

How Stress Kills



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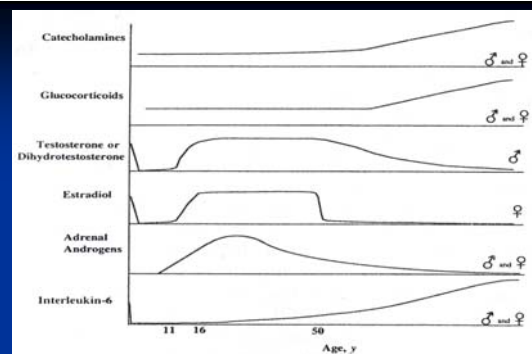


Infection and Trauma Trigger the Immune System's Inflammatory Response

- Inflammatory mechanisms are critical to resolving infections and repairing tissue damage
- Proinflammatory cytokines attract immune cells to sites of infection or injury, and activate the cells to respond to the insult

Proinflammatory cytokines

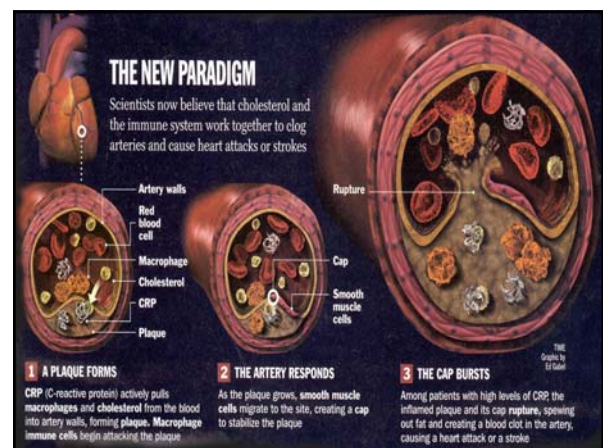
- Tumor necrosis factor- α (TNF- α)
- Interleukin 1 (IL-1)
- Interleukin 6 (IL-6)
- IL-6 directly promotes production of C-reactive protein (CRP), an important risk factor for cardiovascular disease



Changes in circulating hormones and IL-6 with aging
Papanicolaou et al. *Annals of Internal Medicine* 1998

AGE-ASSOCIATED DISEASES LINKED TO PROINFLAMMATORY CYTOKINES

- Cardiovascular disease
- Osteoporosis
- Arthritis
- Type 2 diabetes
- Certain cancers (including multiple myeloma, non-Hodgkin's lymphoma, and chronic lymphocytic leukemia)
- Periodontal disease
- Frailty and functional decline



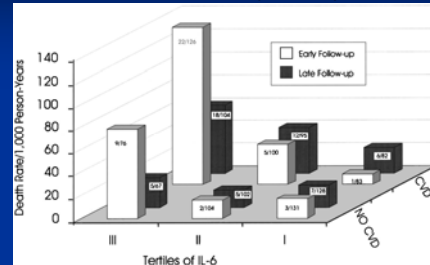
Higher plasma concentrations of IL-6 increase the risk of future myocardial infarction among apparently healthy men



- Prospective study involving 14,916 apparently healthy men with a 6-year follow-up
- Baseline IL-6 in 202 participants who subsequently had an MI and 202 matched for age and smoking status who did not report vascular disease during follow-up
- Men in the highest quartile at entry had a relative risk 2.3 times higher than those in the lowest quartile (95% CI 1.3 to 4.3, $P=0.005$); for each quartile increase in IL-6, there was a 38% increase in risk ($P=0.001$). The relationship persisted after controlling for CRP.

Ridker et al. *Circulation* 2000

IL-6 is prospectively associated with both all-cause and cardiovascular disease mortality over 3 years



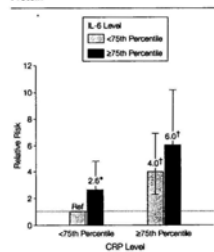
Age-adjusted all-cause mortality rates/1000 person-years by IL-6 levels, history of CVD, and follow-up period. Figures on bars are numbers of deaths and subjects at risk.

Volpato et al., 2001. *Circulation*

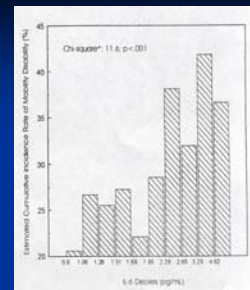
- Elevated levels of CRP and IL-6 predicted the development of type 2 diabetes after adjustments for BMI, family history of diabetes, smoking, exercise, alcohol, and hormone replacement therapy.

■ Pradhan et al. *JAMA* 2001

Figure 1. Relative Risk of Type 2 Diabetes Mellitus in Women According to Baseline Levels of Interleukin 6 and C-Reactive Protein



IL-6 indicates interleukin 6; CRP, C-reactive protein. Groups were formed based on the 75th percentile cut-point value of each biomarker (>0.61 mg/dL for CRP and >2.09 pg/mL for IL-6) using the distributions among control subjects in the study population. Risk estimates were matched on age and fasting status. Error bars indicate 95% confidence intervals. Asterisk indicates $P=0.002$ and dagger, $P<0.001$ for comparisons with the reference group (Ref).

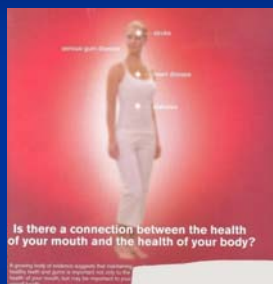


Incidence rates of mobility-disability over the 4-year follow-up according to deciles of IL-6 serum level.

Ferrucci et al; *J Amer Geriatrics Soc* 1999

Chronic or Recurring Infections Can Provoke Pathological Changes

- Low levels of persistent inflammation may be provoked by chronic infectious processes including
 - ⚡ periodontal disease
 - ⚡ urinary tract infections
 - ⚡ chronic pulmonary disease
 - ⚡ chronic renal disease



Inflammation Impacts Cancer: Pathways

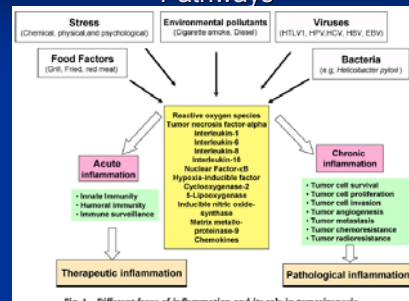


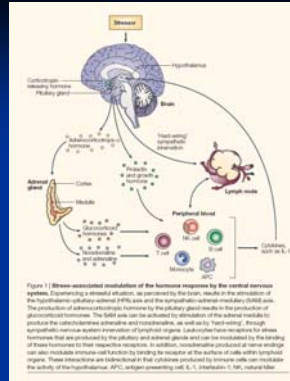
Fig. 1 - Different faces of inflammation and its role in tumorigenesis.

Aggarwal et al. Inflammation and cancer: How not is the link? *Biochem Pharmacol*. 2006



"Oh, that's Edward and his fight or flight mechanism."

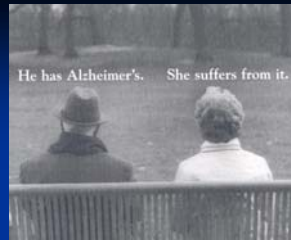
Glaser & Kiecolt-Glaser: *Nature Reviews Immunology*, 2005



Hormone	Expression of receptors by immune cells	Examples of effects on cell function	References
Glucocorticoids	T and B cells, neutrophils, monocytes and macrophages	Inhibit inflammation; inhibit the production of IL-12 by antigen-presenting cells; induces a shift from production of T_H1 to T_H2 cytokines	87,88
Substance P	T and B cells, eosinophils, mast cells, monocytes and macrophages	Stimulates mitogen-induced blastogenesis; increases trafficking of cells from lymph nodes to peripheral blood; stimulates monocytes to produce several cytokines, such as IL-1, IL-6 and TNF	89
Neuropeptide Y	T and B cells, dendritic cells, monocytes and macrophages	Can downregulate antibody production to T-cell-dependent antigens by its effect on dendritic cells, and T and B cells	90
Corticotropin-releasing hormone	T cells, monocytes and macrophages	Increases production of IL-1 by monocytes; evidence for autocrine and/or paracrine modulation of inflammation	91
Prolactin	T and B cells, granulocytes, monocytes and macrophages	Can stimulate lymphoid cell clonal expansion; might function as an auto-co-mitogen for NK cells and macrophages	92,93
Growth hormone	T and B cells, NK cells, monocytes and macrophages	Helps to maintain competence of T and B cells, and macrophages; stimulates antibody production and NK-cell activity	94
Catecholamines, adrenaline and noradrenaline	T and B cells, NK cells, monocytes and macrophages	Induce a shift to a T_H2 response, including antigen-presenting cells and T_H1 cells	95
Sex steroids	T and B cells, NK cells, monocytes and macrophages	Modulates the synthesis of IFN- γ by NK cells; stimulates the production of IL-16 (a chemotactic factor) by T cells	96

IFN- γ , interferon- γ ; IL-1, interleukin-1; NK, natural killer; T_H2 , T-helper 2; TNF, tumor necrosis factor

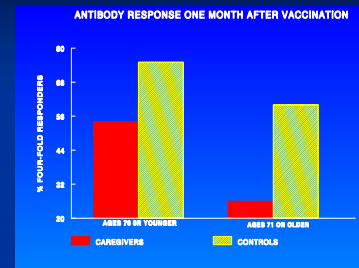
Dementia family caregiving as a chronic stressor



"The 88-year old former airline pilot had been ill for five years, during which time his cognitive abilities had steadily deteriorated. At times he did not recognize his wife and accused her of being a stranger. He could only follow the most simple of commands. His moods were extremely labile, with frequent crying spells and occasional outbursts of anger. He often followed his wife around the house, interrupting her activity with questions and demands. Urinary incontinence and night time agitation were bothersome problems. He often paced at night, occasionally leaving the house and becoming lost... His wife of forty years now devoted her time to looking for her husband. She rarely left home except to do necessary shopping or bill paying, and rarely invited neighbors or friends to her home because of embarrassment about her husband's behavior."

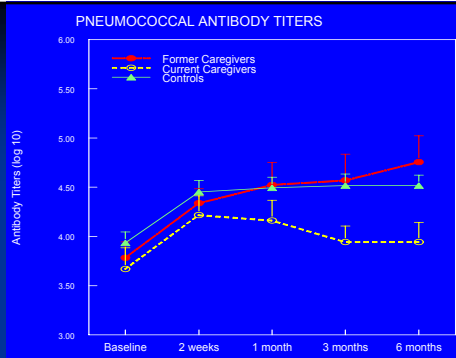
Barnes et al., *Journal of the American Geriatrics Society*, 1981

CAREGIVERS' ANTIBODY RESPONSES TO INFLUENZA VACCINE WERE SUBSTANTIALLY POORER THAN WELL-MATCHED NON-CAREGIVERS



Kiecolt-Glaser, Glaser, Gravenstein, Malarkey, Sheridan: *Proceedings of the National Academy of Sciences, USA*, 1996

Replication with caregivers published by Vedhara et al. in *Lancet*, 1999



Glaser, Sheridan, Malarkey, MacCallum, & Kiecolt-Glaser: Chronic stress modulates the immune response to a pneumococcal pneumonia vaccine. *Psychosomatic Medicine*, 2000

Recent study with younger parental caregivers: Gallagher, Phillips, Drayson, Carroll

Vaccine responses provide a proxy for responses to infectious disease

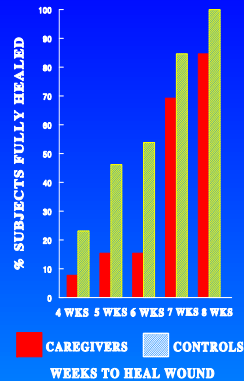
- Multiple labs have shown stress-related alterations in both antibody and T-cell responses to viral and bacterial vaccines including
 - Hepatitis B
 - Influenza virus
 - Pneumococcal pneumonia
 - Rubella
 - Meningitis C conjugate
 - Tetanus

STRESS SLOWS WOUND HEALING

Caregivers took an average of 24% longer than well-matched controls to heal the same small, standardized wound.



Kiecolt-Glaser, Marucha, Malarkey, Mercado, & Glaser: *Lancet*, 1995



ORAL WOUND HEALING AND ACADEMIC STRESS

SUBJECTS: 11 dental students (9 men, 2 women, average age=24.36)

DESIGN: Two punch biopsy wounds were placed on the hard palate

- at the end of summer vacation
- three days before the first major exam of the term (contralateral side)

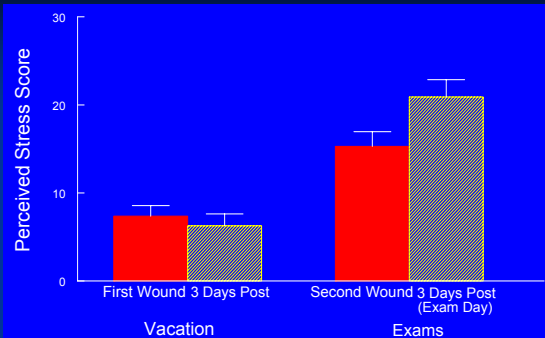
Psychological and health behavior data were collected at each time point

Each student served as her or his own control

Pain/discomfort ratings were low, with 95% reporting mild or no discomfort

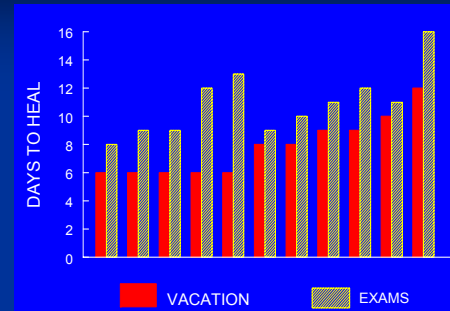
Marucha, Kiecolt-Glaser, & Favagehi: *Psychosomatic Medicine*, 1998

Differences in stress between the two wounds



Marucha, Kiecolt-Glaser, & Favagehi: *Psychosomatic Medicine*, 1998

Days to heal for each of the 11 students; no student healed as rapidly during exams, with the average student taking 40% (3 days) longer

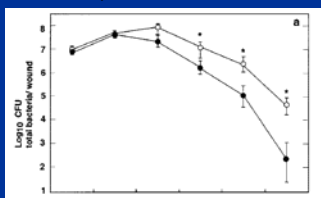


Marucha, Kiecolt-Glaser, & Favagehi: *Psychosomatic Medicine*, 1998

Stress increases susceptibility to wound infections:

Compared to control mice, restraint stress delayed wound healing by 30% and caused a 2-5 log increase in opportunistic bacteria (e.g., *Staphylococcus aureus*)

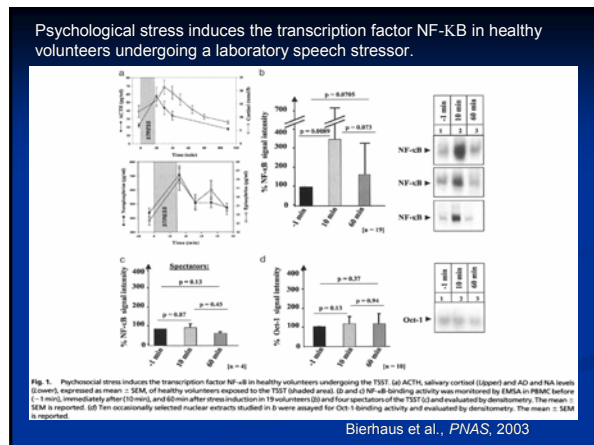
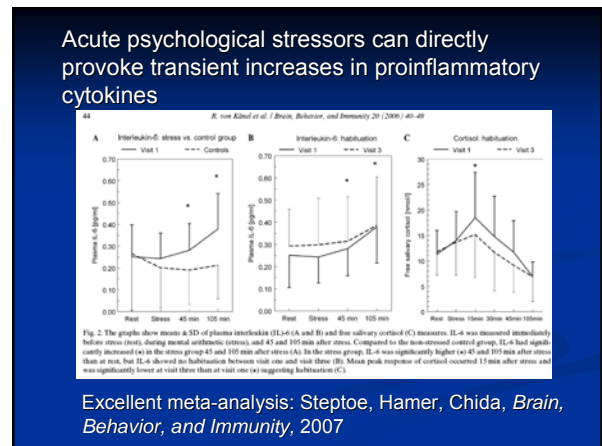
By day 7, 85.4% of restraint-stress mice had bacterial counts predictive of infection, vs. 27.4 of controls



Rojas, Padgett, Sheridan, Marucha: *Brain, Behavior, and Immunity* 2002

Infection and Trauma Trigger the Inflammatory Response

- Inflammatory mechanisms are critical to resolving infections and repairing tissue damage
- Proinflammatory cytokines attract immune cells to sites of infection or injury, and activate the cells to respond to the insult
- Chronic infectious processes or chronic wounds can provoke persistent inflammation (e.g., periodontal disease, urinary tract infections)

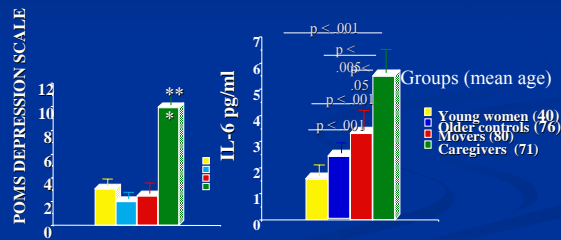


- Acute stress enhances systemic proinflammatory cytokine production while inhibiting local production at wound sites
- Couples were admitted twice to the GCRC (hospital research unit), 24 hours each admission
 - First admission: Structured social support interaction
 - Second admission: Structured conflict (marital disagreement)
 - Both admissions: blister wounds before the interaction, wore wound chambers 22 hours, assessed proinflammatory cytokine secretion at wound sites and plasma
 - Couples' blister wounds healed more slowly, and local cytokine production (IL-6, TNF- α , IL-1 β) was also lower at wound sites following marital conflicts than after social support interactions.
 - Hostile couples showed larger increases in plasma IL-6 and TNF- α the morning after a conflict than after a social support interaction compared to less hostile couples, and hostile couples took an average of two days longer to heal wounds.
- Kiecolt-Glaser, Loving, Stowell, Malarkey, Lemeshow, Dickinson & Glaser (2005). *Archives of General Psychiatry*.



- STRESS/DEPRESSION ENHANCE PROINFLAMMATORY CYTOKINE PRODUCTION
- Major depression \rightarrow increased secretion of proinflammatory cytokines; treatment with antidepressants decreases secretion (Maes; Irwin; Miller)
 - Depressive symptoms were linked to increased IL-6 in community samples of older adults (Dentino et al., 1999)
 - Depressed mood was associated with higher levels of serum IL-6, TNF- α , and CRP among older adults ages 70-79 (Penninx et al., 2003)
 - Chronic stressors like caregiving have been associated with heightened IL-6 compared to noncaregiving controls (Lutgendorf et al.; Kiecolt-Glaser et al.; Glaser et al.)
 - Acute stressors enhance production of proinflammatory cytokines (Goebel et al., 2000; Steptoe et al., 2001)

Interleukin-6 in 4 Groups of Community Women



Lutgendorf et al., Journals of Gerontology, 1999

Longitudinal community study spanning 6 years:

119 caregivers
106 noncaregivers
(mean age at study entry, 71)

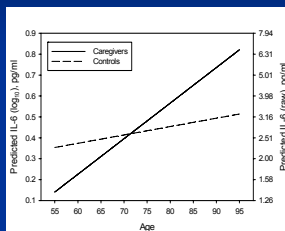
Key Measures:

IL-6 in frozen plasma samples, 2x/year
Health behaviors associated with IL-6
Depressive symptoms

Kiecolt-Glaser, Preacher, MacCallum, Atkinson, Malarkey, & Glaser (2003). *Proceedings of the National Academy of Sciences, USA*

MODELED CHANGE IN IL-6 IN CAREGIVERS VS. NONCAREGIVERS

IL-6 is represented as a linear function of age; each individual's pattern of change is represented by a straight line defined by an intercept (predicted level of IL-6 at age 55) and slope (predicted change in IL-6 per year).



>3.19 = upper quartile, epidemiologic studies

Caregivers' average rate of increase in IL-6 was about *four times as large* as that of noncaregivers, and the two slopes were significantly different from one another, $p = .01$.

WHAT HAPPENS WHEN CAREGIVING ENDS?

Normal bereavement: higher incidence of depression and anxiety in widows and widowers within the first several months after the spouse's death compared to nonbereaved controls.

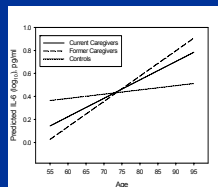
These group differences are typically not significant in follow-up data collected one to two years later (Harlow et al., 1991; Lund et al., 1989; Thompson et al., 1991).

Thus—caregivers should look no different than noncaregivers ~2 years after the spouse's death

On entry into this portion of the longitudinal study, 28 of the caregivers' spouses had already died; an additional 50 of the 119 spouses died during the 6 years of this study.

Selected for this analysis: continuous caregivers vs. those bereaved 3 or more years

Former caregivers' average rate of annual change in IL-6 did not differ from that of current caregivers, even several years after the death of the impaired spouse



Kiecolt-Glaser, Preacher, MacCallum, Atkinson, Malarkey, & Glaser (2003). *Proceedings of the National Academy of Sciences, USA*

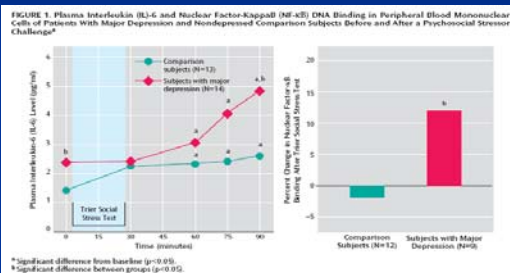
Stress and depression can prime the inflammatory response, promoting larger cytokine increases in response to subsequent stressors and/or minor infectious challenges

Prior stress produces exaggerated proinflammatory cytokine responses to infection. Compared to nonstressed controls, LPS exposure resulted in larger and more rapid proinflammatory cytokine induction in stressed rats. Johnson et al. *Brain, Behavior, and Immunity*, 2002

Among women who had just given birth, those with a prior history of major depression showed greater increases in serum IL-6 and the soluble IL-6 receptor after delivery than women without a similar depression history. Maes et al. *Journal of Affective Disorders*, 2001

Higher levels of depressive symptoms were associated with higher levels of IL-6, as well as an amplified and prolonged inflammatory response following influenza vaccination. Glaser et al. *Arch Gen Psychiatry* 2003

Patients with major depression vs. nondepressed controls:
Greater stress-induced IL-6 and NF- κ B activation
More depressive symptoms=greater change



Pace et al. *Am J Psychiatry* 2006

STRESS PROMOTES POOR HEALTH BEHAVIORS THAT ENHANCE PROINFLAMMATORY CYTOKINE PRODUCTION

- Diet high in saturated fat
- Less exercise
- Poorer sleep
- Smoking

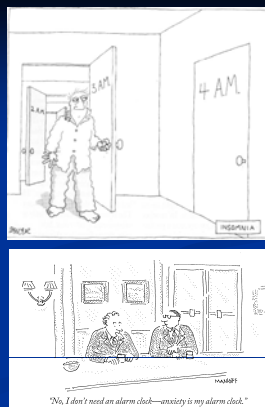


Sleep deprivation alters normal nocturnal increases in IL-6, contributing to immune system dysregulation.

Redwine et al.: *J Clin Endocrinol Metab*, 2000

“...a good night's sleep is associated with decreased daytime secretion of IL-6 and a good sense of well-being...”

Vgontzas et al.: *J Clin Endocrinol Metabol*, 1999



HEALTH-RELATED BEHAVIORS MAKE A DIFFERENCE:

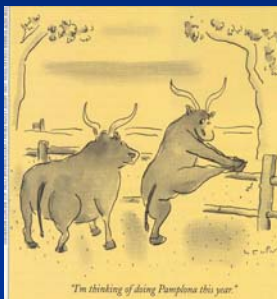
Individuals with a higher body mass index have higher levels of IL-6 and CRP; Papanicolaou, Wilder, Manolagas, Chrousos: *Ann Intern Med*, 1998



Abdominal adipose tissue may secrete up to three times as much IL-6 as other subcutaneous fat tissues; Browning: *Proc Nutri Soc* 2003

Central adiposity may be associated with larger stress-induced cytokine responses; Brydon, Wright, O'Donnell, Zachary, Wardle, Steptoe, *Int J Obes* 2007

Moderate Physical Activity May Help Attenuate Chronic Inflammation



- Lower basal IL-6 and CRP in well-trained swimmers than healthy controls (Espersen et al. *Scand J Med Sci Sports* 1996)
- High levels of recreational activity were associated with lower IL-6 and CRP in healthy older adults (Reuben et al. *J Am Geriatrics Soc* 2003)
- 35% reduction in CRP after 6 months of supervised moderate exercise in men and women at risk for future heart attack (Smith et al. *JAMA* 1999)

Acute stress enhances systemic proinflammatory cytokine production while inhibiting local production at wound sites


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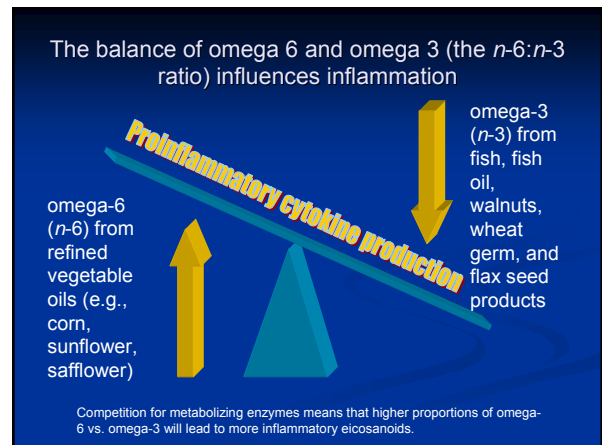
■ *Interventions that diminish stress or depression and/or inflammation may enhance health in part through their positive impact on immune and endocrine regulation*

Nutritional Neuroscience and Psychoneuroimmunology: Interdisciplinary Science at the Crossroads

- Omega-3 and omega-6: implications for depression, cardiovascular disease, and inflammation



"I ate all the wrong things today."



Historical Dietary Changes: Leaves to Seeds

- Omega-3 ($n-3$) polyunsaturated fatty acids (PUFAs): Fish, fish oil, walnuts, wheat germ, flax seed products
- Arachidonic acid (AA) derived (omega-6 or $n-6$) eicosanoids (primarily from refined vegetable oils such as corn, soy, sunflower, and safflower)
- Before early 1900s, dietary $n-6 : n-3$ ratios were ~ 2:1 or 3:1...then processed vegetable seed oils entered the diet
- Contemporary North American dietary ratios: 15-17:1; Europe, UK: 14:1

A NEW ERA FOR SCIENCE IN NUTRITION

Hunter-Gathering Agricultural Industrial


Calories from fat

Time (years)

Leaf A, Weber PC: Am J Clin Nutr 1987

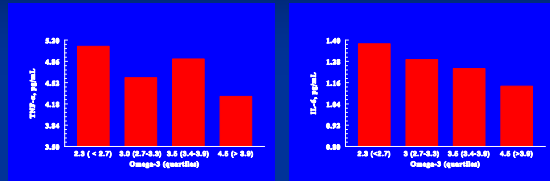
Leaves to Seeds

- In addition to the broad adoption of refined vegetable oils...
 - Industrial meat is raised on more seeds and fewer leaves than preindustrial meat, so it has more omega-6 and less omega-3
 - Official dietary advice since the 1970s has promoted polyunsaturated vegetable oils (particularly corn and soy), high in omega-6
 - When oils are hydrogenated to make them more stable, omega-3s are eliminated



"I ate all the wrong things today."

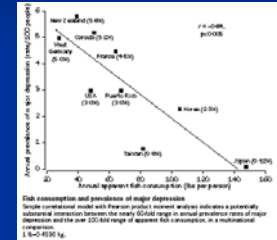
1123 people, ages 20-98, Tuscany, Italy



Ferrucci et al. Relationship of plasma polyunsaturated fatty acids to circulating inflammatory markers. *J Clinical Endocrinology and Metabolism*, 2006

Possible consequences of the historical dietary shift from leaves to seeds: increased depression and cardiovascular disease

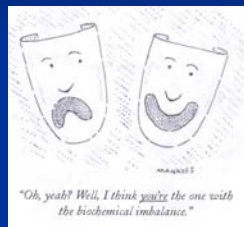
- Significant inverse relationships between annual fish consumption and prevalence of major depression
- Very similar pattern for cardiovascular disease



Hibbeln: Fish consumption and major depression. *Lancet* 1998

Depression and omega-3 intake

- Depressed patients have lower average plasma levels of omega-3 PUFAs than nondepressed controls
- Significant relationships between lower omega-3 plasma levels and greater negative mood have been documented in psychiatric and nonpsychiatric populations
- Clinical trials (mostly add-on designs) are encouraging, but not unanimously positive



Possible Biological Mechanisms: Impact of Omega-3 on Psychiatric Disorders

- Increased serotonergic neurotransmission
- Alterations in dopaminergic function
- Regulation of corticotropin-releasing factor
- Inhibition of protein kinase C
- Suppression of phosphatidylinositol-associated second messenger activity
- Modulation of heart rate variability via vagal mechanisms
- Increased dendritic arborization and synapse formation
- Prevention of neuronal apoptosis
- Improved cerebral blood flow
- Competition of EPA with AA for enzymatic activity and resultant reduction of the inflammatory response

From: Freeman et al. *J Clin Psychiatry*, 2006

Depression, cardiovascular disease, and omega-3

- Large epidemiological studies: inverse relationships between omega-3 levels and cardiovascular disease
- Major depression is associated with lower omega-3 fatty acid levels in patients with recent acute coronary syndromes

Frasure-Smith et al. *Biol Psychiatry*, 2004



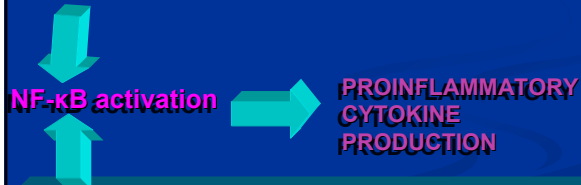
Possible Mechanisms

- Preventing arrhythmias
- Decreasing platelet aggregation
- Decreasing plasma triglycerides
- Moderately decreasing blood pressure
- Reducing atherosclerosis
- Small increase in HDL cholesterol
- Modulating endothelial function
- Decreasing proinflammatory eicosanoids

National Academy of Sciences, 2002

HOW DIET INTERACTS WITH STRESS AND DEPRESSION: NF- κ B ACTIVATION

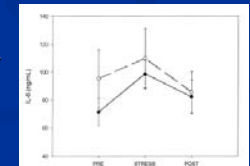
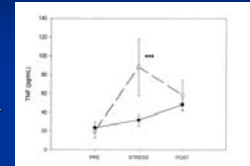
Key omega-3 PUFAs, EPA and DHA, *reduce* NF- κ B activation
Zhao 2004 *J Am Coll Nutr*



Depression and psychological stress *promote* NF- κ B activation
Bierhaus et al. 2003, *PNAS*; Pace et al. 2006, *Am J Psychiatry*

EVIDENCE: DIETARY INTAKE INTERACTS WITH EXAM STRESS

- Students who had higher $n-6:n-3$ ratios (above the mean) before exams demonstrated greater TNF- α production by LPS and mitogen-stimulated PBLs during exams than those with lower ratios
 - Maes et al., 2000, *Biol Psychiatry*
- The data suggest that $n-6:n-3$ ratios influence the proinflammatory response to stressors.
- Because TNF- α and IL-6 are produced by a variety of types of cells, serum cytokine levels may better reflect the overall inflammatory profile than stimulated PBLs.



IMPLICATIONS

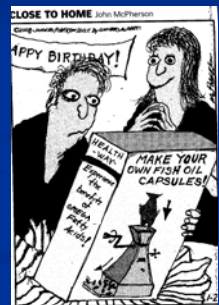
Diets with high omega-6:omega-3 ratios may enhance risk for both depression and inflammatory diseases, particularly when individuals already have elevated levels of depressive symptoms.

Randomized controlled trials are needed...!



NIH-funded randomized controlled clinical trials

- Medical students (NCCAM)
- Healthy community adults, ages 40-88 (NIA)



Interdisciplinary teams that assess psychological and biological outcomes are essential

Janice K. Kiecolt-Glaser, Ph.D.
Ronald Glaser, Ph.D.
William B. Malarkey, M.D.
Stanley Lemeshow, Ph.D.
Martha Belury, Ph.D.