The Hygiene Hypothesis

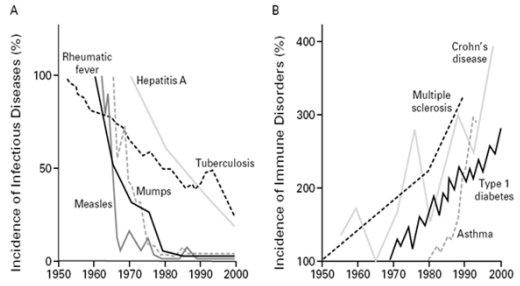
Too Clean or Not Too Clean?

Overview

- What is the Hygiene Hypothesis?
- History
- Immunological Mechanisms
- Case study
- Helminthic Therapy

The Hygiene Hypothesis

- There has been a steep increase in the diagnosis of autoimmune diseases and allergies since 1950
  - Cannot be explained by change in genetic background
- At the same time, cleanliness has increased, and incidence of parasites has decreased.



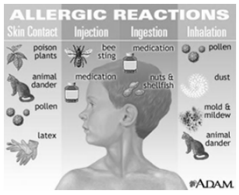
Bach J.F. 2002. New England Journal of Medicine. 347: 911-920

Hygiene Hypothesis definition

- lack of early childhood exposure to infectious agents
  - symbiotic microorganisms (e.g., gut flora or probiotics)
  - parasites
  - endotoxins
- increased susceptibility to allergic diseases by suppressing natural development of the immune system

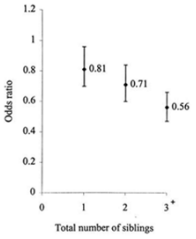
Allergies and Autoimmune diseases

- Allergies: inappropriate immune response of the body against normally harmless substances in the environment
- Autoimmune diseases: inappropriate immune response of the body against substances and tissues normally present in the body
  - Asthma
  - Celiac disease
  - Crohn's disease
  - Multiple sclerosis (MS)



History—The beginning

- Dr. David P. Strachan
  - Used epidemiological data to explain why there was an increase in the prevalence in asthma, hay fever and eczema
- At first it was met with skepticism
  - Prevailing immunological thinking was that infection triggered allergic response
- The prevalence of hay fever was found to be inversely related to the number of siblings of the study subject

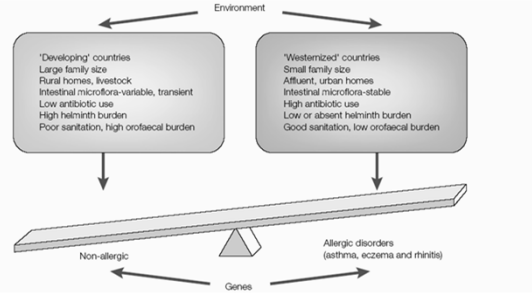


Strachan D.P. 1989. BMJ. 18:1259-1260.

History

"These observations…could be explained if allergic disease were prevented by infection in early childhood, transmitted by unhygienic contact with older siblings, or acquired prenatally…Over the past century declining family size, improved household amenities and higher standards of personal cleanliness have reduced opportunities for cross-infection in young families. This may have result in more widespread clinical expression of atopic disease." David Strachan, BMJ, 1989

Etiological Factors – Gene and Environment



Wills-Karp M, et al. Nature Reviews Immunology; 2001; 1: 69-75

Inverse correlation of type I diabetes and chronic infectious diseases

Zaccone et al. 2006. Parasite Immunology. 28: 515–523.

You are less likely to be allergic if you:

- Had older siblings, especially brothers
- Rarely washed your hands or face as a child
- Lived in a home with bacteria-laden dust
- Were brought up on a farm with animals
- Had a dog
- Had a childhood infection that was transmitted by fecal to oral contamination
- Grew up in Communist, rather than Western, Europe.

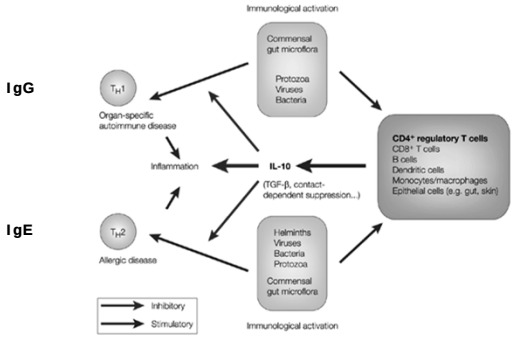
Rook and Brunet. 2002. Biologist (London) 2002:145–149

Mechanisms

1. Counter regulation hypothesis
2. Heavy infection/strong regulatory hypothesis
3. "Old friends" hypothesis

Counter regulation hypothesis

- T lymphocytes are white blood cells (WBC)
  - Play central role in cell mediated immunity
- T-helper (Th) cells assist WBC
  - Th1: promote IgG response. Against microbes
  - Th2: promote IgE response. Against helminthes. Over reactivity can lead to atopy.
- The two kinds of T-helper cells, must be in balance for proper immune system function
- A lack of infections during development leads to lower Th1 and higher Th2, causing more Th2 related reactions such as allergies.

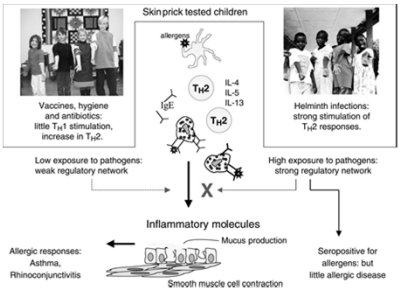


Wills-Karp et al. 2001. Nature Review Immunology. 1: 69–75.

Heavy infection/strong regulatory hypothesis

- There are divergent T_H2 responses
  - Developed countries: vaccines, hygiene, and antibiotics lead to low T_H1 and T_H2 responses.
    - Weak regulatory network
  - Developing countries: helminth and bacterial infections lead to strong T_H2 responses.
    - Strong regulatory network

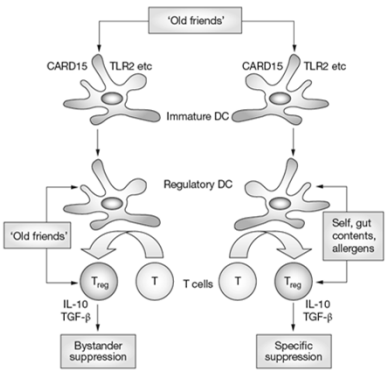
Heavy infection/strong regulatory hypothesis



Old friends hypothesis

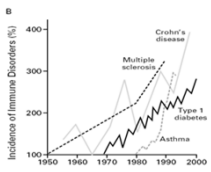
- T regulator cells can only become fully effective if they are stimulated by exposure to organisms with low levels of pathogenicity, and which have coexisted universally with human beings throughout our evolutionary history
  - Helminths
  - Peudocommensals
- These organisms might have been entrusted by evolution with the role of setting up the regulatory circuits of the immune system

Rook, G.A. et al. 2003. Curr. Opin. Allergy Clin. Immunol. 3: 337-42.



Example: Asthma

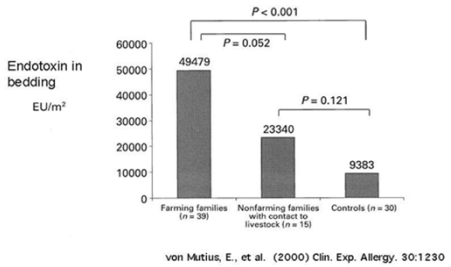
- Disease of chronic airway inflammation
- The most common chronic disorder of childhood
- Over 9 million children in the US have been diagnosed with asthma
- Rapid rise in asthma is greatest in developed countries and urban areas
- Case study



Endotoxins

- Lipopolysaccharides are in the outer cell membrane layer of all gram-negative bacteria
- Endotoxin levels vary widely but tend to be highest in environments where there are farm animals such as cows, horses, and pigs
- Can be inhaled or swallowed and acts as a potent immunostimulant
- Exposure early in life, during the development of the immune system, is most important in providing protection against the development of allergic disease

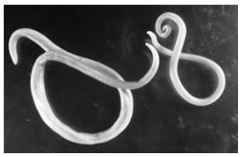
Braun-Fahrländer et al. 2002. New England Journal of Medicine 347:869-77.

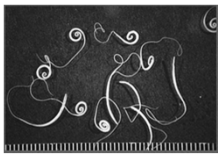


von Mutius, E., et al. (2000) Clin. Exp. Allergy. 30:1230

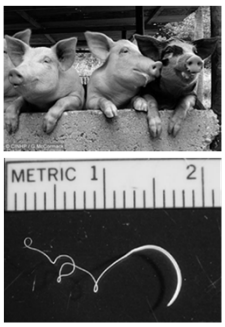
Helminthic Therapies

- Deliberately infect patients with the ova of parasitic worms
  - *Necator americanas*- hookworms
  - *Trichuris suis* - whipworms



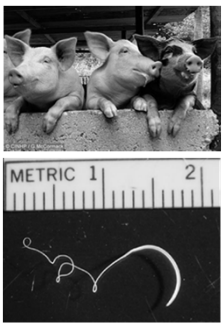


Crohn's disease



- **Several clinical trials using *Trichuris suis* have been conducted to treat human Crohn's disease or ulcerative colitis (autoimmune inflammation of the intestine)**
- ***T. suis* does not productively infect humans yet modulates the immune response**
- **Most studies show clinical improvement for a significant fraction of the patients**

Crohn's disease



- **One example : patients suffering from Crohn's received 2500 *T. suis* ova every 2 weeks for 12 weeks (or placebo)**
- **43.3% of the patients given *T. suis* improved compared with those given placebo (16.7%).**
- **The study also included a 12 week crossover limb where patients in one group were switched: 56.3% of the patients switched to *T. suis* improved compared with 13.3% of patients given placebo (13%).**

Elliot et al. 2007. International Journal for Parasitology 37: 457-464

Multiple Sclerosis (MS)

- A longitudinal study was conducted at an MS clinic, which compared MS outcomes between patients with parasites and those without
- Investigation suggests that parasite infections are associated with clinical, MRI and immunologic benefits
- Longer-term studies in the works

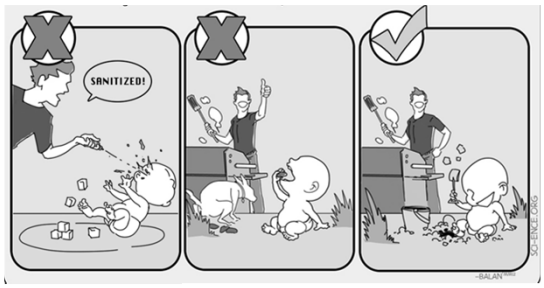
Correale and Farez. 2007. Annals of Neurology 61: 97-108

Conclusions

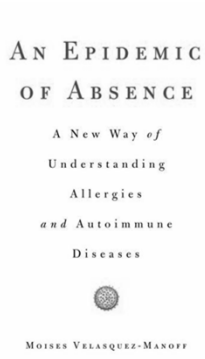
- We should use a little more common sense and let kids be kids

- Stop worrying about most germs
  - Antibacterial soaps
  - Antibacterial toys
- We may have developed a symbiotic relationship with a lot of the microorganisms we are trying to eradicate
- >70% of all bacteria are nonpathogenic

Microban®
antimicrobial product protection

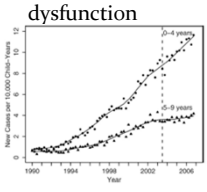


Want to learn more?



Autism

- Disorder of neural development
  - impaired social interaction and communication
  - restricted and repetitive behavior
- Afflict 1 in 88 in developed countries
- Recently, autism has been linked to immune dysfunction



Autism

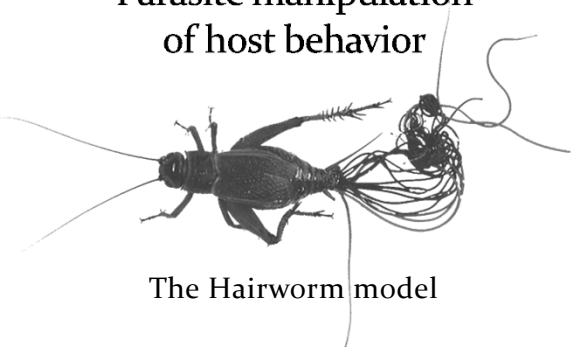
- In autistic individuals the immune system fails at regulation and inflammatory signals dominate
  - Chronic activation
  - Changes brain morphology
- Evidence from a Danish study indicates that the trigger is in the womb
  - Infection during pregnancy
  - Autoimmune disease of mom.

Autism

- The flu: 300% increased chances
- Bacterial infection: 40% increased chances
- Asthma or allergies: 50% increased chances
- Rheumatoid arthritis: 80% increased chances
- Celiac disease: 350% increased chances

- It appears that the mother's inflammatory response is leading to chronic inflammation in her child.

Parasite manipulation of host behavior



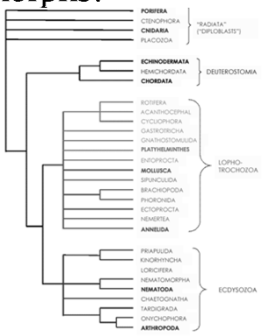
The Hairworm model

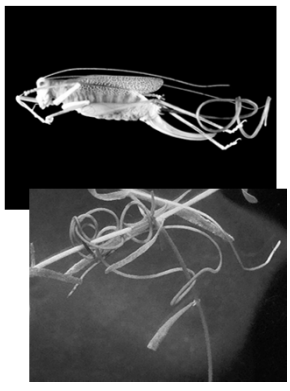
Outline

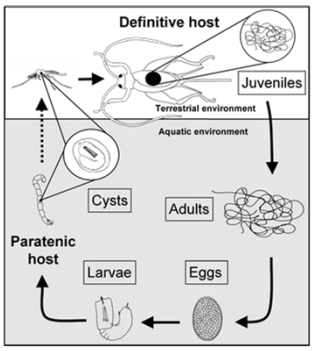
- Background
  - What are hairworms?
  - Life cycle
  - Why study these?
- What have we learned from hairworms?
  - Host behavior
  - Mechanisms of parasite manipulation
- Parasite manipulation in humans!?

What are nematomorphs?

- Phylum of insect and crustacean parasites
  - castrate hosts
- Sister phylum to Nematodes (roundworms)



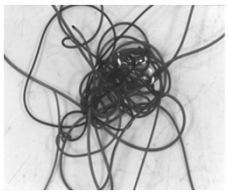




A worm by any other name…

- Horsehair worm
  - Leidy (1870) watched horsehairs placed into water for weeks, without "…having had the opportunity of seeing their vivification…"
- Hair snake
- Violin String worm (Germany)
- Gordian worms

What is a Gordian Knot?



Gordius, king of Phrygia

Why study hairworms?

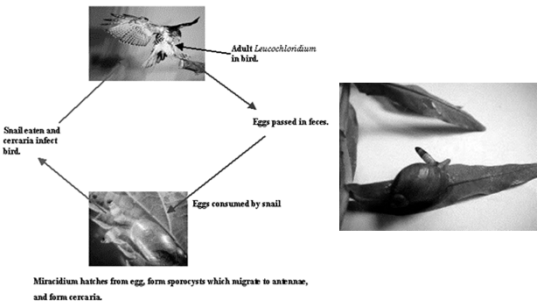
- Model organism
  - *Caenorhabditis elegans*
  - *Drosophila melanogaster*
- Biodiversity indicators
- A great model system to study effects of parasites in humans
  - Host manipulation.

I. Host behavior manipulation

- Some parasitized animals are known to change their behavior
- Behavior increases parasite transmission
- Why?
  - By-product of infection
  - Evolved trait
    - Benefit to parasite

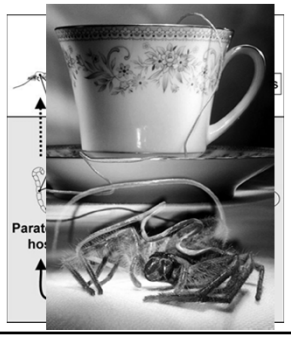
Behavior manipulation

Leucochloridium sp.



Behavior manipulation

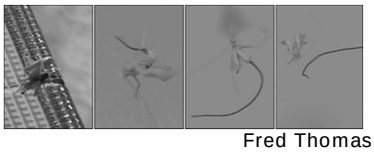
Hairworms



- Adults in crickets
- Crickets are terrestrial
- Worms need water
- How to get worms from cricket into water?
- Is there manipulation?

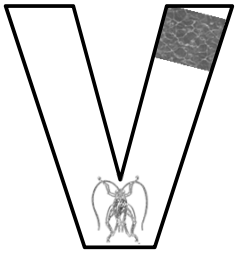
Observations

- Lots of worms are found in water; few on soil
- Lots of hosts seen jumping into water, most contained worms



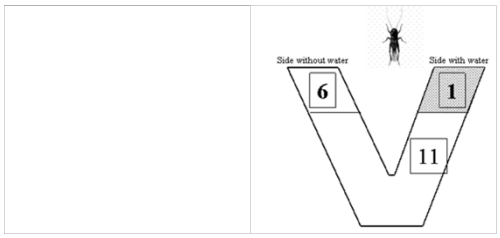
Fred Thomas

Manipulation experiment



- Two groups of crickets
  - Infected
  - Uninfected
- V- maze experiment
  - Water
  - No water
- Do infected (manipulated) hosts preferentially move to water?

Biron *et al*., 2006a



Manipulated Control

- Hosts are not manipulated towards water
- However, they do act 'crazy' when near water.

Biron *et al*., 2006a

Mechanism of manipulation

- What is proximate cause of host manipulation?
  - By-product of Pathology
  - Parasite mediated (evolved) manipulation
- If it is caused by parasite, we should see production of "manipulation factor(s)".

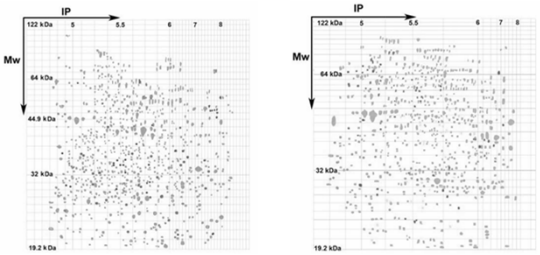
Proteomics

- Large-scale study of proteins, particularly their structures and functions
- Allows for the identification of proteins involved with phenotype

Proteomics

- Determine differential protein expression
  - 2-dimensional gel electrophoresis
- Identify proteins of interest
- 'Feed' into mass spectrometer
  - 'Measures' the aa
  - Reconstruct the peptide
- Compare to database and identify

Proteomics example



Cricket Worm

Biron *et al*., 2006b

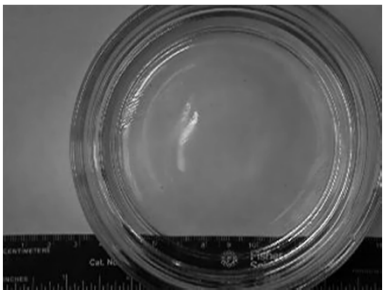
Results

- Manipulated crickets changed expression of genes involved with
  - Neurogenesis
  - Circadian rhythm
  - Neurotransmitter activities
- Manipulative parasite produces Wnt molecules
  - Act on development of cricket central nervous system

Biron *et al*., 2006b

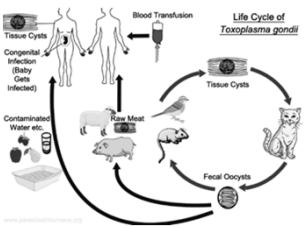
Summary

- Hairworms provide a great model system to study host manipulation
- Proteins produced by worms and host lead to manipulation
  - Suggests that manipulation is evolved trait
- Parasite and host benefit by collaborating.



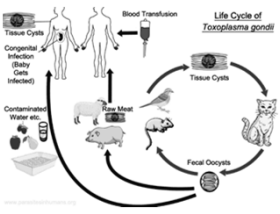
Toxoplasma

- Protozoa that causes toxoplasmosis
- Encysts in brain
- Infection by
  - consuming undercooked meat
  - ingesting feline feces
- 30% of Americans are infected



Toxoplasma

- Normal intermediate host is the mouse
- Needs to get into cat
- Toxoplasma does this by manipulating mouse behavior
  - Decrease motor function
  - Decrease fear of cat smell
- Evolutionary explanation
- And in humans?



Toxoplasma in humans

- Behavioral changes
  - More suspicious
  - Less moralistic
- Impaired motor performance
  - Computerized tests
  - Higher rates of car accidents
- Increased suicidal behaviors and self directed violence
- Link to schizophrenia

Flegr. 2007. Schizophrenia Bulletin 33: 757-760