Infections

- Last 30 years: dramatic increases in illnesses and infections in children
- Otitis media increased from an uncommon illness (1970s) to now affecting 90% children under the age of 2 years
- Increases in hospital admissions for respiratory infections and gastroenteritis, 4.5%/year

‘Big Four’

- Same time frame: significant increases in incidence of
  - Asthma
  - Allergies: now up to 50% children, with 20% children with food allergies
  - ADHD – to 10%, learning difficulties, 20%, speech delay 5%
  - Autism, to 1/100

Infections and Neurodevelopmental disorders

- Association between infections in childhood and speech difficulties in boys
- Espec associations with ear infections (OM), mental health disorders, asthma, allergies
- Recurrent OM infancy also correlated with lower IQ scores, poor school performance, behavioural difficulties
- High correlation between prevalence OM and autism
- Correlation with high levels of antibiotic use and autism

Infections and the Brain

- 4 main mechanisms by which infections can affect brain function
  1. Direct infection –eg viral encephalitis, bacterial meningitis
  2. Toxin-mediated effects
  3. Immune-mediated effects – eg autoantibody formation against brain cells
  4. Disruption of normal metabolism and nutrition
Infections in Autism

Overt Infections

- Overt infection common in history of children prior to diagnosis of autism
- Most common - otitis media (AOM)
- Recent study 206 children under age of 3 with autism: average of nearly 10 bouts of AOM, received an average of 12 courses of antibiotics
- Over 1/3 all given to children under 12 months

Otitis Media

- Previously incidence of respiratory tract infections and otitis media peaked 4 to 6 years old
- Now peak incidence between 6 and 12 months of age.
- Otitis media now said to occur in 48% children within first 6 months
  - In 79% within 1st 12 months
  - In 91% by age 24 months
- Comparing to 1989 figures: 62% of children by 12 months, 83% at age 3 years

Microbiology of AOM

- Bacterial - 75% (aspiration of middle ear fluid)
- *Strep pneumoniae* the major bacterial infecting agent
- Nasopharyngeal carriage of *Strep pneumoniae* extremely common, especially in young children
- Nasal carriage identified as the first step in the acquisition of pneumococci and a necessary first step for invasive pneumococcal disease

Strep Carriage

- Most children become nasal carriers of *Strep pneumoniae* at least once during 5 month period.
- Acute sinusitis in previous 3 months the major associated factor with carriage of *Strep pneumoniae*
- Otitis media in the past 3 months the only associated factor with carriage of antibiotic resistant *Strep pneumoniae*
- Connection between nasal strep, otitis media and sinusitis

Biofilms

- Bacteria live in biofilms
- Sessile colonies surrounded by mucus slime
- Protected from predation by protozoa, antibiotics, antibodies, immune cells
- Antibiotic resistant – factor 1000x
- Cause 80% culture-negative, antibiotic resistant, chronic infections
Gastric Reflux and Infections

- Gastro-oesophageal reflux increasing in incidence in infants in recent years
- Now a major cause of morbidity and failure to thrive, particularly in neurologically impaired children.
- Has been linked with asthma, respiratory infections, childhood chronic sinusitis and AOM
- Reflux part of clinical spectrum of cow’s milk allergy and intolerance

Infections, Reflux, Gastric Acid Inhibitors and Autism

(2006 study) - correlation with use of GAH in infancy for GORD and increased rates of gastroenteritis (2x, to 50%) or pneumonia (5x, to 12%)

 increased risk might result from overgrowth of intestinal pathogens (in the stomach) in a low-acid environment”,
- intestinal pathogens might colonise the oral space and subsequently be aspirated
- anecdotally- rates of reflux, diagnosed GORD and use of PPI’s high in infancy in children who later develop autism

Antibiotics

- Recurrence rate otitis media post antibiotics 2-6x cf placebo
- Antibiotic treatment for otitis media increases nasopharyngeal colonisation of non-pneumococcal alpha-haemolytic streptococcus at 2 months
- Antibiotic resistance in these strains is becoming very common
- Implications for respiratory infections not clear but increasing incidence respiratory infections and gastroenteritis (4%/year)

Antibiotics and Allergy

- Several studies linking prenatal and early infancy antibiotics to later allergy and asthma
- Meta-analysis of 8 studies
- receiving at least 1 antibiotic before 12 months old doubled risk of childhood asthma
- risk dose dependant
- each extra course during first year of life increases risk 1.16 times

Antibiotics and Allergy

- Maternal antibiotics in utero induces Th2 response in infant (immune shift)
- dose-related response
- increased risk asthma, hayfever, eczema, respiratory tract infections, candida

Antibiotics and the Immune System

- Antibiotics promote TH2 immune shift
- Change in gastrointestinal flora thought to be major reason behind TH2 immune system shift documented post antibiotics.
- Antibiotics can induce increased intestinal permeability
INFECTIONS, ANTIBIOTICS, DYSBIOSIS & MINDDD: Feeding Our Bugs
Dr Robyn E Cosfird

Developing Picture:
- Early infancy – frequent breastfeeding difficulties, early formula
- Reflux, colic, treated with PPI
- Recurrent infections, biofilms
- Frequent antibiotic use, change in gastrointestinal flora, further Th2 shift
- Failure maturation of Th1 response, Th2 immune skewing
- Gut-based immune based dysfunction
- Loss of oral tolerance
- Food allergies, sensitivities

Immune System in Autism

Gut-Based Immune Responses in Autism
- Levels of all TH2 cytokines significantly ↑: (IL-4,5,13)
- ↑ activation both TH1, TH2; TH2 predominance, no compensatory ↑ regulatory IL-10
- Th2 shift - ↑ CD4+ T cells, ↑ IL-4, TNF, ↓ IFN-gamma, ↑ IgA, ↑ IgE (Gupta S)
- ↑ antibody-producing B cells 20%, ↑ NK cells 40%
- ↑ total serum proteins
- ↑ albumin, gammaglobulins, partic IgG, IgG2, IgG4

Gut Immune Responses in Autism
- Recent study 100 autistic children on unrestricted diets, 77 on restricted diets, (controls with and without food sensitivities or restricted diets):
  - challenge with bacterial toxins or milk proteins resulted in strong pro-inflammatory response, less able to down-regulate
  - “may indicate the intrinsic natures of dyregulated innate immune responses in autism spectrum disorder children (with gastrointestinal involvement)

Autoantibodies in Autism
- High levels of autoantibodies against myelin basic protein (MBP) and neuron axonal filamentary protein in children with autism (60 to 70%) correlate with raised measles, HHV-6 antibody titres
- Various other antibodies to neuronal tissue also documented
- Recent study of 171 autistic individuals found ‘high levels of antibodies against brain tissue’, one particular unidentified protein apparently involved
- Antibodies also to frontal cortex, 5HT1A receptors, cerebellar neurofilament

Immunology in Autism: autoantibodies (cont)
- Antibodies to 3 cross reactive peptides also raised in children with autism cf controls (IgG, IgA, IgM)
  - chlamydia pneumoniae (CPP)
  - streptococcal M protein (STM6P)
  - milk butyrophilin (BTN)
- Streptococcal spp and enterococcal spp have been found to be elevated in large scale studies assessing common pathogens in cow’s milk
The Gut in Autism

“All diseases begin in the gut”
Hippocrates 460-370 BC

Gut Dysfunction in Autism

Gut abnormalities documented:
- gastric hypochlorhydria with resultant raised pH (Horvath)
- duodenitis (Horvath)
- reduced intestinal disaccharidase enzyme function (Horvath) & dipeptidase function
- colitis, lymphoid hyperplasia (Wakefield,Krigsman)
- increased intestinal permeability (Eufemia)

Infancy, Immunity and Gut Microflora

- Children born with Th2 dominance
- Th1 responses mature in first year or so of life
- mainly in response to microbial antigens
- dependant on growth of symbiotic gut bacteria
- reduction symbiotic bacteria (lactobacilli) in Caesarian-born infants

Gut Microflora and Gut Immunity

- Recent studies indicate that antigen-presenting cells (APC) - dendritic cells, M cells - routinely sample intestinal microflora
- APC release appropriate cytokine stimulus, dependant on strain of bacteria
- induce either inflammatory (IL-2,4,5,6,13) or anti-inflammatory (IL-10, TGF-beta) response

GALT

- Oral tolerance - mechanism by which immunological tolerance to food antigens and commensal gut flora is induced
- typically, absence of appropriate oral tolerance results in exaggerated Th2 responses
Intestinal Flora

- Intestinal flora affects gut permeability
- Failure of the mucosal immune system usually begins with various changes to the population of microflora & subsequent dysfunction of the mucosal epithelial barriers
- In the absence of beneficial intestinal microflora, disturbance in intestinal absorption of macromolecules is more severe than in its presence.

Intestinal Flora

- Healthy adult gut - av 1.5-2kg bacteria, 10-50% total body cells, over 600 species
- Symbiotics, indigenous - Bifidobacteria, Lactobacilli, Propionibacteria, physiological strains Ecoli, Peptostreptococci, Enterococci
- Opportunistic - Bacteroides, Peptococci, Staphylococci, Streptococci, Bacilli, Clostridia, Yeasts (esp Candida), Enterobacter (Proteus, Citrobacter Klebsiella), etc
- Transitional - us gram-negative bacilli

Alterations of the faecal aerobic microbial flora in patients with autism (n=36)

Butt HL, Cosford RE, Roberts TK, Dunstan NR, McGregor NR, Ellis L
University of Newcastle Bioscreen

The mean % distribution of Escherichia coli for the autistic patients was 56.3% of the total aerobic flora, compare to 80% in healthy control subjects (p=0.03).

Enterococcus/Streptococcus spp., was significantly higher in counts (40.1%) in the autistic patients than in healthy subjects (5%). (p<0.0007)

Colonization pattern of faecal aerobes:
NSW autistic patients (n=27) and control subjects (n=117)

Butt HL, Emms TM, Cosford RE, Duff J, Patterson D
Bio21, Molecular Science & Biotechnology Institute, & Bioscreen, University of Melbourne;
Northern Beaches Care Centre, Mona Vale, NSW; Behavioural Neurotherapy Clinic, Doncaster, VIC;
Secretsane Medical Center, Runaway Bay, QLD.

Autism

Retrospective, multi-center (NSW, VIC, & QLD), comparative study of the faecal microbial flora of Autistic Patients and control subjects in, 2004-2005

Butt H.L. \ Emms T.M. \ Cosford, R. \ Duff, J. \ & Patterson D
Bio21, Molecular Science & Biotechnology Institute, & Bioscreen, University of Melbourne;
Northern Beaches Care Centre, Mona Vale, NSW;
Behavioural Neurotherapy Clinic, Doncaster, VIC;
Secretsane Medical Center, Runaway Bay, QLD.
Colonization pattern of faecal aerobes: VIC autistic patients (n=16) and control subjects (n=117)

- Predominant Streptococcal overgrowth seen was non-pneumococcal alpha-haemolytic or non-haemolytic streptococcus
- Similar to pattern of overgrowth seen in nasal carriage post antibiotic therapy
- ?Swallowed down to gut in biofilm

The importance of *E. coli* metabolites in human metabolism

- *E. coli* and other Gram negative bowel organisms produce chorismate

Chorismate is the precursor for:
- 4-aminobenzoate
- 4-OH-benzoate
- anthranilate
- prephenate
- isochorismate

![Chemical structures]

- folic acid
- ubiquinol (CoQ10)
- tryptophan
- tyrosine, phenyalanine
- menaquinone (Vit K)

Functions of Symbiotic Flora

- Antibacterial action:
  - bifidobacteria lactis -ability stimulate innate & Th1 activity, enhance production antimicrobial cytokines (IL-1, IL-2, IFN-gamma, TNF-alpha), helps restore Th1 deficiency
  - eg normal human flora capable of permanently eradicating *Clostridia difficile* from gut
  - Lactobacillus (GG) also been demonstrated to reduce *Clostridia* in gut

Gastrointestinal Dysfunction in Autism

- Inflammation, increased gastrointestinal permeability
- reduction in enzyme function
- gastrointestinal dysbiosis with colonosis
- predominant overgrowth of streptococcal/enterococcal species, loss of E coli, lactobacilli sp
- disruption of normal gastrointestinal flora function & metabolites

Gut-Brain Axis

- “gut-brain interactions may be central to abnormal neural development and the subsequent expression of aberrant behaviors”
  - Andrew Wakefield

www.mindd.org
Gut-Brain Axis

4 mechanisms
1. Enteric nervous system, neurotransmitters
2. Gut-Associated-Lymphoid Tissue, gut-based immune responses
3. Increased intestinal permeability, increased direct toxin and antigen passage
4. Disruption usual gut bacterial metabolism and nutrients

Metabolic Abnormalities

- Numerous metabolic abnormalities demonstrated in children with autism
- “often manifest complex biochemical, metabolic and immunologic abnormalities that a primary genetic cause cannot readily account for”

Enterococcus/Streptococcus and D-Lactic Acid

- Facultative anaerobes
- Homofermentative- produce only lactic acid from glucose fermentation
- (NMR exo-metabolic profiling) – significantly more lactic acid than Ecoli
- Predominantly D-lactic acid
- Associated with decreased faecal pH

D-Lactic Acidosis

- Short Bowel Syndrome (SBS) – adult patients – headaches, weakness, cognitive impairment, fatigue, pain, severe lethargy related to D-lactic acidosis
- Increased intestinal permeability reported in these cases believed to be due to increased colonisation of lactic acid producing Gram-positive bacteria Increased permeability would likely result in increased absorption of microbial metabolites including D-lactic acid
- D-lactate poorly metabolised in humans as lack enzyme D-lactate dehydrogenase

Organic Acidosis in Autism

- 1/3 low bicarb
- 2/3 raised anion
- Consistent with chronic metabolic organic acidosis,
  Streptococcal/enterococcal overgrowth usual in cases organic acidosis
- (personal data)

Gastrointestinal Dysfunction in Autism

- Increased gastrointestinal permeability
- gastrointestinal dysbiosis with colonosis
- predominant overgrowth of streptococcal/enterococcal species
- markers for malabsorption
- disruption of normal gastrointestinal flora function and metabolites
- metabolic sequelae- organic acidosis
Streptococcus and Autoantibodies

Strep and Autoimmune Disease

- Strep species known to be associated with neuro-psychiatric disorders, abnormal movements, auto-immune phenomenon
  - scarlet fever
  - post-streptococcal glomerulonephritis
  - rheumatic fever
  - Sydenham’s chorea (associated with rheumatic fever)

PANDAS

- PANDAS- Paediatric Auto-Immune Neuro-psychiatric Disease Associated with Streptococcus
- OCD exacerbations follow GAS infections, assoc abnormal behaviours, emotional lability, separation anxiety, attentional difficulties
- associated psychiatric disturbances - OCD, generalised anxiety, depression, conduct disorders, hyperkinetic disorders

PANDAS- ADHD, Autism

- Raised strep titres (ASOT, antiDNAse B) correlate with ADHD” (excluding OCD, tics)
- raised ASOT correlated with increased basal ganglia volumes
- raised strep titres common in ASD (c. 50%), often when no recent history of overt streptococcal infection (personal data)

Streptokinase and DPPIV

- Streptococcal enzyme (streptokinase) binds to DPPIV enzyme, more strongly than gluten, casein
- DPPIV is proline endopeptidase – tissue enzyme which hydrolyses bonds containing proline: necessary for degradation of gluten and casein
- DPPIV also CD26 lymphocyte marker;
- Plays key role in growth & differentiation of lymphocytes; T cell mediated immune responses and cytokine production
- Potential for streptococcal overgrowth to interfere with degradation of gluten and casein and immune function
- ‘dysfunctional membrane peptidases and autoantibody production may result in neuroimmune dysregulation and autoimmunity’

Streptococcus and Immune Responses

- Strongly immunogenic, antibodies frequently cross react with human tissue
- Generally, bacteria elicit B cell response, viruses T cell response
- All streptococci - ability to nonspecifically stimulate T cell as well as B cell response
- Inadequate T cell or B cell response could result in chronic streptococcal infection,
- Could then result in depletion both B cell and T cell immune mediators
Streptococcal toxins
- Known strep toxins - eg erythrogenic toxin of scarlet fever, b-haemolytic membrane toxin of b-haem strep
  - streptokinase - ↑inflammatory mediators TNF alpha, IL1, IL6 (T cells)
  - neuraminidase - aid in establishment of viral infections
  - NADase- depletes NAD, necessary for recycling glutathione
  - glutathione peroxidase - necessary for virulence

Streptococcal Toxins & effects
- Streptococcal pyogenic toxins - ‘superantigens’ - stimulate certain T cells to proliferate without processing the toxin
  - ↑ TNF alpha reduces lymphocyte glutathione
  - ↑ TNF alpha implicated in Tourette’s, facial tics, ACD, schizophrenia

Chronic Streptococcal Effects
- In gut:
  - damage to intestinal wall - glycosaminoglycans
  - stimulation of mucus
  - increased intestinal permeability
  - reduction in disaccharidase enzymes (glycosidases)
  - XS antigenic stimulation of GALT
  - reduction symbiotic bacteria
  - reduction of bacterial metabolites
  - bacterial toxins - cross gut wall
  - XS D-lactate

Chronic Streptococcal Infection
- antibody production, cross-reactions human tissue (kidney, heart valves, ?brain)
- systemic inflammation (TNF-alpha, IL-1, IL-6)
- depletion of NAD (numerous enzyme systems including recycling glutathione)
- direct depletion of glutathione (via glutathione peroxidase)
- reduces resistance to viral infections (neuraminidase)

Streptococcus - colonisation
- *Strep pneum* normally colonises nasopharynx 20-40% children
- prepubertal girls, vulvovaginitis strep sp is common
- vulvovaginitis GABHS seen only in this age group (vulvovaginal candidiasis not found in prepubertal girls)
- GBHS vaginal swabs 1/3 pregnant women

A Theoretical Model for the Aetiology of Autism
- Recurrent infections, commonly streptococcal, treated with antibiotics, result in loss of protective gut flora and overgrowth of predominantly streptococcal species in gut
  - loss of beneficial bacteria disrupts normal gut function and production and digestion of nutrients
overgrowth of streptococcal species inflammatory to gut wall, resulting in loss of sulphated GAGs in intestinal mucus and increased gastrointestinal permeability

increased permeability results in passage of macromolecules, abnormal immune activation and food intolerances, peptiduria

- streptococci toxigenic, resulting in numerous metabolic blocks, membrane dysfunctions
- possibly neurotoxic, resulting in abnormal neurological manifestations of Autism, ADHD, CFS
- immune response to streptococci result in cross-reactions with neuronal tissue, with further intracerebral irritation and inflammation
- excitotoxicity (NMDA receptor, glutamate), free radical damage (reduced glutathione) final common pathway in brain for these effects

Chronic Streptococcal Infection and Autism

- Results in widespread metabolic disruption
- inflammation, free radical damage
- auto-immune reactions
- affect brain development and function

Management

Basis: initially: nourish gut, cells, brain

- reduce inflammation, oxidation
- Organic whole food 'Primitive' Traditional alkaline diet
- 5 P's gut: probiotics, colostrum, glucosamine, glutamine etc
- organic whole food-based supplements (high inherent antioxidant capacity, immune support phytonutrients)
- EFA
- Nutrients specific to nervous system (eg Mg, Zn, B6, SAME, glycine, gingko etc)
- Nutrients to enhance detoxification (eg selenium, N-acetyl cysteine, silymarin, glycine, zeolites etc)
- homeopathy

FOODS AND STREP

- Strep feeds on milk
- Strep feeds on sugar
- Strep produces enzymes which can interfere with breakdown of milk and wheat
- Strep can cause inflammation in the small intestine which can disrupt enzyme production (disaccharidases) and affect breakdown of carbohydrates
- Probiotics and fermented foods compete with strep in gut
- High alkaline diet counter acidosis produced by strep strep

Further

- 'chelation' – nutrient based, homeopathy
  Also supportive therapies
  - Primitive reflex integration – homeopathy, kinesiology, chiropractic, movement therapies
  - Auditory/visual pathway integration – sound therapies, biofeedback
  Always treat the whole child not just the symptoms
INFECTIONS, ANTIBIOTICS, DYSBIOSIS & MINDDD: Feeding Our Bugs
Dr Robyn E Cosfird

Neuro-Immune Gastrointestinal Dysfunction Syndrome

- Theory: ADHD, Autism - auto-immune and toxin-mediated disease processes in conjunction with malnutrition secondary to gastrointestinal dysfunction, in genetically susceptible individuals
- Toxins - associated with streptococcal infections, streptococcal and food-derived toxins traversing intestinal barrier, other environmental toxins (mercury)
- Streptococcal and milk antigens activate gut-associated lymphoid tissue (GALT) and cross-react with neuronal antigens

Summary

- Multisystem disorder: neurological immunological gastrointestinal biochemical
- Metabolic Immunological Neurological Digestive Disorder MINDDD

Dr Robyn Cosford
MBBS(Hons)FACNEM
Conjoint Lecturer,
School of Biological Sciences,
University of Newcastle.

Northern Beaches Care Centre
56 Garden St
Narrabeen, NSW 2101
02 99133744, Fx 0299132788
reception@nbcc.net.au

www.mindd.org