttaactta ttgtcatgct tcaaacgaca tgaaaaccaa catgacgact 1980
acttacgctc ctgcgtaaaac aacgtatgca gaaatcaagg gtgattgggg taacgattgt 2040
ttcggagtcg agcttggtgc aactgtgcct atccaaacag aatcttctct cctattcgat 2100
atgtactcac ctttcctgaa gtttcaactt gtgcatacgc accaagatga ctttaaggaa 2160
aacaatagcg atcagggaag atacttcgaa agcagcaatc tcaccaacct ttctctgcct 2220
atcggcatca agtttgagag atttgctaac aacgatacag cttcttatca tgcactgct 2280
gcttattctc ctgatatcgt aagaagtaac cctgactgta ctacttctct gttagtaagc 2340
cccgactctg ctgtctgggt aacgaaagcc aacaaccttg cggaagcgc cttcatgcta 2400
caagcaggaa actactgtgc tttaagtcac aacatagaaa tcttcagcca gtcgggtttc 2460
gagctcaggg gatcttcacg aacctataac gtatgtctcg gatcgaagat ccagttctaa 2520

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<210> SEQ ID NO 23

<211> LENGTH: 839

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 23

```

Met Lys His Pro Val Tyr Trp Phe Leu Ile Ser Ser Ser Leu Phe Ala
1           5           10           15

```

```

Ser Asn Ser Leu Ser Phe Ala Asn Asp Ala Gln Thr Ala Leu Thr Pro
20           25           30

```

```

Ser Asp Ser Tyr Asn Gly Asn Val Thr Ser Glu Glu Phe Gln Val Lys
35           40           45

```

```

Glu Thr Ser Ser Gly Thr Thr Tyr Thr Cys Glu Gly Asn Val Cys Ile
50           55           60

```

```

Ser Phe Ala Gly Lys Asp Ser Gly Leu Lys Lys Ser Cys Phe Ser Ala
65           70           75           80

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Thr Asp Asn Leu Thr Phe Leu Gly Asn Gly Tyr Thr Leu Cys Phe Asp

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85					90					95					
Asn	Ile	Thr	Thr	Thr	Ala	Ser	Asn	Pro	Gly	Ala	Ile	Asn	Val	Gln	Gly
			100					105					110		
Gln	Gly	Lys	Thr	Leu	Gly	Ile	Ser	Gly	Phe	Ser	Leu	Phe	Ser	Cys	Ala
		115					120					125			
Tyr	Cys	Pro	Pro	Gly	Thr	Thr	Gly	Tyr	Gly	Ala	Ile	Gln	Thr	Lys	Gly
	130						135					140			
Asn	Thr	Thr	Leu	Lys	Asp	Asn	Ser	Ser	Leu	Val	Phe	His	Lys	Asn	Cys
	145			150							155				160
Ser	Thr	Ala	Glu	Gly	Gly	Ala	Ile	Gln	Cys	Lys	Gly	Ser	Ser	Asp	Ala
			165						170					175	
Glu	Leu	Lys	Ile	Glu	Asn	Asn	Gln	Asn	Leu	Val	Phe	Ser	Glu	Asn	Ser
		180						185					190		
Ser	Thr	Ser	Lys	Gly	Gly	Ala	Ile	Tyr	Ala	Asp	Lys	Leu	Thr	Ile	Val
		195					200					205			
Ser	Gly	Gly	Pro	Thr	Leu	Phe	Ser	Asn	Asn	Ser	Val	Ser	Asn	Gly	Ser
	210						215				220				
Ser	Pro	Lys	Gly	Gly	Ala	Ile	Ser	Ile	Lys	Asp	Ser	Ser	Gly	Glu	Cys
	225					230					235				240
Ser	Leu	Thr	Ala	Asp	Leu	Gly	Asp	Ile	Thr	Phe	Asp	Gly	Asn	Lys	Ile
			245						250					255	
Ile	Lys	Thr	Ser	Gly	Gly	Ser	Ser	Thr	Val	Thr	Arg	Asn	Ser	Ile	Asp
		260						265					270		
Leu	Gly	Thr	Gly	Lys	Phe	Thr	Lys	Leu	Arg	Ala	Lys	Asp	Gly	Phe	Gly
		275					280					285			
Ile	Phe	Phe	Tyr	Asp	Pro	Ile	Thr	Gly	Gly	Gly	Ser	Asp	Glu	Leu	Asn
	290						295				300				
Ile	Asn	Lys	Lys	Glu	Thr	Val	Asp	Tyr	Thr	Gly	Lys	Ile	Val	Phe	Ser
	305			310							315				320
Gly	Glu	Lys	Leu	Ser	Asp	Glu	Glu	Lys	Ala	Arg	Ala	Glu	Asn	Leu	Ala
			325						330					335	
Ser	Thr	Phe	Asn	Gln	Pro	Ile	Thr	Leu	Ser	Ala	Gly	Ser	Leu	Val	Leu
		340						345					350		
Lys	Asp	Gly	Val	Ser	Val	Thr	Ala	Lys	Gln	Val	Thr	Gln	Glu	Ala	Gly
		355					360					365			
Ser	Thr	Val	Val	Met	Asp	Leu	Gly	Thr	Thr	Leu	Gln	Thr	Pro	Ser	Ser
	370						375				380				
Gly	Gly	Glu	Thr	Ile	Thr	Leu	Thr	Asn	Leu	Asp	Ile	Asn	Ile	Ala	Ser
	385			390					395						400
Leu	Gly	Gly	Gly	Gly	Gly	Thr	Ser	Pro	Ala	Lys	Leu	Ala	Thr	Asn	Thr
			405						410					415	
Ala	Ser	Gln	Ala	Ile	Thr	Ile	Asn	Ala	Val	Asn	Leu	Val	Asp	Ala	Asp
			420					425					430		
Gly	Asn	Ala	Tyr	Glu	Asp	Pro	Ile	Leu	Ala	Thr	Ser	Lys	Pro	Phe	Thr
		435					440					445			
Ala	Ile	Val	Ala	Thr	Thr	Asn	Ala	Ser	Thr	Val	Thr	Gln	Pro	Thr	Asp
	450						455					460			
Asn	Leu	Thr	Asn	Tyr	Val	Pro	Pro	Thr	His	Tyr	Gly	Tyr	Gln	Gly	Asn
	465			470							475				480
Trp	Thr	Val	Thr	Trp	Asp	Thr	Glu	Thr	Ala	Thr	Lys	Thr	Ala	Thr	Leu
			485						490					495	

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Thr Trp Glu Gln Thr Gly Tyr Ser Pro Asn Pro Glu Arg Gln Gly Pro
 500 505 510
 Leu Val Pro Asn Thr Leu Trp Gly Ala Phe Ser Asp Leu Arg Ala Ile
 515 520 525
 Gln Asn Leu Met Asp Ile Ser Val Asn Gly Ala Asp Tyr His Arg Gly
 530 535 540
 Phe Trp Val Ser Gly Leu Ala Asn Phe Leu His Lys Ser Gly Ser Asp
 545 550 555 560
 Thr Lys Arg Lys Phe Arg His Asn Ser Ala Gly Tyr Ala Leu Gly Val
 565 570 575
 Tyr Ala Lys Thr Pro Ser Asp Asp Ile Phe Ser Ala Ala Phe Cys Gln
 580 585 590
 Leu Phe Gly Lys Asp Lys Asp Tyr Leu Val Ser Lys Asn Asn Ala Asn
 595 600 605
 Ile Tyr Ala Gly Ser Leu Tyr Tyr Gln His Ile Ser Tyr Trp Ser Ala
 610 615 620
 Trp Gln Asn Leu Leu Gln Asn Thr Ile Gly Ala Glu Ala Pro Leu Val
 625 630 635 640
 Leu Asn Ala Gln Leu Thr Tyr Cys His Ala Ser Asn Asp Met Lys Thr
 645 650 655
 Asn Met Thr Thr Thr Tyr Ala Pro Arg Lys Thr Thr Tyr Ala Glu Ile
 660 665 670
 Lys Gly Asp Trp Gly Asn Asp Cys Phe Gly Val Glu Leu Gly Ala Thr
 675 680 685
 Val Pro Ile Gln Thr Glu Ser Ser Leu Leu Phe Asp Met Tyr Ser Pro
 690 695 700
 Phe Leu Lys Phe Gln Leu Val His Thr His Gln Asp Asp Phe Lys Glu
 705 710 715 720
 Asn Asn Ser Asp Gln Gly Arg Tyr Phe Glu Ser Ser Asn Leu Thr Asn
 725 730 735
 Leu Ser Leu Pro Ile Gly Ile Lys Phe Glu Arg Phe Ala Asn Asn Asp
 740 745 750
 Thr Ala Ser Tyr His Val Thr Ala Ala Tyr Ser Pro Asp Ile Val Arg
 755 760 765
 Ser Asn Pro Asp Cys Thr Thr Ser Leu Leu Val Ser Pro Asp Ser Ala
 770 775 780
 Val Trp Val Thr Lys Ala Asn Asn Leu Ala Arg Ser Ala Phe Met Leu
 785 790 795 800
 Gln Ala Gly Asn Tyr Leu Ser Leu Ser His Asn Ile Glu Ile Phe Ser
 805 810 815
 Gln Phe Gly Phe Glu Leu Arg Gly Ser Ser Arg Thr Tyr Asn Val Asp
 820 825 830
 Leu Gly Ser Lys Ile Gln Phe
 835

<210> SEQ ID NO 24

<211> LENGTH: 1039

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 24

aaacgttttc atattaatgg ggttcctgaa tggcttttat ctacgcctta ttctcttgct

60

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atgggggtata atatcttggc tacgggagtg cagatgggta aagcctatgc cattcttgcc 120
aacgggtggtt atgatgtgcg ccctaccttg ataaaaaaaa tagtcactac ttctggaaaa 180
gagtacgtgt tgcacctca agttcgtgga gaaagaattc tttctcagga cattgtggat 240
gagggtattga aagctacgcg ttttactacc tatcctggag gaacgggatt tcgggctgcg 300
cctaaaaaagc attccagtgc agggaaaaca ggaacaacag aaaagctagt tcatggaaag 360
tatgataagc atcggcataat ttcttcattt atagggtatca cgccgatata cccttcggca 420
ggggggagtg ttcttttggt catgcttgtc tctatcagtt atacgaccga caacggtagt 480
caagtgtacg tcgttcaatt gcgacatgag ggtatcgaaa tctgtcgtca attcgtccat 540
gttaacctaa ttgtgtggtc attatcgctt tctttatact acttaccgta gttcctacgg 600
atactagcaa aaagtcttgc tctttgcgtt gctctttgaa cagcatactg tactttttaa 660
aagtctgcta aattttcccg ttctccattc ctatctgaga agtagagaag ggctctatct 720
aacacttctt ctccagaaga cacccaattg accatcttac gggcaacgga ctcggtgtct 780
tcttcttttt tggtttgtaa gttttgttgc gtatgctcag ctatatcatt cagatcacca 840
ttgattaaat caatgatcac actgacagct tcaaaatggt cttgcgatag tttattttga 900
tcttgttgta gagtggattg tgcaccccat aaacgctctt ttagattggt tatttgcctc 960
ttcagctctt ccgaatctaa cgcctcttcc agttcaggat cgataatggt agagtttctg 1020
tcttgcatca tcgccatag 1039

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<210> SEQ ID NO 25

<211> LENGTH: 196

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 25

```

Lys Arg Phe His Ile Asn Gly Val Pro Glu Trp Ser Leu Ser Thr Pro
1           5           10           15
Tyr Ser Leu Ala Met Gly Tyr Asn Ile Leu Ala Thr Gly Val Gln Met
20        25        30
Val Lys Ala Tyr Ala Ile Leu Ala Asn Gly Gly Tyr Asp Val Arg Pro
35        40        45
Thr Leu Ile Lys Lys Ile Val Thr Thr Ser Gly Lys Glu Tyr Val Leu
50        55        60
His Pro Gln Val Arg Gly Glu Arg Ile Leu Ser Gln Asp Ile Val Asp
65        70        75        80
Glu Val Leu Lys Ala Thr Arg Phe Thr Thr Tyr Pro Gly Gly Thr Gly
85        90        95
Phe Arg Ala Ala Pro Lys Lys His Ser Ser Ala Gly Lys Thr Gly Thr
100       105       110
Thr Glu Lys Leu Val His Gly Lys Tyr Asp Lys His Arg His Ile Ser
115       120       125
Ser Phe Ile Gly Ile Thr Pro Ile Tyr Pro Ser Ala Gly Gly Ser Val
130       135       140
Pro Leu Val Met Leu Val Ser Ile Ser Tyr Thr Thr Asp Asn Gly Ser
145       150       155       160
Gln Val Tyr Val Val Gln Leu Arg His Glu Gly Ile Glu Ile Cys Arg
165       170       175

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Gln Phe Val His Val Asn Leu Ile Val Trp Ser Leu Ser Leu Ser Leu
 180 185 190

Tyr Tyr Leu Pro
 195

<210> SEQ ID NO 26

<211> LENGTH: 1950

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 26

```

atgaatcacc gtaaatgctt aaccatgatt acctatggag ttctgctctc ctattctttc    60
ctgatcatatc ggtattataa aattcagatt tgtgaggaga aacgttgggc agcagaagct    120
ttaggacaac atgaatttcg agtaaaggac ccttttcgta gggggacgtt tttttctcag    180
atgaatttac gtaagggaga ttcagagcaa cgacaagctc tggccgtgga cattacgaaa    240
tttcatcttt gtttagatgc tgtagctgtt cctgaagaac accgtgatgt gattgctaag    300
aaagttttta gtctcattgg agaagggtgat tatgacaaac tccgtgcgga gtttgataaa    360
aaatctcgct atcgaaagtt atttctttgg ttagatcgtg cggatcatga ccgcatectg    420
tcttggtggc gggggtagcgc agcaaatctt aaaataccct cgaatgcttt gtttttcatg    480
accgactatc aaagatctta tccctttggc aaacttttag gccaatgtct acatactctg    540
agagaagtca aggatgagaa aacaggcaaa gctttcccta caggagggtt agaagcctat    600
tttaaccacg tccttgaagg agagccagga gaacggaaat tcctacgttc tcctttaaat    660
cgtttagatc tagataaagt cacaagattt cctagggatg gttcggatat ttatctcaca    720
gtcaatccct gtatacagac tatagcggaa gaggaattag aaaaaggggt aaaggaagcc    780
aaagctaaag gtgggcgtct aattttaatg aatgcttata caggcgagat tcttgcttta    840
gcacagtatc ctttctttaa tccttcggaa tacaaggaat ttttcaatga taaggaaaaa    900
atagagcaca caaaagtaac atcagtcagt gatgtgtttg aaccggctc tatcatgaaa    960
cctctgactc tggctatagc gttgctggcc aacgaagaga tggtgaaaag atcaggaaaag   1020
cccttatttg atcctaataa acctatagat gtaacccgca ggattttccc aggaagaaaag   1080
caatttcgcg ttaaggatat ctcacgaatc cggcgtttaa atatgtacat ggcgattcaa   1140
aagtcttcga acgtttatgt agcgcaactt gctgatctta tagtgcaaca tctagggaac   1200
cactggtatg aagacaagtt attgttatta ggatttggtt aaaagacggg gatagaattg   1260
ccaggggaag cgtcaggatt ggtaccttca cctaaacggt ttcataatga tggggttcct   1320
gaatggtctt tatctacgcc ttattctctt gctatggggt ataatatctt ggctacggga   1380
gtgcagatgg ttaaagccta tgccattctt gccaacggtg gttatgatgt gcgccctacc   1440
ttgataaaaa aaatagtcac tacttctgga aaagagtacg tgttgcatcc tcaagttcgt   1500
ggagaaagaa ttctttctca ggacattgtg gatgaggtat tgaaagctac gcgttttact   1560
acctatcctg gaggaacggg atttcgggct gcgcctaaaa agcattccag tgcaggaaa   1620
acaggaacaa cagaaaagct agttcatgga aagtatgata agcatcgga tatttcttca   1680
tttataggtt tcacgccgat atacccttcg gcagggggga gtgttccttt ggtcatgctt   1740
gtctctatag atgatcctga tcattgtgtt cgcgaggatg gaacaaagaa ctatatggga   1800
ggccgatgtg ccgccctgt atttggcaga gttgcggatc gtgttttatc ttatctagga   1860

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gttccccgaag ataaagaaaa atacagttat cagagtgagg tggctgctat gaaagctttg 1920

tatgaggaat ggaatcgttc ggggaaataa 1950

<210> SEQ ID NO 27

<211> LENGTH: 649

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 27

Met Asn His Arg Lys Cys Leu Thr Met Ile Thr Tyr Gly Val Leu Leu
1 5 10 15

Ser Tyr Ser Phe Leu Ile Ile Arg Tyr Tyr Lys Ile Gln Ile Cys Glu
20 25 30

Glu Lys Arg Trp Ala Ala Glu Ala Leu Gly Gln His Glu Phe Arg Val
35 40 45

Lys Asp Pro Phe Arg Arg Gly Thr Phe Phe Ser Gln Met Asn Leu Arg
50 55 60

Lys Gly Asp Ser Glu Gln Arg Gln Ala Leu Ala Val Asp Ile Thr Lys
65 70 75 80

Phe His Leu Cys Leu Asp Ala Val Ala Val Pro Glu Glu His Arg Asp
85 90 95

Val Ile Ala Lys Lys Val Phe Ser Leu Ile Gly Glu Gly Asp Tyr Asp
100 105 110

Lys Leu Arg Ala Glu Phe Asp Lys Lys Ser Arg Tyr Arg Lys Leu Phe
115 120 125

Leu Trp Leu Asp Arg Ala Asp His Asp Arg Ile Leu Ser Trp Trp Arg
130 135 140

Gly Tyr Ala Ala Lys Ser Lys Ile Pro Ser Asn Ala Leu Phe Phe Met
145 150 155 160

Thr Asp Tyr Gln Arg Ser Tyr Pro Phe Gly Lys Leu Leu Gly Gln Val
165 170 175

Leu His Thr Leu Arg Glu Val Lys Asp Glu Lys Thr Gly Lys Ala Phe
180 185 190

Pro Thr Gly Gly Leu Glu Ala Tyr Phe Asn His Val Leu Glu Gly Glu
195 200 205

Pro Gly Glu Arg Lys Phe Leu Arg Ser Pro Leu Asn Arg Leu Asp Leu
210 215 220

Asp Lys Val Thr Lys Ile Pro Arg Asp Gly Ser Asp Ile Tyr Leu Thr
225 230 235 240

Val Asn Pro Cys Ile Gln Thr Ile Ala Glu Glu Glu Leu Glu Lys Gly
245 250 255

Val Lys Glu Ala Lys Ala Lys Gly Gly Arg Leu Ile Leu Met Asn Ala
260 265 270

Tyr Thr Gly Glu Ile Leu Ala Leu Ala Gln Tyr Pro Phe Phe Asn Pro
275 280 285

Ser Glu Tyr Lys Glu Phe Phe Asn Asp Lys Glu Lys Ile Glu His Thr
290 295 300

Lys Val Thr Ser Val Ser Asp Val Phe Glu Pro Gly Ser Ile Met Lys
305 310 315 320

Pro Leu Thr Leu Ala Ile Ala Leu Leu Ala Asn Glu Glu Met Val Lys
325 330 335

Arg Ser Gly Lys Pro Leu Phe Asp Pro Asn Glu Pro Ile Asp Val Thr

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340					345					350					
Arg	Arg	Ile	Phe	Pro	Gly	Arg	Lys	Gln	Phe	Pro	Leu	Lys	Asp	Ile	Ser
		355					360					365			
Ser	Asn	Arg	Arg	Leu	Asn	Met	Tyr	Met	Ala	Ile	Gln	Lys	Ser	Ser	Asn
	370					375					380				
Val	Tyr	Val	Ala	Gln	Leu	Ala	Asp	Leu	Ile	Val	Gln	His	Leu	Gly	Asn
	385					390					395				400
His	Trp	Tyr	Glu	Asp	Lys	Leu	Leu	Leu	Gly	Phe	Gly	Lys	Lys	Thr	
			405						410					415	
Gly	Ile	Glu	Leu	Pro	Gly	Glu	Ala	Ser	Gly	Leu	Val	Pro	Ser	Pro	Lys
			420					425					430		
Arg	Phe	His	Ile	Asn	Gly	Val	Pro	Glu	Trp	Ser	Leu	Ser	Thr	Pro	Tyr
		435					440					445			
Ser	Leu	Ala	Met	Gly	Tyr	Asn	Ile	Leu	Ala	Thr	Gly	Val	Gln	Met	Val
	450					455					460				
Lys	Ala	Tyr	Ala	Ile	Leu	Ala	Asn	Gly	Gly	Tyr	Asp	Val	Arg	Pro	Thr
	465					470					475				480
Leu	Ile	Lys	Lys	Ile	Val	Thr	Thr	Ser	Gly	Lys	Glu	Tyr	Val	Leu	His
				485					490					495	
Pro	Gln	Val	Arg	Gly	Glu	Arg	Ile	Leu	Ser	Gln	Asp	Ile	Val	Asp	Glu
			500					505					510		
Val	Leu	Lys	Ala	Thr	Arg	Phe	Thr	Thr	Tyr	Pro	Gly	Gly	Thr	Gly	Phe
		515					520					525			
Arg	Ala	Ala	Pro	Lys	Lys	His	Ser	Ser	Ala	Gly	Lys	Thr	Gly	Thr	Thr
		530				535					540				
Glu	Lys	Leu	Val	His	Gly	Lys	Tyr	Asp	Lys	His	Arg	His	Ile	Ser	Ser
	545					550					555				560
Phe	Ile	Gly	Ile	Thr	Pro	Ile	Tyr	Pro	Ser	Ala	Gly	Gly	Ser	Val	Pro
			565					570						575	
Leu	Val	Met	Leu	Val	Ser	Ile	Asp	Asp	Pro	Asp	His	Cys	Val	Arg	Glu
			580					585					590		
Asp	Gly	Thr	Lys	Asn	Tyr	Met	Gly	Gly	Arg	Cys	Ala	Ala	Pro	Val	Phe
		595					600					605			
Gly	Arg	Val	Ala	Asp	Arg	Val	Leu	Ser	Tyr	Leu	Gly	Val	Pro	Glu	Asp
		610					615					620			
Lys	Glu	Lys	Tyr	Ser	Tyr	Gln	Ser	Glu	Val	Ala	Ala	Met	Lys	Ala	Leu
	625					630					635				640
Tyr	Glu	Glu	Trp	Asn	Arg	Ser	Gly	Lys							
				645											

<210> SEQ ID NO 28

<211> LENGTH: 960

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 28

atgttcaata agctcattga aacagcacag aaacgggtgg aagcaagaaa ctatactatt	60
cgaaagcata ctcttgagta tgacgatgtt atgaataggc aaaggcagac gatctatgct	120
tttcgtaatg acgttatccg ctctgaagat atctttgggt tagctaagga agcaatatct	180
catgttgcat taatgatcgc ttcgttgata gtgagccgtg atcatcctac agggaattct	240
cttcctaggc tggaagaatg gatgaactat tctttccac tgcaattgaa tattgaagaa	300

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ttgaaaagat tgaagtctat agatgccatt gccgaacggg ttgctgatga tctcatagaa    360
gttttccaga ataagtttgc ttctatggtg caggaaatta ccgaagcagc cggagaaaaa    420
gtcgatgcta atggtgtctg taaagatgtt attcgctcgg tcatgattat gcatatcgat    480
gagcagtgga aaattcatct tgtagatatg gatttattac gtagtgaagt aggtttacgt    540
actgtcggtc agaaagaccc tcttatcgaa tttaaacaatg agtcgttctt actattcgaa    600
agtcttattc gcgatattcg tattgctatt gtaaagcatt tgttccgttt agagttgacg    660
atgactagag aacagcggcc tcaaaatgtc gtgcctgttg ttgccacatc ttccaaaaat    720
aatgaaaatt tcggtccttt ggaactcaca gttatcagtg attctgacga tgaataaaaa    780
gagcttagg gctgggctag cttccagcct ttcccttac gttattgatt tatagtttta    840
aataaatacg gaccactcag accaggattg tgtgtcgtgg tggcgatcc aaaatgttct    900
gtgattatcc tcaatcagaa attgtacatg atgatcgcg ttgcgtgttg tcatgcaaat    960

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<210> SEQ ID NO 29

<211> LENGTH: 258

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 29

```

Met Phe Asn Lys Leu Ile Glu Thr Ala Gln Lys Arg Val Glu Ala Arg
 1           5           10          15
Asn Tyr Thr Ile Arg Lys His Thr Leu Glu Tyr Asp Asp Val Met Asn
 20          25          30
Arg Gln Arg Gln Thr Ile Tyr Ala Phe Arg Asn Asp Val Ile Arg Ser
 35          40          45
Glu Asp Ile Phe Gly Leu Ala Lys Glu Ala Ile Ser His Val Ala Leu
 50          55          60
Met Ile Ala Ser Leu Ile Val Ser Arg Asp His Pro Thr Gly Asn Ser
 65          70          75          80
Leu Pro Arg Leu Glu Trp Met Asn Tyr Ser Phe Pro Leu Gln Leu
 85          90          95
Asn Ile Glu Glu Leu Lys Arg Leu Lys Ser Ile Asp Ala Ile Ala Glu
100         105         110
Arg Val Ala Asp Asp Leu Ile Glu Val Phe Gln Asn Lys Phe Ala Ser
115         120         125
Met Val Gln Glu Ile Thr Glu Ala Ala Gly Glu Lys Val Asp Ala Asn
130         135         140
Gly Val Cys Lys Asp Val Ile Arg Ser Val Met Ile Met His Ile Asp
145         150         155         160
Glu Gln Trp Lys Ile His Leu Val Asp Met Asp Leu Leu Arg Ser Glu
165         170         175
Val Gly Leu Arg Thr Val Gly Gln Lys Asp Pro Leu Ile Glu Phe Lys
180         185         190
His Glu Ser Phe Leu Leu Phe Glu Ser Leu Ile Arg Asp Ile Arg Ile
195         200         205
Ala Ile Val Lys His Leu Phe Arg Leu Glu Leu Thr Met Thr Arg Glu
210         215         220
Gln Arg Pro Gln Asn Val Val Pro Val Val Ala Thr Ser Phe Gln Asn
225         230         235         240

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Asp Glu

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<210> SEQ ID NO 30
<211> LENGTH: 697
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci
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<400> SEQUENCE: 30

gggtttgatt	atctcagaga	taattctatt	gcaacttctg	tggatgagca	ggtgggacgt	60
gggttttatt	ttgctattat	cgatgaagtc	gactcgattt	taattgatga	agccagaact	120
cctttaatta	tttctggctc	tggggaaaaa	cataatcctg	tgtatttcga	actcaaagat	180
aaagtggctg	acctcgttca	gttacaaagg	gagttatgta	accagttagc	tcttgaagct	240
agacggggac	tagaattggt	cttggatatg	gatattcttc	ctaaggataa	aaaagttatc	300
gaagctatct	ccgaattttg	ccgtagctta	tggttagtta	gtaagggaat	gcctttaaat	360
cgtgttttgc	gtagagtgcg	cgaacaccca	gatttgcgag	ccatgataga	taaatgggat	420
acttattatc	atgctgagca	aaataaaaga	gagagtatat	agaagctatc	tcagctgtgat	480
atcattgttg	atgaacataa	taacgatttt	gaattgacag	atcgtggcat	gcaacaatgg	540
gtggataaag	ctggagggtc	tgctgaagat	tttgtcatga	tggacatggg	gcatgaatat	600
gctcttatag	atggtgacga	taccttatca	ccgacagaga	aatcaatag	aaaaatagct	660
atttccgaag	aagatacgaq	gaqaaaaqct	cgaqctc			697

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<210> SEQ ID NO 31
<211> LENGTH: 232
<212> TYPE: PRT
<213> ORGANISM: Chlamydia psittaci
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<400> SEQUENCE: 31

Gly 1	Phe	Asp	Tyr	Leu 5	Arg	Asp	Asn	Ser	Ile 10	Ala	Thr	Ser	Val	Asp 15	Glu
Gln	Val	Gly	Arg 20	Gly	Phe	Tyr	Phe	Ala 25	Ile	Ile	Asp	Glu	Val	Asp 30	Ser
Ile	Leu	Ile	Asp 35	Glu	Ala	Arg	Thr 40	Pro	Leu	Ile	Ile	Ser 45	Gly	Pro	Gly
Glu	Lys 50	His	Asn	Pro	Val	Tyr 55	Phe	Glu	Leu	Lys	Asp 60	Lys	Val	Ala	Asp
Leu 65	Val	Gln	Leu	Gln	Arg 70	Glu	Leu	Cys	Asn	Gln 75	Leu	Ala	Leu	Glu	Ala 80
Arg	Arg	Gly	Leu	Glu 85	Leu	Phe	Leu	Asp	Met 90	Asp	Ile	Leu	Pro	Lys 95	Asp
Lys	Lys	Val	Ile 100	Glu	Ala	Ile	Ser	Glu 105	Phe	Cys	Arg	Ser	Leu 110	Trp	Leu
Val	Ser	Lys 115	Gly	Met	Pro	Leu	Asn 120	Arg	Val	Leu	Arg	Arg 125	Val	Arg	Glu
His 130	Pro	Asp	Leu	Arg	Ala 135	Met	Ile	Asp	Lys	Trp	Asp 140	Thr	Tyr	Tyr	His
Ala 145	Glu	Gln	Asn	Lys	Glu 150	Glu	Ser	Ile	Glu	Lys 155	Leu	Ser	Gln	Leu	Tyr 160
Ile	Ile	Val	Asp	Glu	His	Asn	Asn	Asp	Phe	Glu	Leu	Thr	Asp	Arg	Gly

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	165							170					175					
Met	Gln	Gln	Trp	Val	Asp	Lys	Ala	Gly	Gly	Ser	Ala	Glu	Asp	Phe	Val			
			180					185					190					
Met	Met	Asp	Met	Gly	His	Glu	Tyr	Ala	Leu	Ile	Asp	Gly	Asp	Asp	Thr			
		195					200					205						
Leu	Ser	Pro	Thr	Glu	Lys	Ile	Asn	Arg	Lys	Ile	Ala	Ile	Ser	Glu	Glu			
		210				215					220							
Asp	Thr	Arg	Arg	Lys	Ala	Arg	Ala											
225					230													
<210> SEQ ID NO 32																		
<211> LENGTH: 2910																		
<212> TYPE: DNA																		
<213> ORGANISM: Chlamydia psittaci																		
<400> SEQUENCE: 32																		
atgttagatt	ttcttaa	acg	tttcttt	gga	tcttctca	ag	agcg	cac	ctt	aaaaaaaa	attt				60			
caaaaacttg	tgataag	gt	caacctct	at	gatgagat	gc	tagctc	cttt	gtctgat	gag					120			
gagttacgta	ataaaac	agc	agagtta	aaa	aagcgt	tatc	aggacg	gcga	atccttag	at					180			
gatatgcttc	ccgagg	ccta	tgccgt	tagt	aaaaat	gtat	gcaggc	gttt	aacagga	act					240			
cctgtagaag	tgtcggg	gta	tcatcaa	aat	tgggac	atg	ttccct	atga	tgtgcag	gtt					300			
ctcggtgcta	tagctat	gca	taaggg	cttt	ataacc	gaga	tgca	gacagg	agagggg	aaa					360			
actctcaccg	ctgttat	gcc	tctatat	ttta	aatgc	attga	caggca	agcc	tgtgc	attta					420			
gtcacagtga	atgattat	ct	cgctcaa	aag	gattgt	gagt	gggtcg	gctc	tatat	tgct					480			
tggttaggtt	taactacc	gg	agtatt	gata	tcagg	atcgc	ctttag	aaaa	aagaaa	agac					540			
atttatcg	ttgtgac	gtt	ctacgg	taca	gcac	agagt	tcggg	tttga	ttatct	caga					600			
gataattcta	ttgcaact	tc	tgtggat	gag	caggtg	ggac	gtggg	tttta	ttttg	ctatt					660			
atcgatgaag	tcgactcg	at	tttaatt	gat	gaagcc	agaa	ctcctt	ta	tatttc	tggt					720			
cctggggaaa	aacataat	cc	tgtgtatt	tc	gaactc	aaag	ataa	ag	tg	gcctc	gtt				780			
cagttacaaa	gggagtt	atg	taaccag	tta	gctctt	gaag	ctagac	gggg	actaga	aattg					840			
ttcttg	gata	tattct	tccta	aggat	aaaaa	agtta	tcga	agctat	ctccga	attt					900			
tgccgtag	ct	tatggttag	tagta	aggga	atgc	ctttta	atcgt	gtttt	gcgtag	agt					960			
cg	gaacacc	cagatt	tg	cg	agccat	gata	gataa	atggg	atactt	atta	tc	atg	ctg	gag	1020			
caaaaata	aaag	aagag	agtat	agaga	agcta	tctc	agctgt	atatc	attgt	tgat	ga	acat			1080			
aataac	gatt	ttga	attgac	agat	cgtggc	atgc	aaca	at	gggtg	gataa	ggctg	gaggt			1140			
tctg	ctgaag	attttg	tc	at	gtgacat	g	ggc	atga	at	gtctt	at	agatg	tg	gac	1200			
gatac	cttat	caccg	acaga	gaaa	atca	at	agaaaa	atag	ctattt	ccga	aga	agata	c		1260			
aggag	aaaag	ctc	gagct	ca	tggtt	gcgc	caact	attaa	gagc	gcatct	tctt	atg	gaa		1320			
cg	catg	ttg	attat	attgt	tcg	ta	atgat	caaat	ttgt	ca	tcatt	gacga	acata	cgggc	1380			
cg	cccg	caac	caggt	cg	tc	gt	gt	ccga	ggact	gc	atc	aagcc	ataga	agca	aaagaa	1440		
cat	gtc	acta	tc	cg	taagga	atc	acaaa	c	ttg	ctacag	ttac	ctt	taca	gaattt	cttc	1500		
cg	t	ctg	t	atg	ta	g	acg	ac	aa	acg	aa	ttac	gga	atc	taa	agagttt	1560	
aa	agag	at	ttt	ata	atctt	ta	tg	attg	ca	g	tgccc	ac	gt	ttaa	aga	aatg	tttgcgtgta	1620

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gatcacaatg acgaatttta tatgacagag cgtgaaaagt accacgcgat tgtaaaggaa 1680
attgcccgtg tacatgccgt agggaaacccg attctcatag gaacggagtc ttagagggtt 1740
tctgagaaac tttctcgat tttgagacaa aatcgcatag aacatacagt gttaaagtcg 1800
aaaaatcatg ctcaagaagc agagatcatt gcagcagcag gaaagctggg agctgtgact 1860
gtagctacca atatggctgg ccgtggtaca gatattaagc tggatgaaga agctgtagtt 1920
gttgagggtc tccatgttat tggtagcagt cggcaccaat cagcccgat agataggcag 1980
ttgcgcgggc gttgcgcacg tttaggagat cctgggttcg cgaaattttt cctatctttt 2040
gaagatcgcc tgatgcgctt atttgcacg cccaagttaa atgccttgat tcgtcatttc 2100
cgtcctcctg aaggagaggc tatgtcggat cctatgttca ataagctcat tgaacagca 2160
cagaaacggg tggaaagca aaactatact attcgaaagc atactcttga gtatgacgat 2220
gttatgaata ggcaaaggca gacgatctat gcttttcgta atgacgttat ccgctctgaa 2280
gatatctttg gtttagctaa ggaagcaata tctcatgttg cattaatgat cgcttcgttg 2340
atagtgaagc gtgatcatcc tacagggaat tctcttccta ggctggaaga atggatgaac 2400
tattctttcc cactgcaatt gaatattgaa gaattgaaaa gattgaagtc tatagatgcc 2460
attgccgaac gggttgtctg tgatctcata gaagttttcc agaataagtt tgcttctatg 2520
gtgcaggaaa ttaccgaagc agccggagaa aaagtcgatg ctaatggtgt ctgtaaagat 2580
gttattcgct cggtcatgat tatgcatac gatgagcagt ggaatttca tctttagat 2640
atggatttat tacgtagtga agtaggttta cgtactgtcg gtcagaaaga cctcttatac 2700
gaatttaaac atgagtcgtt ctactatttc gaaagtctta ttcgcgatat tcgtattgct 2760
attgtaaagc atttgttccg tttagagttg acgatgacta gagaacagcg gcctcaaaat 2820
gtcgtgcctg ttgttgccac atctttccaa aataatgaaa atttcggtcc tttggaactc 2880
acagttatca gtgattctga cgatgaataa 2910

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<210> SEQ ID NO 33

<211> LENGTH: 969

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 33

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Met  Leu  Asp  Phe  Leu  Lys  Arg  Phe  Phe  Gly  Ser  Ser  Gln  Glu  Arg  Thr
  1      5      10      15
Leu  Lys  Lys  Phe  Gln  Lys  Leu  Val  Asp  Lys  Val  Asn  Leu  Tyr  Asp  Glu
      20      25      30
Met  Leu  Ala  Pro  Leu  Ser  Asp  Glu  Glu  Leu  Arg  Asn  Lys  Thr  Ala  Glu
      35      40      45
Leu  Lys  Lys  Arg  Tyr  Gln  Asp  Gly  Glu  Ser  Leu  Asp  Asp  Met  Leu  Pro
      50      55      60
Glu  Ala  Tyr  Ala  Val  Val  Lys  Asn  Val  Cys  Arg  Arg  Leu  Thr  Gly  Thr
      65      70      75      80
Pro  Val  Glu  Val  Ser  Gly  Tyr  His  Gln  Asn  Trp  Asp  Met  Val  Pro  Tyr
      85      90      95
Asp  Val  Gln  Val  Leu  Gly  Ala  Ile  Ala  Met  His  Lys  Gly  Phe  Ile  Thr
      100     105     110
Glu  Met  Gln  Thr  Gly  Glu  Gly  Lys  Thr  Leu  Thr  Ala  Val  Met  Pro  Leu
      115     120     125

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Tyr	Leu	Asn	Ala	Leu	Thr	Gly	Lys	Pro	Val	His	Leu	Val	Thr	Val	Asn
130						135					140				
Asp	Tyr	Leu	Ala	Gln	Arg	Asp	Cys	Glu	Trp	Val	Gly	Ser	Ile	Leu	Arg
145					150					155					160
Trp	Leu	Gly	Leu	Thr	Thr	Gly	Val	Leu	Ile	Ser	Gly	Ser	Pro	Leu	Glu
			165					170						175	
Lys	Arg	Lys	Asp	Ile	Tyr	Arg	Cys	Asp	Val	Val	Tyr	Gly	Thr	Ala	Ser
			180					185					190		
Glu	Phe	Gly	Phe	Asp	Tyr	Leu	Arg	Asp	Asn	Ser	Ile	Ala	Thr	Ser	Val
	195					200						205			
Asp	Glu	Gln	Val	Gly	Arg	Gly	Phe	Tyr	Phe	Ala	Ile	Ile	Asp	Glu	Val
210					215					220					
Asp	Ser	Ile	Leu	Ile	Asp	Glu	Ala	Arg	Thr	Pro	Leu	Ile	Ile	Ser	Gly
225					230					235					240
Pro	Gly	Glu	Lys	His	Asn	Pro	Val	Tyr	Phe	Glu	Leu	Lys	Asp	Lys	Val
			245					250						255	
Ala	Asp	Leu	Val	Gln	Leu	Gln	Arg	Glu	Leu	Cys	Asn	Gln	Leu	Ala	Leu
		260						265					270		
Glu	Ala	Arg	Arg	Gly	Leu	Glu	Leu	Phe	Leu	Asp	Met	Asp	Ile	Leu	Pro
	275					280					285				
Lys	Asp	Lys	Lys	Val	Ile	Glu	Ala	Ile	Ser	Glu	Phe	Cys	Arg	Ser	Leu
290					295						300				
Trp	Leu	Val	Ser	Lys	Gly	Met	Pro	Leu	Asn	Arg	Val	Leu	Arg	Arg	Val
305					310					315					320
Arg	Glu	His	Pro	Asp	Leu	Arg	Ala	Met	Ile	Asp	Lys	Trp	Asp	Thr	Tyr
			325					330						335	
Tyr	His	Ala	Glu	Gln	Asn	Lys	Glu	Glu	Ser	Ile	Glu	Lys	Leu	Ser	Gln
		340					345						350		
Leu	Tyr	Ile	Ile	Val	Asp	Glu	His	Asn	Asn	Asp	Phe	Glu	Leu	Thr	Asp
	355					360					365				
Arg	Gly	Met	Gln	Gln	Trp	Val	Asp	Lys	Ala	Gly	Gly	Ser	Ala	Glu	Asp
	370					375					380				
Phe	Val	Met	Met	Asp	Met	Gly	His	Glu	Tyr	Ala	Leu	Ile	Asp	Gly	Asp
385				390						395				400	
Asp	Thr	Leu	Ser	Pro	Thr	Glu	Lys	Ile	Asn	Arg	Lys	Ile	Ala	Ile	Ser
			405					410					415		
Glu	Glu	Asp	Thr	Arg	Arg	Lys	Ala	Arg	Ala	His	Gly	Leu	Arg	Gln	Leu
		420					425					430			
Leu	Arg	Ala	His	Leu	Leu	Met	Glu	Arg	Asp	Val	Asp	Tyr	Ile	Val	Arg
	435					440					445				
Asn	Asp	Gln	Ile	Val	Ile	Ile	Asp	Glu	His	Thr	Gly	Arg	Pro	Gln	Pro
	450				455						460				
Gly	Arg	Arg	Phe	Ser	Glu	Gly	Leu	His	Gln	Ala	Ile	Glu	Ala	Lys	Glu
465				470						475				480	
His	Val	Thr	Ile	Arg	Lys	Glu	Ser	Gln	Thr	Phe	Ala	Thr	Val	Thr	Leu
			485					490					495		
Gln	Asn	Phe	Phe	Arg	Leu	Tyr	Glu	Lys	Leu	Ala	Gly	Met	Thr	Gly	Thr
		500					505					510			
Ala	Ile	Thr	Glu	Ser	Lys	Glu	Phe	Lys	Glu	Ile	Tyr	Asn	Leu	Tyr	Val
	515					520					525				
Leu	Gln	Val	Pro	Thr	Phe	Lys	Glu	Cys	Leu	Arg	Val	Asp	His	Asn	Asp

[illegible]

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Val Ala Thr Ser Phe Gln Asn Asn Glu Asn Phe Gly Pro Leu Glu Leu
945 950 955 960

Thr Val Ile Ser Asp Ser Asp Asp Glu
965

<210> SEQ ID NO 34
<211> LENGTH: 577
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 34

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gttgatgctg cagttattcc agggaaacttc gccattgcag ggggaatctg tccgtataaa    60
aacagtctat acctagaaga tgtccgtact tcccaataca ccaatgtcgt tgcatacgt    120
gctgaagata tggaagactc gagaatgcat aaactaaaac agctattgca aagcagttct    180
gtgcaggatt tctttaatac gaaatataag gggatctttt tatcgtagta acacatctgg    240
atggcttagg gaagagttga gccaccccggt tctccgtagg tttaaggcat attgggaaac    300
gattttcttg aattttttga aaaactttga ctgtttttct tttgattatt cgaagcagat    360
gtatgtcgag tatggcggtt ttagggccca gaggtccttt cagttctcct ttacatggt    420
ctctataccc aaccaccta aaaatgcact tgctagggtc cattcctata gttggcatat    480
acattggagc gaagcgata gccgcggtg ctcaatatca tagaatgtgt agagcgaata    540
caggagtgtc tcaggtgatt attcaggatt caggatt                                577
```

<210> SEQ ID NO 35
<211> LENGTH: 76
<212> TYPE: PRT
<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 35

Val Asp Ala Ala Val Ile Pro Gly Asn Phe Ala Ile Ala Gly Gly Ile
1 5 10 15
Cys Pro Tyr Lys Asn Ser Leu Tyr Leu Glu Asp Val Arg Thr Ser Gln
20 25 30
Tyr Thr Asn Val Val Val Ile Arg Ala Glu Asp Met Glu Asp Ser Arg
35 40 45
Met His Lys Leu Lys Gln Leu Leu Gln Ser Ser Ser Val Gln Asp Phe
50 55 60
Phe Asn Thr Lys Tyr Lys Gly Ile Phe Leu Ser Gln
65 70 75

<210> SEQ ID NO 36
<211> LENGTH: 804
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 36

```
atgaaaaaaa tcacaatact ctcgttactt gcttagcca tctctttaac aggttggtgc    60
aagaattcag aaggagtctt gcggattgcg gcgagtccca cgccacatgc agagcttctt    120
tatagtttag aaaaggaggc tcaatocctt ggattgcaat tgaaaatact tcccatagat    180
gattaccgtg tacctaaccg ttgctttta gataagcaaa tagaggcaaa ttatttccaa    240
catgaagatt tcttaaaaga tgagtgtgct cggtagcaat gcgaaggaaa acttgcgatt    300
```

-continued

```

ttggctaagg tacatttaga acctatgggt ttatatattcta ataaaaccca gtctctcgaa 360
gagcttaaag tcaaggaaca gctacgtata gcggttccta tagatagaac aaacgaacaa 420
cgtgcgctag acttatttgcg agactgcaat ttgattagtt acaagaagc ttctcatcta 480
gatatcaccg caaaagatgt ctttggttgt ggagggaaaa aggtaacgat tatagagatg 540
gcagcacctt tattagtatc ttctttacca gacgttgatg ctgcagttat tccagggaac 600
ttcgccattg cagggggaat ctgtccgtat aaaaacagtc tatacctaga agatgtccgt 660
acttcccaat acaccaatgt cgttgtcata cgtgctgaag atatggaaga ctcgagaatg 720
cataaactaa aacagctatt gcaaagcagt tctgtgcagg atttctttaa tacgaaatat 780
aaggggatct ttttatcgca gtaa 804

```

<210> SEQ ID NO 37

<211> LENGTH: 267

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 37

```

Met  Lys Lys Ile Thr Ile Leu Ser Leu Leu Ala Leu Ala Ile Ser Leu
   1              5              10              15

Thr  Gly Cys Cys Lys Asn Ser Glu Gly Val Leu Arg Ile Ala Ala Ser
      20              25              30

Pro  Thr Pro His Ala Glu Leu Leu Tyr Ser Leu Glu Lys Glu Ala Gln
      35              40              45

Ser  Leu Gly Leu Gln Leu Lys Ile Leu Pro Ile Asp Asp Tyr Arg Val
      50              55              60

Pro  Asn Arg Leu Leu Leu Asp Lys Gln Ile Glu Ala Asn Tyr Phe Gln
      65              70              75              80

His  Glu Asp Phe Leu Lys Asp Glu Cys Ala Arg Tyr Gln Cys Glu Gly
      85              90              95

Lys  Leu Ala Ile Leu Ala Lys Val His Leu Glu Pro Met Gly Leu Tyr
      100             105             110

Ser  Asn Lys Thr Gln Ser Leu Glu Glu Leu Lys Val Lys Glu Gln Leu
      115             120             125

Arg  Ile Ala Val Pro Ile Asp Arg Thr Asn Glu Gln Arg Ala Leu Asp
      130             135             140

Leu  Leu Arg Asp Cys Asn Leu Ile Ser Tyr Lys Glu Ala Ser His Leu
      145             150             155             160

Asp  Ile Thr Ala Lys Asp Val Phe Gly Cys Gly Gly Lys Lys Val Thr
      165             170             175

Ile  Ile Glu Met Ala Ala Pro Leu Leu Val Ser Ser Leu Pro Asp Val
      180             185             190

Asp  Ala Ala Val Ile Pro Gly Asn Phe Ala Ile Ala Gly Gly Ile Cys
      195             200             205

Pro  Tyr Lys Asn Ser Leu Tyr Leu Glu Asp Val Arg Thr Ser Gln Tyr
      210             215             220

Thr  Asn Val Val Val Ile Arg Ala Glu Asp Met Glu Asp Ser Arg Met
      225             230             235             240

His  Lys Leu Lys Gln Leu Leu Gln Ser Ser Ser Val Gln Asp Phe Phe
      245             250             255

Asn Thr Lys Tyr Lys Gly Ile Phe Leu Ser Gln
      260             265

```

-continued

<210> SEQ ID NO 38
 <211> LENGTH: 402
 <212> TYPE: DNA
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 38

```
catgtatattt acgcaaaaaa taaacggtat aactcctgct tacaagccgc gctataccac    60
aataatatcc cgacaaccgt gtacacaaac cttattgata tcgtgaagaa aaattcttca    120
ctaatacagc agtacttttc catcaacaa cgatgcttaa atctaaaaga tttccatttt    180
tatgatgttt atgctccctt aagtcagtcc aaagagaaaa aatatacgtt ccaagaagct    240
gtggatctta tctatactag cctttctcct cttagaacgg aatacatgta taccttaaaa    300
cagggggtta caactcaagg ctgggtagat aaatacgaat atcttaataa acgctccgga    360
gcctattctt cgggatgtta cgatagccac ccttatgtcc tc                        402
```

<210> SEQ ID NO 39
 <211> LENGTH: 134
 <212> TYPE: PRT
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 39

```
His Val Phe Tyr Ala Lys Asn Lys Arg Tyr Asn Ser Cys Leu Gln Ala
 1      5      10      15
Ala Leu Tyr His Asn Asn Ile Pro Thr Thr Val Tyr Thr Asn Leu Ile
 20      25      30
Asp Ile Val Lys Lys Asn Ser Ser Leu Ile Thr Lys Tyr Phe Ser Ile
 35      40      45
Lys Gln Arg Cys Leu Asn Leu Lys Asp Phe His Phe Tyr Asp Val Tyr
 50      55      60
Ala Pro Leu Ser Gln Ser Lys Glu Lys Lys Tyr Thr Phe Gln Glu Ala
 65      70      75      80
Val Asp Leu Ile Tyr Thr Ser Leu Ser Pro Leu Gly Thr Glu Tyr Ile
 85      90      95
Asp Thr Leu Lys Gln Gly Leu Thr Thr Gln Gly Trp Val Asp Lys Tyr
100     105     110
Glu Asn Leu Asn Lys Arg Ser Gly Ala Tyr Ser Ser Gly Cys Tyr Asp
115     120     125
Ser His Pro Tyr Val Leu
130
```

<210> SEQ ID NO 40
 <211> LENGTH: 1827
 <212> TYPE: DNA
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 40

```
atgagcgtag aattcaacaa gcaacaagtc cgtccaagaa gtgaaatttc ccctcaagat    60
tgttgggata tcacccctt atactctaat agaaaagcat ggaaagcaga tcttgattct    120
ttcgatttaa aaacagacgg ctcacctagc tggcccgtc ttcaagcaac gcaataccaa    180
ctggacaact cagaatctct actatcctta ttaactactc tcttctctat tgagagaaaa    240
ttaaacaaac tctacgttta cgctcatctg actcatgatc aggatattac aaatcaagaa    300
```

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```

ggcatcgag atcttaaattc tatcacgcat ctacatacct tatttgccga agaaacctct 360
tggtgacaac ccgctttaac cagcctatcg gaatctctca ttgctcagca cctatcagct 420
ccctgtttag ctcccttatag attctattta gagaaaatct ttagactatc tatacacaca 480
ggcactcctg gagaagaaaa aattctcgct tccgccttta ctctcttga agtagccagt 540
aaggcatttt ctctcttaag tgactctgaa attccctttg ggcaagctac agactcagaa 600
ggaaactctc acccgctttc tcatgcactg gcttcattgt atatgcaatc cacagatcgg 660
gaattacgaa aaacatccta cctagcaciaa tgtgaaagat atcatagtta ccgacatacc 720
tttgctaact tactcaatgg gaaaatccaa gcccatgtat tttagcaciaa aaataaacgg 780
tataactcct gttacaaagc cgcgtatata cacaataata tcccgacaac cgtgtacaca 840
aaccttattg atatcgtgaa gaaaaattct tcactaatca cgaagtactt ttccatcaaa 900
caacgatgct taaatctaaa agatttccat ttttatgatg tttatgctcc cctaagtcag 960
tccaaagaga aaaaatatac gttccaagaa gctgtggatc ttatctatac tagcctttct 1020
cctctaggaa cggaatacat tgatacctta aaacaggggt taacaactca aggctgggta 1080
gataaatacg aaaatcttaa taaacgctcc ggagcctatt cttagggatg ttacgatagc 1140
cacccttatg tcctcctaaa ctatacaggc accctgtatg atgtatccgt cattgcccac 1200
gaaggcgagc acagtatgca ctctgatttt agtaggaagc atcaaccttt ccatgacgct 1260
caatataccta ttttccttgc tgaaattgct totaccttaa atgaaatgct tcttatggat 1320
tccatgctga aggagagcga ctcaaaagaa gagaaaatca ccattctgac acgatgtttg 1380
gataccatct tctctacact attccgtcag gtattattcg cctcttttga atacgatatt 1440
catcacgcag cagaacatgg gggtcctcta actgaagaat acctatcctc aacttacaag 1500
aatttacaaa atgagtttta cggagaaatt atcacatttg atgtcctgtc gaacatagaa 1560
tggtgcaagaa ttctctatct ctattacaat ttctacgtat accaatatgc aacgggcatt 1620
atagccgccc tgtgcttttt agaaaaaatt cttacaacg aagataacgc tcttaactcc 1680
tatctcaact ttttaaaaag tgggtgatca gatttccctc tagaaatctt aaaaaaatca 1740
ggattagata tgggcacagt tgagccaatc caaaaagctt tttgctttat cgagaaaaaa 1800
atccaggagc tatcatcttt aatttga 1827

```

<210> SEQ ID NO 41

<211> LENGTH: 608

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 41

```

Met  Ser Val Glu Phe Asn Lys Gln Gln Val Arg Pro Arg Ser Glu Ile
  1      5      10      15

Ser  Pro Gln Asp Cys Trp Asp Ile Thr Pro Leu Tyr Leu Asn Arg Lys
    20      25      30

Ala  Trp Lys Ala Asp Leu Asp Ser Phe Gly Leu Lys Thr Asp Gly Ser
    35      40      45

Pro  Thr Trp Pro Ala Leu Gln Ala Thr Gln Tyr Gln Leu Asp Asn Ser
    50      55      60

Glu  Ser Leu Leu Ser Leu Leu Thr Thr Leu Phe Ser Ile Glu Arg Lys
    65      70      75      80

Leu  Asn Lys Leu Tyr Val Tyr Ala His Leu Thr His Asp Gln Asp Ile

```

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	85					90					95				
Thr	Asn	Gln	Glu	Gly	Ile	Ala	Asp	Leu	Lys	Ser	Ile	Thr	His	Leu	His
			100					105					110		
Thr	Leu	Phe	Ala	Glu	Glu	Thr	Ser	Trp	Val	Gln	Pro	Ala	Leu	Thr	Ser
	115						120					125			
Leu	Ser	Glu	Ser	Leu	Ile	Ala	Gln	His	Leu	Ser	Ala	Pro	Cys	Leu	Ala
	130					135					140				
Pro	Tyr	Arg	Phe	Tyr	Leu	Glu	Lys	Ile	Phe	Arg	Leu	Ser	Ile	His	Thr
145					150					155					160
Gly	Thr	Pro	Gly	Glu	Glu	Lys	Ile	Leu	Ala	Ser	Ala	Phe	Thr	Pro	Leu
			165					170						175	
Glu	Val	Ala	Ser	Lys	Ala	Phe	Ser	Ser	Leu	Ser	Asp	Ser	Glu	Ile	Pro
			180					185					190		
Phe	Gly	Gln	Ala	Thr	Asp	Ser	Glu	Gly	Asn	Ser	His	Pro	Leu	Ser	His
	195						200					205			
Ala	Leu	Ala	Ser	Leu	Tyr	Met	Gln	Ser	Thr	Asp	Arg	Glu	Leu	Arg	Lys
	210					215					220				
Thr	Ser	Tyr	Leu	Ala	Gln	Cys	Glu	Arg	Tyr	His	Ser	Tyr	Arg	His	Thr
225					230					235					240
Phe	Ala	Asn	Leu	Leu	Asn	Gly	Lys	Ile	Gln	Ala	His	Val	Phe	Tyr	Ala
			245						250					255	
Lys	Asn	Lys	Arg	Tyr	Asn	Ser	Cys	Leu	Gln	Ala	Ala	Leu	Tyr	His	Asn
			260					265					270		
Asn	Ile	Pro	Thr	Thr	Val	Tyr	Thr	Asn	Leu	Ile	Asp	Ile	Val	Lys	Lys
		275					280					285			
Asn	Ser	Ser	Leu	Ile	Thr	Lys	Tyr	Phe	Ser	Ile	Lys	Gln	Arg	Cys	Leu
	290					295					300				
Asn	Leu	Lys	Asp	Phe	His	Phe	Tyr	Asp	Val	Tyr	Ala	Pro	Leu	Ser	Gln
305					310					315					320
Ser	Lys	Glu	Lys	Lys	Tyr	Thr	Phe	Gln	Glu	Ala	Val	Asp	Leu	Ile	Tyr
			325						330					335	
Thr	Ser	Leu	Ser	Pro	Leu	Gly	Thr	Glu	Tyr	Ile	Asp	Thr	Leu	Lys	Gln
		340					345						350		
Gly	Leu	Thr	Thr	Gln	Gly	Trp	Val	Asp	Lys	Tyr	Glu	Asn	Leu	Asn	Lys
	355					360						365			
Arg	Ser	Gly	Ala	Tyr	Ser	Ser	Gly	Cys	Tyr	Asp	Ser	His	Pro	Tyr	Val
	370					375					380				
Leu	Leu	Asn	Tyr	Thr	Gly	Thr	Leu	Tyr	Asp	Val	Ser	Val	Ile	Ala	His
385					390					395					400
Glu	Gly	Gly	His	Ser	Met	His	Ser	Tyr	Phe	Ser	Arg	Lys	His	Gln	Pro
			405						410					415	
Phe	His	Asp	Ala	Gln	Tyr	Pro	Ile	Phe	Leu	Ala	Glu	Ile	Ala	Ser	Thr
		420						425					430		
Leu	Asn	Glu	Met	Leu	Leu	Met	Asp	Ser	Met	Leu	Lys	Glu	Ser	Asp	Ser
	435						440					445			
Lys	Glu	Glu	Lys	Ile	Thr	Ile	Leu	Thr	Arg	Cys	Leu	Asp	Thr	Ile	Phe
	450					455					460				
Ser	Thr	Leu	Phe	Arg	Gln	Val	Leu	Phe	Ala	Ser	Phe	Glu	Tyr	Asp	Ile
465					470					475					480
His	His	Ala	Ala	Glu	His	Gly	Val	Pro	Leu	Thr	Glu	Glu	Tyr	Leu	Ser
			485					490						495	

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Ser Thr Tyr Lys Asn Leu Gln Asn Glu Phe Tyr Gly Glu Ile Ile Thr
500 505 510

Phe Asp Val Leu Ser Asn Ile Glu Trp Ala Arg Ile Pro His Phe Tyr
515 520 525

Tyr Asn Phe Tyr Val Tyr Gln Tyr Ala Thr Gly Ile Ile Ala Ala Leu
530 535 540

Cys Phe Leu Glu Lys Ile Leu Asn Asn Glu Asp Asn Ala Leu Asn Ser
545 550 555 560

Tyr Leu Asn Phe Leu Lys Ser Gly Gly Ser Asp Phe Pro Leu Glu Ile
565 570 575

Leu Lys Lys Ser Gly Leu Asp Met Gly Thr Val Glu Pro Ile Gln Lys
580 585 590

Ala Phe Cys Phe Ile Glu Lys Lys Ile Gln Glu Leu Ser Ser Leu Ile
595 600 605

<210> SEQ ID NO 42

<211> LENGTH: 1517

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 42

```

gcgtaggatt cggaagagct gaaagagcaa ataaacaatc taaaagagcg tttatgggat      60
gcacaatcca ctctacaaca agatcaaaaat aaactatcgc aagaacattt tgaagctgtc    120
agtgtgatca ttgatttaat caatgggtgat ctgaatgata tagctgagca tacgcaacaa    180
aacttacaaa ccaaaaaaga agaagaacac gaggccgttg cccgtaagat ggtcaattgg    240
gtgtcttctg gagaagaagt gttaaataga gcccttctct acttctcaga taggaatgga    300
gaacgggaaa atttagcaga ctttttaaaa gtacagtatg ctgttcaaag agcaacgcaa    360
agagcagaac tttttgctag tatcgtagga actacggtaa gtagtataaa gacgataatg    420
accacacaat taggttaaca tggacgaatt gacgacagat ttcgataccc tcatgtcgca    480
attgaacgac gtacacttga ctaccgttgt cggtcgtata actgaagtcg tcggtatggt    540
aattaaagct gtcgttccca atgtacgcgt tggggaggtg tgcttagtta aacgttatgg    600
tatggagccg ctcgtagccg aagtcgctcg cttcacacaa aatttcgctt ttttatcgcc    660
actaggagaa ctactggag tcagcccttc ttcagagggt attcccacag gctcgccttt    720
gtatatccgt gcaggtaacg gtccttttag tcgtgtattg aatggtcttg gagaacctat    780
cgactccgag atcaaaggac ctttggttga tgttaacgaa acctaccctg tgtttcgcgc    840
tccaccagat ccattgcata gagaaaaatt aagaacaatt ttatccaccg gcgtgcggtg    900
tatcgacggg atgctcacag tcgccagagg ccagcgtata ggcatTTTTG ctggagctgg    960
ggtagggtaaa tcgtctctct tgggaatgat cgctagaaac gcggaagaag ccgatgtcaa   1020
tgtgattgct ctcacgag agcggggccg agaggttcgt gaatttatcg agggcgatct   1080
cggagaagaa ggaatgaaac gttcgggtgat cgtcgtctct acttcagatc aatcctcaca   1140
gttgcgatta aatgctgctt acgtaggcac cgctatagca gagtattttc gtgatcaggg   1200
caaaaccgta gttttgatga tggattctgt caccgattt gcccgagccc taagagaagt   1260
cgggtctagct gccggagaac cgccagctcg aggaggatac acaccttctg tattctcaac   1320
tttgcttagg ttattagaac gttccggagc ttcggataaa ggaacaatca cagcctttta   1380

```

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```

cacagtactt gttgcggggg atgatatgaa tgaaccggtc gctgatgaag ttaaactgat 1440
tcttgatggt cacgttgtct tgtctaacgc tttagctcag gcataccatt atcctgctat 1500
tgatgtctta gcatcta 1517

```

```

<210> SEQ ID NO 43
<211> LENGTH: 145
<212> TYPE: PRT
<213> ORGANISM: Chlamydia psittaci

```

```

<400> SEQUENCE: 43

```

```

Ala  Leu Asp Ser Glu Glu Leu Lys Glu Gln Ile Asn Asn Leu Lys Glu
  1          5          10         15
Arg  Leu Trp Asp Ala Gln Ser Thr Leu Gln Gln Asp Gln Asn Lys Leu
      20          25          30
Ser  Gln Glu His Phe Glu Ala Val Ser Val Ile Ile Asp Leu Ile Asn
      35          40          45
Gly  Asp Leu Asn Asp Ile Ala Glu His Thr Gln Gln Asn Leu Gln Thr
      50          55          60
Lys  Lys Glu Glu Glu His Glu Ser Val Ala Arg Lys Met Val Asn Trp
      65          70          75          80
Val  Ser Ser Gly Glu Glu Val Leu Asn Arg Ala Leu Leu Tyr Phe Ser
      85          90          95
Asp  Arg Asn Gly Glu Arg Glu Asn Leu Ala Asp Phe Leu Lys Val Gln
      100         105         110
Tyr  Ala Val Gln Arg Ala Thr Gln Arg Ala Glu Leu Phe Ala Ser Ile
      115         120         125
Val  Gly Thr Thr Val Ser Ser Ile Lys Thr Ile Met Thr Thr Gln Leu
      130         135         140
Gly
145

```

```

<210> SEQ ID NO 44
<211> LENGTH: 669
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci

```

```

<400> SEQUENCE: 44

```

```

atggtagatc ctttgaagct tttcccaaag ctagactccg agaaagaaac agcttctata 60
cagaagcctt taggaactcc tttagccagt gagttacata aggaagttcc tgcattttct 120
ttagggacgg cagcagactc cttgaataaa aatatagagg atgtcaagcc taaccctatg 180
gcgatgatgc aagacagaaa ctctaacatt atcgatcctg aactggaaga ggcgttagat 240
tcggaagagc tgaagagaca aataaacaat ctaaaagagc gtttatggga tgcacaatcc 300
actctacaac aagatcaaaa taaactatcg caagaacatt ttgaagctgt cagtgtgatc 360
attgatttaa tcaatggtga tctgaatgat atagctgagc atacgcaaca aaacttacia 420
accaaaaaag aagaagaaca cgagtcggtt gcccgtaaga tggtaattg ggtgtcttct 480
ggagaagaag tgtaaatag agcccttctc tacttctcag ataggaatgg agaacgggaa 540
aatttagcag actttttaaa agtacagtat gctgttcaaa gagcaacgca aagagcagaa 600
ctttttgcta gtatcgtagg aactacggta agtagtataa agacgataat gaccacacaa 660
ttaggttaa 669

```

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<210> SEQ ID NO 45
 <211> LENGTH: 222
 <212> TYPE: PRT
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 45

```

Met  Val Asp Pro Leu Lys Leu Phe Pro Lys Leu Asp Ser Glu Lys Glu
  1              5              10              15

Thr  Ala Ser Ile Gln Lys Pro Leu Gly Thr Pro Leu Ala Ser Glu Leu
      20              25              30

His  Lys Glu Val Pro Ala Phe Ser Leu Gly Thr Ala Ala Asp Ser Leu
      35              40              45

Asn  Lys Asn Ile Glu Asp Val Lys Pro Asn Pro Met Ala Met Met Gln
      50              55              60

Asp  Arg Asn Ser Asn Ile Ile Asp Pro Glu Leu Glu Glu Ala Leu Asp
      65              70              75              80

Ser  Glu Glu Leu Lys Glu Gln Ile Asn Asn Leu Lys Glu Arg Leu Trp
      85              90              95

Asp  Ala Gln Ser Thr Leu Gln Gln Asp Gln Asn Lys Leu Ser Gln Glu
      100             105             110

His  Phe Glu Ala Val Ser Val Ile Ile Asp Leu Ile Asn Gly Asp Leu
      115             120             125

Asn  Asp Ile Ala Glu His Thr Gln Gln Asn Leu Gln Thr Lys Lys Glu
      130             135             140

Glu  Glu His Glu Ser Val Ala Arg Lys Met Val Asn Trp Val Ser Ser
      145             150             155             160

Gly  Glu Glu Val Leu Asn Arg Ala Leu Leu Tyr Phe Ser Asp Arg Asn
      165             170             175

Gly  Glu Arg Glu Asn Leu Ala Asp Phe Leu Lys Val Gln Tyr Ala Val
      180             185             190

Gln  Arg Ala Thr Gln Arg Ala Glu Leu Phe Ala Ser Ile Val Gly Thr
      195             200             205

Thr  Val Ser Ser Ile Lys Thr Ile Met Thr Thr Gln Leu Gly
      210             215             220

```

<210> SEQ ID NO 46
 <211> LENGTH: 1329
 <212> TYPE: DNA
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 46

```

atggacgaat tgacgacaga ttctgatacc ctcatgtcgc aattgaacga cgtacacttg      60
actaccgttg tcggtcgat aactgaagtc gtcggtatgt taattaaagc tgcgtttccc      120
aatgtacgcy ttggggagggt atgcttagtt aaacgttatg gtatggagcc gctcgtgacc      180
gaagtcgtcg gcttcacaca aaatttcgct tttttatcgc cactaggaga acttactgga      240
gtcagccctt cttcagaggt tattccaca ggtctgcctt tgtatatccg tgcagtaac      300
ggtcttttag gtcgtgtatt gaatggtctg ggagaacctt tcgactccga gatcaaagga      360
cctttggttg atgttaacga aacctaccct gtgtttcgcg ctccaccaga tccattgcat      420
agagaaaaat taagaacaat tttatccacc ggcgtgcggt gtatcgacgg tatgctcaca      480
gtcgccagag gccagcgat aggcattttt gctggagctg gggtggttaa atcgtctctc      540

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ttgggaatga tcgctagaaa cgcggaagaa gccgatgtca atgtgattgc tctcatcgga    600
gagcggggcc gagaggttcg tgaatttata gagggcgatc tcggagaaga aggaatgaaa    660
cgttcgggtga tcgtcgtctc tacttcagat caatcctcac agttgcgatt aaatgctgct    720
tacgtaggca ccgctatagc agagtatttt cgtgatcagg gcaaaaccgt agttttgatg    780
atggattctg tcacccgatt tgcccagacc ctaagagaag tcggtctagc tgccggagaa    840
ccgccagctc gaggaggata cacaccttct gtattctcaa ctttgcctag gttattagaa    900
cgttccggag cttcggataa aggaacaatc acagcctttt acacagtact tgttgccggg    960
gatgatatga atgaaccggt cgctgatgaa gttaaatacga ttcttgatgg tcacgttgtc   1020
ttgtctaacg ctttagctca ggcataccat tatcctgcta ttgatgtctt agcatctatc   1080
agccgattgc tgacagcaat tgttcctgag gaacaacgac gcatcatagg aaaagcccga   1140
gaggtgctgg caaaatacaa agcaaacgaa atgcttatac gtattggaga atatcgccga   1200
gggtccgatac gtgaagtgga ttttgctata gatcacattg ataaattgaa cagattctta   1260
aagcaagata ttcataaaaa aacaaattac gaggaagcct cgcaacagct tcgggctatt   1320
ttccgataa                                     1329

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<210> SEQ ID NO 47

<211> LENGTH: 442

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 47

```

Met  Asp Glu Leu Thr Thr Asp Phe Asp Thr Leu Met Ser Gln Leu Asn
  1      5      10      15
Asp  Val His Leu Thr Thr Val Val Gly Arg Ile Thr Glu Val Val Gly
      20      25      30
Met  Leu Ile Lys Ala Val Val Pro Asn Val Arg Val Gly Glu Val Cys
      35      40      45
Leu  Val Lys Arg Tyr Gly Met Glu Pro Leu Val Thr Glu Val Val Gly
      50      55      60
Phe  Thr Gln Asn Phe Ala Phe Leu Ser Pro Leu Gly Glu Leu Thr Gly
      65      70      75      80
Val  Ser Pro Ser Ser Glu Val Ile Pro Thr Gly Leu Pro Leu Tyr Ile
      85      90      95
Arg  Ala Gly Asn Gly Leu Leu Gly Arg Val Leu Asn Gly Leu Gly Glu
      100     105     110
Pro  Ile Asp Ser Glu Ile Lys Gly Pro Leu Val Asp Val Asn Glu Thr
      115     120     125
Tyr  Pro Val Phe Arg Ala Pro Pro Asp Pro Leu His Arg Glu Lys Leu
      130     135     140
Arg  Thr Ile Leu Ser Thr Gly Val Arg Cys Ile Asp Gly Met Leu Thr
      145     150     155     160
Val  Ala Arg Gly Gln Arg Ile Gly Ile Phe Ala Gly Ala Gly Val Gly
      165     170     175
Lys  Ser Ser Leu Leu Gly Met Ile Ala Arg Asn Ala Glu Glu Ala Asp
      180     185     190
Val  Asn Val Ile Ala Leu Ile Gly Glu Arg Gly Arg Glu Val Arg Glu
      195     200     205
Phe  Ile Glu Gly Asp Leu Gly Glu Glu Gly Met Lys Arg Ser Val Ile

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-continued

	210	215	220	
Val	Val Ser Thr Ser Asp Gln Ser Ser Gln Leu Arg Leu Asn Ala Ala			
225		230	235	240
Tyr	Val Gly Thr Ala Ile Ala Glu Tyr Phe Arg Asp Gln Gly Lys Thr			
	245	250	255	
Val	Val Leu Met Met Asp Ser Val Thr Arg Phe Ala Arg Ala Leu Arg			
	260	265	270	
Glu	Val Gly Leu Ala Ala Gly Glu Pro Pro Ala Arg Gly Gly Tyr Thr			
	275	280	285	
Pro	Ser Val Phe Ser Thr Leu Pro Arg Leu Leu Glu Arg Ser Gly Ala			
	290	295	300	
Ser	Asp Lys Gly Thr Ile Thr Ala Phe Tyr Thr Val Leu Val Ala Gly			
305		310	315	320
Asp	Asp Met Asn Glu Pro Val Ala Asp Glu Val Lys Ser Ile Leu Asp			
	325	330	335	
Gly	His Val Val Leu Ser Asn Ala Leu Ala Gln Ala Tyr His Tyr Pro			
	340	345	350	
Ala	Ile Asp Val Leu Ala Ser Ile Ser Arg Leu Leu Thr Ala Ile Val			
	355	360	365	
Pro	Glu Glu Gln Arg Arg Ile Ile Gly Lys Ala Arg Glu Val Leu Ala			
	370	375	380	
Lys	Tyr Lys Ala Asn Glu Met Leu Ile Arg Ile Gly Glu Tyr Arg Arg			
385		390	395	400
Gly	Ser Asp Arg Glu Val Asp Phe Ala Ile Asp His Ile Asp Lys Leu			
	405	410	415	
Asn	Arg Phe Leu Lys Gln Asp Ile His Glu Lys Thr Asn Tyr Glu Glu			
	420	425	430	
Ala	Ser Gln Gln Leu Arg Ala Ile Phe Arg			
	435	440		

<210> SEQ ID NO 48

<211> LENGTH: 477

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 48

```

cttcttgacg atgccgactc tgtcaacctt gcaactggat tcaacggctc cactagtga 60
actttcaatg ttaaacaac agataatgct gacgggacaa catatattct aggcagcgcg 120
atcacctttg aacacataaa tcaattaaaa ccagcaaaca ctactgtgtt tgctaataca 180
gctggagatc taacgtttac tgggaatcga cgacttctct atttcaataa tatttcatca 240
acagcgaaag gtgccgctat cagcacaact gcggatggta agacactcac aatatccggg 300
gctctacaac tgattttcta catgtcgcca agattggcca cgggaaatgg cgtcatttat 360
tctaatagct ctgtactcat cgagaacaat totcaaggta gctcgggact gaataagtct 420
gcagggaaag gcgtctttat ttgttgtagg aaaagtacgg atgtgggagc tacatca 477

```

<210> SEQ ID NO 49

<211> LENGTH: 159

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 49

-continued

Leu 1 Leu Ala Asp 5 Ala Asp Ser Val Asn 10 Leu Ala Thr Gly Phe Asn 15 Gly
 Ser Thr Ser Glu Thr Phe Asn Val Lys 25 Gln Thr Asp Asn 30 Ala Asp Gly
 Thr Thr Tyr Ile Leu Gly Ser 40 Ala Ile Thr Phe Glu His 45 Ile Asn Gln
 Leu Lys Pro Ala Asn Thr Ser Cys Phe 55 Ala Asn Thr 60 Ala Gly Asp Leu
 Thr 65 Phe Thr Gly Asn Arg Arg Leu Leu Tyr Phe 75 Asn Asn Ile Ser Ser
 Thr Ala Lys Gly Ala Ala Ile Ser Thr Thr 90 Ala Asp Gly Lys Thr 95 Leu
 Thr Ile Ser Gly Ala Leu Gln Leu Ile Phe Tyr Met Ser Pro Arg Leu 110
 Ala Thr Gly Asn Gly Val Ile Tyr Ser Asn Ser Ser Val Leu Ile Glu 125
 Asn Asn Ser Gln Gly Ser Ser Gly Leu Asn Lys Ser Ala Gly Lys Gly 140
 Val Phe Ile Cys Cys Glu Lys Ser Thr Asp Val Gly Ala Thr Ser 155

<210> SEQ ID NO 50
 <211> LENGTH: 591
 <212> TYPE: DNA
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 50

```

acctttgaac acataaatca attaaaacca gcaaactacta gctgttttgc taatacagct      60
ggagatctaa cgtttactgg gaatcgacga cttctctatt tcaataatat ttcatacaaca    120
gcgaaaggtg ccgctatcag cacaactgcg gatggtaaga cactcacaat atccgggggt      180
ctacaactga ttttctacat gtcgccaaga ttggccacgg gaaatggcgt catattattct    240
aatagctctg tactcatcga gaacaattct caaggtagct cgggactgaa taagtctgca     300
gggaaagcgg tctttatttg ttgtgagaaa agtacggatg tgggagctac atcaccgaca     360
ttaatcatac ggaataacgg agagtttctt actgtaggta atgcagctac tagctctgga     420
ggagcgattt atgcggagaa aatgatctta tcctcaggag gatatacaaa atttcaatcc     480
aatgttagct atgatcaagg tggggccatt gccattgctc ctaatggaga aattagtctc     540
tccgcggata aaggaaatat cgtctttgaa agaaacctta aaattgcca c                591
  
```

<210> SEQ ID NO 51
 <211> LENGTH: 197
 <212> TYPE: PRT
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 51

Thr 1 Phe Glu His Ile Asn Gln Leu Lys 10 Pro Ala Asn Thr Ser Cys Phe 15
 Ala Asn Thr Ala Gly Asp Leu Thr Phe Thr Gly Asn Arg Arg Leu Leu 30
 Tyr Phe Asn Asn Ile Ser Ser Thr Ala Lys Gly Ala Ala Ile Ser Thr 45
 Thr Ala Asp Gly Lys Thr Leu Thr Ile Ser Gly Ala Leu Gln Leu Ile

-continued

50					55					60					
Phe	Tyr	Met	Ser	Pro	Arg	Leu	Ala	Thr	Gly	Asn	Gly	Val	Ile	Tyr	Ser
65					70					75				80	
Asn	Ser	Ser	Val	Leu	Ile	Glu	Asn	Asn	Ser	Gln	Gly	Ser	Ser	Gly	Leu
			85						90					95	
Asn	Lys	Ser	Ala	Gly	Lys	Gly	Val	Phe	Ile	Cys	Cys	Glu	Lys	Ser	Thr
			100					105					110		
Asp	Val	Gly	Ala	Thr	Ser	Pro	Thr	Leu	Ile	Ile	Arg	Asn	Asn	Gly	Glu
		115					120					125			
Phe	Leu	Thr	Val	Gly	Asn	Ala	Ala	Thr	Ser	Ser	Gly	Gly	Ala	Ile	Tyr
	130					135					140				
Ala	Glu	Lys	Met	Ile	Leu	Ser	Ser	Gly	Gly	Tyr	Thr	Lys	Phe	Gln	Ser
145					150					155					160
Asn	Val	Ser	Tyr	Asp	Gln	Gly	Gly	Ala	Ile	Ala	Ile	Ala	Pro	Asn	Gly
			165					170					175		
Glu	Ile	Ser	Leu	Ser	Ala	Asp	Lys	Gly	Asn	Ile	Val	Phe	Glu	Arg	Asn
			180					185					190		
Leu	Lys	Ile	Ala	Asn											
			195												

<210> SEQ ID NO 52

<211> LENGTH: 2040

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 52

```

atgcagggaa tactaatgaa aaactctatt tatggggttt tactgttttc ctcttttgcc      60
ttatccactg ctaccaaact tcttgcagat gccgactctg tcaaccttgc aactggattc      120
aacggctcca ctagtgaaac tttcaatgtt aaacaaacag ataatgctga cgggacaaca      180
tatattctag gcagcgcgat cacctttgaa cacataaatc aattaaaacc agcaaact      240
agctgttttg ctaatacagc tggagatcta acgtttactg ggaatcgacg acttctctat      300
ttcaataata ttcatcaaac agcgaaaggt gccgctatca gcacaactgc ggatggtaag      360
aactcacao tatccggggc tctacaactg attttctaca tgcgcgaag attggccacg      420
ggaaatggcg tcatttattc taatagctct gtactcatcg agaacaattc tcaaggtagc      480
tcgggactga ataagtctgc agggaaaggc gtctttattt gttgtgagaa aagtacggat      540
gtgggagcta catcaccgac attaatcata cggaataacg gagagtttct tactgtaggt      600
aatgcagcta ctagctctgg aggagcgatt tatgcggaga aatgatctt atcctcagga      660
ggatatacaa aatttcaatc caatgttagc tatgatcaag gtggggccat tgccattgct      720
cctaattggag aaattagtct ctccgcggat aaaggaaata tcgtctttga aagaaacctt      780
aaaattgcca acaaacaaaa tactcccaat gccattcacc taggagacaa tgcgaaattt      840
cttcaattac gtgctgctaa caacaaagcc atattttttt atgaccgat tacaaccacg      900
ggatctgtgg cagatcggct aattattaat aactcgcaag gagaagcctc gacttacgat      960
ggggcgattg tattttctag tctcaactta ttactcatt cccctgaatg taaactctct      1020
tcattttctc aaggctctac tttagcggca ggatcattag ttttagaaga gggggtatgt      1080
gtacaagctc cgtcttttga tcaacgtgct cactcccaac tattcatgaa tcctgggacg      1140
aagttaacag ctaccagaa catctcggt aagaatctcc atctcaatct taatagaata      1200

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-continued

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gcagaagagc cggcgatat caccacaaca gacgatgctt ctagtgtgga catttgcgga 1260
cctgtagtta tgcatataga tgatgagatc ttctataatc agacagtatt agcaaatgag 1320
ttgtctgtag agtggtttaa tctcagttct ccacatctcg ataatacac tattgatgac 1380
gttcccgagc tgcctatcat gacgtagaa tcgcatcgtg gttatcaagg tacatgggaa 1440
atctcttgga aagagcaacc taaacttacc ttgggaagg cgactatcgc gcctaataag 1500
cagatgcacc ttatttgga accttctggt tacgttcctt tctcaggggg aactggagag 1560
tttacgacat ctttagtgcc taatagctta tggaatctct ttttagatac acgtttttct 1620
caacaagcga ttgagaaca tgctgtatct tcaggtaacg gtatatggat ttctccatg 1680
accaattctt ttctcaagg ttctacgaac aacaaccacg gctttcgtca taagagttca 1740
ggatataccg cagggggaaa aatacaaca cttcaagatg atatctttag tgtcagtttt 1800
tctcagctat ttgggagatc taaggatttt ggatctgcca catctaagga tacattccta 1860
tcgggctcta tctatgctca gcattcgaga cgcttacttc ctataatgag attccttgca 1920
ggaacatcaa catatagacc gcgactctta ctgagtattc ccaagaatct tcctatcaat 1980
tttgatgttc ttgtgagtta cagctatgac agtaaccaca tgaaagtaca aaaattctaa 2040

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<210> SEQ ID NO 53

<211> LENGTH: 679

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 53

```

Met  Gln Gly Ile Leu Met Lys Asn Ser  Ile Tyr Gly Val Leu Leu Phe
  1      5      10      15

Ser  Ser Phe Ala Leu Ser Thr Ala Thr Lys Leu Leu Ala Asp Ala Asp
    20      25      30

Ser  Val Asn Leu Ala Thr Gly Phe Asn Gly Ser Thr Ser Glu Thr Phe
    35      40      45

Asn  Val Lys Gln Thr Asp Asn Ala Asp Gly Thr Thr Tyr Ile Leu Gly
    50      55      60

Ser  Ala Ile Thr Phe Glu His Ile Asn Gln Leu Lys Pro Ala Asn Thr
    65      70      75      80

Ser  Cys Phe Ala Asn Thr Ala Gly Asp Leu Thr Phe Thr Gly Asn Arg
    85      90      95

Arg  Leu Leu Tyr Phe Asn Asn Ile Ser Ser Thr Ala Lys Gly Ala Ala
    100     105     110

Ile  Ser Thr Thr Ala Asp Gly Lys Thr Leu Thr Ile Ser Gly Ala Leu
    115     120     125

Gln  Leu Ile Phe Tyr Met Ser Pro Arg Leu Ala Thr Gly Asn Gly Val
    130     135     140

Ile  Tyr Ser Asn Ser Ser Val Leu Ile Glu Asn Asn Ser Gln Gly Ser
    145     150     155     160

Ser  Gly Leu Asn Lys Ser Ala Gly Lys Gly Val Phe Ile Cys Cys Glu
    165     170     175

Lys  Ser Thr Asp Val Gly Ala Thr Ser Pro Thr Leu Ile Ile Arg Asn
    180     185     190

Asn  Gly Glu Phe Leu Thr Val Gly Asn Ala Ala Thr Ser Ser Gly Gly
    195     200     205

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Ala	Ile	Tyr	Ala	Glu	Lys	Met	Ile	Leu	Ser	Ser	Gly	Gly	Tyr	Thr	Lys
210						215					220				
Phe	Gln	Ser	Asn	Val	Ser	Tyr	Asp	Gln	Gly	Gly	Ala	Ile	Ala	Ile	Ala
225						230					235				240
Pro	Asn	Gly	Glu	Ile	Ser	Leu	Ser	Ala	Asp	Lys	Gly	Asn	Ile	Val	Phe
			245						250					255	
Glu	Arg	Asn	Leu	Lys	Ile	Ala	Asn	Lys	Gln	Asn	Thr	Pro	Asn	Ala	Ile
			260					265					270		
His	Leu	Gly	Asp	Asn	Ala	Lys	Phe	Leu	Gln	Leu	Arg	Ala	Ala	Asn	Asn
	275						280					285			
Lys	Ala	Ile	Phe	Phe	Tyr	Asp	Pro	Ile	Thr	Thr	Thr	Gly	Ser	Val	Ala
	290					295					300				
Asp	Arg	Leu	Ile	Ile	Asn	Asn	Ser	Gln	Gly	Glu	Ala	Ser	Thr	Tyr	Asp
305					310					315					320
Gly	Ala	Ile	Val	Phe	Ser	Ser	Leu	Asn	Leu	Phe	Thr	His	Ser	Pro	Glu
			325						330					335	
Cys	Lys	Leu	Ser	Ser	Phe	Ser	Gln	Gly	Leu	Thr	Leu	Ala	Ala	Gly	Ser
		340						345					350		
Leu	Val	Leu	Glu	Glu	Gly	Val	Cys	Val	Gln	Ala	Pro	Ser	Phe	Asp	Gln
	355						360					365			
Arg	Ala	His	Ser	Gln	Leu	Phe	Met	Asn	Pro	Gly	Thr	Lys	Leu	Gln	Ala
	370					375					380				
Thr	Gln	Asn	Ile	Ser	Val	Lys	Asn	Leu	His	Leu	Asn	Leu	Asn	Arg	Ile
385					390					395					400
Ala	Glu	Glu	Pro	Ala	Tyr	Ile	Thr	Thr	Thr	Asp	Asp	Ala	Ser	Ser	Val
			405						410					415	
Asp	Ile	Cys	Gly	Pro	Val	Val	Met	His	Ile	Asp	Asp	Glu	Ile	Phe	Tyr
		420					425					430			
Asn	Gln	Thr	Val	Leu	Ala	Asn	Glu	Leu	Ser	Val	Glu	Cys	Leu	Asn	Leu
		435					440					445			
Ser	Ser	Pro	His	Leu	Asp	Asn	Ile	Thr	Ile	Asp	Asp	Val	Pro	Ala	Val
	450					455					460				
Pro	Ile	Met	Thr	Leu	Glu	Ser	His	Arg	Gly	Tyr	Gln	Gly	Thr	Trp	Glu
465				470						475					480
Ile	Ser	Trp	Lys	Glu	Gln	Pro	Lys	Leu	Thr	Phe	Gly	Lys	Ala	Thr	Ile
			485						490					495	
Ala	Pro	Asn	Lys	Gln	Met	His	Leu	Ile	Trp	Lys	Pro	Ser	Gly	Tyr	Val
			500					505					510		
Pro	Phe	Ser	Gly	Gly	Thr	Gly	Glu	Phe	Thr	Thr	Ser	Leu	Val	Pro	Asn
	515						520					525			
Ser	Leu	Trp	Asn	Leu	Phe	Leu	Asp	Thr	Arg	Phe	Ser	Gln	Gln	Ala	Ile
	530					535					540				
Glu	Lys	His	Ala	Val	Ser	Ser	Gly	Asn	Gly	Ile	Trp	Ile	Ser	Ser	Met
545						550				555					560
Thr	Asn	Ser	Phe	Leu	Gln	Gly	Ser	Thr	Asn	Asn	Asn	His	Gly	Phe	Arg
			565						570					575	
His	Lys	Ser	Ser	Gly	Tyr	Thr	Ala	Gly	Gly	Lys	Ile	Gln	Thr	Leu	Gln
			580					585					590		
Asp	Asp	Ile	Phe	Ser	Val	Ser	Phe	Ser	Gln	Leu	Phe	Gly	Arg	Ser	Lys
	595						600					605			
Asp	Phe	Gly	Ser	Ala	Thr	Ser	Lys	Asp	Thr	Phe	Leu	Ser	Gly	Ser	Ile

-continued

610	615	620	
Tyr Ala Gln His Ser Arg Arg Leu Leu Pro Ile Met Arg Phe Leu Ala			
625	630	635	640
Gly Thr Ser Thr Tyr Arg Pro Arg Leu Leu Leu Ser Ile Pro Lys Asn			
	645	650	655
Leu Pro Ile Asn Phe Asp Val Leu Val Ser Tyr Ser Tyr Asp Ser Asn			
	660	665	670
His Met Lys Val Gln Lys Phe			
675			

<210> SEQ ID NO 54
 <211> LENGTH: 487
 <212> TYPE: DNA
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 54

acctcgagag aggattctct tagtgtggct ttctgtcagt tatttgcaa agataaagac	60
taccttgtaa gcaagaacgc cgcaaacgtc tatgcgggtt ctgtatatta tcagcatgtg	120
agcaagtttg atgatctcac gcggttattt aatgggccta acacgtgttg ttcagggttt	180
tctaaagaga ttcttatttt ctgggatgca caaattacct attgccacac ggccaacaac	240
atgacaacgt cctatacaga ctatcctgaa gtgaaagggt cttggggtaa tgataccctg	300
ggcttaactt tgttactag cgtacctatc ccggtattta gttcttctat ctttgatagt	360
tatgcaccgt ttgcaaaatt acaagttgtc tatgcgcacc aagatgactt taaagaacca	420
acaacagaag gccgggtctt tgaaagcagc gatcttctca acgtttctgt acctataggt	480
ataaaat	487

<210> SEQ ID NO 55
 <211> LENGTH: 162
 <212> TYPE: PRT
 <213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 55

Thr Ser Arg Glu Asp Ser Leu Ser Val Ala Phe Cys Gln Leu Phe Ala	
1 5 10 15	
Lys Asp Lys Asp Tyr Leu Val Ser Lys Asn Ala Ala Asn Val Tyr Ala	
20 25 30	
Gly Ser Val Tyr Tyr Gln His Val Ser Lys Phe Asp Asp Leu Thr Arg	
35 40 45	
Leu Phe Asn Gly Pro Asn Thr Cys Cys Ser Gly Phe Ser Lys Glu Ile	
50 55 60	
Pro Ile Phe Leu Asp Ala Gln Ile Thr Tyr Cys His Thr Ala Asn Asn	
65 70 75 80	
Met Thr Thr Ser Tyr Thr Asp Tyr Pro Glu Val Lys Gly Ser Trp Gly	
85 90 95	
Asn Asp Thr Leu Gly Leu Thr Leu Ser Thr Ser Val Pro Ile Pro Val	
100 105 110	
Phe Ser Ser Ser Ile Phe Asp Ser Tyr Ala Pro Phe Ala Lys Leu Gln	
115 120 125	
Val Val Tyr Ala His Gln Asp Asp Phe Lys Glu Pro Thr Thr Glu Gly	
130 135 140	
Arg Val Phe Glu Ser Ser Asp Leu Leu Asn Val Ser Val Pro Ile Gly	

-continued

145	150	155	160
Ile Lys			
<210> SEQ ID NO 56			
<211> LENGTH: 2781			
<212> TYPE: DNA			
<213> ORGANISM: Chlamydia psittaci			
<400> SEQUENCE: 56			
atgaggcctt ctttatataa gattttaata tcgtcgacgc tgacgttacc aatatctttt	60		
cacttctcgc aattgcatgc agaagtggct ttaactcaag aatctattct cgatgcaa	120		
ggagcattca gtccgcaatc tacaagcact gcgggaggaa cgatttaca cgtcgagagt	180		
gatatttcta ttgtagatgt aggacagaca gcggctcttg cttcctcagc tttgttcag	240		
actgcagaca acctaacttt caaagggaac aaccatagct tatccataac gaacgcgaat	300		
gccggagcta atcctgcggg aattaacggt aacactgccg ataagattct tacgctgaca	360		
gatttttcta agttgagctt taaggaatgc ccatcttctc tagtgaatac tggaaaagg	420		
gctatgaaat ccggaggagc attaaactta gcgaataatg ccagtattct gtttgatcag	480		
aactattccg ctgagaatgg tggagccatc tcttgcaaag ctttttctct aaccggctcg	540		
agcaaagaaa tcagcttcac cactaactct actgcgaaaa aggtggagc gattgctgct	600		
acgggaatag ctcactcttc ggacaaccaa ggcacaatca gattttctgg gaacactgct	660		
gtgaattctg ggggagcagt atattcagaa gcttctatga cgattgcagg taacaaccac	720		
gttgctttta gcaacaatgc tgtttccggt tcactctgatg gttgcggtgg agctatccat	780		
tgtagcaaaa caggttcagc accgaccctt actataagag ataacaaagt cttgattttt	840		
gaggaaaata cttcttcagc aaaagggtga gcgatttaca ccgataaact catattgact	900		
tctggtgggc ctacggcatt tatcaataac aaagttaccc atgctacacc taagggtgga	960		
gctattggta ttgctgcaa tggagaatgt agcttaaccg ctgaacatgg ggatattact	1020		
tttgataata acctgatggc cacacaagac aatgctacaa taaaaagaaa tgccattaac	1080		
attgaaggca atggtaaatt cgtcaactta cgtgcagcgt ctggaaagac gatttctttc	1140		
tatgaccta tcacagtga aggtaatgct gctgatcttc tcactttgaa taaagctgag	1200		
ggtgataaaa cgtataatgg aagaattatt ttttcaggag aaaagctcac tgaagaacaa	1260		
gctgctgttg cgataaacct aaagacaaca ttacacagc ctatcacttt agctgctggt	1320		
gaacttggtg tacgcagcgg tgtggaagta gaagcaaaaa cagtcgtgca aacagcagga	1380		
tccttgattc tgatggatgc aggcacaaag ttatccgcaa aaacagaaga tgctacactg	1440		
acgaatctgg ctattaatcc gaataacctt gatgggaaaa aattcgccgt agtcgatgcc	1500		
gttgctgctg ggaagaatgt gactttatca ggtgctattg gcgttattga tcctacagg	1560		
aagttttatg aaaaccataa gctaaatgat acgttagctt taggaggaat tcaactttct	1620		
gggaaagggt cggtgacaac aaccaacgtg cctagtcatg ttgttggtgt tgctgaaacc	1680		
cactatgggt atcaaggaaa ctggtctgtc agttgggtca aagataataa ctctgacct	1740		
aaaacacaaa cagcaatctt tacctggaat aaaacaggat atgttccaaa tcctgaacgt	1800		
cgtgctccgc tagtactcaa tagcctttgg ggatccttta tagatttacg ttctattcaa	1860		
gatgtcttgg aacgtagtgt tgatagtatt cttgagacac gtcgtggttt gtgggtctct	1920		

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ggaattggga acttcttcca taaagatcgg aatgctgaaa atcgcaaatt ccgtcatatc 1980
agttcgggat atgtgttagg agccacaaca aatacctcga gagaggattc tcttagtgtg 2040
gctttctgtc agttatttgc aaaagataaa gactaccttg taagcaagaa cgccgcaaac 2100
gtctatgcgg gttctgtata ttatcagcat gtgagcaagt ttgatgatct cacgcgggta 2160
tttaatgggc ctaacacgtg ttgttcaggg ttttctaaag agattcctat tttcttgat 2220
gcacaaatta cctattgcc aacggccaac aacatgacaa cgtcctatac agactatcct 2280
gaagtgaag gttcttgggg taatgatacc ctgggcttaa ctttgtctac tagcgtacct 2340
atcccgggat ttagttcttc tatctttgat agttatgcac cgtttgcaa attacaagtt 2400
gtctatgcgc accaagatga ctttaagaa ccaacaacag aagccgggt ctttgaaagc 2460
agcgatcttc tcaacgtttc tgtacctata ggtataaaat ttgagaaact ctcctatgga 2520
gagagaagtg cttatgatct tacactgatg tatatacctg atgtgtaccg tcataatcca 2580
agctgtatga caggattggc gatcaatgac gtttcctggt taaccacagc tacgaatctt 2640
gctagacaag ctttcatagt tcgcgcgggt aaccatattg ccttaacctc tgggtgtgag 2700
atgttcagtc agtttggttt cgaattacga agctcttcaa gaaattataa cgtagatctt 2760
ggcgctaagg tcgcgttcta a 2781

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<210> SEQ ID NO 57

<211> LENGTH: 926

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 57

```

Met  Arg Pro Ser Leu Tyr Lys Ile Leu Ile Ser Ser Thr Leu Thr Leu
  1          5          10          15

Pro  Ile Ser Phe His Phe Ser Gln Leu His Ala Glu Val Ala Leu Thr
      20          25          30

Gln  Glu Ser Ile Leu Asp Ala Asn Gly Ala Phe Ser Pro Gln Ser Thr
      35          40          45

Ser  Thr Ala Gly Gly Thr Ile Tyr Asn Val Glu Ser Asp Ile Ser Ile
      50          55          60

Val  Asp Val Gly Gln Thr Ala Ala Leu Ala Ser Ser Ala Phe Val Gln
      65          70          75          80

Thr  Ala Asp Asn Leu Thr Phe Lys Gly Asn Asn His Ser Leu Ser Ile
      85          90          95

Thr  Asn Ala Asn Ala Gly Ala Asn Pro Ala Gly Ile Asn Val Asn Thr
      100         105         110

Ala  Asp Lys Ile Leu Thr Leu Thr Asp Phe Ser Lys Leu Ser Phe Lys
      115         120         125

Glu  Cys Pro Ser Ser Leu Val Asn Thr Gly Lys Gly Ala Met Lys Ser
      130         135         140

Gly  Gly Ala Leu Asn Leu Ala Asn Asn Ala Ser Ile Leu Phe Asp Gln
      145         150         155         160

Asn  Tyr Ser Ala Glu Asn Gly Gly Ala Ile Ser Cys Lys Ala Phe Ser
      165         170         175

Leu  Thr Gly Ser Ser Lys Glu Ile Ser Phe Thr Thr Asn Ser Thr Ala
      180         185         190

Lys  Lys Gly Gly Ala Ile Ala Ala Thr Gly Ile Ala His Leu Ser Asp
      195         200         205

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Asn	Gln	Gly	Thr	Ile	Arg	Phe	Ser	Gly	Asn	Thr	Ala	Val	Asn	Ser	Gly
210						215					220				
Gly	Ala	Val	Tyr	Ser	Glu	Ala	Ser	Met	Thr	Ile	Ala	Gly	Asn	Asn	His
225					230					235					240
Val	Ala	Phe	Ser	Asn	Asn	Ala	Val	Ser	Gly	Ser	Ser	Asp	Gly	Cys	Gly
				245					250					255	
Gly	Ala	Ile	His	Cys	Ser	Lys	Thr	Gly	Ser	Ala	Pro	Thr	Leu	Thr	Ile
		260						265					270		
Arg	Asp	Asn	Lys	Val	Leu	Ile	Phe	Glu	Glu	Asn	Thr	Ser	Ser	Ala	Lys
	275					280						285			
Gly	Gly	Ala	Ile	Tyr	Thr	Asp	Lys	Leu	Ile	Leu	Thr	Ser	Gly	Gly	Pro
290						295					300				
Thr	Ala	Phe	Ile	Asn	Asn	Lys	Val	Thr	His	Ala	Thr	Pro	Lys	Gly	Gly
305					310					315					320
Ala	Ile	Gly	Ile	Ala	Ala	Asn	Gly	Glu	Cys	Ser	Leu	Thr	Ala	Glu	His
				325					330					335	
Gly	Asp	Ile	Thr	Phe	Asp	Asn	Asn	Leu	Met	Ala	Thr	Gln	Asp	Asn	Ala
		340						345					350		
Thr	Ile	Lys	Arg	Asn	Ala	Ile	Asn	Ile	Glu	Gly	Asn	Gly	Lys	Phe	Val
	355						360					365			
Asn	Leu	Arg	Ala	Ala	Ser	Gly	Lys	Thr	Ile	Ser	Phe	Tyr	Asp	Pro	Ile
370						375					380				
Thr	Val	Glu	Gly	Asn	Ala	Ala	Asp	Leu	Leu	Thr	Leu	Asn	Lys	Ala	Glu
385					390					395					400
Gly	Asp	Lys	Thr	Tyr	Asn	Gly	Arg	Ile	Ile	Phe	Ser	Gly	Glu	Lys	Leu
				405				410						415	
Thr	Glu	Glu	Gln	Ala	Ala	Val	Ala	Asp	Asn	Leu	Lys	Thr	Thr	Phe	Thr
			420					425						430	
Gln	Pro	Ile	Thr	Leu	Ala	Ala	Gly	Glu	Leu	Val	Leu	Arg	Ser	Gly	Val
		435					440					445			
Glu	Val	Glu	Ala	Lys	Thr	Val	Val	Gln	Thr	Ala	Gly	Ser	Leu	Ile	Leu
450						455					460				
Met	Asp	Ala	Gly	Thr	Lys	Leu	Ser	Ala	Lys	Thr	Glu	Asp	Ala	Thr	Leu
465					470					475					480
Thr	Asn	Leu	Ala	Ile	Asn	Pro	Asn	Thr	Leu	Asp	Gly	Lys	Lys	Phe	Ala
				485					490					495	
Val	Val	Asp	Ala	Val	Ala	Ala	Gly	Lys	Asn	Val	Thr	Leu	Ser	Gly	Ala
			500					505						510	
Ile	Gly	Val	Ile	Asp	Pro	Thr	Gly	Lys	Phe	Tyr	Glu	Asn	His	Lys	Leu
		515					520						525		
Asn	Asp	Thr	Leu	Ala	Leu	Gly	Gly	Ile	Gln	Leu	Ser	Gly	Lys	Gly	Ser
					535						540				
Val	Thr	Thr	Thr	Asn	Val	Pro	Ser	His	Val	Val	Gly	Val	Ala	Glu	Thr
545					550					555					560
His	Tyr	Gly	Tyr	Gln	Gly	Asn	Trp	Ser	Val	Ser	Trp	Val	Lys	Asp	Asn
				565					570					575	
Asn	Ser	Asp	Pro	Lys	Thr	Gln	Thr	Ala	Ile	Phe	Thr	Trp	Asn	Lys	Thr
			580					585						590	
Gly	Tyr	Val	Pro	Asn	Pro	Glu	Arg	Arg	Ala	Pro	Leu	Val	Leu	Asn	Ser
		595					600						605		

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Leu	Trp	Gly	Ser	Phe	Ile	Asp	Leu	Arg	Ser	Ile	Gln	Asp	Val	Leu	Glu
610						615					620				
Arg	Ser	Val	Asp	Ser	Ile	Leu	Glu	Thr	Arg	Arg	Gly	Leu	Trp	Val	Ser
625					630				635						640
Gly	Ile	Gly	Asn	Phe	Phe	His	Lys	Asp	Arg	Asn	Ala	Glu	Asn	Arg	Lys
			645					650						655	
Phe	Arg	His	Ile	Ser	Ser	Gly	Tyr	Val	Leu	Gly	Ala	Thr	Thr	Asn	Thr
			660				665						670		
Ser	Arg	Glu	Asp	Ser	Leu	Ser	Val	Ala	Phe	Cys	Gln	Leu	Phe	Ala	Lys
		675				680						685			
Asp	Lys	Asp	Tyr	Leu	Val	Ser	Lys	Asn	Ala	Ala	Asn	Val	Tyr	Ala	Gly
	690					695					700				
Ser	Val	Tyr	Tyr	Gln	His	Val	Ser	Lys	Phe	Asp	Asp	Leu	Thr	Arg	Leu
705				710					715						720
Phe	Asn	Gly	Pro	Asn	Thr	Cys	Cys	Ser	Gly	Phe	Ser	Lys	Glu	Ile	Pro
			725						730					735	
Ile	Phe	Leu	Asp	Ala	Gln	Ile	Thr	Tyr	Cys	His	Thr	Ala	Asn	Asn	Met
		740					745						750		
Thr	Thr	Ser	Tyr	Thr	Asp	Tyr	Pro	Glu	Val	Lys	Gly	Ser	Trp	Gly	Asn
		755				760						765			
Asp	Thr	Leu	Gly	Leu	Thr	Leu	Ser	Thr	Ser	Val	Pro	Ile	Pro	Val	Phe
	770				775					780					
Ser	Ser	Ser	Ile	Phe	Asp	Ser	Tyr	Ala	Pro	Phe	Ala	Lys	Leu	Gln	Val
785				790					795						800
Val	Tyr	Ala	His	Gln	Asp	Asp	Phe	Lys	Glu	Pro	Thr	Thr	Glu	Gly	Arg
			805					810						815	
Val	Phe	Glu	Ser	Ser	Asp	Leu	Leu	Asn	Val	Ser	Val	Pro	Ile	Gly	Ile
		820					825						830		
Lys	Phe	Glu	Lys	Leu	Ser	Tyr	Gly	Glu	Arg	Ser	Ala	Tyr	Asp	Leu	Thr
	835					840						845			
Leu	Met	Tyr	Ile	Pro	Asp	Val	Tyr	Arg	His	Asn	Pro	Ser	Cys	Met	Thr
	850				855						860				
Gly	Leu	Ala	Ile	Asn	Asp	Val	Ser	Trp	Leu	Thr	Thr	Ala	Thr	Asn	Leu
865				870					875						880
Ala	Arg	Gln	Ala	Phe	Ile	Val	Arg	Ala	Gly	Asn	His	Ile	Ala	Leu	Thr
			885					890					895		
Ser	Gly	Val	Glu	Met	Phe	Ser	Gln	Phe	Gly	Phe	Glu	Leu	Arg	Ser	Ser
		900					905					910			
Ser	Arg	Asn	Tyr	Asn	Val	Asp	Leu	Gly	Ala	Lys	Val	Ala	Phe		
		915					920					925			

<210> SEQ ID NO 58

<211> LENGTH: 559

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 58

tggtgttcatt ctttagcagg agttgcattt acgttggttc tctgtgagca tatgtttacc	60
aatatgcttg cttcttctta ttttaaggaa ggcagtgggt ttgttcagtt agtgagcaaa	120
tttcatcaga ttcttggtct gaagatcata gaaattgttt ttttagccct accgtttact	180
tgtcacgcta tcctaggtat tttctatctt tttcaagcgc aaactaatc acgggcttct	240

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gacggcagaa aacccgcggt aatctatgcg agaaatcttg cctatacttg gcagagaaga 300
actgcttgga ttttactttt cggctcttatt tttcacgtag ttcagtttcg ttttcttcgt 360
tatectattc atgtagagct gcatgggcaa acatactatg ttgtcgatat tgacgcttct 420
cggtatgcgg cgatagtgcg gggtacacaa ggatttttta ctataaattt ttcagctcct 480
caacttgaaa cgattcgttt ggataaagag gatcttgacg gcagcgcagt ttctcaatta 540
ttagacagaa aagcgtatc 559

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<210> SEQ ID NO 59
<211> LENGTH: 186
<212> TYPE: PRT
<213> ORGANISM: Chlamydia psittaci

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```

<400> SEQUENCE: 59

```

```

Cys  Val His Ser Leu Ala Gly Val Ala Phe Thr Leu Phe Leu Cys Glu
  1          5          10          15
His  Met Phe Thr Asn Met Leu Ala Ser Ser Tyr Phe Lys Glu Gly Ser
          20          25          30
Gly  Phe Val Gln Leu Val Ser Lys Phe His Gln Ile Pro Gly Leu Lys
          35          40          45
Ile  Ile Glu Ile Val Phe Leu Ala Leu Pro Phe Thr Cys His Ala Ile
          50          55          60
Leu  Gly Ile Phe Tyr Leu Phe Gln Ala Gln Thr Asn Ser Arg Ala Ser
        65          70          75          80
Asp  Gly Arg Lys Pro Ala Leu Ile Tyr Ala Arg Asn Leu Ala Tyr Thr
          85          90          95
Trp  Gln Arg Arg Thr Ala Trp Ile Leu Leu Phe Gly Leu Ile Phe His
          100         105         110
Val Val Gln Phe Arg Phe Leu Arg Tyr Pro Ile His Val Glu Leu His
          115         120         125
Gly Gln Thr Tyr Tyr Val Val Asp Ile Asp Ala Ser Arg Tyr Ala Ala
          130         135         140
Ile Val Arg Gly Thr Gln Gly Phe Phe Thr Ile Asn Phe Ser Ala Pro
          145         150         155         160
Gln Leu Glu Thr Ile Arg Leu Asp Lys Glu Asp Leu Asp Gly Ser Ala
          165         170         175
Val Ser Gln Leu Leu Asp Arg Lys Ala Tyr
          180         185

```

```

<210> SEQ ID NO 60
<211> LENGTH: 687
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci

```

```

<400> SEQUENCE: 60

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```

atgatgaatg aaaaggaatc atgttctgag gctactcaga ggtcatggaa gtactacact 60
agctttgttt tacgttgtgt tcattcttta gcaggagttg catttacgtt gtttctctgt 120
gagcatatgt ttaccaatat gcttgcttct tcttatttta aggaaggcag tggttttgtt 180
cagttagtga gcaaatttca tcagattcct ggtctgaaga tcatagaaat tgttttttta 240
gccctaccgt ttacttgtca cgctatccta ggtattttct atctttttca agcgcaaaact 300
aattcacggg cttctgacgg cagaaaaccc gcgttaatct atgcgagaaa tcttgcctat 360

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acttggcaga gaagaactgc ttggatttta cttttcggtc ttatttttca cgtagttcag 420
tttcgttttc ttcgttatcc tattcatgta gagctgcatg ggcaaacata ctatgttgtc 480
gatattgacg cttctcggta tgcggcgata gtgcggggta cacaaggatt ttttactata 540
aatttttcag ctcctcaact tgaaacgatt cgtttggata aagaggatct tgacggcagc 600
gcagtttctc aattattaga cagaaaagcg tatctcctga ctcctaattgt tggaccgctt 660
ttctttatgt tgttcgggat tcttttag 687

```

```

<210> SEQ ID NO 61
<211> LENGTH: 228
<212> TYPE: PRT
<213> ORGANISM: Chlamydia psittaci

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```

<400> SEQUENCE: 61

```

```

Met  Met Asn Glu Lys Glu Ser Cys Ser Glu Ala Thr Gln Arg Ser Trp
  1      5      10      15
Lys  Tyr Tyr Thr Ser Phe Val Leu Arg Cys Val His Ser Leu Ala Gly
      20      25      30
Val  Ala Phe Thr Leu Phe Leu Cys Glu His Met Phe Thr Asn Met Leu
      35      40      45
Ala  Ser Ser Tyr Phe Lys Glu Gly Ser Gly Phe Val Gln Leu Val Ser
      50      55      60
Lys  Phe His Gln Ile Pro Gly Leu Lys Ile Ile Glu Ile Val Phe Leu
      65      70      75      80
Ala  Leu Pro Phe Thr Cys His Ala Ile Leu Gly Ile Phe Tyr Leu Phe
      85      90      95
Gln  Ala Gln Thr Asn Ser Arg Ala Ser Asp Gly Arg Lys Pro Ala Leu
      100     105     110
Ile  Tyr Ala Arg Asn Leu Ala Tyr Thr Trp Gln Arg Arg Thr Ala Trp
      115     120     125
Ile  Leu Leu Phe Gly Leu Ile Phe His Val Val Gln Phe Arg Phe Leu
      130     135     140
Arg  Tyr Pro Ile His Val Glu Leu His Gly Gln Thr Tyr Tyr Val Val
      145     150     155     160
Asp  Ile Asp Ala Ser Arg Tyr Ala Ala Ile Val Arg Gly Thr Gln Gly
      165     170     175
Phe  Phe Thr Ile Asn Phe Ser Ala Pro Gln Leu Glu Thr Ile Arg Leu
      180     185     190
Asp  Lys Glu Asp Leu Asp Gly Ser Ala Val Ser Gln Leu Leu Asp Arg
      195     200     205
Lys  Ala Tyr Leu Leu Thr Pro Asn Val Gly Pro Leu Phe Phe Met Leu
      210     215     220
Phe Gly Ile Leu
225

```

```

<210> SEQ ID NO 62
<211> LENGTH: 1329
<212> TYPE: DNA
<213> ORGANISM: Chlamydia psittaci

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```

<400> SEQUENCE: 62

```

```

atgactctac aaccctacca agcatcctct agaaagtacc gtccacaaat ctttcgagaa 60
attctaggtc agagctctgt tgtcgtgtga ttaaaaaatg ccttggtctt caaccgagcc 120

```

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gccacgcct atctattttc tggaattcgt ggtacagga aaaccacact agctcgcat 180
ttagcaaaa ctctgaactg cgtgcatctt agcgaggatg gcgagccctg caaccagtgt 240
ttttcttgta aagagattgc ttcaggatcc tctttagacg ttttagaaat tgacggagcc 300
tcccaccgtg gtatcgaaga tatccgtcaa attaataaaa ctgtattatt cactcctgta 360
aaagcaaagt ttaaaattta tatcatagat gaagttcata tgctcactaa ggaagccttc 420
aatgctttat tgaagacttt agaagagcct ccacaacatg taaaattttt ctttgcaact 480
acagaaatcc ataaaattcc cggaactatt ttaagtcgtt gtcaaaaaat gcactttcaa 540
aggattcctg aaaaacgat cctggagaag ctatcgctta tggctcaaga tgaccatatt 600
gaggcgctgc aagaagcatt ggcgccgac gcccgtcag cacaaggaag cttgcgtgat 660
gcagaatctc tttatgacta cgtaatatct ttatttccta aatctctctc tcccgacacg 720
gttgcccaag ctttaggctt tgcttcccaa gattctctcc ggactttaga caatgcgatt 780
cttcaaaggg actatgcgac agccttaggg atcgtaacgg acttcttaaa ttctggggta 840
gcacctgtca catttctcca tgacctaca ttattttatc gtaatcttct tcttacgaat 900
tctacaacaa gcaagttcag ctctcagtat aagacggagc agcttctaga aatcatagat 960
ttccttgag aatctgctaa gcacctacaa aataccatct tcgaacagac atttttagaa 1020
accgtcatca ttcatatcat tcgcatctat caaaggcctg ttttatcaga gttgatctct 1080
tctattaaga gtcggcagtt tgaagggtt cgcaatatta aggagccac cttgacgcag 1140
caagtatcag ctctcaacc tcagcccacc tacaagaac agagtttttt agagaaaaa 1200
aatcaacctg ctgcggaagg taaaattata tctgtagaag ttaaaagctc agcttcaata 1260
aaatctgcag ctgtagacac attattacag ttgctgttg tagaattttc aggaatttta 1320
agacaataa 1329

```

<210> SEQ ID NO 63

<211> LENGTH: 442

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 63

```

Met  Thr Leu Gln Pro Tyr Gln Ala Ser Ser Arg Lys Tyr Arg Pro Gln
  1          5          10          15

Ile  Phe Arg Glu Ile Leu Gly Gln Ser Ser Val Val Ala Val Leu Lys
    20          25          30

Asn  Ala Leu Val Phe Asn Arg Ala Ala His Ala Tyr Leu Phe Ser Gly
    35          40          45

Ile  Arg Gly Thr Gly Lys Thr Thr Leu Ala Arg Ile Leu Ala Lys Ala
    50          55          60

Leu  Asn Cys Val His Leu Ser Glu Asp Gly Glu Pro Cys Asn Gln Cys
    65          70          75          80

Phe  Ser Cys Lys Glu Ile Ala Ser Gly Ser Ser Leu Asp Val Leu Glu
    85          90          95

Ile  Asp Gly Ala Ser His Arg Gly Ile Glu Asp Ile Arg Gln Ile Asn
   100          105          110

Glu  Thr Val Leu Phe Thr Pro Val Lys Ala Lys Phe Lys Ile Tyr Ile
   115          120          125

Ile  Asp Glu Val His Met Leu Thr Lys Glu Ala Phe Asn Ala Leu Leu

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130	135	140
Lys Thr Leu Glu Glu Pro Pro Gln His Val Lys Phe Phe Phe Ala Thr		
145	150	155 160
Thr Glu Ile His Lys Ile Pro Gly Thr Ile Leu Ser Arg Cys Gln Lys		
	165	170 175
Met His Leu Gln Arg Ile Pro Glu Lys Thr Ile Leu Glu Lys Leu Ser		
	180	185 190
Leu Met Ala Gln Asp Asp His Ile Glu Ala Ser Gln Glu Ala Leu Ala		
	195	200 205
Pro Ile Ala Arg Ala Ala Gln Gly Ser Leu Arg Asp Ala Glu Ser Leu		
	210	215 220
Tyr Asp Tyr Val Ile Ser Leu Phe Pro Lys Ser Leu Ser Pro Asp Thr		
	225	230 235 240
Val Ala Gln Ala Leu Gly Phe Ala Ser Gln Asp Ser Leu Arg Thr Leu		
	245	250 255
Asp Asn Ala Ile Leu Gln Arg Asp Tyr Ala Thr Ala Leu Gly Ile Val		
	260	265 270
Thr Asp Phe Leu Asn Ser Gly Val Ala Pro Val Thr Phe Leu His Asp		
	275	280 285
Leu Thr Leu Phe Tyr Arg Asn Leu Leu Leu Thr Asn Ser Thr Thr Ser		
	290	295 300
Lys Phe Ser Ser Gln Tyr Lys Thr Glu Gln Leu Leu Glu Ile Ile Asp		
	305	310 315 320
Phe Leu Gly Glu Ser Ala Lys His Leu Gln Asn Thr Ile Phe Glu Gln		
	325	330 335
Thr Phe Leu Glu Thr Val Ile Ile His Ile Ile Arg Ile Tyr Gln Arg		
	340	345 350
Pro Val Leu Ser Glu Leu Ile Ser Ser Ile Lys Ser Arg Gln Phe Glu		
	355	360 365
Gly Leu Arg Asn Ile Lys Glu Pro Thr Leu Thr Gln Gln Val Ser Ala		
	370	375 380
Pro Gln Pro Gln Pro Thr Tyr Lys Glu Gln Ser Phe Leu Glu Lys Lys		
	385	390 395 400
Asn Gln Pro Ala Ala Glu Gly Lys Ile Ile Ser Val Glu Val Lys Ser		
	405	410 415
Ser Ala Ser Ile Lys Ser Ala Ala Val Asp Thr Leu Leu Gln Phe Ala		
	420	425 430
Val Val Glu Phe Ser Gly Ile Leu Arg Gln		
	435	440

<210> SEQ ID NO 64

<211> LENGTH: 1479

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 64

atgtatcgat atagtgcctt agaattagca aaagctgtga ctttagggga actgacagcc	60
acaggggtga ctcaacattt ttttcataga atagaagaag ctgaggggca ggtaggtgcc	120
tttatttcct tgtgtaagga acaagcttta gaacaggcag agctcataga taaaaagcgt	180
tcgcgtggag aacctttagg aaaactcgca ggtgttcctg taggaattaa agataatatt	240
cacgttacag gcctgaagac aacatgcgcc tctcgtgtgc tcgagaatta tcaaccaccg	300

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tttgatgcta ctgtttaga aagaatcaaa aaagaagatg ggattatctt aggcaaactc 360
aatatggatg agtttgctat gggatcaaca acgctatatatt ctgcttttca tcctacccac 420
aaccctggg atttatctcg tgttcctgga ggttcttcag ggggatctgc ggcgcagtt 480
tctgctagat tttgtcccgt agccctagga tcagataccg gaggatccat ccgtcagccc 540
gcagcatttt gtggtgttgt aggttttaag ccttcctacg gagccgtttc gcgttacggg 600
ctttagcctt ttgcctcttc gctagatcaa atcggtcctt tagccaatac tgtagaagac 660
gtcgccttaa tgatggatgt gttttctggt agagatccta aagatgcaac ctcaagagag 720
ttttccgtg attcttttat gagcaagttg tctacggagg ttcctaaagt gattggggtg 780
cctagaacat ttttagaggg actccgtgat gatattaggg agaatttctt ctcttcatta 840
gccattttt aaggagaagg aaccatctt gtggatgtgg agttggatat tctcagccac 900
gctgtatcta tatattacat tttagcatct gctgaagctg ccacgaattt agcaaggttc 960
gatggggtgc gttatggata tcgttctcct caagcgcata ccatcagcca actctacgat 1020
ctctcacgtg gagaaggatt tggcaaaag gtcatgcgca gaatcctctt agggaaactat 1080
gtcttgctg cgagagaca gaatgtttat tataagaaag ctacggcagt gcgtgctaag 1140
attgtaaaag catttagaac tgcatttgaa aagtgtgaaa tcttagccat gcccgctctgt 1200
tctagccccg cgtttgaaat aggagaaatt ctagatcctg tgactttata tctacaggat 1260
atctatactg tagctatgaa tttagcgtat cttcctgccca ttgccgtacc ctctggattt 1320
tctaaggagg gcctgccctt aggcctacag attatcggac agcaaggaca agaccaacaa 1380
gtgtgccaa ggggttacag tttccaagag catgcgcaa ttaagcaatt gttttctaag 1440
agatatgcca aaagtgttgt tctaggaggt caatcatga 1479

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<210> SEQ ID NO 65

<211> LENGTH: 492

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 65

```

Met  Tyr  Arg  Tyr  Ser  Ala  Leu  Glu  Leu  Ala  Lys  Ala  Val  Thr  Leu  Gly
 1          5          10          15

Glu  Leu  Thr  Ala  Thr  Gly  Val  Thr  Gln  His  Phe  Phe  His  Arg  Ile  Glu
 20          25          30

Glu  Ala  Glu  Gly  Gln  Val  Gly  Ala  Phe  Ile  Ser  Leu  Cys  Lys  Glu  Gln
 35          40          45

Ala  Leu  Glu  Gln  Ala  Glu  Leu  Ile  Asp  Lys  Lys  Arg  Ser  Arg  Gly  Glu
 50          55          60

Pro  Leu  Gly  Lys  Leu  Ala  Gly  Val  Pro  Val  Gly  Ile  Lys  Asp  Asn  Ile
 65          70          75          80

His  Val  Thr  Gly  Leu  Lys  Thr  Thr  Cys  Ala  Ser  Arg  Val  Leu  Glu  Asn
 85          90          95

Tyr  Gln  Pro  Pro  Phe  Asp  Ala  Thr  Val  Val  Glu  Arg  Ile  Lys  Lys  Glu
100          105          110

Asp  Gly  Ile  Ile  Leu  Gly  Lys  Leu  Asn  Met  Asp  Glu  Phe  Ala  Met  Gly
115          120          125

Ser  Thr  Thr  Leu  Tyr  Ser  Ala  Phe  His  Pro  Thr  His  Asn  Pro  Trp  Asp
130          135          140

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Leu	Ser	Arg	Val	Pro	Gly	Gly	Ser	Ser	Gly	Gly	Ser	Ala	Ala	Ala	Val
145					150				155						160
Ser	Ala	Arg	Phe	Cys	Pro	Val	Ala	Leu	Gly	Ser	Asp	Thr	Gly	Gly	Ser
			165					170					175		
Ile	Arg	Gln	Pro	Ala	Ala	Phe	Cys	Gly	Val	Val	Gly	Phe	Lys	Pro	Ser
		180					185					190			
Tyr	Gly	Ala	Val	Ser	Arg	Tyr	Gly	Leu	Val	Ala	Phe	Ala	Ser	Ser	Leu
	195					200					205				
Asp	Gln	Ile	Gly	Pro	Leu	Ala	Asn	Thr	Val	Glu	Asp	Val	Ala	Leu	Met
	210				215					220					
Met	Asp	Val	Phe	Ser	Gly	Arg	Asp	Pro	Lys	Asp	Ala	Thr	Ser	Arg	Glu
225					230				235						240
Phe	Phe	Arg	Asp	Ser	Phe	Met	Ser	Lys	Leu	Ser	Thr	Glu	Val	Pro	Lys
			245					250					255		
Val	Ile	Gly	Val	Pro	Arg	Thr	Phe	Leu	Glu	Gly	Leu	Arg	Asp	Asp	Ile
		260					265					270			
Arg	Glu	Asn	Phe	Phe	Ser	Ser	Leu	Ala	Ile	Phe	Glu	Gly	Glu	Gly	Thr
	275					280					285				
His	Leu	Val	Asp	Val	Glu	Leu	Asp	Ile	Leu	Ser	His	Ala	Val	Ser	Ile
	290					295				300					
Tyr	Tyr	Ile	Leu	Ala	Ser	Ala	Glu	Ala	Ala	Thr	Asn	Leu	Ala	Arg	Phe
305				310					315					320	
Asp	Gly	Val	Arg	Tyr	Gly	Tyr	Arg	Ser	Pro	Gln	Ala	His	Thr	Ile	Ser
			325					330					335		
Gln	Leu	Tyr	Asp	Leu	Ser	Arg	Gly	Glu	Gly	Phe	Gly	Lys	Glu	Val	Met
		340					345					350			
Arg	Arg	Ile	Leu	Leu	Gly	Asn	Tyr	Val	Leu	Ser	Ala	Glu	Arg	Gln	Asn
		355				360					365				
Val	Tyr	Tyr	Lys	Lys	Ala	Thr	Ala	Val	Arg	Ala	Lys	Ile	Val	Lys	Ala
	370				375					380					
Phe	Arg	Thr	Ala	Phe	Glu	Lys	Cys	Glu	Ile	Leu	Ala	Met	Pro	Val	Cys
385				390					395					400	
Ser	Ser	Pro	Ala	Phe	Glu	Ile	Gly	Glu	Ile	Leu	Asp	Pro	Val	Thr	Leu
			405					410					415		
Tyr	Leu	Gln	Asp	Ile	Tyr	Thr	Val	Ala	Met	Asn	Leu	Ala	Tyr	Leu	Pro
		420				425						430			
Ala	Ile	Ala	Val	Pro	Ser	Gly	Phe	Ser	Lys	Glu	Gly	Leu	Pro	Leu	Gly
	435					440						445			
Leu	Gln	Ile	Ile	Gly	Gln	Gln	Gly	Gln	Asp	Gln	Gln	Val	Cys	Gln	Val
	450				455					460					
Gly	Tyr	Ser	Phe	Gln	Glu	His	Ala	Gln	Ile	Lys	Gln	Leu	Phe	Ser	Lys
465				470					475					480	
Arg	Tyr	Ala	Lys	Ser	Val	Val	Leu	Gly	Gly	Gln	Ser				
			485					490							

<210> SEQ ID NO 66

<211> LENGTH: 1962

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 66

atgagctacc gtaaactgtc gactctaatt gttctaggag tgtttgctct ttatgctctt

60

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ctagtattgc gttattataa aattcaaatt tgtgaaggag accactgggc cgcagaagct	120
ctcgggcaac acgaattttg tgtccgtgat ccttttcgaa ggggcacctt ttttgctaac	180
acgacagtac gtaagggaga caaagacctt cagcagcctt tcgctgtcga tattacaaaa	240
tttcaccttt gtgcagatcc tttagctatt cccgaatgtc atcgtgatga gatcatccaa	300
gggattctcc aattttattga ggggcagacc tacgacgacc tctccctaaa gttagataag	360
aaatctcggg attgtaagct gtatccttta ttagatgttt ctgtccatga ccggctatcc	420
ctttggtgga aaggatatgc aacaaagcat cgcttaccaa caaacgccct attttttatt	480
acggactacc aacgctcgta tccttttggg aagctccttg gacaagtctt ccatacctta	540
agagaaatta aggatgagaa aacaggaaaa gcctttccca caggcgggat ggaggcgtag	600
tttaatcata ttctggaagg ggacgttgga gagagaaagc tgttgcgctt tcctttgaac	660
cgtttagata cgaatcgtgt tatcaaaactg cctaaagatg gctctgatat ctaccttacg	720
atcaatcctg tgatccagac cattgcagag gaagaactcg aacggggcgt gctagaagct	780
aaagcccagg ggggtaggct cattctaagt aactcccaaa caggagagat tcttgcactg	840
gctcaatatc cgtttttcga tcccacaaat tataaggaat acttcaataa caaagagcgc	900
atcgaacata cgaaggtatc ttttgtgagc gatgttttg aacccgggtc gatcatgaaa	960
cctttgactg tggcgattgc tttaacagct aacgaagagg cttagcttaa atcgcagaaa	1020
aagatttttg atcctgaaga acctatcgat gtgaccagga cactcttccc tggacgaaaa	1080
ggatctccgc ttaaggatat ttctagaaac tctcaattga atatgtacat ggctatccag	1140
aaatcttcga atgtctatgt agctcagctg gctgaccgca tcatacaatc tttaggagtg	1200
gcctgggtacc aacagaagtt gctagctctg ggatttgaa gaaaaacagg gatcgagctt	1260
cccagtgagg cctctggttt ggtgccttct ccccatcggt tccatattaa tggttccctg	1320
gaatggtcct tatctactcc atattctttg gctatgggat ataataatctt ggcaacaggg	1380
atacaaatgg ttcaagccta cgctatcctt gcaaacggag gttatgccgt ccggccact	1440
ttagtaaaaa agatcgcttc tgcttcagga gaggaatatc atcttcctac taaagagaag	1500
acacgactct tttcagaaga aattactaga gaagttgttc gtgccatgcg ttttacaacg	1560
ttacccggag gttcgggatt tcgagcctct cctaagcatc actctagtgc tgggaaaaca	1620
ggaactacag aaaagatgat tcatggaaaa tatgataaac gccgtcatat tgcttctttt	1680
ataggtttta ctcccgtaga gagctcggag ggaaatttcc cacctttagt gatgctcgtc	1740
tccatagatg atcctgaata tggtttgca gccgacggca cgaaaaatta tatggggggg	1800
cgttggtcgg caccattttt ttctagggtt gctgaccgca cactcctcta tttagggatt	1860
cttcagaca agaagctaag aaattgcgac gaagaagctg ctgcattaaa gcgtctctat	1920
gaagaatgga atcgttctcc gaaacaaggg ggaacgaggt ga	1962

<210> SEQ ID NO 67

<211> LENGTH: 653

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 67

Met	Ser	Tyr	Arg	Lys	Arg	Ser	Thr	Leu	Ile	Val	Leu	Gly	Val	Phe	Ala
1				5				10						15	

Leu	Tyr	Ala	Leu	Leu	Val	Leu	Arg	Tyr	Tyr	Lys	Ile	Gln	Ile	Cys	Glu
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----

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	20					25					30				
Gly	Asp	His	Trp	Ala	Ala	Glu	Ala	Leu	Gly	Gln	His	Glu	Phe	Cys	Val
	35						40					45			
Arg	Asp	Pro	Phe	Arg	Arg	Gly	Thr	Phe	Phe	Ala	Asn	Thr	Thr	Val	Arg
	50					55					60				
Lys	Gly	Asp	Lys	Asp	Leu	Gln	Gln	Pro	Phe	Ala	Val	Asp	Ile	Thr	Lys
65					70					75				80	
Phe	His	Leu	Cys	Ala	Asp	Pro	Leu	Ala	Ile	Pro	Glu	Cys	His	Arg	Asp
				85					90					95	
Glu	Ile	Ile	Gln	Gly	Ile	Leu	Gln	Phe	Ile	Glu	Gly	Gln	Thr	Tyr	Asp
			100					105					110		
Asp	Leu	Ser	Leu	Lys	Leu	Asp	Lys	Lys	Ser	Arg	Tyr	Cys	Lys	Leu	Tyr
	115						120					125			
Pro	Leu	Leu	Asp	Val	Ser	Val	His	Asp	Arg	Leu	Ser	Leu	Trp	Trp	Lys
	130					135					140				
Gly	Tyr	Ala	Thr	Lys	His	Arg	Leu	Pro	Thr	Asn	Ala	Leu	Phe	Phe	Ile
145					150					155					160
Thr	Asp	Tyr	Gln	Arg	Ser	Tyr	Pro	Phe	Gly	Lys	Leu	Leu	Gly	Gln	Val
			165						170					175	
Leu	His	Thr	Leu	Arg	Glu	Ile	Lys	Asp	Glu	Lys	Thr	Gly	Lys	Ala	Phe
			180					185					190		
Pro	Thr	Gly	Gly	Met	Glu	Ala	Tyr	Phe	Asn	His	Ile	Leu	Glu	Gly	Asp
	195						200					205			
Val	Gly	Glu	Arg	Lys	Leu	Leu	Arg	Ser	Pro	Leu	Asn	Arg	Leu	Asp	Thr
	210					215					220				
Asn	Arg	Val	Ile	Lys	Leu	Pro	Lys	Asp	Gly	Ser	Asp	Ile	Tyr	Leu	Thr
225					230					235					240
Ile	Asn	Pro	Val	Ile	Gln	Thr	Ile	Ala	Glu	Glu	Glu	Leu	Glu	Arg	Gly
			245						250					255	
Val	Leu	Glu	Ala	Lys	Ala	Gln	Gly	Gly	Arg	Leu	Ile	Leu	Met	Asn	Ser
		260					265						270		
Gln	Thr	Gly	Glu	Ile	Leu	Ala	Leu	Ala	Gln	Tyr	Pro	Phe	Phe	Asp	Pro
	275						280					285			
Thr	Asn	Tyr	Lys	Glu	Tyr	Phe	Asn	Asn	Lys	Glu	Arg	Ile	Glu	His	Thr
	290					295					300				
Lys	Val	Ser	Phe	Val	Ser	Asp	Val	Phe	Glu	Pro	Gly	Ser	Ile	Met	Lys
305					310					315					320
Pro	Leu	Thr	Val	Ala	Ile	Ala	Leu	Gln	Ala	Asn	Glu	Glu	Ala	Ser	Leu
			325						330					335	
Lys	Ser	Gln	Lys	Lys	Ile	Phe	Asp	Pro	Glu	Glu	Pro	Ile	Asp	Val	Thr
			340					345					350		
Arg	Thr	Leu	Phe	Pro	Gly	Arg	Lys	Gly	Ser	Pro	Leu	Lys	Asp	Ile	Ser
	355						360					365			
Arg	Asn	Ser	Gln	Leu	Asn	Met	Tyr	Met	Ala	Ile	Gln	Lys	Ser	Ser	Asn
	370					375					380				
Val	Tyr	Val	Ala	Gln	Leu	Ala	Asp	Arg	Ile	Ile	Gln	Ser	Leu	Gly	Val
385					390					395					400
Ala	Trp	Tyr	Gln	Gln	Lys	Leu	Leu	Ala	Leu	Gly	Phe	Gly	Arg	Lys	Thr
			405						410					415	
Gly	Ile	Glu	Leu	Pro	Ser	Glu	Ala	Ser	Gly	Leu	Val	Pro	Ser	Pro	His
			420					425					430		

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Arg Phe His Ile Asn Gly Ser Leu Glu Trp Ser Leu Ser Thr Pro Tyr
 435 440 445
 Ser Leu Ala Met Gly Tyr Asn Ile Leu Ala Thr Gly Ile Gln Met Val
 450 455 460
 Gln Ala Tyr Ala Ile Leu Ala Asn Gly Gly Tyr Ala Val Arg Pro Thr
 465 470 475 480
 Leu Val Lys Lys Ile Val Ser Ala Ser Gly Glu Glu Tyr His Leu Pro
 485 490 495
 Thr Lys Glu Lys Thr Arg Leu Phe Ser Glu Glu Ile Thr Arg Glu Val
 500 505 510
 Val Arg Ala Met Arg Phe Thr Thr Leu Pro Gly Gly Ser Gly Phe Arg
 515 520 525
 Ala Ser Pro Lys His His Ser Ser Ala Gly Lys Thr Gly Thr Thr Glu
 530 535 540
 Lys Met Ile His Gly Lys Tyr Asp Lys Arg Arg His Ile Ala Ser Phe
 545 550 555 560
 Ile Gly Phe Thr Pro Val Glu Ser Ser Glu Gly Asn Phe Pro Pro Leu
 565 570 575
 Val Met Leu Val Ser Ile Asp Asp Pro Glu Tyr Gly Leu Arg Ala Asp
 580 585 590
 Gly Thr Lys Asn Tyr Met Gly Gly Arg Cys Ala Ala Pro Ile Phe Ser
 595 600 605
 Arg Val Ala Asp Arg Thr Leu Leu Tyr Leu Gly Ile Leu Pro Asp Lys
 610 615 620
 Lys Leu Arg Asn Cys Asp Glu Glu Ala Ala Leu Lys Arg Leu Tyr
 625 630 635 640
 Glu Glu Trp Asn Arg Ser Pro Lys Gln Gly Gly Thr Arg
 645 650

<210> SEQ ID NO 68

<211> LENGTH: 819

<212> TYPE: DNA

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 68

```

atgaaaaaaa aattatcatt acttgtaggt ttaatttttg ttttgagttc ttgccataag      60
gaagatgctc agaataaaat acgtattgta gccagtcgga cacctcatgc ggaattattg      120
gagagtttac aggaagaggc taaagatcct ggaatcaagc tgaaaatact tccagtagat      180
gattatcgta ttctaatcgc ttgtcttttg gataaacaag tagatgcaaa ttactttcaa      240
catcaagcct ttcttgatga cgaatgcgag cgttatgatt gtaaggggtga attagttggt      300
atcgctaaag ttcatttgga acctcaagca atttattcta agaaacattc ttctttagag      360
cgcttaaaaa gccagaagaa actgactata gogattcctg tggatcgtag gaatgctcag      420
cgtgctctac acttggttaga agagtgcgga ctcatgtgtt gcaaagggcc tgctaattta      480
aatatgacag ctaaagatgt ctgtgggaaa gaaaatagaa gtatcaacat attagaggtg      540
tcagctcctc ttctgtcggg atctcttcct gacgttgatg ctgctgtcat tcctggaaat      600
tttgctatag cagcaaacct ttctccaaag aaagatagtc tttgtttaga ggatctttcg      660
gtatctaagt atacaaacct tggtgtcatt cggtctgaag acgtagggtc tcctaaaatg      720
ataaaattac agaagctgtt tcaatctcct tctgtacaac atttttttga tacaaaatat      780

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catgggaata ttttgacaat gactcaagac aatgggttag

819

<210> SEQ ID NO 69

<211> LENGTH: 272

<212> TYPE: PRT

<213> ORGANISM: Chlamydia psittaci

<400> SEQUENCE: 69

```

Met  Lys Lys Lys Leu Ser Leu Leu Val Gly Leu Ile Phe Val Leu Ser
 1      5      10      15

Ser  Cys His Lys Glu Asp Ala Gln Asn Lys Ile Arg Ile Val Ala Ser
      20      25      30

Pro  Thr Pro His Ala Glu Leu Leu Glu Ser Leu Gln Glu Glu Ala Lys
      35      40      45

Asp  Leu Gly Ile Lys Leu Lys Ile Leu Pro Val Asp Asp Tyr Arg Ile
      50      55      60

Pro  Asn Arg Leu Leu Leu Asp Lys Gln Val Asp Ala Asn Tyr Phe Gln
      65      70      75      80

His  Gln Ala Phe Leu Asp Asp Glu Cys Glu Arg Tyr Asp Cys Lys Gly
      85      90      95

Glu  Leu Val Val Ile Ala Lys Val His Leu Glu Pro Gln Ala Ile Tyr
      100     105     110

Ser  Lys Lys His Ser Ser Leu Glu Arg Leu Lys Ser Gln Lys Lys Leu
      115     120     125

Thr  Ile Ala Ile Pro Val Asp Arg Thr Asn Ala Gln Arg Ala Leu His
      130     135     140

Leu  Leu Glu Glu Cys Gly Leu Ile Val Cys Lys Gly Pro Ala Asn Leu
      145     150     155     160

Asn  Met Thr Ala Lys Asp Val Cys Gly Lys Glu Asn Arg Ser Ile Asn
      165     170     175

Ile  Leu Glu Val Ser Ala Pro Leu Leu Val Gly Ser Leu Pro Asp Val
      180     185     190

Asp  Ala Ala Val Ile Pro Gly Asn Phe Ala Ile Ala Ala Asn Leu Ser
      195     200     205

Pro  Lys Lys Asp Ser Leu Cys Leu Glu Asp Leu Ser Val Ser Lys Tyr
      210     215     220

Thr  Asn Leu Val Val Ile Arg Ser Glu Asp Val Gly Ser Pro Lys Met
      225     230     235     240

Ile  Lys Leu Gln Lys Leu Phe Gln Ser Pro Ser Val Gln His Phe Phe
      245     250     255

Asp  Thr Lys Tyr His Gly Asn Ile Leu Thr Met Thr Gln Asp Asn Gly
      260     265     270

```

1. A vaccine comprising:

(a) a pharmaceutically acceptable carrier, and

(b) at least one polynucleotide having a *Chlamydia* sequence.

2. The vaccine of claim 1, wherein the at least one polynucleotide has a *Chlamydia psittaci* sequence.

3. The vaccine of claim 1, wherein the at least one polynucleotide has a *Chlamydia pneumoniae* sequence.

4. The vaccine of claim 1, wherein the at least one polynucleotide has a sequence isolated from a *Chlamydia psittaci* genomic DNA expression library.

5. The vaccine of claim 4, wherein the at least one polynucleotide has a sequence isolated from a *Chlamydia psittaci* genomic DNA expression library.

6. The vaccine of claim 1, wherein the at least one polynucleotide has a sequence of SEQ ID NO: 6, SEQ ID NO: 8, SEQ ID NO: 10, SEQ ID NO: 12, SEQ ID NO: 14, SEQ ID NO: 16, SEQ ID NO: 18, SEQ ID NO: 20, SEQ ID

NO: 22, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 28, SEQ ID NO: 30, SEQ ID NO: 32, SEQ ID NO: 34, SEQ ID NO: 36, SEQ ID NO: 38, SEQ ID NO: 40, SEQ ID NO: 42, SEQ ID NO: 44, SEQ ID NO: 46, SEQ ID NO: 48, SEQ ID NO: 50, SEQ ID NO: 52, SEQ ID NO: 54, SEQ ID NO: 56, SEQ ID NO: 58, SEQ ID NO: 60, or fragment thereof.

7. The vaccine of claim 6, wherein the at least one polynucleotide has a sequence of SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, or SEQ ID NO:26, or fragment thereof.

8. The vaccine of claim 6, wherein the at least one polynucleotide has a sequence of SEQ ID NO:6, SEQ ID NO:10, SEQ ID NO:14, SEQ ID NO:20, or SEQ ID NO:24, or fragment thereof.

9. The vaccine of claim 1, wherein at least one polynucleotide has a sequence of SEQ ID NO:62, SEQ ID NO:64, SEQ ID NO:66, or SEQ ID NO:68 or fragment thereof.

10. The vaccine of claim 1, comprising a polynucleotide encoding an antigen having a sequence of SEQ ID NO:7, SEQ ID NO:7, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, or SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31, SEQ ID NO:33, SEQ ID NO:35, SEQ ID NO:37, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:43, or SEQ ID NO:45, SEQ ID NO:47, SEQ ID NO:49, SEQ ID NO:51, SEQ ID NO:53, SEQ ID NO:55, SEQ ID NO:57, SEQ ID NO:59, SEQ ID NO:61, or an antigenic fragment thereof.

11. The vaccine of claim 1, comprising a polynucleotide encoding an antigen having a sequence of SEQ ID NO:63, SEQ ID NO:65, SEQ ID NO:67, or SEQ ID NO:69, or an antigenic fragment thereof.

12. The vaccine of claim 1, wherein the polynucleotide is comprised in generic immunization vector.

13. The vaccine of claim 12, wherein the vector comprises a gene encoding a mouse ubiquitin fusion polypeptide.

14. The vaccine of claim 12, wherein the vector comprises a promoter operable in eukaryotic cells.

15. The vaccine of claim 14, wherein the promoter is a CMV promoter.

16. The vaccine of claim 1, wherein the polynucleotide is cloned into a viral expression vector.

17. The vaccine of claim 16, wherein the viral expression is selected from the group consisting of adenovirus, adeno-associated virus, retrovirus and herpes-simplex virus.

18. The vaccine of claim 1, comprising at least a first polynucleotide having a *Chlamydia psittaci* sequence and a second polynucleotide having a sequence, wherein the first polynucleotide and the second polynucleotide have different sequences.

19. The vaccine of claim 6, wherein the first polynucleotide has a sequence of SEQ ID NO: 50.

20. A vaccine comprising:

(a) a pharmaceutically acceptable carrier; and

(b) at least one *Chlamydia* antigen.

21. The vaccine of claim 20, wherein the at least one *Chlamydia* antigen has a sequence of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO: 15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31, SEQ ID NO:33, SEQ ID NO:35, SEQ ID NO:37, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:43, SEQ ID NO:45, SEQ ID NO:47, SEQ ID NO:49, SEQ ID NO:51, SEQ ID NO:53, SEQ ID NO:55, SEQ ID NO:57, SEQ ID NO:59, or SEQ ID NO:61, or an antigenic fragment thereof.

22. The vaccine of claim 20, where the at least one *Chlamydia* antigen has a sequence of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO: 15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, or SEQ ID NO:27, or an antigenic fragment thereof.

23. The vaccine of claim 20, wherein the at least one *Chlamydia* antigen has a sequence of SEQ ID NO:7, SEQ ID NO:11, SEQ ID NO:15, SEQ ID NO:21, or SEQ ID NO:25, or an antigenic fragment thereof.

24. The vaccine of claim 20, wherein the at least one *Chlamydia* antigen has a sequence of SEQ ID NO:63, SEQ ID NO:65, SEQ ID NO:67, or SEQ ID NO:69, or an antigenic fragment thereof.

25-73. (canceled)

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