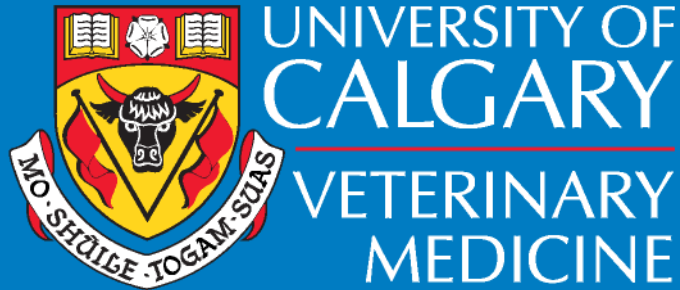


Johne's Disease in cattle and Crohn's Disease in humans – linked diseases??



**Herman Barkema, Mariá Negrón, Kevin Rioux,
Jeroen De Buck, Karin Orsel, Subrata Ghosh and
Gilaad Kaplan**

Alberta Inflammatory Bowel Disease Consortium

Slowly progressive disease of the intestines

Can affect all ruminants

Slowly progressive

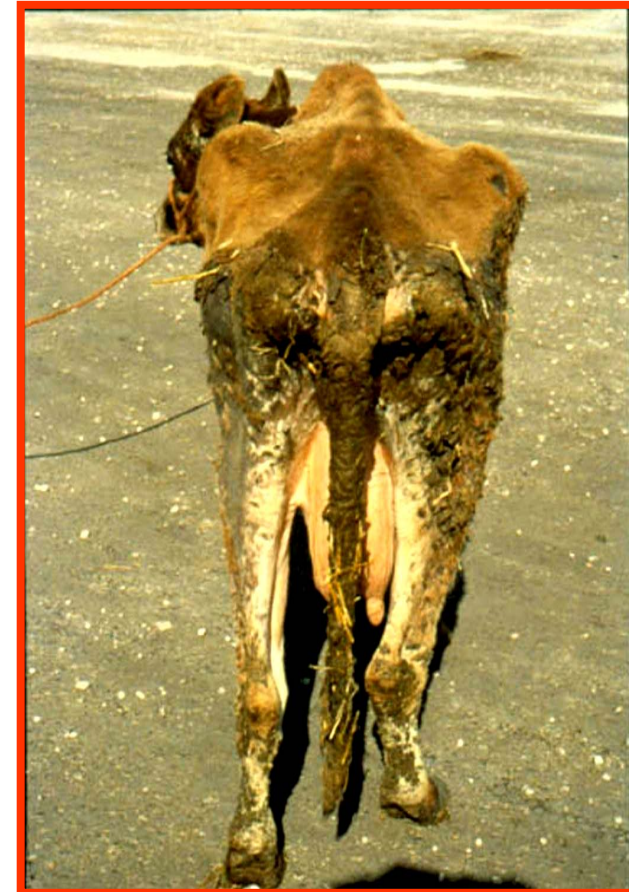
Intermittent chronic diarrhea

Weight loss despite appetite

Apparent 3-6 yrs

Onset associated with stress

No successful treatment



***Mycobacterium avium* subspecies *paratuberculosis* (MAP)**

- **bacterial disease**
- **far‘relative’ of TBC**
- **grows very slowly**



Infected the intestines of all ruminants

Excreted in faeces and milk



Prevalence of MAP infection

MAP infection estimated to cost the Canadian dairy industry \$15-90 million annually and the American dairy industry \$200-250 million annually

Herd seroprevalence AB = 40-74%

Herd-level prevalence based on fecal culture = 28-57%

NAHMS Dairy 2007 study: 69% of U.S dairies infected



MAP is present also in...



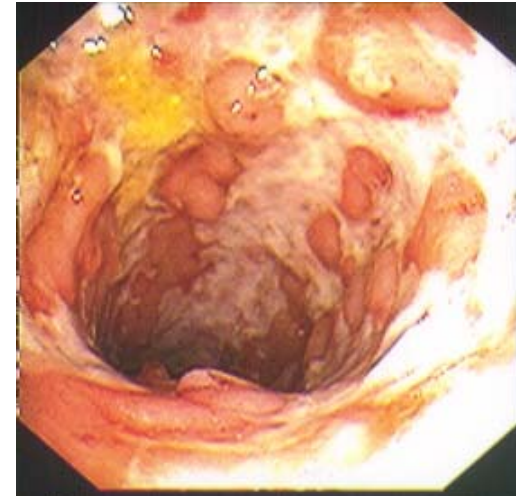
An inflammatory bowel disease

Peak of onset – 15-25 yrs

Chronic urgent diarrhea

Nausea, vomiting and fevers

Weight loss and lack of energy

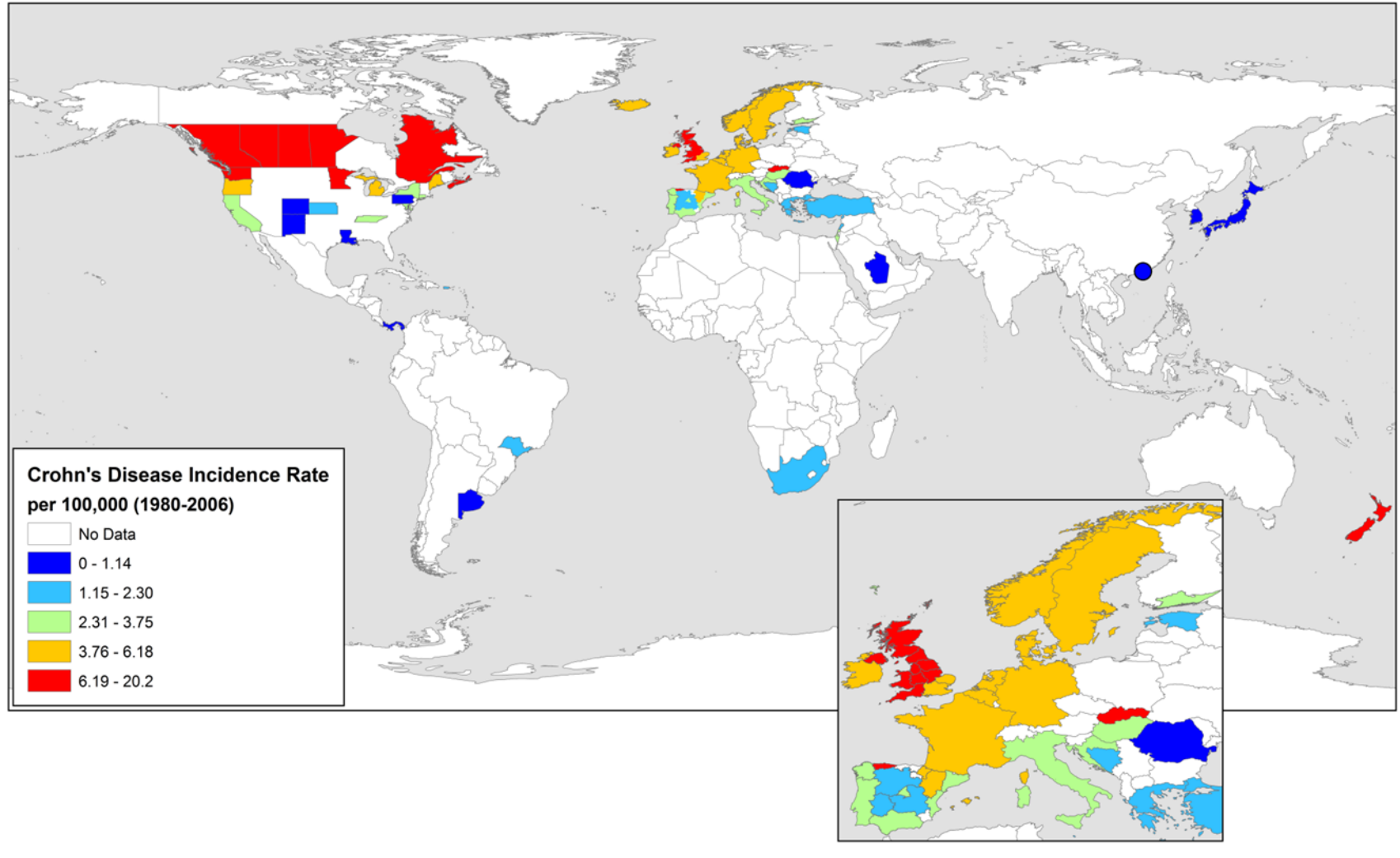


50,000-100,000 patients in Canada

Over a half million patients in US

Increasing incidence (4000% since 1930)





Molodecky et al. The incidence of the inflammatory bowel diseases across time and geography: A systematic review of international trends



Environmental factors (diet, smoking, stress, infectious agents)

Family clustering without a clear pattern of simple Mendelian inheritance

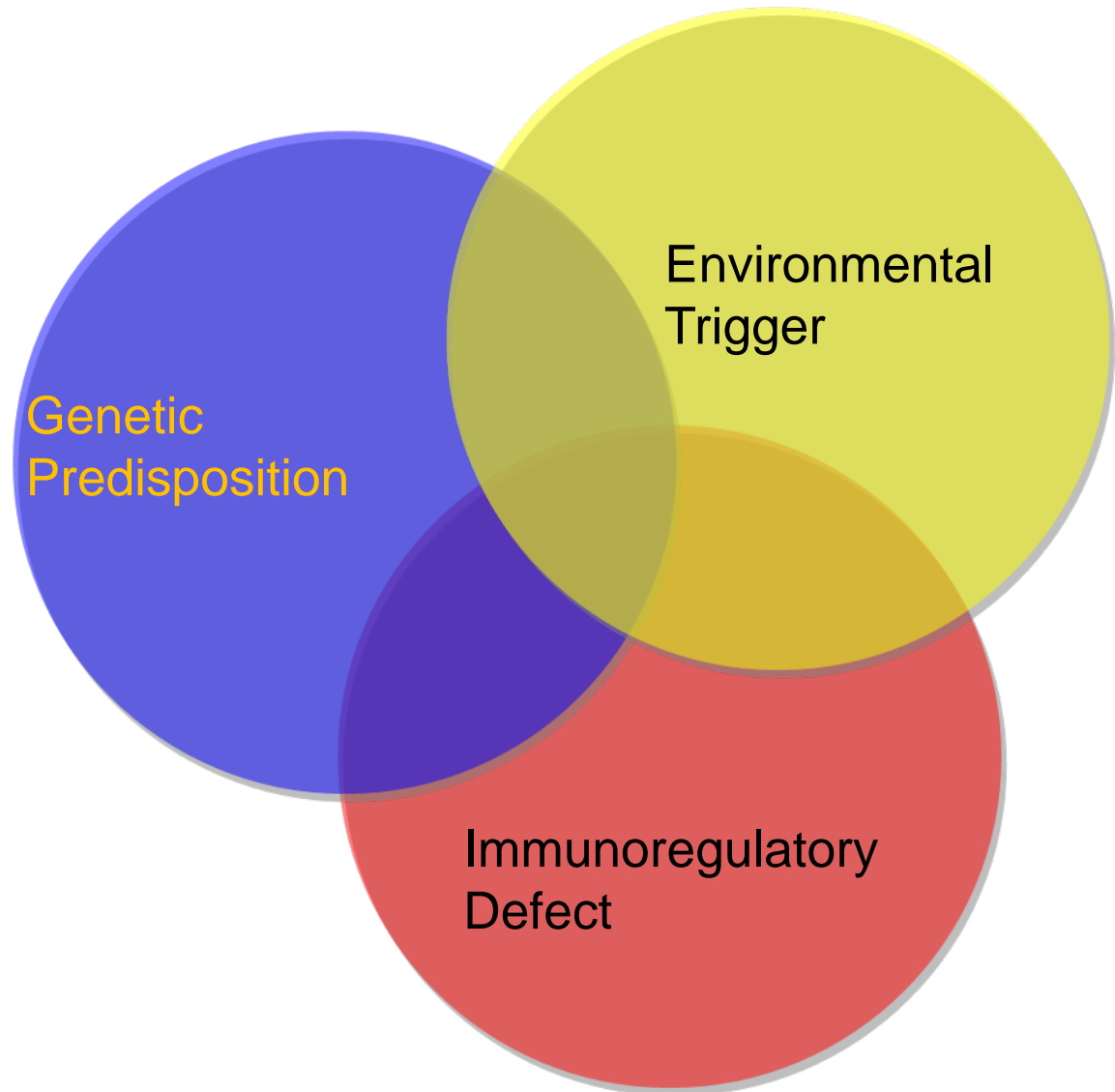
→ Crohn's disease results from a complex interaction between genetic susceptibility and environmental exposures:

Genome – Microbe – Environment

Research focus of AB IBD Consortium



Etiology



Pathogenesis: Genetics

Barriere Function

- PTGER4, ITLN-1, MUC19

Innate Immunity

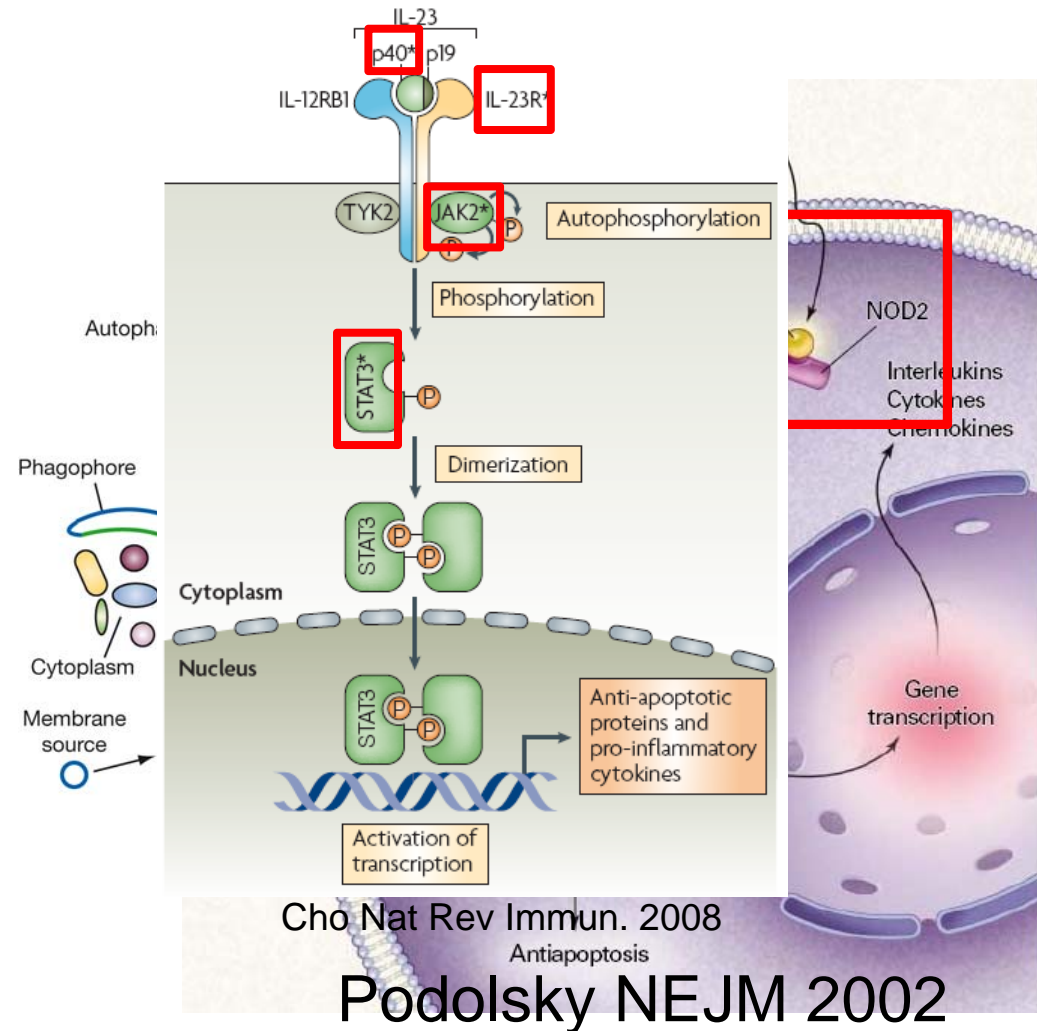
- NOD2

Autophagy

- ATG16L1, IRGM

Adaptive Response

- IL23R, IL12B (p40)
- JAK2/STAT3



Barrett *Nature Genetics* 2008



5-20% of CD patients have a familial history of IBD

All “susceptibility genes” proposed are associated with the immune system

No single gene mutation is sufficient/necessary to cause disease

NOD2/CARD15

ATG16L1

Defective immune response impairs the clearance of luminal antigens and/or pathogens and leads to the development of chronic intestinal inflammation



Bacteria, viruses and parasitic pathogens have been implicated either with the development or relapse of IBD

Bacteria

MAP

E. coli

Campylobacter spp.

Salmonella spp.

Identified either by culture or PCR in biopsy specimens, blood and/or stool samples from CD patients



Evidence for

Evidence against

Conclusions (?)



Crohn's first described in 1913 by Dalziel who also wrote:

“Tissue characteristics from Johne's and Crohn's patients are so similar as to justify a proposition that the diseases may be the same”



Researchers have been trying since 1952 to grow mycobacteria from surgically removed Crohn's tissue

In 1984, Chiodini cultured MAP from the gut wall of children with Crohn's Disease



MAP in Crohn's disease

Method	MAP detectie (%)		P value	Reference
	CD	Controls		
Culture	86	5.6		Schwartz <i>et al.</i> 2000
	63	10	<0.0001	Sechi <i>et al.</i> 2005
	42	9	0.0019	Bull <i>et al.</i> 2003
	50	0	0.0005	Naser <i>et al.</i> 2004
IS900 PCR	45	6.3	<0.05	Collins <i>et al.</i> 2000
	40	0		Ryan <i>et al.</i> 2002
	92	26	0.0002	Bull <i>et al.</i> 2003
	83	17	<0.005	Romero <i>et al.</i> 2005
	52	5	0.0001	Autschbach <i>et al.</i> 2005
<i>In situ</i> Hx	40	0		Hulten <i>et al.</i> 2001
	73	0		Sechi <i>et al.</i> 2001
	67	0	<0.005	Romero <i>et al.</i> 2005



Group	Mean MAP detection	Range
Crohn's	57.6%	(30-92%)
Controls	5.1%	(0-17.9%)



Some studies MAP-positives in UC patients

Discordance between IS900 en culture

High sero-reactivity against MAP antigen in Crohn's but lack of specificity

Inability to demonstrate cellular immune response to MAP antigens



MAP not more frequently found in patients with other bowel inflammations

Other mycobacteria found as often in Crohn's patients and bowels of people that don't have Crohn's disease

Cattle strains also found in humans, while sheep strains are not



Recently, two excellent meta-analyses

Pooled OR = 7.01 in 18 studies

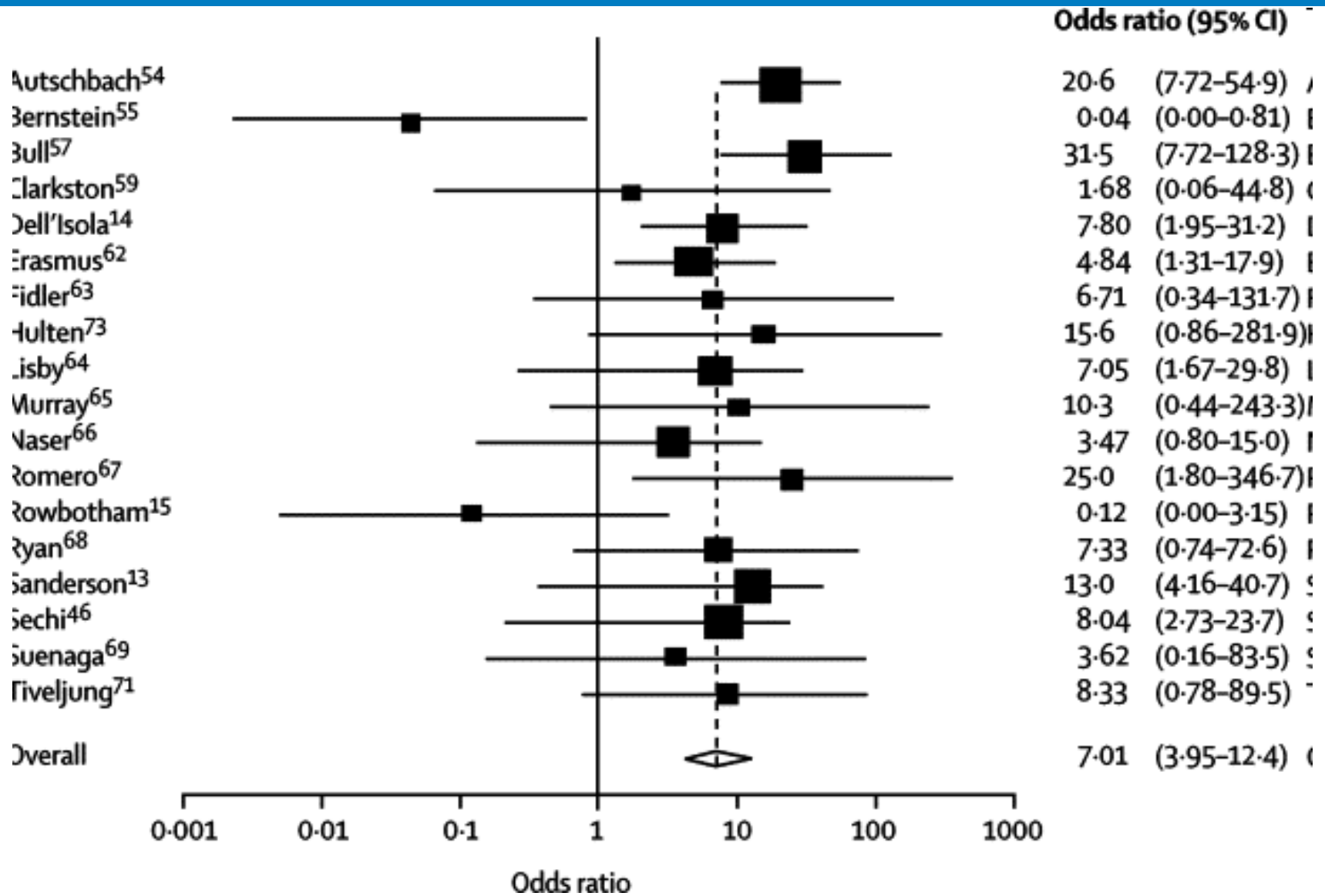
When using UC as comparison

OR = 4.13

Feller et al., 2007 and Abubakar et al., 2007



Meta-analysis Feller et al. 2007 PCR



No proof of causality...

Infection

Colonization

Contaminant

Opportunist

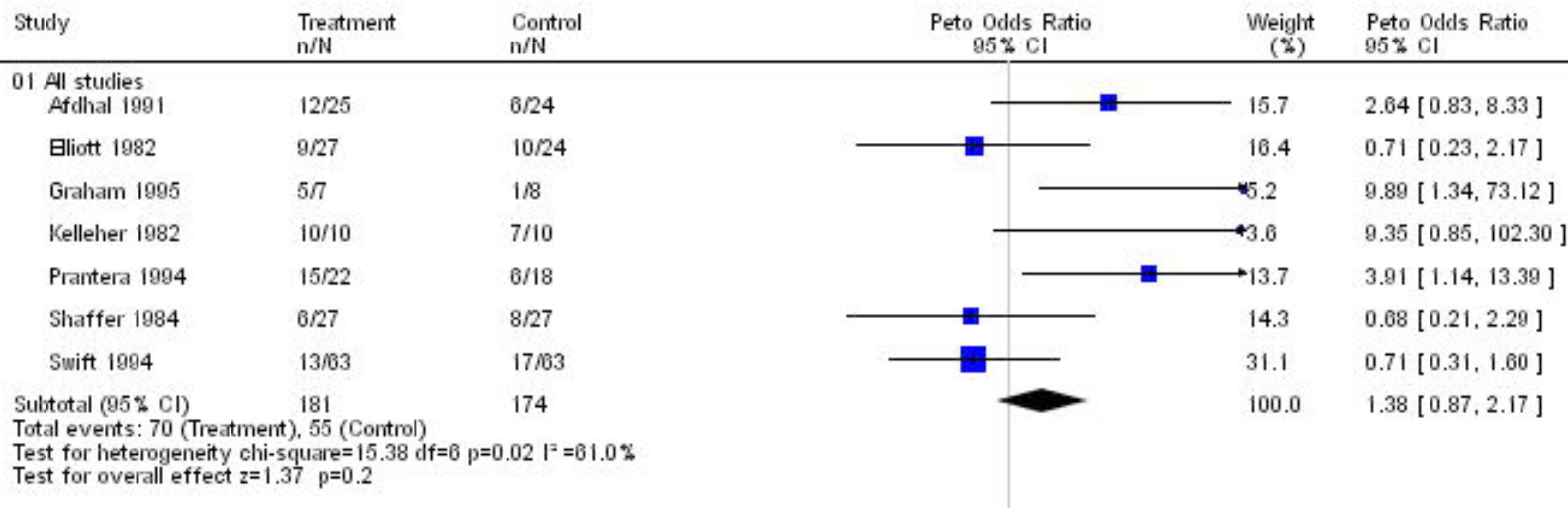


What happens to CD if you try
eliminate MAP?



Anti-TB therapy in Crohn's disease

Review: Anti-tuberculous therapy for maintenance of remission in Crohn's disease
 Comparison: 01 Anti-tuberculous therapy
 Outcome: 01 Maintenance of remission



These clinical trials used agents ineffective in treating MAP



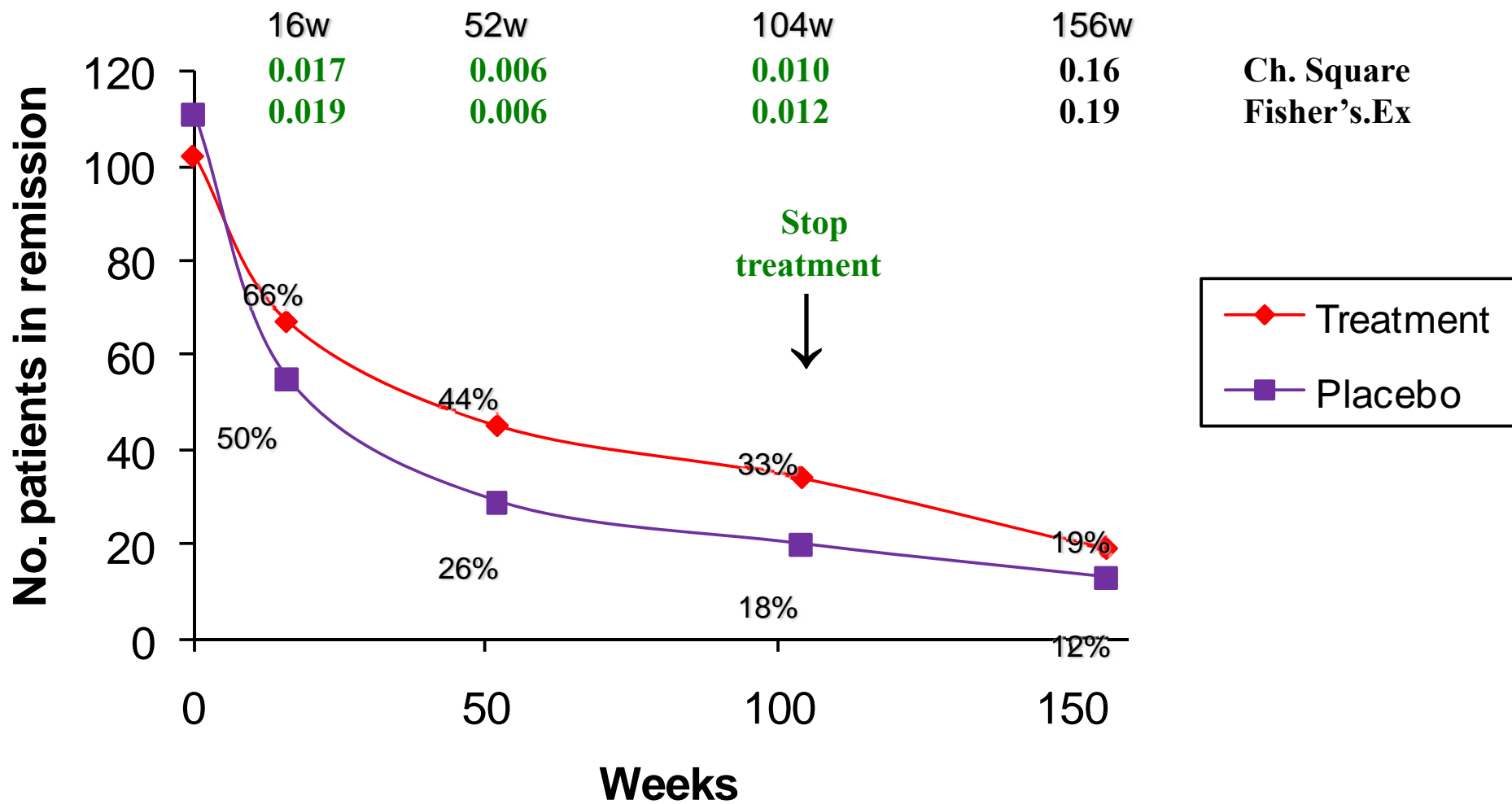
Macrolide-based therapy

Author	N	Design	Length (mo.)	Treatment	Result
Gui 1997	52	Open	6-35	Rifabutin Clarithromycin	89% remission
Douglass 1999	20	Open	6	Rifabutin Clarithromycin Clofazimine	50% remission
Shafran 2002	36	Open	4-17	Rifabutin Clarithromycin probiotics	58% response
Borody 2005	52	Open	6-108	Rifabutin Clarithromycin Clofazimine	62% remission
Selby 2005	213	RCT	16-104	Clarithromycin Clofazimine	NS at 40 mo.

No trial correlated response with elimination of MAP



Australian RCT of R/C/C



Didn't try to document presence and eradication of MAP!

Selby *et al.* 2005

Cannot see MAP

Failure of antimicrobial therapy

Effectivity of immune modulators

Identification of Crohn's related genes

Lacks plausible mechanism of tissue injury

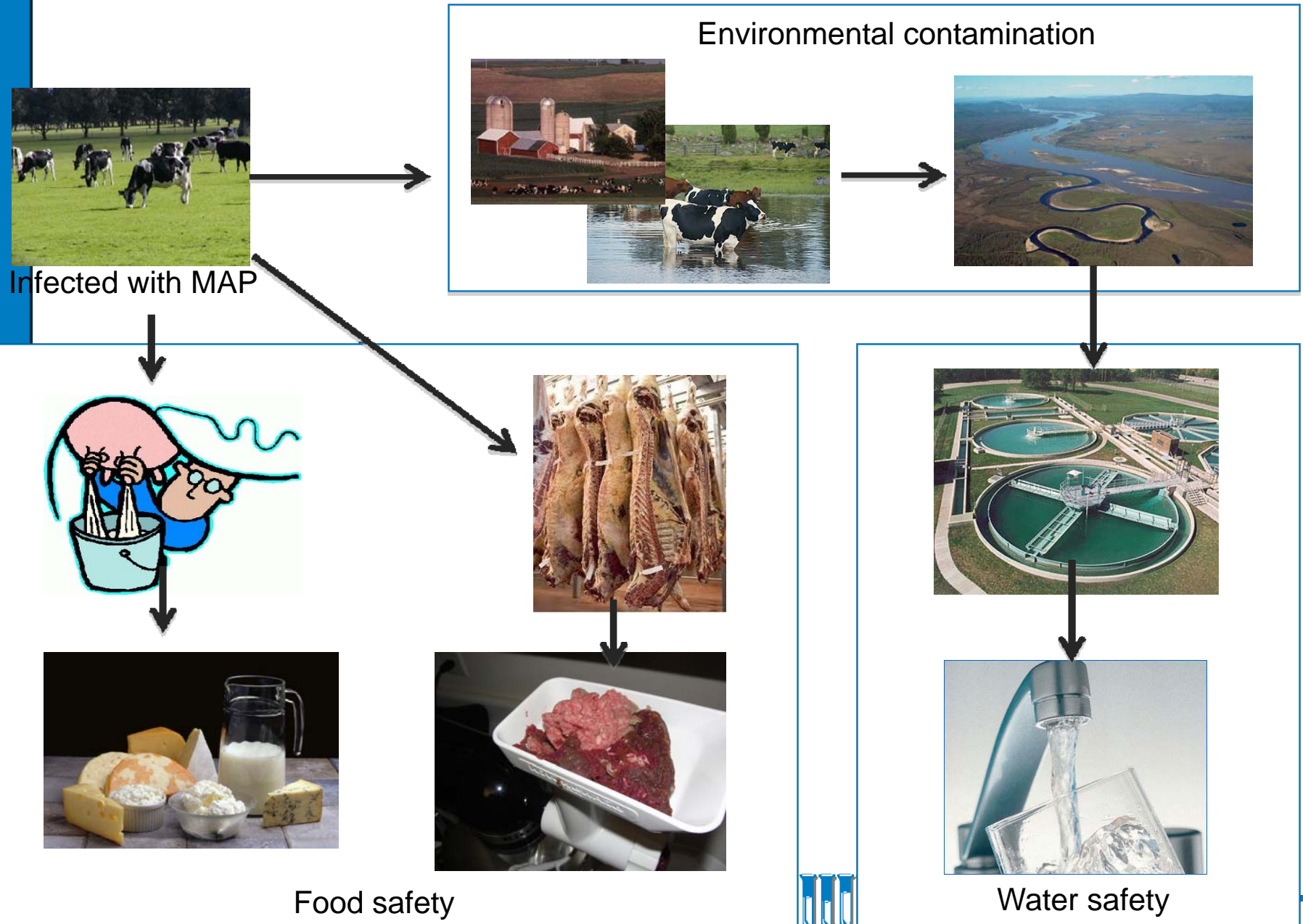
Sweden has a high incidence of Crohn's disease but no Johne's disease

Farmers do not have Crohn's disease more often

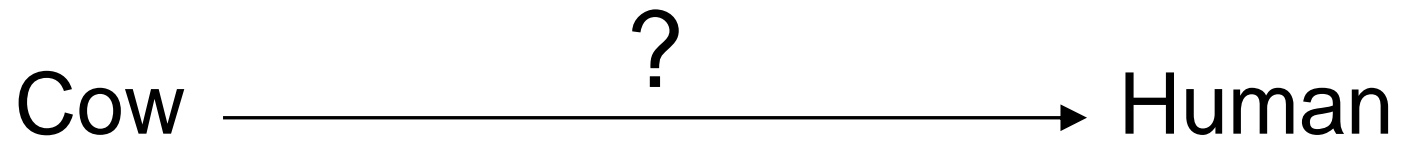


Suggested spread of MAP

Adapted from: Paratuberculosis: Organism, Disease Control Book



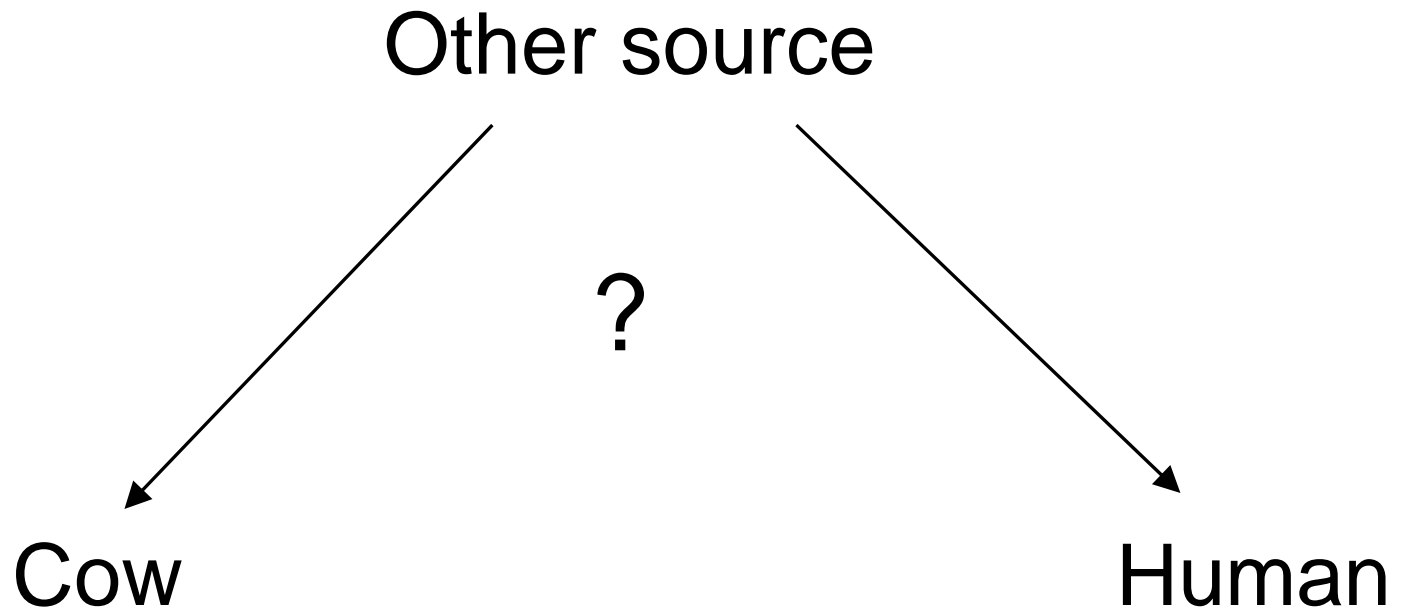
Association or causation?



Milk?
Beef?
Water?



Association or causation?



So, ??

The role of MAP in Crohn's disease is very possible, but not proven

Role of other bacteria?

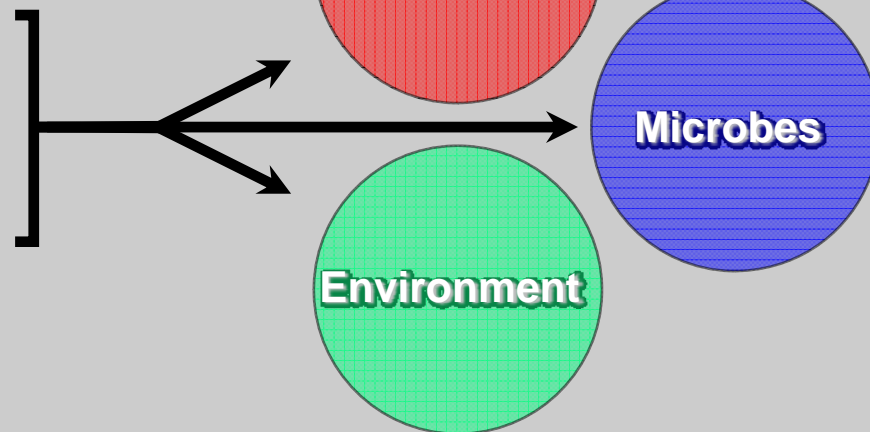
MAP not a necessary or sufficient cause of CD?

Infection of Crohn's patients through milk is possible, but not proven

Other ways of infection (e.g. water and meat) are not well studied



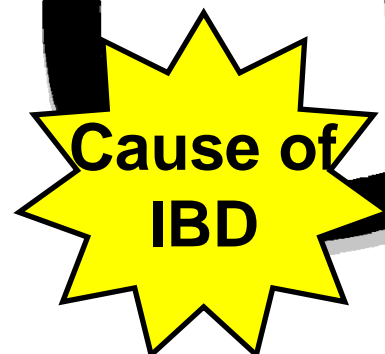
TIER 1



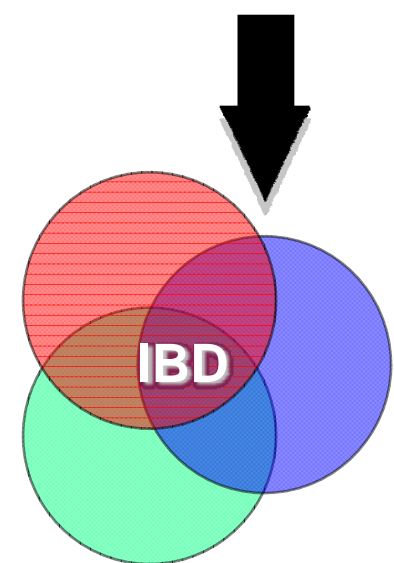
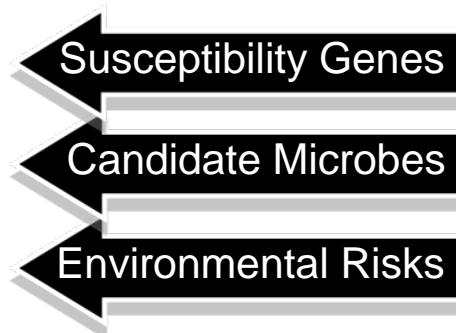
Integration of research

Findings and Data Shared

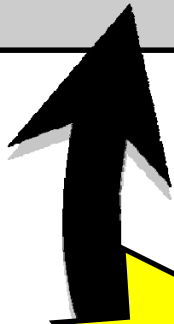
Training



Mechanisms
Discovered through a variety of methods



TIER 2



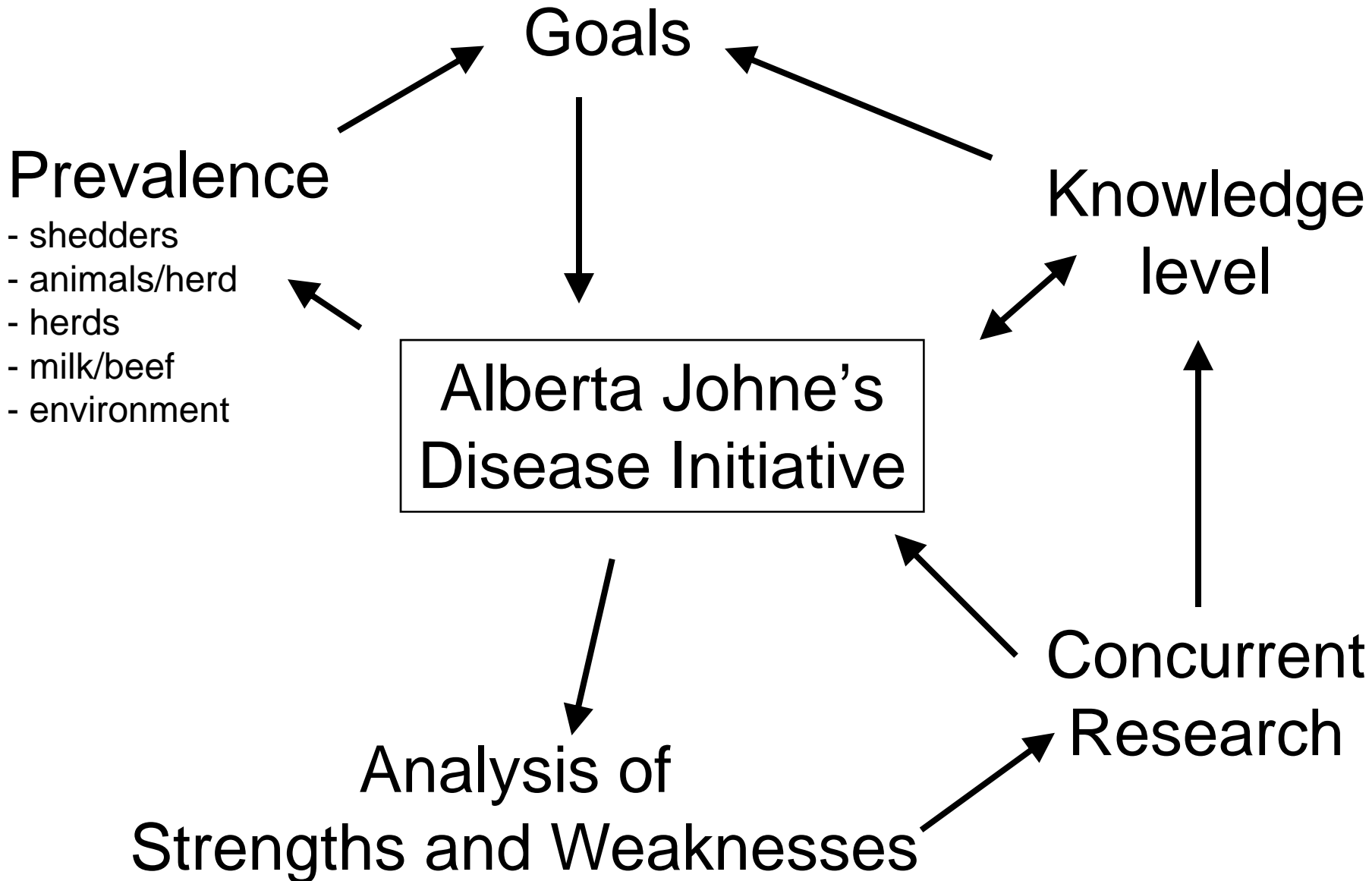
Post-operative cohort

Mesenteric lymph node and creeping fat study

GIS study on association between farming activities, bacteria in drinking water and occurrence of IBD



MAP research in Alberta



Rienske Mortier

‘Age- and dose dependent susceptibility of cattle for MAP-infection’

Nick MacKenzie

‘PE and PPE proteins as targets in Johne's Disease immunodiagnosis’

Joel David

‘Typing of MAP using HRMA technique’

Maria Negron

‘The role of bacteria in Inflammatory Bowel Disease’



Robert Wolf

‘Prevalence of MAP and adoption of BMP’s on Alberta dairy farms’

Mathieu Pruvot

‘The role of the interaction between beef cattle and North American elk in the epidemiology of economically important infectious diseases in SW Alberta’

Taya Forde

‘Molecular diagnosis of MAP in Wood Bison’

Yasmin Khalil

‘Cytokine profiling and macrophage homing to the gut in MAP infection’



Aito Ueno

‘Immune cell profiling after MAP infection’

Christina Ahlstrom

‘Transmission pattern profiling of MAP between and within Canadian dairy herds by fast and discriminating strain typing’

Jillian Steele

‘Herd characteristics to describe differences in MAP infection in Caribou’



Questions?



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