Case Summaries

Venue: Larwill Hotel, Melbourne (accommodation and meeting rooms)
Dates: Sunday 24 July to Thursday 28th July 2016
International faculty: Dr Graham Davies, Dr Andrew Gennery.
Local faculty: Dr Andrew McLean-Tooke, Dr Joanne Smart, Dr Melanie Wong, Dr Theresa Cole, Prof Stuart Tangye. Guest speaker: Dr Sharon Choo

Delegates (22):
Dr Anthea Anantharajah, Dr Abigail Cheung, Dr Joseph DeLuca, Dr Luke Droney, Dr Jocelyn Xin Jiang, Dr Narinder Kaur, Dr Elizabeth Klinken, Dr Jessie Lee, Dr Monique Lee, Dr Paxton Loke, Dr Leigh Mackey, Dr Paul Mansfield, Dr Birgit Marchand, Dr Melissa Norman, Dr Alberto Pinzon, Dr Phillipa Pucar, Dr Stephanie Richards, Dr Sarah Sasson, Dr Sabeena Selvarajah, Dr Bella Shadur, Dr Mark Taylor, Dr Grace Thompson.

Support: Unrestricted educational grants provided by CSL Behring, Grifols and Shire

Contents:
- Anthea Anantharajah - Adult-onset spastic paraparesis in VODI with immunodeficiency
- Abigail Cheung - A surviving tale of Schimke Immuno-osseous dysplasia (SIOD)
- Joseph De Luca - CTLA-4 Deficiency presenting as a PID
- Luke Droney – Adult onset CGD
- Jocelyn Xin Jiang - Immunodeficiency and autoimmunity
- Narinder Kaur - UNClear to diagnosis and BMT (FHL)
- Elizabeth Klinken – A likely case of GATA-2 deficiency
- Jessie Lee - AD ectodermal dysplasia and immunodeficiency in a mother and daughter with a NFKBIA mutation
- Monique Lee - The X factor in a blue moon
- Paxton Loke – Relapsing-remitting granulomatous disease in 22q11 deletion syndrome
- Leigh Mackey – Looking for the shoe that fits
- Paul Mansfield – Hyper IgE Syndrome (HIES)
- Birgit Marchand - Long-term piece of mind
- Melissa Norman - LRBA Deficiency – getting to the gut of the problem
- Alberto Pinzon – Roifman syndrome
- Phillipa Pucar - A case of monogenic immune deficiency and autoimmunity
- Stephanie Richards - Reticular dysgenesis – SCID
- Sarah Sasson - Rapamycin reduces massive splenomegaly and lymphadenopathy in CVID
- Sabeena Selvarajah – A term infant girl
- Bella Shadur - A Case of TTC7A Deficiency
- Mark Taylor – CVID-like disease in two siblings of consanguineous parentage
- Grace Thompson – IPEX syndrome – not just a paediatric condition
APID 2016 CASE SUMMARY: ADULT ONSET SPASTIC PARAPARESIS IN VENO-OCCCLUSIVE DISEASE (VODI) WITH IMMUNODEFICIENCY

Presented by Dr Anthea Anantharajah

Case Summary:

- Female born to non-consanguinous parents diagnosed with VODI at 9 months after presenting with PR bleeding and hepatomegaly
- Two affected siblings (both now deceased), one healthy sibling
- Remained well whilst in paediatric care apart from mild bronchiectasis and hypogammaglobulinaemia – managed with Bactrim and intravenous immunoglobulin
- Present at the age of 18 with bilateral leg weakness (spastic paraparesis) and double incontinence
- MRI of the brain and spinal cord revealed a communicating hydrocephalus and multiple hypointense T1 foci in the perivascular spaces, some of which were enhancing, in addition to enhancement of the entire spinal cord, more marked in the medulla and cervical spine. The exact aetiology of the lesions was unclear, possible infection with Toxoplasma or Cryptococcus was considered and the patient received empiric treatment
- CSF demonstrated an elevated WCC with a predominance of monocytes with normal protein and glucose
- Serology and nucleic acid testing excluded JC virus, HSV, enterovirus, mycobacteria, HIV, hepatitis A, B and C, toxoplasmosis, syphilis, Epstein-Barr virus, human T-lymphotropic virus 1, coccidiomycosis, histoplasmosis, cryptococcosis, schistosomiasis and hydatid disease.
- Underwent brain biopsy which demonstrated no evidence of viral inclusion bodies, malignancy, vasculitis or demyelination, prolonged culture and extensive NAT was negative
- No definitive diagnosis was found but patient improved over an 18 month period with intensive rehabilitation

Discussion:

Veno-occlusive disease with immunodeficiency (VODI) is an autosomal recessive combined immunodeficiency that results from mutations in the gene encoding PML nuclear body protein (SP110)(1). The function of SP110 remains unclear although it may influence apoptosis, gene regulation, antiviral responses, proteolysis, DNA repair, and tumor suppression. Similarly, the mechanism by which mutations in SP100 result in the VODI phenotype remains unknown. This condition was first described in Australian Lebanese patients and further cases have been reported in patients of Hispanic, Arabic and Italian ancestry (approximately 38 cases in total). Clinically, patients present in the first year of life with complications from hepatic veno-occlusive disease or infections resulting from T cell dysfunction or antibody deficiency. The Australian cohort spans about 30 years and few patients have survived to adulthood; therefore this case provides unique insights into the complications and presentations that may occur in adulthood. Although a final diagnosis was not made in this case, it seems likely that this patient developed leukodystrophy, which has been described in a number of VODI patients. Treatment strategies have included immunoglobulin replacement, which may also prevent the development of hepatic veno-occlusive disease and prophylactic antibodies. There is limited data on haematopoietic transplantation – an Israeli study reported cure in 3/5 patients (2).
References


APID2016 CASE SUMMARY: SCHIMKE-IMMUNO-OSSEOUS DYSPLASIA (SIOD)

Presented by Dr Abigail Cheung

We describe a 4 year old boy, of consanguineous parents, with Schimke Immuno-osseous Dysplasia. At the age of 2 years, he was found to have a deletion of 2q35 with deletion of exons 13 and 14 of SMARCAL-1 gene on microarray after being investigated for facial dysmorphism, solitary right kidney with nephrotic range proteinuria and hypertension, bicuspid aortic valve & mitral stenosis, skeletal dysplasia with short stature and developmental delay. He also had a history of prematurity (ex-25 weeks gestation) with intra-uterine growth restriction.

He had an older brother, born at 26 weeks gestation, who died at 5 months of age with unclear cause, and an older sister who was well. At presentation to immunology, he had a mild infection history with approximately 2 upper respiratory tract infections and 1 gastroenteritis episode per year. He had tolerated his vaccines including live vaccines.

Immune investigations showed T cell lymphopenia with reduced naïve T-cells and reduced anti-CD3 proliferation. Immunoglobulin levels were normal with poor pneumovax response. He had progressive decline in T cell function with increasing frequency of infections. He was commenced on immunoglobulin replacement with marked reduction in frequency of infections.

More recently, he presented with acute kidney injury. While some initial improvement with conservative management, his renal function continued to decline until he passed away with complications of renal failure.

Schimke Immuno-osseous dysplasia (SIOD) is an autosomal recessive condition, caused by a mutation in the gene encoding SMARCAL1, with over 40 different known mutations. The prevalence is unknown, estimated at 1:1,000,000. The SMARCAL-1 protein encodes a chromatin remodelling enzyme which participates in gene regulation. The genotype does not predict disease severity or outcome with reports of intra-familial variation.

There is a wide spectrum of clinical manifestations but is characterized by spondyloepiphyseal dysplasia with short stature, nephropathy and T cell deficiency. Almost all affected individuals have proteinuria with the majority (86%) with progressive steroid-resistant nephropathy, which generally evolves into end-stage renal disease. Patients with SIOD may have neurological disease, most commonly migraines and cerebral ischaemic events.

Approximately 80% of affected individuals have a T-cell deficiency with reduction in both CD4 and CD8 T cells. T cells are predominantly of a memory surface phenotype, rather than a naïve phenotype. Analysis of thymic capacities in individuals with SIOD has shown that those affected have reduced TREC and T-cell receptor rearrangements. SIOD has also been associated with a lack of interleukin 7 receptor alpha expression on the T cells of patients with SIOD. These findings indicate that there is an underlying defect in thymus histology with reduction in thymic output and activity, but also defective T cell development.

B cell numbers are usually normal to slightly elevated with normal IgG in 70% of patients. About 20% of individuals with SIOD have features of autoimmune disease including immune thrombocytopenia, haemolytic anaemia, enteropathy, pericarditis and Evans syndrome.
There is large variation in severity of SIOD, ranging from in utero growth restriction and early death to a slowly progressive course with survival into adulthood. The mean age of death is 11 years, with the most common causes of death including infection (23%), stroke (13%), pulmonary hypertension and congestive cardiac failure (13%) and renal failure (11%). Treatment is largely supportive.

There have been five reports of haematopoietic stem cell transplant in SIOD, one of which survived, at last report, to the age of 21 years. This patient developed renal deterioration post HSCT, and received subsequent renal transplantation from the same donor, now with good bone marrow and renal function. The other four patients died of complications including sepsis, pulmonary oedema and cardiac tamponade, pulmonary embolism and cerebral ischaemia and haemorrhage.

SIOD is a multi-system disorder with a wide variable phenotype. Unfortunately, it is associated with a poor prognosis with limited treatment options. While HSCT, with or without renal transplantation, may be an option of treatment, there is a reported high mortality rate.

References:

APID2016 CASE SUMMARY: CTLA-4 DEFICIENCY PRESENTING AS A PID

Presented by Dr Joseph De Luca

Case summary
19yo male who was transferred to our care from the local Children’s Hospital. He was initially referred to the Immunology Department at the children’s hospital at the age of 16yo with a chronic moist cough, persistent lymphopenia and panhypogammaglobulinemia. (1gG 1.43g/L, IgA <0.05g/L, IgM 0.24 g/L). He had no other symptoms and was noted to have some mild splenomegaly on examination. Both parents are alive, he has one well brother with no significant past medical history and there is no family history of immunodeficiency. Our patient had a significant history of autoimmune haemolytic anaemia that was not responsive to steroids or IVIG, so he was treated with Rituximab with good response.

On testing organised by the Immunology department at the children's hospital, our patient was found to have normal numbers of T cells, B cells and NK cells but reduced switched and marginal zone memory B cells with reduced naïve CD4 and CD8 T cells. He had evidence of poor specific antibody production to polysaccharide vaccines, with seroconversion to 0/15 Pneumovax serotypes. He was promptly started on immunoglobulin replacement. On further investigation of his cough, he was found to have innumerable solid and ground glass nodules in both lungs on CT Chest, with a restrictive pattern on RFTs. Lung biopsy of one of these nodules revealed changes consistent with Lymphocytic Interstitial Pneumonitis (LIP), with focal infiltration of mostly CD3+ Lymphocytes. He was started on Sirolimus as management of the LIP.

He then went on to have Genetic testing, and on SNP microarray was found to have a deletion in chromosome region 2q33.2-2q33.3 in the region of the ICOS and CTLA4 genes. Further sequencing of his second ICOS gene was found to be normal, which means his presentation and genetic findings were consistent with a CTLA4 Haploinsufficiency. In January of 2016, he was admitted to our hospital with a right lower quadrant visual field defect in his right eye with normal visual acuity and evidence of papilloedema. MRI showed changed consistent with an inflammatory pseudotumour involving his right medial rectus and optic nerve. He was pulsed with IV Methylprednisolone and his Sirolimus dose was uptitrated.

CTLA4 Haploinsufficiency
Cytotoxic T-lymphocyte Antigen 4 (CTLA-4) is the high affinity receptor for CD80/86 ligand and acts as a ‘costimulator’ with a critical inhibitory function. It is constitutively expressed by CD4+FOXP3+ Regulatory T cells (TReg), but becomes dramatically upregulated following activation. CTLA-4 is also expressed on regular CD4+ T cells following activation.

There have been 2 major papers published on patients presenting with primary immunodeficiency (PID) in which CTLA4 Haploinsufficiency is the underlying genetic defect leading to their phenotype. Kuehn et al 2014 described 4 unrelated families with unique mutations and 6 affected individuals between them, whilst Shubert et al 2014 described 6 families with unique mutations and 14 affected individuals between them. There was evidence of incomplete penetrance with unaffected members of the family carrying the same mutation. The clinical phenotype amongst the affected individuals were similar, with hypogammaglobulinemia and autoimmune manifestations like enteropathy, autoimmune haemolytic anaemia, splenomegaly and lymphocytic tissue infiltration (Lung, Bone Marrow, Liver, Brain). The immune phenotype of these individuals were also similar between the two papers, with reduced naïve T cells, reduced switched memory B cells, evidence of increased lymphocyte exhaustion and persistent immune activation. Kuehn et al was able to demonstrate
reduced CTLA-4 expression and mRNA production in affected patients compared to healthy controls.

**Treatment options**
The treatment of patients with **CTLA4** haploinsufficiency to date has been replacement of immunoglobulin to mitigate their infection risk, as well as the use of systemic immunosuppression to manage their autoimmune manifestations. As a lack of CTLA-4 is the underlying pathological cause of disease in patients with **CTLA4** haploinsufficiency, it was postulated that replacement of CTLA4 in these patients would reverse the effects of their underlying mutation. Kuehn et al showed that in vitro addition of CTLA4-Ig to CD4+ and CD8+ T cells lead to reduced activation.

Abatacept is a commercially produced CTLA4-Ig that has historically been used for the treatment of rheumatoid arthritis. Unfortunately, there have been no trials examining the utility of Abatacept in the treating individuals with **CTLA4** Haploinsufficiency. There has been a paper published, however, on the use of Abatacept in vivo in patients with LRBA deficiency, a disease that presents like **CTLA4** Haploinsufficiency as LRBA is a critical control point for lysosomal turnover of CTLA-4. In a paper by Lo et al 2015, they described the treatment of 3 individuals with LRBA Deficiency with Abatacept. They were revealed to have prompt resolution of their autoimmune and tissue infiltration manifestations of their disease after the introduction of Abatacept, as well as an improvement in their FEV1 and DLCO.

**Summary:**
- CTLA-4 has a critical inhibitory function in the immune system
- Patients with **CTLA4** Haploinsufficiency display features of severe immune dysregulation, with a combination of immunodeficiency and autoimmune tissue infiltration and destruction
- This is consistent with their immunophenotype with reduced naïve T cells, switched memory B cells and features of immune hyperproliferation
- Treatment options include systemic immunosuppression, directed therapy with CTLA4-Ig (Abatacept)

**References**
APID2016 CASE SUMMARY: ADULT ONSET CGD

Presented by Dr Luke Droney

Case: A mosaic of suspicious infections

A 66 year-old gentleman presented with *Serratia marcescens* urosepsis on a background of distant *Burkholderia cepacia* urosepsis and disseminated *Nocardia transvalensis* infection. There was no family history suggestive of primary immune deficiency.

Neutrophil oxidative burst testing (by DHR-flow cytometric method) was significantly abnormal and, on first impressions, consistent with X-linked CGD. However, a small (6%) population of neutrophils appeared to burst normally. Genetic sequencing of the CYBB gene identified a novel missense mutation that was predicted to result in a functionally deficient protein. The patient was treated as having Chronic Granulomatous disease (CGD) with prophylactic antibiotic therapy and interferon gamma.

Adult-onset CGD is seldom described in the literature. A PubMed literature search revealed six published cases. Of these six, 4 patients presented in their 20s and 2 presented in their 60s. All cases involved mutations of the CYBB gene, in particular splice sites. 2 patients were females with evidence of skewed lyonisation.

The patient’s daughter came to the attention of our clinical team due to concerns of propagation of the X-linked CYBB mutation. However, the patient’s daughter had a normal DHR-NOB and did not have the CYBB mutation identified in her father. The patient’s daughter was quite confident of paternity and this was confirmed by further formal genetic analysis.

The absence of the mutation in the patient’s daughter and his attenuated phenotype with a residual population of functionally normal neutrophils led to the conclusion that the patient demonstrated somatic mosaicism at the CYBB locus.

Mosaicism is defined as the presence of two or more cellular populations with distinct genotypes in one individual. Mosaicism is more common in males and with increasing age. The genetic size of mosaicism may be variable and thus, in some instances, it may not be detected by low resolution cytogenetic/genetic analysis (i.e karyotyping, microarrays). Unsurprisingly, mosaicism is associated with malignancy, particularly haematologic.

Mosaicism has been described in one case of adult-onset CGD (Wolach et al, 2005). This female patient began to develop typical infections at the age of 66. Analysis revealed a mosaic CYBB mutation and skewing of X-inactivation toward the CYBB-mutant neutrophil population.

Our patient remained well on prophylactic therapies until approximately 2 years after his diagnosis. He was diagnosed with minimally-differentiated Acute Myeloid Leukaemia and passed away soon after. Whilst there is no established relationship between CGD and acute myeloid leukaemia, as mentioned above there is an association with mosaicism.

This case demonstrates that CGD should be considered as a diagnosis in adult patients who present with infections with organisms that are highly associated with this primary immune deficiency. Furthermore, the demonstration of CYBB mosaicism raises the possibility that mosaicism at other loci may be an important cause of adult-onset immune deficiency.

Reference:

APID2016 CASE SUMMARY: AUTOIMMUNITY AND IMMUNODEFICIENCY

Presented by Dr Jocelyn Jiang

Case 1
- 42 year old woman referred for lymphopenia (incidental finding on routine bloods)
  - No history of recurrent infections, examination essentially normal
  - No recent vaccinations; no history of blood transfusions; Never immunosuppressed
- Other investigations
  - CD4 and CD8 lymphopenia; CD19 B cells in normal limits but lacking memory B cells
  - Normal serum electrophoresis, serum free light chains and complement levels
  - Serum immunoglobulins within normal limits and preserved vaccination responses
- Bilateral hilar and mediastinal lymphadenopathy on CT chest; ACE 45 U/L
  - Biopsy confirmed sarcoidosis;
- Subtle increase in lymphocyte count after commencement of steroids and Plaquenil

Case 2
- Severe autoimmune haemolytic anaemia and hypogammaglobulinaemia with low specific antibody responses
- No history of severe infection but has had monthly upper respiratory tract infection and occasional sinus infection. Constant sore throat/cough
  - CVID panel: Friburg Ia; MB0, Euroclas: B=smB-CD210lo/Tr-lo
- Commenced on IVIg for CVID: Symptoms of constant URTI completely resolved
- Recurrent thrombocytopenia
  - failed steroids, cyclophosphamide, rituximab, splenectomy, romiplostim
- Good initial response to eltrombopag but recent relapse

Discussion: Immune dysregulation
- The association between immunodeficiency and autoimmunity is well described in the literature: failure to respond to non-self pathogens while reacting to self
- Central tolerance: deletion of self reactive T-cells in the thymus e.g. AIRE mutation, DiGeroge syndrome with mild-mod lymphopenia
- Peripheral tolerance: self reactive T cells that escape central tolerance check points remain unresponsive in peripheral organs E.g. IPEX, CD25 deficiency, CTLA4 deficiency
- Other mechanisms include:
  - Lymphopenia and impaired natural selection process for lymphocytes (RAG def)
  - Failure of apoptosis (ALPS: FAS-deficiency)
  - STAT1 GOF/STAT3 GOF mutations
  - Hyper activation of lymphocytes:
  - Breakdown of B cell tolerance
- In CVID
  - reduced numbers of switched memory B cells and an increased proportion of CD21lo cells have increased proportion of splenomegaly and autoimmunity
  - role of T cells ?? - Increased naïve T cellsDecreased Tregs
  - abnormalities of cytokines
- Lessons from systemic autoimmunity
  - Considerable overlap between genes associated with increased rheumatoid arthritis risk and PID
    - SLE associated with failure to clear apoptotic debris, issues with Interferon-alpha pathways
Familial Haemophagocytic Lymphohistiocytosis (FHL) is a rare multisystem congenital disorder characterized by excessive immune activation causing inflammation and tissue damage. Diagnosis is challenging especially in a neonate because it mimics severe sepsis by demonstrating fevers, hepatosplenomegaly, coagulopathy and cytopenias. The hallmark of haemophagocytic lymphohistiocytosis (HLH) is impaired or absent function of natural killer (NK) cells. Five genetic subtypes (FHL1, FHL2, FHL3, FHL4 and FHL5) are described. PRF1, UNC13D and STX11 gene defects contribute to 40-50% of primary cases.

Our patient was born prematurely at 35 weeks gestation and presented at birth with respiratory distress requiring mechanical ventilation; hydrops with ascites, pericardial and pleural effusions; coagulopathy; conjugated hyperbilirubinemia with liver dysfunction; fevers; sepsis and HLH (high ferritin 8649 µg/L, low fibrinogen 0.4 g/L and high triglycerides 3.4 mmol/L). NK cell chromium release cytotoxicity and degranulation flow cytometry was absent. Treatment with Prednisone, Cyclosporin, Immunoglobulins and Etopside was commenced. The immunogenetic testing showed UNC13D c.118-308C>T mutation which abrogates transcription of the protein MUNC13-4 in lymphocytes consistent with FHL 3 diagnosis. He had multiple intensive care admissions with respiratory distress and sepsis requiring ventilation and intravenous antibiotics and persistent hypertension. The family history was positive, with demise of one of his siblings at 3 months of age with “cold” and bleeding. Our patient received a Haplo-identical Bone Marrow Transplant from his non-carrier sister at 6 months of age. The post-transplant period was complicated by transplant related issues, hypertension, developmental delay and possibility of another genetic condition.

Recent clinical observations of increased recognition of HLH in older children and adults, sometimes in association with classic disease-associated mutations, is challenging the traditional view of HLH as either a distinctly familial or secondary disorder. FHL is fatal with a median survival of 2 months if left untreated. Chemo immunotherapy followed by hematopoietic stem cell transplantation (HSCT) improves disease outcome. Survival is better with matched donor, controlled disease and reduced intensity chemotherapy before HSCT.
APID2016 CASE SUMMARY: A LIKELY CASE OF GATA-2 DEFICIENCY

Presented by Dr Elizabeth Klinken

We describe the case of a woman who first presented at 28 years of age with a granulomatous panuveitis. Her history was significant for recurrent bacterial infections from the age of 16 years. Infections have included abscesses, severe lower limb cellulitis, a single episode of septic arthritis, a number of hospital admissions for pneumonia and a Listeria monocytogenes brain abscess. Staphylococcus aureus and Streptococcus pyogenes have commonly been isolated. Further history was significant for extensive human papilloma virus (HPV) infection from 25 years of age and a recent diagnosis (at 44 years of age) of a multi-focal invasive squamous cell carcinoma (SCC). She has also had recurrent cutaneous SCCs and basal cell carcinomas. Our patient was well as a young child, has no family history of immunodeficiency and has three well children from three pregnancies. Investigations have shown a persistent mild lymphopaenia with undetectable monocytes.

Lymphocyte immunophenotyping has revealed low normal T cell subsets, a mild NK lymphopaenia and undetectable B cells. Lymphocyte proliferation to ConA and PHA is impaired as is NK cell cytotoxicity. The immunoglobulin profile shows a markedly elevated total IgG, with a normal IgM and IgA and undetectable IgE. A persistent, polyclonal elevation of IgG1 (up to 47.5 g/L), with a normal IgG3, low IgG2 and undetectable IgG4 has been demonstrated. This B cell differentiation defect is accompanied by a non-sustained IgG2 response to pneumococcal antibodies and a low MBL. In the context of this, intravenous immunoglobulin (IVIg) replacement therapy was commenced with a significant reduction in infection frequency. A number of diagnoses have been considered over the years, including GATA2 deficiency; however, whole exome sequencing did not identify a putative disease-associated mutation. WHIM syndrome was also considered but sequencing of CXCR4 was normal. In the context of the recent invasive SCC, a staging PET scan demonstrated FDG-avid abdominal and hilar lymph nodes, biopsy positive for Mycobacterium avium. The diagnosis of GATA2 deficiency was re-visited and a heterozygous intronic variant GATA2:c.1017-572C>T was identified. This case highlights the importance of reconsidering diagnoses and being aware of developments in genetics. Relevant disease-associated mutations may be missed on whole exome sequencing alone.

GATA2 is a haematopoetic transcriptional regulator that plays a key role in the maintenance and survival of the haematopoetic stem cell pool. It is also important in the formation of early blood and lymphatic vessels. A deficiency of GATA2 has been associated with MonoMAC (characterised by monocytopenia and mycobacterial infections), DCML deficiency (DC, monocyte, B and NK cell deficiency), familial myelodysplastic syndromes (MDS) / acute myeloid leukaemias (AML), Emberger syndrome, type I classical NK cell deficiency and some paediatric neutropaenia and aplastic anaemia. The immunological phenotype evolves over time, with an extended age of onset from 5 to 55 years. Significantly, it is estimated that up to 90% of individuals with a GATA2 mutation will develop a myelodysplastic syndrome by 60 years of age. Other clinical manifestations include recurrent viral and disseminated mycobacterial infections, pulmonary alveolar proteinosis, lymphoedema, deep vein thrombosis, miscarriage and sensorineural hearing loss. Almost one hundred gene defects have been described, including two discrete mutations in the intron 5 enhancer of the GATA2 gene. Management options include prophylactic anti-microbials, vaccination against HPV and IVIg as appropriate. Ultimately, haematopoetic stem cell transplantation (HSCT) is the only effective definitive therapy. The majority of transplant data comes from patients with MDS/AML; however, there are increasing reports of patients being successfully transplanted earlier in the disease course, prior to the development of haematological malignancy. Given the nature of the defect, a myeloablative conditioning regime is likely to be required for optimal engraftment and reduced risk of relapse. HSCT is currently being considered in our patient.
Mrs MD is a 73 year old lady who presented to the emergency department with increasing
dyspnoea and hypoxia. In the preceding five weeks, she had recurrent lower respiratory tract
infections, for which she had received multiple courses of oral and parenteral azithromycin,
moxifloxacin and ciprofloxacin. Following negative blood and sputum cultures and a negative
bronchoalveolar lavage, she was also commenced on prednisone 25mg daily. On admission, chest
imaging demonstrated right middle lobe consolidation with small bilateral pleural effusions, and
blood tests demonstrated elevated inflammatory markers with leucocytosis of 13 x 10^9/L and raised
CRP of 125.2mg/mL. Blood cultures isolated *Burkholderia cepacia* complex for which she was
commenced on tobramycin, ceftazidime and Bactrim. Voriconazole was also commenced as a
fungal process was not excluded.

Despite treatment, she deteriorated, with the development of fevers, rash, hypotension and acute
respiratory failure. She developed new cytopenias with Hb 84g/L, platelets 86 x 10^9/L, and
increasingly deranged liver function with ALP 345U/L, GGT 776U/L, ALT 126U/L and AST 197U/L.
She had also developed a hypertriglyceridaemia of 2.3mmol/L, with hyperferritinaemia of 10
93ug/L and elevated CRP of 299.7mg/L. She was admitted to the intensive care unit and
administered vasopressors and high flow nasal prong oxygen. A bone marrow biopsy demonstrated
hypercellular marrow with prominent haemophagocytosis, and a diagnosis of macrophage activation
syndrome was made, likely secondary to *B. cepacia* infection. She was treated with three doses of
pulse methylprednisolone 1g, followed by prednisone 50mg daily with a prolonged taper. She was
also given IVIG induction at 2g/kg and cyclosporine 1.5mg/kg. Filgrastim and blood transfusions
were required to support the neutropenia and anaemia respectively. She responded well, and was
discharged home on cyclosporin, prophylactic Bactrim, itraconazole and weaning prednisone.

Mrs MD is a known X-linked carrier for chronic granulomatous disease, diagnosed at age 33 years
following the deaths of two sons at 9 and 14 months, from overwhelming pyogenic sepsis. Genetic
studies demonstrated c. 764hetdelA; p.Lys255Argfs*14 on *CYBB*, with an arginine deletion resulting
in a frameshift mutation and stop codon 13 positions downstream. She had had several previous
mild lower respiratory tract infections, persistent mediastinal lymphadenopathy and deranged liver
function tests for which a biopsy demonstrated non-caseating granuloma. NBT/DHR demonstrated
extreme lyonisation with only 2% neutrophils with normal respiratory burst after PMA stimulation.
She had been reviewed by our service but refused prophylactic Bactrim. Her other background
include polymyalgia rheumatic, hyperparathyroidism and previous biliary sepsis.

This is an interesting case of extreme lyonisation in an elderly X-linked CGD carrier, with resultant
pathologic phenotype, together with macrophage activation syndrome secondary to *B. cepacia*.
CGD occurs at an incidence of 1/ 250,000, with the majority of cases being X-linked, resulting from
a wide range of mutations in gp91 phox on the *CYBB* gene. X-linked carriers are known to have
immune dysregulation, with higher risk of autoimmunity and granulomatous organ involvement, as
in this case. Increasing lyonisation occurs with age, and X-linked CGD carriers manifest typical
infections when less than 5% of neutrophils are capable of respiratory burst. The level of superoxide
production is one of the steady determinants of mortality and residual NADPH oxidase activity leads
to attenuated disease and better prognosis.

*B. cepacia* complex is a motile gram negative bacillus that is involved in 3-7% of pneumonia and
18% of deaths in CGD. It has been shown to interfere with phagosome maturation by reducing
NADPH oxidase activity, in order to enable survival in phagocyte vacuoles. There have been
several case reports of macrophage activation syndrome in CGD patients with most of these
associated with infections including *B. cepacia, leishmania, stenotrophomonas, staphylococcus,
enterobacter, HHV6, BCG and pseudomonas*. Thus, CGD results in a state of hyperactive
inflammation exacerbated by impaired pathogen killing due to defective NADPH activity, which is
further impeded by *B. cepacia*. NADPH deficiency has also been shown to favour production of TH1 cytokines in a murine model, further inducing macrophage activation, leading to systemic hyperinflammation and multiorgan damage. Treatment options include IVIG, corticosteroids, cyclosporin and G-CSF together with targeted and prophylactic antimicrobials. Haematopoietic stem cell transplant is potentially curative. IVIG and pathogen-specific therapy has resulted in good outcomes in one series, with mortalities resulting from inadequate pathogen specific therapy and immunosuppressive therapy.
APID 2016 CASE SUMMARY:
RELAPSING-REMITTING GRANULOMATOUS DISEASE IN 22Q11 DELETION SYNDROME

Presented by Dr Paxton Loke

Summary:
The case involves a 13 year old girl with 22q11 deletion syndrome (DiGeorge syndrome) who was diagnosed in the context of developmental delay by FISH at the age of 3, and subsequently confirmed by microarray 8 years later. At the age of 8, she was referred for an immunological opinion due to a long standing history of sino-pulmonary infections and bronchiectasis. Investigations revealed a mild CD8 T cell lymphopenia, markedly reduced naïve T cells, reduced lymphocyte proliferation to PHA and anti-CD3, panhypogammaglobulinaemia, reduced memory B cells and poor vaccine responses (to both polysaccharide and protein antigens). She was diagnosed with concurrent antibody deficiency, and was started on immunoglobulin replacement therapy.

Four years ago in 2013, she presented with recurrent fevers, splenomegaly and cytopenias, and investigations revealed granulomatous disease involving her lungs, lymph nodes and retroperitoneal regions. She was initially treated with steroids with improvement but relapse the following year. Following that, rituximab and mycophenolate were used as combination therapy but she relapsed a second time when her B cells returned in 2015. She was again treated with steroids, rituximab and mycophenolate to ensure control of her disease. In 2016, she represented with symptoms suggestive of inflammatory meningitis (no organisms found on extensive infective screens) with cerebral vasculitis, and was treated with steroids and commenced on sirolimus.

Discussion:
This case presents an overview of the different immunological phenotypes of DiGeorge syndrome (DGS) which encompasses complete DGS, atypical complete DGS, and partial DGS (with or without autoimmunity) and highlights the difficulty of treatment in the subgroup of partial DGS with both autoimmunity and granulomatous disease. A single study has shown that it is possible that partial DGS with higher naïve T cells may be protective against immune dysregulation leading to autoimmunity. Treatment options for both autoimmunity and granulomatous disease inferred from the literature were mainly from experience with patients with CVID and ALPS. There are also limited case reports from the literature of patients with both DGS and granulomatous disease (6 in total) where morbidity and mortality is high.

Learning points include the need to continue searching for a second mutation in addition to the patient’s 22q11 deletion (as one of the above patients in the case reports was discovered to have another gene mutation) and managing the patient’s symptoms and the rest of the patient’s co-morbidities to ensure a good quality of life.
APID2016 CASE SUMMARY: LOOKING FOR THE SHOE THAT FITS

Presented by Dr Leigh Mackey

I presented a case of an undifferentiated PID presenting in a young infant with multiple congenital abnormalities. Although the diagnosis remains unknown a treatment regime (SCIG) was put in place that has kept him well.

During the case presentation I was stopped several times and asked questions around further information that might help lead to a clue in diagnosis. Everyone feels more comfortable when the diagnosis is known, however everyone manages patients in whom the exact nature of the condition remains unknown. Clinical assessment and reassessment of progress is required in these patients. Some patients never have a firm diagnosis, however others do after many years of follow up. In either case you treat the patient in front of you and maintain a high index of suspicion.

A recurring theme throughout the week was that a presenting phenotype does not always match the corresponding genotype. In the patient I presented a variant of unknown significance was identified in the NBN gene which is associated with Nijmegan breakage syndrome. Despite the fact that we had not considered this diagnosis for our patient before this information was presented, it was suggested that we should complete out investigation for this disease as a possibility as perhaps this could be a case that the phenotype does not match the genotype.

The use of genetic testing for patients with PID can be very useful, especially when the patient is found to have a well-known condition with the associated genetic abnormality. However, it was discussed during the course that genetic testing can also reveal conditions or potential for disease that the patient or family did not want to know. There may also be further implications that families had not considered, for example insurance implications. These important ethical concerns need to be discussed thoroughly with the patient and their family before proceeding with genetic testing.
APID2016 CASE SUMMARY: HYPER IGE SYNDROME (HIES)

Presented by Dr Paul Mansfield

Case history:
AP is a 23 year old male with autosomal dominant (AD)-HIES. He developed eczema and recurrent skin infections in the neonatal period. At three years of age, he required surgical release of optic nerve compression secondary to craniosynostosis (fused cranial bones). This complication led to the diagnosis of HyperIgE syndrome. His IgE at the time of diagnosis was ~2000kU/L. In 2007, he was confirmed to have a Stat 3 mutation on genetic testing, a de novo mutation. His predominant problem has been of recurrent pneumonias, which occur once to twice a year on average, despite being on intravenous immunoglobulin replacement. He is on continuous Bactrim and starts antibiotics when feeling unwell; resistant organisms have not been a problem. Fortunately, he has not developed bronchiectasis despite the frequent pneumonias. Even when he has a dense lobar pneumonia he has only a limited CRP (up to 30mg/L) and neutrophil response (up to 11x10^9/L). He has had minimal skin infections with the use of cetaphil wash. However, mucocutaneous candidiasis has been very problematic, and he has persistent oral and penile thrush and candida onychomycosis despite continuous fluconazole 200mg daily. He also has the complication of osteoporosis, and had his first fracture at 8 years of age. He has now had 9 minimal trauma fractures despite pamidronate and then zoledronic acid. AP also has retained primary dentition, resulting in two rows of teeth. He has consulted an oral surgeon, but the decision was not to proceed with primary dentition removal, in part because of nervousness about the long history of bisphosphonate use.

AP has many specialties involved in his care and has frequent hospital admissions. Nonetheless, he has full-time employment, a partner and actively participates in sport, including tennis and the gym. He recently enjoyed a three month holiday to Europe.

HIES:
AD-HIES (previously called Job’s syndrome) is typically due to mutations in Stat3. It is rare, with an estimated prevalence of 1:100,000 - 1:500,000. It is typified by immunologic and connective tissue abnormalities. The classical triad is of:
   a. an eczematoid rash and skin abscesses (Staph), starting within ~1 month of birth
   b. recurrent sino-pulmonary infections; and
   c. an elevated IgE, usually over >2,000IU/mL +/- eosinophilia

The connective tissue abnormalities include retained primary dentition, aberrant wound healing leading to pneumatoceles after pneumonia, osteopenia and minimal trauma fractures, scoliosis and joint hyper-mobility, a characteristic facies and vascular abnormalities, including of dilatations and aneurysms. Similar manifestations occur in animal models of Stat3 deficiency. Scoring indices have been developed to predict the likelihood of finding a Stat 3 mutation, and have five key features that predict a Stat 3 mutation with a sensitivity of 87.5% and a specificity of 80.6%. These features are pneumonia, newborn rash, pathologic fractures, the characteristic face of Job syndrome and a cathedral palate.

By comparison, autosomal recessive (AR)-HIES is mainly due to mutations in DOCK8 and tends to occur in consanguineous families. Patients are particularly prone to viral skin infections, such as with Molluscum contagiosum, Herpes simplex and zoster and human papilloma virus. They also tend not to have the connective tissue manifestations of AD-HIES.

Patients with AD-HIES frequently have normal serum levels of IgG, but the quality of the IgG responses can be sub-optimal, hence the immunoglobulin replacement. It should also be noted that an elevated serum IgE is not required for diagnosis; particularly in adults the IgE may be within the...
normal range. AD-HIES patients generally do well with intensive therapy and supportive care. Bone marrow transplantation (BMT) can be considered for the infective complications but of course will not correct the connective tissue abnormalities. This contrasts to the situation in AR-HIES, in whom a BMT is curative.

**Sources:**
1. Up-to-date
2. Mutations in Stat3 and diagnostic guidelines for hyper IgE Syndrome, JACI, 125 (2)
APID2016 CASE SUMMARY: LONG TERM PIECE OF MIND (XLT/WAS)

Presented by Dr Birgit Marchand

Case Vignette:
Patient TE was found to have a WAS gene mutation which confers upon him an XLT phenotype rather than an overt WAS phenotype; 5.6% WAS protein expression and WAS Ochs score equal to 2. Presence of moderate thrombocytopenia, easy to control eczema, and on testing, immune function is essentially normal. His case was discussed at a TAPID meeting, and it was recommended that he undergo a bone marrow transplant; in his favour he had a HLA matched older sibling. The parents decided to face the upfront risks of transplant to effect a cure, rather than face the medium term risk of ICH and the longer term risks of treating XLT conservatively.

Discussion:
Whether patient TE’s particular mutation and his extremely low WASP expression can accurately predict his outcome in life is controversial. His mutation is described in a total of 29 patients found in 3 sentinel articles. Albert et al reported on the long term follow-up of 173 patients with XLT in Blood in 2010, the largest series of XLT patients to date. Their conclusion was that mutation was not predictive of overall survival or onset of events. Imai et al on the other hand, in an earlier publication on the clinical course of patients with the WASP gene mutation concluded that there was a strong geno-phenotype correlation in 50 Japanese patients.

What is the significance of his 5% WASP expression? In the Albert paper, WASP expression had no influence on overall survival or event free survival. But in the Imai paper, there was a strong protein expression / phenotype correlation. Long term complications of XLT are best described in the Albert paper which described very long term follow-up of 173 children with the XLT phenotype and did not include those with WAS. The most relevant points are: Overall survival of 96% at 30 years and 81% at 60 years, which is equivalent to the survival of the overall German male population. Event free survival of 74% at 15 years, 56% at 30 years and 27% at 60 years i.e. by the age of 30, 54% had had a significant adverse effect. Intracranial hemorrhage occurred in 10% (of whom 3 died). 10% of patients had significant bleeding in other sites – namely bowel, lung, and ENT. Median onset of bleeding was 5.7 years. In summary, 25% had an event by the age of 15, rising to 73% by age 60. Notably there was no correlation with platelet counts. Infection occurred in 10%, cancer developed in 5%, and autoimmune disease developed in 15%. Historically the most information regarding transplantation for a child with a mutation in the WAS gene has been for children with the WAS phenotype.

In 2008, Ozsahin et al reported on the long-term follow-up of 96 WAS patients who received transplants between 1979 and 2001 and who had survived at least 2 years following transplant. Of the 96 survivors, 45 received matched sibling donor transplants and had event free survival of 88% at 7 years. Subsequent paper by Moratto et al in 2011, which included 66 of Ozsahin’s original 96 patients, there were 39 patients with WAS who received a matched sibling donor transplant and they had a 95% overall survival.

In 2015, Oshima published the outcome of stem cell transplantation for 24 XLT patients in the journal of Clinical Immunology: 24 patients transplanted over 21 years in 14 centres in 5 countries. Oshima et al concluded that HSCT following myeloablative transplant is curative and associated with acceptable risks as a treatment option for XLT.

Are there any other treatments? Splenectomy mostly increases platelet count but it is not guaranteed and more importantly is accompanied by a life long risk of overwhelming post-splenectomy infection. A treatment algorithm for XLT, proposed by Worth and Thrasher 2015, favour splenectomy. However they acknowledge the risk of subsequent sepsis and lifelong prophylactic antibiotic therapy. Worth and Thrasher also suggest HSCT in XLT patients who go on to progress to a clinical phenotype more typical of WAS. Gene therapy: Autologous gene modified
HSCT is an experimental option for WAS children without a suitable donor for allogeneic transplantation. Thrombopoietin receptor agonist: Gerrits et al trialed the effect of thrombopoietic agent Eltrombopag, and found that it increased platelet counts, but did not improve platelet activation in WAS/ XLT patients.

Arguments in favour of BMT for XLT are the following; strongly supported during APID discussion

1) BMT will most likely increase the platelet count and prevent intracranial hemorrhage. If our patient were to sustain an ICH, there are three possible outcomes: death, alive with significant morbidity or total recovery
2) Reduction in anxiety and improvement in peace of mind in the setting of managing a child with a low-platelet count at risk of serious life threatening bleeding
3) BMT will reduce the risk of onset of infections
4) BMT will probably prevent the development of autoimmune disease in XLT
5) There is a risk of developing cancer due to XLT, however this may be counterbalanced by the risk of secondary cancer as a consequence of receiving chemotherapy for BMT
6) Our patient's WASP expression could be viewed as effectively 0 however, and a number of publications report this as being predictive of a difficult life.
7) Although our patient's WAS Ochs score now is 2, scoring at the age of 2 years maybe unreliable and may change as he gets older.
8) The results are best when performed in the younger years, before complications develop. Our patient is in this age group
9) If one is considering a transplant, the results are best for a matched sibling transplant, and our patient has MSD
10) Eminent immunologists in Australia, Great Britain and France have emphatically recommended it, including members of the TAPID group

Arguments against Transplant:
1) The overall survival of patients with XLT in the Albert series was the same as that of the general population
2) WASP expression and its relevance to the XLT phenotype is controversial
3) At the age of 2 years it is hard to predict his future on what is known about his mutation. There are some long term survivors recorded to age 55. His score may or may not change over time.
4) There is a transplant mortality rate of 5% attached to having a matched sibling donor transplant
5) There is a risk of rejection
6) There is a risk of delayed mixed donor chimerism, which may results in ongoing thrombocytopenia, delayed immune reconstitution and development of autoimmune disease. One example of the latter is the development of ITP and this would be total irony.
7) Infertility with the dose of cyclophosphamide is highly likely
8) GVHD can become a chronic illness in its own right that requires treatment, sometimes for several years. It is of absolutely no benefit to him, and increases his risk of infection, autoimmune disease and death
9) Secondary cancer is a rare but well documented consequence of receiving chemotherapy for a BMT. However balanced against this is the risk of the development of cancer if his XLT remains untreated.
10) The welfare of his potential donor also needs to be considered.
11) There are also variables such as experience of transplant centre, health status of patient, donor selection, conditioning regimen and GVHD prophylaxis that need to be considered for ensuring a most favourable outcome.
APID2016 CASE SUMMARY: LRBA DEFICIENCY, GETTING TO THE GUT OF THE PROBLEM

Presented by Dr Melissa Norman

Here is described a case of a 7 year old boy of Turkish heritage, born to non-consanguineous parents presenting with signs of recurrent infection and systemic auto-inflammation.

His initial presentation was at the age of 3 with recurrent episodes of thrombocytopenia, treated with IVIG and high dose methylprednisolone. He also had chronic diarrhoea which was initially thought to be post infectious after salmonella was detected on stool. He had recurrent otitis media with a chronic perforation on the R. At 4yo he developed anaemia and a fluctuating low grade neutropaenia, and was diagnosed with Evan's syndrome. Bone marrow biopsy showed a normocellular marrow with increased megakaryocytes and his DAT on serum was positive. This was consistent with peripheral destruction and he was maintained on low dose (</= 0.5mg/kg) oral prednisolone. He experienced a life threatening haemolysis event requiring ICU admission 8 days post Pneumovax23 vaccine administration.

On further evaluation he had evidence of multi-organ inflammation with hepatosplenomegaly and significant lymphadenopathy. He had a chronic cough and CT/PET of the chest showed inflammatory nodules. Lung biopsy showed patchy fibroblastic foci with chronic inflammatory change. He continued to have diarrhoea and abdominal pain and was failing to thrive. On endoscopy/colonoscopy he was found to have eosinophilic oesophagitis, and widespread colitis with an inflammatory infiltrate and candida species present on biopsy. He had chronic carriage of both Clostridium difficile which was both antigen and toxin positive) on repeat stool culture.

Immune work up revealed low B cells and a normal IgG (8) with low IgM (0.2) and very low IgA (0.08). He had normal responses to protein vaccines with low polysaccharide responses to pneumococcal serotypes (3.3) after routine vaccination. He had normal T cells subsets and functional response to PHA. He had raised double negative T cells (8%) with 4% α/β T cells, 4% γ/δ T cells. ALPS gene testing had been previously negative (FAS, FAS L, CASP10, CASP8, PRKCD). NK cell numbers were normal with normal degranulation but reduced lysis.

Whole genome sequencing led to discovery of a compound heterozygous gene defect in the LRBA (lipopolysaccharide-responsive vesicle trafficking, beach-and-anchor-containing) protein encoding gene (chr4:g151604817CA>C Deletion/chr4g:g151356773G>A SNP). One mutation is a frameshift deletion and the other a single nucleotide change which induces a stop codon. One copy was found in either parent, and both would be expected to truncate the protein and impair function.

The LRBA protein co-localises with CTLA4 (inhibitory checkpoint protein) in endosome and is involved in its trafficking to cell surface of activated T cells and Tregs. CTLA4 is a competitive inhibitor of CD28 for CD80/86 ligand which results in down-regulation of T cell responses. In LRBA deficiency, there is increased CTLA4 turnover and reduced expression on cell surface which results in uncontrolled T cell inflammatory responses.\(^i\)

He has been treated with Ig replacement, high dose steroids, and several immuno-modulatory medications which have led to significant symptomatic improvement. Abatacept is a CTLA4 Ig fusion drug. This replaces the missing CTLA4 function and blocks activating interaction with CD28 on T cells and Tregs.\(^ii\) Sirolimus is a mTOR inhibitor which, through blocking this pathway, limits T cell activation and function and increases the activity of Tregs.\(^iii\) Hydroxychloroquine inhibits
lysosomal degradation, and can increase CTLA4 at the cell surface to enhance its inhibitory function.2

Haematopoietic Stem Cell Transplant has been performed in several patients with LRBA deficiency and there are 5 published cases3,v,vI. All were conditioned with reduced intensity regimens and received matched related donor transplants. Follow up outcomes suggest improvement in cytopenias, immune function, inflammatory bowel symptoms and growth. Two patients have ongoing thrombocytopenia and there has been one reported death.

In summary, this case represents a presentation typical of the LRBA spectrum. Knowledge of the specific gene defect has allowed for the use of targeted therapy which has led to significant symptomatic improvements for this patient. HSCT remains a treatment option and as experience with this condition increases, prognostication and risk assessment should become more clear in deciding the therapeutic path to take.

References


1 Lo, B. Patients with LRBA deficiency show CTLA4 loss and immune dysregulation responsive to abatacept therapy. Science. 2015 Jul 24;349(6246):436-40


APID 2016 CASE SUMMARY: ROIFMAN SYNDROME

Presented by Dr Alberto Pinzon

Roifman Syndrome is a rare congenital disorder characterized by growth retardation, cognitive delay, spondyloepiphyseal dysplasia and antibody deficiency. In spite of some variability, all subjects share remarkably identical dysmorphic, skeletal and immunological features which are the clues to diagnosis.

Dysmorphism

All individuals have typical facial features including a markedly long philtrum with a thin upper lip, a narrow, tubular and upturned nose with hypoplastic alae nasi, widely spaced eyes with long palpebral fissures and prominent lashes.

Skeletal Dysplasia

All cases also present highly characteristic skeleton abnormalities. The proximal epiphyses of the femora demonstrate symmetric delayed ossification, as well as mild flattening and irregularity; unlike Schimke immuneosseous dysplasia, the acetabulae are normal. Similar but less pronounced changes are seen in the other epiphyses of the axial skeleton: the vertebrae are ‘bullet’ shaped or biconvex at an age one would expect them to be ‘squarer’. In addition, all cases have been reported to have brachydactyly, most also have transverse palmar creases and clinodactyly of the fifth digit.

Immunological abnormalities

While serum immunoglobin levels are variable, all patients have been shown to be unable to produce specific antibodies. Circulating B-cell number is on the lower end of the normal range, with mature B cell and memory B-cell numbers within normal ranges. T-cell number and function are completely normal. Summary of immunological investigations in all cases described (table below)

<table>
<thead>
<tr>
<th>Lymphocyte Markers</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
</tr>
</thead>
<tbody>
<tr>
<td>CD3</td>
<td>1.6</td>
<td>2.3</td>
<td>2.2</td>
<td>3.1</td>
<td>2.4</td>
<td>2.66 (0.66-2.41)</td>
</tr>
<tr>
<td>CD4</td>
<td>1.0</td>
<td>1.4</td>
<td>1.7</td>
<td>1.4</td>
<td>1.6</td>
<td>1.03 (0.43-1.62)</td>
</tr>
<tr>
<td>CD8</td>
<td>0.6</td>
<td>0.9</td>
<td>0.4</td>
<td>1.2</td>
<td>0.7</td>
<td>1.05 (1.5-1.01)</td>
</tr>
<tr>
<td>CD4/20</td>
<td>0.07</td>
<td>0.08</td>
<td>0.04</td>
<td>0.04</td>
<td>0.39</td>
<td>0.07 (0.08-0.58)</td>
</tr>
<tr>
<td>CD16/96</td>
<td>0.2</td>
<td>0.29</td>
<td>0.18</td>
<td>0.32</td>
<td>0.58</td>
<td>0.11 (0.05-0.52)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Mitogen Response</th>
<th>PHA (counts x 10^9)</th>
<th>SAC</th>
<th>Immunoglobulin(sg/L)</th>
<th>IgG</th>
<th>IgA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>123.95</td>
<td>3.31</td>
<td>7.99 (6.7-7.17)</td>
<td>19.06 (6.7-7.17)</td>
<td>9.506 (7.17-13.5)</td>
</tr>
<tr>
<td></td>
<td>119.117</td>
<td>2.64</td>
<td>19.06 (6.7-7.17)</td>
<td>19.06 (6.7-7.17)</td>
<td>9.506 (7.17-13.5)</td>
</tr>
<tr>
<td></td>
<td>105.130</td>
<td>4.12</td>
<td>19.06 (6.7-7.17)</td>
<td>19.06 (6.7-7.17)</td>
<td>9.506 (7.17-13.5)</td>
</tr>
<tr>
<td></td>
<td>Normal</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>Normal</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Antibodies</th>
<th>0.2</th>
<th>0.37</th>
<th>0.37</th>
<th>0.37</th>
<th>0.41 (0.43)</th>
<th>0.60 (0.5-1.6)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anti-Tetanus (U/L)</td>
<td>&lt;0.01 (0.04)</td>
<td>&lt;0.01 (0.04)</td>
<td>&lt;0.01 (0.04)</td>
<td>&lt;0.01 (0.04)</td>
<td>0.27</td>
<td>&lt;0.1 (0.16)</td>
</tr>
<tr>
<td>Anti-Diphtheria (U/L)</td>
<td>&lt;1.8 (8/16)</td>
<td>&lt;1.8 (8/16)</td>
<td>&lt;1.8 (8/16)</td>
<td>&lt;1.8 (8/16)</td>
<td>0.23</td>
<td>&lt;0.1 (0.15)</td>
</tr>
<tr>
<td>Anti-Haemophilus Influenza (U/L)</td>
<td>0.23</td>
<td>1.02</td>
<td>0.23</td>
<td>1.02</td>
<td>0.23</td>
<td>1.02</td>
</tr>
<tr>
<td>Anti-Pneumococcus (fold increase)</td>
<td>NR(&gt;3)</td>
<td>NR(&gt;3)</td>
<td>NR(&gt;3)</td>
<td>NR(&gt;3)</td>
<td>0.23</td>
<td>1.02</td>
</tr>
</tbody>
</table>

- Page 22 of 38 -
Retinal Dystrophy

50% of individuals have been shown to have retinal dystrophy with extensive degeneration of the rod and cone systems.

Genetics

While it was previously proposed that Roifman Syndrome was a novel ciliopathy with immunodeficiency, because of retinal dystrophy and some early and transient bone changes, this could not be confirmed despite extensive investigation. X-linked inheritance was also suspected because most reported cases were males. Candidate gene studies using targeted sequencing were unsuccessful in identifying causal variants. Only until recently and through whole-genome sequencing it has been found that all Roifman Syndrome patients are compound heterozygous rare variants of the RNU4ATAC small nuclear RNA gene, a minor spliceosome component that is essential for minor intron splicing.

To understand the potential effect that disruption of the minor spliceosome could have on cellular, immune and skeletal functions and the phenotype of patients with Roifman syndrome display, however, it is essential to understand the role of the small spliceosome. As DNA is transcribed into RNA and then into the various proteins that perform the functions of life, non-coding gene sequences (introns) need to be removed from the transcribed RNA strand and the remaining gene sequences (exons) joined together. This is the job of specialized molecular machinery called the spliceosome. There are two varieties of spliceosomes, the so-called major and minor. The major spliceosome is by far the most abundant, such that the role of its minor counterpart is often disregarded. With each type of spliceosome recognizing different splicing cues, the major spliceosome acts on the vast majority of introns (>200,000) and the minor one splices the several hundred minor-type introns. Moreover, the evolutionary persistence and role of the minor spliceosome has remain puzzling since the minor introns it targets are far outnumbered by the major introns handled by the major spliceosome. Interestingly, the mRNAs produced from genes that have a minor intron are not ready until all their introns, both major and minor, are spliced. Thus a single inefficiently spliced minor intron can hold up expression – mRNA and protein production – for an entire gene.

Roifman syndrome patients as well as patients with a similar condition called micropcephalic osteodysplastic primordial dwarfism-1 (MOPID-1) have demonstrated the important concept that when minor spliceosome activity is reduced, the minor introns are retained in the mRNA. This, unfortunately signals the mRNA for degradation, limiting the expression of genes that contain minor introns. Therefore, expression of genes that are “minor-intron-rich” is proposed to be responsible for the plethora of clinical characteristics of these patients. See graph below
References


APID2016 CASE SUMMARY:
A CASE OF MONOGENIC IMMUNE DEFICIENCY AND AUTOIMMUNITY

Presented by Dr Phillipa Pucar

A 40 year old female was referred for investigation of a possible primary immunodeficiency. She had an 18 month history of recurrent sino-pulmonary infections with underlying bronchiectasis as a young adult. She had chronic oral herpes simplex, shingles, oral and vaginal candidiasis, chronic hepatitis B on lamivudine, and prior peripartum *Listeria* sepsis. 6 years earlier, she had a splenectomy for refractory idiopathic thrombocytopaenia purpura and remained on penicillin prophylaxis. She was treated with sulfasalazine for psoriatic arthritis, which was stopped owing to profound thrombocytopaenia which did not resolve. A single dose of Romiplostim caused an impressive platelet response from <10 to 1300. Our patient migrated from Burma, was married with two well children and the youngest of 9 with short stature but no other dysmorphic features.

Initial assessment revealed hypogammaglobulinaemia (IgG 5.7g/L, IgA 0.43g/L, normal IgM), with suboptimal vaccine responses. She had a lymphocytosis (6.34x10^9) with reduced switched and unswitched memory B cells and increased CD21-lo cells placing her in the Freiberg IA category of common variable immunodeficiency with a predisposition to autoimmune presentations. She responded well to immunoglobulin replacement therapy. Whole exome sequencing identified a heterozygous missense mutation in signal transducer and activation of transcription (STAT) 3, resulting in a methionine-to-threonine substitution at position 394 (M394T) in the DNA binding domain.

STATs are important intracellular signaling molecules downstream of multiple cytokine receptors, and influence gene transcription.1 Mutations in the JAK-STAT pathway are associated with immunodeficiency phenotypes including STAT1 deficiency (viral and mycobacterial infections) and STAT3 loss of function (staphylococcal boils, candidiasis, eczema and dysmorphic features through loss of Th17 differentiation).2

Activating mutations in STAT3 have only recently been described, first by Flanagan et al;3 who reported 4 different de novo germline mutations in highly conserved STAT3 domains in 5 patients with early onset autoimmune disease (diabetes, enteropathy, interstitial lung disease, juvenile arthritis, hypothyroidism), short stature and eczema. Muted STAT3 responses to IL-6 stimulation were noted relative to wild type, and regulatory T cells (Tregs) were reduced. Milner et al4 identified 9 germline activating mutations in 13 patients with lymphadenopathy, autoimmune cytopenias, multiorgan autoimmunity, recurrent infections and short stature. Hypogammaglobulinaemia, T cell lymphopaenia, expansion of double negative T cells, reduced STAT1 and STAT5 phosphorylation on stimulation and a reduction in Tregs was noted. One patient was transplanted attaining complete remission, and another treated successfully with IL-1 blockade. Haapaniemi et al5 described 3 patients with an immunodeficiency phenotype and multisystem autoimmunity, where the first reported case of the M394T mutation was reported. This patient had mild autoimmunity (lymphocytic colitis, ITP and eczema), recurrent respiratory tract infections, lymphopoenia, hypogammaglobulinaemia with a Freiberg IA phenotype, reduced Tregs and disseminated mycobacterial disease with intact IL-12-IFNy signalling.

In summary, germline activating mutations in STAT3 cause dysregulated cytokine signaling, a reduction in Tregs and altered Th17 cells resulting in autoimmunity and immunodeficiency as demonstrated by our case. The M394T mutation predisposes to mycobacterial infectious however the
cause for this is not known. STAT signaling is complex, as demonstrated by STAT3 loss of function and activating mutations which do not cause opposing phenotypes. These findings have important implications for the understanding of lymphocyte tolerance and the pathogenesis of systemic autoimmunity.

References

APID 2016 CASE SUMMARY:
RETICULAR DYSGENESIS – A CASE OF SEVERE COMBINED IMMUNODEFICIENCY

Presented by Dr Stephanie Richards

A male infant born at 36 weeks gestation to non-consanguineous parents developed temperature instability and presumed sepsis on day 1 of life. A routine full blood count demonstrated marked neutropaenia (0.02x10^9/L) and lymphopaenia (0.3x10^9/L), and he was transferred to the Royal Children’s Hospital, Melbourne, for further investigation and management. Subsequent investigations confirmed agranulocytosis with marked reduction in lymphocytes and monocytes on bone marrow aspirate. Peripheral blood flow cytometry demonstrated undetectable T cells with essentially normal numbers of B and NK cells, with virtually absent naïve T cells, consistent with a diagnosis of reticular dysgenesis severe combined immunodeficiency. He has subsequently had genetic confirmation of a homozygous mutation in exon 1 of the AK2 gene, and has undergone a successful matched unrelated donor haematopoietic stem cell transplant.

Reticular dysgenesis SCID (RD-SCID) was first described in 1959 and is one of the more rare forms of SCID. It is characterised by granulocytopenia, impaired lymphoid maturation and sensorineural deafness. Affected infants are extremely susceptible to infections and early death, and subsequently the majority of patients present in the first week of life.

The gene defect underlying RD-SCID, mutations in mitochondrial adenylate kinase 2 (AK2), was identified in 2009. Mitochondrial oxidative phosphorylation is the process by which cellular energy is generated, in the form of ATP. Multiple enzymes within a cell are required to maintain the balance of energy, of which AK2 is the primary ADP-generating enzyme. Leukocytes express AK2 but little or no alternative protein for the generation of ADP that is required for mitochondrial oxidative phosphorylation. This differential expression of AK2 describes why leukocytes may be particularly susceptible to the defects caused by lack of AK2 in RD-SCID. AK2 is also required for neutrophil differentiation, but not differentiation of other myeloid lineages, which may underlie the effect of AK2 mutations of neutrophil development and contribute to the granulocytopenia that is characteristic of RD-SCID.

To date there are 30 published cases of RD-SCID. The only current curative therapy is haematopoietic stem cell transplant. Use of a myeloablative-conditioning regimen is associated with better survival outcomes, likely due to the high rates of maternal-foetal engraftment in these patients. G-CSF is not effective in these patients. More recently, the use of antioxidant therapy prior to HSCT has been proposed as a therapeutic option, due to reduction in reactive oxygen species production and cellular damage.
APID2016 CASE SUMMARY:
RAPAMYCIN REDUCES SPLENOMEGALY AND LYMPHADENOPATHY IN COMMON VARIABLE IMMUNODEFICIENCY (CVID): A CASE REPORT

Presented by Dr Sarah Sasson

Case Report:
A 54 year old female was hospitalised with anti-microbial-sensitive *H. influenza* pneumonia, her first major infection. Past medical history included prior motor vehicle accident, 160 pack year history of smoking and a recent diagnosis of chronic obstructive pulmonary disease (COPD). Medications on admission were Budesonide-Efomoterol, terbinafine and bupropion. There was no history of allergy, recent travel, sick contacts, recreational drug use or pets in the home. The patient was treated with 1.2g of IV Benzylpenicillin QID and a weaning dose of oral prednisone beginning at 50mg PO OD. She failed to clinically improve by Day 10 prompting further investigations.

A progress chest X-ray showed bilateral lower zone infiltrates while computer tomography (CT) of the chest, abdomen and pelvis showed evidence of endobronchial infection as well as hepatosplenomegaly and widespread axillary, mediastinal and Para aortic lymphadenopathy. Differential diagnoses at this stage included underlying malignancy, sarcoidosis or connective tissue disease.

Laboratory investigations including FBC, EUC, LFT, autoimmune serology and investigations for infections including HIV, viral hepatitis, CMV, EBV, aspergillus, mycobacteria and pertussis were all unremarkable. The inflammatory markers CRP and ESR were elevated at 64 and 28, respectively. The most remarkable finding was of a panhypogammaglobulinemia with low levels of all IgG (0.3g/L) IgA (<0.3g/L) and IgM (0.5g/L). An axillary lymph node biopsy performed to investigate for sarcoidosis and malignancy showed reactive lymphoid hyperplasia.

A provisional diagnosis of common variable immunodeficiency (CVID) was made. The patient received 0.4mg/kg of intravenous immunoglobulin (IVIG) as an inpatient and was discharged with a plan for monthly outpatient IVIG. A bone marrow aspirate and trephine showed sparse plasma cells, which is consistent with CVID, and no evidence of malignancy.

Over the following twelve months the patient’s lymphadenopathy and splenomegaly worsened with splenic capsular pain becoming a major complaint. A positron electron tomography (PET) scan showed wide-spread avid lesions above and below the diaphragm reported as consistent with sarcoidosis or lymphoma. A second axillary lymph node biopsy showed atypical lymphoid strictures and lymphoid hyperplasia with no evidence of malignancy. Immunophenotyping demonstrated a B+SMB+CD21low phenotype by EuroClass criteria, which has a higher association with splenomegaly and granulomatous disease. Additionally all of the patients CD4 and CD8 T-cells displayed a CD45RA-CCR7- activated/terminally differentiated phenotype.

The role of the mTOR pathway was investigated by using an HRP-conjugated antibody to pS6 (a protein downstream of mTOR) and immunohistochemistry on the previously obtained lymphoid tissue. Immunohistochemistry found up-regulated expression of pS6 in the patients follicular B and T-cells compared with normal tonsillar tissue. Additionally, there was minimal EBER staining (<10% of follicles).

Given this provisional evidence of up-regulated mTOR activity, the patient was commenced in a trial of rapamycin (an mTOR inhibitor) 6mg PO STAT followed by 2mg PO OD aiming for a trough of 5-10microg/L. After two months of treatment the patient displayed marked clinical improvement with reduction of lymphadenopathy and splenomegaly and associated pain. A progress PET scan

---

1 Wehr et al Blood 2008
confirmed decreased tracer uptake into lymph nodes, marked reduction in hepatosplenomegaly and decreased SUVmax of the spleen from 4.4 to 2.9.

Summary and Discussion:
This patient represents a late presentation and diagnosis of CVID in a patient whose clinical course was not characterised by recurrent infections. Patients with CVID have decreased survival compared with the general population over 40 years\(^2\) and patients with the non-infectious complications of CVID have the highest mortality. Unfortunately good evidence for the treatment and management of such complications is lacking.

Rapamycin has been used successfully in autoimmune lymphoproliferative syndrome (ALPS)\(^3\) a condition that generally presents in the paediatric population. This condition is characterised by defective lymphocyte apoptosis, secondary to mutations in Fas/FasL, and lymphoid hyper-proliferation mediated by the mTOR pathway. ALPS commonly presents with marked cervical lymphadenopathy. Histopathologically, ALPS is driven by Epstein Barr virus (EBV) and is associated with a characteristic expansion of CD4-8- double negative T-cells, which were not features in this case.

A number of single gene mutations have been associated with primary immunodeficiencies that may be relevant to this patient. Mutations in AID and UNG have been associated with autosomal recessive hyper-IgM syndrome\(^4\) with low IgG, IgA and enlarged lymph nodes and germinal centres. However in this condition serum IgM levels are generally normal to high and not low, as in this case. PI3K gain of function mutations have been described in fourteen patients with sino-pulmonary infections, lymphadenopathy, nodular lymphoid hyperplasia and CMV or EBV viremia\(^5\). In these patients naïve and central memory T-cells were deficient and senescent T-cells were over-represented. Over-activation of the mTOR pathway was demonstrated in these patients and addition of rapamycin \textit{in vitro} partially restored T-cell defects.

Mutations in ICOS have been associated with variable age of onset recurrent bacterial infections, splenomegaly, autoimmunity, intestinal lymphoid hyperplasia and poor germinal centre formation\(^6\). Finally, polymorphisms in CTLA-4 have been associated with autoimmunity and granulomatous disease with or without lymphoid hyperplasia in approximately one third of patient\(^7\). Our patient is currently undergoing whole exome sequencing to investigate for these aforementioned mutations and polymorphisms and also to investigate for novel gene mutations.

Conclusions:
This case suggests the mTOR pathway may be up-regulated in some cases of CVID-associated splenomegaly and lymphadenopathy. In such cases mTOR inhibitors may be useful therapy, however the optimal duration of such therapy is unclear. Whole exome sequencing of this patient’s DNA is currently underway in the hope of identifying a previously reported or novel molecular diagnosis.

Acknowledgements:
Dr Adrienne Morey, A/Prof William Sewell, Mr Sandy Smith, Dr Elissa Deenick, Prof Stuart Tangye Dr Mark D'Anta, Dr Louise Emmett, Dr Keith Fay, Prof Tony Kelleher, Dr Tri Phan

\(^2\) Resnick \textit{et al} Blood 2012  
\(^3\) Dragana \textit{et al} Pediatric Blood Cancer 2009  
\(^4\) Lee \textit{et al} Blood 2005  
\(^5\) Lucas \textit{et al} Nature Immunology 2014  
\(^6\) Grimbacher \textit{et al} Nature Immunology 2003  
\(^7\) Knight \textit{et al} Journal of clinical Immunology 2007
APID2016 CASE SUMMARY: A TERM INFANT GIRL

Presented by Dr Sabeena Selvarajah

A term infant girl presented at 2 months of age with eczema, intermittent diarrhoea, failure to thrive, persistent oral candida and functional asplenia. A colonoscopy was largely unremarkable and she was managed with prophylactic antibiotics and additional vaccinations. At 6 months of age she had a prolonged admission with PJP complicated by pulmonary hypertensions and an Enterobacter line infection. Post discharge she had ongoing issues with eczema, drug allergies/atypical drug eruptions and severe failure to thrive.

Whilst a primary immune deficiency was suspected there was discordance between the clinical phenotype and the laboratory markers. The infant had a persistent mild CD3/CD8/CD19 lymphopenia with an eosinophilia. T cell proliferation (with Concanavalin A and PHA) was normal on two separate occasions. Immunoglobulin replacement was commenced at six months due to worsening hypogammaglobinemia, though this could have reflected the nadir of maternal immunoglobulin protection. IgE was briefly raised and then normalised. A variety of diagnoses were considered ranging from hypomorphic SCID or Omenn's syndrome, to a combined immune deficiency such as ZAP-70 or Card 9. Immune deficiencies associated with immune dysregulation were also considered (e.g. IPEX-like disorders) however, given the inconsistencies, the case was difficult to categorise.

A decision was made to transplant based on clinical phenotype rather than risk another life limiting infection and the associated reduction in morbidity/mortality outcomes. The post-transplant course was complicated by atypical cutaneous Graft versus Host disease requiring ongoing immunosuppression. This has led to a number of medication side effects including a minimal trauma fracture. Extensive targeted next gene ration sequencing has yet to identify a pathogenic mutation.

The case highlights the importance of transplanting based on clinical phenotype rather than awaiting a molecular diagnosis. It also encourages further discussion of the changing diagnostic landscape of primary immune deficiencies. Targeted sequencing of genes associated with primary immune deficiencies is being gradually being replaced by whole exome/genome sequencing. Targeted sequencing has the advantage of greater accuracy, fewer incidental findings and less expense. However, with increasing numbers of monogenic primary immune deficiencies being identified and the growing acknowledgment of the clinical heterogeneity within these disorders, whole exome (WES) and whole genome sequencing (WGS) has gained favour for complex cases. It is also easier to identify novel mutations using the latter approach.

However WES/WGS relies on accurate interpretation and the ability to separate common polymorphisms from pathogenic mutations. Some studies have found a false positive rate as high as 36%. Whole exome sequencing is also unable to detect intronic mutations. Whilst the majority of pathogenic mutations are within the exome, there are case reports of intronic mutations including for UNC13D, NEMO, SH2D1A and ZAP-70 deficiency. Finally, whole exome/genome sequencing is associated with an increased risk of incidental findings. In 2013 the American Society of Genomics and Genetics published a list of 56 mutations, recommending they be reported “regardless of the clinical indication for sequencing.” These included mutations associated with breast & ovarian cancer, cardiomyopathy, Marfan’s, long QT and polyposis syndromes. Many of these conditions would not manifest until adulthood. The recommendation caused consternation, particularly amongst paediatricians. The majority of paediatricians polled by the American college of Paediatrics
did not agree with this approach. Whilst there is no consensus within Australia, the delegates at the meeting agreed this is an area which needs further consideration.

References


APID2016 CASE SUMMARY: A CASE OF TTC7A DEFICIENCY

Presented by Dr Bella Shadur

Case summary

- 4/12 male
- Non-consanguineous, Anglo-Saxon heritage
- Presented with severe failure to thrive and diarrhoea
  - Initially worked up for cow’s milk protein intolerance
  - No improvement with various HA formulas
- Colonoscopy: severe inflammation of the entire colon
- Referred to immunology: could this be IPEX or IL10/IL10R deficiency?
- Baseline immune work-up demonstrated:
  - IgG 0.29 (↓), IgA 0.29 (normal), IgM 0.08 (↓), IgE < 5
  - Normal lymphocyte subsets
  - T cell phenotype: normal proliferation to PHA and anti-CD3, STAT5p, TREC, naïve T cells
  - Neutrophils: normal NBT and DHR
  - IPEX: normal FoxP3, CD25 staining, endocrine autoantibodies NAD
  - HLH: ferritin 50, SAP, XIAP, perforin, NK cell degranulation normal
- Enrolled in NEOPICS study (Inter-National Early Onset Paediatric IBD Cohort Study)
  - WES: compound heterozygous for two novel mutations in the TTC7A gene
  - Segregated in parents (both of whom asymptomatic)
  - Both predicted to be pathogenic, although one required confirmation via the creation of a DNA ladder
- Patient stabilised on subcutaneous immunoglobulin and total parenteral nutrition
- Ongoing monitoring of immune parameters, particularly T cell function
- For consideration of bone marrow transplant

Learning points

- TTC7A deficiency = Tetratricopeptide repeat domain 7A
- Located on chromosome 2p21, autosomal recessive
- Deficiency leads to severe pathology of the gut and the immune system
  - Gut disease can range from inflammatory bowel disease to multiple intestinal atresia
  - Immune system defects can range from combined immune deficiency to SCID
  - The balance of these defects determines the priorities of management
- Poor genotype-phenotype correlation, even within families
- Presents soon after birth and is almost universally fatal within the first 1-2yrs of life
- Pathogenesis is still being discovered but two pathways appear affected:
  - PI4KA-TTC7A-EFRB3 pathway
  - TTC7A-RhoA Kinase (ROCK) inhibition
  - Both pathways appear important for cytoskeletal development, cell adhesion/polarization/migration; defects lead to increased cell apoptosis
  - Impaired intestinal epithelial polarity impacts on the secretory role of the gut
    - Remains unclear if gut disease is solely intrinsic gut pathology or partially immune-mediated as well
  - Impaired adhesion of T cells and dendritic cells
- Treatment: TPN, microbial prophylaxis, immunoglobulin replacement, immunosuppression, transplant
- 7 HSCTs reported in the literature, only one survived but has recurrent multiple intestinal atresia
References


APID2016 CASE SUMMARY:  
CVID-LIKE DISEASE IN TWO SIBLINGS OF CONSANGUINEOUS PARENTAGE

Presented by Dr Mark Taylor

Two siblings of consanguineous Lebanese parentage (first cousins) with primary immunodeficiency that resembles common variable immunodeficiency (CVID) are presented. The proband is a 39 year old female who was diagnosed with CVID at age 39. She has a history of recurrent upper and lower respiratory tract and otic infections since childhood, daily watery diarrhoea and upper abdominal pain for the previous 6 years, and lethargy with 6–8 kg of unintentional weight loss, fevers, chills, and drenching night sweats in the 12 months prior to diagnosis. She suffered painless loss of all secondary molar teeth in early adolescence, and alopecia totalis at age 18, with regrowth of scalp hair around age 25 and subsequent alopecia areata of the scalp. In recent years she has experienced bilateral MCP and PIP joint arthralgia, sicca symptoms, Raynaud's phenomenon, and intermittent parotid gland swelling and tenderness. Laboratory investigations (Table 1) are significant for hypogammaglobulinaemia and absent specific IgG response to vaccination, and lymphopenia across B, CD4+, CD8+ and NK subsets with a marked B lymphopenia, reduced switched memory B cells, and expansion of CD21lo B cells, consistent with CVID Freiburg class 1a. Detailed T and B cell immunophenotyping and functional studies are in lab at present. Radiological and biopsy investigations have demonstrated sterile granulomatosus hepatomegaly, massive splenomegaly, multiple pulmonary nodules, hilar and subcarinal lymphadenopathy, with no evidence of clonal lymphoproliferation or mycobacterial disease on bone marrow or hepatic biopsy. Preliminary genetic analysis of PIK3CD exons 10, 11, 24 and CTLA4 exons 1 – 4 has demonstrated no mutations. The patient has been commenced on immunoglobulin replacement therapy and awaits whole genome sequencing.

The proband's brother is a 35 year old male with a background of obesity class III and obstructive sleep apnoea, who is currently being investigated for primary immunodeficiency. He has a history of recurrent upper and lower respiratory tract infections since childhood, including infective exacerbations of asthma that have resulted in ICU admission. He underwent incision and drainage of a perianal abscess at age 11. In adulthood, he has had two separate ICU admissions for management of sepsis of unknown source, and has suffered from recurrently infected left leg and foot ulcers, requiring extended courses of oral antibiotics annually. He has previously experienced urinary tract infections, oral candidiasis and tinea pedis. There is neither a history of chronic diarrhoea nor of clinically overt autoimmune symptoms. Laboratory investigations (Table 1) are significant for borderline hyogammaglobulinaemia, with combined IgG subclass, IgA and IgM deficiency. Pre-vaccination titres of specific IgG to pneumococcus, Haemophilus influenzae type B, diphtheria, and tetanus are either undetectable or below the protective titres for adults. Post vaccination studies are pending. Investigations are significant for lymphopenia across B, CD4+, CD8+ and NK subsets, with reduced switched memory B cells, and expansion of CD21lo B cells. He also has leukopenia and thrombocytopenia. CT chest-abdomen-pelvis in 2013 demonstrated splenomegaly, posterior mediastinal lymphadenopathy and mild left upper lobe bronchiectasis. The proband and her brother have two parents aged 62 and 69 years, and four living siblings aged 24–36, who have not yet attended for Immunology review, but who apparently do not share a history of recurrent infections. One sibling is reported to have died at age 6 years from leukaemia. There is an anecdotal history of recurrent infections within previous generations of this family.
While these two patients await whole exome sequencing, their consanguineous parentage raises the possibility of an underlying monogenic cause for their primary immunodeficiency. As of 2016, a monogenic cause has been demonstrated for approximately 2–10% of patients with CVID-like disease, and Bogaert and co-workers provide an excellent overview of monogenic causes of CVID-like diseases. Rivoisy and co-workers have studied a subgroup of 24 CVID patients with consanguineous parents from a larger group of 436 European patients with CVID. Compared to CVID patients with non-consanguineous parents, Rivoisy and co-workers demonstrated that CVID patients with consanguineous parents had significantly higher frequency of splenomegaly (62.5% vs 29%), granulomatous disease (29% vs 12%), bronchiectasis (58% vs 29%) and a higher incidence of opportunistic infections (29% vs 5%). Within a similar study group of 313 European CVID patients, Malphettes and co-workers identified a subset of 28 patients with late-onset combined immune deficiency (LOCID), which they defined by the occurrence of an opportunistic infection and/or CD4+ T cell count < 200 /µL. Compared to the remaining CVID patients, the LOCID patients more frequently belonged to consanguineous families (29% vs 8%), had a higher frequency of splenomegaly (64% vs 31%), granuloma (43% vs 10%), gastrointestinal disease (75% vs 42%), and lymphoma (29% vs 4%). The B cell, CD4+ and CD8+ T cell compartments were each significantly decreased in the LOCID compared to remaining CVID patients, with median B cell count 20 vs 102 /µL, median CD4+ T cell count 158 vs 604 /µL, and median CD8+ T cell count 221 vs 466 /µL. While the proband patient presented herein does not meet the current criteria for LOCID, as to date serial CD4+ T cell counts have been above 200 /µL and there have been no proven opportunistic infections, her overall clinical presentation and laboratory studies suggest a milder presentation of LOCID.

Table 1. Summary of laboratory findings for the patients presented.

<table>
<thead>
<tr>
<th>Subject</th>
<th>P1 (39 year old female)</th>
<th>P2 (35 year old male)</th>
</tr>
</thead>
<tbody>
<tr>
<td>IgG (g/L)</td>
<td>3.9 (6.6 – 15.6)</td>
<td>6.9 (6.6 – 15.6)</td>
</tr>
<tr>
<td>IgA (g/L)</td>
<td>&lt; 0.25 (0.75 – 3.8)</td>
<td>0.36 (0.75 – 3.8)</td>
</tr>
<tr>
<td>IgM (g/L)</td>
<td>&lt; 0.25 (0.40 – 3.1)</td>
<td>&lt; 0.10 (0.40 – 3.1)</td>
</tr>
<tr>
<td>Spec IgG pre- and post-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>vaccination pre-vaccine</td>
<td>&lt; 3.3</td>
<td>&lt; 3.3 (&gt; 39)</td>
</tr>
<tr>
<td>Pneumococcal IgG (µg/mL)</td>
<td>0.15</td>
<td>0.19 (&gt; 0.01)</td>
</tr>
<tr>
<td>Diphtheria IgG (IU/mL)</td>
<td>0.26</td>
<td>0.28 (&gt; 0.16)</td>
</tr>
<tr>
<td>Tetanus IgG (IU/mL)</td>
<td></td>
<td>0.14 (&gt; 0.16)</td>
</tr>
<tr>
<td>Hb (g/L) / platelet count (x</td>
<td>128</td>
<td>149</td>
</tr>
<tr>
<td>10^9/L)</td>
<td></td>
<td>180</td>
</tr>
<tr>
<td>Total lymphocyte count (µL)</td>
<td>440 (1000 – 4000)</td>
<td>550 (1000 – 4000)</td>
</tr>
<tr>
<td>CD3+ (µL)</td>
<td>348 (650 – 1820)</td>
<td>369 (650 – 1820)</td>
</tr>
<tr>
<td>CD3+CD4+ (µL)</td>
<td>273 (380 – 1390)</td>
<td>226 (380 – 1390)</td>
</tr>
<tr>
<td>CD3+CD8+ (µL)</td>
<td>70 (200 – 690)</td>
<td>143 (200 – 690)</td>
</tr>
<tr>
<td>CD3+CD16+CD56+ (µL)</td>
<td>70 (85 – 500)</td>
<td>82 (85 – 500)</td>
</tr>
<tr>
<td>CD19+ (µL)</td>
<td>13 (80 – 430)</td>
<td>72 (80 – 430)</td>
</tr>
<tr>
<td>CD19+CD27+ (% of CD19+)</td>
<td>7.9% (&gt; 11%)</td>
<td>9.7% (&gt; 11%)</td>
</tr>
<tr>
<td>CD19+CD27+IgD+ (% of CD19+)</td>
<td>0.08% (&gt; 0.4%)</td>
<td>0.07% (&gt; 0.4%)</td>
</tr>
<tr>
<td>CD19+CD21+ (% of CD19+)</td>
<td>54% (&lt; 20%)</td>
<td>28% (&lt; 20%)</td>
</tr>
<tr>
<td>Treg (% of CD4+ cells)</td>
<td>3.9% (4.5 – 12.3%)</td>
<td>–</td>
</tr>
<tr>
<td>Genetic analysis to date</td>
<td>PIK3CD exons 10, 11, 24: NAD</td>
<td>CTLA4 exons 1–4: NAD</td>
</tr>
<tr>
<td>------------------------</td>
<td>-------------------------------</td>
<td>----------------------</td>
</tr>
<tr>
<td></td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>Microbiology</td>
<td>Sputum: Bordetella pertussis DNA</td>
<td>H1N1 +</td>
</tr>
<tr>
<td></td>
<td>Sputum: Human metapneumovirus</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Serum: EBV DNA detected</td>
<td></td>
</tr>
</tbody>
</table>

References


APID2016 CASE SUMMARY: IPEX SYNDROME- NOT JUST A PAEDIATRIC CONDITION

Presented by Dr Grace Thompson

I described a case of a 61 yr. old gentleman diagnosed with IPEX syndrome, the oldest patient reported with the condition in the literature to date. His diagnosis was made following his grandson being diagnosed after presenting with neonatal diabetes without any other features of IPEX syndrome.

Following this diagnosis a full family history was taken and his grandfather and cousin were subsequently also diagnosed. His grandfather reported a history of severe eczema from 6 weeks of age, enteropathy from 6 months of age which was diagnosed at the time as being ulcerative colitis. Despite treatment with immunosuppression at the time he remained symptomatic requiring a colectomy at 15 yrs of age. He then remained entirely well for the following 35 yrs when at age 50 he developed chronic autoimmune haemolytic anaemia, massive splenomegaly and intestinal varices which resolved following splenectomy. His other grandson was also subsequently diagnosed at age 10 years having only isolated eczema. Further investigations revealed positive tTg antibodies and a subsequent endoscopy diagnosing coeliac disease, he also has positive IA2 antibodies but clinically has not developed diabetes.

All of our patients had normal numbers of T regulatory cells in peripheral blood. Given the mild phenotype in our patient the grandfather and in his grandsons they were discussed at TAPID and decision was made not to transplant them. They are currently not on any therapy and remain under close monitoring.

Immune dysregulation polyendocrinopathy x linked (IPEX) syndrome is a rare life threatening condition first described in 1982 in a family in which 17 males infants died in the first few years of life from a syndrome of diarrhoea, polyendocrinopathy and fatal infection. In 2000 it was linked to mutations in the FOXP3 gene which encodes the FOXP3 transcription factor which is critical for T regulatory cell function. Loss of T regulatory cell function in these patients leads to the cardinal features of enterophy, eczema and endocrinopathy often diabetes. Patients may also have other autoimmune phenomena and often have an increased susceptibility to infections thought to be due to impaired barrier function of the skin and gut and immunosuppressive therapy. There have been 148 cases described in the literature to date of which more than half have been described in the last few years. This condition is thought to be universally fatal within the first few years of life without treatment. The oldest case in the literature reported to date was from the original paper describing IPEX. He had enteropathy, failure to thrive; splenomegaly, lymphadenopathy and died of diabetes related complications at 30 years of age.

There is significant heterogeneity in the clinical phenotype of cases reported to date and there is no clear correlation between genotype, protein expression, functional studies and the phenotype. This is highlighted in our family who display a varied phenotypes. The mutation R347H detected in this family resides in the forkhead binding domain which is where the majority of mutations are located. This is the region which interacts with IL2 promoter and other target genes essential for most aspects of T reg function. There are 6 other cases reported in the literature of this specific mutation which again appear to have a varied phenotype. Seidel et al described two brothers who both had the R347H, one which died at 9 months of age with severe enteropathy and eczema, the other remains well and alive at 29 yrs of age off all immunosuppression.

Bone marrow transplantation is the only curative treatment and early transplantation is generally thought of as the treatment of choice. Of those that have been transplanted there is about a 20% mortality rate, although there are increasing reports of success with reduced intensity regimens. Disease remission is also reported with mixed donor chimerism suggesting that wild type T regulatory cells may be sufficient to control disease and hence gene therapy may be a therapeutic option in the future. Immunosuppressive therapy has varied outcomes, the most commonly used
agents being prednisolone with cyclosporine or tacrolimus but there have been increasing reports in the literature of success with sirolimus.

This case illustrates the clinical heterogeneity in IPEX syndrome which is being increasingly recognized in the literature. This case should increase our awareness of IPEX syndrome occurring in milder phenotypes. Overall the natural history of this condition in milder phenotypes remains unknown and as a result the ideal management is unclear. Management is particularly challenging as there is an absence any reliable biomarkers to use to predict prognosis and help guide treatment decision.


