BMJ Open

Prolonged versus single dose in penicillin oral challenge testing – Protocols for a pilot and definitive randomised controlled trial (PROSPECTOR Studies)

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2024-094712
Article Type:	Protocol
Date Submitted by the Author:	07-Oct-2024
Complete List of Authors:	Ng, Irvin; Austin Health, Infectious Diseases and Immnology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases James, Fiona; Austin Health, Department of Infectious Diseases; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases Copaescu, Ana; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases Vogrin, Sara; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases Mitri, Elise; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases; National Allergy Centre of Excellence Rose , Morgan; Austin Health, Department of Infectious Diseases; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases; Peter MacCallum Cancer Centre Sullivan, Richard; St George Hospital, Infectious Diseases Lane, Michael; Royal Brisbane and Women's Hospital Legg, Amy; Royal Brisbane and Women's Hospital, Pharmacy Department Godsell, Jack; Austin Health, Infectious Diseases and Immunology; The Royal Melbourne Hospital, Allergy and Immunology Fernando, Suran; Royal North Shore Hospital, Clinical Immunology and Allergy Garvey, Lene Heise; University of Copenhagen; Herlev and Gentofte Hospital, Allergy Clinic, Department of Dermatology and Allergy Sabato, Vito; University of Antwerp, Department of Immunology, Allergology and Rheumatology; University Hospital Antwerp, Department of Immunology, Allergology and Rheumatology Li, Philip; Hong Kong University, Division of Rheumatology and Clinical Immunology, Department of Medicine Peter, Jonathan; University of Cape Town, Division of Allergy and Immunology, Department of Medicine Trubiano, Jason ; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases; National Allergy Centre of Excellence
Keywords:	Clinical Trial, Antibiotics < Anti-Bacterial Agents, PUBLIC HEALTH, Protocols & guidelines < HEALTH SERVICES ADMINISTRATION & MANAGEMENT, IMMUNOLOGY

Title: Prolonged versus single dose in penicillin oral challenge testing – Protocols for a pilot and definitive randomised controlled trial (PROSPECTOR Studies)

Authors: Ng I[1,2], James F[1,2], Copaescu A[1,2], Vogrin S[1,2], Mitri E[1,2,3], Rose MT[1,2,4], Sullivan RP[5], Lane MJ[6], Legg A[7], Godsell J[1,8], Fernando S[9], Garvey LH[10], Sabato V[11], Li PH[12], Peter J[13], Trubiano JA[1,2,3].

Affiliations:

1. Centre for Antibiotic Allergy and Research, Department of Infectious Diseases and Immunology, Austin Health, Victoria, Australia
2. Department of Infectious Diseases, The Peter Doherty Institute for Infection and Immunity, University of Melbourne, Victoria, Australia.
3. National Allergy Centre of Excellence (NACE), Parkville, Victoria, Australia.
4. National Centre for Infection in Cancer, Department of Infectious Diseases, Peter MacCallum Cancer Centre, Australia
5. Department of Infectious Diseases, St George Hospital, School of Clinical Medicine, UNSW Medicine and Health, Kogarah, NSW, Australia
6. Royal Brisbane and Women's Hospital, University of Queensland, Queensland, Australia
7. Department of Pharmacy, Royal Brisbane and Women's Hospital, Brisbane, Queensland, Australia
8. Department of Allergy and Immunology, Royal Melbourne Hospital, University of Melbourne, Parkville, Australia
9. Department of Clinical Immunology and Allergy, Royal North Shore Hospital, Sydney, Australia
10. Allergy Clinic, Department of Dermatology and Allergy, Herlev and Gentofte Hospital and University of Copenhagen, Copenhagen, Denmark.
11. Department of Immunology, Allergology and Rheumatology, University of Antwerp, Antwerp University Hospital, Antwerpen, Belgium; Allergology Unit, AZ Jan Palfijn, Ghent, Belgium; Infla-Med Centre of Excellence, University of Antwerp, Antwerpen, Belgium
12. Division of Rheumatology and Clinical Immunology, Department of Medicine, Queen Mary Hospital, The University of Hong Kong, Hong Kong SAR, China.

13. Division of Allergy and Clinical Immunology, Department of Medicine, University of Cape Town, Cape Town, South Africa

Trial Sponsor:

Prof Jason Trubiano

Centre for Antibiotic Allergy and Research

Department of Infectious Diseases and Immunology, Austin Health

145 Studley Road, Heidelberg VIC 3121

AUSTRALIA

Funding: The work is supported by an NHMRC Emerging Leadership Fellowship (EL2) awarded to Professor Jason Trubiano and Austin Health

Corresponding author:

Prof Jason Trubiano

Department of Infectious Diseases and Immunology, Austin Health

P: +61 3 94966676

E: jason.trubiano@austin.org.au

Author contributions:

JAT and SV planned the study design and wrote the study protocols.

Ethics submission: IN and FJ

Patient recruitment: JAT, AC, EM, RMT, RPS, MJL, AL, JG, SF, LHG, VS, PHL and JP

Statistical design and analysis: SV

All authors reviewed the protocol and this manuscript.

Protocol version:

PROSPECTOR 1 Protocol: Version 7 dated 22nd August 2023

PROSPECTOR 2 Protocol: Version 1.1 dated 28th August 2024

Word count: 2983

ABSTRACT

Introduction: Penicillin allergy labels (PALs) are reported in 1 in 10 hospitalized patients globally and associated with inferior patient, hospital and microbiological outcomes, however the majority are incorrect and should be removed. Direct oral penicillin challenge has been demonstrated to be a safe and effective method for the removal of PALs. However, the question of whether a single dose is sufficient to ascertain true allergy status remains unanswered, with some studies suggesting that extended challenges of 3 or more days are superior for the exclusion of delayed immune reactions. The aim of the PROSPECTOR studies is to determine the feasibility (PROSPECTOR 1) of a definitive trial (PROSPECTOR 2) to evaluate the safety and effectiveness of prolonged oral challenge (i.e. 5-day) versus single dose oral challenge in patients with a delayed or unknown penicillin allergy phenotype (PROSPECTOR-2).

Methods and analysis: A pair of double-blind two-arm parallel placebo-controlled trials will be undertaken - **PRO**longed versus **S**ingle dose in **PE**nicillin oral **C**hallenge **T**esting double-blind parallel group randomised placebo-c**O**ntrolled t**R**ial (PROSPECTOR Studies). Patients with a reported delayed or unknown timing penicillin allergy that have passed a supervised single dose oral amoxicillin challenge (with or without prior skin testing/single or split dose) will be recruited. Informed patient consent will be granted for sites to recruit patients and collect routine clinical data. PROSPECTOR-1 will assess the safety and feasibility of a placebo-controlled trial for single dose amoxicillin challenge versus 5-day prolonged oral challenge. PROSPECTOR-2 will assess the superiority of 5-day prolonged oral challenge compared with single dose amoxicillin challenge in excluding a delayed immune reaction. PROSPECTOR-2 will commence immediately post completion of PROSPECTOR-1 in a vanguard design, with adjustments to the projected sample size for superiority made following completion of PROSPECTOR-1. PROSPECTOR-2 will commence recruitment immediately following closure of PROSPECTOR-1, however data from each trial will be analysed separately.

Ethics and dissemination: These studies were reviewed and approved by the Austin Health Human Research Ethics Committee (PROSPECTOR-1: HREC/99740/Austin-2023 & PROSPECTOR-2: HREC/109785/Austin-2024). Results will be published in peer-reviewed journals and presented at relevant conferences.

Registration: PROSPECTOR-1: ACTRN12623001242617 & PROSPECTOR-2: ACTRN12624001107516

Keywords: penicillin allergy, beta-lactam allergy; penicillin provocation, penicillin challenge, drug allergy, antimicrobial stewardship

Strengths and limitations of this study:

1. These studies are among the first double-blind randomised placebo-controlled trials in antibiotic allergy investigation
2. The pilot phase, randomised experimental design and recruitment of patients from existing inpatient or outpatient settings will minimise the opportunity for selection bias
3. The definitive trial is international and multicentre, allowing for increased sample heterogeneity and generalisability of results
4. The pilot study does not aim to explore the ideal number of days for prolonged challenge in eliciting true delayed allergy, so conclusions will be limited to comparison with a prolonged 5-day challenge while there is still variability in practice globally
5. A twice-daily 500mg dose of penicillin is set as the intervention, however variability in preferred dosage remains in prolonged challenge practice globally

INTRODUCTION

Background and rationale

Penicillin allergy labels (PALs) are commonly documented in patient electronic medical records (EMRs) (1). At the higher end, the prevalence has been estimated at 10% for hospitalised Australians (2) and 9.9% of inpatients in Montreal, Canada (3). A Danish study found 5% of hospital inpatients carried a PAL (4), while at the lower end the prevalence is estimated at 3.2% in hospital inpatients in South Africa (5) and 2% for all beta-lactam allergy in Hong Kong patients (6). This figure may be even higher among vulnerable patients such as the immunocompromised (7), geriatric and rheumatology populations (8, 9). Those patients that carry a PAL are more likely to receive an inappropriate antibiotic, suffer a hospital-associated adverse event and acquire a multidrug resistant organism (10-13). PALs are also associated with increased hospital length-of-stay (LOS), higher readmission rates, increased hospital costs and mortality rates (10, 14, 15). At a public health level, they are associated with inappropriate prescribing and antimicrobial resistance (1, 16, 17).

Despite their omnipresence, the majority of PALs are assessed as "low-risk" and can be safely removed by penicillin allergy testing (18-23). Oral penicillin challenge with or without preceding skin testing is considered the gold standard for delabelling (24), however clinical equipoise remains regarding the superiority of single dose or prolonged (i.e. multiple-day dosing) oral challenge for patients that report a delayed or unknown timing penicillin allergy phenotype. The current Drug Allergy Practice Parameters recommend "against the routine use of multiple-day challenges in the evaluation of penicillin allergy", providing a "strong recommendation" but with "low certainty of evidence" (25). The European guidelines reviewed the literature of over 6,484 patients, demonstrating a 2.3% positive rate following the initial challenge and 5.5% during the varied prolonged challenges. They concluded there is no consensus on a preferred procedure and could not provide a recommendation for or against prolonged oral challenge (26).

Results from a mixture of European observational and retrospective studies suggest that prolonged challenges ranging from 3 to 10 days may be superior to single dose challenges at eliciting delayed

immune reactions. However, the reported prevalence of delayed reactions is highly variable (5-12% of patients), and many were reliant on patient self-reporting (27-34). In a recent retrospective single-centre Danish study of 3,179 low-risk patients, 2.6% were positive on day 1 of challenge and 7.2% after day 1 (35). This contrasts with the North American experience, where prolonged challenges have been associated with low rates of delayed reactions (0-1.8%) (36-39). A paediatric study demonstrated that delayed reactions may occur <7 days following a single challenge (40). This gap between guideline recommendations and evidence, and differing results across geographical regions, highlights the need for robust evidence to inform practice with clinical certainty.

Randomised controlled trials (RCTs) have not routinely been used to answer questions regarding best practice in penicillin allergy research, despite their ability to provide high-quality evidence. The PROSPECTOR (**PRO**longed versus **S**ingle dose in **PE**nicillin oral **C**hallenge **T**esting - Double-blind parallel-group randomised placebo-c**O**ntrolled t**R**ial) studies will utilise a double-blind, parallel-group, placebo-controlled RCT study design. PROSPECTOR-1 is an external pilot trial which will assess the feasibility of conducting a blinded placebo-controlled trial of single dose versus prolonged dose oral challenge in patients with a documented or reported PAL. It will also inform the sample size of a definitive full-scale trial which will follow directly on from PROSPECTOR-1, with adjustments made as required based on effectiveness and feasibility outcome data. PROSPECTOR-2 is the definitive trial which will assess whether a prolonged oral challenge is superior to a single dose challenge for ascertaining true immune-mediated penicillin allergy.

Objectives

PROSPECTOR 1: To evaluate the feasibility of a placebo-controlled trial and inform the design of a definitive trial evaluating whether prolonged oral challenge (5 days) is superior to single dose oral challenge in patients reporting penicillin allergy with delayed or unknown timing phenotype to ascertain a true immune-mediated adverse reaction. PROSPECTOR 2: To evaluate the effectiveness of prolonged oral penicillin challenge (5 days) over single dose penicillin challenge for ascertainment of true penicillin allergy (i.e. immune-mediated allergy).

Trial Design

The PROSPECTOR studies are multi-centre, prospective, double-blinded, placebo-controlled, parallel-group, randomised controlled trials: PROSPECTOR-1 is an Australian external pilot study for a future definitive, international, Phase III trial, PROSPECTOR-2.

METHODS: Participants, interventions and outcomes

The study design for PROSPECTOR-1 and PROSPECTOR-2 is outlined in Figure 1.

Study Setting

PROSPECTOR-1 will be undertaken at 4 tertiary hospital centres in Australia, including Austin Health (Victoria), Peter MacCallum Cancer Centre (Victoria), St George Hospital (New South Wales) and Royal Brisbane and Women's Hospital (Queensland). PROSPECTOR-2 will be expanded to 14 tertiary hospital centres across Australia, Asia, North America, Africa and Europe (a complete list may be viewed in Supplementary Table 1).

Eligibility criteria

Adult patients referred to inpatient or outpatient allergy services for a suspected immune-mediated penicillin allergy with history of delayed or unknown timing will be eligible for inclusion in the study. Participants will then be risk-assessed using the PEN-FAST tool (41) and if confirmed to be low-risk (PEN-FAST score < 3), receive a 250-500mg (PROSPECTOR-1) or 500 mg (PROSEPCTOR-2) amoxicillin challenge (with or without prior skin testing). Participants who pass this initial challenge without an observed immune-mediated adverse event (1-2 hours post dose) will proceed to randomisation. The inclusion and exclusion criteria are listed in **Table 1**.

Interventions

Intervention

Participants randomised to the intervention arm will receive a prolonged, 5-day course of oral penicillin (500mg amoxicillin, administered twice daily, to commence the day following the initial single-dose amoxicillin oral challenge).

Control

Participants randomised to the control arm will receive a prolonged, 5-day course of oral placebo (microcrystalline cellulose MCC, prepared to be visually identical to the intervention, administered twice daily, to commence the day following the initial single-dose amoxicillin oral challenge).

Preparation and Dispensing

A qualified pharmacy staff member will dispense the study medication in unique container numbers to a member of the investigator team. A second staff member will verify the dispensing. The participant or their caregiver should be instructed to maintain the product in the bottles provided throughout the course of dosing and return the bottles to the site at either a study visit or via return registered post.

Study compliance

Participants will be asked to record the date and time of each dose of study medication using an electronic or paper diary. Compliance will be assessed at each scheduled visit by study staff who will ask the participant to count the number of capsules remaining. The following non-compliance cases will be recorded as protocol deviations:

- Study medication missed for ≥ 2 consecutive doses
- Study medication compliance < 80%

Outcomes

The primary outcomes for the PROSPECTOR studies are outlined below, with secondary outcomes listed in **Table 2**.

PROSPECTOR-1: Compliance with the intervention (proportion of participants (n, %) taking at least 80% of the doses), need for unblinding (proportion of participants (n, %) being intentionally or unintentionally unblinded) and recruitment to eligibility ratio (proportion of participants (n, %) consented to the study from eligible participants). Subgroup analyses for admission setting (inpatient vs outpatient), risk (PEN-FAST score < 3 vs ≥ 3) and severity (RegiSCAR score < 2 vs ≥ 2) will be performed in PROSPECTOR-1 study.

PROSPECTOR-2: Proportion of positive oral challenges (i.e. immune-mediated reaction up to and including day 7 following the first test dose, as adjudged by an independent blinded panel), n (%). We will perform subgroup analyses for the following parameters: admission setting (inpatient vs outpatient), index reaction phenotype – delayed vs unknown timing, risk (PEN-FAST score 0 vs 1-2 vs ≥ 3), immunocompromised status, sex, region (Australia vs Europe vs Asia vs North America vs Africa), clinic type (specialized allergy clinic vs non-allergy clinic), index reaction severity (RegiSCAR < 2 vs ≥ 2), index reaction phenotype – Severe MPE (RegiSCAR score 1 or 2) vs RegiSCAR score < 1 vs RegiSCAR score > 2, and index reaction timing – delayed exanthema < 5 years post index reaction vs delayed exanthema > 5 years post index reaction.

Participant timeline

The participant timeline is outlined in a schedule of enrolment, interventions and assessments for both studies (**Table 3**). After randomisation on Day 0, participants will be provided with the study medication and a telehealth review will be scheduled for Days 1, 5, 7 and 14 by a specialist allergy healthcare provider. If patients are inpatients at the time, then this review will be performed at the patient bedside. At each telehealth review, compliance will be recorded by reporting the number of doses taken and participants will be asked about any other concurrent antibiotic therapy. If a positive oral challenge is reported, a summary of the patient reported symptoms utilising a standardised questionnaire and clinical photography of any rash, cutaneous or mucosal changes will be sent to an independent review panel consisting of an allergist and dermatologist blinded to the intervention to ascertain if the reported reaction is an "immune-mediated adverse drug reaction".

At Day 30 and Day 90 post-randomisation, a telephone questionnaire and assessment of the medical record will be undertaken to assess for secondary outcomes including antibiotic-associated diarrhoea, *Clostridioides difficile* infection or acquisition of a multi-drug resistant organism. Patients at Day 90 follow up will be unblinded if preferred by the site principal investigator and those in the control arm offered a prolonged oral challenge.

Sample Size

PROSPECTOR-1: A total of 120 participants are planned for inclusion (60 per arm). This sample size was chosen to provide a precise estimate of feasibility outcomes with width of confidence interval being < 20% for any proportion. Such a sample size would also likely provide a reliable estimate of effectiveness as it has been shown that with binary outcomes, gain in precision is smaller once each group reaches 60 participants [42]. This sample size also likely represents >9% of the definitive trial's sample size (to detect 5% difference assuming 8% event rate with 90% power and 5% significance level, a total of almost 900 participants would be required) [42].

PROSPECTOR-2: A total of 830 participants are planned for inclusion (415 per arm). The incidence of delayed reactions after single dose oral challenge in the current literature is approximately 3% [24], while the incidence of delayed reactions after prolonged oral challenge is approximately 8% [31, 46]. To detect a 5% difference with 85% power and 5% significance level, 372 participants would need to be randomised to each arm. To account for 10% loss to follow-up, a total of 830 participants will be recruited.

As the stipulated 5% difference reported in the literature and observed in our pilot data is not regarded as clinically relevant by many drug allergy specialists, we will also evaluate the non-inferiority of single dose challenge. Given the lesser severity of an adverse event, a clinically relevant non-inferiority margin was determined to be 10% among investigators of this study. The planned sample size will enable us to evaluate a non-inferiority of the risk difference between study arms (secondary outcome) with double-sided 95% CI with 82% power (assuming real difference between arms being

5%) [48]. The sample size for PROSPECTOR-2 will be updated prior study start based on the estimates observed in PROSPECTOR-1.

Recruitment

Recruitment will be undertaken by appropriately trained and delegated study investigators at participating sites in both the ambulatory clinic setting and inpatient setting. The central study team will monitor and encourage recruitment by regularly engaging with participating site staff, providing strategies for boosting enrolment and troubleshooting solutions. Recruitment number updates will be communicated through a regular study newsletter.

Consent

Eligible patients will be provided with a verbal explanation of the project by a delegated study investigator and a paper or electronic consent form to read through. They will be encouraged to ask questions and discuss their participation with family, friends or a trusted family doctor if helpful. A thorough assessment of the participant's capacity to make a valid informed decision will be made by the study investigator prior to the patient being recruited and documented informed consent being obtained.

METHODS: Assignment of interventions

Allocation

Permuted block design randomisation will be used, stratified by the hospital site and setting (inpatient vs outpatient). While block design might result in larger treatment imbalances, such design is preferred to overcome logistical difficulties. Randomization will be performed by the unblinded pharmacy dispensing team via REDCap just prior to the intervention. The allocation sequence will be concealed until the time of the randomisation.

Blinding

Participants (and their caregivers if applicable), as well as study investigators and research staff will be blinded to the assigned intervention. Adverse Event Review Panel members will be blinded to the assigned intervention. Clinical trials pharmacists will be unblinded.

Participants may be unblinded during the 7-day follow up period in the event of a serious adverse event (SAE) or Grade 3 or 4 adverse event and if the site principal investigator deems this appropriate. If a participant's assignment is revealed, the Sponsor and Coordinating Principal Investigator will be notified within 24 hours of unblinding. The date and reason for the blind broken must be recorded in the source documentation and case report form. All participants may be unblinded at the 90-day follow-up to allow for those in the control arm to receive a prolonged challenge if that is the site's practice or preference.

METHODS: Data collection, management and analysis

Data collection methods

De-identified clinical data will be stored in a secure electronic REDCap database, hosted by the University of Melbourne. Each participating centre will only have access to their own patient data. All electronic and paper data will be retained for a period of 15 years after which all data will be destroyed according to hospital policy in place at the time.

Progression from pilot to definitive trial

PROSPECTOR-2 will proceed immediately upon completion of PROSPECTOR-1, provided the following criteria are met:

1. Compliance with study medication is ≥ 80%
2. Unblinding is ≤ 10%
3. Recruitment to eligibility ratio ≥ 80%

If these criteria are not met, appropriate amendments to the study design of PROSPECTOR-2 will be made. If compliance is under 80%, additional reminders will be scheduled for participants. If unblinding exceeds 10%, the trial will be converted to open-label. If the recruitment to eligibility ratio is lower than 80%, additional strategies for recruitment may be considered.

Statistical Methods

PROSPECTOR-1: Results will be presented according to CONSORT guidelines for feasibility studies (43). Patient characteristics and penicillin allergy history will be presented by arm using median (interquartile range) for continuous variables and count (percentage) for categorical variables. Binary outcomes will be presented as count and percentage with 95% exact confidence intervals. All outcomes (where feasible) will be presented as overall, by study arm and by setting. Exploratory efficacy outcomes will also be presented as absolute (risk difference) and relative difference (risk ratio) with 95% confidence intervals. No statistical tests will be performed. The amount and pattern of missing data will be explored.

PROSPECTOR 2: Results will be presented according to CONSORT guidelines (44). Patient characteristics and penicillin allergy history will be presented by arm using median (interquartile range) for continuous variables and count (percentage) for categorical variables. The primary analysis will be on intention-to-treat basis. A generalized linear model with binomial family will be used to calculate the risk difference (identity link) and risk ratio (log link) between intervention and control. Results will be presented with two-sided 95% confidence intervals. Models will be adjusted for stratification variables (clinical site and setting). Subgroup analysis will be performed by an inclusion of interaction term between subgroup and arm. The primary analysis will be also performed in per-protocol population. Time to adverse reaction will be evaluated using Kaplan-Meier method and Cox proportions hazards regression. A detailed statistical analysis plan will be prepared and uploaded to the ANZCTR registry listing prior to study completion.

Oversight and monitoring

Adverse Event Review Panel

A blinded independent review panel consisting of an allergist and a dermatologist will be established to review reported adverse drug reactions for both PROSPECTOR studies. A summary of the patient's reaction will be compiled, comprising a standardised symptom questionnaire and clinical photography of any rash, cutaneous or mucosal change. The review panel will provide a determination of whether the reported reaction is to be classified as an "immune-mediated adverse drug reaction". This classification will be provided to the data safety monitoring board (DSMB) for further deliberation in addition to the stipulated reports.

Data safety monitoring board (DSMB)

A DSMB will be established to review trial data and monitor the progress of each trial. The DSMB will monitor adherence to the protocol, participant recruitment, outcomes and participant safety data. They will also monitor the assumptions underlying sample size calculations for the study and alert the investigators if an increased recruitment effort is required. The DSMB will make recommendations as to whether the study should continue or be terminated, consider participant safety or other circumstances as grounds for early termination, including either compelling internal or external evidence of treatment differences or feasibility of addressing the study hypotheses (e.g. poor participant enrolment).

Ethics and dissemination

These studies were reviewed and approved by the Austin Health Human Research Ethics Committee (References Numbers: PROSPECTOR-1: HREC/99740/Austin-2023; PROSPECTOR-2: HREC/109785/Austin-2024). Additional approvals will be sought for international PROSPECTOR-2 sites prior to their participation in the study. Results will be published in peer-reviewed journals and presented at relevant conferences. The final dataset will be

Harms

Adverse events will be recorded from time of randomisation until Day 90. Patients will be supplied with a prescription for oral corticosteroids and second-generation antihistamines upon hospital discharge, only to be used in the event of an immune-mediated positive oral challenge, as instructed by the site investigators at time of the day 1, 5,7 and 14 reviews. Participants will be instructed by site investigators to fill this script at their own expense if required.

Patient and public involvement

No patient and/or public were involved in the study development.

Abbreviations: AAL, Antibiotic Allergy Label; BID, Twice Daily; LOS, Length of Stay; SAE, Serious Adverse Events

Competing Interests: None declared

REFERENCES

1. Trubiano JA, Cairns KA, Evans JA, et al. The prevalence and impact of antimicrobial allergies and adverse drug reactions at an Australian tertiary centre. *BMC Infect Dis*. 2015;15:572. Published 2015 Dec 16. doi:10.1186/s12879-015-1303-3

2. Trubiano JA, Chen C, Cheng AC, Grayson ML, Slavin MA, Thursky KA, et al. Antimicrobial allergy 'labels' drive inappropriate antimicrobial prescribing: lessons for stewardship. J Antimicrob Chemother. 2016;71(6):1715-22.

3. Picard M, Bégin P, Bouchard H, Cloutier J, Lacombe-Barrios J, Paradis J, Des Roches A, Laufer B, Paradis L. Treatment of patients with a history of penicillin allergy in a large tertiary-care academic hospital. J Allergy Clin Immunol Pract. 2013 May-Jun;1(3):252-7. doi: 10.1016/j.jaip.2013.01.006. Epub 2013 Feb 14. PMID: 24565481.

4. Borch JE, Andersen KE, Bindslev-Jensen C. The prevalence of suspected and challenge-verified penicillin allergy in a university hospital population. *Basic Clin Pharmacol Toxicol*. 2006;98(4):357-362. doi:10.1111/j.1742-7843.2006.pto_230.x

5. Day C, Mendelson M, Peter J; ADvISE study group . Low self-reported penicillin allergy in South Africa-implications for global public health response. *JAC Antimicrob Resist*. 2023;5(1):dlad015. Published 2023 Feb 21. doi:10.1093/jacamr/dlad015

6. Li PH, Yeung HHF, Lau CS, Au EYL. Prevalence, Incidence, and Sensitization Profile of β-lactam Antibiotic Allergy in Hong Kong. *JAMA Netw Open*. 2020;3(5):e204199. Published 2020 May 1. doi:10.1001/jamanetworkopen.2020.4199

7. Trubiano JA, Grayson ML, Thursky KA, Phillips EJ, Slavin MA. How antibiotic allergy labels may be harming our most vulnerable patients. Med J Aust. 2018;208(11):469-70.

8. Li PH, Chung HY, Lau CS. Epidemiology and outcomes of geriatric and non-geriatric patients with drug allergy labels in Hong Kong. Hong Kong Med J. 2021 Jun;27(3):192-197. doi: 10.12809/hkmj208716. PMID: 34168086.

9. Chan SCW, Yeung WWY, Wong JCY, Chui ESH, Lee MSH, Chung HY, Cheung TT, Lau CS, Li PH. Prevalence and Impact of Reported Drug Allergies among Rheumatology Patients. Diagnostics (Basel). 2020 Nov 9;10(11):918. doi: 10.3390/diagnostics10110918. PMID: 33182278; PMCID: PMC7695245.

10. Macy E, Contreras R. Health care use and serious infection prevalence associated with penicillin "allergy" in hospitalized patients: A cohort study. The Journal of allergy and clinical immunology. 2014;133(3):790-6.

11. MacFadden DR, LaDelfa A, Leen J, Gold WL, Daneman N, Weber E, et al. Impact of Reported Beta-Lactam Allergy on Inpatient Outcomes: A Multicenter Prospective Cohort Study. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2016;63(7):904-10.

12. Blumenthal KG, Lu N, Zhang Y, Li Y, Walensky RP, Choi HK. Risk of methicillin resistant Staphylococcus aureus and Clostridium difficile in patients with a documented penicillin allergy: population based matched cohort study. Bmj. 2018;361:k2400.

13. Blumenthal KG, Ryan EE, Li Y, Lee H, Kuhlen JL, Shenoy ES. The Impact of a Reported Penicillin Allergy on Surgical Site Infection Risk. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2018;66(3):329-36.

14. Knezevic B, Sprigg D, Seet J, Trevenen M, Trubiano J, Smith W, et al. The revolving door: antibiotic allergy labelling in a tertiary care centre. Internal medicine journal. 2016;46(11):1276-83.

15. Trubiano JA, Leung VK, Chu MY, Worth LJ, Slavin MA, Thursky KA. The impact of antimicrobial allergy labels on antimicrobial usage in cancer patients. Antimicrobial resistance and infection control. 2015;4:23.

16. Blumenthal KG, Peter JG, Trubiano JA, Phillips EJ. Antibiotic allergy. Lancet. 2019;393(10167):183-98.

17. Shi W, Liu N, Huang JX, Xiao H, Meng J, Li PH. Penicillin Allergy in China: Consequences of Inappropriate Skin Testing Practices and Policies. Clin Exp Allergy. 2024 Jul 25. doi: 10.1111/cea.14546. Epub ahead of print. PMID: 39053914.

18. Copaescu AM, Vogrin S, James F, Chua KYL, Rose MT, De Luca J, et al. Efficacy of a Clinical Decision Rule to Enable Direct Oral Challenge in Patients With Low-Risk Penicillin Allergy: The PALACE Randomized Clinical Trial. JAMA internal medicine. 2023;183(9):944-52.
19. Trubiano JA, Thursky KA, Stewardson AJ, Urbancic K, Worth LJ, Jackson C, et al. Impact of an Integrated Antibiotic Allergy Testing Program on Antimicrobial Stewardship: A Multicenter Evaluation. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2017;65(1):166-74.
20. Li PH, Wong JCY, Chan JMC, et al. Hong Kong Drug Allergy Delabelling Initiative (HK-DADI) consensus statements for penicillin allergy testing by nonallergists. *Front Allergy*. 2022;3:974138. Published 2022 Sep 5. doi:10.3389/falgy.2022.974138
21. Li PH, Thong BY, Pawankar R, et al. APAAACI clinical pathway on direct provocation testing for penicillin allergy delabeling. *Asia Pac Allergy*. 2023;13(4):142-147. doi:10.5415/apallergy.0000000000000122
22. Kan AKC, Hui HKS, Li TS, et al. Comparative Effectiveness, Safety, and Real-World Outcomes of a Nurse-Led, Protocol-Driven Penicillin Allergy Evaluation From the Hong Kong Drug Allergy Delabelling Initiative (HK-DADI). *J Allergy Clin Immunol Pract*. 2023;11(2):474-480.e2. doi:10.1016/j.jaip.2022.08.052
23. Wong JCY, Kan AKC, Chik TSH, et al. Prospective, Multicenter, Head-to-Head Comparison Between Allergists Versus Nonallergists in Low-Risk Penicillin Allergy Delabeling: Effectiveness, Safety, and Quality of Life (HK-DADI2). *J Allergy Clin Immunol Pract*. 2024;12(7):1801-1808.e2. doi:10.1016/j.jaip.2024.04.010
24. Wrenn RH, Trubiano JA. Penicillin Allergy Impact and Management. Infectious disease clinics of North America. 2023;37(4):793-822.
25. Khan DA, Banerji A, Blumenthal KG, Phillips EJ, Solensky R, White AA, et al. Drug allergy: A 2022 practice parameter update. The Journal of allergy and clinical immunology. 2022.
26. Barbaud A, Garvey LH, Torres M, Laguna JJ, Arcolaci A, Bonadonna P, et al. EAACI/ENDA position paper on drug provocation testing. Allergy. 2023.
27. Hjortlund J, Mortz CG, Skov PS, Eller E, Poulsen JM, Borch JE, et al. One-week oral challenge with penicillin in diagnosis of penicillin allergy. Acta Derm Venereol. 2012;92(3):307-12.
28. Hjortlund J, Mortz CG, Skov PS, Bindslev-Jensen C. Diagnosis of penicillin allergy revisited: the value of case history, skin testing, specific IgE and prolonged challenge. Allergy. 2013;68(8):1057-64.
29. Mori F, Cianferoni A, Barni S, Pucci N, Rossi ME, Novembre E. Amoxicillin allergy in children: five-day drug provocation test in the diagnosis of nonimmediate reactions. The journal of allergy and clinical immunology In practice. 2015;3(3):375-80 e1.
30. Ratzon R, Reshef A, Efrati O, Deutch M, Forschmidt R, Cukierman-Yaffe T, et al. Impact of an extended challenge on the effectiveness of beta-lactam hypersensitivity investigation. Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology. 2016;116(4):329-33.
31. Fransson S, Mosbech H, Kappel M, Hjortlund J, Poulsen LK, Kvisselgaard AD, et al. The Importance of Prolonged Provocation in Drug Allergy - Results From a Danish Allergy Clinic. The journal of allergy and clinical immunology In practice. 2017;5(5):1394-401.
32. Lezmi G, Alrowaishdi F, Bados-Albiero A, Scheinmann P, de Blic J, Ponvert C. Non-immediate-reading skin tests and prolonged challenges in non-immediate hypersensitivity to beta-lactams in children. Pediatric allergy and immunology : official publication of the European Society of Pediatric Allergy and Immunology. 2018;29(1):84-9.
33. Borch JE, Bindslev-Jensen C. Full-course drug challenge test in the diagnosis of delayed allergic reactions to penicillin. International archives of allergy and immunology. 2011;155(3):271-4.
34. PMID: 35108710
35. Boysen J, Garvey, L. Retrospective evaluation of the PEN-FAST tool in Patients Undergoing Prolonged Drug Provocation. EAACI2023.
36. Mendelson LM, Ressler C, Rosen JP, Selcow JE. Routine elective penicillin allergy skin testing in children and adolescents: study of sensitization. The Journal of allergy and clinical immunology. 1984;73(1 Pt 1):76-81.

37. Pichichero ME, Pichichero DM. Diagnosis of penicillin, amoxicillin, and cephalosporin allergy: reliability of examination assessed by skin testing and oral challenge. The Journal of pediatrics. 1998;132(1):137-43.
38. Solensky R, Earl HS, Gruchalla RS. Lack of penicillin resensitization in patients with a history of penicillin allergy after receiving repeated penicillin courses. Archives of internal medicine. 2002;162(7):822-6.
39. Dorman SM, Seth S, Khan DA. Risk of Allergic Reactions to Recurrent Intravenous Penicillin Administration in Penicillin Skin Test Negative Patients. The journal of allergy and clinical immunology In practice. 2018;6(1):196-200.
40. Garcia Rodriguez R, Moreno Lozano L, Extremera Ortega A, Borja Segade J, Galindo Bonilla P, Gomez Torrijos E. Provocation Tests in Nonimmediate Hypersensitivity Reactions to beta-Lactam Antibiotics in Children: Are Extended Challenges Needed? The journal of allergy and clinical immunology In practice. 2019;7(1):265-9.
41. Trubiano JA, Vogrin S, Chua KYL, et al. Development and Validation of a Penicillin Allergy Clinical Decision Rule. *JAMA Intern Med*. 2020;180(5):745-752. doi:10.1001/jamainternmed.2020.040342. Teare MD, Dimairo M, Shephard N, Hayman A, Whitehead A, Walters SJ. Sample size requirements to estimate key design parameters from external pilot randomised controlled trials: a simulation study. *Trials*. 2014;15:264. Published 2014 Jul 3. doi:10.1186/1745-6215-15-264
43. Eldridge SM, Chan CL, Campbell MJ, Bond CM, Hopewell S, Thabane L, et al. CONSORT 2010 statement: extension to randomised pilot and feasibility trials. Pilot Feasibility Stud. 2016;2:64.
44. Schulz KF, Altman DG, Moher D, Group C. CONSORT 2010 statement: updated guidelines for reporting parallel group randomized trials. Annals of internal medicine. 2010;152(11):726-32.
45. Kulalert P, Phinyo P, Chiriac AM, Demoly P, Saokaew S, Kanchanaphoomi K, et al. Is a Prolonged Drug Provocation Test Better Than a Single-Day Drug Provocation Test? A Systematic Review and Meta-Analysis. The journal of allergy and clinical immunology In practice. 2023.
46. Trubiano J, Phillips E. Antimicrobial stewardship's new weapon? A review of antibiotic allergy and pathways to 'de-labeling'. Curr Opin Infect Dis. 2013;26(6):526-37.
47. Mitri E, Reynolds G, Hornung CJ, Trubiano JA. Low-risk penicillin allergy delabeling: a scoping review of direct oral challenge practice, implementation, and multi-disciplinary approaches. Expert Rev Anti Infect Ther. 2023:1-11.
48. Baur D, Gladstone BP, Burkert F, Carrara E, Foschi F, Dobele S, et al. Effect of antibiotic stewardship on the incidence of infection and colonisation with antibiotic-resistant bacteria and Clostridium difficile infection: a systematic review and meta-analysis. Lancet Infect Dis. 2017;17(9):990-1001.
49. Baiardini I, Braido F, Fassio O, Calia R, Giorgio WC, Romano A, et al. Development and validation of the Drug Hypersensitivity Quality of Life Questionnaire. Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology. 2011;106(4):330-5.

Figure legends:

Figure 1: Study Design for PROSPECTOR-1 and 2 studies.

[a]penicillin "unspecified", penicillin VK, penicillin G, amoxicillin, ampicillin, flucloxacillin, dicloxacillin, cloxacillin, mecillinam, pivmecillinam, pivampicillin

Abbreviations: BID, twice daily

Table 1. Inclusion and Exclusion Criteria

Inclusion Criteria
1. Adults patients referred to the inpatient or outpatient allergy services for a suspected penicillin allergy with an immune-related allergy history of delayed (> 6 hours after first dose of drug administration) or unknown timing, who tolerate first single-dose of an oral amoxicillin challenge
2. Willing and able to give consent and undergo telehealth/telephone review
Exclusion Criteria
1. Patient age is < 18 years
2. Any other illness that, in the investigator's judgement, will substantially increase the risk associated with subject's participation in this study
3. Stevens-Johnson Syndrome or Toxic Epidermal Necrolysis to beta-lactam
4. Inpatients concurrently receiving or likely to receive a beta-lactam antibiotic therapy during the 14-day study period
5. Concurrent use of antihistamines and systemic steroid therapy (i.e. > 10mg daily) **[PROSPECTOR-2]**

Table 2. Secondary outcome measures for PROSPECTOR studies

	PROSPECTOR-1	**PROSPECTOR-2**
Positive Oral Challenge	Up to day 7 following the first hospital administered single dose	Up to day 14 following the first hospital administered single dose
Feasibility	- Recruitment rate per site (recruitment/site/month) - Randomisation to recruitment ratio (n, %) - Withdrawal (n, %) - Loss to follow-up (n, %) - Missing data - Protocol compliance (n, %)	Not applicable
Safety	- Severe adverse reaction (n, %) - Immune-mediated adverse event or severe adverse drug reaction (n, %) - Non-immune mediated adverse event (n, %) - Any cutaneous adverse reaction (n, %)	- Immediate severe adverse reaction (anaphylaxis or death) (n, %) - Delayed adverse reaction (severe cutaneous adverse reaction) (n, %) - Non-immune mediated adverse event (n, %) - Grade 3 or 4 adverse reactions as defined by World Allergy Organization (ref: Sanchez-Borges et al, 2019) (n, %)

		- Any cutaneous adverse reactions (n, %)
Efficacy	- *C. difficile* infection at day-30 and -90 (n, %) - Isolation of a multidrug resistant infection at day-30 and -90 (n, %)	- *C. difficile* infection at day-30, -90 and -120 (n, %) - Multidrug resistant infection at day-30, -90 and -120 (n, %) - Multidrug resistant colonisation at day-30, -90 and -120 (n, %)
Cost Effectiveness	cost effectiveness of placebo versus open label trial	Cost effectiveness analysis of prolonged versus single dose oral challenge
Quality of Life	Not applicable	Health-related quality of life outcome, measure by shortened Drug Hypersensitivity Quality of Life Questionnaire (DrHy-Q) (**Supplementary Table 2**) at day 0 and day 90.

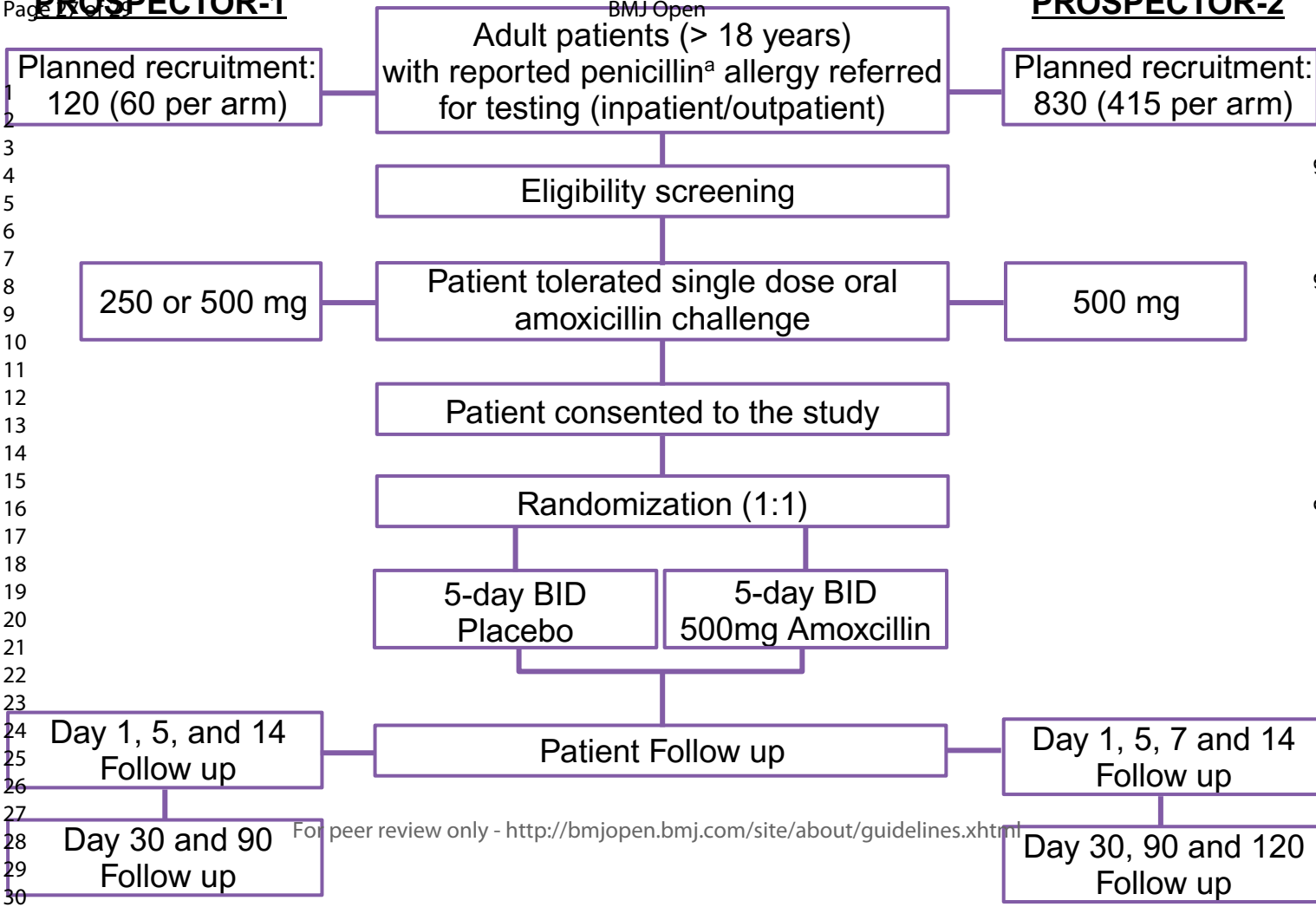
Table 3. Schedule of enrolment, interventions and assessments

	Screening/Enrolment*	Treatment period		Follow up period					Notes
		Day 1	Day 5	Day 7	Day 14	Day 30	Day 90	Day 120	
Visit window	Day -5 to 0	(0)	(+/-1)	(+/-1)	(+/- 1)	(+/-3)	(+/-3)	(+/-3)	
Informed consent	X								Informed consent must occur prior to all study activities
Baseline demographics	X								
Baseline medical and allergy history	X								
Concomitant medications	X	X	X	X	X				Record any new medications started during the study period or within 5 days of enrolment
DrHy-Q Questionnaire	X								
Vital signs measurement	X	X							Vital signs to be measured at baseline on Day 0 (prior to single dose oral challenge) and post-dose (frequency at investigator's discretion)
Single dose oral penicillin challenge	X								Challenge and 1-hour post-dose observation period to be completed prior to randomisation
Review of eligibility criteria	X								Review of eligibility criteria must be performed prior to randomisation
Randomisation	X								Day 0

Dispense study medication	X							Study medication is dispensed on the day of randomisation (Day 0)
Study drug administration		Day 1 – 5 (10 doses total)						Participant to self-administer study drug twice daily starting Day 1
Participant completes dosing log		Day 1 – 5						Participant to complete dosing log. Note: If dose 1 is administered in the evening or PM of Day 1, treatment will continue until Day 6.
Study drug compliance check		X	X					Site research staff to perform compliance check and review dosing log
Adverse event monitoring		X	X	X	X	X	X	Site research staff to perform follow-up phone calls with participants to monitor AEs
Telehealth/telephone follow-up		X	X	X	X			
Email outcomes survey						X	X	
Retrieval of unused medications				X				

*Screening procedures may occur on Days -5 to Day 0, i.e. may be performed on the day of randomisation if preferred by site investigators

**Use of Shortened DrHY-Q - Drug Allergy Quality of Life Questionnaire [49]



SUPPLEMENTARY Materials

Supplementary Table 1. List of study centres for PROSPECTOR-1 and PROSPECTOR-2

Site	Address	Site PI	PROSPECTOR-1	PROSPECTOR-2
Austin Health	145 Studley Road, Heidelberg VIC 3084, Australia	Prof Jason Trubiano	X	X
Peter MacCallum Cancer Centre	305 Grattan Street, Melbourne, VIC 3000, Australia	Dr Morgan Rose	X	X
Royal Melbourne Hospital	300 Grattan Street, Parkville VIC 3052, Australia	Dr Jack Godsell		X
St George Hospital	Gray Street, Kogarah, NSW 2217, Australia	Dr Richard Sullivan	X	X
Royal North Shore Hospital	Reserve Road, St Leonards NSW 2065, Australia	Prof Suran Fernando		X
Royal Brisbane and Women's Hospital	Butterfield St, Herston QLD 4006, Australia	Dr Michael Lane	X	X
Royal Adelaide Hospital	Port Rd, Adelaide SA 5000, Australia	A/Prof William Smith		X
Sir Charles Gairdner Hospital	Hospital Ave, Nedlands WA 6009, Australia	Prof Michaela Lucas		X
Montreal General Hospital	1650 Cedar Ave, Montreal, Quebec H3G 1A4, Canada	Dr Ana Copaescu		X
Allergy Clinic, Herlev and Gentofte Hospital	Gentofte Hospitalsvej 8 1. Floor, 2900 Hellerup, Denmark	Prof Lene H Garvey		X
Groot Schuur Hospital	Main Road, Observatory, Cape Town, 7935, South Africa	Prof Jonathan Peter		X

Queen Mary Hospital, University of Hong Kong	Pokfulam Road, Pokfulam, Hong Kong	Dr Philip Li		X
Antwerp University Hospital	Drie Eikenstraat 655, 2650 Edegem, Belgium	Prof Vito Sabato		X

Supplementary Table 2. Drug Hypersensitivity Quality of Life Questionnaire (Mak HWF et al, JACI: In Pract 2024)

	Not at all				**Very much**
	0	1	2	3	4
1. The problem of adverse reaction to drugs affects my life					
2. The fact that I cannot use medication safely made me feel different from others					
3. I feel anxious due to my problem of allergy reaction					
4. I feel anguished due to my problem of allergy reaction					
5. The idea of taking a medicine makes me feel anxious					

Total Score: _____

BMJ Open

Prolonged versus single dose in penicillin oral challenge testing – Protocols for a pilot and definitive randomised controlled trial (PROSPECTOR Studies)

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2024-094712.R1
Article Type:	Protocol
Date Submitted by the Author:	19-Jan-2025
Complete List of Authors:	Ng, Irvin; Austin Health, Infectious Diseases and Immnology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases James, Fiona; Austin Health, Department of Infectious Diseases; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases Copaescu, Ana; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases Vogrin, Sara; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases Mitri, Elise; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases; National Allergy Centre of Excellence Rose , Morgan; Austin Health, Department of Infectious Diseases; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases; Peter MacCallum Cancer Centre Sullivan, Richard; St George Hospital, Infectious Diseases Lane, Michael; Royal Brisbane and Women's Hospital Legg, Amy; Royal Brisbane and Women's Hospital, Pharmacy Department Godsell, Jack; Austin Health, Infectious Diseases and Immunology; The Royal Melbourne Hospital, Allergy and Immunology Fernando, Suran; Royal North Shore Hospital, Clinical Immunology and Allergy Garvey, Lene Heise; University of Copenhagen; Herlev and Gentofte Hospital, Allergy Clinic, Department of Dermatology and Allergy Sabato, Vito; University of Antwerp, Department of Immunology, Allergology and Rheumatology; University Hospital Antwerp, Department of Immunology, Allergology and Rheumatology Li, Philip; Hong Kong University, Division of Rheumatology and Clinical Immunology, Department of Medicine Peter, Jonathan; University of Cape Town, Division of Allergy and Immunology, Department of Medicine Trubiano, Jason ; Austin Health, Infectious Diseases and Immunology; The Peter Doherty Institute for Infection and Immunity, Infectious Diseases; National Allergy Centre of Excellence
Primary Subject Heading:	Immunology (including allergy)
Secondary Subject Heading:	Infectious diseases

Keywords:	Clinical Trial, Antibiotics < Anti-Bacterial Agents, PUBLIC HEALTH, Protocols & guidelines < HEALTH SERVICES ADMINISTRATION & MANAGEMENT, IMMUNOLOGY

Title: Prolonged versus single dose in penicillin oral challenge testing – Protocols for a pilot and definitive randomised controlled trial (PROSPECTOR Studies)

Authors: Ng I[1,2], James F[1,2], Copaescu A[1,2], Vogrin S[1,2], Mitri E[1,2,3], Rose MT[1,2,4], Sullivan RP[5], Lane MJ[6], Legg A[7], Godsell J[1,8], Fernando S[9], Garvey LH[10], Sabato V[11], Li PH[12], Peter J[13], Trubiano JA[1,2,3].

Affiliations:

1. Centre for Antibiotic Allergy and Research, Department of Infectious Diseases and Immunology, Austin Health, Victoria, Australia
2. Department of Infectious Diseases, The Peter Doherty Institute for Infection and Immunity, University of Melbourne, Victoria, Australia.
3. National Allergy Centre of Excellence (NACE), Parkville, Victoria, Australia.
4. National Centre for Infection in Cancer, Department of Infectious Diseases, Peter MacCallum Cancer Centre, Australia
5. Department of Infectious Diseases, St George Hospital, School of Clinical Medicine, UNSW Medicine and Health, Kogarah, NSW, Australia
6. Royal Brisbane and Women's Hospital, University of Queensland, Queensland, Australia
7. Department of Pharmacy, Royal Brisbane and Women's Hospital, Brisbane, Queensland, Australia
8. Department of Allergy and Immunology, Royal Melbourne Hospital, University of Melbourne, Parkville, Australia
9. Department of Clinical Immunology and Allergy, Royal North Shore Hospital, Sydney, Australia
10. Allergy Clinic, Department of Dermatology and Allergy, Herlev and Gentofte Hospital and University of Copenhagen, Copenhagen, Denmark.
11. Department of Immunology, Allergology and Rheumatology, University of Antwerp, Antwerp University Hospital, Antwerpen, Belgium; Allergology Unit, AZ Jan Palfijn, Ghent, Belgium; Infla-Med Centre of Excellence, University of Antwerp, Antwerpen, Belgium
12. Division of Rheumatology and Clinical Immunology, Department of Medicine, Queen Mary Hospital, The University of Hong Kong, Hong Kong SAR, China.

13. Division of Allergy and Clinical Immunology, Department of Medicine, University of Cape Town, Cape Town, South Africa

Trial Sponsor:

Prof Jason Trubiano

Centre for Antibiotic Allergy and Research

Department of Infectious Diseases and Immunology, Austin Health

145 Studley Road, Heidelberg VIC 3121

AUSTRALIA

Funding: The work is supported by an NHMRC Emerging Leadership Fellowship (EL2) awarded to Professor Jason Trubiano and Austin Health (Grant number GNT2008071)

Corresponding author:

Prof Jason Trubiano

Department of Infectious Diseases and Immunology, Austin Health

P: +61 3 94966676

E: jason.trubiano@austin.org.au

Author contributions:

JAT and SV planned the study design and wrote the study protocols.

Ethics submission: IN and FJ

Patient recruitment: JAT, AC, EM, RMT, RPS, MJL, AL, JG, SF, LHG, VS, PHL and JP

Statistical design and analysis: SV

All authors reviewed the protocol and this manuscript.

JAT is responsible for the overall content as guarantor

Protocol version:

PROSPECTOR-1 Protocol: Version 7 dated 22nd August 2023

PROSPECTOR-2 Protocol: Version 1.1 dated 28th August 2024

Word count: 2993

ABSTRACT

Introduction: Penicillin allergy labels (PALs) are reported in 1 in 10 hospitalized patients globally and associated with inferior patient, hospital and microbiological outcomes, however the majority are incorrect and should be removed. Direct oral penicillin challenge has been demonstrated to be a safe and effective method for the removal of PALs. However, the question of whether a single dose is sufficient to ascertain true allergy status remains unanswered, with some studies suggesting that extended challenges of 3 or more days are superior for the exclusion of delayed immune reactions. The aim of the PROSPECTOR studies is to determine the feasibility (PROSPECTOR-1) of a definitive trial (PROSPECTOR-2) to evaluate the safety and effectiveness of prolonged oral challenge (i.e. 5-day) versus single dose oral challenge in patients with a delayed or unknown penicillin allergy phenotype (PROSPECTOR-2).

Methods and analysis: A pair of double-blind two-arm parallel placebo-controlled trials will be undertaken - **PRO**longed versus **S**ingle dose in **PE**nicillin oral **C**hallenge **T**esting double-blind parallel group randomised placebo-c**O**ntrolled t**R**ial (PROSPECTOR Studies). Patients with a reported delayed or unknown timing penicillin allergy that have passed a supervised single dose oral amoxicillin challenge (with or without prior skin testing/single or split dose) will be recruited. Informed patient consent will be granted for sites to recruit patients and collect routine clinical data. PROSPECTOR-1 will assess the safety and feasibility of a placebo-controlled trial for single dose amoxicillin challenge versus 5-day prolonged oral challenge. PROSPECTOR-2 will assess the superiority of 5-day prolonged oral challenge compared with single dose amoxicillin challenge in excluding a delayed immune reaction. PROSPECTOR-2 will commence immediately post completion of PROSPECTOR-1 in a vanguard design, with adjustments to the projected sample size for superiority made following completion of PROSPECTOR-1. PROSPECTOR-2 will commence recruitment immediately following closure of PROSPECTOR-1, however data from each trial will be analysed separately.

Ethics and dissemination: These studies were reviewed and approved by the Austin Health Human Research Ethics Committee (PROSPECTOR-1: HREC/99740/Austin-2023 & PROSPECTOR-2: HREC/109785/Austin-2024). Results will be published in peer-reviewed journals and presented at relevant conferences.

Registration: PROSPECTOR-1: ACTRN12623001242617 & PROSPECTOR-2: ACTRN12624001107516

Keywords: penicillin allergy, beta-lactam allergy; penicillin provocation, penicillin challenge, drug allergy, antimicrobial stewardship

Strengths and limitations of this study:

1. These studies are among the first double-blind randomised placebo-controlled trials in antibiotic allergy investigation
2. The pilot phase, randomised experimental design and recruitment of patients from existing inpatient or outpatient settings will minimise the opportunity for selection bias
3. The definitive trial is international and multicentre, allowing for increased sample heterogeneity and generalisability of results
4. The pilot study does not aim to explore the ideal number of days for prolonged challenge in eliciting true delayed allergy, so conclusions will be limited to comparison with a prolonged 5-day challenge while there is still variability in practice globally
5. A twice-daily 500mg dose of penicillin is set as the intervention, however variability in preferred dosage remains in prolonged challenge practice globally

INTRODUCTION

Background and rationale

Penicillin allergy labels (PALs) are commonly documented in patient electronic medical records (EMRs) [1]. At the higher end, the prevalence has been estimated at 10% for hospitalised Australians [2] and 9.9% of inpatients in Montreal, Canada [3]. A Danish study found 5% of hospital inpatients carried a PAL [4], while at the lower end the prevalence is estimated at 3.2% in hospital inpatients in South Africa [5] and 2% for all beta-lactam allergy in Hong Kong patients [6]. This figure may be even higher among vulnerable patients such as the immunocompromised [7], geriatric and rheumatology populations [8, 9]. Those patients that carry a PAL are more likely to receive an inappropriate antibiotic, suffer a hospital-associated adverse event and acquire a multidrug resistant organism [10-13]. PALs are also associated with increased hospital length-of-stay (LOS), higher readmission rates, increased hospital costs and mortality rates [10, 14, 15]. At a public health level, they are associated with inappropriate prescribing and antimicrobial resistance [1].

Despite their omnipresence, the majority of PALs are assessed as "low-risk" and can be safely removed by penicillin allergy testing [16-19]. Oral penicillin challenge with or without preceding skin testing is considered the gold standard for delabelling [20], however clinical equipoise remains regarding the superiority of single dose or prolonged (i.e. multiple-day dosing) oral challenge for patients that report a delayed or unknown timing penicillin allergy phenotype. The current Drug Allergy Practice Parameters recommend "against the routine use of multiple-day challenges in the evaluation of penicillin allergy", providing a "strong recommendation" but with "low certainty of evidence" [21]. The European guidelines reviewed the literature of over 6,484 patients, demonstrating a 2.3% positive rate following the initial challenge and 5.5% during the varied prolonged challenges. They concluded there is no consensus on a preferred procedure and could not provide a recommendation for or against prolonged oral challenge [22].

Results from a mixture of European observational and retrospective studies suggest that prolonged challenges ranging from 3 to 10 days may be superior to single dose challenges at eliciting delayed immune reactions. However, the reported prevalence of delayed reactions is highly variable (5-12% of patients), and many were reliant on patient self-reporting [23-30]. In a recent retrospective single-centre Danish study of 3,179 low-risk patients, 2.6% were positive on day 1 of challenge and 7.2% after day 1 [31]. This contrasts with the North American experience, where prolonged challenges have been associated with low rates of delayed reactions (0-1.8%) [32-35]. A paediatric study demonstrated that delayed reactions may occur <7 days following a single challenge [36]. This gap between guideline recommendations and evidence, and differing results across geographical regions, highlights the need for robust evidence to inform practice with clinical certainty.

Randomised controlled trials (RCTs) have not routinely been used to answer questions regarding best practice in penicillin allergy research, despite their ability to provide high-quality evidence. The PROSPECTOR (**PRO**longed versus **S**ingle dose in **PE**nicillin oral **C**hallenge **T**esting - Double-blind parallel-group randomised placebo-c**O**ntrolled t**R**ial) studies will utilise a double-blind, parallel-group, placebo-controlled RCT study design. PROSPECTOR-1 is an external pilot trial which will assess the feasibility of conducting a blinded placebo-controlled trial of single dose versus prolonged dose oral challenge in patients with a documented or reported PAL. It will also inform the sample size of a definitive full-scale trial which will follow directly on from PROSPECTOR-1, with adjustments made as required based on effectiveness and feasibility outcome data. PROSPECTOR-2 is the definitive trial which will assess whether a prolonged oral challenge is superior to a single dose challenge for ascertaining true immune-mediated penicillin allergy.

Objectives

PROSPECTOR-1: To evaluate the feasibility of a placebo-controlled trial and inform the design of a definitive trial evaluating whether prolonged oral challenge (5 days) is superior to single dose oral challenge in patients reporting penicillin allergy with delayed or unknown timing phenotype to ascertain a true immune-mediated adverse reaction. PROSPECTOR-2: To evaluate the effectiveness

of prolonged oral penicillin challenge (5 days) over single dose penicillin challenge for ascertainment of true penicillin allergy (i.e. immune-mediated allergy).

Trial Design

The PROSPECTOR studies are multi-centre, prospective, double-blinded, placebo-controlled, parallel-group, randomised controlled trials: PROSPECTOR-1 is an Australian external pilot study for a future definitive, international, Phase III trial, PROSPECTOR-2.

METHODS: Participants, interventions and outcomes

The study design for PROSPECTOR-1 and PROSPECTOR-2 is outlined in Figure 1.

Study Setting

PROSPECTOR-1 will be undertaken at 4 tertiary hospital centres in Australia, including Austin Health (Victoria), Peter MacCallum Cancer Centre (Victoria), St George Hospital (New South Wales) and Royal Brisbane and Women's Hospital (Queensland). PROSPECTOR-2 will be expanded to 14 tertiary hospital centres across Australia, Asia, North America, Africa and Europe (a complete list may be viewed in Supplementary Table 1).

Eligibility criteria

Adult patients referred to inpatient or outpatient allergy services for a suspected immune-mediated penicillin allergy with history of delayed or unknown timing will be eligible for inclusion in the study. Participants will then be risk-assessed using the PEN-FAST tool [37] and RegiSCAR score. Participant will receive a single dose of 250-500mg (PROSPECTOR-1) or 500 mg (PROSEPCTOR-2) amoxicillin challenge (with or without prior skin testing). Participants who pass this initial challenge without an observed immune-mediated adverse event (1-2 hours post dose) will proceed to randomisation. The inclusion and exclusion criteria are listed in **Table 1**.

Interventions

Intervention

Participants randomised to the intervention arm will receive a prolonged, 5-day course of oral 500mg amoxicillin, administered twice daily, to commence the day following the initial single-dose amoxicillin oral challenge.

Control

Participants randomised to the control arm will receive a prolonged, 5-day course of oral placebo (microcrystalline cellulose MCC, prepared to be visually identical to the intervention, administered twice daily, to commence the day following the initial single-dose amoxicillin oral challenge).

Preparation and Dispensing

A qualified pharmacy staff member will dispense the study medication in unique container numbers to a member of the investigator team. A second staff member will verify the dispensing. The participant or their caregiver should be instructed to maintain the product in the bottles provided throughout the course of dosing and return the bottles to the site at either a study visit or via return registered post.

Study compliance

Participants will be asked to record the date and time of each dose of study medication using an electronic or paper diary. Compliance will be assessed at each scheduled visit by study staff who will ask the participant to count the number of capsules remaining. The following non-compliance cases will be recorded as protocol deviations:

- Study medication missed for ≥ 2 consecutive doses
- Study medication compliance < 80%

Outcomes

The primary outcomes for the PROSPECTOR studies are outlined below, with secondary outcomes listed in **Table 2**.

PROSPECTOR-1: Compliance with the intervention (proportion of participants (n, %) taking at least 80% of the doses), need for unblinding (proportion of participants (n, %) being intentionally or unintentionally unblinded) and recruitment to eligibility ratio (proportion of participants (n, %) consented to the study from eligible participants). Subgroup analyses for admission setting (inpatient vs outpatient), risk (PEN-FAST score < 3 vs ≥ 3) and severity (RegiSCAR score < 2 vs ≥ 2) will be performed in PROSPECTOR-1 study.

PROSPECTOR-2: Proportion of positive oral challenges (i.e. immune-mediated reaction up to and including day 7 following the first test dose, as adjudged by an independent blinded panel), n (%). We will perform subgroup analyses for the following parameters: admission setting (inpatient vs outpatient), index reaction phenotype – delayed vs unknown timing, risk (PEN-FAST score 0 vs 1-2 vs ≥ 3), immunocompromised status, sex, region (Australia vs Europe vs Asia vs North America vs Africa), clinic type (specialized allergy clinic vs non-allergy clinic), index reaction severity (RegiSCAR < 2 vs ≥ 2), index reaction phenotype – Severe MPE (RegiSCAR score 1 or 2) vs RegiSCAR score < 1 vs RegiSCAR score > 2, and index reaction timing – delayed exanthema < 5 years post index reaction vs delayed exanthema > 5 years post index reaction.

Participant timeline

The participant timeline is outlined in a schedule of enrolment, interventions and assessments for both studies (**Supplementary Table 2**). After randomisation on Day 0, participants will be provided with the study medication and a telehealth review will be scheduled for Days 1, 5, 7 and 14 by a specialist allergy healthcare provider (i.e. board-certified allergist, clinical immunologist and other clinicians with specialized training in allergy and immunology). If patients are inpatients at the time, then this review will be performed at the patient bedside. At each telehealth review, compliance will be recorded by reporting the number of doses taken and participants will be asked about any other

concurrent antibiotic therapy. If a positive oral challenge is reported, a summary of the patient reported symptoms utilising a standardised questionnaire and clinical photography of any rash, cutaneous or mucosal changes will be sent to an independent review panel consisting of an allergist and dermatologist blinded to the intervention to ascertain if the reported reaction is an "immune-mediated adverse drug reaction".

At Day 30 and Day 90 post-randomisation, a telephone questionnaire and assessment of the medical record will be undertaken to assess for secondary outcomes including antibiotic-associated diarrhoea, *Clostridioides difficile* infection or acquisition of a multi-drug resistant organism. Patients at Day 90 follow up will be unblinded if preferred by the site principal investigator and those in the control arm offered a prolonged oral challenge.

Sample Size

PROSPECTOR-1: A total of 120 participants are planned for inclusion (60 per arm). This sample size was chosen to provide a precise estimate of feasibility outcomes with width of confidence interval being < 20% for any proportion. Such a sample size would also likely provide a reliable estimate of effectiveness as it has been shown that with binary outcomes, gain in precision is smaller once each group reaches 60 participants [38]. This sample size also likely represents >9% of the definitive trial's sample size (to detect 5% difference assuming 8% event rate with 90% power and 5% significance level, a total of almost 900 participants would be required) [38].

PROSPECTOR-2: A total of 830 participants are planned for inclusion (415 per arm). The incidence of delayed reactions after single dose oral challenge in the current literature is approximately 3% [20], while the incidence of delayed reactions after prolonged oral challenge is approximately 8% [27, 39]. To detect a 5% difference with 85% power and 5% significance level, 372 participants would need to be randomised to each arm. To account for 10% loss to follow-up, a total of 830 participants will be recruited.

As the stipulated 5% difference reported in the literature and observed in our pilot data is not regarded as clinically relevant by many drug allergy specialists, we will also evaluate the non-inferiority of single dose challenge. Given the lesser severity of an adverse event, a clinically relevant non-inferiority margin was determined to be 10% among investigators of this study. The planned sample size will enable us to evaluate a non-inferiority of the risk difference between study arms (secondary outcome) with double-sided 95% CI with 82% power (assuming real difference between arms being 5%) [40]. The sample size for PROSPECTOR-2 will be updated prior study start based on the estimates observed in PROSPECTOR-1.

Recruitment

Recruitment will be undertaken by appropriately trained and delegated study investigators at participating sites in both the ambulatory clinic setting and inpatient setting. The central study team will monitor and encourage recruitment by regularly engaging with participating site staff, providing strategies for boosting enrolment and troubleshooting solutions. Recruitment number updates will be communicated through a regular study newsletter.

Consent

Eligible patients will be provided with a verbal explanation of the project by a delegated study investigator and a paper or electronic consent form to read through (Supplementary Materials 1-2). They will be encouraged to ask questions and discuss their participation with family, friends or a trusted family doctor if helpful. A thorough assessment of the participant's capacity to make a valid informed decision will be made by the study investigator prior to the patient being recruited and documented informed consent being obtained.

METHODS: Assignment of interventions

Allocation

Permuted block design randomisation will be used, stratified by the hospital site and setting (inpatient vs outpatient). While block design might result in larger treatment imbalances, such design is preferred to overcome logistical difficulties. Randomization will be performed by the unblinded pharmacy dispensing team via REDCap just prior to the intervention. The allocation sequence will be concealed until the time of the randomisation.

Blinding

Participants (and their caregivers if applicable), as well as study investigators and research staff will be blinded to the assigned intervention. Adverse Event Review Panel members will be blinded to the assigned intervention. Clinical trials pharmacists will be unblinded.

Participants may be unblinded during the 7-day follow up period in the event of a serious adverse event (SAE) or Grade 3 or 4 adverse event and if the site principal investigator deems this appropriate. If a participant's assignment is revealed, the Sponsor and Coordinating Principal Investigator will be notified within 24 hours of unblinding. The date and reason for the blind broken must be recorded in the source documentation and case report form. All participants may be unblinded at the 90-day follow-up to allow for those in the control arm to receive a prolonged challenge if that is the site's practice or preference.

METHODS: Data collection, management and analysis

Data collection methods

De-identified clinical data will be stored in a secure electronic REDCap database, hosted by the University of Melbourne. Each participating centre will only have access to their own patient data. All electronic and paper data will be retained for a period of 15 years after which all data will be destroyed according to hospital policy in place at the time.

Progression from pilot to definitive trial

PROSPECTOR-2 will proceed immediately upon completion of PROSPECTOR-1, provided the following criteria are met:

1. Compliance with study medication is ≥ 80%
2. Unblinding is ≤ 10%
3. Recruitment to eligibility ratio ≥ 80%

If these criteria are not met, appropriate amendments to the study design of PROSPECTOR-2 will be made. If compliance is under 80%, additional reminders will be scheduled for participants. If unblinding exceeds 10%, the trial will be converted to open-label. If the recruitment to eligibility ratio is lower than 80%, additional strategies for recruitment may be considered.

Statistical Methods

PROSPECTOR-1: Results will be presented according to CONSORT guidelines for feasibility studies [41]. Patient characteristics and penicillin allergy history will be presented by arm using median (interquartile range) for continuous variables and count (percentage) for categorical variables. Binary outcomes will be presented as count and percentage with 95% exact confidence intervals. All outcomes (where feasible) will be presented as overall, by study arm and by setting. Exploratory efficacy outcomes will also be presented as absolute (risk difference) and relative difference (risk ratio) with 95% confidence intervals. No statistical tests will be performed. The amount and pattern of missing data will be explored.

PROSPECTOR 2: Results will be presented according to CONSORT guidelines [42]. Patient characteristics and penicillin allergy history will be presented by arm using median (interquartile range) for continuous variables and count (percentage) for categorical variables. The primary analysis will be on intention-to-treat basis. A generalized linear model with binomial family will be used to calculate the risk difference (identity link) and risk ratio (log link) between intervention and control.

Results will be presented with two-sided 95% confidence intervals. Models will be adjusted for stratification variables (clinical site and setting). Subgroup analysis will be performed by an inclusion of interaction term between subgroup and arm. The primary analysis will be also performed in per-protocol population. Time to adverse reaction will be evaluated using Kaplan-Meier method and Cox proportions hazards regression. A detailed statistical analysis plan will be prepared and uploaded to the ANZCTR registry listing prior to study completion.

Oversight and monitoring

Adverse Event Review Panel

A blinded independent review panel consisting of an allergist and a dermatologist will be established to review reported adverse drug reactions for both PROSPECTOR studies. A summary of the patient's reaction will be compiled, comprising a standardised symptom questionnaire and clinical photography of any rash, cutaneous or mucosal change. The review panel will provide a determination of whether the reported reaction is to be classified as an "immune-mediated adverse drug reaction". This classification will be provided to the data safety monitoring board (DSMB) for further deliberation in addition to the stipulated reports.

Data safety monitoring board (DSMB)

A DSMB will be established to review trial data and monitor the progress of each trial. The DSMB will monitor adherence to the protocol, participant recruitment, outcomes and participant safety data. They will also monitor the assumptions underlying sample size calculations for the study and alert the investigators if an increased recruitment effort is required. The DSMB will make recommendations as to whether the study should continue or be terminated, consider participant safety or other circumstances as grounds for early termination, including either compelling internal or external evidence of treatment differences or feasibility of addressing the study hypotheses (e.g. poor participant enrolment).

Ethics and dissemination

These studies were reviewed and approved by the Austin Health Human Research Ethics Committee (References Numbers: PROSPECTOR-1: HREC/99740/Austin-2023; PROSPECTOR-2: HREC/109785/Austin-2024). Additional approvals will be sought for international PROSPECTOR-2 sites prior to their participation in the study. Results will be published in peer-reviewed journals and presented at relevant conferences.

Harms

Adverse events will be recorded from time of randomisation until Day 90. Patients will be supplied with a prescription for oral corticosteroids and second-generation antihistamines upon hospital discharge, only to be used in the event of an immune-mediated positive oral challenge, as instructed by the site investigators at time of the day 1, 5,7 and 14 reviews. Participants will be instructed by site investigators to fill this script at their own expense if required.

Patient and public involvement

No patient and/or public were involved in the study development.

Abbreviations: AAL, Antibiotic Allergy Label; BID, Twice Daily; LOS, Length of Stay; SAE, Serious Adverse Events

Competing Interests: None declared

REFERENCES

1. Trubiano, J.A., et al., *The prevalence and impact of antimicrobial allergies and adverse drug reactions at an Australian tertiary centre.* BMC Infect Dis, 2015. **15**: p. 572.
2. Trubiano, J.A., et al., *Antimicrobial allergy 'labels' drive inappropriate antimicrobial prescribing: lessons for stewardship.* J Antimicrob Chemother, 2016. **71**(6): p. 1715-22.
3. Picard, M., et al., *Treatment of patients with a history of penicillin allergy in a large tertiary-care academic hospital.* J Allergy Clin Immunol Pract, 2013. **1**(3): p. 252-7.
4. Borch, J.E., K.E. Andersen, and C. Bindslev-Jensen, *The prevalence of suspected and challenge-verified penicillin allergy in a university hospital population.* Basic Clin Pharmacol Toxicol, 2006. **98**(4): p. 357-62.
5. Day, C., M. Mendelson, and J. Peter, *Low self-reported penicillin allergy in South Africa-implications for global public health response.* JAC Antimicrob Resist, 2023. **5**(1): p. dlad015.
6. Li, P.H., et al., *Prevalence, Incidence, and Sensitization Profile of β-lactam Antibiotic Allergy in Hong Kong.* JAMA Netw Open, 2020. **3**(5): p. e204199.
7. Trubiano, J.A., et al., *How antibiotic allergy labels may be harming our most vulnerable patients.* Med J Aust, 2018. **208**(11): p. 469-470.
8. Li, P.H., H.Y. Chung, and C.S. Lau, *Epidemiology and outcomes of geriatric and non-geriatric patients with drug allergy labels in Hong Kong.* Hong Kong Med J, 2021. **27**(3): p. 192-197.
9. Chan, S.C.W., et al., *Prevalence and Impact of Reported Drug Allergies among Rheumatology Patients.* Diagnostics (Basel), 2020. **10**(11).
10. Macy, E. and R. Contreras, *Health care use and serious infection prevalence associated with penicillin "allergy" in hospitalized patients: A cohort study.* J Allergy Clin Immunol, 2014. **133**(3): p. 790-6.
11. MacFadden, D.R., et al., *Impact of Reported Beta-Lactam Allergy on Inpatient Outcomes: A Multicenter Prospective Cohort Study.* Clin Infect Dis, 2016. **63**(7): p. 904-910.
12. Blumenthal, K.G., et al., *Risk of meticillin resistant Staphylococcus aureus and Clostridium difficile in patients with a documented penicillin allergy: population based matched cohort study.* BMJ, 2018. **361**: p. k2400.
13. Blumenthal, K.G., et al., *The Impact of a Reported Penicillin Allergy on Surgical Site Infection Risk.* Clin Infect Dis, 2018. **66**(3): p. 329-336.
14. Knezevic, B., et al., *The revolving door: antibiotic allergy labelling in a tertiary care centre.* Intern Med J, 2016. **46**(11): p. 1276-1283.
15. Trubiano, J.A., et al., *The impact of antimicrobial allergy labels on antimicrobial usage in cancer patients.* Antimicrob Resist Infect Control, 2015. **4**: p. 23.
16. Copaescu, A.M., et al., *Efficacy of a Clinical Decision Rule to Enable Direct Oral Challenge in Patients With Low-Risk Penicillin Allergy: The PALACE Randomized Clinical Trial.* JAMA Intern Med, 2023. **183**(9): p. 944-952.
17. Trubiano, J.A., et al., *Impact of an Integrated Antibiotic Allergy Testing Program on Antimicrobial Stewardship: A Multicenter Evaluation.* Clin Infect Dis, 2017. **65**(1): p. 166-174.
18. Kan, A.K.C., et al., *Comparative Effectiveness, Safety, and Real-World Outcomes of a Nurse-Led, Protocol-Driven Penicillin Allergy Evaluation From the Hong Kong Drug Allergy Delabelling Initiative (HK-DADI).* J Allergy Clin Immunol Pract, 2023. **11**(2): p. 474-480.e2.
19. Wong, J.C.Y., et al., *Prospective, Multicenter, Head-to-Head Comparison Between Allergists Versus Nonallergists in Low-Risk Penicillin Allergy Delabeling: Effectiveness, Safety, and Quality of Life (HK-DADI2).* J Allergy Clin Immunol Pract, 2024. **12**(7): p. 1801-1808.e2.
20. Wrenn, R.H. and J.A. Trubiano, *Penicillin Allergy Impact and Management.* Infect Dis Clin North Am, 2023. **37**(4): p. 793-822.
21. Khan, D.A., et al., *Drug allergy: A 2022 practice parameter update.* J Allergy Clin Immunol, 2022.
22. Barbaud, A., et al., *EAACI/ENDA position paper on drug provocation testing.* Allergy, 2023.

23. Hjortlund, J., et al., *One-week oral challenge with penicillin in diagnosis of penicillin allergy.* Acta Derm Venereol, 2012. **92**(3): p. 307-12.
24. Hjortlund, J., et al., *Diagnosis of penicillin allergy revisited: the value of case history, skin testing, specific IgE and prolonged challenge.* Allergy, 2013. **68**(8): p. 1057-64.
25. Mori, F., et al., *Amoxicillin allergy in children: five-day drug provocation test in the diagnosis of nonimmediate reactions.* J Allergy Clin Immunol Pract, 2015. **3**(3): p. 375-80 e1.
26. Ratzon, R., et al., *Impact of an extended challenge on the effectiveness of beta-lactam hypersensitivity investigation.* Ann Allergy Asthma Immunol, 2016. **116**(4): p. 329-33.
27. Fransson, S., et al., *The Importance of Prolonged Provocation in Drug Allergy - Results From a Danish Allergy Clinic.* J Allergy Clin Immunol Pract, 2017. **5**(5): p. 1394-1401.
28. Lezmi, G., et al., *Non-immediate-reading skin tests and prolonged challenges in non-immediate hypersensitivity to beta-lactams in children.* Pediatr Allergy Immunol, 2018. **29**(1): p. 84-89.
29. Borch, J.E. and C. Bindslev-Jensen, *Full-course drug challenge test in the diagnosis of delayed allergic reactions to penicillin.* Int Arch Allergy Immunol, 2011. **155**(3): p. 271-4.
30. Fransson, S., et al., *Safe De-Labeling of Patients at Low Risk of Penicillin Allergy in Denmark.* Int Arch Allergy Immunol, 2022. **183**(6): p. 640-650.
31. Boysen, J., Garvey, L., *Retrospective evaluation of the PEN-FAST tool in Patients Undergoing Prolonged Drug Provocation*, in *EAACI*. 2023.
32. Mendelson, L.M., et al., *Routine elective penicillin allergy skin testing in children and adolescents: study of sensitization.* J Allergy Clin Immunol, 1984. **73**(1 Pt 1): p. 76-81.
33. Pichichero, M.E. and D.M. Pichichero, *Diagnosis of penicillin, amoxicillin, and cephalosporin allergy: reliability of examination assessed by skin testing and oral challenge.* J Pediatr, 1998. **132**(1): p. 137-43.
34. Solensky, R., H.S. Earl, and R.S. Gruchalla, *Lack of penicillin resensitization in patients with a history of penicillin allergy after receiving repeated penicillin courses.* Arch Intern Med, 2002. **162**(7): p. 822-6.
35. Dorman, S.M., S. Seth, and D.A. Khan, *Risk of Allergic Reactions to Recurrent Intravenous Penicillin Administration in Penicillin Skin Test Negative Patients.* J Allergy Clin Immunol Pract, 2018. **6**(1): p. 196-200.
36. Garcia Rodriguez, R., et al., *Provocation Tests in Nonimmediate Hypersensitivity Reactions to beta-Lactam Antibiotics in Children: Are Extended Challenges Needed?* J Allergy Clin Immunol Pract, 2019. **7**(1): p. 265-269.
37. Trubiano, J.A., et al., *Development and Validation of a Penicillin Allergy Clinical Decision Rule.* JAMA Intern Med, 2020. **180**(5): p. 745-752.
38. Teare, M.D., et al., *Sample size requirements to estimate key design parameters from external pilot randomised controlled trials: a simulation study.* Trials, 2014. **15**: p. 264.
39. Mitri, E., et al., *Low-risk penicillin allergy delabeling: a scoping review of direct oral challenge practice, implementation, and multi-disciplinary approaches.* Expert Rev Anti Infect Ther, 2024. **22**(1-3): p. 59-69.
40. Baur, D., et al., *Effect of antibiotic stewardship on the incidence of infection and colonisation with antibiotic-resistant bacteria and Clostridium difficile infection: a systematic review and meta-analysis.* Lancet Infect Dis, 2017. **17**(9): p. 990-1001.
41. Eldridge, S.M., et al., *CONSORT 2010 statement: extension to randomised pilot and feasibility trials.* Pilot Feasibility Stud, 2016. **2**: p. 64.
42. Schulz, K.F., et al., *CONSORT 2010 statement: updated guidelines for reporting parallel group randomized trials.* Ann Intern Med, 2010. **152**(11): p. 726-32.
43. Baiardini, I., et al., *Development and validation of the Drug Hypersensitivity Quality of Life Questionnaire.* Ann Allergy Asthma Immunol, 2011. **106**(4): p. 330-5.

Figure legends:

Figure 1: Study Design for PROSPECTOR-1 and 2 studies.

[a]penicillin "unspecified", penicillin VK, penicillin G, amoxicillin, ampicillin, flucloxacillin, dicloxacillin, cloxacillin, mecillinam, pivmecillinam, pivampicillin

Abbreviations: BID, twice daily

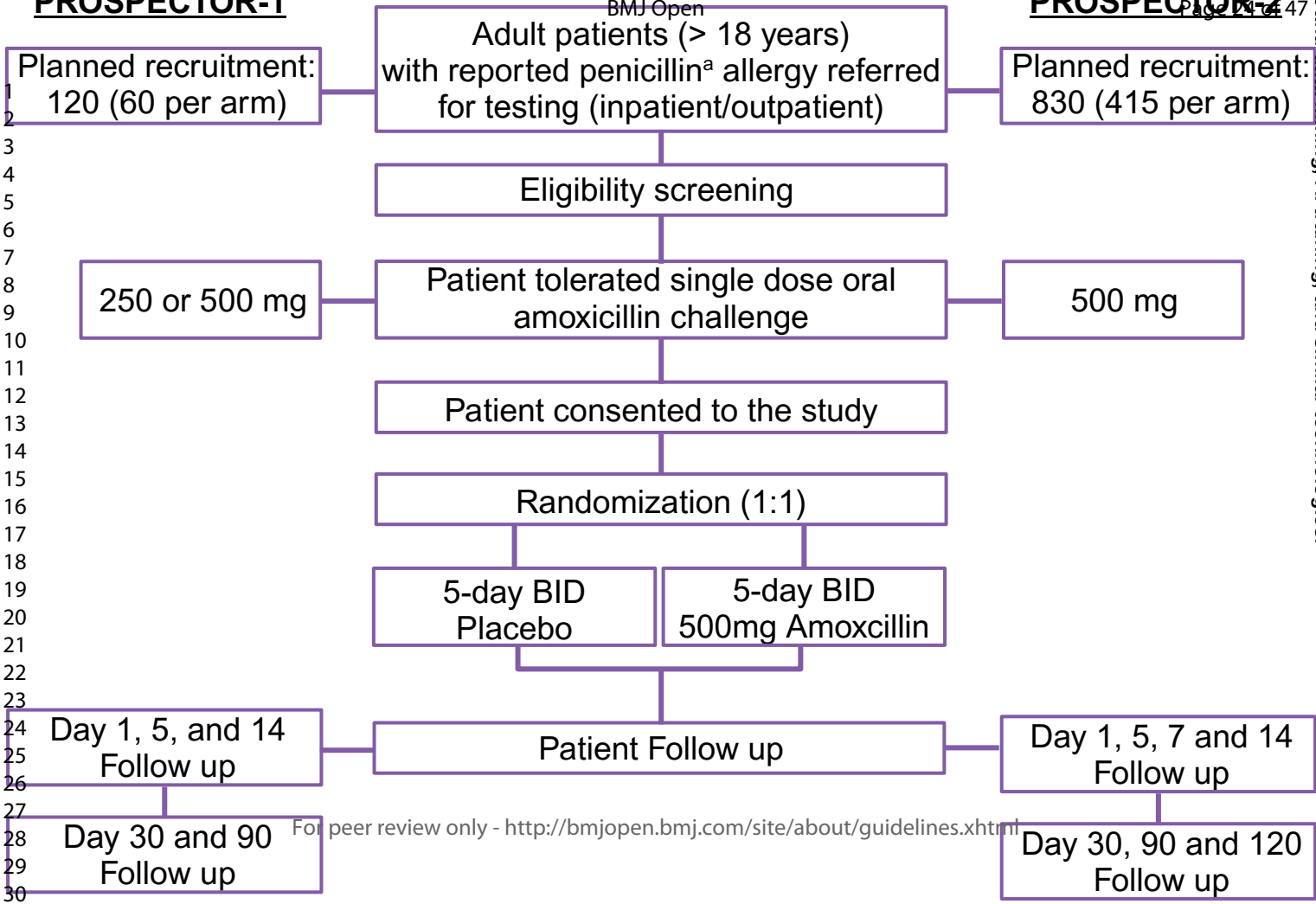
Table 1. Inclusion and Exclusion Criteria

Inclusion Criteria
1. Adults patients referred to the inpatient or outpatient allergy services for a suspected penicillin allergy with an immune-related allergy history of delayed (> 6 hours after first dose of drug administration) or unknown timing, who tolerate first single-dose of an oral amoxicillin challenge
2. Willing and able to give consent and undergo telehealth/telephone review
Exclusion Criteria
1. Patient age is < 18 years
2. Any other illness that, in the investigator's judgement, will substantially increase the risk associated with subject's participation in this study
3. Stevens-Johnson Syndrome or Toxic Epidermal Necrolysis to beta-lactam
4. Inpatients concurrently receiving or likely to receive a beta-lactam antibiotic therapy during the 14-day study period
5. Concurrent use of antihistamines and systemic steroid therapy (i.e. > 10mg daily) **[PROSPECTOR-2]**

Table 2. Secondary outcome measures for PROSPECTOR studies

	PROSPECTOR-1	**PROSPECTOR-2**
Positive Oral Challenge	Up to day 7 following the first hospital administered single dose	Up to day 14 following the first hospital administered single dose
Feasibility	- Recruitment rate per site (recruitment/site/month) - Randomisation to recruitment ratio (n, %) - Withdrawal (n, %) - Loss to follow-up (n, %) - Missing data - Protocol compliance (n, %)	Not applicable
Safety	- Severe adverse reaction (n, %) - Immune-mediated adverse event or severe adverse drug reaction (n, %) - Non-immune mediated adverse event (n, %) - Any cutaneous adverse reaction (n, %)	- Immediate severe adverse reaction (anaphylaxis or death) (n, %) - Delayed adverse reaction (severe cutaneous adverse reaction) (n, %) - Non-immune mediated adverse event (n, %) - Grade 3 or 4 adverse reactions as defined by World Allergy Organization (ref: Sanchez-Borges et al, 2019) (n, %) - Any cutaneous adverse reactions (n, %)

Efficacy	- *C. difficile* infection at day-30 and -90 (n, %) - Isolation of a multidrug resistant infection at day-30 and -90 (n, %)	- *C. difficile* infection at day-30, -90 and -120 (n, %) - Multidrug resistant infection at day-30, -90 and -120 (n, %) - Multidrug resistant colonisation at day-30, -90 and -120 (n, %)
Cost Effectiveness	cost effectiveness of placebo versus open label trial	Cost effectiveness analysis of prolonged versus single dose oral challenge
Quality of Life	Not applicable	Health-related quality of life outcome, measure by shortened Drug Hypersensitivity Quality of Life Questionnaire (DrHy-Q) (**Supplementary Table 3**) at day 0 and day 90.



SUPPLEMENTARY Materials

Supplementary Table 1. List of study centres for PROSPECTOR-1 and PROSPECTOR-2

Site	Address	Site PI	PROSPECTOR-1	PROSPECTOR-2
Austin Health	145 Studley Road, Heidelberg VIC 3084, Australia	Prof Jason Trubiano	X	X
Peter MacCallum Cancer Centre	305 Grattan Street, Melbourne, VIC 3000, Australia	Dr Morgan Rose	X	X
Royal Melbourne Hospital	300 Grattan Street, Parkville VIC 3052, Australia	Dr Jack Godsell		X
St George Hospital	Gray Street, Kogarah, NSW 2217, Australia	Dr Richard Sullivan	X	X
Royal North Shore Hospital	Reserve Road, St Leonards NSW 2065, Australia	Prof Suran Fernando		X
Royal Brisbane and Women's Hospital	Butterfield St, Herston QLD 4006, Australia	Dr Michael Lane	X	X
Royal Adelaide Hospital	Port Rd, Adelaide SA 5000, Australia	A/Prof William Smith		X
Sir Charles Gairdner Hospital	Hospital Ave, Nedlands WA 6009, Australia	Prof Michaela Lucas		X
Montreal General Hospital	1650 Cedar Ave, Montreal, Quebec H3G 1A4, Canada	Dr Ana Copaescu		X
Allergy Clinic, Herlev and Gentofte Hospital	Gentofte Hospitalsvej 8 1. Floor, 2900 Hellerup, Denmark	Prof Lene H Garvey		X
Groot Schuur Hospital	Main Road, Observatory, Cape Town, 7935, South Africa	Prof Jonathan Peter		X
Queen Mary Hospital,	Pokfulam Road, Pokfulam, Hong Kong	Dr Philip Li		X

University of Hong Kong				
Antwerp University Hospital	Drie Eikenstraat 655, 2650 Edegem, Belgium	Prof Vito Sabato		X

Supplementary Table 2. Drug Hypersensitivity Quality of Life Questionnaire (Mak HWF et al, JACI: In Pract 2024)

Table 3. Schedule of enrolment, interventions and assessments

	Screening/Enrolment*	Treatment period		Follow up period					Notes
		Day 1	Day 5	Day 7	Day 14	Day 30	Day 90	Day	
Visit window	Day -5 to 0	(0)	(+/-1)	(+/-1)	(+/- 1)	(+/-3)	(+/-3)	(+	
Informed consent	X								Informed consent must occur prior to all study activities
Baseline demographics	X								
Baseline medical and allergy history	X								
Concomitant medications	X	X	X	X	X				Record any new medications started during the study period or within 5 days of enrolment
DrHy-Q Questionnaire	X						X		
Vital signs measurement	X	X							Vital signs to be measured at baseline on Day 0 (prior to single dose oral challenge) and post-dose (frequency at investigator's discretion)
Single dose oral penicillin challenge	X								Challenge and 1-hour post-dose observation period to be completed prior to randomisation
Review of eligibility criteria	X								Review of eligibility criteria must be performed prior to randomisation

Randomisation	X								Day 0
Dispense study medication	X								Study medication is dispensed on the day of randomisation (Day 0)
Study drug administration		Day 1 – 5 (10 doses total)							Participant to self-administer study drug twice daily starting Day 1
Participant completes dosing log		Day 1 – 5							Participant to complete dosing log. Note: If dose 1 is administered in the evening or PM of Day 1, treatment will continue until Day 6.
Study drug compliance check		X	X						Site research staff to perform compliance check and review dosing log
Adverse event monitoring		X	X	X	X	X	X	X	Site research staff to perform follow-up phone calls with participants to monitor AEs
Telehealth/telephone follow-up		X	X	X	X				
Email outcomes survey						X	X	X	
Retrieval of unused medications				X					

*Screening procedures may occur on Days -5 to Day 0, i.e. may be performed on the day of randomisation if preferred by site investigators

**Use of Shortened DrHY-Q - Drug Allergy Quality of Life Questionnaire [43]

Supplementary Table 3. Drug Hypersensitivity Quality of Life Questionnaire (Mak HWF et al, JACI: In Pract 2024)

	Not at all				**Very much**
	0	1	2	3	4
1. The problem of adverse reaction to drugs affects my life					
2. The fact that I cannot use medication safely made me feel different from others					
3. I feel anxious due to my problem of allergy reaction					
4. I feel anguished due to my problem of allergy reaction					
5. The idea of taking a medicine makes me feel anxious					

Total Score: _____

Supplementary Material 1. PROSPECTOR-1 Patient Information and Consent Form

PATIENT INFORMATION AND CONSENT FORM

Adult providing own consent

Title	Prolonged versus single dose in penicillin oral challenge testing randomized control trial – PROSPECTOR Study
Short Title	PROSPECTOR Study
Protocol Number	PROSPECTOR Version 7 dated 22 August 2023
Coordinating Principal Investigator	Professor Jason A Trubiano
Site Principal Investigator	*[Principal Investigator]*
Associate Investigator(s)	*[Associate Investigator(s)]*
Study Site	*[Location]*

Part 1 What does my participation involve?

Between 5-15% of patients in developed countries have an allergy to penicillin (Blumenthal, 2019; Trubiano 2015). However, many of these allergies disappear over time and more than 90% are not present when tested with oral challenge (with or without prior skin testing).

Among patients reported to having a delayed onset of penicillin allergy, it is not clear whether a single oral challenge (i.e. single test dose) or a prolonged challenge (i.e. multiple day test dose) is the best in revealing one's allergy status. In this study, participants will be randomised to either a single oral challenge given as routine clinical practice or a prolonged oral challenge (5 days).

1 Introduction

You are invited to take part in this research project because you have either of the following:

- a reported penicillin allergy that is delayed in onset (e.g. after 2 hours post the dose), or
- unknown in timing of allergy onset and have passed a single dose oral challenge in clinic.

A research personnel will ask for your allergy and medical history. Your healthcare team will guide you through a single-dose oral challenge during a clinic appointment. This will be completed as part of routine clinical practice. Only when you pass this challenge, you will be randomly assigned to:

Group 1: Placebo

5 days of placebo* capsules twice a day with no amoxicillin

*The placebo looks the same as an amoxicillin capsule but will not contain amoxicillin. It is a sugar-filled capsule.

Group 2: Penicillin

5 days of amoxicillin capsules twice a day

You will not know if you have the placebo or amoxicillin as both are in the same capsule form.

Upon discharge, you will be supplied with a prescription for oral corticosteroids and antihistamines to be used in the setting of an immune-mediated positive oral challenge. A site investigator will instruct you to fill this script at your own expense if required.

This *Participant Information Sheet/Consent Form* tells you about the research project. It explains the tests and treatments involved. Knowing what is involved will help you decide if you want to take part in the research.

Please read this information carefully. Ask questions about anything that you don't understand or want to know more about. Before deciding whether or not you can take part, you might want to talk about it with a relative, friend or your local doctor.

Participation in this research is voluntary. If you don't wish to take part, you don't have to. You will receive the best possible care whether or not you participate.

If you decide you want to take part in the research project, you will be asked to sign the consent section. By signing it you are telling us that you:

- Understand what you have read;
- Consent to taking part in the research project;
- Consent to having the tests and treatments that are described;
- Consent to the use of your personal and health information as described.

You will be given a copy of this *Participant Information and Consent Form* to keep.

2 What is the purpose of this research?

The purpose of this study is to determine if a single dose of penicillin or prolonged course is required to accurately diagnose a penicillin allergy.

This study has been initiated by the study doctor, Prof Jason Trubiano. It is being conducted by Austin Health, Peter MacCallum Cancer Centre, St George Hospital, Royal Brisbane and Womens, Hospital McGill University Health Centre (MUHC), Groote Schuur Hospital and Herlev and Gentofte Hospital. There are no pharmaceutical or commercial sponsorships.

3 What does participation in this research involve?

This study will compare two ways of testing penicillin allergy in patients with a delayed allergy or unknown timing. Sometimes, we do not know which method is best for managing a condition. To find out, we need to compare different methods. In this study, we will put people into two groups in order to test two different methods. To try to make the groups the same, each participant is put into a group by chance (randomly).

Your allergy and medical history will be taken by a study investigator and a validated penicillin allergy assessment tool will be completed. If you have an identified penicillin allergy and have a negative single dose challenge (i.e. least likely to be a true allergy), you will be able to participate in this study.

If you are randomly assigned to the **"treatment" or test group** you will take a 5 day course of oral amoxicillin 500mg twice per day. The longer test dose procedure has been done in hundreds of hospitals in many countries, and has been able to safely prove allergy status.

If you are randomly selected to go into the **"no treatment" or control group,** you will receive a placebo capsule for 5 days which does not include penicillin. A single dose of penicillin only is used in many countries to prove or disprove penicillin allergy.

Regardless of the group you are in, the capsules will look the same. In the **"no treatment"** or **control group** this will be a **"placebo"** which does not contain penicillin. In the **"treatment"** or intervention group this will be amoxicillin 500mg. The doctors looking after you will not know which group you have been allocated to.

Participants in both groups will be followed up on day 1, 5 and 14 post testing with a short telephone call. You will also be asked to complete a questionnaire about your allergy 30 days and 90 days after you start.

There are no additional costs associated with participating in this research project, nor will you be paid. All capsules, tests and medical care done as part of the research project will be provided to you free of charge. If you decide to participate in this research project, the study doctor will inform your local doctor of the results.

4 What do I have to do?

You do not have to do anything in particular or stop anything you might be doing. You can still take most regular medications and will not need to change your lifestyle in other ways. You can still donate blood if you want to and take part in other studies.

5 Other information about the research project

We estimate that 120 participants will be taking part in this research. There are four Australian Hospitals involved (Austin Health, Peter MacCallum Cancer Centre, St George, Royal Brisbane and Womens Hospital), one Canadian center (McGill University Health Centre (MUHC), one South African centre: Groote Schuur Hospital (Capetown, SA) and one Danish hospital: Herlev and Gentofte Hospital (Copenhagen, Denmark).

6 Do I have to take part in this research project?

Participation in any research project is voluntary. If you do not wish to take part, you do not have to. If you decide to take part and later change your mind, you are free to withdraw from the project at any stage. If you do decide to take part, you will be given this *Participant Information and Consent Form* to sign and you will be given a copy to keep. Your decision to take part or not, or to take part and then withdraw, will not affect your routine treatment, your relationship with those treating you or your relationship with *[Study Site].*

7 What are the alternatives to participation?

You do not have to take part in this research project to receive treatment at this hospital. Other options are available; these include receiving a single dose challenge or prolonged challenge depending on the local site practice. The study doctor will discuss these options with you before you decide whether you can take part in this research project. You can also discuss the options with your local doctor.

8 What are the possible benefits of taking part?

We cannot guarantee or promise that you will receive any benefits from this research; however, possible benefits may include not requiring additional testing prior to the drug challenge and avoiding multiple dose provocation.

9 What are the possible risks and disadvantages of taking part?

Medical treatments often cause side effects. You may have none, some or all of the effects listed below, and they may be mild, moderate or severe. If you have any of these side effects, or you are worried about them, talk with study doctor. The study doctor will also be looking out for side effects. There may be side effects that the researchers do not expect or do not know about and that may be serious. Tell the study doctor immediately about any new or unusual symptoms.

Many side effects go away shortly after treatment ends. However, sometimes side effects can be serious, long lasting or permanent. If a severe side effect or reaction occurs, your doctor may need to stop the treatment. The doctor will discuss with you the best way of managing any side effects.

The types of side effects include allergic reactions such as mild rash (i.e. 2 in 100) or anaphylaxis (i.e.1 in 10,000). Anaphylaxis is a serious allergic response, which usually involves more than one system in the body. Symptoms such as hives, swelling and trouble breathing usually begin 5 - 30 minutes after exposure to an allergen and may lead to anaphylactic shock which can be fatal if not treated immediately. Serious reactions such as anaphylaxis have not been reported with this test dose procedure but are theoretically possible. Other side effects such as itch, nausea, vomiting, diarrhea, abnormalities of liver or kidney tests are also possible, although very unlikely.

10 What if new information arises during this research project?

Sometimes during the course of a research project, new information becomes available about the treatment that is being studied. If this happens, your study doctor will tell you about it and discuss whether you want to continue to participate in the research project. If you decide to withdraw, your study doctor will plan for your regular health care to continue. If you decide that you can continue in the research project you will be asked to sign an updated consent form.

If new information does become available, your study doctor might consider it to be best for you to stop the research project. If this happens, the doctor will explain the reasons and arrange for your regular health care to continue.

11 Can I have other treatments during this research project?

You may not be able to take some or all of the medications or treatments you have been taking for your condition or for other reasons. It is important to tell your study doctor and the study staff about any treatments or medications that you may be taking, including over-the-counter medications, vitamins or herbal remedies, acupuncture or other alternative treatments. You should also tell your study doctor about any changes to these during your participation in the study. Your study doctor will also explain which treatments or medications you may need to stop when you are in the study.

12 What if I withdraw from this research project?

If you decide to withdraw from the study, please let one of research team know. This means that they can discuss any health risks or special requirements that may come with withdrawing.

If you do withdraw during the study, the study doctor and relevant study staff will not collect additional personal information from you, although personal information already collected will be retained to ensure that the results of the research project can be measured properly and to comply with law. You should be aware that data collected by the researchers up to the time you withdraw will be part of the research project results. If you do agree with this, you must tell the research team before you join the research project.

13 Could this research project be stopped unexpectedly?

This research project may be stopped unexpectedly for a variety of reasons. These may include reasons such as:

- Unacceptable side effects;
- The drug/treatment/device being shown not to be effective;
- The drug/treatment/device being shown to work and not need further testing;
- Decisions made in the commercial interests of the sponsor or by local regulatory/health authorities.

14 What happens when the research project ends?

When the study is finished, a report will be completed and published in the scientific literature. If you would like a report of the findings, please let one of the research team know.

Part 2 How is the research project being conducted?

15 What will happen to the information about me?

By signing the consent form you agree to the study doctor and relevant research staff collecting and using personal information about you for the research. Any information that can identify you will remain confidential. Your information will be entered into a secure database as a study number with no personally identifiable information. Only study investigators will have access to it. Your information will only be used for the purpose of this research project and it will only be disclosed with your permission, except as required by law.

Information about you may be obtained from your health records held at this hospital and other health services for the purpose of this research. By signing the consent form you agree to the study team accessing health records if they are relevant to participation in this research project. Your health records and any information collected and stored by the study doctor during the research project may be reviewed for the purpose of verifying the procedures and the data. By signing the Consent Form, you give permission for the release of, or access to, this confidential information to the study team.

We expect that the results of this research project will be published and or presented in a number of ways. In any publication and/or presentation, information will be provided in such a way that you cannot be identified, except with your permission. No identifying information about the people who took part will be included. Information about participation in this research project may be recorded in your health records.

In accordance with relevant Australian and/or Victorian privacy and other relevant laws, you have the right to request access to the information collected and stored by the study team. You also have the right to request that any information with which you disagree be corrected. Please contact the study team member named at the end of this document if you would like to access this information.

16 Who is organising and funding the research?

This research project is being conducted by Prof Jason Trubiano of Austin Health. In addition, if knowledge leant through this research leads to discoveries that are of commercial value to the study doctors or their institutions, there will be no financial benefit to you or your family from these discoveries. No member of the research team will receive a personal financial benefit from your involvement in this research project (other than their ordinary wages).

17 Who has reviewed the research project?

All research in Australia involving humans is reviewed by an independent group of people called a Human Research Ethics Committee (HREC). The ethical aspects of this research project have been approved by the HREC of Austin Health. This project will be carried out according to the *National Statement on Ethical Conduct in Human Research (2007)*. This statement has been developed to protect the interests of people who agree to participate in human research studies.

18 Further information and who to contact

The person you may need to contact will depend on the nature of your query.

If you want any further information concerning this project or if you have any medical problems that may be related to your involvement in the project (for example, any side effects), you can contact the principal study doctor, *[Site Study Doctor]* at *[contact details]*, quoting "PROSPECTOR Study" in the email title or any of the following people:

Clinical contact person

Name	*[Name]*
Position	*[Position]*

Telephone	*[Contact number]*
Email	*[Email]*

For matters relating to research at the site at which you are participating, the details of the local site complaints person are:

Complaints contact person

Position	*[Position]*
Telephone	*[Contact Number]*
Email	*[Email]*

If you have any complaints about any aspect of the project, the way it is being conducted or any questions about being a research participant in general, then you may contact:

Reviewing HREC approving this research and HREC Executive Officer details

Reviewing HREC name	Austin Health Human Research Ethics Committee
HREC Executive Officer	Manager, Office for Research
Telephone	+61 3 9496 4035
Email	research@austin.org.au

Consent Form

Title	Prolonged versus single dose in penicillin oral challenge testing randomized control trial – PROSPECTOR Study
Short Title	PROSPECTOR Study
Protocol Number	PROSPECTOR Version 6 dated 16 July 2023
Coordinating Principal Investigator	Professor Jason A Trubiano
Site Principal Investigator	*[Site Principal Investigator]*
Associate Investigator(s)	*[Associate Investigators]*
Study Site	*[Study Site]*

Declaration by Participant

I have read the *Participant Information Sheet* or someone has read it to me in a language that I understand.

I understand the purposes, procedures and risks of the research described in the project.

I have had an opportunity to ask questions and I am satisfied with the answers that I have received.

I believe that my participation in this study is not contrary to my best interests/my preferences and values and my social wellbeing.

I freely agree to participate in this research project as described and understand that I am free to withdraw at any time during the project without affecting my future health care.

I understand that I will be given a signed copy of this document to keep.

I give permission for my doctors, other health professionals, hospitals or laboratories outside this hospital to release information to Austin Health concerning my condition and treatment for the purposes of this project. I understand that such information will remain confidential.

Name of Participant (please print) _____

Signature _____ Date _____

Declaration - for participants unable to read the information and consent form

Witness to the informed consent process*

Name (please print) _____

Signature _____ Date _____

*Witness is not to be the Investigator, a member of the study team or their delegate. Witness must be 18 years or older.

Declaration by Study Doctor/Senior Researcher†

I have given a verbal explanation of the research project; its procedures and risks and I believe that the participant has understood that explanation.

Name of Study Doctor/

Senior Researcher† (please print) _____

Signature _____ Date _____

† A senior member of the research team must provide the explanation of, and information concerning, the research project.

Note: All parties signing the consent section must date their own signature.

Form for Withdrawal of Participation

Title	Prolonged versus single dose in penicillin oral challenge testing randomized control trial – PROSPECTOR Study
Short Title	PROSPECTOR Study
Protocol Number	PROSPECTOR Version 6 dated 16 July 2023
Coordinating Principal Investigator	Professor Jason A Trubiano
Site Principal Investigator	*[Site Principal Investigator]*
Associate Investigator(s)	*[Associate Investigators]*
Study Site	*[Study Site]*

Declaration by Participant

I wish to withdraw from participating in the above research project and understand that this withdrawal will not affect my routine treatment, my relationship with the medical staff or with the treating hospital.

Name of Participant (please print) _____

Signature _____ Date _____

Declaration by Study Doctor/Senior Researcher†

I have given a verbal explanation of the implications of withdrawal from the research project and I believe that the participant has understood that explanation.

Name of Study Doctor/

Senior Researcher† (please print) _____

Signature _____ Date _____

† A senior member of the research team must provide the explanation of, and information concerning, the research project.

Note: All parties signing the consent section must date their own signature

Supplementary Material 2. Patient Information and Consent Form for PROSPECTOR-2

Participant Information and Consent Form

Short Name of Project	PROSPECTOR-2
Full Name of Project	PROlonged versus Single dose in PEnicillin oral Challenge Testing double-blind parallel randomized placebo cOntrolled tRial
Coordinating Principal Investigator	Professor Jason Trubiano
Site Principal Investigator	*[Site Principal Investigator]*
Site Name	*[Name of site]*

What am I being invited to do?

We invite you to take part in a project that is looking at whether a single dose of penicillin or a longer course is needed to work out if someone is allergic to penicillin. You have been invited to take part because you have had either a delayed onset allergy to penicillin or an allergy of unknown timing after receiving a single dose of amoxycillin as a test dose in an oral challenge.

Around 830 people will take part in this project. They will be from hospitals around Australia and overseas.

Please read this information and feel free to ask any questions. You can take some time to make up your mind and decide if this project is right for you. You can also talk to someone you trust, like a family member, friend, or your local doctor.

What is the purpose of this project?

In this project, we will ask you about your allergy and medical history and give you a single dose of amoxicillin (with or without prior skin testing (which take approximately 30 minutes)) to see if you develop a reaction. If you do, you will be given the standard treatment by the doctor looking after you. If you don't develop a reaction, you will then be able to take part in the study if you would like to. If you decide to take part, you will be randomly assigned to one of these two groups:

Group 1: Penicillin	Group 2: Placebo
5 days of amoxicillin capsules twice a day	5 days of placebo* capsules twice a day with no amoxicillin **The placebo looks the same as an amoxicillin capsules but will not contain amoxicillin. It is a sugar-filled capsule.*

Neither you or your study doctor will know if you have the placebo or amoxicillin as both are in the same capsule form. At the end of the study period, there is a possibility that you will be unblinded (if deemed appropriate by the site principal investigator).

Upon discharge, you will be supplied with a prescription for oral corticosteroids and antihistamines to be used in the setting of an immune-mediated positive oral challenge. A site investigator will instruct you to fill this script at your own expense if required.

We have already completed a small study to see if this approach is safe.

If you decide you want to take part in the research project, you will be asked to sign the consent section. By signing it you are telling us that you:

- Understand what you have read;
- Consent to taking part in the research project;
- Consent to having the tests and treatments that are described;
- Consent to the use of your personal and health information as described.

You will be given a copy of this *Participant Information and Consent Form* to keep.

Do I have to take part and can I change my mind?

Taking part is up to you

You can decide whether you take part in this project. You can say no if you want to. Your decision won't affect your relationship with your doctor or *[Study Site]*.

You can change your mind at any time

If you do take part, you can stop at any time. If you want to stop, please tell someone in the project team. You do not have to tell us the reason.

Once you stop taking part, we will not collect any more information about you. We will keep the information we have already collected to make sure the results of the project can be measured properly.

The project might stop for other reasons

We might need to stop the project while you are taking part. If this happens, we will explain the reasons to you.

We may also ask you to stop taking part in the project if it is no longer in your best interest. If this happens, we will discuss this with you.

What do I have to do if I take part?

If you take part in this project, you will be in it for four months.

This table below outlines what you need to do in this project. For more information, please ask a member of the project team.

What part of the project?	What do I have to do?
Consenting to take part in this project	If you are happy to take part in this project, you will be asked to sign a consent form
During the project	For the first five days you will take the capsules twice a day We will contact you by telephone/telehealth four times during the study – on days 1, 5, 7 and 14 to complete a short electronic questionnaire. You will be asked to complete a questionnaire on day 30, 90 and 120 via email/telephone. We will ask you to complete a questionnaire before you start the study and again on day 90. This will take about 5 minutes

By taking part, you will help the researchers understand more about penicillin allergies. This knowledge may help people in the future.

You may not directly benefit from taking part in this project. It is possible but unknown whether this study will help determine what testing is required to accurately diagnose penicillin allergies.

If I take part, what are the possible risk?

Medical treatments often cause side effects. You may have none, some or all of the effects listed below, and they may be mild, moderate or severe. If you have any of these side effects, or you are worried about them, talk with study doctor. The study doctor will also be looking out for side effects. There may be side effects that the researchers do not expect or do not know about and that may be serious. Tell the study doctor immediately about any new or unusual symptoms.

Many side effects go away shortly after treatment ends. However, sometimes side effects can be serious, long lasting or permanent. If a severe side effect or reaction occurs, your doctor may need to stop the treatment. The doctor will discuss with you the best way of managing any side effects.

The types of side effects include allergic reactions such as mild rash (i.e. 2 in 100) or anaphylaxis (i.e.1 in 10,000). Anaphylaxis is a serious allergic response, which usually involves more than one system in the body. Symptoms such as hives, swelling and trouble breathing usually begin 5 - 30 minutes after exposure to an allergen and may lead to anaphylactic shock which can be fatal if not treated immediately. Serious reactions such as anaphylaxis have not been reported with this test dose procedure but are theoretically possible. Other side effects such as itch, nausea, vomiting, diarrhea, abnormalities of liver or kidney tests are also possible, although very unlikely.

If I take part, what will happen to my information and samples?

Collecting your information

We will collect information for the project from your medical records and directly from you.

Keeping your information safe

To keep your information safe, we will:

follow all relevant privacy requirements
keep it securely on an electronic database (University of Melbourne REDCap)
take steps to prevent anyone from accessing information that identifies you unless they need to, for example, to check it in an audit
give it a code and keep it separate from anything that could easily identify you, like your name or contact information.

You can ask us to tell you what information we have collected about you as part of this project. If your information is not correct, you can also ask us to change it.
We will keep your information for 15 years. After this, we will destroy it in accordance with hospital policy.

Sharing your information with others

We will share some of your information with others.

Sharing information with other researchers: we will share certain information from this project so that other researchers can use it in the future. These researchers may be in Australia or overseas. We will only share information that has been de-identified, no personal identifiable information will be shared.

Getting more information

If you would like to know more about how we will collect, store and share your information as part of this project, please ask one of the research team.

Who is running and paying for this project?

This project is being run by Prof Jason Trubiano of Austin Health. In addition, if knowledge leant through this research leads to discoveries that are of commercial value to the study doctors or their institutions, there will be no financial benefit to you or your family from these discoveries. No member of the research team will receive a personal financial benefit from your involvement in this research project (other than their ordinary wages).

Who has approved this project?

The Austin Health Human Research Ethics Committee has approved this project. This committee makes sure that this project meets Australian ethical standards for research that involves people.

Complaints about how this project is being run

If you have any complaints about how this project is being run, please contact:

Name: Manager, Discovery and Innovation Unit

Contact details: Tel: (03) 9496 4090 Email: feedback@austin.org.au

Where can I find more information?

Thank you for taking the time to read this information about our project. The person you may need to contact will depend on the nature of your query. If you want any further information concerning this project or if you have any medical problems which may be related to your involvement in the project (for example, any side effects), You can contact a member of the project team at any time to ask questions.

Name *Role* *Contact details (phone number preferred)*

Name *Role* *Contact details (phone number preferred)*

Signature Page

Short Name of Project	PROSPECTOR-2
Full Name of Project	PROlonged versus Single dose in PEnicillin oral Challenge Testing double-blind parallel randomized placebo cOntrolled tRial
Coordinating Principal Investigator	Professor Jason Trubiano
Site Principal Investigator	*[Site Principal Investigator]*
Site Name	*[Name of site]*

Consent to take part in this project

By signing this consent form, I acknowledge that:
I freely agree to take part in this project
I understand that I can stop taking part in the project at any time
I have read, or have had read to me, the information provided about this project and understand what is involved
I have had the opportunity to consider the information, ask questions and am satisfied with the answers I received
I give permission for my medical records to be accessed for the purposes of this project

Person taking part in the project

Signature: _____ Date: _____

Name: _____

Person conducting the informed consent discussion

I have explained the research project, its procedures and risks to the participant and I believe they have understood that explanation.

Signature: _____ Date: _____

Name: _____

Each person must sign and personally date this consent form

Form of Withdrawal of Participation

Short Name of Project	PROSPECTOR-2
Full Name of Project	PROlonged versus Single dose in PEnicillin oral Challenge Testing double-blind parallel randomized placebo cOntrolled tRial
Coordinating Principal Investigator	Professor Jason Trubiano
Site Principal Investigator	*[Site Principal Investigator]*
Site Name	*[Name of site]*

Declaration by Participant

I wish to withdraw from participating in the above research project and understand that this withdrawal will not affect my routine treatment, my relationship with the medical staff or with the treating hospital.

Signature: _____ Date: _____

Name: _____

Declaration by Study Doctor/Senior Researcher†

I have given a verbal explanation of the implications of withdrawal from the research project and I believe that the participant has understood that explanation.

Signature: _____ Date: _____

Name: _____

Each person must sign and personally date this withdrawal of participation form