

## Butyrylcholinesterase in Relation to Gulf War Illness:

### Preliminary Evidence of Gene-Exposure Interaction in 1991 Gulf War Veterans



Lea Steele, Ph.D.  
Director, Veterans Health Research  
Baylor Institute of Biomedical Studies

RAC-GWVI  
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## 1990-1991 Gulf War:

### Operations Desert Shield/Desert Storm



Aug 2, 1990 - Iraq invaded Kuwait

Jan 16, 1991 - Air strikes began

Feb 24, 1991 - Ground combat began

Feb 28, 1991 - Cease fire declared

- 6 weeks air strikes, 4 day ground war

- 37 countries in Allied Coalition

~ 700,000 U.S. troops

- Decisive victory; relatively few casualties



**1991: Returning Veterans Report Difficult Symptoms,  
Not Explained by Familiar Medical or Psych Diagnoses**

**"Gulf War Syndrome:" Widespread reports of persistent health problems**

- *Chronic headaches*
- *Widespread pain*
- *Memory and concentration difficulties*
- *Persistent, unexplained fatigue*
- *Chronic diarrhea*
- *Respiratory problems*
- *Mood changes*
- *Unusual skin rashes*

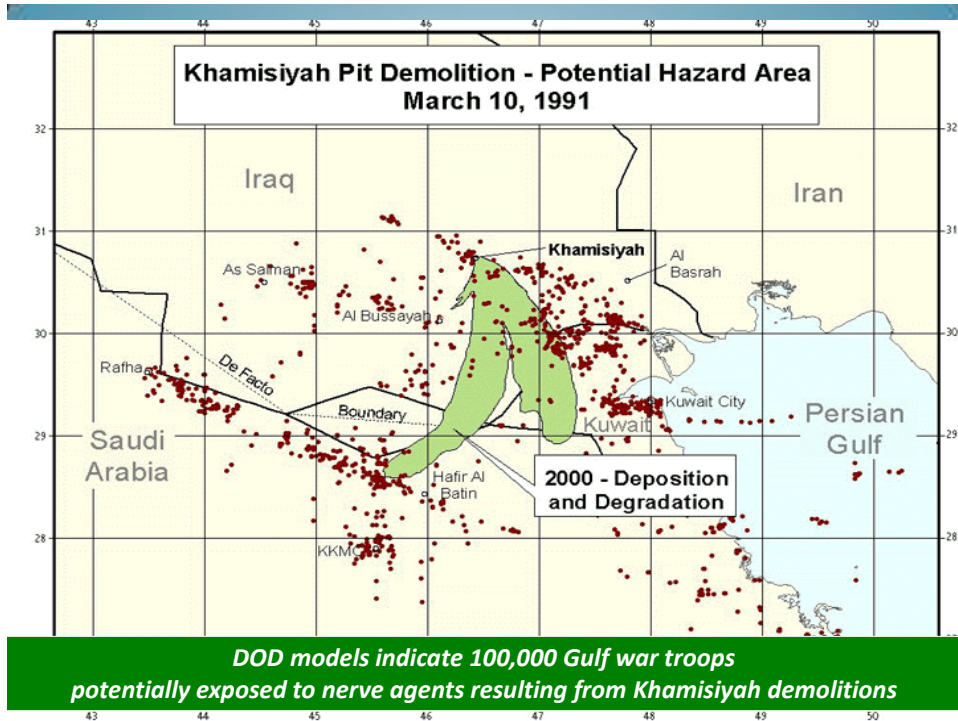


***What Caused Gulf War Illness?***

*Early on, many suspected causes/contributors*









## Widespread use, overuse of pesticides and insect repellants

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

Compound	Use	Chemical Class	Purpose	Application
<i><u>Pesticides and Repellants Used by the General Military Population</u></i>				
DEET, 33% cream, stick	Personal use repellent	Dialkylamide	Repel flies and mosquitoes	By hand to skin
DEET, 75% liquid	Personal use repellent	Dialkylamide	Repel flies and mosquitoes	By hand to skin, uniform, netting
Permethrin, 0.5% spray	Personal use repellent	Pyrethroid	Repel flies and mosquitoes	Sprayed on uniforms
d-Phenothrin, 0.2% aerosol	Area use repellent	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
<i><u>Pesticides Used by Pesticide Applicators</u></i>				
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
<i><u>Delousing Pesticide</u></i>				
Lindane, 1% powder	Delouser	Organochlorine	Kill lice, other insects	Dusted on prisoners, also for personal use

Source: DOD Environmental Exposure Report: Pesticides (2003)<sup>152</sup>



### Large number of Gulf War-related experiences/exposures of potential concern

- *Psychological stress, trauma*
- *Chemical weapons*
- *Oil well fires*
- *Munitions containing depleted uranium*
- *Heavy use of insecticides/repellants*
- *PB pills (pyridostigmine bromide)*
- *Vaccines*
- *Infectious diseases*
- *Tent heaters*
- *Particulates*
- *Fuel exposures*
- *Solvents, CARC paint*



### Association of Deployment Experiences/Exposures with Gulf War Illness

#### Synthesis of Evidence from Epidemiologic Studies of Gulf War Veterans

- Psychological stressors	Evidence indicates <u>no</u> association
- Pyridostigmine bromide (PB) - Pesticides	Evidence consistently indicates a <u>significant</u> association
- Low-level nerve agents - Sustained oil well smoke - Large number of vaccines - Combinations of exposures	Unclear, association cannot be ruled out; evidence is inconsistent or limited in important ways
- Depleted uranium - Anthrax vaccine - Fuels, solvents - Sand, particulates - Other	Little evidence of association; unlikely to have been primary contributing factor for the majority of affected veterans



**24 Years After Desert Storm:  
What Have We Learned About the Cause(s) of Gulf War Illness?**

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- Etiology appears complex; important to evaluate health outcomes in veteran subgroups
- Studies most consistently implicate anti-nerve gas pills (PB), and excess use of pesticides
- low-level exposure to chemical weapons, oil fires, chemical combinations not ruled out

*Chemical risk factors of greatest concern are toxic to the brain/nervous system. Many are in single class of toxicants (AChE inhibitors)*

**Question: Why did some veterans develop GWI, while others remained well?**

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**Individual Differences in Vulnerability to Adverse Effects of Neurotoxicants?**

- A number of circulating enzymes protect us from adverse effects of some neurotoxicants (e.g. carbamates, OPs); long speculation that GWI risk may be associated with **genetic variability in protective enzymes**
- Paraoxonase (PON1): Previous studies have suggested possible association of GWI with PON1 enzyme activity levels or genotype
- Butyrylcholinesterase (BChE): Enzyme that binds acetylcholinesterase inhibitors, protects from adverse effects

## Paraoxonase (PON1) in Relation to GWI

### Studies Mixed; Overall Picture Difficult to Compare, Interpret

- **Haley et al 1999: Small study of Navy Seabees: PON1 activity (paraoxon) nonsign. higher in cases; type Q arylesterase activity sign. lower in cases. More cases than controls had PON1<sub>192</sub> R allele.**
- **Mackness et al 2000: 152 U.K. Gulf War veterans had lower PON1 activity (in paraoxon) than nondeployed comparison group; no genetic differences**
- **Hotopf et al 2003: 210 U.K. Gulf War veterans had lower PON1 activity than nondeployed veterans; no association with illness, no genetic differences**
- **Concato et al 2007: no PON1 activity differences in GWI cases vs. controls**



## Is GWI Associated with BChE Enzyme Activity or Genotype?

Steele et al. *Environmental Health* 2015, 14:4  
<http://www.ehjournal.net/content/14/1/4>



RESEARCH

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### Butyrylcholinesterase genotype and enzyme activity in relation to Gulf War illness: preliminary evidence of gene-exposure interaction from a case-control study of 1991 Gulf War veterans

Lea Steele<sup>1\*</sup>, Oksana Lockridge<sup>2</sup>, Mary M Gerkovich<sup>3</sup>, Mary R Cook<sup>4</sup> and Antonio Sastre<sup>5</sup>

#### Abstract

**Background:** Epidemiologic studies have implicated wartime exposures to acetylcholinesterase (AChE)-inhibiting chemicals as etiologic factors in Gulf War illness (GWI), the multisymptom condition linked to military service in the 1991 Gulf War. It is unclear, however, why some veterans developed GWI while others with similar exposures did not. Genetic variants of the enzyme butyrylcholinesterase (BChE) differ in their capacity for metabolizing AChE-inhibiting chemicals, and may confer differences in biological responses to these compounds. The current



**Collaborators:**

**Midwest Research Institute (now MRI Global)**

**Antonio Sastre, PhD  
Mary Cook, PhD  
Mary Gerkovich, PhD**

**Eppley Institute, University of Nebraska Medical School**

**Oksana Lockridge, PhD**

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**Butyrylcholinesterase (BChE): Overview**

- BChE: present at > 10X level of AChE in blood; physio functions not well understood. Acts as scavenger, binds organophosphates, carbamates, other compounds
- Early pharmacogenetics: Patients with abnormal response to succinylcholine (e.g. given for surgery) found to have inherited BChE deficiency. Produces prolonged paralysis/unable to breathe after dose that normally acts for few minutes
- One BChE gene, multiple variations: Wild type "U" allele most common; then K, A, F
- Reduced serum BChE activity routinely used as a biomarker for pesticide exposure
- BChE currently being developed as a prophylactic measure to protect against effects of nerve agents (safer alternative to PB)

## Is GWI Associated with BChE?

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### Background

- Lowenstein-Lichtenstein (1995) [Soreq lab]: Case report of Israeli soldier—severe symptoms with PB pills during the Gulf War; found to be BChE AA homozygote, poor BChE affinity for carbamates
- Lockridge: 1999 DOD project: Nebraska GW veterans: most (73%) carriers of A, F alleles had reported they had Gulf War syndrome (vs < 30% overall)

## Butyrylcholinesterase in Relation to Gulf War illness

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### Study Design

- Assessed both BChE enzyme activity and genotype in 304 1991 Gulf War veterans
- Population-based sample: 144 GWI cases (KS case def), 160 GW veteran controls
- Determined if risk associated with “cholinergic exposures” (e.g. pesticides, PB, possible nerve agent exposure) differed with BChE genotype

### Study: Butyrylcholinesterase in Relation to Gulf War illness

- **BChE subgroup analyses: Compared variant subgroups: “common” genotypes (UU and UK), “LCV--Less Common Variants” (K/K, U/AK, U/A, A/F, A/K,F)**
  - *“Normal” BChE variants: enzyme effective at neutralizing AChE inhibitors*
  - *“Less common” variants: enzyme acts more slowly, less effective at neutralizing some chemicals*
- **Overall, BChE enzyme activity and genotype of GWI cases were similar to controls**



### Results: Butyrylcholinesterase Enzyme Activity Levels

	n	BChE Mean (SD) Activity*
All veterans in study	304	1.10 (0.26)
<b>BChE genotype</b>		
U/U	189	1.19 (0.24)
U/K	87	1.01 (0.21)
K/K	13	0.80 (0.15)
U/AK	10	0.76 (0.18)
U/A	3	1.03 (0.12)
A/F	1	0.92
AK/F	1	0.69
Common variants combined (U/U and U/K)	276	1.13 (0.24)
Less common variants combined (K/K, U/AK, U/A, A/F, AK/F)	28	0.81 (0.17) <sup>†</sup>

\*Enzyme activity expressed in umoles benzoylcholine hydrolyzed per minute per ml. serum



### Results: Butyrylcholinesterase Enzyme Activity Levels

<u>Gulf War illness case status</u>	n	BChE Mean (SD) Activity
<b>Gulf War illness cases</b>	<b>144</b>	<b>1.10 (0.24)</b>
<b>Gulf War veteran controls</b>	<b>160</b>	<b>1.10 (0.27)</b>

Enzyme activity expressed in umoles benzoylcholine hydrolyzed per minute per ml. serum



### Results: Distribution of Butyrylcholinesterase Genotype

BChE Genotype	Total Sample (n = 304)		GWI Cases (n = 144)		Controls (n = 160)	
	n	(%)	n	(%)	n	(%)
U/U	189	(62%)	89	(62%)	100	(62%)
U/K	87	(29%)	41	(28%)	46	(29%)
K/K	13	(4%)	7	(5%)	6	(4%)
U/AK	10	(3%)	5	(3%)	5	(3%)
U/A	3	(1%)	1	(<1%)	2	(1%)
A/F	1	(<1%)	0	(0%)	1	(<1%)
AK/F	1	(<1%)	1	(<1%)	0	(0%)
Less common variants combined (K/K, U/AK, U/A, A/F, AK/F)	28	(9%)	14	(10%)	14	(9%)

### Association of Gulf War Illness with Cholinergic Exposures, by BChE Genetic Subgroup

Experience/Exposure	All Gulf War Veterans
Took PB pills	OR = 3.21*
Wore pesticide-treated uniforms	OR = 3.72*
Used pesticides on skin	OR = 2.89*
Living area sprayed with pesticides	OR = 1.33
Heard chemical alarms sounded	OR = 1.31

\*p < 0.001  
 Ref: Steele et al (2015) Environ Health 14:4



### Association of Gulf War Illness with Cholinergic Exposures, by BChE Genetic Subgroup

Experience/Exposure	All Gulf War Veterans	BChE Common Variants (normal activity) UU and U/K (n=276)
Took PB pills	OR = 3.21*	OR = 2.68*
Wore pesticide-treated uniforms	OR = 3.72*	OR = 3.63*
Used pesticides on skin	OR = 2.89*	OR = 3.14*
Living area sprayed with pesticides	OR = 1.33	OR = 1.30
Heard chemical alarms sounded	OR = 1.31	OR=1.26

\*p < 0.001  
 Ref: Steele et al (2015) Environ Health 14:4



### Association of Gulf War Illness with Cholinergic Exposures, by BChE Genetic Subgroup

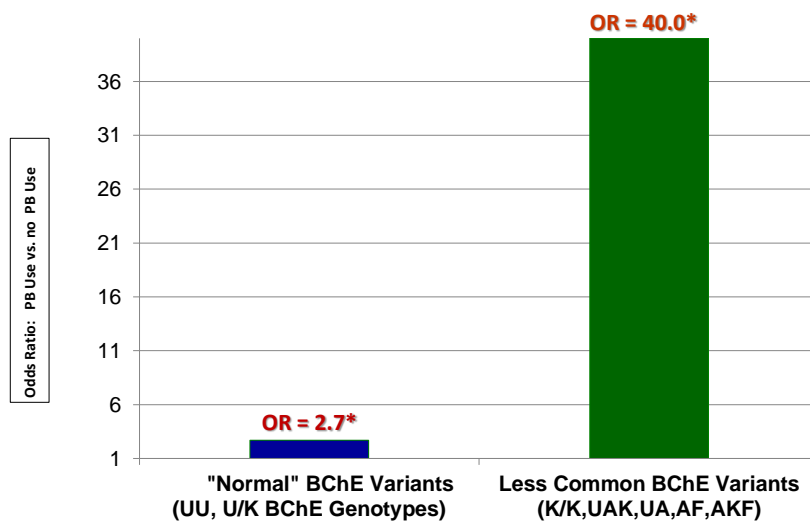
Experience/Exposure	All Gulf War Veterans	BChE Common Variants (normal activity) UU and U/K (n=276)	BChE Less Common Variants (slow-acting) KK,UAK,UA,AF,AKF (n=28)
Took PB pills	OR = 3.21*	OR = 2.68*	OR = 40.00*
Wore pesticide-treated uniforms	OR = 3.72*	OR = 3.63*	OR = 4.80
Used pesticides on skin	OR = 2.89*	OR = 3.14*	OR = 1.33
Living area sprayed with pesticides	OR = 1.33	OR = 1.30	OR = 1.64
Heard chemical alarms sounded	OR = 1.31	OR=1.26	OR = 1.80

\*p < 0.001  
 Ref: Steele et al (2015) Environ Health 14:4



#### Preliminary Indication of Significant Gene-Exposure Interaction

#### Risk of Gulf War Illness in Relation to PB Use, by BChE Genetic Subgroup



Ref: Steele et al (2015) Environ Health 14:4



### Can Apparent BChE x PB Interaction be Explained by Study Factors?

- **Did BChE-LCV subgroup differ from other Gulf War veterans in other ways?**
  - Demographically (sex, age, education) almost identical
  - Exposures: nearly identical (e.g. 58% vs. 57% reported PB use)
  
- **Was association with BChE an anomaly of KS GWI case definition?**
  - Test by reassigning case/control status using CDC criteria
  - Yields 187 CMI cases, 117 controls



### Association of PB with Gulf War Illness in BChE Genetic Subgroups

#### Evaluated Using Two Case Definitions

Case Definition	All Gulf War Veterans	BChE Common Variants (normal activity) UU and U/K (n=276)	BChE Less Common Variants (slow-acting) KK,UAK,UA,AF,AKF (n=28)
GWI—Kansas Case Definition	OR = 3.21*	OR = 2.68*	OR = 40.00*
CMI—Fukuda/CDC Case Def	OR = 1.99*	OR = 1.73*	OR = 11.37*

\*p < 0.01  
 Ref: Steele et al (2015) Environ Health 14:4



## Context

- **Study provides preliminary evidence that BChE-LCV subgroup (K homozygotes; A,F heterozygotes) who used PB were at substantially increased risk for GWI**
- **Consistent with:**
  - 2 preliminary reports of BChE-associated risk in Gulf War veterans
  - 2 rat studies indicate delayed neuro effects of low-dose PB only in strain with low BChE enzyme activity

### Caveats:

- **Small sample (n=304) included only 28 with “at risk” genotypes**
- **Hard to estimate the total number GWV affected by this interaction; based on our study, this interaction would, at most, explain ~ 10 % of GWI cases**

## Conclusions

- **Study provides preliminary evidence of significant gene-exposure interaction; subgroup of Gulf War veterans with less active genetic forms of BChE who used PB were at significantly greater risk for GWI, compared to other veteran subgroups**
- **Highly significant association, OR = 40 for BChE LCV subgroup**
  - Small (but representative) sample: association not explained by study factors
  - Requires replication in larger sample
- **Analytic methods important: Factors related to possible genetic “vulnerability” only apparent when exposures considered**

### Additional BChE questions:

- **“Situational” reduced BChE activity in theater associated with GWI for other veterans?**
- **Implications for animal models: species differences in detoxicating enzymes, including BChE**



**Approach used in BChE study may provide a foundation/model for further evaluation of gene-exposure interactions in GWI**

- **As in other areas of Gulf War research, essential to evaluate appropriate subgroups**
- **Genetic variants that confer reduced ability to neutralize certain chemicals would not be expected to yield poor health outcomes in the absence of those exposures**
- **Prior PON1 studies did not evaluate association of GWI with genes in relation to wartime exposures; evaluated genotype, enzyme activity in:**
  - *GW deployed vs. nondeployed*
  - *Veterans with GWI vs healthy controls (either GW vets or nondeployed)*

## Paraoxonase (PON1)

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### Are differences in PON1 genotype associated with Gulf War Illness?

#### A Different Approach

## Paraoxonase (PON1): Background

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- **PON1: Hydrolyzes (inactivates) organophosphate (OP) compounds (AChE inhibitors), including insecticides and nerve agents**
- **PON1 differences thought to confer differing sensitivity to specific OP compounds:**

**PON1<sub>192</sub> - Q variant most effective at hydrolyzing nerve agents**

**PON1<sub>192</sub> - R variant most effective at hydrolyzing some pesticides (e.g. chlorpyrifos, parathion)**

### Exploratory GWI - PON1 Evaluation in a Case-Control Study

- **Population-based sample (n=72); "high risk" 1991 Gulf War veterans**
  - 40 GW veterans with GWI (GWI cases)
  - 18 GW veteran controls
  - 14 nondeployed/era veteran controls
  - No case/control differences by sex, age, race, education
  
- **GW sample: GW veterans at highest risk for GWI**
  - All served in Army, enlisted ranks, forward deployed
  - In units identified as potentially exposed to nerve agents
  
- **Highly exposed cohort; GWI sign. associated with degree of exposure to PB, pesticides**



### Association of GWI with GW Exposures, by PON1<sub>192</sub> Genotype

**ORs for GW Cases (n=40) vs. GW controls (n=18)**

Experience/Exposure	All Gulf War Veterans			
Chemical alarms sounded	OR = 1.90 (ns)			
Used skin pesticides	OR = 1.03 (ns)			
Wore pesticide-treated uniforms	OR = 3.10 (ns)			
Took PB 1 week or longer	<b>OR = 3.41*</b>			

\*statistically significant, p<0.05

**Association of GWI with GW Exposures, by PON1<sub>192</sub> Genotype**

**ORs for GW Cases (n=40) vs. GW controls (n=18)**

Experience/Exposure	All Gulf War Veterans	PON1 QQ Genotype (n=31)	PON1 QR/RR Genotypes (n=27)	sign. intrxn?
Chemical alarms sounded	OR = 1.90 (ns)	OR = 0.75 (ns)	<b>OR = 7.56*</b>	~
Used skin pesticides	OR = 1.03 (ns)	OR = 2.33 (ns)	OR = 0.20 (ns)	~
Wore pesticide-treated uniforms	OR = 3.10 (ns)	<b>OR = 21.0*</b> <small>[logit est]</small>	OR = 0.72 (ns)	<b>0.02*</b>
Took PB 1 week or longer	<b>OR = 3.41*</b>	<b>OR = 11.20*</b>	OR = 0.74 (ns)	<b>0.04*</b>

\*statistically significant, p<0.05

**Summary**

- **Preliminary Evidence: Suggests Association of GWI with Exposures May Differ in Relation to PON1 Genotype**
  - ✓ Recall PON1 R variant has better capacity for hydrolyzing some pesticides, but provides poorer hydrolysis of sarin
  - ✓ Veterans with R allele were at sign. greater risk for GWI if they heard chemical alarms (QQ homozygotes were not)
  - ✓ Use of pesticides and PB were sign. associated with GWI only in PON1 QQ homozygotes (not in veterans with PON1 R allele)



**Preliminary Indication:  
Association of GWI with GW Exposures May Differ in Relation to PON1**

- Supports approach described for BChE study, i.e. that understanding impact of genetic vulnerability to neurotoxicants requires evaluation of GWI-exposure interaction in PON1 genetic subgroups
- Caution: Small sample size; findings are exploratory
- Identified subgroup associations are in directions that might be expected in relation to PON1 genotype



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## - EXTRA SLIDES

### **24 Years After Desert Storm: What Have We Learned About Gulf War Illness (GWI)?**

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- Affects 25 – 30 % of the 700,000 U.S. personnel who served in 1991 Gulf War
- GWI not due to stress or psych disorder; PTSD rates low in GW vets
- No widespread “GWI”-type problem in OIF, OEF veterans
- Higher GWI rates in ground troops, enlisted personnel, forward-deployed
- Few veterans have recovered over time; many have been sick for ~ 24 yrs

Table 3 Association of Gulf War illness with exposures with potential cholinergic effects, by butyrylcholinesterase genetic subgroup

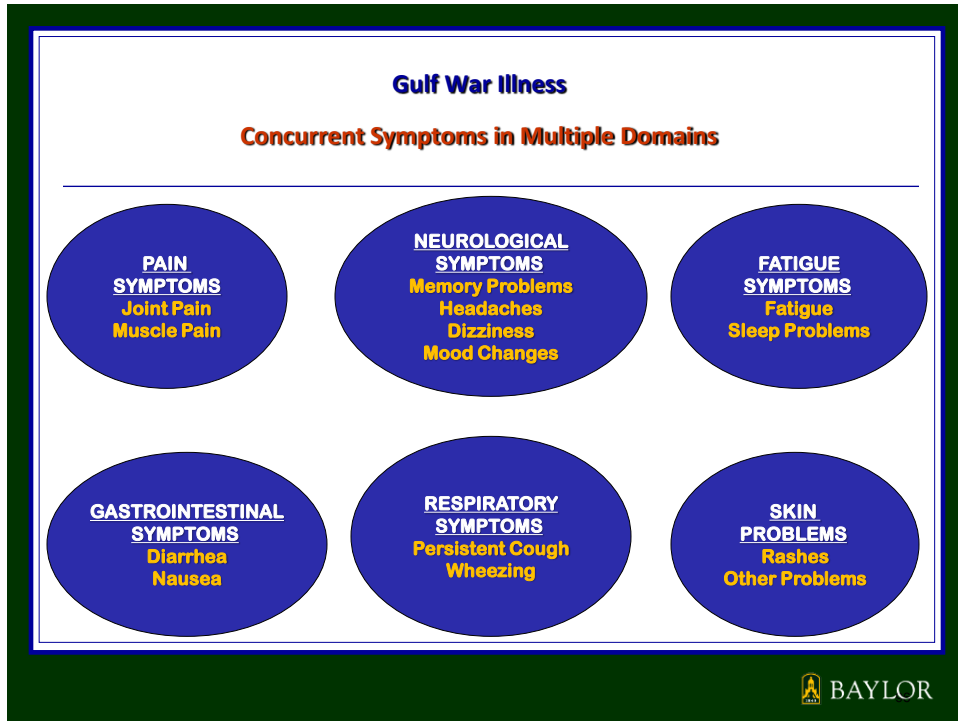
Experience/Exposure	Butyrylcholinesterase Genetic Subgroup								
	U/U Homozygotes (n = 89 GWI cases, 100 controls)			U/K Heterozygotes (n = 41 GWI cases, 46 controls)			Less Common BChE Variants (K/K, U/AK, U/A, A/F, AK/F) (n = 14 GWI cases, 14 controls)		
	% Cases Exposed	% Controls Exposed	OR <sup>†</sup> (95% C.I.)	% Cases Exposed	% Controls Exposed	OR <sup>†</sup> (95% C.I.)	% Cases Exposed	% Controls Exposed	OR <sup>†</sup> (95% C.I.)
Took PB (pyridostigmine bromide) pills	67%	48%	2.25 (1.23-4.11)	74%	41%	3.98 (1.57-10.1)	92%	23%	<b>40.0<sup>†</sup></b> (3.58-447)
Wore uniforms treated with pesticides	22%	8%	3.07 (1.27-7.44)	37%	11%	4.92 (1.59-15.2)	29%	8%	4.80 (0.46-50.2)
Used pesticide cream or spray on skin	55%	31%	2.75 (1.50-5.02)	62%	28%	4.23 (1.71-10.5)	50%	43%	1.33 (0.31-5.91)
Living area sprayed with pesticides	21%	18%	1.23 (0.59-2.57)	24%	17%	1.47 (0.51-4.29)	21%	14%	1.64 (0.23-11.7)
Heard chemical alarms sounded	57%	57%	1.02 (0.57-1.82)	62%	44%	2.00 (0.84-4.79)	64%	50%	1.80 (0.40-8.18)

BChE=butyrylcholinesterase, GWI=Gulf War illness, OR=prevalence odds ratio, C.I.=confidence interval  
<sup>†</sup>OR compares GWI cases with controls within each genetic subgroup

Table 4 Association of pyridostigmine bromide use with chronic illness in BChE subgroups: evaluation using two case definitions

Case Definition	Use of PB by Butyrylcholinesterase Genetic Subgroups								
	Use of PB by All Gulf War Veterans (n = 304)			Common BChE Variants (U/U and U/K) (n=276)			Less Common BChE Variants (K/K, U/AK, U/A, A/F, AK/F) (n=28)		
	% Cases Exposed	% Controls Exposed	OR (95% C.I.)	% Cases Exposed	% Controls Exposed	OR (95% C.I.)	% Cases Exposed	% Controls Exposed	OR (95% C.I.)
GWI (Kansas case definition) <sup>†</sup>	72%	44%	3.21 (1.97-5.24)	69%	46%	2.68 (1.62-4.44)	92%	23%	<b>40.00</b> (3.58-447)
CMI (CDC case definition) <sup>†</sup>	63%	46%	1.99 (1.23-3.22)	62%	49%	1.73 (1.05-2.84)	78%	22%	<b>11.37</b> (1.65-78.4)

BChE=butyrylcholinesterase, PB=pyridostigmine bromide, CDC=U.S. Centers for Disease Control and Prevention, OR=prevalence odds ratio, C.I.=confidence interval  
<sup>†</sup>Gulf War illness, as defined in Steele [1].  
 Chronic multisymptom illness, as defined in Fukuda et al [32].



**Summary: Association of GWI with Deployment Experiences/Exposures Across GW Veteran Populations**

Epidemiologic Studies of Gulf War Veterans:  
 Association of Deployment Exposures With Multisymptom Illness

	<i>Preliminary Analyses* (no controls for other exposures)</i>		<i>Adjusted Analyses* (controlling for effects of other exposures)</i>		
	GWV populations in which association was assessed <sup>a</sup>	GWV populations in which association was sign. <sup>b</sup>	GWV populations in which association was assessed <sup>a</sup>	GWV populations in which association was sign. <sup>b</sup>	Dose-response effect identified?
Pyridostigmine bromide	11	10	7	7	Yes
Pesticides	11	11	7	6	Yes
Psychological stressors	15	14	8	1	
Chemical weapons	18	15	7	4	
Oil well fires	10	9	5	3	Yes
Number of vaccines	2	2	1	1	Yes
Anthrax vaccine	5	5	2	1	
Tent heater exhaust	5	4	2	1	
Sand/particulates	3	3	3	1	
Depleted uranium	6	4	2	0	
Solvents	4	4	1	0	
Fuel exposures	5	4	2	0	
CARC paint	4	3	1	0	

Abbreviations: GWV = Gulf War veterans, sign. = statistically significant, HPA = hypothalamic-pituitary-adrenal axis, CARC = chemical agent resistant coating