The squalene controversy has been multifaceted, raising such diverse questions as whether the government illegally used an experimental vaccine adjuvant during the Gulf War, whether squalene adjuvants cause the types of symptoms seen in Gulf War veterans, and whether the tests used to detect squalene antibodies are reliable. The Committee reviewed available evidence concerning these and other questions. For the Committee's purposes, however, a single question is primary, that is, whether there is evidence that symptomatic Gulf War veterans have abnormally high levels of circulating squalene antibodies. Despite the attention given to the squalene issue over the past decade by scientists and government agencies, no clear answer to this question has emerged.

Squalene antibodies in ill veterans. Although Dr. Asa is reported to first have broached the subject of adjuvant-induced illness in Gulf War veterans with military officials in 1994,[977,1675] the first study that addressed the issue was published in 2000. In the study, Dr. Asa and colleagues at Tulane University tested for IgG antibodies to squalene in two Gulf War veteran groups, using an assay developed at Tulane. In the first group, blinded analyses were conducted on sera from 38 sick and 12 healthy Gulf War veterans. Symptomatic Gulf War veterans in the study all met the CDC definition of chronic multisymptom illness, but more than a third also had serious diagnosed conditions such as ALS, lupus, and multiple sclerosis.[69] Results indicated that 36 of the 38 sick veterans (95%) tested positive for squalene antibodies, but none of the 12 healthy veterans tested positive. An additional six symptomatic Gulf War era veterans who had received vaccines for deployment, but had not actually deployed, also tested positive.

The second set of analyses were not blinded. The samples tested included 86 Gulf War veterans of undetermined health status and 48 blood donors from the general population. Sixty-nine percent of the Gulf War veterans tested positive for squalene antibodies, but only five percent of the community blood donors were positive. By comparison, few patients from separate groups of lupus patients and breast implant recipients tested positive for squalene antibodies.[69]

In a second paper, Dr. Asa and her Tulane collaborators reported elevated levels of squalene antibodies among military personnel who had received the anthrax vaccine after 1997, as part of DOD's anthrax vaccine immunization program (AVIP). About half of the 25 vaccine recipients in the sample were symptomatic, although case status was not explicitly defined. Thirty-two percent of vaccine recipients tested positive for squalene antibodies, compared to only 16 percent of matched, nonmilitary controls. Further analyses indicated that all individuals with squalene antibodies had received anthrax vaccine from one of five specific lots. Among the 17 individuals who received vaccine from those lots, 76 percent were symptomatic and 47 percent had squalene antibodies. None of the eight veterans vaccinated with other lots were symptomatic or had squalene antibodies.[70]

These studies were reported to have been done with no external funding and, in a number of ways, were not optimally designed.[1572] Samples were small, self-selected, and poorly defined. For example, some Gulf War multisymptom illness cases in the first study had concurrent autoimmune diseases, while some controls had fibromyalgia or chronic fatigue; some findings came from unblinded analyses of sera obtained from poorly characterized patients. Still, study results were intriguing and raised a hypothesis that could be further evaluated in more definitive studies. Identification of an objective test that distinguished a sizable proportion of symptomatic from healthy Gulf War veterans was potentially of great importance as a diagnostic tool, and for providing possible insights concerning pathophysiological processes and even treatments for ill veterans.

Squalene antibody research at Walter Reed Army Institute of Research (WRAIR). After its publication, the Asa/Tulane squalene antibody research was criticized by government scientists and panels.[45,679,1572] Critics questioned the idea that squalene, when injected, acted as an antigen, and whether the assay used had actually detected antibodies to squalene. Within months, however, investigators at Walter Reed Army Institute of Research (WRAIR) published research showing that squalene can act as

an antigen and that antibodies could be detected in a model system.[981] A WRAIR high throughput squalene antibody assay was developed by 2002,[979] and subsequently used to evaluate squalene antibodies in mice and humans. Human blood samples from three populations were evaluated: (1) retired laboratory workers from Fort Detrick who had received multiple vaccines, including AVA, over many years, (2) similarly aged community controls who had never received AVA, and (3) samples from an Army blood center at Fort Knox, which primarily contained serum from young recruits.[980]

Squalene antibodies were detected in all of the groups tested. IgM antibodies were found in about one third of both the Fort Detrick and community cohorts (37%, 32% respectively), but in significantly fewer blood center samples (19%). IgG antibodies were found less frequently in the Fort Detrick and community samples (7% and 15%, respectively), and were not detected at all in the Army blood center samples. Investigators suggested that the prevalence of squalene antibodies increases with age, since the mean age of Fort Detrick volunteers was 68, and the blood center samples were predominantly from individuals 18-21 years of age. This was supported by studies in mice demonstrating a significant increase in circulating squalene antibodies with age.[980]

The WRAIR studies have been extremely useful, developing a well-validated assay and demonstrating that squalene antibodies are detectable in human serum at rates that may increase with age. However, this research did not address the core issue raised by the Asa/Tulane studies concerning Gulf War illness. That is, the WRAIR studies did not measure IgG squalene antibodies in individuals who received vaccines postulated to contain squalene. Nor did they compare squalene antibody levels in symptomatic versus healthy Gulf War veterans. It is important to note, also, that the WRAIR and Asa/Tulane studies provided comparable results concerning "background" rates of squalene IgG antibodies in humans. The WRAIR study identified IgG antibodies in 7-15 percent of individuals in the Fort Detrick and community samples, similar to the 5-16 percent range identified in control populations in the two Asa studies.

Did anthrax vaccine used in the Gulf War contain a squalene adjuvant? Both DOD and NIH have sponsored multiple animal studies and human trials of vaccines with experimental adjuvants containing squalene and squalane, a hydrogenated variant of squalene. This includes DOD-sponsored animal studies of anthrax vaccine formulations with squalene-containing adjuvants begun in 1987, and two small DOD-sponsored human trials of a malaria vaccine that were underway before the Gulf War.[1675]

It has been argued that, given the deadly threat posed by anthrax, the addition of a potent adjuvant to AVA at the time of the Gulf War might have been considered a responsible decision, to enhance the immunogenicity provided by the vaccine in the limited time available in the run up to the war. Facing similar time constraints, British officials opted to use the pertussis vaccine to adjuvanate the U.K. anthrax vaccine. But U.S. government reports have consistently maintained that DOD officials decided against using novel adjuvants with the anthrax vaccine because of restrictions and delays required for FDA licensure of an altered vaccine formulation.[1622,1675]

Still, given the concern surrounding the issue, several investigations were undertaken to determine whether or not there was squalene in the U.S. anthrax vaccine. The Department of Defense commissioned Stanford Research Institute International (SRI) to develop an assay capable of detecting squalene in anthrax vaccine. The initial assay could detect squalene at levels of 140 parts per billion, equal to 70 ng. of squalene per 0.5 ml. AVA dose. SRI then tested samples from 17 lots of AVA, including the five lots identified in the 2002 Asa study as being associated with excess levels of squalene antibodies. No squalene was found in any of the lots tested.[1459]

In 2000, however, U.S. Food and Drug Administration (FDA) officials announced that, using a more sensitive assay, they had detected "trace" amounts of squalene, ranging from 10 to 83 parts per billion, in each of five lots of anthrax vaccine tested, and also in lots of tetanus and diphtheria vaccines.[984] The FDA report verified that manufacturing records did not indicate that squalene had been added to the anthrax

vaccine formulation, and concluded that the low levels detected were likely related to natural occurrence or low-level contamination.[1665] Subsequently, SRI developed a more sensitive assay, capable of identifying squalene at the level of 1 ng. per 0.5 ml. dose of vaccine, and tested samples from 38 lots of AVA, including seven lots reported to have been used during the Gulf War. All lots tested had been produced by MDPH or Bioport, with expiration dates between 1982 and 2001. No squalene was detected in 43 of the 44 lots tested, but an extremely low level was detected in three samples from lot FAV008 (1-9 ng. per ml.).[1458] In contrast to the FDA findings, none of the suspect lots identified in the Asa/Tulane studies were found to contain squalene.

Reports indicate that, when squalene-containing adjuvants are used as part of a vaccine formulation, squalene is present at a concentration of 0.2 – 5.0 percent, or 1-25 mg. in a 0.5 ml. dose of vaccine.[1458,1665] This is more than a million times the level of squalene detected in anthrax vaccines by FDA and SRI testing. Whatever the source of the detected squalene, the FDA and SRI studies support the government's assertion that squalene-containing adjuvants were not added to the tested vaccine lots at levels generally used for adjuvants. Some have speculated that even these very minute levels of squalene, less than the level normally found in human blood, might be capable of stimulating reactions,[977] but no studies have evaluated this contention.

Health effects of squalene. Squalene is an oily substance that naturally occurs in plants and animals. It is found in a variety of foods, lotions, and cosmetics. It is also used as a food supplement and has been postulated to provide therapeutic benefits.[225,787] In humans, squalene is synthesized by the liver as a precursor to cholesterol, and circulates in the blood. Many substances that are ingested or found in the blood, however, cannot be safely injected. Squalene, when injected, stimulates a nonspecific immune response, making it a useful component of vaccine adjuvants.[41,1218] The original theory relating anthrax vaccine and squalene adjuvant to Gulf War illness suggested that veterans' symptoms were autoimmune in nature, that is, they were associated with autoantibodies stimulated by receipt of the vaccine. Associations between autoimmune conditions and vaccines have long been postulated, including vaccines against diptheria, tetanus, polio, and hepatitis B.[1148,1404] In animal models, squalene has been used to induce antibodies and precipitate diseases that simulate human autoimmune conditions, including lupus[847,1356] rheumatoid arthritis,[213,618] and multiple sclerosis.[114]

No squalene-containing adjuvants are licensed for use in the U.S., but a number have been evaluated in clinical studies. These include trials of vaccines for influenza, herpes simplex, and HIV.[92,518,867,972,1675] MF59, an oil-in-water microemulsion, is the most widely used squalene-containing adjuvant. An influenza vaccine containing MF59 has been licensed for use in European countries since 1997 and is considered to have a good safety record.[1217]

A recent collaborative study between Italian and U.S. investigators suggests that receipt of squalene-containing adjuvants may not stimulate elevated levels of squalene antibodies in humans.[328] Using an ELISA squalene antibody assay, investigators identified low-levels of squalene IgG antibodies in 79 percent of a sample of U.S. adults, and 26 percent of European adults who had not received vaccines containing squalene. IgG antibodies to squalene were also detected at low levels in 94-100 percent of European adults 65 years of age and over who participated in influenza vaccine trials, whether or not the vaccine received in the trial contained MF59. Investigators concluded that low levels of circulating squalene antibodies are commonly found in adults, and that squalene antibody levels are not affected by receipt of vaccines containing squalene. Although interesting, these findings raise more questions than they answer. No clear explanation is offered, for example, for the substantial differences in rates of squalene antibody positivity in the three cohorts evaluated. Nor is there any explanation for why results differed so dramatically from the two previous U.S. studies, which found IgG antibodies in fewer than 20 percent of adults studied.[69,980]

Conclusions of special committees. Previous government committees and scientific panels have focused on different aspects of the squalene issue. The theory was sometimes dismissed without detailed consideration, based on DOD assertions that squalene was not added to the anthrax vaccine.[1227,1690] Little published research was available to earlier panels, so conclusions were usually based on commonsense observations. The Presidential Special Oversight Board pointed out, for example, that low levels of squalene had been found in diphtheria and tetanus vaccines, but no "Gulf War syndrome" issues had been raised in relation to these vaccines.[1232] An Institute of Medicine panel pointed out shortcomings in the Asa/Tulane squalene antibody studies, and concluded they did not provide persuasive evidence that squalene antibodies had been detected.[679] The World Health Organization issued a statement on the safety of squalene-containing adjuvants, citing the safety record of the 22 million doses of the Italian influenza vaccine distributed since 1997.[1812]

Other adjuvants in Gulf War vaccines. Adjuvant issues relating to the anthrax vaccine have also made headlines in the U.K. The British MOD acknowledged, in 1997, that pertussis vaccine had been used to adjuvanate the anthrax vaccine given to British Gulf War troops, a decision that has also been controversial.[203,1551,1756] MOD officials have maintained that no vaccines containing squalene were given to British troops. Tests conducted by an independent laboratory in 2001, sponsored by MOD, found no squalene in 11 lots of the U.K. anthrax vaccine, nor in other types of vaccines given to British troops during the Gulf War.[1568]

The U.S. Department of Defense has consistently maintained that all adjuvants in vaccines used during the Gulf War were aluminum-based and FDA approved. Immunizations given to Gulf War veterans that contained aluminum adjuvants included anthrax, BT, and tetanus-diptheria vaccines.[1622] Aluminum adjuvants have been used in vaccines for over 60 years, and are considered to have a good safety record.[711,909] Vaccines containing aluminum adjuvants have been extensively studied in humans and animals for both effectiveness and adverse effects, but very little research has specifically looked at neurological effects of vaccine adjuvants, an area of particular interest in relation to Gulf War illness.

A recent Canadian study evaluated long-term effects of both squalene and aluminum hydroxide adjuvants on behavior and central nervous system tissues in a mouse model.[1202] Using dosages comparable to those used in human vaccines, animals received two injections, two weeks apart, of one of the adjuvants, both adjuvants combined, or placebo. They were then evaluated using a variety of neurobehavioral tests over a six month period, followed by histochemical analyses of brain and spinal cord tissues. Anti-squalene antibodies were found in 20% of animals injected with placebo, 27% of those injected with aluminum, 40% of those injected with squalene, but only 10% of those injected with both adjuvants. Overall, the aluminum adjuvant produced more adverse effects than placebo, squalene, or the combined adjuvants. After six months, mice injected with the aluminum adjuvant exhibited significant declines in muscle strength and endurance, and increased indicators of anxiety, compared to placebo. Aluminum adjuvant was also associated with indicators of increased central nervous system inflammation and motor neuron loss, as reflected by a significant increase (350%) in the number of reactive astrocytes in the lumbar spinal cord and neuronal apoptosis in the motor cortex and spinal cord. Investigators concluded that their findings were consistent with an association between aluminum adjuvants and neurological deficits, including ALS. By contrast, squalene adjuvant was associated with fewer changes in brain and behavior, none of which were statistically significant.

Vaccines containing aluminum hydroxide adjuvant have also been associated with the development of macrophagic myofasciitis.[486] This recently-identified condition is characterized by macrophage infiltration of muscle tissue after receipt of vaccines.[239,485] Patients develop arthromyalgias and fatigue, among other symptoms, with one report indicating that about half of macrophagic myofasciitis patients meet criteria for chronic fatigue syndrome.[76]

Although squalene has been the primary adjuvant issue raised in relation to Gulf War veterans, no studies have specifically linked receipt of squalene-containing adjuvants to biological processes or chronic symptoms that parallel those affecting Gulf War veterans. In contrast, there are preliminary indicators, from both human and animal studies, that aluminum hydroxide adjuvant may be associated with neurological damage and chronic symptoms potentially relevant to the health of Gulf War veterans.

Primary Gulf War squalene antibody question unanswered. Eleven years after the squalene controversy was first publicly raised, studies have addressed several related questions. Two laboratories have assessed levels of squalene in selected lots of the U.S. anthrax vaccine, both yielding results that support government assertions that a squalene-containing adjuvant was not added to those lots. Additional studies have provided insights on detection of antibodies to squalene in humans and animals, but have provided little indication that receipt of squalene-containing adjuvants results in chronic production of squalene antibodies.

The observation from the Asa/Tulane studies that is most relevant to the health of Gulf War veterans, however, has not been further evaluated. Their initial study, reported in 2000, indicated that symptomatic Gulf War veterans had detectable levels of IgG antibodies to squalene, but that healthy veterans did not. This raised a testable hypothesis concerning an objective measure of an immunological abnormality that distinguished ill from healthy veterans. The Asa/Tulane studies may have correctly identified excess rates of squalene antibodies in ill veterans, whether or not they were caused by vaccines, by vaccine contamination, or by clandestine use of an unapproved adjuvant. It is important to determine whether the observed association between squalene antibodies and Gulf War illness is supported, or refuted, by more definitive research.

Health Effects of Receiving Multiple Vaccines

Most studies conducted to establish vaccine safety in humans evaluate vaccines individually. Studies that evaluate receipt of more than one vaccine at the same time are generally concerned with changes in vaccine effectiveness, but may also report on short term adverse effects. The Committee identified little research that provides information on long-term effects of specific vaccine combinations, or numerous vaccines received in a brief time span.[512,675] Receipt of multiple vaccines together is fairly common, however. Multiple immunizations are routinely given to infants and young children. Adults traveling overseas also commonly receive multiple immunizations. Studies have assessed short term side effects related to receipt of vaccine combinations in childhood and have generally reported little or no increase in short-term reactogenicity.[236,1453,1540] In civilians traveling to foreign countries, receipt of multiple vaccines has also been reported as being well-tolerated, although the number who experience acute side effects increases with the number of vaccines received.[156,426] For many years, multiple vaccinations have also been given to new military recruits and to troops preparing for overseas deployment.[1523] Surprisingly little information is available, either from studies or from monitoring programs, that quantifies short or long-term adverse effects resulting from specific combinations of vaccines, or a large number of vaccines received concurrently.

Some insights regarding effects of multiple vaccines received over a prolonged period of time have been provided by a series of follow-up evaluations of laboratory workers at Fort Detrick, Maryland. These workers received multiple immunizations as participants in vaccine studies conducted for the U.S. biological weapons development and countermeasures program between 1943 and 1969.[1211] The first three assessments were conducted in 1958, 1965, and 1974. No abnormal excess of diagnosed diseases affected the workers, but several indicators of a chronic inflammatory process were identified. These included increased rates of leukocytosis and lymphocytosis, alterations in ratios of alpha and beta globulins, and higher mean serum levels of hexosamine.[1189,1190,1770]

The most recent assessment, reported in 2004, evaluated 155 former workers who participated in a Fort Detrick laboratory alumni gathering in 1996, and 265 age and gender-matched controls.[1212] Fort Detrick participants were evaluated an average of 43 years after their first program immunization. Individuals had received different types and numbers of vaccines—a median of 154 each—during their years in the program. For example, 142 of the 155 Fort Detrick workers had received multiple doses of anthrax vaccine, an average of 23 doses per subject. Results indicated no differences in clinically diagnosed diseases between workers and community controls. Fort Detrick workers reported their overall health status to be slightly worse, however, and a higher proportion reported being fatigued. Several serum parameters also differed between the groups. Most significantly, unspecified monoclonal proteins were detected at elevated rates in the Fort Detrick workers. These gammopathies were not further characterized and were not associated with identifiable disease, but have been linked in other studies with the development of serious conditions, including multiple myeloma.[851,852]

The Fort Detrick studies provide an interesting look at the long-term health of selected individuals many years after receiving a large number of vaccines. Overall, the studies indicate that receipt of repeated doses of multiple vaccines over an extended period of time was not associated with identifiable disease, but may produce persistent immune alterations in a subset of individuals. For several reasons, however, these studies have limited relevance for understanding effects of multiple vaccines received for Gulf War deployment. Of the over 3,000 original participants in the Fort Detrick program, those evaluated in follow-up studies were a select group of volunteers. They would not have included, for example, individuals who did not tolerate multiple vaccinations and withdrew from the program, those who had died, or those not healthy enough to attend a social gathering many years after retirement.[1211,1212] Unlike the Fort Detrick program, Gulf War veterans received multiple vaccines over a brief period of time, vaccines that differed from those given to Fort Detrick workers in the 1940s, 1950s, and 1960s.

Animal studies of effects of vaccines combined with other Gulf War exposures. As part of its Gulf War research effort, the British Ministry of Defence (MOD) sponsored a series of studies that evaluated effects of multiple vaccines given to U.K. military personnel in the Gulf War, combined with pyridostigmine bromide (PB), in several animal models. A number of abnormalities resulting from these exposures were identified, which differed according to the study design and animal model used. Relatively high-dose combinations of anthrax and pertussis vaccines produced observable illness and weight loss in mice, with milder effects associated with more dilute vaccines. Toxicity effects in the mouse model were attributed mainly to the pertussis vaccine.[1285] A separate study found that PB, given at levels comparable to those used in the Gulf War, had no effect on humoral immunity in mice.[535] A second series of experiments evaluated effects of 10 vaccines, given at various doses, alone and with PB, in a guinea pig model. After 72 days, all animals appeared generally healthy. The only observable effect was slight weight loss in the animals who received the highest-dose vaccine regimen.

The most comprehensive series of studies evaluated diverse health parameters in the marmoset, a small primate, following receipt of vaccines and/or PB. Evaluations included effects on general health, cognitive function, muscle function, sleep patterns, electroencephalograms (EEGs), immune function, and adrenal function. Animals received one-fifth the human dose of all 10 vaccines, along with boosters, over a 51 day period, and were infused for 28 days with PB at the dose required to reduce serum cholinesterase levels by 30 percent. Outcomes were monitored over a 21 month period. The marmosets exhibited no obvious behavioral or health changes over that time, and no differences in weight or muscle function.

Animals that received PB and/or vaccines, however, had significantly higher error rates on two of the eight measures of cognitive function (new learning and compound reversal) that lessened over the period of observation.[1486] Animals treated with PB exhibited significantly reduced EEG alpha wave activity early in the study, and reduced beta 2 waves at various times over the observation period. Pyridostigmine bromide was also associated with reduced levels of rapid eye movement (REM) sleep and fewer REM periods. In contrast, animals who received multiple vaccines, had fewer waking periods early in the

observation period, with improved sleep efficiency and more REM periods.[1789] Urinary cortisol levels did not vary by treatment group and there were no indicators of compromised immunity following treatment with multiple vaccines and/or PB.[632]

Overall, the U.K. studies found no interactive or synergistic effects between PB and receipt of multiple vaccines on any of the parameters studied. Individually, receipt of multiple vaccines, as well as PB, produced a limited number of significant neurobehavioral effects. Both also produced significant, but different, effects on sleep patterns. But neither PB nor multiple vaccines had detectable effects on muscle function, peripheral immune response, or cortisol levels.

Results from the marmoset studies have parallel observations in Gulf War veterans—both in what was found and in what was not found. A large British epidemiologic study reported no significant interaction between PB and receipt of multiple vaccinations in relation to Gulf War illness.[1698] Gulf War veterans have also been reported to be similar to controls with respect to resting cortisol levels,[502] and *in vitro* tests of cellular and humoral immunity.[422] Gulf War veterans commonly report sleep and cognitive difficulties, and studies have identified several domains of measurable cognitive impairment in subsets of symptomatic veterans.[1709,1779]

As will be described in a separate section, the Committee has reviewed evidence suggesting that immune alterations related to veterans' persistent symptoms might more likely be found in the brain than in the peripheral circulation. A study conducted by investigators at Boston University School of Medicine provides a preliminary indication that combined exposure to vaccines, stress, and PB may affect brain processes associated with central immune activation and inflammation.[917,1752] Immune stimulation by KLH, used as a vaccine analog in the Boston study, significantly enhanced and prolonged the production of stress-activated kinases regionally in the mouse brain. This effect was further enhanced and prolonged by PB. Investigators suggested that vaccines and PB may act synergistically to dysregulate processes normally associated with immune activation and inflammation in the brain, processes that mediate neuronal damage following exposure to stress and toxic chemicals.

Studies Evaluating the Health of Gulf War Veterans in Relation to Vaccines

Association of Gulf War illness with individual vaccines. As detailed in Appendix A-12a, Gulf War studies have frequently reported significant associations between Gulf War illness and receipt of individual vaccines (e.g., BT, meningococcal, anthrax, plague, typhoid) using analyses that did not take into account effects of other exposures in theater. This includes novel findings from a large study of British Gulf War veterans that tetanus and cholera vaccines were not associated with Gulf War illness if veterans received them before deployment, but were problematic for veterans who received them during deployment.[641] It is not possible to reliably interpret these findings, however, in light of confounding potentially introduced by concurrent exposures, as previously described.

Only two Gulf War studies have assessed effects of individual vaccines while adjusting for the effects of other exposures in theater. The large U.S. study of Navy Seabees queried veterans about their receipt of five vaccines and immune globulin. All were significantly associated with Gulf War illness in unadjusted analyses. After controlling for effects of other exposures, however, only meningococcal vaccine was associated with Gulf War illness, presenting a significant, but only slightly elevated risk (OR = 1.3).[527] In the Fort Devens cohort, receipt of the anthrax vaccine was a significant risk factor for Gulf War illness after adjusting for other exposures in theater. The increased risk for anthrax vaccine was also modest (OR = 1.5) and the study did not assess contributions of other vaccines.[1804]

Health effects of anthrax vaccine in Gulf War veterans. Although it has often been suggested that anthrax vaccine is a cause of Gulf War illness there is relatively little reliable evidence to support this

view. No efforts were made during the Gulf War to monitor short or long-term health problems following receipt of AVA. Epidemiologic studies have generally not identified the anthrax vaccine to be a prominent risk factor for Gulf War illness. As indicated, anthrax vaccine has been associated with increased rates of symptoms, Gulf War illness, and poor health status in several studies, using analyses that did not take into account effects of other exposures in theater.[161,511,1371,1374,1698] The magnitude of risk identified for the anthrax vaccine in these studies, however, was similar to that for most other vaccines, and lower than for other types of exposures in theater. But more informative research on this issue is extremely limited. Only two studies have evaluated the association of anthrax vaccine with Gulf War illness, adjusting for effects of other exposures. Anthrax vaccine was identified as a significant, albeit modest, risk factor in one of those studies.[1804]

There has been some concern that, since many Gulf War veterans might not have known if they received the anthrax vaccine, inaccurate reporting could markedly affect studies evaluating effects of the anthrax vaccine. Two studies have compared health outcomes in Gulf War veterans with self-reported versus documented receipt of the vaccine. In a large study of U.K. Gulf War veterans, associations between anthrax vaccine and health outcomes were similar in individuals who did and did not have their vaccine records, as well as for anthrax vaccine received prior to and after deployment.[641,1698] In all subgroups, receipt of the anthrax vaccine was associated with a 1.3 to 1.5 times greater risk of Gulf War illness, with no adjustments made for other types of exposures in theater.

The U.S. DOD has identified over 7,000 Gulf War veterans who are known to have received the anthrax vaccine, based on available documents. Investigators from the Washington, DC, VAMC identified 352 of those individuals among the 11,441 Gulf War veterans previously interviewed for the U.S. national survey of Gulf War veterans.[957] Gulf War veterans documented to have received the anthrax vaccine reported a number of medical conditions and symptoms at higher rates than veterans who said they did not receive the anthrax vaccine. These included significantly higher rates of dermatitis, gastritis, diarrhea, joint pain, fatigue, mood changes, sleep abnormalities, and indigestion. A still greater number of symptoms and health problems were significantly associated with self-reported, but undocumented, receipt of the anthrax vaccine. These results indicate that, while the anthrax vaccine is potentially associated with excess symptoms in Gulf War veterans, self-reported data introduced a bias that led to an overestimate of the vaccine's adverse effects.

Gulf War illness and receipt of multiple vaccines. An additional vaccine-related question of importance is whether receipt of multiple vaccinations together, rather than any single vaccine alone, contributed to the development of Gulf War illness. This issue has been of particular interest in the U.K and has been investigated in studies of British and Australian Gulf War veterans, but not U.S. veterans. In 1997, Professors Rook and Zumla of University College in London hypothesized that receipt of multiple vaccines for the Gulf War could have precipitated an immunological shift that resulted in an unbalanced production of Th2-type cytokines (associated with humoral immunity) relative to Th1-type cytokines (associated with cell-mediated immunity).[1306] They suggested that this shift may have resulted from the many TH2-inducing vaccines given to Gulf War personnel, exacerbated by stress and perhaps also by pesticide exposures. This idea, referred to as the Rook hypothesis, provided a testable theory for explaining veterans' diverse symptoms.

In 1999, investigators from King's College in London reported that Gulf War veterans who received the largest number of vaccines for the war had significantly worse health, on multiple measures, than veterans who received fewer vaccines.[1698] Additional analyses among the 923 study veterans with vaccination records appeared, initially, to indicate that receipt of multiple vaccines *before* deployment was not problematic. However, veterans who received five or more vaccines *during* deployment had a significantly elevated rate of Gulf War illness (OR = 5).[641] Commentators identified possible explanations for this difference, pointing out, for example, that the types of vaccines received in theater

differed from those administered prior to deployment.[153,638,679] The study investigators suggested that the difference was attributable to receiving multiple vaccines in conjunction with the stress of deployment.[641]

Further analyses, however, indicated that receiving multiple vaccines in theater was not more problematic than multiple vaccines received before deployment. In response to suggestions from scientific colleagues, the King's College investigators revised their analytic approach and found no significant differences between effects of multiple vaccines administered before and during deployment.[638] Their final conclusion, then, was that their data supported an overall association between multiple vaccines and ill health in Gulf War veterans that was not specific to post-deployment vaccines.[638] A study of Australian Gulf War veterans also reported higher symptom rates among those who received the largest number of immunizations for the Gulf War.[789] Results of both the King's College and Australian studies are difficult to interpret, however, since neither study assessed effects of multiple vaccines, adjusted for other types of exposures in theater.

A second British study provides a more informative look at this issue. Controlling for effects of multiple exposures during deployment, investigators at the University of Manchester reported that the number of inoculations received by British Gulf War veterans was significantly correlated with overall symptom severity, and with symptoms of peripheral neuropathy.[241] The Manchester study also indicated that there were no differences between effects of vaccines received prior to and during deployment. No specific information was provided, however, on types of vaccines or vaccine combinations linked to veterans' ill health.

There are two components of the original Rook hypothesis. The first is that receipt of multiple vaccinations contributed to veterans' persistent symptoms after the war. This has not been evaluated in U.S. veterans, but is supported by one well-analyzed study of British veterans, with suggestive evidence provided by two additional studies. The other component of the Rook hypothesis is that veterans' ill health resulted from a Th1-Th2 shift in cytokine production. Several studies have tested this directly by assessing Th1 and Th2-related immune parameters in Gulf War veterans.[1420,1835] As will be described in more detail in a later section of the report, findings have not supported a bias towards production of Th2-type cytokines in ill Gulf War veterans. It is not possible to know if such a shift occurred, temporarily, at the time of the war, but a Th1-Th2 shift is not evident in veterans evaluated years after their return from theater.[1182]

Other health effects of vaccines received during the Gulf War. Very few other Gulf War-related health outcomes have been assessed in relation to vaccines. Two studies have identified significant associations between acute adverse reactions to vaccines received for deployment and poor health outcomes after the war.[1374,1698] Military hospitalization records indicate that there were 58 hospitalizations in theater for adverse reactions to vaccines. The largest number of cases attributed to one vaccine was tetanus. One hospital admission, for seizures, was attributed to receipt of the anthrax vaccine.[1622] A report on U.S. military personnel who participated in DOD's Gulf War registry program, the CCEP, indicated that veterans who reported receiving BT, but not anthrax, had a significantly higher rate of hospitalization after the war.[1435] In studies of British Gulf War veterans, cancer rates were not associated with receiving biological warfare vaccines (anthrax, pertussis, plague).[943] Self-reported receipt of the anthrax vaccine was associated with a small, nonsignificant, increase in overall mortality among British veterans (mortality rate ratio = 1.2).[944]

Vaccines and the health of veterans who did not serve in the Gulf War. Epidemiologic studies have generally assessed Gulf War-related health outcomes by comparing the health of Gulf War veterans to personnel who served in the military during the war, but did not deploy to the Gulf War theater. A potential problem of using nondeployed Gulf War era veterans as a comparison group is that they may have received some or all of the vaccines given to Gulf War veterans. There are several reports

of military personnel with symptoms that resemble Gulf War illness who received vaccines in preparation for service in the Gulf War, but did not actually deploy.[69,918,1684]

Very little research has assessed health problems in relation to vaccines received by Gulf War-era veterans who did not serve in the Gulf War. The Kansas study asked nondeployed Gulf War-era veterans if they had received any vaccines during the time of the Gulf War. Nondeployed veterans who reported getting vaccines during that time had significantly higher rates of symptoms in several domains (chronic somatic pain, neurological, and gastrointestinal problems) and a nearly four-fold higher rate of Gulf War illness than nondeployed veterans who did not receive vaccines. Veterans who served in theater, by comparison, had Gulf War illness symptoms at 11 times the rate of nondeployed veterans who did not receive vaccines.[1476] These findings provide support for the idea that military vaccines contributed to the development of chronic symptoms in Gulf War era veterans. But the findings are preliminary and are nonspecific, that is, no information is provided on the types or number of vaccines received by nondeployed personnel.

Findings from the King's College study of U.K. veterans may also have relevance to this issue. The study compared the health of U.K. Gulf War veterans with Bosnia veterans, and asked both groups about vaccines they had received. Few Bosnia veterans reported receiving biological warfare vaccines: anthrax, pertussis, or plague. The total number of vaccines received was significantly associated with multisymptom illness in Gulf War veterans, but not in Bosnia veterans.[1698] Information provided did not allow for a clear interpretation of these findings, however. They could indicate that the multiple vaccine effect observed in British Gulf War veterans is related to specific vaccines given to Gulf War, but not Bosnia, personnel. Alternatively, they could indicate that the multiple vaccine effect resulted from confounding by other risk factors associated with Gulf War service. For example, the large Manchester study of British Gulf War veterans found that the number of vaccines received by Gulf War veterans was significantly correlated with other exposures in theater, such as the number of days PB was used.[241]

Accuracy of self-reported vaccine data. Most Gulf War studies have assessed veterans' health in relation to vaccines based on veterans' own reports of immunizations they received for the war. Self-reported information on the number and types of vaccines received for Gulf War deployment is potentially more problematic than for some other self-reported exposures. In addition to usual problems related to accurate recall, veterans might not have known what vaccines they received at the time they received them, as has been reported for anthrax and BT vaccines. Several studies have provided useful insights related to the accuracy of vaccine reporting by veterans. Department of Veterans Affairs investigators were able to evaluate the accuracy of veterans' self-reported receipt of the anthrax vaccine in 352 veterans interviewed for the U.S. national Gulf War survey. When questioned, three-fourths of the 352 veterans with DOD-documented receipt of the anthrax vaccine reported that they had, in fact, received it. Only 10 percent reported they did not receive the anthrax vaccine and 16 percent didn't know.[957]

Two studies of U.K. Gulf War veterans found that veterans who used their shot records in reporting immunizations tended to report more vaccines than veterans who did not have shot records.[241,1698] But misreporting and underreporting of vaccines appears to have had little effect on health findings related to vaccines. Both large national studies of British Gulf War veterans found that associations between vaccines and health outcomes were similar in veterans who did have vaccine records compared to veterans who did not have their records.[241,1698]

Summary. Vaccines and Gulf War illness. Gulf War veterans received multiple immunizations for deployment. These included the anthrax vaccine, which was given to a large number of military personnel for the first time during the Gulf War. Diverse issues have been raised in relation to the anthrax vaccine's potential for causing adverse health effects. Due to changes in production methods and quality control measures between 1990 and 2001, it is not known if the safety profile of the anthrax vaccine in

current use is the same as that of the vaccine given to Gulf War personnel. Recent studies have indicated that the current anthrax vaccine is associated with high rates of acute adverse reactions, particularly in women. No information is available on rates of persistent symptoms or multisymptom illness following receipt of the anthrax vaccine. Studies have not identified excess hospitalizations or outpatient visits for diagnosed diseases in the weeks and months following receipt of the vaccine. Limitations in the types of information provided by these studies, however, indicate a continued need for long-term follow up, to determine whether excess rates of diagnosed or undiagnosed conditions occur in anthrax vaccine recipients.

An excess of circulating antibodies to the natural substance squalene was reported in symptomatic Gulf War veterans in 2000, and investigators suggested this could have been caused by an unapproved vaccine adjuvant in the anthrax vaccine. Testing of potentially suspect vaccine lots by two laboratories identified only trace amounts of squalene, far below levels usually used for vaccine adjuvants. The observed association between Gulf War illness and elevated levels of squalene antibodies was not contingent on anthrax vaccine being the source of this abnormality, however, and has not yet been independently evaluated.

Gulf War epidemiologic studies have not identified any individual vaccine, including the anthrax vaccine, to be a prominent risk factor for Gulf War illness. Several studies have provided indications that personnel who received a larger number of vaccines for deployment have had higher rates of persistent symptoms since the war. Few Gulf War studies have adequately analyzed data collected in relation to vaccines received for deployment, however, to determine whether individual vaccines or combinations of vaccines are independent risk factors for persistent health problems in Gulf War veterans.

Recommendations

Diverse concerns have been raised in relation to vaccines received for the Gulf War, but relatively little reliable information has implicated individual vaccines as prominent risk factors for Gulf War illness. Several issues related to vaccines received by Gulf War veterans have not been adequately addressed by existing research. These include the need for more thorough evaluation of vaccines as risk factors for chronic health problems in epidemiologic studies, a definitive study to conclusively evaluate the previously-observed association between squalene antibodies and Gulf War illness, and the need for longer-term evaluation of symptoms and diagnosed diseases following receipt of the anthrax vaccine.

The Committee therefore recommends the following research:

- In previously-conducted and future epidemiologic studies of Gulf War veterans, analyze associations between Gulf War illness and individual vaccines, combinations of vaccines, and total number of vaccines received using methods that control for potential confounding by other Gulf War-related exposures.

- Commission a case-control study to provide clear answers concerning possible associations between Gulf War illness and squalene antibodies. The study should, at minimum, analyze blinded samples from well-characterized symptomatic and healthy Gulf War veterans for the presence of squalene antibodies using each of the assays developed for this purpose. It should also assess whether there is an identifiable link between levels of squalene antibodies in ill Gulf War veterans and receipt of the anthrax vaccine or vaccines more generally. The project should be organized and overseen by qualified investigators not affiliated with the federal government or civilian scientists whose initial work raised the squalene issue in relation to Gulf War illness.

- Evaluate the association of anthrax vaccine adsorbed (AVA) with chronic symptoms, Gulf War illness, and diagnosed diseases in personnel known to have received the anthrax vaccine during the Gulf War. These health outcomes should also be assessed at least five years after vaccination in deployment and era subgroups of personnel in the Millenium Cohort study as well as other groups vaccinated in association with the military's anthrax vaccine immunization program and federal anthrax vaccine trials.

Cholinergic and Related Neurotoxicants: Pyridostigmine Bromide, Pesticides, and Nerve Agents

> Vesser [Acting Special Assistant to the Secretary of Defense for Gulf War illnesses LTG Dale Vesser] remarked that although Saddam Hussein didn't use nuclear, biological, or chemical agents against coalition forces during the war, 'it never dawned on us … that we may have done it to ourselves.'
>
> … 'We know that at least 40,000 American troops may have been overexposed to pesticides,' Vesser said, adding that more than 250,000 American troops took the small, white pyridostigmine bromide pills. …. Both of these substances may cause symptoms that are consistent with the symptoms that some Gulf War veterans have.'
>
> --Armed Forces Press Service, 2001[491]

Many classes of chemicals are neurotoxicants, that is, exposure to these compounds can have adverse biological and physical effects on the nervous system. Three types of neurotoxicant exposures encountered by Gulf War military personnel during deployment are chemically related. They include chemical nerve agents, many of the pesticides used during the Gulf War, and pyridostigmine bromide (PB), the drug given to troops as a protective measure in the event of nerve gas attack. In its 2004 report, the Committee provided an overview of information concerning veterans' exposures to these toxicants, what was known about their health effects, and what had been learned from studies of Gulf War veterans. The report concluded that available evidence supported a probable link between Gulf War illness and exposure to these compounds.

Chemical nerve agents, PB, and many of the pesticides to which Gulf War veterans were exposed belong to a class of chemicals known as acetylcholinesterase (AChE) inhibitors. They share a common toxic mechanism of action, that is, they inactivate the enzyme AChE, which is essential for breaking down the nerve signaling chemical (or neurotransmitter) acetylcholine. Inhibition of AChE leads to the buildup of acetylcholine in the brain and peripheral nerve endings, and over stimulation of cholinergic nerve receptors. Acetylcholinesterase-inhibiting medications and pesticides can be used safely at recommended levels. Adverse effects can occur with excessive exposure, and are also seen at lower doses in individuals who are particularly sensitive to these compounds.

The acute symptoms of excess exposure to AChE inhibitors relate to the different types of cholinergic receptors affected by acetylcholine buildup. Excess cholinergic stimulation of muscarinic receptors of the parasympathetic autonomic nervous system results in increased salivation and respiratory secretions, nausea, abdominal cramping, diarrhea, and excess sweating. Effects on autonomic nicotinic receptors include increased heart rate and blood pressure. Excess stimulation of nicotinic receptors in skeletal muscles leads to muscle twitching, cramps, weakness, tremors, and paralysis. Excess stimulation of acetylcholine receptors in the brain produces fatigue, mental confusion, headache, poor concentration and general weakness and, at higher exposures, convulsions and coma.[387] At sufficient doses, exposure to AChE inhibiting chemicals can result in respiratory arrest and death.

This section of the report provides information on what is known about Gulf War veterans' exposure to these chemicals, what is known about their health effects overall, and what has been learned about their effects from studies of Gulf War veterans. It also includes information on additional pesticides of concern that are not AChE inhibitors and information from research on effects of exposure to combinations of PB, pesticides, and nerve agents.

Exposure to Cholinergic and Related Neurotoxicants During Gulf War Deployment

> My unit arrived in the Gulf the day before the air war started. We first spent about a month in Dhahran in Saudi Arabia. Our chemical alarms went off several times during that month, and we had to go to MOPP-level four, which meant we had to put on chemical suits, masks, gloves, and boots. While we were still in Dhahran, we started taking pyridostigmine bromide pills, which were supposed to protect us against exposures to nerve gas. About three days after I started taking the pills, my eyes were jittery, my vision was jumping, and I was seeing double, and I was nauseated. By the fourth day, I was vomiting a little blood, so I went to sick-call. They told me to cut the dose in half and said there was nothing to worry about. At least I no longer vomited blood after I reduced the dosage. Many other people in the unit reported having similar vision problems.
>
> --SSgt PB, Gulf War veteran[716]

Military personnel serving in the 1990-1991 Gulf War were exposed to a variety of substances that have the potential to adversely affect the central nervous system. These include multiple types of anticholinesterase compounds—pyridostigmine bromide (PB) pills, pesticides, and for some veterans, low-level exposure to chemical nerve agents. But not all personnel were exposed to the same compounds at the same dosages and in the same combinations. Although records are not available that document individual exposures to these compounds, government investigations have provided considerable information on the extent and patterns of use of PB and pesticides, and have modeled nerve agent exposures in relation to the largest verified nerve agent release incident.

Table 1. Veteran-Reported Exposures to Neurotoxicants During Gulf War Deployment

	U.S. National Survey[751]	*U.S. Army Veterans*[1804]	*U.S. Navy Seabees*[524]	*U.K. National Survey*[1698]
Took pyridostigmine bromide pills	49 %	66 %	33 %	82 %
Used personal pesticides	48 %	46 %	35 %	69 %
Exposed to nerve gas/chemical agents	10 %	19 %	3 %	9 %

Population-based surveys of Gulf War veterans have also provided consistent information on veteran-reported exposures during deployment. Table 1 summarizes responses to survey questions from studies of Gulf War veterans in the U.S. and U.K. concerning their use of PB and pesticides, and whether they thought they were exposed to chemical weapons. About half of all U.S. Gulf War veterans, and a higher proportion of U.K. Gulf War veterans, report using PB and pesticides during deployment. Nearly two out of three Gulf War veterans in the U.S. national survey reported that they had heard chemical alarms sound or put on their MOPP gear (mission oriented protective posture, protective garments worn in a possible chemical event) during deployment,[751] but only 10 percent believed that they were exposed to nerve agents or other chemical weapons in theater. Overall, Army veterans report greater exposure to PB, pesticides, and nerve agents than Navy veterans. A large survey of British Gulf War veterans also found that PB and pesticide use were reported by more U.K. Army personnel than those in the Royal Air Force or Navy.[241]

Pyridostigmine bromide (PB) use in the Gulf War

Pyridostigmine bromide (PB) is a compound that reversibly binds to, and temporarily inactivates, AChE. It is the active ingredient in the nerve agent pyridostigmine pretreatment (NAPP) pills that were distributed to military personnel in the Gulf War as part of a three drug regimen to protect troops from poisoning by nerve agents. The small white PB pills were intended for use *before* a nerve gas attack, to establish blood levels adequate to temporarily bind about 30 percent of circulating AChE. If exposed to nerve agents, soldiers were to inject themselves (or their buddy) with two antidotes, atropine and 2-pralidoxime chloride (2-PAM), using prepackaged autoinjectors. These measures were intended to protect cholinergic receptors from excess acetylcholine buildup, and "rescue" AChE in order to restabilize cholinergic nerve transmission after the attack. Orders for initiating PB pretreatment were issued by unit commanders. The NAPPs were contained in blister packs of 21 pills, 30 mg. each. Each pack provided the number of pills needed for one week at the recommended dosage of one 30 mg. pill every eight hours.[951,1604]

The 1990-1991 Gulf War was the first time the U.S. military had used PB on a widespread basis as a nerve agent pretreatment. In 1990, PB was not licensed for this purpose by the U.S. Food and Drug Administration (FDA) but had been approved, since 1955, for treatment of myasthenia gravis. As a nerve agent protective measure, PB was considered an investigational new drug (IND). At the request of DOD, FDA granted a temporary waiver, in December 1990, that allowed use of PB in theater, in situations involving combat or the threat of combat, without the usual IND requirement for informed consent.[1275] The waiver was granted in light of the threat posed by Iraqi chemical weapons and the long history of PB safety in the treatment of myasthenia gravis.[781,951,1275,1604,1667] A number of problems occurred in implementation of the use of PB under this agreement, however, which prominently included insufficient information provided to troops in theater, and failure to keep adequate records of PB distribution and use.[462,1275,1604,1667]

Pyridostigmine bromide has now been approved by FDA for use as a pretreatment measure against exposure to the nerve agent soman.[1664] Research in animal models indicates that PB pretreatment enhances the effectiveness of the two antidotes that are used after exposure to soman, which permanently inactivates AChE within minutes. Pyridostigmine is not useful as a pretreatment in the event of sarin exposure, since sarin's effects on AChE can be mitigated by the post exposure antidotes over a period of several hours.[504,830,951,1793,1810] There have been no reports indicating that soman was present in theater during the 1990-1991 Gulf War, however. Available documents suggest that during the war, PB pretreatment was directed in anticipation of nerve agent exposure more generally, rather than specifically in relation to soman.[781,1588,1604,1690]

Epidemiologic studies indicate that about half of U.S. Gulf War veterans report using PB during deployment,[692,751] with greatest use among Army personnel.[458,1804] The DOD Office of the Special Assistant for Gulf War Illnesses (OSAGWI) commissioned the RAND National Defense Research Institute to undertake an in-depth evaluation of pesticide and PB use patterns by ground troops during the Gulf War.[458] Investigators conducted detailed interviews of over 2,000 Gulf War veterans. Results indicated that slightly more than half of Army and Navy/Marine Corps personnel serving on the ground used PB, but only 23 percent of Air Force personnel used PB. Among individuals who used PB, the number of pills taken was highly variable, with an average of 26 pills used in a given month. Most individuals reported taking three or fewer pills per day for 30 days or less, but a small percentage reported taking substantially more.[458] Overall, troops living in the open desert took PB at twice the rate of troops in tent cities, who in turn took PB at twice the rate of personnel living in buildings.[458] Results from the population-based Iowa study also suggested that active duty personnel took more pills, overall, than reservists.[692]

Pesticide use and exposures in the Gulf War

> On a nightly basis, we would spray our uniforms with pesticides. There was a chemical spray that they gave us to spray our uniforms. We had to hang them outside so that the excess spray would dissipate in the air, I guess. We weren't supposed to put them on immediately after spraying them. The sand fleas were a problem. We used to put flea collars around the legs of our cots or we would put flea powder on the floor around our cots to try to keep the sand fleas away from us while we were sleeping. We slept with nets over us to keep the flies off. The flies were ungodly.
>
> --SSgt TS, Gulf War veteran[716]

The desert environment was home to large numbers of flying and biting insects, and other pests, which posed a risk of disease to troops in theater. Control of disease-carrying pests is an important part of force protection and readiness during military deployments. Troops serving in the region in earlier campaigns had been affected by high rates of pest-borne diseases[475,596,1632] and the U.S. military implemented extensive measures to limit this problem in the Gulf War. Individuals were issued pesticide creams, liquids, and sprays, to use on their skin, their uniforms, and their bedding, and pest strips, bait, and sprays to use in their living quarters. Military preventive medicine specialists and field sanitation teams also conducted extensive operations to control pests in areas where people lived, ate, and worked with environmental fogging and surface spraying. These efforts were largely successful, as demonstrated by the low rate of vector-transmitted infections identified during the war.[664,1632]

Similar to other exposures in the Gulf War, no records were kept in relation to pesticide use or exposure for different areas, units, or individuals. After the war, when concerns were raised about the possible contribution of pesticides to veterans' unexplained illness, DOD undertook a number of assessments to determine the types of pesticides to which Gulf War veterans were exposed, the amounts used in theater, and patterns of pesticide use among individuals in the general military population and by pest control personnel.

In its final Environmental Exposure Report on pesticide use in the Gulf War, issued in 2003, DOD reported that U.S. personnel serving in the Gulf War used or had available for use, at least 64 pesticides and related products, containing 37 active ingredients.[1632] Of these, 15 were identified as "pesticides of potential concern" based on what was known about the use and toxic effects of these compounds. The 15 pesticides are listed in Table 2, and include seven organophosphates, three carbamates, two pyrethroids, one organochlorine, and two forms of the insect repellant DEET.

The most commonly used personal repellants were DEET, which was primarily to be used on the skin, and permethrin, which was to be sprayed onto uniforms. Some personnel are known to have acquired personal use pesticides in addition to those supplied by the military, including the commercial product OFF, citronella products, and flea collars. Military environmental pesticide control measures included surface spraying and environmental fogging using the organophosphates chlorpyrifos, diazinon, and malathion, in varying concentrations, as well as the carbamates propoxur and bendiocarb. The organochlorine lindane powder was used by military police and other personnel for delousing in the processing of the more than 87,000 enemy prisoners captured in the war. Lindane was also issued to troops for their personal use, primarily to Army personnel.[1632] In addition, environmental pest control was commonly provided by local pest control services in host nations, either under contract with the military, or supplied by health departments of local Saudi Arabian municipalities. Relatively little information is available concerning the types of compounds used or the frequency and patterns of spraying done by local pesticide services.[1632]

Gulf War epidemiologic studies queried veterans about pesticide use in theater in diverse ways that ranged from a simple question about whether or not the veteran had used "pesticides" during deployment, to more detailed questions about specific types used and the extent of their use. The U.S. national survey

Table 2. Pesticides and Insect Repellants Identified as Pesticides of Potential Concern by the Deployment Health Support Directorate

Compound	*Use*	*Chemical Class*	*Purpose*	*Application*
Pesticides and Repellants Used by the General Military Population				
DEET, 33% cream, stick	Personal use repellant	Dialkylamide	Repel flies and mosquitoes	By hand to skin
DEET, 75% liquid	Personal use repellant	Dialkylamide	Repel flies and mosquitoes	By hand to skin, uniform, netting
Permethrin, 0.5% spray	Personal use repellant	Pyrethroid	Repel flies and mosquitoes	Sprayed on uniforms
d-Phenothrin, 0.2% aerosol	Area use repellant	Pyrethroid	Knock down, kill flies and mosquitoes	Sprayed in tents, other enclosed areas
Methomyl 1% crystals	Fly bait	Carbamate	Attract and kill flies	Placed in pans outside latrines, tents
Azamethiphos, 1% crystals	Fly bait	Organophosphate	Attract and kill flies	Placed in pans outside latrines, tents
Dichlorvos, 20% pest strip	Pest strip	Organophosphate	Attract and kill mosquitoes	Hung in tents, working areas, dumpsters
Pesticides Used by Pesticide Applicators				
Chlorpyrifos, 45% liquid	Sprayed liquid	Organophosphate	Kill flies, mosquitoes, flying insects	Sprayed in corners, cracks, crevices
Diazinon, 48% liquid	Sprayed liquid	Organophosphate	Kill flies, mosquitoes, flying insects	Sprayed in corners, cracks, crevices
Malathion, 57% liquid	Sprayed liquid	Organophosphate	Kill flies, mosquitoes, flying insects	Sprayed in corners, cracks, crevices
Propoxur, 14.7% liquid	Sprayed liquid	Carbamate	Kill flies, mosquitoes, flying insects	Sprayed in corners, cracks, crevices
Bendiocarb, 19% liquid	Sprayed powder	Carbamate	Kill flies, mosquitoes, flying insects	Sprayed in corners, cracks, crevices
Chlorpyrifos, 19% liquid	Fog	Organophosphate	Kill flies, mosquitoes	Large area fogging
Malathion, 91% liquid	Fog	Organophosphate	Kill flies, mosquitoes	Large area fogging
Delousing Pesticide				
Lindane, 1% powder	Delouser	Organochlorine	Kill lice, other insects	Dusted on prisoners, also for personal use

Source: DOD Environmental Exposure Report: Pesticides (2003)[1632]

indicated that about half of all Gulf War veterans reported using personal pesticides,[751] with additional studies suggesting that pesticide use was more common in Army than Navy personnel.[524,1782] The Iowa study also indicated that reservists reported pesticide use more commonly (63%) than active duty personnel (44%).[692]

The RAND investigation of pesticide use among ground troops during the Gulf War reported considerable diversity in patterns of pesticide use in theater. Survey respondents were often unable to recall the specific chemicals used during deployment, but could identify the form of pesticides used (e.g., spray, liquid, powder) and how it was used (e.g., on clothing, skin, in tent), from which investigators imputed the most likely compound.[458] Personnel living in the desert used more pesticide sprays and liquids than those who lived in buildings. Officers reported less use of pesticide lotions and flea collars than enlisted personnel, and senior enlisted personnel reported greatest use of pesticide sprays and powders.

Overall, 62 percent of ground troops interviewed reported some form of pesticide use. Forty-four percent used pesticide sprays, a median of 30 times per month, and 26 percent used pesticide lotions a median of 20 times per month. Investigators estimated that the most commonly used compound was DEET, used by half of all personnel, a median of 30 times per month. Permethrin was used by fewer personnel[458,1632] but was used an average of almost 30 times per month. This raises concerns, since the permethrin label indicated that uniforms were to be sprayed only once every six weeks, or after six launderings. In contrast, DOD reports indicate that guidance issued to some Army personnel directed them to "apply a light coat of permethrin every four or five days."[1632]

The RAND investigation indicates that overuse of pesticides was most apparent for permethrin, d-phenothrin, lindane, and flea collars, although fewer individuals used these than the more commonly used DEET. No-pest strips were also frequently used in greater density than recommended, particularly in eating areas and latrines. Some pesticide overuse was extreme. About 13 percent of veterans reported using pesticide sprays more than 50 times per month, and about five percent reported using pesticide liquids or lotions more than 100 times in a given month, or more than three times per day.[458]

> It also seems reasonable that people in environments with large numbers of insects, such as in the Persian Gulf, would be tempted to use whatever means was available to remove the pests, including using products in ways that were not recommended.
>
> --RAND National Defense Research Institute, *Pesticide Use During the Gulf War*[458]

RAND investigators reported that personnel who reported frequent use of one type of personal pesticide were also more likely to report frequent use of multiple pesticides, suggesting exposure to a "cocktail of pesticides." [458] Use of personal pesticides was also significantly correlated with the number of PB pills taken in a given month. Over one in four veterans serving on the ground reported they had applied pesticides from 51 to over 120 times in a given month, and had also used an average of 15-19 PB pills in the same month.[458] By comparison, ground troops who reported no use of pesticides took, on average, only six PB pills in a given month.

The DOD final environmental exposure report on pesticides in the Gulf War included a health risk assessment that relied on information from the RAND survey, as well as interviews conducted with preventive medicine personnel knowledgeable about field pesticide use. The report concluded that "at least 41,000 Gulf War service members may have been overexposed to pesticides" and that "overexposure to pesticides, particularly organophosphates and carbamates, may have contributed to the unexplained illnesses reported by some Gulf War veterans." The figure of 41,000 was provided as a minimum figure, and did not consider effects of "overexposure" potentially resulting from combinations of organophosphate and carbamate pesticides with concurrent exposures to DEET, permethrin, PB, or low-level nerve agents or pesticide exposures resulting from pest control services provide by host nations.[1632]

Although comprehensive information on pesticide use in current deployments to Iraq and Afghanistan has not yet been reported, it appears that improved pesticide use and oversight have been among the important lessons learned from the 1991 Gulf War.[36] In 1993, the Deputy Undersecretary of Defense issued three pest management "Measures of Merit" that established objectives for improved pest management planning, a 50 percent reduction in the amount of pesticides used on military installations, and improved training and certification of pesticide applicators.[442] The military has now established improved standards and practices that include expanded use of trained preventive medicine field teams that monitor environmental hazards, training and printed materials for military personnel on the need for proper use of pesticides and insect repellants, as well as some changes in the specific pesticide products used.[1074,1583,1632]

There are multiple indications that pesticide usage in Operation Iraqi Freedom has differed from that in the 1990-1991 Gulf War. DEET formulations currently provided by the military contain 20-33 percent DEET; the 75 percent DEET liquid issued during the Gulf War is no longer in use.[62] Lindane, the organochlorine issued for delousing prisoners and for personal use during the Gulf War is no longer used for either purpose by the military.[63] In addition, troops in current deployments have had access to uniforms that were pretreated with permethrin, and permethrin treatment kits that reduce risks associated with uniform spraying, and sometimes over spraying, as occurred in the Gulf War.[62,1771]

Reports indicate that Iraq War troops were more educated about pesticides,[36] but sometimes did not have sufficient access to repellants, at least in the first years of the war.[399,1771] A survey of 870 service members at camps in Kuwait in 2004 indicated that most personnel had received medical briefings on why and how insect repellants were to be properly used. However, only 36 percent had been issued any DEET product and 48 percent had received permethrin products.[1771] A substantially larger number of cutaneous leishmaniasis cases have occurred among Iraq War troops, compared to the 1991 Gulf War,[399] which a National Defense University report suggests may be related to reduced used of pesticides.[1537]

Therefore it appears that pesticide usage by troops in Operation Iraqi Freedom is decreased, or from another perspective, improved and more judicious, compared to pesticide usage in the 1991 Gulf War. This can be attributed to a number of factors, including improved pest management policies, improved education of pesticide applicators and the general military population, expanded placement of preventive medicine field sanitation teams, differences in living conditions, discontinued use of lindane and 75 percent DEET, and, in some cases, inadequate supplies of repellant products.

Exposure to chemical weapons in the Gulf War

> In late January 1991, while assigned to an area between Rafha and Naryian about six miles south of the Iraqi border, BM recorded in his journal and on videotape that chemical 'false alarms' were going off almost every day. At first, according to BM, the alarms were explained as being caused by vapors coming off the sand. Later, since the alarms kept going off and troops no longer believed they were being caused by vapors, BM said he was informed by both his battalion commander and the battalion NBC NCO that the alarms were sounding because of 'minute' quantities of nerve agent in the air, released by the coalition bombing of Iraqi chemical weapons facilities. The troops were assured that there was no danger.
>
> --1994 Senate Committee report on Gulf War veteran, 18th Airborne Corps[1688]

Among the many challenging issues related to understanding levels and effects of hazardous exposures in the Gulf War, those surrounding troop exposures to chemical weapons in theater are the most complex and controversial. Multiple accounts of chemical alerts during the war, positive readings on chemical detection tests, and incidents involving unusual vapors and unexplained symptoms were reported in the

media and documented in Congressional reports. Seventeen years after the war, after numerous government and special committee investigations, research studies, and reports, significant questions remain about the extent to which military personnel were exposed to low levels of chemical warfare agents during the Gulf War.

For the first five years after Desert Storm, DOD maintained that no troops had been exposed to chemical agents. The Iraqis were known to have chemical weapons and to have used them against Iranians and their own citizens in the 1980s. The Department of Defense consistently affirmed, however, that Iraq had not used chemical weapons offensively in the Gulf War and that none had been positioned in areas of Iraq that were penetrated by Coalition forces. In preparing for the Gulf War air offensive, U.S. military planners had identified multiple Iraqi targets where chemical weapons were believed to be manufactured or stored.[1748] Most of these chemical targets had been successfully destroyed during the air campaign.[320] But DOD indicated that any chemical agents released with the bombing of Iraqi targets had occurred a great distance from Coalition troop locations, too far away to have affected U.S. or allied personnel.

In June of 1996, DOD announced that U.S. troops had potentially been exposed to low levels of nerve agents after the cease fire in March of 1991, when Army personnel detonated large caches of munitions stored at a massive compound near Khamisiyah, in southeastern Iraq. This announcement proved to be a turning point in the federal response to Gulf War health issues. It triggered an expanded effort to analyze and address Gulf War health issues overall, stimulated multiple investigations into chemical weapons exposures in theater, and led to a military research program aimed at better understanding effects of low-dose exposure to chemical warfare agents.[1102]

Chemical warfare agent exposure and detection in the Gulf War theater. Understanding the likely extent of chemical agent exposures in the Gulf War has been complicated by a number of factors. These include long-time official denials that chemical releases and exposures had occurred in theater, the postwar disappearance of the U.S. Central Command's records of reported chemical events during the Gulf War, the limited capabilities of chemical monitoring equipment in theater, controversy surrounding government conclusions about chemical releases at Khamisiyah and other locations, and the limited degree to which military personnel could have known if they had been exposed to low levels of chemical agents.

There are multiple scenarios in which chemical agent exposures could potentially have occurred. These include Iraqi offensive use of chemical weapons, downwind drift of chemical agents released by Coalition aerial bombing of Iraqi targets, local exposure and downwind drift following ground destruction of chemical munitions, and exposure of individuals who entered bunkers or other areas contaminated by chemical weapons. The Department of Defense has maintained that there was no offensive use of chemical weapons in theater, and has only verified that troops were exposed to nerve agents in one case, as a result of the ground destruction of chemical weapons at Khamisiyah. Multiple reports of chemical detections and other incidents that potentially involved chemical exposures have long fueled speculation, however, that additional exposures may have occurred, exposures that were either not identified or not verified by DOD.[388,1560,1683,1685,1688]

In the years since the war, an extensive number of reports from DOD, the U.S. Central Intelligence Agency (CIA), the United Nations, and other sources have provided information on the types and amounts of chemical agents stored, deployed, and destroyed in different locations in the Gulf War theater. Previous advisory panels, government agencies, and Congressional committees have been tasked with reviewing available information on these issues. Their reports have identified a variety of different issues and produced different, sometimes contradictory, findings.[1231,1232,1595,1683,1688,1690] Detailed analysis of the many intelligence reports, modeling protocols, incident reports, and investigations related to possible chemical releases and exposures in theater was beyond the scope of the present report. Instead, the

Committee broadly reviewed information provided by government investigations and issues raised by earlier panels to determine what has been learned about chemical agent exposures during the Gulf War.

It is important to note that there have been no reports during or after the war of high-level chemical exposure incidents in which large numbers of personnel experienced clear signs and symptoms of chemical agent poisoning. Available information indicates that the major unanswered questions about exposure to chemical weapons during the Gulf War relate to: (1) whether more limited or lower level chemical agent exposures occurred in theater that were undetected, unreported, or unverified by the government, and (2) if modeled plume estimates for the Khamisiyah demolitions usefully reflect chemical exposures that resulted from releases at that site.

Limitations and problems in detecting chemical warfare agents. At the most fundamental level, identifying chemical agent exposures during the Gulf War depended on reliable detection of those agents. Concerns about the inability of chemical monitoring systems used in the Gulf War to detect lower levels of chemical agents in the field were raised in 1994 by the Senate Banking Committee, and again in 1996 and 1997 by the Presidential Advisory Committee on Gulf War Veterans' Illnesses.[1227,1231,1690]

During the Gulf War, the U.S. military used a multilevel system for detecting and verifying chemical agents. With an initial detection, an alarm alerted personnel to the possible presence of a chemical agent and troops donned protective gear until results from a second type of test either verified a positive detection or permitted an "all clear" notification. The primary early warning system for airborne chemical agents was the M8A1 chemical alarm. These alarms could be placed upwind from the unit's position to monitor for VX and G series nerve agents, including sarin. The M8A1 could only detect nerve agents at levels that can also cause symptoms, and could not detect blister agents such as mustard gas at any level.[1231,1595,1605,1690] Handheld chemical agent monitors (CAMs) could detect airborne vapors of both nerve and blister agents but were not primarily used as an open air warning device. They were more commonly used to determine if personnel or surfaces had been contaminated, by assessing and roughly quantifying vapors emanating from liquid agent.[961,1595] Liquid chemical agent hazards could also be detected by M8 and M9 papers, issued to individual service members, and by a specialized kit (M272) used by NBC (nuclear, biological, chemical) personnel to identify chemical agents in water.[1595]

The most widely used system for verifying airborne chemical agents was the M256A1 Chemical Agent Detector Kit. Testing was conducted by trained NBC personnel and involved a sequence of steps that required 20-25 minutes to complete.[1595,1613] Although not useful as an early warning monitor, the M256A1 kits were more sensitive to nerve agents than the alarms, and less prone to false positives.

The U.S. military also fielded 60 armored FOX NBC Reconnaissance vehicles, provided by Germany during Operation Desert Shield.[1606] The Fox vehicles were considered the most technologically advanced chemical equipment used by the U.S. in the Gulf War. This mobile unit could conduct chemical and radiation reconnaissance in different settings, with capabilities to sample for, detect, and verify chemical agents. Fox vehicles were equipped with the M43A1 chemical agent detector and an MM-1 mobile mass spectrometer. They were designed primarily to identify ground contaminated areas and were most sensitive and specific for detecting liquid chemical agents. The Fox was not considered a suitable first warning device when used in air sampling mode, since a relatively high concentration of nerve or blister agent vapor was required for detection.[1231,1595,1606,1628,1690] The MM-1 spectrometer provided detailed chemical agent verification capability, but identified only the compound present at the highest concentration.[1228,1628] If, for example, nerve agent was present, but at a lower concentration than a compound in oil fire smoke, only the oil fire compound would be identified.

The multilevel chemical agent detection system used by the military during the Gulf War was intended to detect chemical agents at levels that could cause acute harm to troops in the field, and provide warning to allow them to take protective action. This was consistent with the understanding at the time that subacute

Table 3. Chemical Agent Vapor Detection Capabilities of Equipment Used in the Gulf War, and Current Chemical Weapons Air Exposure Guidelines and Standards

	Sarin	*Mustard*
Vapor Detection Capabilities of Chemical Detection Equipment Used by the U.S. Military in the Gulf War		
M8A1 chemical alarms	0.1 – 0.2 mg/m^3	no capability
Portable chemical agent monitors	$\leq$ 0.1 mg/m^3	$\leq$ 0.1 mg/m^3
M256A1 detector kits	0.005 mg/m^3	2 mg/m^3
Fox vehicle M43A1 alarm	0.2 mg/m^3	no capability
Fox vehicle MM-1 mass spectrometer monitor	62 – 100 mg/m^3	
Chemical Weapons Air Standards and Guidelines Currently Used by the U.S. Military		
Air exposure limits:		
Immediate danger to life and health (1 time exposure)	0.1 mg/m^3	0.7 mg/m^3
Short term exposure limit (occasional 15 minute exposure)	0.0001 mg/m^3	0.003 mg/m^3
Acute exposure guideline levels (1 time exposure)		
Level 1 (potential for noticeable effects, minor discomfort)		
- 10 minutes	0.0069 mg/m^3	0.400 mg/m^3
- 1 hour	0.0028 mg/m^3	0.067 mg/m^3
Level 2 (more obvious effects, potential impact on function)		
- 10 minutes	0.087 mg/m^3	0.600 mg/m^3
- 1 hour	0.035 mg/m^3	0.100 mg/m^3
Level 3 (potentially life threatening)		
- 10 minutes	0.38 mg/m^3	3.900 mg/m^3
- 1 hour	0.13 mg/m^3	2.100 mg/m^3

Chemical Agent Detection Capabilities Sources: Defense Science Board Task Force on Persian Gulf War Health Effects,[1597] National Research Council,[1007] U.S. Department of Defense[1605,1606,1613]
Air Standards and Guidelines Source: U.S. Army Center for Health Promotion and Preventive Medicine[1581,1582]

exposure to chemical agents did not pose a serious health threat. In recent years, growing concern about possible adverse effects of lower level exposures have prompted federal agencies, including DOD, to revise chemical agent exposure standards and adopt guidelines that are more conservative than those in place during the Gulf War, that is, standards that consider lower exposure levels to be potentially problematic.[1574,1755] The Department of Defense has also replaced the M8A1 alarms with next generation alarms that have expanded capabilities and are less prone to false alarm.[1605]

Information on detection capabilities of equipment used by the U.S. in the Gulf War for airborne sarin and mustard is provided in Table 3. The table also provides current toxicity standards and guidelines used by the military for exposure to these agents. Because the toxicity of chemical agents varies with the concentration and duration of exposure,[86] limits are provided for immediate and short term exposures, and guidelines identify levels at which mild, more serious, and life threatening health effects may acutely occur.[1581,1582] As shown, the M8A1 alarms could have detected sarin at levels that pose an immediate danger to life and health with a one time exposure. The alarms would not have detected sarin present at levels capable of producing limited symptoms after only 10 minutes exposure, identified as a Level 1 exposure in the table. The M8A1 alarms also might not have detected a Level 2 exposure, associated with

more significant symptoms and signs with 10 minutes exposure. The M256A1 detection kits could potentially have identified chemical agents at considerably lower levels, but would not likely have been used in the absence of an initial warning alarm.

Overall, monitoring capabilities for chemical agent vapors were insufficient to detect levels that could cause limited symptoms with relatively brief exposures, or more pronounced problems with sustained exposures. And, as previously indicated, the M8A1/M43A1 alarm systems would not have detected nerve agents at levels too low to cause any symptoms, or blister agents at any level.

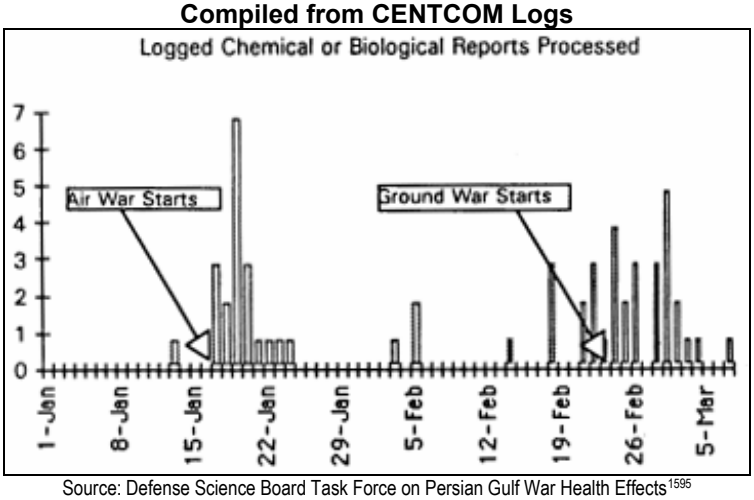
> From the Saudi berm north, the air was heavy with oil smoke. This smoke deposited an oily residue on the alarms' paddles which tripped the alarms. On the average, the alarms activated every 20 to 30 minutes. … The M8A1s were useless in the smoky, dusty desert environment.
>
> --March 1991 memo, 2nd Light Armored Infantry Battalion[1381]

Another major concern related to chemical detections were problems caused by the repeated sounding of alarms deemed to be false. Department of Defense reports indicate that M8A1 alarms were widely used in theater, and that false alarms were triggered by factors such as fuel vapors and engine exhaust, oily smoke, blowing sand, and low batteries on the units.[961,1605] Surveys of Gulf War veterans consistently report that most ground troops heard chemical alarms one or more times during deployment, indicating that a very large number of alarms sounded in theater. But overall, it is not known how many chemical alarms sounded over the course of the war, in what areas they occurred, or how many were followed up with additional testing. Repeated false alarms in some locations led some units to ignore or disable their alarms.[1228,1684] Such problems might have led some personnel to believe they were exposed to chemical agents when they had not been, or others to believe they had not been exposed when they might have been.

Chemical incidents during the war that were communicated to Central Command (CENTCOM) forward headquarters in Riyadh, Saudi Arabia, were recorded in logs maintained by NBC desk officers. In 1996, DOD announced that the NBC logs on which chemical incidents had been recorded during Operations Desert Shield and Desert Storm were missing. The Pentagon's Office of the Inspector General (IG) investigated the logs' disappearance and issued its report in 1997. The IG report indicated that the NBC logs had initially been generated in hard copy, entered into computer files, and backed up on portable disks by the NBC desk in Riyadh.[1602] After the war, hard copies, laptop computers, and disks containing the logs were shipped to CENTCOM Headquarters in Tampa, Florida, and stored in safes. It was never determined with certainty what happened to them, but the IG report found it likely that, contrary to DOD policy, they had inadvertently been disposed of when the NBC office was relocated in 1994. Duplicate disks had also been sent to Aberdeen Proving Ground after the war, but were also not locatable.

The loss of the NBC logs made it impossible to ascertain what chemical incidents had been reported to CENTCOM and what determinations had been made. It also raised a great deal of public skepticism concerning the reliability of DOD reporting on chemical exposures in theater. The IG report indicated that copies of 32 of the approximately 150 pages of the missing NBC logs had been provided to assist the Defense Science Board Task Force on Persian Gulf War Health Effects in 1993, whose 1994 report contained information compiled from those pages. The graph in Figure 1 is taken from the 1994 report, and depicts an unspecified group of chemical reports logged by CENTCOM between January 1 and March 7, 1991. The largest number were recorded in the early days after the air war began, and again in the days surrounding the ground war.

Figure 1. Logged Chemical or Biological Reports Processed: Compiled from CENTCOM Logs



Source: Defense Science Board Task Force on Persian Gulf War Health Effects[1595]

As previously described, verification of a chemical agent detection required that an initial detection be retested and confirmed by a second type of test that used different technology. The Presidential Advisory Committee pointed out that verification of chemical detections using the Fox vehicle was often not possible on the battlefield, as described below:

> Doctrine required that following an initial alarm for CW [chemical warfare] agent(s), the Fox vehicle's full spectrum capability should be engaged. To complete the full spectrum analysis, however, required that U.S. military personnel stop the Fox vehicle, return to the site where the MM-1 alarmed, and then perform a 20-minute process. Fox vehicle personnel recognized the danger that stopping in the midst of battle would pose to themselves and their fellow service members, and so they did not. As a consequence, full spectrum analyses rarely were performed during the Gulf War. Yet doctrine is clear it is impossible to confirm a detection without a full spectrum. Because doctrine did not accommodate the actual conditions of use, a post-incident evaluation of an incident that lacks a full spectrum cannot be validated.
>
> --Presidential Advisory Committee on Gulf War Veterans' Illnesses, 1997[1231]

The Department of Defense has consistently maintained that no U.S. chemical agent detections during the Gulf War were verified as positive. This is commonly interpreted to mean that all alarms that sounded in theater were false alarms, and that troops were not exposed to chemical agents, except in relation to the Khamisiyah demolitions. But there are limitations in what can and cannot be determined from chemical alarm detections. It can only be said that DOD has not verified any U.S. chemical detections based on its own criteria which, at a minimum, required evidence of positive detections using two types of tests.[1230] As indicated, blister agents and lower level exposures to nerve agents would not have been detected by M8A1 alarms or the M43A1 alarm used on Fox vehicles. And detections that did trigger alarms were not always followed up with additional testing.

It is relevant to note that at least one chemical alarm sounded on March 4, 1991, during the first munitions demolitions at Khamisiyah. Initial follow up tests with M256A1 kits were inconclusive or negative, and repeat tests were negative, leading NBC personnel to conclude that the alarm had been false, and that no chemical agents were present. Therefore, no additional actions were taken and no chemical incident report was submitted up the chain of command.[1638] Years later, DOD confirmed that there were multiple definite releases at the site and that about 100,000 troops may have been exposed to low levels of nerve

agent as a result. But based on routine field criteria, even at close proximity to the chemical agent release, the alarm was determined to be a false alarm, and the detection not credible.

Exposure to nerve agents in relation to the Khamisiyah demolitions. In early March of 1991, just days after the U.S. declared a cease fire, U.S. soldiers began operations to destroy enemy munitions at a large weapons compound near Khamisiyah, about 100 km. from the Kuwaiti border in southeastern Iraq. The Khamisiyah Ammunition Supply Point was a massive Iraqi weapons storage area, covering nearly 40 square km., which included approximately 100 ammunition storage bunkers, 88 ammunition storage warehouses, and many additional buildings.[1630] It is now known that chemical agents were located at this site and were destroyed and scattered during the demolitions operations, potentially exposing large numbers of U.S. personnel to low levels of sarin and cyclosarin. An enormous amount has been written about these events, including details of the demolitions operations, dissemination of intelligence on chemical agents to units responsible for destroying munitions, and efforts to determine who may have been exposed to chemical agents, and at what levels.[1590,1630]

Although attention has focused on demolition events at Khamisiyah following the cease fire, there was considerable activity at the site during the Coalition air and ground offensives. A 2002 DOD report indicates that during the period of active hostilities, Coalition aircraft made 40 air strikes against Khamisiyah on six different dates between January 19 and February 25, 1991. These attacks reportedly destroyed 45 warehouses and at least four bunkers.[1630] Units of the XVIII Airborne Corps had attacked and occupied the sector of Iraq in which Khamisiyah was located during the ground war. On February 26, 1991, the XVIII Airborne Tactical Operations Center sent a message that they may have hit chemical munitions near a site referred to as Objective Gold, a primary target of the ground offensive that was located about five km. from Khamisiyah.[1590,1630] No further investigations have been reported concerning possible chemical releases during the air and ground wars either at Khamisiyah or at Objective Gold. The 24th Infantry Division is reported to have pushed through the Khamisiyah weapons site on February 26, but not to have occupied the site at that time.

Before the ground war began in February, 1991, Army Central Command had directed the XVIII Airborne and VII Corps to destroy all enemy munitions within their respective sectors, in an effort to eliminate Iraq's military capabilities.[1630] After the ceasefire, units in the XVIII Airborne's 82nd Airborne Division, along with supporting units, conducted their initial reconnaissance in and around Khamisiyah. Troops wore protective MOPP gear and had M8A1 alarms and M256A1 test kits when they entered the bunkers to survey the site, and chemical officers later reported that no chemical weapons had been detected. Army directives available to the XVIII Airborne at that time indicated that Iraqi chemical weapons could be identified by certain characteristic markings. Later information that munitions carrying chemical agents were not clearly or consistently marked was not provided until after the demolitions.[1590,1630]

After their initial survey of the site, combat engineering units set charges in preparation for the initial large-scale demolitions on March 4, 1991. All personnel and civilians were cleared from the area. The troops conducting the demolitions moved back, at least three miles from the site, to observe the explosions when the charges were detonated. Reports indicate that the massive explosions were visible for miles around, with debris flying out to great distances, some dropping in areas where demolitions personnel were observing the explosions. M8A1 alarms were operational during this time, and at least one is reported to have sounded from an observation location, causing unit members to go into MOPP4. Confirmation tests using M256 detection kits were negative, and the alarm or alarms were determined to be false. Explosions continued for hours after the detonations.[1630,1638]

Additional large-scale demolitions were conducted on March 10, when bunkers, warehouses, and stacks of crated rockets in an area known as "the Pit" were destroyed. On March 20, more than 400 earth berm