Capsule

Modeling the mitochondrial cardiomyopathy of Barth syndrome with induced pluripotent stem cell and heart-on-chip technologies

Study of monogenic mitochondrial cardiomyopathies may yield insights into mitochondrial roles in cardiac development and disease. Wang and collaborators combined patient-derived and genetically engineered induced pluripotent stem cells (iPSCs) with tissue engineering to elucidate the pathophysiology underlying the cardiomyopathy of Barth syndrome (BTHS), a mitochondrial disorder caused by mutation of the gene encoding tafazzin (TAZ). Using BTHS iPSC-derived cardiomyocytes (iPSC-CMs), the authors defined metabolic, structural and functional abnormalities associated with TAZ mutation. BTHS iPSC-CMs assembled sparse and irregular sarcomeres, and engineered BTHS 'heart-on-chip' tissues contracted weakly. Gene replacement and genome editing demonstrated that TAZ mutation is necessary and sufficient for these phenotypes. Sarcomere assembly and myocardial contraction abnormalities occurred in the context of normal whole-cell ATP levels. Excess levels of reactive oxygen species mechanistically linked TAZ mutation to impaired cardiomyocyte function. This study provides new insights into the pathogenesis of Barth syndrome, suggests new treatment strategies, and advances iPSC-based in vitro modeling of cardiomyopathy.

Nature Med 2014; 20: 616

Eitan Israeli

Capsule

Inhibition of miR-25 improves cardiac contractility in the failing heart

Heart failure is characterized by a debilitating decline in cardiac function, and recent clinical trial results indicate that improving the contractility of heart muscle cells by boosting intracellular calcium handling might be an effective therapy. MicroRNAs (miRNAs) are dysregulated in heart failure but whether they control contractility or constitute therapeutic targets remains speculative. Using high-throughput functional screening of the human microRNAome, Wahlquist et al. identified miRNAs that suppress intracellular calcium handling in heart muscle by interacting with messenger RNA encoding the sarcoplasmic reticulum calcium uptake pump SERCA2a (also known as ATP2A2). Of 875 miRNAs tested, miR-25 potently delayed calcium uptake kinetics in cardiomyocytes in vitro and was upregulated in heart failure, both in mice and humans. Whereas adeno-associated virus 9 (AAV9)-mediated overexpression of miR-25 in vivo resulted in a significant loss of contractile function, injection of an antisense oligonucleotide (antagomiR) against miR-25 markedly halted established heart failure in a mouse model, improving cardiac function and survival relative to a control antagomiR oligonucleotide. These data reveal that increased expression of endogenous miR-25 contributes to declining cardiac function during heart failure and suggest that it might be targeted therapeutically to restore function.

Nature 2014; 508: 531

Eitan Israeli

Capsule

Cellular response to the growth hormone

The receptor for growth hormone is a well-studied representative of a family of cytokine receptors through which binding of hormone molecules at the cell surface is converted into a biochemical signal within the cell. Brooks and group used a combination of crystal structures, biophysical measurements, cell biology experiments with modified receptors, and molecular dynamics and modeling to decipher how the receptor actually transmits the information that a hormone molecule is bound. The results suggest that the receptors exist in inactive dimeric complexes in which two associated JAK2 protein kinase molecules interact in an inhibitory manner. Binding of growth hormone causes a structural change in the receptor that results in movement of the receptor intracellular domains apart from one another. This relieves the inhibition of the JAK2 molecules and allows them to activate one another, thus initiating the cellular response to the hormone.

Science 2014; 344:1249783

Eitan Israeli

Capsule

Essential role for autophagy in the maintenance of immunological memory against influenza infection

Vaccination has been the most widely used strategy to protect against viral infections for centuries. However, the molecular mechanisms governing the long-term persistence of immunological memory in response to vaccines remain unclear. Chen et al. show that autophagy has a critical role in the maintenance of memory B cells that protect against influenza virus infection. Memory B cells displayed elevated levels of basal autophagy with increased expression of genes that regulate autophagy initiation or autophagosome maturation. Mice with B cell-specific deletion of Atg7 (B/Atg7−/− mice) showed normal primary antibody responses after immunization against influenza but failed to generate protective secondary antibody responses when challenged with influenza viruses, resulting in high viral loads, widespread lung destruction and increased fatality. These results suggest that autophagy is essential for the survival of virus-specific memory B cells in mice and the maintenance of protective antibody responses required to combat infections.

Nature Med 2014; 30: 503

Eitan Israeli

Capsule

Intrinsic autoimmune capacities of hematopoietic cells from female New Zealand hybrid mice

Most systemic autoimmune diseases occur more frequently in females than in males. This is particularly evident in Sjögren's syndrome, systemic lupus erythromatosis (SLE) and thyroid autoimmunity, where the ratio of females to males ranges from 20:1 to 8:1. Our understanding of the etiology of SLE implies important roles for genetics, environmental factors and sex hormones, but the relative significance of each remains unknown. Using the New Zealand hybrid mouse model system of SLE, we present here a new fetal liver chimera-based system in which we can segregate effects of immune system genes from that of sex hormones in vivo. David and collaborators show that female hematopoietic cells express an intrinsic capacity to drive lupus-like disease in both male and female recipient mice, suggesting that this capacity is hormone independent. Particularly, only chimeric mice with a female hematopoietic system showed significantly increased numbers of germinal center B cells, memory B cells and plasma cells followed by a spontaneous loss of tolerance to nuclear components and hence elevated serum antinuclear autoantibodies. A protective effect of testosterone was noted with regard to disease onset, but not disease incidence. Thus, genetic factors encoded within the female hematopoietic system can effectively drive lupus-like disease even in male recipients.

Genes Immunity 2014; 15: 153

Eitan Israeli

Capsule

Endosomes are specialized platforms for bacterial sensing and NOD2 signaling

Despite the fact that NOD2 is well understood to have a key role in regulating innate immune responses and that mutations at the NOD2 locus are a common risk factor in inflammatory bowel disease and possibly other chronic inflammatory states, little is known about how its ligands escape from endosomes. Nakamura et al. show that two endo-lysosomal peptide transporters, SLC15A3 and SLC15A4, are preferentially expressed by dendritic cells, especially after TLR stimulation. The transporters mediate the egress of bacterially derived components, such as the NOD2 cognate ligand muramyl dipeptide (MDP), and are selectively required for NOD2 responses to endosomally derived MDP. Enhanced expression of the transporters also generates endosomal membrane tubules characteristic of dendritic cells, which further enhanced the NOD2-dependent response to MDP. Finally, sensing required the recruitment of NOD2 and its effector kinase RIPK2 to the endosomal membrane, possibly by forming a complex with SLC15A3 or SLC15A4. Thus, dendritic cell endosomes are specialized platforms for both the lumenal and cytosolic sensing of pathogens.

Nature 2014; 509: 240

Eitan Israeli

Capsule

MHC associations with clinical and autoantibody manifestations in European SLE

Systemic lupus erythematosus (SLE) is a clinically heterogeneous disease affecting multiple organ systems and characterized by autoantibody formation to nuclear components. Although genetic variation within the major histocompatibility complex (MHC) is associated with SLE, its role in the development of clinical manifestations and autoantibody production is not well defined. Morris and group conducted a meta-analysis of four independent European SLE case collections for associations between SLE sub-phenotypes and MHC single-nucleotide polymorphism genotypes, human leukocyte antigen (HLA) alleles and variant HLA amino acids. Of the 11 American College of Rheumatology criteria and 7 autoantibody sub-phenotypes examined, anti-Ro/SSA and anti-La/SSB antibody subsets exhibited the highest number and most statistically significant associations. HLA-DRB1*03:01 was significantly associated with both sub-phenotypes. The authors found evidence of associations independent of MHC class II variants in the anti-Ro subset alone. Conditional analyses showed that anti-Ro and anti-La subsets are independently associated with HLA-DRB1*0301, and that the HLA-DRB1*03:01 association with SLE is largely but not completely driven by the association of this allele with these sub-phenotypes. These results provide strong evidence for a multilevel risk model for HLA-DRB1*03:01 in SLE, where the association with anti-Ro and anti-La antibody-positive SLE is much stronger than SLE without these autoantibodies.

Genes Immunity 2014; 15: 210

Eitan Israeli

Capsule

Origins of tumor macrophages

To help the immune system fight cancer, it is important to understand the origins and functions of immune cells in tumors and the surrounding tissues. One type of immune cells, macrophages, is present both in tumors and in nearby non-cancerous tissue, but the relationship between these two cell populations is unclear. Franklin and team found that tumor-associated macrophages in mouse mammaries differed in form, function, and origin from macrophages found in nearby non-cancerous mammary tissue. Moreover, when they removed macrophages from the tumors but not the other mammary tissue, tumors shrank and cytotoxic T cells – another kind of immune cell that kills tumor cells – infiltrated the tumors. Tumor-associated macrophages may thus be an important therapeutic target.

Science 2014; 344: 921

Eitan Israeli

Capsule

Developmental pathway for potent V1V2-directed HIV-neutralizing antibodies

Antibodies capable of neutralizing HIV-1 often target variable regions 1 and 2 (V1V2) of the HIV-1 envelope, but the mechanism of their elicitation has been unclear. Doria-Rose and group have defined the developmental pathway by which such antibodies are generated and acquire the requisite molecular characteristics for neutralization. Twelve somatically related neutralizing antibodies (CAP256-VRC26.01–12) were isolated from donor CAP256 [from the Centre for the AIDS Programme of Research in South Africa (CAPRISA)]; each antibody contained the protruding tyrosine-sulphated, anionic antigen-binding loop [complementarity-determining region (CDR) H3] characteristic of this category of antibodies. Their unmutated ancestor emerged between weeks 30 and 38 post-infection with a 35-residue CDR H3, and neutralized the virus that superinfected this individual 15 weeks after initial infection. Improved neutralization breadth and potency occurred by week 59 with modest affinity maturation, and was preceded by extensive diversification of the virus population. Human immunodeficiency virus IV-1 V1V2-directed neutralizing antibodies can thus develop relatively rapidly through initial selection of B cells with a long CDR H3, and limited subsequent somatic hypermutation. These data provide important insights relevant to HIV-1 vaccine development.

Nature 2014; 509: 55

Eitan Israeli

Capsule

Trogocytosis by *Entamoeba histolytica* contributes to cell killing and tissue invasion

Entamoeba histolytica is the causative agent of amoebiasis, a potentially fatal diarrheal disease in the developing world. The parasite was named "histolytica" for its ability to destroy host tissues, which is probably driven by direct killing of human cells. The mechanism of human cell killing has been unclear, although the accepted model was that the parasites use secreted toxic effectors to kill cells before ingestion. Ralston and colleagues report the discovery that amoebae kill by ingesting distinct pieces of living human cells, resulting in intracellular calcium elevation and eventual cell death. After cell killing, amoebae detach and cease ingestion. Ingestion of human cell fragments is required for cell killing and also contributes to invasion of intestinal tissue. The internalization of fragments of living human cells is reminiscent of trogocytosis (from Greek trogo, nibble) observed between immune cells, but amoebic trogocytosis differs because it results in death. The ingestion of live cell material and the rejection of corpses illuminate a stark contrast to the established model of dead cell clearance in multicellular organisms. These findings change the model for tissue destruction in amoebiasis and suggest an ancient origin of trogocytosis as a form of intercellular exchange.

Nature 2014; 508: 526

Eitan Israeli

Capsule

Bacteria breach intestinal barriers

In an ironic complication of liver cirrhosis, beneficial microbes can escape from the gut and cause serious infections – or even death. Balmer et al. show that the blood vessels of the healthy liver form a barrier to runaway gut bacteria. However, in animal models of liver disease and gut dysfunction and in patients with non-alcoholic liver disease, the liver is unable to capture these escapees. The bacteria then leak into the blood system, activating immune responses that break down the mutualistic relationship between the gut microbes and the host. This type of breakdown is an important complication of liver disease.

Sci Transl Med 2014; 6: 237ra66

Eitan Israeli

Capsule

Breaking down the blood-brain barrier in stroke

Ischemic stroke, one of the most common causes of death and disability, occurs when a blood vessel supplying oxygen and nutrients to the brain becomes obstructed. Besides injuring brain cells, a stroke disrupts the function of endothelial cells in the blood-brain barrier (BBB), which exacerbates brain damage. The cellular mechanisms underlying BBB breakdown during a stroke are poorly understood. To study this, Knowland et al. created transgenic mice expressing a fluorescent reporter gene in endothelial cells and then, with the help of fluorescent dyes, used two-photon microscopy to image BBB function in the mice after an experimentally induced stroke. In contrast to a prevailing theory emphasizing the primary role played by a diffusion barrier called the tight junction, the imaging study revealed that the initial cause of BBB breakdown (occurring 6 hours after the stroke) was aberrant upregulation of transcytosis, a process by which molecules are transported across the endothelial cell. It was not until 24 to 48 hours after the stroke that tight junctions showed structural defects. Understanding this sequence of events may lead to therapies that limit the brain damage caused by a stroke.

Neuron 2014;10.1016/j.neuron.2014.03.003

Eitan Israeli

Capsule

Disrupted development in autism

Socioemotional difficulties and abnormal overgrowth of the brain are apparent early in childhood for those with autism. Although the brain overgrowth has resolved by adulthood, the difficulties remain. Stoner et al. analyzed the expression of a variety of genes that relate to the identification of neuron and glial subtypes, as well as a handful of genes linked to autism in postmortem samples of brains from unaffected children and children with autism. Multiple readouts were assembled computationally to reconstruct the three-dimensional pattern of gene expression. Samples from children with autism showed small patches, 5 to 7 mm in length, in which the expression of several genes was abnormally reduced. The expression of genes related to excitatory neurons was most affected in these patches, genes related to interneurons less so, and genes related to glia even less affected. No one subset of genes or specific locations characterized all the samples. Neurons were present, however, in patches of reduced gene expression. The diversity in locations of the disrupted patches may reflect the diversity in how autism affects children, so that, depending on where a disruption happened to land, different brain functions could be affected.

N Engl J Med 2014; 370: 1209

Eitan Israeli

Capsule

Plasmodium genetic loci linked to host cytokine and chemokine responses

Both host and parasite factors contribute to disease severity of malaria infection; however, the molecular mechanisms responsible for the disease and the host-parasite interactions involved remain largely unresolved. To investigate the effects of parasite factors on host immune responses and pathogenesis, Pattaradilokrat and team measured levels of plasma cytokines/chemokines (CCs) and growth rates in mice infected with two *Plasmodium yoelii* strains having different virulence phenotypes and in progeny from a genetic cross of the two parasites. Quantitative trait loci (QTL) analysis linked levels of many CCs, particularly IL-1β, IP-10, IFNγ, MCP-1 and MIG, and early parasite growth rate to loci on multiple parasite chromosomes, including chromosomes 7, 9, 10, 12 and 13. Comparison of the genome sequences spanning the mapped loci revealed various candidate genes. The loci on chromosomes 7 and 13 had significant ($P < 0.005$) additive effects on IL-1β, IL-5 and IP-10 responses, and the chromosome 9 and 12 loci had significant ($P = 0.017$) interaction. Infection of knockout mice showed critical roles of MCP-1 and IL-10 in parasitemia control and host mortality. These results provide important information for a better understanding of malaria pathogenesis and can be used to examine the role of these factors in human malaria infection.

Genes Immunity 2014; 15: 145

Eitan Israeli

Capsule

Interleukin-35 induces regulatory B cells that suppress autoimmune disease

Interleukin-10 (IL-10)-producing regulatory B (Breg) cells suppress autoimmune disease, and increased numbers of Breg cells prevent host defense against infection and promote tumor growth and metastasis by converting resting CD4+ T cells to regulatory T (Treg) cells. The mechanisms mediating the induction and development of Breg cells remain unclear. Wang and colleagues show that IL-35 induces Breg cells and promotes their conversion to a Breg subset that produces IL-35 as well as IL-10. Treatment of mice with IL-35 conferred protection from experimental autoimmune uveitis (EAU), and mice lacking IL-35 (p35 knockout (KO) mice) or defective in IL-35 signaling (IL-12Rβ2 KO mice) produced fewer Breg cells endogenously or after treatment with IL-35 and developed severe uveitis. Adoptive transfer of Breg cells induced by recombinant IL-35 suppressed EAU when transferred to mice with established disease, inhibiting pathogenic T helper type 17 (TH17) and TH1 cells while promoting Treg cell expansion. In B cells, IL-35 activates STAT1 and STAT3 through the IL-35 receptor comprising the IL-12Rβ2 and IL-27Rα subunits. As IL-35 also induced the conversion of human B cells into Breg cells, these findings suggest that IL-35 may be used to induce autologous Breg and IL-35+ Breg cells and treat autoimmune and inflammatory disease.

Nature Med 2014; 20: 633

Eitan Israeli

Capsule

Tumor endothelium FasL establishes a selective immune barrier promoting tolerance in tumors

Motz and co-authors describe a new mechanism regulating the tumor endothelial barrier and T cell infiltration into tumors. The authors detected selective expression of the death mediator Fas ligand (FasL, also called CD95L) in the vasculature of human and mouse solid tumors but not in normal vasculature. In these tumors, FasL expression was associated with scarce CD8+ infiltration and a predominance of FoxP3+ T regulatory (Treg) cells. Tumor-derived vascular endothelial growth factor A (VEGF-A), interleukin 10 (IL-10) and prostaglandin E2 (PGE2) cooperatively induced FasL expression in endothelial cells, which acquired the ability to kill effector CD8+ T cells but not Treg cells because of higher levels of c-FLIP expression in Treg cells. In mice, genetic or pharmacologic suppression of FasL produced a substantial increase in the influx of tumor-rejecting CD8+ over FoxP3+ T cells. Pharmacologic inhibition of VEGF and PGE2 produced a marked increase in the influx of tumor-rejecting CD8+ over FoxP3+ T cells that was dependent on attenuation of FasL expression and led to CD8-dependent tumor growth suppression. Thus, tumor paracrine mechanisms establish a tumor endothelial death barrier, which has a critical role in establishing immune tolerance and determining the fate of tumors.

Nature Med 2014; 20: 607

Eitan Israeli

Capsule

Cell competition is a tumor suppressor mechanism in the thymus

Cell competition is an emerging principle underlying selection for cellular fitness during development and disease. Competition may be relevant for cancer, but an experimental link between defects in competition and tumorigenesis is elusive. In the thymus, T lymphocytes develop from precursors that are constantly replaced by bone marrow-derived progenitors. Martins et al. show that in mice this turnover is regulated by natural cell competition between 'young' bone marrow-derived and 'old' thymus-resident progenitors that, although genetically identical, execute differential gene expression programs. Disruption of cell competition leads to progenitor self-renewal, up-regulation of Hmga1, transformation, and T cell acute lymphoblastic leukemia (T-ALL) resembling the human disease in pathology, genomic lesions, leukemia-associated transcripts, and activating mutations in Notch1. Hence, cell competition is a tumor suppressor mechanism in the thymus. Failure to select fit progenitors through cell competition may explain leukemia in X-linked severe combined immune deficiency patients who showed thymus-autonomous T cell development after therapy with gene-corrected autologous progenitors.

Nature 2014; 509: 465

Eitan Israeli

Capsule

The pathology of severe schistosomiasis

Schistosomiasis-causing blood flukes infect hundreds of millions of people in tropical regions, but the occurrence of pathology is highly variable, with 5 to 10% of infections becoming severe. Likewise, schistosome infections take very different courses in different strains of mice, a phenomenon that relates to their relative ability to generate lymphocytes classified as CD4+ T helper 17 (TH17) cells. Sick children with blood flukes have also been found to have higher percentages of CD4+ TH17 cells. Ponichtera and colleagues have now discovered that the antigen-presenting dendritic cells of a mouse strain that develops severe hepatic granulomatous responses to schistosome eggs have a many times greater expression of a C-type lectin receptor called CD209a (a homolog of human ICAM-3-grabbing nonintegrin) on their cell surfaces as compared with a mouse strain that shows little pathology. CD209a is essential for the induction of the cytokines interleukin-1β and interleukin-23 that stimulate CD4+ TH17 cell development. Possibly the pathology of severe schistosomiasis is caused by elevated CD209a levels in some people sensitizing recognition of the fucose-rich glycans that coat the parasites' egg surface.

J Immunol 2014; 10.4049/ *jimmunol*.1400121

Eitan Israeli

Capsule

Protective mucosal immunity mediated by epithelial CD1d and IL-10

The mechanisms by which mucosal homeostasis is maintained are of central importance to inflammatory bowel disease. Critical to these processes is the intestinal epithelial cell (IEC), which regulates immune responses at the interface between the commensal microbiota and the host. CD1d presents self and microbial lipid antigens to natural killer T (NKT) cells, which are involved in the pathogenesis of colitis in animal models and human inflammatory bowel disease. As CD1d crosslinking on model IECs results in the production of the important regulatory cytokine interleukin (IL)-10, decreased epithelial CD1d expression – as observed in inflammatory bowel disease – may contribute substantially to intestinal inflammation. Olszak et al. show in mice that whereas bone marrow-derived CD1d signals contribute to NK-cell-mediated intestinal inflammation, engagement of epithelial CD1d elicits protective effects through the activation of STAT3 and STAT3-dependent transcription of IL-10, heat shock protein 110 (HSP110, also known as HSP105), and CD1d itself. All of these epithelial elements are critically involved in controlling CD1d-mediated intestinal inflammation. This is demonstrated by severe NKT cell-mediated colitis upon IEC-specific deletion of IL-10, CD1d, and its critical regulator microsomal triglyceride transfer protein (MTP), as well as deletion of HSP110 in the radio-resistant compartment. Our studies thus uncover a novel pathway of IEC-dependent regulation of mucosal homeostasis and highlight a critical role of IL-10 in the intestinal epithelium, with broad implications for diseases such as inflammatory bowel disease.

Nature 2014; 509: 497

Eitan Israeli