**The effect of biologic medication on patients with nasal polyp eosinophilia**

**Sponsor: Investigator initiated**

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**Summary**

|  |  |
| --- | --- |
| **Study Title** | The effect of biologic medication on patients with nasal polyp eosinophilia |
|  |  |
| **Objectives** | Primary:  1. Assess the tissue histopathological changes in response to mepolizumab and/or dupilumab therapy in patients with nasal polyp eosinophilia  Secondary:  2. Assess the effectiveness of Mepolizumab and/or Dupilumab in nasal polyp eosinophilia based on patient reported outcome measures, blood eosinophilia and recorded endoscopy  3. Assess changes in serum/blood markers in response to mepolizumab and/or dupilumab therapy in patients with nasal polyp eosinophilia  4. Assess the relationship between serum/blood markers and tissue markers  5. Define changes in IL-4, IL-5, IL-13 -related eosinophil, mast cell biology and mucosal type 2 inflammatory process, in response to mepolizumab and/or dupilumab therapy, using GeoMxTM multiplex digital spatial profiling of tissue samples  6. Assess factors that may contribute to treatment failure / non-response |
| **Study design** | Phase 2 clinical trial |
| **Planned sample size** | 25 |
| **Selection criteria** | Adult patients with nasal polyp eosinophilia whose condition is not fully managed by current standard of care Not currently receiving Dupilumab, Mepolizumab or Benralizumab treatment.  Patients who do not meet the PBS criteria for severe lower airway disease  Body weight: A minimum body weight >=40 kilograms (kg) at Visit 1  Gender: Male or female.  Informed consent: Capable of giving signed written informed consent and willingness to participate to and comply with the study |
| **Study procedures** | At the initial baseline visit, prior to initiation of Mepolizumab and/or Dupilumab therapy, the following set of data will be recorded: demographic data, tissue biopsy of mucosa and polyp, serum sample, smell test (threshold and identification components of the commercially available Sniffin Sticks Kit), patient reported outcome scores including SNOT-22, ACQ, smell questionnaire, FeNO and recorded endoscopy.  Patients will then receive their first course of Mepolizumab and/or Dupilumab therapy. They will then return for routine visits for ongoing Mepolizumab and/or Dupilumab provided at the clinic. There will be a tissue biopsy undertaken at every 2nd visit following initiation of therapy. In addition, there will be 2 weekly assessment of the following outcomes: blood sample, SNOT-22, ACQ, FeNo, recorded endoscopy. At visit 4, smell assessment will also be included.  At the end of the 6 month treatment period, an overall assessment of treatment response / non-response would be assessed, including repeating the smell assessment.  Patients will return 3 months following completion of therapy to have a post completion tissue biopsy and serum sample. |
| **Statistical considerations** | As there is limited published data regarding the effect of Mepolizumab and/or Dupilumab in nasal polyp eosinophilia, sample size calculation is not available. This study is a pilot study / phase 2 clinical trial. |
| **Study duration** | 5 years |

1. BACKGROUND 7

1.1. Disease Background\* 7

1.2. Rationale for Performing the Study\* 7

2. STUDY OBJECTIVES\* 8

2.1. Primary Objective\* 8

2.2. Secondary objectives 8

3. STUDY Design\* 8

3.1. Design\* 8

3.2. Study Groups 9

3.3. number of participants\* 9

3.4. number of SITES 9

3.5. duration 9

4. Participant section 9

4.1. Inclusion Criteria\* 9

4.2. Exclusion Criteria\* 9

5. STUDY Outline\* 9

5.1. Study Flow Chart 9

5.2. Investigation plan\* 10

5.3. Study Procedure Risks\* 12

5.4. Recruitment and Screening\* 13

5.5. Informed Consent Process\* 13

5.6. Enrolment Procedure\* 13

5.7. Randomisation Procedure 13

6. TISSUE CoLLECTION/BIOBANKING 13

7. SAFETY\* 13

7.1. Adverse Event Reporting\* 13

7.2. Serious Adverse Event Reporting 14

7.3. Data Safety and Monitoring Board 14

7.4. Early Termination 14

8. BLINDING AND UNBLINDING 14

9. OUTCOMES AND FUTURE PLANS 14

10. STATISTICAL CONSIDERATIONS\* 14

11. CONFIDENTIALITY AND STORAGE AND ARCHIVING OF STUDY DOCUMENTS\* 15

12. Other study documents 15

13. RESOURCES 15

14. REFERENCES\* 15

1. BACKGROUND

## Disease Background\*

Eosinophilic chronic rhinosinusitis (eCRS) is an emerging classification of chronic rhinosinusitis (CRS), thought to more accurately reflect the underlying pathophysiology. eCRS is thought to be a disease of chronic sinus inflammation rather than infection, linked with skewed T-helper 2 (TH2) allergic responses, driven by eosinophilic inflammation and a dysregulated sinus mucosa (Chin & Harvey, 2013). eCRS is diagnosed by histopathological assessment of eosinophilic infiltration within sinus tissue. There is wide variation within the literature and no consensus currently exists regarding the cut-off for diagnosis of eCRS however, an absolute eosinophil count of >10 per high power field (HPF) is generally associated with poorer outcomes and overall prognosis.

## Rationale for Performing the Study\*

Mepolizumab and/or dupilumab is an anti-IL-4, IL-5, IL-13 -monoclonal antibody, currently used for the management of severe eosinophilic asthma and has been acclaimed as a potential treatment in the management of eCRS. The standard cut-off applied for Australian government subsidised provision of mepolizumab and dupilumab for eosinophilic asthma is a blood eosinophil count of 0.3x109/L.

Our anecdotal experience with mepolizumab and/or dupilumab in a well defined group of chronic rhinosinusitis patients, with eosinophilic inflammation, has been very positive. We believe mepolizumab and/or dupilumab has been successful in this population due to their well defined mucosal eosinophilia, that we characterize as part of our broader clinical research program (Snidvongs et al. 2012).

We don’t rely on serum eosinophil levels as a monitoring tool for sinus patients (as is done in mepolizumab and/or dupilumab for asthma) but instead use nasendoscopy. The nasal endoscopy of patients, who have had prior surgery, allows direct examination of the sinus cavity and mucosa. It is very obvious when therapy is influencing the disease, such as in corticosteroid therapy, as the mucosa often normalizes during such therapy. Such changes have been observed in patients on mepolizumab and dupilumab therapy.

Patients with eCRS very often suffer from smell loss, as a result of increased sinonasal concentration of eosinophils which mediate damage to olfactory epithelium and neurons (Chung et al., 2015; Wu et al., 2018; Zhang et al., 2019). Treatment with mepolizumab and/or dupilumab has been suggested to improve patient reported sense of smell (Bachert et al., 2017), however objective data has not been reported. Subjective assessments do not always correlate with functional olfaction tests, thus, the true impact of mepolizumab and/or dupilumab on smell function in eCRS is yet to be determined (Langstaff et al., 2019).

One of the challenges of defining successful mepolizumab and/or dupilumab therapy, will be demonstrating that respiratory mucosal eosinophilic inflammation decreases with treatment, and not simply the serum eosinophil level. Blocking the IL-4, IL-5, IL-13 -receptor also inhibits mast cell activity. Few studies have detailed the exact mechanism of action for Mepolizumab and/or Dupilumab on IL-4, IL-5, IL-13 -induced eosinophil biology in eCRS. There are many reasons why some patients may not have an immediate clinical benefit: intercurrent infection, mucosal remodelling and fixed changes, including nasal polyps in the upper airway. Remodelling changes have been demonstrated in chronic rhinosinusitis (Barham et al., 2015). Furthermore, it is uncertain whether Mepolizumab and/or Dupilumab treatment exerts inhibition across mucosal inflammatory processes beyond IL-4, IL-5, IL-13 -related eosinophil biology, such as the recently described specialised IL-4, IL-5, IL-13 -+ expressing CD4+ T-helper 2 subset, that has enhanced effector function (Yin et al., 2017), that may explain the inter-individual differences in Mepolizumab and/or Dupilumab responses (Roufosse, 2018). GeoMx™ digital spatial profiling allows for the simultaneous analysis in-situ of up to 1600 immune genes and 52 immune protein targets in their tissue-based morphological context from formalin fixed paraffin embedded samples to provide comprehensive information on inflammatory cells, pathways and molecules mediating biological activity (Hoang, et al., 2019; Merritt et al., 2019). GeoMx™ profiling enables highly multiplexed characterisation of the pathobiology of disease and biological response to treatment. GeoMx™ multiplex spatial profiling has been used to identify immune correlates of response to immunotherapy in high risk resectable melanoma (Amaria et al., 2018) and to determine the inflammatory mediators of checkpoint inhibitor-associated immune adverse events (Johnson et al., 2019).

Thus, this study would provide evidence of the direct tissue response to mepolizumab and/or dupilumab, including changes in IL4, IL-5, IL-13 related eosinophil and mast cell biology, and mucosal type 2 inflammatory processes. It will also provide a relationship between tissue and serum/blood marker levels and might provide prognostic information to responders to therapy, the effects of mepolizumab and/or dupilumab on oncogenes, on immune cells and pathways associated with immune-related adverse events. This could reveal novel data of the mechanisms of action of Mepolizumab and/or Dupilumab and help guide the application of this biological in type 2 inflammatory diseases.

# STUDY OBJECTIVES\*

## Primary Objective\*

To identify and assess the tissue histopathological changes in response to mepolizumab and/or dupilumab therapy in patients with nasal polyp eosinophilia

## Secondary objectives

Assess the effectiveness of Mepolizumab and/or Dupilumab in eosinophilic nasal polyp eosinophilia

Assess changes in serum/blood markers in response to mepolizumab and/or dupilumab therapy in patients with nasal polyp eosinophilia

Assess the relationship between serum/blood markers and tissue markers

Assess factors that may contribute to treatment failure / non-response to Mepolizumab and/or Dupilumab

Assess the effect of Mepolizumab and/or Dupilumab on IL4, IL-5, IL-13 immune gene expression signalling in eosinophil and mast cell biology, IL4, IL-5, IL-13 dependent immune protein abundance in eosinophil biology and type 2 inflammation.

Identify a preliminary population who may benefit from Mepolizumab and/or Dupilumab therapy

# STUDY Design\*

## Design\*

Phase 2 clinical trial with single group of non-blinded patients with open label therapy. At the initial baseline visit, prior to initiation of Mepolizumab and Dupilumab therapy, the following set of data will be recorded: demographic data, tissue biopsy of mucosa and polyp, serum sample, smell test (threshold and identification components of the commercially available Sniffin Sticks Kit) patient reported outcome scores including SNOT-22, ACQ and smell questionnaire, FeNO and recorded endoscopy.

Patients will then receive their first course of Mepolizumab and/or Dupilumab therapy. They will then return every 4 weeks for routine visit for ongoing Mepolizumab and/or Dupilumab provided at the clinic. There will be a tissue biopsy undertaken at 6 months and potentially every 2nd visit following initiation of therapy. In addition, there will be 4 weekly assessment of the following outcomes: blood sample, SNOT-22, ACQ, FeNo, recorded endoscopy.

At the end of the 6 month treatment period, an overall assessment of treatment response / non-response would be assessed.

Patients will return 3 months following completion of therapy to have a post completion tissue biopsy and serum sample.

Each biopsy sample will be assessed and reported using a standardized synoptic histopathology profile and the resulting formalin-fixed, paraffin-embedded sections will be further analysed using Nanostring GeoMx™ multiplex digital spatial profiling system and a mixOmics toolkit.

## Study Groups

Single group: 25 patients. Non-randomised trial. Open (masking not used)

## number of participants\*

25 patients

## number of SITES

Sydney Ear, Nose and Throat Clinic (67 Burton Street, Darlinghurst)

- Expected number of participants: 25

## duration

Feb 2018 – Aug 2026. Patients will undergo 6 month treatment duration. Expected recruitment phase will be 18 months depending on ability to recruit patients to the study.

# Participant section

## Inclusion Criteria\*

* Adult patients with nasal polyp eosinophilia whose condition is not fully managed by current standard of care
* Not currently receiving Mepolizumab, Dupilumab or Benralizumab treatment
* Patients who do not meet the PBS criteria for severe lower airway disease
* Body weight: A minimum body weight >=40 kilograms (kg) at Visit 1
* Gender: Male or female.
* Informed consent: Capable of giving signed written informed consent and willingness to participate to and comply with the study
* Age >18 Years

## Exclusion Criteria\*

* Subjects with known hypersensitivity to mepolizumab and/or dupilumab
* Subjects with other conditions that could lead to elevated eosinophils such as Hypereosinophilic Syndromes, including Churg-Strauss Syndrome, or Eosinophilic Esophagitis.
* Subjects with known immunodeficiency
* Subjects with cystic fibrosis
* Pregnant subjects or subjects currently lactating as the effect on human pregnancy is unknown.

# STUDY Outline\*

## Study Flow Chart

Every patient of the investigators, who fulfils the inclusion criteria, and is undergoing assessment of chronic rhinosinusitis with nasal polyp eosinophilia, will be assessed for eligibility. Those who are eligible, willing to participate, and can provide written informed consent will be enrolled.

## Investigation plan 1\*

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Visit number | V1 | V2 | V3 | V4 | V5 | V6 | V7 | V8 | V9 | V10 | V11 | V12 | V13 | Final Study Visit |
| Number of weeks | 0 | 2 | 4 | 6 | 8 | 10 | 12 | 14 | 16 | 18 | 20 | 22 | 24 | 36 |
| Informed Consent | ü |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Inclusion / Exclusion  criteria | ü |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Demographic Data | ü |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Questionnaires | ü |  | ü |  | ü |  | ü |  | ü |  | ü |  | ü | ü |
| FENO | ü |  | ü |  | ü |  | ü |  | ü |  | ü |  | ü | ü |
| Endoscopy | ü |  | ü |  | ü |  | ü |  | ü |  | ü |  | ü | ü |
| Blood/serum sample | ü |  | ü |  | ü |  | ü |  | ü |  | ü |  | ü | ü |
| Tissue biopsy | ü |  | ü |  | ü |  |  |  | ü |  |  |  | ü | ü |
| Adverse Event & Serious Adverse  Event Assessment | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü |
| Administration of  dupilumab | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü | ü |  |  |
| Overall assessment  of treatment response |  |  |  |  |  |  |  |  |  |  |  |  | ü | ü |

**\***Dupilumab

## Investigation plan 2#

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Interventions | Enrolment Visit  (Visit 1) | Visit 2  (4 weeks) | Visit 3  (8 weeks) | Visit 4 (12 weeks) | Visit 5  (16 weeks) | Visit 6  (20 weeks) | Visit 7  (24 weeks) | Final Study Visit |
| Informed Consent | ✓ |  |  |  |  |  |  |  |
| Inclusion / Exclusion criteria | ✓ |  |  |  |  |  |  |  |
| Demographic Data | ✓ |  |  |  |  |  |  |  |
| Questionnaires | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ |
| FENO | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ |
| Smell Assessment | ✓ |  |  | ✓ |  |  | ✓ |  |
| Endoscopy | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ |
| Blood/serum sample | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ |
| Tissue biopsy | ✓ | ✓ | ✓ |  | ✓ |  | ✓ | ✓ |
| Adverse Event & Serious Adverse Event Assessment | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ |
| Administration of Mepolizumab | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ |  |  |
| Overall assessment of treatment response |  |  |  |  |  |  | ✓ | ✓ |

#Mepolizumab

**Data collection**

Blood/serum samples will be obtained at a recognised pathology provider (SydPath). All other data collection / procedures will occur at the Sydney ENT clinic. Data will be collected and stored in a de-identified excel sheet which will be password protected on the co-investigator’s computer.

* Patient demographic data collection: age, gender, presence of nasal polyps, past history of asthma, smoking status
* Blood/serum sample collection: venepuncture by accredited phlebotomists at a pathology provider
  + Eosinophil count, full blood count
* Recorded nasal endoscopy: a rigid nasoendoscope will be used for endoscopy following topical local anaesthetic spray.
* Tissue biopsy: performed under local anaesthetic spray and endoscopic guidance using biopsy forceps by trained individuals. The tissue will be sent to SydPath (pathology provider) for histopathological assessment and reported according to a synoptic report
* Patient questionnaires: Sino-Nasal Outcome Test (SNOT-22), Asthma Controlled Questionnaire (ACQ) and smell questionnaire (including the Individual Importance of Olfaction Questionnaire and Questionnaire for Olfactory Disorders) are validated surveys of patient reported outcome measures.
* Levels of nitric oxide were measured by NIOX VERO machine.
* Patient olfactory performance: as measured by the threshold and identification components of the Sniffin Sticks Kit (Burghart, Germany)

## Study Procedure Risks\*

Mepolizumab and Dupilumab

* Trade name: Nucala and Dupixent
* Manufacturer: GlaxoSmithKline and Sanofi
* Supplier of drug/device: manufacturer and pharmacy
* Approved therapeutic indication: severe asthma
* Believed mode of action: Mepolizumab and dupilumab is a humanised monoclonal antibody (IgG1, kappa), which targets human IL4, IL-5, IL-13 -with high affinity and specificity IL4, IL-5, IL-13 is the major cytokine responsible for the growth and differentiation, recruitment, activation and survival of eosinophils. Mepolizumab and dupilumab inhibits the bioactivity of IL4, IL-5, IL-13 with nanomolar potency by blocking the binding of IL4, IL-5, IL-13 to the alpha chain of the IL4, IL-5, IL-13 receptor complex expressed on the eosinophil cell surface, thereby IL4, IL-5, IL-13 signalling and reducing the production and survival of eosinophils.
* Dosage regimen: Mepolizumab - 100mg subcutaneous injection once every 4 weeks, Dupilumab - 300mg subcutaneous injection once every 2 weeks
* Mode of excretion: Mepolizumab and dupilumab is a humanised IgG1 monoclonal antibody degraded by proteolytic enzymes which are widely distributed in the body and not restricted to hepatic tissue.
* Known adverse events:
  + Side effects 1 in 10: Headache.
  + Side effects 1 in 100: injection site reaction (pain, skin redness, swelling, itching, and burning sensation of the skin near where the injection was given), Back Pain, Pharygitis (sore throat), Lower respiratory tract infection (congestion, cough), Nasal congestion (stuffy nose), Upper absdominal pain (stomach pain or discomfort in the upper area of the stomach), Eczema (itchy red patches on the skin), Urinary tract infection (blood in urination, painful and frequent urination, fever, pain in lower back)and Fever (high temperature).
  + Side effects 1 in 10,000: Hypersensitivity (allergic reaction) including anaphylaxis (an allergic reaction that can be life threatening).
* Known contra-indications or warnings
  + Do not use if allergic to mepolizumab and dupilumab or any of the other ingredients of the medicine (dibasic sodium phosphate heptahydrate, sucrose, polysorbate 80)
* Blood serum collection
  + Pain at venepuncture site during collection
  + Haematoma causing discomfort and pain
* Sinus biopsy
  + Short term minimal local nasal bleeding following biopsy
  + Potential discomfort
  + Appropriate medical care will be given in the event of nasal bleeding.

## Recruitment and Screening\*

Patients attending the investigator’s clinic at Sydney ENT Clinic will be recruited. Initial contact will be made by the investigators. Patients will be screened for the study based on their medical histories and clinical evidence of chronic rhinosinusitis with nasal polyp eosinophilia.

## Informed Consent Process\*

Prospective participants will be invited to read an information sheet in simple, non-technical language. This form incorporates the patient consent. Potential subjects will have up to two weeks to decide whether to take part in the study. Potential participants will have adequate opportunity to discuss the proposed trial with friends/relatives. If a patient is unable to give informed consent because of age, mental illness, dementia, communication difficulties or other reasons, they will be excluded from the study.

## Enrolment Procedure\*

Each participant will be enrolled into the study after screening has verified that the participant meets all the inclusion criteria and none of the exclusion criteria. Upon enrolment, the informed consent process will be completed. The participant will receive a study enrolment number and this will be documented in the participant’s medical record and on all study documents.

## Randomisation Procedure

N/A

# TISSUE CoLLECTION/BIOBANKING

Biopsy tissue will be collected during clinic appointments. Blood samples will be taken at accredited pathology services. Blood samples and tissue biopsies will be processed by routine pathology services (SydPath). Tissue disposal/destruction will be as per routine pathology service protocol. Tissue will be re-identifiable.

# SAFETY\*

Adverse events are expected to be minimal. An adverse event is defined here as any untoward medical occurrence in a participant which does not necessarily have a causal relationship with the study treatment. An adverse even can therefore be any unfavourable or unintended sign, symptom or condition, and/or an observation that may or may not be related to the study treatment.

## Adverse Event Reporting\*

Under the guidelines of the St Vincent’s Hospital Human Research Ethics Committee, adverse events will be reported to the Committee according to the Adverse Event Reporting Policy. Adverse events related to the administration of the Mepolizumab and Dupilumab will be reported to the Therapeutic Goods Administration and St Vincent’s Hospital Human Research Ethics Committee in accordance with the requirements of the National Health and Medical Research Council, Position Statement “*Safety monitoring and reporting in clinical trials involving therapeutic goods (September 2016).*

**Adverse event**

An adverse event for medicines is also referred to as an adverse experience, any untoward medical occurrence in a patient or clinical investigation participant administered a pharmaceutical product and which does not necessarily have a causal relationship with this treatment.

An adverse event can therefore be any unfavourable and unintended sign, symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

## Serious Adverse Event Reporting

**Serious adverse event (SAE)**:

**For medicines, also referred to as serious adverse drug reaction**, any untoward medical occurrence that at any dose:

* results in death;
* is life-threatening;
* requires in-patient hospitalisation or prolongation of existing hospitalisation;
* results in persistent or significant disability/incapacity;
* is a congenital anomaly/birth defect; or
* is a medically important event or reaction.

NOTE: The term 'life-threatening' in the definition of 'serious' refers to an event in which the patient was at risk of death at the time of the event; it does not refer to an event/reaction which hypothetically might have caused death if it were more severe.

## Data Safety and Monitoring Board

Co-investigator Raquel Alvarado has been appointed to monitor the data collection for this study. All patients will be monitored for eligibility and consent. All patients experiencing any adverse events or serious adverse event will be monitored.

## Early Termination

Should early termination of the study be necessary, for example in the case of a serious adverse event suggestive of an unacceptable benefit risk profile, the principal investigator will inform patients and the HREC.

# BLINDING AND UNBLINDING

Unblinded study

# OUTCOMES AND FUTURE PLANS

The plan will be for publication of project outcomes and presentation at conferences. The research will be available within the medical literature as well as in the form of a student thesis. The results from this research may form the basis of other research projects.

# STATISTICAL CONSIDERATIONS\*

As there is limited published data regarding the effect of Mepolizumab and/or Dupilumab in nasal polyp eosinophilia, sample size calculation is not available. This study is a pilot study / phase 2 clinical trial. Paired T-testing would be used to identify changes following Mepolizumab and Dupilumab therapy.

Comparative assessment of responders vs. non-responders. Assessing tissue and serum responses to changes in endoscopy and symptom scores.

All immune gene expression and immune protein data analysis will be conducted using the Advanced Analysis Module in nSolver 4.0 (Nanostring, Seattle, USA) and with customised R scripts in the R Statistical Computing environment for differential gene expression, pathway and pathview plots, and immune cell profiling between pre- and post-treatment. Genes and proteins with counts below 50 in greater than 75% of samples will be excluded from analysis. Immune cell scores will be determined using cell specific gene and protein expression from The Cancer Genome Atlas, as previously described (Danaher et al., 2017; West et al., 2019). Data will be visualised with volcano plots and heatmaps from unsupervised hierarchical clustering to examine for distinct patterns of expressions.

To assess the impact of immune gene and protein expression on the primary and secondary clinical and haematological outcomes a multivariate approach using methods implemented in the mixOmics toolkit will be adopted. The DIABLO (Data Integration Analysis for Biomarker discovery using Latent variable approaches for ‘Omics studies) method will be used to predict intervention responses to a biomarker.

# CONFIDENTIALITY AND STORAGE AND ARCHIVING OF STUDY DOCUMENTS\*

Consent forms will be kept in the patient files on the Sydney Ear Nose and Throat Clinic database, which is kept on a secure server with restricted access. The data custodian is Professor Richard Harvey. Patient data will be de-identified. Only the chief investigator will have access to the full data set. Data collected will be stored as an encrypted file on the co-investigator’s computer.

In accordance with the NHMRC National Statement, NSW Supplement (2009), data will be retained for 15 years, at which time it will be securely destroyed.

# Other study documents

* Consent form
* eCTN study details form

# RESOURCES

Funding/support being sought: Rhinology and Skull Base Research Group Trust Fund

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