

NUTRITION, IMMUNE FUNCTIONS AND HEALTH

▷ MUCOSAL ADAPTIVE IMMUNITY: IMPACT OF EXOGENOUS STIMULI AND FEEDING

The vast majority of antigenic challenges confronting the body make contact with mucosal surfaces. To maintain homeostasis in the extensive and vulnerable mucosae, they are protected by specialized anti-inflammatory immune components / processes such as secretory IgA (sIgA) antibodies and tolerance against innocuous antigens, including nutrients and components of commensal bacterial flora. The induction of mucosal immunity is highly dependent on exogenous stimuli and the neonatal period is critical. Both the mucosal barrier with its reinforcement by sIgA and the immunoregulatory network require successful adaptation after birth, depending on appropriate microbial colonization as well as adequate timing and dose of dietary antigens when first introduced. Breastfeeding can in several ways have a beneficial effect on the developing immuno-phenotype of the infant.

Per BRANDTZAEG, *Rikshospitalet University Hospital, Oslo, Norway*

▷ NUTRITIONAL MODULATION OF EARLY THYMIC DEVELOPMENT

The thymus has long been known to be exquisitely sensitive to malnutrition. Such effects may be most potent in early life and may 'programme' the future capacity for replenishment of the circulating T-cell pool. Thymic development starts early in gestation and the organ reaches its maximum size in proportion to total body mass shortly after birth. Ultrasound measurements have shown that nutrition and/or infectious exposures in pregnancy significantly impact on thymic size at birth. Breast-fed babies have larger thymuses than formula-fed babies suggesting that breast-milk may contain thymic trophic factors. Preliminary evidence suggests that breast-milk IL-7 may be a key mediator. Our studies in Gambia suggest that impaired thymic development may explain a profound season-of-birth effect on mortality in young adulthood.

Andrew PRENTICE, *London School of Hygiene and Tropical Medicine, UK*

▷ MODULATION OF ORAL TOLERANCE IN RATS BY PERINATAL SUPPLY OF ESSENTIAL FATTY ACIDS

Essential fatty acids (EFA) influence the immunological response by regulating the Th1/Th2 ratio. There are controversial findings of serum fatty acid patterns in allergy and it has been discussed if the increased prevalence of allergy lately may be related to such environmental factors as the dietary fatty acid intake. The Western diet has resulted in a recommendation of low saturated fat content but an increase in the supply of n-6 fatty acids, increasing the ratio of n-6/n-3 fatty acids 3 to 10 fold over recommendations. We have found that a reduction of EFA in the rat dams during pregnancy and lactation resulted in suppression of the serum antibody levels and delayed hypersensitivity reactivity (DTH) against ovalbumin (OA) in the adult offspring exposed via the milk at 10-16 days to OA, indicating oral tolerance. Higher amount of TGF- β mRNA in the draining intestinal lymph nodes suggested a role of regulatory T cells. In a parallel study the dams were supplied with different ratios of n-6/n-3 EFA, either of a ratio of 0.4, which resulted in tolerance to OA and an unrelated protein, presumably as an effect of TGF- β production; or of 9, which did not give any immunological tolerance; or very high, up to 200, which resulted in tolerance specific for OA, presumably due to T cell anergy. The studies suggest that the maternal supply of EFA, both qualitatively and quantitatively has effects on the development of immunological tolerance in the off-spring. The results may be important in the context of increased allergy in many communities.

Birgitta STRANDVIK, *Göteborg University, Sweden*

▷ MOTHER-CHILD BREASTFEEDING

The newborn has a tiny immune system. It expands fast after birth, mainly due to exposure to the colonizing gut flora, preferably coming from the mother. While the immune system of the infant develops it is to be protected by the trans-placental IgG antibodies from the mother. But such inflammation-inducing antibodies require complement and phagocytes to defend and those systems are deficient in early infancy. The milk contains numerous defence factors which all protect immediately from birth by stopping infectious agents already on the mucosa, without inducing inflammation. The milk secretory IgA antibodies are produced in the mammary gland by lymphocytes from the mother's gut and are directed against the microbes in the mother's milieu. Numerous signals via the milk seem to enhance the infant's protection, also long-term.

Lars A. HANSON, *Göteborg University, Sweden*

▷ TRACE ELEMENTS AND IMMUNITY

Major trace elements such as iron, zinc and copper are essential minerals for energy production and cellular proliferation processes. They modulate several components of the immune system and influence the predisposition of the host to pathogenic microorganism-driven processes. The effect of individual trace element is difficult to evaluate due to frequent concomitant deficits in other micronutrients. We have used a human model of pure iron deficiency – adult women with gynecologic bleeding – to study some mechanisms of impaired immune response in these subjects. The effect of mineral supplementation trials on immuno-competence and frequency of infectious processes in deficient individuals, and its relevance in public health will be reviewed.

Carlos MUNOZ, *INTA, University of Chile, Santiago, Chile*

▷ NUCLEOTIDES AND IMMUNO-COMPETENCE

Nucleotides are normal components of the human diet from birth onwards. The immune system is not able to fulfil the needs of cell nucleotides exclusively by *de novo* synthesis and they preferentially utilize the salvage pathway recovering nucleosides and nucleo-bases coming from blood and the diet. Nucleotides have been involved in different aspects of the systemic immune response, such as lymphocyte maturation, activation and proliferation, modification of lymphocyte subpopulations, modulation of the phagocytic activity of macrophages, modulation of the delayed hypersensitivity and allograft and tumour responses and modulation of immunoglobulin production, as well as in the modulation of gut-associated immune responses. Dietary nucleotides appear to influence protein biosynthesis as well as signal cell membrane transduction which may contribute to modulate the expression of a number of genes, some of which can directly affect the levels of intestinal cytokines.

Angel GIL, *University of Granada, Spain*

▷ NUTRITIONAL REGULATION OF NO SYNTHESIS AND ITS IMPLICATIONS FOR HEALTH

Nitric oxide (NO), a product of L-arginine catabolism, plays crucial roles in virtually every cellular and organ function in the body. This discovery has unified the diverse areas of life science research, including nutrition, physiology, immunology, neuroscience and pathology. A wide array of dietary factors, includ-

ing amino acids, glucose, fructose, cholesterol, fatty acids, vitamins, minerals, phyto-estrogens, ethanol, and polyphenols, are either beneficial to health or contribute to the pathogenesis of chronic diseases partially through modulation of NO production. New knowledge about nutritional regulation of NO synthesis is very beneficial for improving health and preventing disease in both humans and animals.

Guoyao WU, *Texas A&M University, College Station, USA*

▷ BIOACTIVE MOLECULES IN MILK: HOMEOSTATIC FUNCTION IN THE INTESTINAL MUCOSA

Breast-fed newborns experience a lower incidence of infectious processes, inflammation and allergy. Immunomodulatory molecules in milk may modulate host reactivity during neonatal colonization. In recent years, we have worked on the following molecules:

1. TGF- β 2 downregulates immune activation of intestinal epithelial cells and lamina propria immune cells.

2. Soluble CD14, a pattern recognition receptor for bacteria, can play a sentinel function for mucosal cells and may be involved in modulating innate and adaptive immune response.

3. Soluble Toll-like receptor 2 found in milk and serum may play a role in regulating microbial-induced toll-like receptor stimulation.

Eduardo SCHIFFRIN, *Nestec Ltd, Nestlé Research Center, Lausanne, CH*

▷ NUTRITION AND INFECTION: CURRENT ISSUES OF GLOBAL IMPORTANCE

Malnutrition is the largest single risk factor for death and disability in the world, especially in less developed countries. The evidence for this will be reviewed, along with examples of current issues confronting researchers. Examples will illustrate the need for basic research linked to questions of public health importance.

Rebecca J. STOLTZFUS, *Cornell University, Ithaca, USA*

▷ IMMUNOCOMPETENCE ASSESSMENT: A USEFUL TOOL TO EVALUATE NUTRITIONAL STATUS IN EATING DISORDERS AND IN ATHLETES

Nutritional deprivation often causes immunodeficiency, leading to increased frequency and severity of infection. The patients with anorexia nervosa (AN) frequently show immune impairments, although they are less severe than would be expected considering their nutritional deficiencies; AN patients even seem to be surprisingly free of infectious complications. It is readily noticeable that some of the consequences of heavy physical training and inadequate intakes of elite athletes are very similar to the symptoms and complications of AN. Therefore, our aim is to find out the usefulness of assessing immunocompetence in order to evaluate the nutritional status of both groups.

Ascension MARCOS, *Consejo Superior de Investigaciones Científicas, Madrid, Spain*

▷ MOLECULES AT THE INTERFACE BETWEEN IMMUNE AND METABOLIC REGULATION: A MAJOR ROLE FOR LEPTIN?

Over the last few years a series of molecules known to play a function in the metabolic function have also been shown to play an important role in the regulation of the immune response. In this context, the adipocyte-derived hormone leptin has been shown to regulate the immune response both in normal as well as in pathological conditions. More specifically, it has been shown that conditions of reduced leptin production are associated with increased infection susceptibility. Conversely, immune-mediated disorders such as autoimmune disorders are associated with increased secretion of leptin and production of pro-inflammatory pathogenic cytokines. In this context, leptin could represent the "missing link" between immune response, metabolic function and nutritional status. Strategies aimed at interfering with the leptin axis could represent innovative therapeutic tools for infections and autoimmune disorders.

Giuseppe MATARESE, *Istituto di Endocrinologia e Oncologia Sperimentale, Napoli, Italy*

▷ POLYUNSATURATED FATTY ACIDS, INFLAMMATION AND IMMUNITY

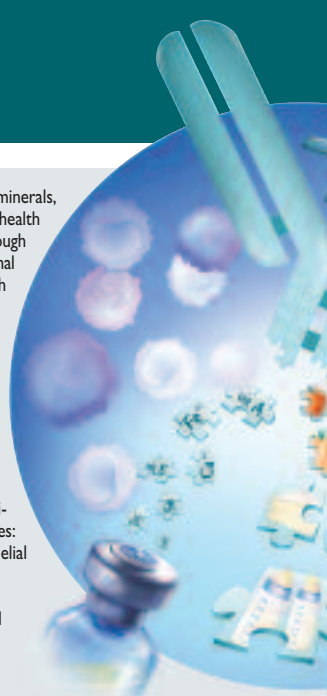
In most human diets consumption of n-6 polyunsaturated fatty acids (PUFAs) exceeds that of n-3 PUFAs. The n-6 PUFA arachidonic acid gives rise to the eicosanoid family of inflammatory mediators (prostaglandins, leukotrienes and related metabolites) and through these regulates the activities of inflammatory cells, the production of cytokines and the various balances within the immune system. Fish oil and oily fish are good sources of long chain n-3 PUFAs. Consumption of these fatty acids decreases the amount of arachidonic acid in cell membranes and so available for eicosanoid production. Thus, n-3 PUFAs act as arachidonic acid antagonists. Components of both natural and acquired immune effectors, including the production of key inflammatory cytokines, can be affected by n-3 PUFAs. Although some of the effects of n-3 fatty acids may be brought about by modulation of the amount and types of eicosanoids made, it is possible that these fatty acids might elicit some of their effects by eicosanoid-independent mechanisms. Such n-3 fatty acid-induced effects may be of use for protection against and therapy of acute and chronic inflammatory processes, and for disorders which involve an inappropriately-activated immune response.

Philip C. CALDER, *University of Southampton, UK*

▷ REGULATION OF IMMUNE RESPONSES TO ORAL IMMUNIZATION BY DIETARY FAT EMULSIONS

Oral tolerance has been nominated for many decades as a prophylactic strategy of food allergy; however, there is a scanty evidence that proves the feasibility even in animal studies because of difficulties in oral immunization. We hypothesize that food components other than proteins might play a critical role in determining immunological consequences, and dietary fat was found to be a determinant. Repeated feeding of emulsified bovine β -lactoglobulin with dietary oil at tolerogenic dose elicited a systemic humoral immune response in mice, whereas the same dose of the aqueous antigen molecule failed even against the booster feeding of the emulsified antigen.

Tetsuo KANEKEO, *Food Science Institute, Meiji Dairies Corporation, Kanagawa-ken, Japan*





▷ ENTERAL NUTRITION AS A TOOL FOR IMMUNOMODULATION IN CRITICALLY ILL PATIENTS

The immune system, especially the innate immune system, is malfunctioning in people with "Western lifestyle", especially in elderly, persons with chronic diseases and in critically ill. About 80% of the immune system is located in the gastrointestinal tract. This offers unique possibilities to enforce the immune functions through enteral nutrition. Several studies support the efficacy of aggressive enteral nutrition compared to total parenteral nutrition to reduce morbidity, especially various infections such as chest – located infectious processes in critically ill patients. The efficacy of various tools such as antioxidants, dietary fibres and probiotic bacteria are analysed and discussed. Special focus is given to recent experience with synbiotic treatment in postoperative and critically ill patients.

Stig BENGMARK, *University College, London, UK*

▷ GLUTAMINE IN THE CRITICALLY ILL

Glutamine as such an abundant amino acid was traditionally considered non-essential in the diet. Over the last forty years the important role that glutamine plays in metabolism has come to be realised and that in severe illness it may even become indispensable. Its omission from conventional amino acid solutions for pharmaceutical reasons in retrospect was a mistake. The development of a conditional deficiency may be sufficient to compromise survival in some intensive care patients. Progress in the application of glutamine and glutamine dipeptides in clinical settings will be discussed and the outcome benefits reviewed.

Richard D. GRIFFITHS, *University of Liverpool, UK*

▷ N-3 FATTY ACIDS IN SEPTIC PATIENTS

Modern clinical nutrition focuses not only on caloric support but introduces immune-modulation by feeding a patient. Conventional lipid emulsions provide building blocks for cell membranes but also supply precursors for inflammatory lipid mediators thereby increasing the inflammatory processes in septic patients or aggravating pulmonary dysfunction in acute lung injury. In contrast, inclusion of n-3 fatty acids into nutritional regimes provides competing substrates to be metabolized to alternative immune-modulating lipid mediators. They may be used to positively influence leukocyte-endothelial interactions, vasomotor response, and organ failure. Given via enteral or parenteral route, n-3 fatty acids improved intestinal perfusion and lung function in septic models. Furthermore, modulation of immunity, shortened ventilation, and reduced infection rates were shown in critical ill patients. These positive effects may translate in introducing n-3 fatty acids as adjunct therapy for septic patients.

Konstantin MAYER, *University of Giessen, Germany*

▷ IMPACT OF VITAMIN E ON IMMUNE FUNCTION AND INFECTIOUS DISEASES IN THE AGED

Ageing is associated with dysregulation of immune and inflammatory responses, which is believed to contribute to the higher morbidity and mortality from infection, neoplastic, and inflammatory diseases. Supplementation of vitamin E above the recommended levels has been shown to enhance immune functions and to be associated with increased resistance against several pathogens in the aged. This presentation will focus on the role of vitamin E in modulating immune responses and mechanisms by which vitamin E exerts its effects. Findings from infectious disease models and gene expression studies will be discussed.

Sung Nim HAN, *Tufts University, Boston, USA*

▷ NUTRITIONAL ZINC, OXIDATIVE STRESS AND IMMUNO-SENESCENCE. BIOCHEMICAL, GENETIC AND LIFE STYLE IMPLICATIONS FOR HEALTHY AGEING

Background: Zinc and other antioxidant micronutrients control the development and function of the immune cells, the activity of stress-related proteins (metallothioneins, chaperones, ApoJ, PARP-1, nitric oxide/NO, MsrA) and antioxidant enzyme (SOD) and help to maintain genomic integrity and stability, reflecting diet-gene interactions. During ageing, the intake of zinc decreases due to inadequate diet or/and intestinal malabsorption, thus causing frailty, general disability and increased incidence of age-related degenerative diseases (cancer, infections and atherosclerosis). Therefore there is a potential to improve the health of Europe's ageing population with simple cheap zinc tablets. However, too much zinc can be toxic, and certain groups of people may have adequate zinc even in old age. The Zincage project will clarify the intrinsic biochemical mechanisms and the impact of nutrient zinc on the genome during immunosenescence.

Project Objectives:

1. To establish a comprehensive picture of the links between metallothioneins (MTs), chaperones, nitric oxide, telomere length, cell cycle kinetics, ApoJ, PARP-1 activity, signal transduction and DNA repair in lymphocytes from old and nonagenarian subjects, thus providing potential novel biomarkers for immunosenescence and successful ageing, respectively.
2. To establish the mechanistic basis of the interrelationships among MTs, chaperones, nitric oxide, telomere length, cell cycle kinetics, ApoJ, PARP-1 activity, signal transduction and DNA repair. At the same time, the role played by the proteasome system in eliminating harmful related stress proteins whose accumulation within the cytosol is dangerous, will be carefully examined as well as the function of peptide methionine sulfoxide reductases.
3. To provide a simple genetic screening method in order to identify people at risk of zinc deficiency.

Eugenio MOCCHIGIANI, *INRCA, Ancona, Italy*

▷ THE IMMUNE SYSTEM AS A PREDICTOR OF LONGEVITY. ROLE OF ANTIOXIDANTS PREVENTING IMMUNOSENESCENCE AND INCREASING LIFE SPAN

It is known that a well preserved function of the immune cells is an excellent marker of health. We have confirmed that the functional competence of leucocytes, determined by means of a battery of parameters that change with age, is a marker of "biological age" and a good predictor of longevity. We have also shown that immunosenescence is due to the oxidative stress (linked to raised production of

free radicals and decreased antioxidant defences) that occurs with age. Several studies, performed both in human subjects and experimental animals, have shown that the ingestion of antioxidants by aged individuals causes a "rejuvenation" of the immune system, and we have observed that this is accompanied by an increased longevity in the experimental animals. These results support the oxidation/inflammation theory of ageing, as well as the usefulness of antioxidants for life expectancy increase and the role of the investigated leucocyte functions as markers of health and longevity.

Mónica De La FUENTE, *Complutense University of Madrid, Spain*

▷ NUTRIGENETICS AND THE METABOLIC SYNDROME

The metabolic syndrome is defined as a cluster of clinical and biological abnormalities associated with insulin resistance and with high risk for cardio-vascular diseases. This condition has a multifactorial origin with both a genetic basis and strong effects of environmental factors such as nutrition. Nutrients interact with genes to modulate metabolism and gene variability in various genes involved in nutrient absorption and partitioning associates with the risk for the metabolic syndrome and associated vascular complications. A well established example is the nuclear receptor PPAR gamma, target of the anti-diabetic thiazolidiones, where the effect of the Pro12Ala variant allele is modulated by the saturated fat diet content. Obesity has also a strong effect on the metabolic syndrome modifying the secretion of various adipokines which contribute to a chronic inflammatory state. Among those, the diabetes and atherosclerosis protective hormone adiponectin. There is strong evidence from both genetic and genomic studies that hypoadiponectinemia is a primary determinant of diabetes.

Philippe FROGUEL, *Imperial College London & CNRS 8090 – Institut Pasteur de Lille, France*

▷ HOST-MICROBIAL RELATIONSHIPS IN THE INTESTINE: SENSING COMMENSAL AND PATHOGENIC BACTERIA

Functional genomics studies of the response to mono-association of germ-free mice with *Bacteroides thetaiotaomicron* have revealed that this symbiont modulates expression of genes involved in a broad range of important intestinal functions, including nutrient absorption, angiogenesis, xenobiotic metabolism, and fortification of the innate immune system. To explore the microbial determinants of beneficial versus pathogenic host-bacterial relationships, similar types of functional genomic studies were performed in germ-free mice colonized with *Listeria monocytogenes*, or its closely related non-pathogenic relative, *L. innocua*. The results of functional genomics analyses revealed that the host response to *L. innocua* is very similar to that documented with *B. thetaiotaomicron*. In contrast, *L. monocytogenes* evokes a complex program of host gene expression that includes NF-kappaB-dependent and IFN-responsive pathways. Strikingly, a *L. monocytogenes* mutant for listeriolysin produces a host response that phenocopies that of *L. innocua*. These findings reveal that the presence or absence of a single gene product that allows the microbe to access the cytoplasmic compartment of host cells can profoundly influence the host-bacterial relationship.

Marc LECUIT, *Institut Pasteur & Hôpital Necker, Paris, France*

▷ EFFECT OF COMMENSAL MICROBIOTA ON THE DEVELOPING IMMUNE SYSTEM

There are several indications that diseases caused by exaggerated or dysregulated immune responses are connected to the hygienic lifestyle of affluent Western societies. The commensal microbiota contains the vast majority of all exogenous antigens to which the immune system is exposed. Alterations in intestinal colonization pattern may, thus, underlie the dysregulation of immune responses. We have recently obtained evidence that regulatory T cells are expanded in infants colonized by toxin-producing *Staphylococcus aureus*. These toxins function as superantigens activating a broad range of T cells. The results suggest that poor T cell stimulation may underlie immune dysregulation, including allergy.

Ingerd ADLEBERTH, *Göteborg University, Sweden*

▷ PROBIOTICS IN ATOPIC DISEASE

Atopic disease, manifesting as atopic eczema, allergic rhinitis and asthma, is the most common chronic disease in Westernized world. The steep increase in prevalence during the 20th century has been linked to changes in environmental microbial factors (the hygiene hypothesis). In addition to pathogenic microorganisms, certain strains of indigenous gut microbiota have been shown to have properties that might be beneficial in combat against allergy. Both clinical and scientific aspects of probiotics in atopic disease will be discussed in the presentation.

Marko KALLIOMAKI, *University of Turku, Finland*

▷ PREBIOTIC ACTIVITY OF HUMAN MILK: APPLICATION FOR INFANT FORMULA

Breast milk contains prebiotic substances which selectively promote the growth of *Bifidobacteria* in the digestive microbiota of new-borns. It is considered that these bacteria exert a protective effect through an adjuvant activity on the intestinal immunity. By the use of adult gnotobiotic mice, either adult germ-free mice colonised with the faecal flora of either breast-fed (with bifidobacteria) or formula-fed (without bifidobacteria) babies, and then orally inoculated with a heterologous (nonmurine) simian SA-11 rotavirus strain, it was possible to test the only influence of the digestive microbiota of babies on the intestinal IgA anti-rotavirus antibody response. Results highlight the immuno-enhancing effect of the strain of *Bifidobacterium* present in the digestive microbiota of the breast-fed baby, *Bifidobacterium bifidum*, on the sIgA anti-rotavirus response. These data suggest the relevance of the supplementation of infant formula with prebiotics to promote selectively the growth of intestinal *Bifidobacterium* and consequently to enhance the specific sIgA antibody response towards rotavirus responsible of diarrhoeas in infants.

Marie-Christiane MOREAU, *INRA, Jouy-en-Josas, France*

▷ PROBIOTICS AND IMMUNE REGULATION : EFFECTS OF LACTOBACILLUS CASEI

Probiotics, including Lactobacilli, have been postulated to alleviate allergic and inflammatory diseases but mechanisms by which they exert their actions remain poorly understood. To examine whether anti-inflammatory effect of probiotics could involved immune modulation of effector and regulatory T cells, we examine whether *L. casei* DN-114 001 could affect antigen-specific T cell mediated skin inflammation. In order to get further insights into the mode of action of probiotics on innate immunity, we next investigate how *Lactobacillus casei* DN-114 001 could modulate the inflammatory pathways in intestinal epithelial cells.

Raphaëlle BOURDET-SICARD, *Danone Vitapole, Palaiseau, France*