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Recombinant Antibodies for Biological Warfare Detection

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Recombinant antibodies to biological warfare threat agents including *F. tularensis*, *Y. pestis*, *Brucella spp.*, *V. cholerae* O1 and O139, ricin, staphylococcal enterotoxins B and C, botulinum toxins A, B, and E and cholera toxin have been developed through the use of phage display technology. Both recombinant scFvs and Fabs have been produced. Substitutions of currently available monoclonal antibodies with these recombinant antibodies in immunological based detection systems have been successful. The recombinant antibodies exhibited equal sensitivity and equal or lower background across a number of platforms including ELISA assays, ECL based platforms and hand-held immunochromatographic assays. In addition, incorporation of the recombinant antibodies into current detection systems provides a stable genetic source for maintaining critical immunological reagents. The use of recombinant antibodies has allowed for improved detection and identification of biological warfare agents.

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FINAL REPORT

01 October 1996 – 30 September 2000

GRANT #: N0001499AF00001

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INSTITUTION: Naval Medical Research Center, Biological Defense Research Directorate

GRANT TITLE: Recombinant Antibodies for Biological Warfare Detection

AWARD PERIOD: 01 October 1996 – 30 September 2000

OBJECTIVE: To develop high affinity recombinant antibodies to select biological warfare (BW) agents through the use of recombinant DNA and phage display technologies. Recombinant antibodies will be incorporated into current antibody-based detection formats including antigen capture ELISA assays and hand-held immunochromatographic assays.

APPROACH: Two separate phage display systems for the production of recombinant antibody fragments were utilized to develop antibodies to select BW agents. Combinatorial phage display libraries expressing Fabs or scFvs were constructed via recombinant DNA techniques from immune tissue. Panning and screening the libraries isolated antigen specific clones. Expression and purification of recombinant antibody protein from these clones was followed by incorporation in current antibody-based detection platforms.

ACCOMPLISHMENTS: Recombinant antibodies to biological warfare threat agents including *F. tularensis*, *Y. pestis*, *Brucella spp.*, *V. cholerae* O1 and O139, ricin, staphylococcal enterotoxins B and C, botulinum toxins A, B, and E and cholera toxin have been developed through the use of phage display technology. Both recombinant scFvs and Fabs have been produced. Substitutions of currently available monoclonal antibodies with these recombinant antibodies in immunological based detection systems have been successful. The recombinant antibodies exhibited equal sensitivity and equal or lower background across a number of platforms including ELISA assays, ECL based platforms and hand-held immunochromatographic assays. In addition, incorporation of the recombinant antibodies into current detection systems provides a stable genetic source for maintaining critical immunological reagents. The use of recombinant antibodies has allowed for improved detection and identification of biological warfare agents.

CONCLUSIONS: Rapid, reliable and sensitive methods to detect and identify potential biological warfare agents are essential to defend members of the Armed Forces against biological threats. These methods are indispensable in providing prompt medical intervention and ensuring the success of military operations. The development and incorporation of recombinant antibodies in current antibody-based detection platforms has been the focus of our efforts in support of these needs. Recombinant antibodies developed by this breakthrough technology have allowed the DOD community to standardize BW detection reagents. Unlike traditional polyclonal and monoclonal antibodies, recombinant antibodies are maintained in bacteria, offer a stable genetic source, and can be genetically manipulated. Expression and purification of recombinant antibodies by bacterial fermentation is less expensive, easier to perform and less time consuming

than production of monoclonal antibodies through conventional means. In addition, these recombinant antibodies provide a level of reagent purity and consistency that has not been achieved with either polyclonal sera or monoclonal cell culture systems. The recombinant antibody clones provide a stable genetic source for maintaining critical immunological reagents and the use of these antibodies in current detection platforms has resulted in improved detection and identification of BW agents.

SIGNIFICANCE: The overall goal of this research is to develop and improve techniques for the rapid detection of potential biological warfare threat agents. This research is directed towards meeting the Navy medical requirement for improved methods for the rapid identification of potential biological warfare agents to enable timely medical defense and public health intervention. The ability to rapidly identify a specific biological agent responsible for mission-abortive illness on the battlefield or onboard ships will allow earlier therapeutic intervention, and earlier implementation of appropriate medical and protective measures to minimize the spread of infection to other personnel. In addition, the availability of rapid diagnostic assays such as the hand-held immunochromatographic assay will assist forward medical teams to rapidly assess the spectrum of BW agents threatening deployed troops.

PATENT INFORMATION: N/A

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1. Emanuel, P.A., J. Dang, **J.S. Gebhardt**, J. Aldrich, E.A.E. Garber, H. Kulaga, P. Stopa, J.J. Valdes, and **A. Dion-Schultz**. 2000. Recombinant antibodies: a new reagent for biological agent detection. Biosensors and Bioelectronics 14:751-759.