REGULATORY TOXICOLOGY AND PHARMACOLOGY **24,** S129–S138 (1996)
ARTICLE NO. 0089

Chemical Sensitivities and the Gulf War: Department of Veterans Affairs Research Center in Basic and Clinical Science Studies of Environmental Hazards[1]

NANCY FIEDLER,* HOWARD KIPEN,* BENJAMIN NATELSON,† AND JOHN OTTENWELLER†

**UMDNJ-Robert Wood Johnson Medical School, Environmental & Occupational Health Sciences Institute, 681 Frelinghuysen Road, Piscataway, New Jersey 08855; and †Veterans Affairs Medical Center, 385 Tremont Avenue, East Orange, New Jersey 07017-1095*

Received May 17, 1996

The purpose of the New Jersey Center for Environmental Hazards Research is to define the illness referred to as Persian Gulf Syndrome (PGS). Our preliminary data indicated that more than half of the Persian Gulf Registry (PGR) veterans reported illness characterized by severe fatigue and symptoms consistent with chemical sensitivities. Therefore, our research approach focuses on investigations of veterans with chronic fatigue syndrome (CFS) and multiple chemical sensitivities (MCS). Project 1 is an epidemiological study of 2800 PGR veterans. Symptoms, indices of Chronic Fatigue (CF) and Chemical Sensitivity (CS), and risk factors will be surveyed with mailed questionnaires. Risk factors include demographics, past medical history, psychosocial variables, Gulf War experiences such as prophylactic medication use, occupational and environmental exposures, and pesticide exposures. Symptoms will be clustered to define Gulf War Syndromes. Significant associations between risk factors and these symptom clusters will also be investigated. Subjects identified as CF, CS, or both will be recruited into Projects 2 and 3. In Project 2, healthy veterans will be compared to veterans with CF, CS, and CF concurrent with CS. Veterans will undergo four studies: (1) viral–immunological, (2) psychiatric, psychological, behavioral, and neuropsychological, (3) autonomic dysregulation, and (4) marker of P4501A2 induction resulting from exposure to combusting material. The purpose of Project 3 is to test the autonomic, immunologic, neuropsychologic, and psychologic responses of veterans with CS or CF to two stressors: controlled chemical exposure and exercise. CS subjects will undergo chemical exposures in our Controlled Environment Facility (CEF) to assess their biologic and psychologic response to low-level exposure. CF subjects will undergo a maximal treadmill exercise test. Circadian patterns of catecholamines and axillary temperature, viral burden, and cardiovascular and endocrine reactivity will be measured in response to this physical stressor. Project 4 is an animal study evaluating the interaction between stress and pathology/physiology when rats are predisposed to disease by exposure to Soman or to Dioxin. Two strains of rats that differ in stress reactivity will be used to determine the interaction of hereditary factors and chemical exposure.

In 1990, the United States mobilized its armed forces to help Kuwaitis regain control of their country following the invasion of Iraq. Seven hundred thousand U.S. troops were sent to the Persian Gulf region during this time period. Actual ground fighting lasted only 4 days but troops were stationed in the region from November of 1990 until May of 1991. While troops experienced relatively little combat, the threat of chemical warfare was persistent until the end of ground combat.

Subsequent to this time period and upon returning home, soldiers from the conflict began to express a variety of health complaints, not all of which could be readily explained. In response to these complaints, a registry was established through the Department of Veterans Affairs (DVA); the DVA also requested research proposals to study the illnesses of the Gulf War. The ubiquitous threat of chemical exposures during the war suggested to many civilians and troops that these illnesses could be chemically related. This paper describes the research protocols developed to study the illness of the Gulf war veterans, particularly those veterans who report unusual sensitivities to chemicals following service in the Gulf.

BACKGROUND OF THE PERSIAN GULF REGISTRY

By April of 1995, approximately 36,604 of the 700,000 Persian Gulf veterans had completed the DVA registry

[1] This study was funded by New Jersey Center for Environmental Hazards Research, which is funded as a Research Center in Basic and Clinical Science Studies of Environmental Hazards by the Department of Veterans Affairs.

0273-2300/96 $18.00

TABLE 1
Chronic Fatigue—1988 Case Definition (2)

A. CDC Criteria—Major

1. New onset of fatigue with ⩾50% reduction in activity
2. No other preexisting medical condition

B. CDC Criteria—Minor (8 of 11)

1. Mild fever (99.5–101.5)
2. Sore throat
3. Tender cervical or axillary lymph nodes
4. Muscle weakness
5. Myalgias (pain in extremities)
6. Headaches (new and different)
7. Arthralgia
8. Neuropsychological complaints
9. Sleep disturbance
10. Chills

examination and were entered into the Department of Veterans Affairs Persian Gulf Registry (PGR). These veterans came from active duty, reserve, and guard units. Seventy-six percent of the 31,705 veterans with one or more symptoms received a medical diagnosis from this examination. Approximately 24% received no conventional diagnosis for their symptoms. Nine of the 15 most common diagnoses made by the examining DVA physician were based on symptom reports rather than objective test findings (e.g., joint pain, neurasthenia).

Among the most frequent registry veteran complaints, as recorded by the examining physician, were fatigue (14%), skin rash (15%), headache (HA) (17%), muscle/joint pain (16%), loss of memory (13%), shortness of breath (8%), sleep disturbances (5%), diarrhea and other gastrointestinal symptoms (4%), and chest pain (4%). Fatigue, muscle/joint pain, HA, and loss of memory are among those required in the CDC case definition for diagnosis of chronic fatigue syndrome (1, 2) (see Table 1) and are common among patients with multiple chemical sensitivities (3). Based on the relatively large number of veterans with unexplained or undiagnosed medical symptoms and the similarity in presentation between these complaints and those voiced by patients with CFS and/or MCS, our Research Center was developed to investigate hypotheses of an epidemic of CFS/MCS among Persian Gulf Veterans (PGV).

In addition to the symptomatic similarities cited above, a number of environmental stressors were present that could be associated with the unexplained symptoms presented. Because of the nature of the conflict in the Gulf, its location, the known capability of Iraq to deliver chemical and biological ordnance, the smoke from oil well fires, exposures to fuels and exhausts, sand fleas, pest control agents, depleted uranium, and electromagnetic radiation, it was reasonable to hypothesize that exposure to one or a combination of these environmental agents could be contributing to the veteran complaints.

PREVALENCE OF CFS AND MCS: PRELIMINARY DATA

Prior to the development of the New Jersey Environmental Hazards Research Center, two preliminary surveys were conducted in collaboration with Dr. Han Kang of VACO's environmental epidemiology service. A pilot questionnaire was mailed to 203 Northeastern region veterans whose medical complaints included fatigue, and thereafter, the same questionnaire was mailed to 228 different Northeastern region veterans who were on the registry but who did not list fatigue as a symptom. Results from the first sample revealed that 89% of those responding to the survey reported a fatiguing illness which began in 1991 or 1992. Approximately one-half of those with fatigue reported chemical exposure as a significant event that preceded their onset of fatigue. Thirty-nine percent of all respondents considered themselves especially sensitive to certain chemicals.

Four questions, emphasizing avoidance behaviors due to chemical sensitivities, were used to screen for chemical sensitivities among the PGR veterans (see Table 2) (4). Seven percent of the respondents reported three or more avoidance behaviors based on chemical sensitivities. Thirty-three percent of the respondents considered themselves especially sensitive to car exhaust, and 20% to perfume.

Analysis of the second questionnaire sent to PGR veterans, who were not initially screened for fatigue, revealed similar results. Sixty-three percent reported a fatiguing illness with 20% reporting a chemical exposure that preceded their illness. Thirty percent of all respondents considered themselves sensitive to certain chemicals with 19% sensitive to car exhaust and 11% to perfume.

Thus, a significant prevalence of self-reported fatiguing illness and chemical sensitivities was detected

TABLE 2

	YES	NO
1. Do you now need to follow any special diet because of chemical or food sensitivity?	_____	_____
2. Do you now take special precautions in your home or home furnishings (furniture, drapes, carpets) because of chemical sensitivity?	_____	_____
3. Do you now need to wear particular clothes because of chemical sensitivity?	_____	_____
4. Do you have trouble shopping in stores or eating in restaurants because of chemical sensitivity?	_____	_____

Source. Ref. (4).

TABLE 3

Medical data
Demographics
Past medical and psychiatric history
Review of systems
Psychosocial data
Family support
Social support
Psychological symptoms
Life events
Medical—Gulf War
Immunizations
Prophylactic medications (pyridostigmine)
Medical provider encounters
Therapeutic medications (atropine)
Combat
Wounds
Proximity to exploding ordnance
Under fire
Seeing wounded/dead
Unit leadership
Living conditions
Before, during, and after Gulf War
Environmental risks
Use of chemical protective clothing
Fire, smoke, respiratory irritants
CARC paint
DEET
Depleted uranium
Pest controls

among PGR veterans. This preliminary questionnaire data formed the basis of the proposed studies to investigate the risk factors, clinical presentation, and biological mechanisms of chronic fatigue and chemical sensitivities among PGR veterans.

PROJECT I: THE EPIDEMIOLOGICAL SURVEY

While our preliminary data suggested an outbreak of chronic fatigue and chemical sensitivities among the PGR veteran, it was not known whether these syndromes accounted for the remainder of the unexplained illnesses experienced by the PGR veterans. Further, even if CFS and MCS defined Persian Gulf Syndrome, nothing was known about the risk factors that may have contributed to the outbreak of these illnesses. Therefore, the purpose of our epidemiological survey is to systematically catalog physical and psychological symptoms of the PGR veteran and to survey the chemical, psychosocial, and physical stressors that contributed to this illness.

Approach

Subjects. A random sample of 2800 PGR veterans is being developed in cooperation with Dr. Han Kang of the Veterans Administration.

Questionnaire. A questionnaire which includes the variables in Table 3 is being mailed to the PGR subject pool for completion.

Analysis

A case control analysis between individuals who do and do not qualify for categories and combinations of symptomaticity will be performed to define the Persian Gulf illness and to characterize the relationship between this illness, Chronic Fatigue Syndrome, Multiple Chemical Sensitivities, and other standard psychiatric illness such as Posttraumatic Stress Disorder.

The relationship between several categories of risk factors and clusters of symptoms will be explored. For example, from preliminary questionnaire data, several demographic factors distinguished Registry veterans from the general population of veterans who fought the war. Among the general troops, 83.3% were from active units while 16.6% were from the reserve and national guard, yet 51% of registry veterans came from guard and reserve units, a threefold excess. A 50% excess of women, a fivefold excess of formerly married veterans, and a threefold excess of veterans over 40 years old were on the registry as compared to the general population of veterans in the Gulf. These preliminary data suggested that gender, age, and psychosocial factors may be important risk factors.

With regard to exposures, questions about residence, personal habits, and occupation will be asked in three time frames: pre-Gulf, Persian Gulf, and post-Gulf. A similar strategy will be used to explore psychosocial stressors such as significant life events or traumas and stressors imposed by the war, such as care of children and family separations. Results of the questionnaire will then be used to recruit veterans for study in the remaining projects.

PROJECT 2: CROSS-SECTIONAL STUDY OF THE PERSIAN GULF VETERAN

The purpose of this project is to characterize the chemical and psychosocial exposure status and illness of the veterans on the Persian Gulf Registry. While the nature of the exposures varied between individual veterans and between groups of veterans, it was apparent that soldiers in the Gulf conflict were exposed to a number of known psychological and chemical stressors. However, the impact of these stressors depends on the strength and duration of the exposures and individual moderating variables such as premorbid personality, other psychosocial and physical stressors, biological health status, and psychological coping mechanisms. Animal research has shown that these variables fit into an "equation" which allows one to predict the risk of medical illness following the kind of stressors such as those reported in the Gulf (5).

This model of illness is also consistent with that proposed by Bell *et al.* (6) as an explanatory model for chemical sensitivities. Bell *et al.* (6) cites the time-dependent sensitization literature in which repeated,

low-level chemical exposures over time can produce sensitization and symptoms to a chemical substance at much lower levels than might be expected. This literature suggests that other, nonchemical stressors (e.g., novel environment) can alter the nature and rate of sensitization (7). Thus, the chemical and psychosocial stressors present in the Gulf may have provided a natural experiment in which some susceptible individuals became sensitized to chemical exposures. This sensitization may not produce overt pathology detectable with routine clinical tests. Nevertheless, it can produce symptomatology in response to subsequent exposures.

The interaction of chemical exposures and psychosocial risk factors may account for the symptoms of fatigue and sensitivity to chemicals that resemble CFS and MCS. As currently observed, both CFS and MCS are preponderantly endemic illnesses. If a higher prevalence of these illnesses is observed in the PGR veterans, then this may represent an epidemic of these disorders. This could offer an opportunity to detect risk factor(s) that are difficult to delineate in a more heterogeneous population. Thus, the goals of this project are to characterize and compare the risk factors and the current medical status of PGR veterans with symptomaticity similar to chronic fatigue and chemical sensitivities.

Method

Subjects. Four groups of subjects will be recruited from those who completed the epidemiologic questionnaire from Project 1: veterans with chronic fatigue (CF) and chemical sensitivities (CS); veterans with CF, but no CS; veterans with CS, but no CF; and healthy veterans with no medical complaints. Veterans reporting between 4 and 7 symptoms from Table 1 will be defined to have moderate CF while veterans with 8 or more symptoms will have severe CF. Veterans who regard themselves as unusually sensitive to chemicals and who report having changed at least two of four lifestyle factors because of sensitivity to chemicals (Table 2) will be selected as CS. Subjects may also be classified as CS if they report feeling ill in response to five or more of eight substances/situations (e.g., pesticides, car exhaust, cologne) (Table 4). Due to the higher relative proportion of women on the registry and among community samples of CFS and MCS, 50% of the subject groups will be women.

Veterans with the following conditions will be excluded: underlying diagnosable medical illness to include neurologic disease or brain injury, stroke or cardiovascular disease, serious pulmonary disease, liver or kidney disease, serious gastrointestinal disorders, psychoses, bipolar disorder, bulimia/anorexia, and diabetes. Pregnant or lactating women will not be included.

Standard history and physical/structured psychiatric interview. All subjects will undergo a complete history and physical using the same review of systems as in the epidemiologic questionnaire to validate the questionnaire reports. In addition, extensive exposure history before, during, and after the Gulf war will be collected. The Diagnostic Interview Survey of the DSM-IIIR will be administered by clinical psychologists to determine current and lifetime psychiatric status. The Structured Clinical Interview for Diagnosis of Personality Disorders (SCID-II) will be administered to diagnose Axis II Personality Disorders.

Study 1: Viral–Immunological Factors

Activation of white blood cells to release cytokines can produce fatigue, malaise, myalgia, headache, low grade fever, arthralgia, and cognitive dysfunction, many of the PGV symptoms. Cytokine patterns will be evaluated to determine occult infection (e.g., Leishmania infection) and to contrast cytokines in the PGV with other CFS patients. Cell surface markers will also be evaluated. It is also possible that stress, chemical exposure, viral infection, or some combination of these factors has allowed latent viruses to reactivate. The acute onset of CFS and its flu-like components were responsible for the original belief that CFS was a form of chronic mononucleosis (8, 9). A much greater difference between patients and controls has been reported for HHV-6 (10). Two forms of this virus have now been identified and a new member of this virus family, HHV-7, has also been identified. Since the herpesvirus family is the class of viruses that can reactivate from a latent state, these will be assayed for the presence of these viruses in the peripheral blood mononuclear cells (PBMCs) of our veteran study groups.

Study 2: Psychosocial Stressors

The following stressful events will be assessed for their role in PGV illness: critical childhood trauma, life events occurring before, during, and following the Gulf war (e.g., divorce, death), and traumatic events associated with PTSD. A Combat Exposure Questionnaire, pertinent to the Gulf war experience, will characterize combat and related stressors. Several investigators have reported that a history of abuse (physical/sexual) (11), parental illness during childhood, serious childhood illness, and parental loss during childhood are variables associated with the development of psychiatric disorders including somatoform disorders (12). Assessment of these variables is important to determine potential premorbid risk factors that could interact with the exposures that occurred in the Gulf war. Similarly, stressful life events have been associated with changes in immunity (13).

Psychiatric status and personality. As mentioned, current and lifetime Axis I psychiatric diagnoses will be assessed with the DIS-IIIR. Special emphasis will

TABLE 4

Indicate how often, if at all, the following statements apply. (In these statements "ill" means having symptoms such as upset stomach, headache, dizziness, or muscle/joint pain.)

	Never	Rarely	Sometimes	Often	Always
1. I feel ill from the odor of pesticide.	0	1	2	3	4
2. I feel ill from the odor of car exhaust.	0	1	2	3	4
3. I feel ill from the odor of cologne, aftershave, or perfume.	0	1	2	3	4
4. I feel ill from walking into a room with brand new carpet.	0	1	2	3	4
5. I feel ill from the odor of paint.	0	1	2	3	4
6. I feel ill from walking down the detergent aisle of the grocery store.	0	1	2	3	4
7. I feel ill from walking into a beauty parlor or barber shop.	0	1	2	3	4
8. I feel ill from reading a freshly printed newspaper.	0	1	2	3	4
I consider myself to be especially sensitive to certain chemicals.				TRUE	FALSE

be given to clarification of PTSD since many symptoms overlap those reported by the PGV (e.g., fatigue and somatic complaints, GI disturbances, joint pain, headache) (14). Any traumatic events that are reported by the PGV will be queried, whether or not these events meet DSM-IIIR criteria as traumatic. Several questions about the qualities of the trauma (e.g., perceived and actual threat of physical harm) will be asked for all traumas reported. Finally, the Mississippi PTSD Scale (15), as modified for the Gulf conflict, will be administered.

Several investigators have reported that the personality trait of negative emotionality is a relatively stable attribute associated with health complaints and illness (16–18). The Neuroticism Scale of the Neuroticism, Extroversion, Openness Inventory (19) will be used to assess personality traits in the PGR veteran. The SCID-III-R will assess DSM-IIIR, Axis II disorders. The Toronto Alexithymia Scale (20) will be used to assess characteristics associated with somatization.

Positive psychosocial moderators. Numerous investigators have reviewed and discussed the mitigating effects of positive affect and coping resources on life stressors (18, 21). The Coping Responses Inventory (22) will assess coping responses to stressful situations. This scale assesses behavioral and emotional methods of coping. Social desirability has been shown to influence response to questionnaires and is associated with the tendency to repress emotions. The latter may lead to symptom development and negative health consequences (18, 23). Therefore, the Marlowe Crowne Social Desirability Scale (24) will be administered.

Symptom checklist. The Profile of Mood States (POMS), the Krupp Fatigue Severity Scale, the Functional Status Questionnaire will be administered as measures of fatigue, disability, and affect. These measures are currently in use in ongoing CFS and MCS studies. Therefore, comparisons between our existing groups and the PGR veteran can be made.

Neuropsychologic assessment. One of the primary complaints of the PGR veteran is memory problems. This complaint is common across several diagnoses to include depression, head injury, neurological conditions, PTSD, CFS, and MCS. Our preliminary results with CFS subjects revealed reduced information processing, and hence encoding of the memory trace rather than an impairment in storage and/or retrieval (25). An MRI brain study in CFS found 27% of CFS subjects had documented evidence of CNS pathology compared to 2% of mild head injury controls (56). Our work with MCS patients revealed that relative to normal controls, MCS subjects had a significant deficit on a visual memory signal detection task (26). Thus standardized assessment of neuropsychologic function accompanied by MRI scans will be included in the protocol for PGV (see Table 5).

Study 3: Autonomic Factors in the Genesis of the PGV's Symptoms

Symptoms of fatigue and dizziness may represent autonomic dysregulation. Moreover, autonomic dysregulation could explain the common CFS complaint of markedly increased fatigue after minimal exertion, a complaint reported by 80% (36) PGVs with moderate or severe CF. CFS patients have been shown to be hypovagal during walking and to show reduced recovery of vagal function during rests as compared to sedentary controls. If orthostatic intolerance is demonstrated, it will support the hypothesis of autonomic dysregulation.

Dependent variables include vagal nerve activity, heart rate, blood pressure, total systemic vascular resistance, and stroke volume, respiration rate, and tidal volume. These variables are being studied before, during, and after the following stressors: postural challenge on a tilt table following thermal vasodilation, deep breathing, lying-to-standing test, Valsalva maneuver, speech preparation and talk, application of cold to the forehead, which elicits a vagal response known

TABLE 5

Vocabulary Subtest of WAIS-R	Measure of verbal intelligence (27)
Digit Span Subtest of WAIS-R	Immediate memory span (27)
Block Design Subtest of WAIS-R	Organization and visuospatial skills (27)
Trail Making Test	Psychomotor speed, attention/concentration in the visual modality (28)
Paced Serial Addition Test	Auditory information processing (29)
Continuous Visual Memory Test	Visual memory (30)
California Verbal Learning Test	Verbal memory (31)
Judgment of Line Orientation	Visual spatial function (32)
Booklet Category Test	Complex problem solving and concept formation (33)
Rey-Osterrieth Complex Figure Test	Organizational skills, visuospatial constructional ability and memory as opposed to visual recognition (34)
Logical Memory Subtest of WMS-R	Short-term learning and recall of verbal material (short stories) (35)

as the "diving reflex" (37), and a Paced Auditory Serial Addition Test. Vagal nerve activity is computed by quantifying the respiratory sinus arrhythmia—heart rate variability at the respiratory frequency—by spectrum analysis (38) because this source of variability in heart rate is known to accurately reflect parasympathetic activity (39).

Study 4: The Caffeine Breath Test as a Marker of P4501A2 Induction

Along with a detailed exposure history, the Caffeine Breath Test (CBT) will be used to infer previous exposure to various polyhalogenated organics including dioxins, dibenzofurans, and various pesticides.

Fatigue, weight loss, neurological symptoms, and skin rashes have characterized populations under study after exposure to polyhalogenated hydrocarbons. Exposure to these chemicals occurs daily in developed society, and from a Gulf war point of view may be particularly increased by combustion of fossil fuel, incineration of human waste and refuse, and possibly after explosion of certain ordnance. Immune dysfunction is prominent in animal studies of the effects of these compounds, and has been suggested in one human cohort who suffered a high exposure. The cytosolic AH receptor, with which these agents bind, controls induction of a hepatic enzyme of the cytochrome P450 super gene family of enzymes, cytochrome P4501A2. In animals, cytochrome P4501A2 is the first biological parameter which is altered by these chemicals, and the degree to which a chemical can induce this enzyme predicts the toxicity of the chemical and the susceptibility of the individual to the toxic effects. Dr. Lambert's recent studies in humans have shown induction of P4501A2 and a correlation between degree of induction and human toxicity (40). Induction can also be detected at levels below the serum detection limit for the chemical itself.

Dr. Lambert and others have designed a sensitive and noninvasive method to quantify P4501A2 activity in the human. The method is the caffeine breath test, which monitors P4501A2-dependent 3 N demethylation of caffeine by monitoring caffeine-derived carbon dioxide in the exhaled breath of the subjects using stable nonradioactive isotopes and mass spectroscopic analysis. Cigarette smoking can act synergistically to induce the enzyme, and thus will be controlled.

PROJECT 3

The focus of Project 3 is to test the autonomic, immunologic, neuropsychologic, and psychologic responses of these veterans to two types of stressors reported to produce symptoms among CFS and MCS patients, i.e., exercise and controlled chemical exposures.

Study 1

Specific aims. (1) Assess responses of CS and matched veteran control subjects to subolfactory threshold inhalation of phenylethyl alcohol (PEA) and to PEA delivered dermally. (2) Substantiate the body burden of PEA achieved through inhalation and dermal exposure routes. (3) Assess physical complaints and mood changes, psychophysiologic responses, nasal cellular influx, reaction time in response to PEA exposure. (4) Measure cytokine gene expression in peripheral blood lymphocytes at baseline and 6 hr after the onset of exposure.

Study 2

Specific aims. (1) Determine circadian patterns of cortisol, ACTH, somatomedin, epinephrine, norepinephrine, and axillary temperature: Day 1, baseline; Day 2, when the subject performs a maximal treadmill test; and Day 3, recovery. (2) Determine viral burden in PBMCs of Epstein-Barr virus, human cytomegalovirus, human herpesvirus 6, and human herpesvirus 7 and profiles of gene expressed cytokines in PBLs of IFN-γ, IL-2, IL-4, IL-5, IL-6, TNF-α, and IL-10 before and after a bout of acute exhaustive exercise. (3) Determine if acute exhaustive exercise impairs cognitive performance disproportionately more in patients than in controls. (4) Determine if a bout of acute exhaustive exer-

cise alters cardiovascular and hormonal reactivity to cognitive stressors proportionately more in patients than in controls. (5) Determine if a bout of acute exhaustive exercise produces an increase in fatigue and other symptoms in patients.

Experimental Strategy, Study 1: Chemical Challenge Study

Rationale. A critical question is whether chemical challenges in a controlled environment will produce objective and subjective responses (e.g., symptoms) among chemically sensitive patients and whether these responses can be distinguished from healthy controls. Nineteen percent of the fatigued veterans reported sensitivities to perfumes. This is a common complaint among MCS patients and selected other groups such as asthmatics in other settings (26). Therefore, PEA, a common component of perfumes, soaps, lotions, and food products (41), was chosen as the volatile organic used in the proposed challenge studies.

Chemically sensitive patients may be psychologically reactive to the idea of exposure. Several methods have been used to block odors, such as masking of an odor with another odor (e.g., peppermint or cinnamon mask) (42), using a physical block such as nose clips, or chemical blocks such as capsaicin. None of these methods is completely satisfactory. First, the odor used as a mask may cause reactions among CS patients since they react to many substances to which healthy controls do not. Second, physical blocks are not completely effective since olfactory cues can be perceived via mouth breathing at the back of the throat. Finally, using a chemical block could also cause a response among these sensitive individuals.

Instead of masking odors, two novel exposure modalities will be used to standardize olfactory cues: subthreshold olfactory inhalation exposure and transdermal exposure. One may question whether subthreshold olfactory exposure will be of sufficient concentration to cause an effect. This may be a meaningful exposure level for two reasons. First, the air concentration of PEA in the bottles presented in olfactory threshold testing is an average of 7 ppm. One standard deviation below this average olfactory threshold concentration is approximately 1 ppm. This concentration is consistent with background levels found in ambient environments such as home, office, and stores—all environments in which MCS patients develop symptoms. Exposures will occur for a period of 2 hr at a steady concentration in the air. This exposure is consistent with those used in Danish studies and at USEPA (43, 44). In addition, these levels of exposure are sufficient to allow documentation of body burden from breath samples.

Dermal exposure offers the best alternate exposure route for avoidance of olfactory cues. The concentration used in the patch and the time required to reach a steady state are comparable to that achieved with inhalation and can be calculated based on pharmacokinetic principles. Due to the uncertainty regarding our ability to document a significant effect with subolfactory thresholds of PEA during inhalation, a higher dose of PEA will be administered transdermally. No odor will be present from this method since the patch will be applied while the forearm is inside a glass box, and it will be kept enclosed for the duration of the experiment. Blood PEA levels will be estimated from exhaled breath to document the body burden during both inhalation and dermal exposures.

Beyond control of psychological response to odors, it is also desirable to assess objective, physiological changes which result from exposures. This avoids reliance on relatively subjective symptom reports. By definition, MCS patients report multiple symptoms reflective of multiple organ systems (45). Objective tests were chosen based on symptomatic responses reported most frequently during olfactory testing.

The symptoms most frequently reported during our olfactory testing procedure were those reflective of upper airway irritation (e.g., runny nose, throat irritation). Increasingly, the nose has been identified as a target for damage due to chemical exposure (46–49). Therefore, interest in methods to measure nasal responses to exposure has increased and methods of measurement have improved. For example, Koren *et al.* (48) used nasal lavage, a relatively noninvasive procedure, to document changes in polymorphonuclear neutrophils (PMN) as markers of inflammation in response to 2-hr, 0.4 ppm exposures to ozone. Immediately postexposure, PMN were increased eightfold and mast cell tryptase levels were increased twofold, whereas PGE2, leukotrienes C4, D4, and E4, C3a, and urokinase-type plasminogen activator were not increased. Albumin was increased only at 18 hr postexposure. Bascom *et al.* (46) also studied ozone at 0.5 ppm for 4 hr, and found increases in PMN eosinophil, and mononuclear cells and albumin, all immediately postexposure. Bascom *et al.* (46) evaluated exposure to environmental tobacco smoke in historically ETS-sensitive subjects and found no increases in inflammatory mediators representative of mast cell activation, including histamine, albumin, kinin, or TAME-esterase activity. Based on these data and symptoms reported in our ongoing PEA threshold studies, cellular inflammation and albumin levels in nasal lavage specimens will be measured before, during, and after exposure in the Controlled Environment Facility (CEF).

Extranasal symptoms such as fatigue, nausea, and headache are also reported in our threshold studies with PEA and pyridine. Changes in interleukins are candidate mediators for these nonfocal symptoms. Thus, interleukin production at baseline and 2 hr after exposure ends will be assayed.

Another prominent symptom among MCS patients

are cognitive complaints of poor concentration and memory. The most consistently sensitive tests are those assessing reaction time (47). In addition, under uncontrolled exposure conditions, MCS patients had significant decrements in performance on a signal detection task of visual memory. Therefore, a computerized signal detection, reaction time task will assess cognitive performance before, during, and after exposure. This test is part of a battery of computerized tests applied widely in epidemiological and controlled exposure studies (48). It is also suitable to be repeated. Finally, exposure conditions could act as a stressor to heighten autonomic reactivity among CS subjects. This could influence symptoms reports and neurobehavioral performance independent of exposure condition. Therefore, we will assess heart and respiratory rates, blood pressure, stroke volume, and total systemic vascular resistance.

Design. Two separate groups of CS subjects will undergo two separate double-blind exposure conditions in a Controlled Environmental Facility (CEF). The CEF is a stainless steel, 887-cubic-foot chamber in which air flow, temperature, and humidity are controlled. Twenty CS PG veterans and 20 matched PG controls will be exposed for 2 hr through inhalation on two separate occasions to PEA at subolfactory threshold (approximately 1 ppm) and to placebo (i.e., clean air). A separate group of 20 CS PG veterans and their matched controls will be exposed dermally for 2 hr on two occasions to PEA and placebo. The order in which exposures occur will be counterbalanced such that half of the CS–control pairs will receive PEA first followed by placebo; the reverse order will be administered for the second half. All exposures will occur in the morning to avoid any time-of-day effects. Neurobehavioral symptom reports, autonomic reactivity, and immunological measures will be taken in the CEU at two separate points: baseline, and the second hour of the 2-hr exposure period. The subjects will then be removed from the CEF to an anteroom and will complete the symptoms questionnaire immediately following exposure. One hour after the end of exposure, the subject will repeat all evaluation measures.

Experimental Strategy, Study 2: Mechanism of Resting and Postexertional Fatigue

Rationale. Of the 63 veterans with chronic fatigue who responded to our questionnaire, 44 (80%) noted a significant exacerbation of fatigue after minimal exertion. This is a common complaint for the CFS patient which frequently produces exacerbation of symptoms. Exertion is known to trigger cytokine release and it is the cytokines which are thought to produce the symptoms of achiness, chills, and malaise which occur in healthy people following strenuous exercise (49). Therefore, the CFS patient may have an abnormal pattern of cytokine release or a hyperactive response to relatively mild exertion.

Another possible explanation involves endocrine function. Patients with CFS are mildly hypoadrenal (50). The hypoadrenal state may produce the malaise, fatigue, and general increase in symptoms reported. Finally, adults with growth hormone deficiency have many of the symptoms of both the fibromyalgic and CFS patients (54–56). Again, exercise might exacerbate that deficiency. No data exist to date on catecholamines (CA) in these patients and it is possible that symptoms following exercise could follow either an increased or a decreased CA response.

Another explanation has to do with thermoregulation during and after exercise. It is possible that long-term consequence of chemical exposure (and/or any chronic fatiguing illness) is an abnormality in thermal control during or after exercise. Were abnormal heat transfer to occur during exercise or were temperature to remain elevated following exercise, these signs of dysfunctional thermoregulation might trigger hormone and/or cytokine abnormalities to produce the symptom exacerbation reported.

Thus, the purpose of this experiment is twofold: first, to study the effects of exertion on CF symptoms, and second, to determine the role of hormones and cytokines in producing the increase in symptoms reported to follow exercise.

Design. An a–b design will be used in which control data will be collected on the first day of hospitalization, during a treadmill walking task, and thereafter. Limitations on subject ability to attend two separate 3-day sessions require this design rather than a separate set of veterans who do not exercise. The other major advantage of this design is that it will allow assessment of circadian rhythms of hormones, cytokines, and temperature. This protects from the possibility of sampling at a time when differences due to exercise are not large.

CONCLUSION

Studies described in this paper focus on the parallels between the unexplained illnesses of the PGV and those illnesses in community samples identified as chronic fatigue and chemical sensitivities. An epidemiologic survey will be used to define symptom clusters among a sample of registry veterans. Veterans meeting criteria for fatiguing illness and/or chemical sensitivities will be recruited for studies to characterize their illness. Specific hypotheses have been developed to test reactions of the veterans to physical and chemical stressors. Thus, the proposed research will help define the PGV illness and will test factors presumed important in the exacerbation of symptoms among chronic fatigue and chemical sensitivities patients.

REFERENCES

1. Holmes, G. P., Kaplan, J. E., Gantz, N. M., Komaroff, A. L., Schonberger, L. B., Strauss, S. E., *et al.* Chronic fatigue syndrome: A working case definition. *Ann. Int. Med.* 1988; **108:** 387–389.
2. Fukuda, K., Straus, S. E., Hickie, I., Sharpe, M. C., Dobbins, J. G., Komaroff, A., and the International Chronic Fatigue Syndrome Study Group. The chronic fatigue syndrome: A comprehensive approach to its definition and study. *Am. Coll. Phys.* 1994; **121**(12): 953–959.
3. Buchwald, D., and Garrity, D. Comparison of patients with chronic fatigue syndrome, fibromyalgia, and multiple chemical sensitivities. *Arch. Int. Med.* 1994; **154:** 2049–2053.
4. Simon, G. E., Katon, W. J., and Sparks, P. J. Allergic to life: Psychological factors in environmental illness. *Am. J. Psychiat.* 1990; **147:** 901–906.
5. Tapp, W. N., and Natelson, B. H. Consequences of stress: A multiplicative function of health status. *FASEB J.* 1988; **2:** 2268–2271.
6. Bell, I. R., Schwartz, G. E., Peterson, J. M., Amend, D., and Stini, W. A. Possible time-dependent sensitization to xenobiotics: Self-reported illness from chemical odors, foods, and opiate drugs in an older adult population. *Arch. Environ. Health* 1993; **48**(5): 315–327.
7. Stewart, J., and Badiani, A. Tolerance and sensitization to the behavioral effects of drugs. *Behav. Pharmacol.* 1993; **4:** 289–312.
8. DuBois, R. W., Seeley, J. K., Brus, I., Sakamoto, K., Ballow, M., Harada, S., Bechtold, T. A., Pearson, G., and Purtilo, D. T. Chronic mononucleosis syndrome. *Southern Med. J.* 1984; **77:** 1376–1382.
9. Holmes, G. P., Kaplan, J. E., Stewart, J. A., Hunt, B., Pinsky, P. F., and Schonberger, L. B. A cluster of patients with chronic mononucleosis-like syndrome. *JAMA* 1987; **257:** 2297–2302.
10. Josephs, S. F., Henry, B., Balachandran, N., Strayer, D., Peterson, D., Komaroff, A. L., and Ablashi, D. V. HHV-6 reactivation in chronic fatigue syndrome. *Lancet* 1991; **337:** 1346–1347.
11. Teicher, M. H., Glod, C. A., Surrey, J., and Swett, C. Early childhood abuse and limbic system ratings in adult psychiatric outpatients. *J. Neuropsychiat.* 1993; **5:** 301–306.
12. Harris, T., Brown, G. W., and Bifulco, A. Loss of parent in childhood and adult psychiatric disorder: The role of lack of adequate parental care. *Psychol. Med.* 1986; **16:** 641–659.
13. Herbert, T. B., and Cohen, S. Stress and immunity in humans: A meta-analytic review. *Psychosomat. Med.* 1993; **55:** 364–379.
14. Sutker, P. B., Bugg, F., and Allain, A. N. Psychometric prediction of PTSD among POW survivors. *J. Consult. Clin. Psychol.* 1991; **3:** 105–110.
15. Keane, T. M., Caddell, J. M., and Taylor, K. L. *The Mississippi Scale for Combat-Related PTSD.* VA Medical Center, Boston, MA, 1986.
16. Costa, P. T., and McCrae, R. R. Neuroticism, and aging: When are somatic complaints unfounded? *Am. Psychol.* 1985; **40**(1): 19–28.
17. Friedman, H. S., and Booth-Kewley, S. The "disease-prone personality": A meta-analytic view of the construct. *Am. Psychol.* 1987; **42**(6): 539–555.
18. Watson, D., and Pennebaker, J. W. Health complaints, stress, and distress: Exploring the central role of negative affectivity. *Psychol. Rev.* 1989; **96:** 234–254.
19. Costa, P. T., and McCrae, R. R. The NEO personality inventory. Psychological Assessment Resources, Odessa, FL, 1985.
20. Taylor, G. J., Ryan, D., and Bagby, R. M. Toward the development of a new self-report alexithymia scale. *Psychother. Psychosomat.* 1985; **44:** 191–199.
21. Miller, S. M. Monitoring and blunting: Validation of a questionnaire to assess styles of information seeking under threat. *J. Pers. Soc. Psychol.* 1987; **52:** 345–353.
22. Moos, R. H. *Coping Responses Inventory: Adult Form Manual.* Center for Health Care Evaluation, Stanford University and Dept. of Veterans Affairs Medical Centers, Palo Alto, CA, 1992.
23. Jensen, M. R. *Psychophysiological Factors in the Prognosis and Treatment of Neoplastic Disorders* (Unpublished Doctoral Dissertation, Yale University, 1984.
24. Crowne, D. P., and Marlowe, D. *The Approval Motive: Studies in Evaluation Dependence.* Wiley, New York, 1964.
25. DeLuca, J., Johnson, S. K., and Natelson, B. H. Information processing efficiency in chronic fatigue syndrome and multiple sclerosis. *Arch. Neurol.* 1993; **50:** 301–304.
26. Fiedler, N., Kipen, H., DeLuca, J., Kelly-McNeil, K., and Natelson, B. A controlled comparison of multiple chemical sensitivity and chronic fatigue syndrome. *Psychosomat. Med.* 1996; **58:** 38–49.
27. Wechsler, D. *Wechsler Adult Intelligence Scale—Revised.* The Psychological Corporation, San Antonio, TX, 1991.
28. Trites, R. L. *Neuropsychological Test Manual.* Technolab, Montreal, 1981.
29. Brittain, J. L., LaMarche, J. A., Reeder, K. P., Roth, D. L., and Boll, T. J. Effects of age and IQ on paced auditory serial addition task (PASAT) performance. *Clin. Neuropsychol.* 1991; **5:** 163–175.
30. Trahan, D. E., and Larrabee, G. J. *Continuous Visual Memory Test.* Psychological Assessment Resources, Odessa, FL, 1988.
31. Benton, A. L., deS. Hamsher, K., and Varney, N. R. Judgment of Line Orientation. In *Contributions to Neuropsychological Assessment—A Clinical Manual* (Benton *et al.*, Eds.). Oxford University Press, New York, 1994.
32. Delis, D. C., Kramer, J. H., Kaplan, E., and Ober, B. A. *California Verbal Learning Test.* The Psychological Corporation, San Antonio, TX, 1987.
33. DeFilippis, N. A., and McCampbell, E. *The Booklet Category Test.* Psychological Assessment Resources, Odessa, FL, 1979.
34. Viser, R. S. H. *Manual for the Complex Figure Test.* Sweta & Zeitlinger, The Netherlands, 1985.
35. Wechsler, D. *Wechsler Memory Scale—Revised.* The Psychological Corporation, San Antonio, TX, 1987.
36. Levin, A. S., and Byers, V. S. Environmental illness: A disorder of immune regulation. In *Workers with Multiple Chemical Sensitivities: Occupational Medicine State of the Art Review.* M. R. Cullen, 1994: 669–681.
37. Kawakami, Y., Natelson, B., and Dubois, A. Cardiovascular effects of face immersion and factors affecting diving reflex in man. *J. Appl. Physiol.* 1967; **23:** 964–970.
38. Tapp, W. N., Knox, F. S., III, and Natelson, B. H. The heart rate spectrum in simulated flight: Reproducibility and effects of atropine. *Aviat. Space Environ. Med.* 1990; **61:** 887–892.
39. Akselrod, S., Gordon, D., Madwed, J. B., *et al.* Hemodynamic regulation: Investigation by spectral analysis. *Am. J. Physiol.* 1985; **248:** H867–H875.
40. Kipen, H. M., Fiedler, N., Maccia, C., Turkow, E., Todaro, J., and Laskin, D. Immunological evaluation of chemically sensitive patients. *Toxicol. Ind. Health* 1992; **8**(4): 125–135.
41. Ford, R. A. Metabolic and kinetic criteria for the assessment of reproductive hazard. In *Basic Science in Toxicology* (Volans, G. N., Sims, J., Sullivan, F. M., and Turner, P., Eds.), pp. 59–68. Taylor & Francis, London, 1990.
42. Selner, J. C., and Staudenmayer, H. The practical approach to

the evaluation of suspected environmental exposures: Chemical intolerance. *Ann. Allergy* 1985; **55:** 665–673.

43. Hudnell, H. K., Otto, D. A., House, D. E., and Molhave, L. Exposure of humans to a volatile organic mixture. II. Sensory. *Arch. Environ. Health* 1992; **47:** 31–38.
44. Molhave, L., Bach, B., and Pedersen, O. F. Human reactions to low concentrations of volatile organic compounds. *Environ. Int.* 1986; **12:** 167–175.
45. Cullen, M. R. Workers with multiple chemical sensitivities. Hanley and Belfus, Philadelphia, 1987.
46. Bascom, R., Kulle, T., Kagey-Sobotka, A., and Proud, D. Upper respiratory tract environmental tobacco smoke sensitivity. *Am. Rev. Respir. Dis.* 1991; **143:** 1304–1311.
47. Koren, H. S., and Devlin, R. B. Human upper respiratory tract responses to inhaled pollutants with emphasis on nasal lavage. *Ann. NY Acad. Sci.* 1992; **641:** 215–224.
48. Koren, H. S., Hatch, G. E., and Graham, D. E. Nasal lavage as a tool in assessing acute inflammation in response to inhaled pollutants. *Toxicology* 1990; **60:** 15–25.
49. Witek, T. J. The nose as a target for adverse effects from the environment: Applying advances in nasal physiologic measurements and mechanisms. *Am. J. Ind. Med.* 1993; **24:** 649–657.
50. Gamberale, F. Use of behavioral performance tests in the assessment of solvent toxicity. *Scand. J. Work Environ. Health* 1986; **11:** 65–74.
51. Letz, R., and Baker, E. L. Neurochemical evaluation system. Neurobehavioral Systems, New York, 1988.
52. Cannon, J. G., Fielding, R. A., Fiatarone, M. A., Orencole, S. F., Dinarello, C. A., and Evans, W. J. Increased interleukin 1B in human skeletal muscule after exercise. *Am. J. Physiol.* 1989; **257:** R451–R455.
53. Demitrack, M. A., Dale, J. K., Straus, S. E., Laue, L., Listwak, S. J., Kruesi, M. J. P., Chrousos, G. P., and Gold, P. W. Evidence for impaired activation of the hypothalamic-pituitary-adrenal axis in patients with chronic fatigue syndrome. *J. Clin. Endocrinol. Metab.* 1991; **73:** 1224–1234.
54. Bennett, R. M., Clark, S. R., Campbell, S. M., and Burckhardt, C. S. Low levels of somatomedin C in patients with fibromyalgia syndrome: A possible link between sleep and muscle pain. *Arthritis Rheum.* 1992; **35:** 1113–1116.
55. Bennett, R. M., Clark, S. R., Campbell, S. M., and Burckhardt, C. S. Low levels of somatomedin C in patients with the fibromyalgia syndrome. *Arthritis Rheum.* 1992; **35:** 1117–1121.
56. Cuneo, R. C., Salomon, F., McGauley, G. A., and Sonksen, P. H. The growth hormone deficiency syndrome in adults. *Clin. Endocrinol.* 1992; **37:** 387–397.