SAIIE: A HEALTH INDEX FOR PEOPLE RE-EXPOSED TO WATER-DAMAGED BUILDINGS

IAQA

10/14/07

Sequential Activation of Innate Immune Elements SAIIE

Ritchie C. Shoemaker MD
Margaret S. Maizel
Center for Research of Biotoxin Associated Illnesses
Pocomoke, Maryland

The Problems

- Use of spore counts alone to clear a building for re-occupancy doesn't make sense
- Symptoms, analyzed individually, won't be a reliable marker, especially in litigation
- Since safety for humans underlies testing for microbes and inflammagens in a water-damaged building, do we look at the microbes or do we look at the people?

Repeating IAQ history

- Indoor versus outdoor spores, total
- Indoor versus outdoor spores, by species
- Threshold levels of given kinds of spores
- Ahh, counting fungal particles is far more important than counting fungal spores
- ERMI is so much better!
- What about bacteria, mycobacteria, actinomycetes, VOCs and beta glucans?

How can you measure quality?

- Absence of bad things
- Threshold of bad implies a dose-response
- Dose-response isn't linear in inflammatory illnesses! And don't forget genetics!
- Presence of good things doesn't prevent
- How do you account for differential susceptibility based on genetics, previous exposures, illness, inflammatory status?

What is today's accepted approach?

- Maybe a few spore counts
- Maybe a symptom list
- Maybe a moisture reading
- Maybe a CO_2 reading
- Only a few use human health as a marker
- Welcome to the world of SAIIE!

Symptoms alone are subjective

- Check lists are full of bias, even if recorded by a trained third party
- An individual symptom means little without differential diagnosis
- Upgrade: Cluster analysis looks at grouping of symptoms; gives statistical certainty
- Mega-upgrade: Logistic regression provides much greater statistical certainty

Lab tests would help

- Before and after measures using a prospective design provides causation, *if:*
  - Patient is treated such that his lab results equal those of large numbers controls before re-exposure
  - No additional re-exposure activity elsewhere
  - No other change in health status
  - Patient becomes his own control

What labs are abnormal in WDB illness?

- If allergy is the problem, total IgE is a good start, but IgE doesn't rise with exposure
- Removal from exposure will reduce symptoms but not IgE
- Allergic responses are antibody mediated and as such are part of *acquired immune* response
- *Innate immune* responses have little to do with allergic responses: biotoxins affect innate immunity and not acquired immunity

Illness from WDB Is Not Allergy

Mean IgE, by illness, all patients

		Cases N=	IgE
Controls	No illness	305	38
Mold cases	Confirmed case	672	43
Asthma cases	Inhaled steroids + 1 other med, > 6 months/year	45	973
Nasal allergy	Nasal steroid + 1 other med, > 6 months/year	40	407

WHY NOT LOOK AT HEALTH EFFECTS AFTER RE-OCCUPANCY?

- This idea should be a no-brainer
- We know the genetic basis of susceptibility
- We can study human illness using prospective exposure protocols *if* adequate control of other, ongoing illness is present
- We can use prospective exposure trials only if we can first treat the illness!
- Treating docs have evidence-based data

PARAMETERS	ADULT	
	ILL	WELL
MSH, mean	15.3	23.2
MMP9, mean	506	225
VEGF % < 31	38	0
VEGF % >200	15	0
ADH/osmolality dysfunction	65%	14%
ACTH/cortisol dysfunction	44%	6%
MARCoNS +	80%	3%

PARAMETERS	ADULT	
	ILL	WELL
C3a	> 1100	285
C4a	7287	631
IL-10	10.2	0.5
IL-1B	5.9	0.8
Interferon alpha	398	14
Erythropoietin % < 7.3	25	3
Erythropoietin % > 27.7	9	3

New players in clinical evaluation

- Vasoactive intestinal polypeptide (VIP)
- Suprachiasmatic hypothalamic nuclear agonist
- Inputs from olfactory bulb and retina
- Regulates cytokines peripherally, pulmonary artery pressure responses to exercise
- Stimulates a rise in intracellular cAMP
- Low in >85% of CBAI
- Low in all MCS patients seen to date (N>500)

Von Willebrand's and VIII

- Acquired abnormalities in vWF invariably seen in those with mucus membrane bleeds
- Epistaxis and hemoptysis
- Factor VIII is acute phase reactant
- vWF and ristocetin associated factor fall after day 2-3
- Hemorrhage can be profuse-Rx with DDAVP

TGF-beta

- Part of hypoxia response
- Associated with abnormal collagen cross-linking and wingspan > height
- Major player in abnormalities in T-cell regulation
- Rx with low dose erythropoietin
- Assay not available commercially yet

IL-1ra

- Interleukin-1-receptor antagonist
- Compensatory rise after activation of IL-1
- IL-1beta often found to be elevated, rising in 12 hours, but measuring blood levels alone will ignore autocrine and paracrine activity
- IL-1ra correlates highly with clinical illness and may surpass MMP9 as best indicator of cytokine activity in WDB patients

What labs aren't abnormal?

- CBC, metabolic profile, ESR, CRP, TSH
- ANA, immunoglobulins (includes IgE)
- Lipid profiles, antibody profiles
- All complement except for anaphylatoxins
- All genetic testing except for HLA DR by PCR
- LH, FSH, SHBG, estradiol, estrone, prolactin

What studies show that labs change hyperacutely?

- ASM Biodefense cohort 2006
- NTT cohort 2006
- ISTM cohort 2006

- SAIIE 2007!

C4a-1

- Anaphylatoxin, released when C4 is activated
- Dr. Giclas says, “C4 is an element of complement that acts like it has a big sign on it: Activate me.”
- Stimulates smooth muscle contraction, increases vascular permeability
- Recruits chemokines, degranulates mast cells, basophils
- Normal is < 2830 ng/ml

C4a-2

- Release activated by cell wall components of essentially every pathogenic fungus (Kozel)
- Rises in 4 hours after exposure to WDB
- Rises in 12 hours after a tick bite in those with Lyme disease
- Elevated in CFS, fibromyalgia, dinoflagellate and cyanobacteria illnesses
- Rarely elevated in non-innate immune illnesses

C4a-3

- Highly correlated with symptoms of executive cognitive problems (CDC-IACFS, ASTMH)
- Highly correlated with elevated lactate and low ratio of glutamate to glutamine in frontal lobes and hippocampi
- Capillary hypoperfusion is the currency of C4a
- Correction of C4a corrects cognitive problems, total symptoms and CNS metabolites

Leptin-1

- Adipocytokine
- Agonist of POMC pathway in VLN hypothalamus; long-isoform receptor is primitive gp-130 cytokine receptor
- Inflammatory in its own right
- Disproportionate increase in leptin resistance will make patients fat
- If leptin resistant, forget weight loss by standard means!

Leptin-2

- Rise in leptin seen after 48 hours
- Tie unexplained weight gain to time of onset of MSH deficiency in WDB illness
- Lowered by pioglitazone and high dose omega three fatty acids; PPAR-gamma control
- Must avoid insulin rise to lower leptin pharmacologically; use no-amylose diet
- Dimorphic; levels in women higher than men

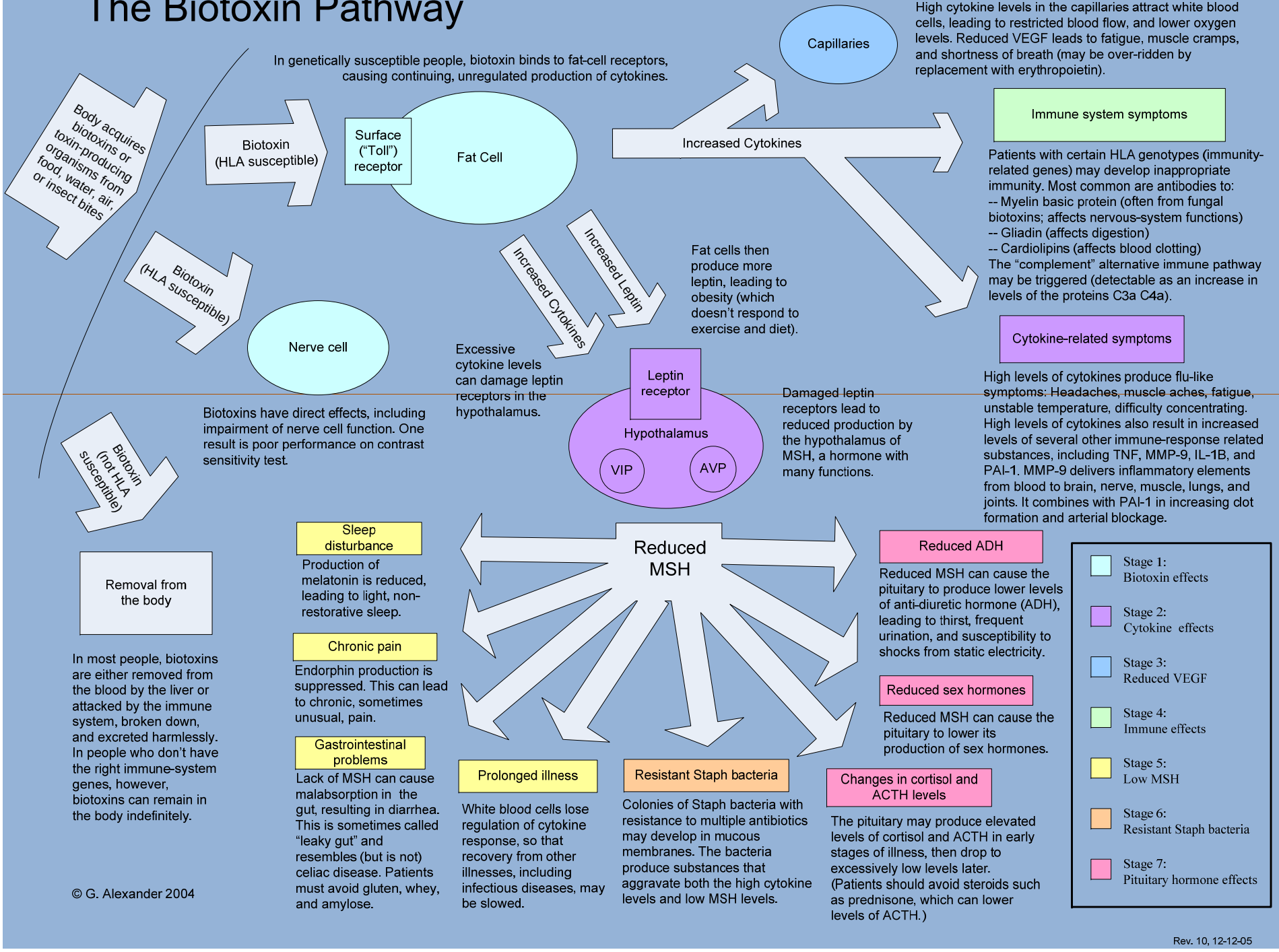
Matrix metalloproteinase-9

- MMP9 is the “delivery van” takes inflammatory elements out of blood and puts them into brain, joint, lung, nerve and muscle
- Normal range is 0-322; LabCorp range is wrong, still uses Esoterix range that combined cases and controls
- Freeze serum quickly after draw; don’t let tube clot!!
- Best measure of cytokine effect on endothelial cells and macrophages

Vascular endothelial growth factor

- Often low (< 31 ng/ml) in biotoxin illnesses
- Fumagillin was first antiangiogenic substance
- Clinical importance of high VEGF in cancer and atherosclerosis
- Response to hypoxia is initial transcription; expect levels to rise hyperacutely
- Second regulatory wave causes a fall of VEGF shortly thereafter

The Biotoxin Pathway



In genetically susceptible people, biotoxin binds to fat-cell receptors, causing continuing, unregulated production of cytokines.

High cytokine levels in the capillaries attract white blood cells, leading to restricted blood flow, and lower oxygen levels. Reduced VEGF leads to fatigue, muscle cramps, and shortness of breath (may be over-ridden by replacement with erythropoietin).

Patients with certain HLA genotypes (immunity-related genes) may develop inappropriate immunity. Most common are antibodies to:
-- Myelin basic protein (often from fungal biotoxins; affects nervous-system functions)
-- Gliadin (affects digestion)
-- Cardiolipins (affects blood clotting)
The "complement" alternative immune pathway may be triggered (detectable as an increase in levels of the proteins C3a C4a).

Fat cells then produce more leptin, leading to obesity (which doesn't respond to exercise and diet).

Excessive cytokine levels can damage leptin receptors in the hypothalamus.

Damaged leptin receptors lead to reduced production by the hypothalamus of MSH, a hormone with many functions.

High levels of cytokines produce flu-like symptoms: Headaches, muscle aches, fatigue, unstable temperature, difficulty concentrating. High levels of cytokines also result in increased levels of several other immune-response related substances, including TNF, MMP-9, IL-1B, and PAI-1. MMP-9 delivers inflammatory elements from blood to brain, nerve, muscle, lungs, and joints. It combines with PAI-1 in increasing clot formation and arterial blockage.

Biotoxins have direct effects, including impairment of nerve cell function. One result is poor performance on contrast sensitivity test.

In most people, biotoxins are either removed from the blood by the liver or attacked by the immune system, broken down, and excreted harmlessly. In people who don't have the right immune-system genes, however, biotoxins can remain in the body indefinitely.

Sleep disturbance
Production of melatonin is reduced, leading to light, non-restorative sleep.

Chronic pain
Endorphin production is suppressed. This can lead to chronic, sometimes unusual, pain.

Gastrointestinal problems
Lack of MSH can cause malabsorption in the gut, resulting in diarrhea. This is sometimes called "leaky gut" and resembles (but is not) celiac disease. Patients must avoid gluten, whey, and amylose.

Prolonged illness
White blood cells lose regulation of cytokine response, so that recovery from other illnesses, including infectious diseases, may be slowed.

Resistant Staph bacteria
Colonies of Staph bacteria with resistance to multiple antibiotics may develop in mucous membranes. The bacteria produce substances that aggravate both the high cytokine levels and low MSH levels.

Changes in cortisol and ACTH levels
The pituitary may produce elevated levels of cortisol and ACTH in early stages of illness, then drop to excessively low levels later. (Patients should avoid steroids such as prednisone, which can lower levels of ACTH.)

Reduced ADH
Reduced MSH can cause the pituitary to produce lower levels of anti-diuretic hormone (ADH), leading to thirst, frequent urination, and susceptibility to shocks from static electricity.

Reduced sex hormones
Reduced MSH can cause the pituitary to lower its production of sex hormones.

Rev. 10, 12-12-05

Treatment of Biotoxin Illnesses

- Follow the innate immune responses
- Remove from exposure
- Cholestyramine protocol first
- Eradicate biofilm formers next
- Correct cytokines with Actos
- Correct hormones: ADH, androgens
- Correct VEGF/epo
- Correct autoantibodies
- Correct C3a, C4a
- Correct acquired pulmonary hypertension
- Correct CNS lactate; glutamate/glutamine

Now you are ready for SAIIE

- Collate symptoms and labs using 7 steps
- Baseline; After Rx (AC-1); Away from building off all meds three days (HOC); into WDB off mall meds three days (BOC-1, BOC-2, BOC-3); Re-Rx (AC-2)
- Used in hundreds of patients safely
- Patient provides informed consent
- Physician can forbid if C4a > 20,000

Time course of Biotoxin Pathway

- Initial detection: cytokines and C3a, C4a
- Binding to Toll, c-linked lectin and dectin receptors activates gene transcription, IL-1b
- Cytokines bind to long isoform of leptin receptor, compensatory rise in leptin at day 2
- Cytokines turn on second wave of gene transcription, MMP9 rises day 2-3
- VEGF rises on Day 1 and crashes on day 3

Case definition-1

FIRST TIER modeled on CDC *Pfiesteria* Case definition from 1998

- Potential for exposure
- Multisystem, multisymptom illness
- Absence of confounders
  - Differential diagnosis key feature here

Case definition-2

SECOND TIER

- Simply stated: what did cases have that controls didn't
- Genetic susceptibility; HLA DR by PCR
- Hypothalamic impairment; low MSH
- Neurotoxic illness; VCS deficit
- Cytokine activation; MMP9 elevation
- Pituitary and peripheral endocrine response dysregulation
  - ACTH/cortisol
  - ADH/osmolality

Case definition set the bar at 100%

- Over 4400 WDB patients
- Over 600 controls
- Given that nothing in biology is ever 100%
  - All cases met criteria
  - No controls met criteria
- Potential for incorrect classification
- Logistic regression shows that number is infinitesimally small

Does the sequence of innate immune events confirm the illness?

- Study design: identify cases and controls
- Treat cases to equal controls at baseline
- Controls don’t have AC-1, HOC
- Record BOC 1, 2, 3 as % of baseline
- Subtract control % from case % BOC 1, 2, 3
- Establish illness effect by time of exposure
- Rate changes as % of 100, assign number 1-5

Using the SAIIE-1

- Prospective trial of 50 buildings
- Add indices from each category: symptoms, leptin, VEGF, MMP9 and C4a
- Cases average SAIIE was 17.1
- Controls 6.1
- Enables additional comparison of human health to environmental sampling
- Any ERMI > 2 was associated with acquisition of illness and SAIIE >13

Using the SAIIE-2

- If any C4a was > 20,000, however, illness recrudesced and SAIIE > 13 if ERMI was as low as NEGATIVE 1!
- No correlation of “safe” spore counts with any human health parameter (ie in the buildings that were cleared by spore counts)
- Not all patients have the identical pattern of response
- 50 buildings and 50 patients aren’t enough

Using the SAIIE-3

- Case in litigation
- Spore counts say the building is just peachy
- 3 volunteers, each met case definition and successfully treated
- Symptoms peak at day 3
- C4a peaks at Day 1, does not fall
- Leptin, VEGF and MMP9 show peaks on time
- Average SAIIE = 19.2

The future-1

- Add vWF and Factor VIII-difficult to get outlying labs to do specimen preparation right
- Genomics using PAX tubes
  - mMRA tells us which genes are activated and when
  - Other biotoxins (ciguatera, cholera) show same pattern
  - Assess gene activation as function of duration
- Use of genomics to assess illness at baseline

The future-2

- Link of SAIIE to ERMI will require much more data
- Early results are eerily similar, however
- If C4a > 20K means no exposure is safe, where do these patients live?
- How can the elevated IL-4, Il-8, IL-10 immuno-paralytic engine be stopped?

For more information

- www.chronicneurotoxins.com
- www.biotoxin.info
- www.moldwarriors.com
- Mold Warriors 2005, 2007
- Desperation Medicine 2001, 2006
- Lose the Weight You Hate 2002, 2005
- Surviving Mold Spring, 2008