Stratifying risk of infection and response to therapy in patients with myeloma: a prognostic study

Ilaria J Chicca,[1†] Jennifer LJ Heaney,[1†] Gulnaz Iqbal,[2] Janet A Dunn,[2] Stella Bowcock,[3] Tim Planche,[4] Guy Pratt,[5] Kwee Yong,[6] Eric Low,[7] Jill Wood,[2] Kerry Raynes,[2] Helen Higgins[2] and Mark T Drayson[1*]

[1]Clinical Immunology Service, University of Birmingham, Birmingham, UK
[2]Warwick Clinical Trials Unit, University of Warwick, Coventry, UK
[3]King's College Hospital NHS Foundation Trust, London, UK
[4]Department of Medical Microbiology, St George's, University of London, London, UK
[5]Department of Haematology, University Hospitals Birmingham NHS Foundation Trust, Birmingham, UK
[6]Department of Haematology, University College London Cancer Institute, London, UK
[7]Patient Advocacy, Myeloma UK, Edinburgh, UK

*Corresponding author M.T.Drayson@bham.ac.uk
†These authors contributed equally

Declared competing interests of authors: Mark T Drayson reports personal fees from Abingdon Health Ltd (York, UK), outside the submitted work.

Published December 2020
DOI: 10.3310/eme07100

Scientific summary

Infection risk and therapy response in myeloma patients

Efficacy and Mechanism Evaluation 2020; Vol. 7: No. 10
DOI: 10.3310/eme07100

NIHR Journals Library www.journalslibrary.nihr.ac.uk

Scientific summary

Background

Multiple myeloma is a cancer of antibody-secreting plasma cells in bone marrow. Multiple myeloma causes anaemia, lytic bone lesions and fractures, kidney damage and severe bacterial infections resultant of profound immunodeficiency. There are 5500 new UK multiple myeloma cases per year, and 10% of patients registered onto UK-based Medical Research Council trials (from 1980 to 2002) die within 60 days of trial entry: almost half of these deaths are attributable to bacterial infection. Antibiotic prophylaxis is an obvious strategy to prevent infection, hospital admission and early death, but ambivalence to antibiotic use exists because of the risks of increasing antibiotic resistance and health-care-associated infections. The Tackling Early Morbidity and Mortality in Myeloma (TEAMM) trial assessed the risks, benefits and health economics of once daily use of levofloxacin for 12 weeks (i.e. antibiotic prophylaxis) in newly diagnosed multiple myeloma patients by a prospective, multicentre, randomised, double-blind, placebo-controlled trial. Interactions between the triad of multiple myeloma disease activity, immunity and infection were central to the TEAMM trial (*Figure a*), which aimed to prevent infection and early death in multiple myeloma patients. Active multiple myeloma profoundly suppresses both innate immunity, including phagocyte function, and adaptive immunity, including antibody production, causing a heavy burden of infection-associated morbidity and mortality. In addition, infections delay administration of anti-multiple myeloma therapy, and inflammation associated with infection is thought to nurture multiple myeloma activity and resistance to anti-multiple myeloma therapy.

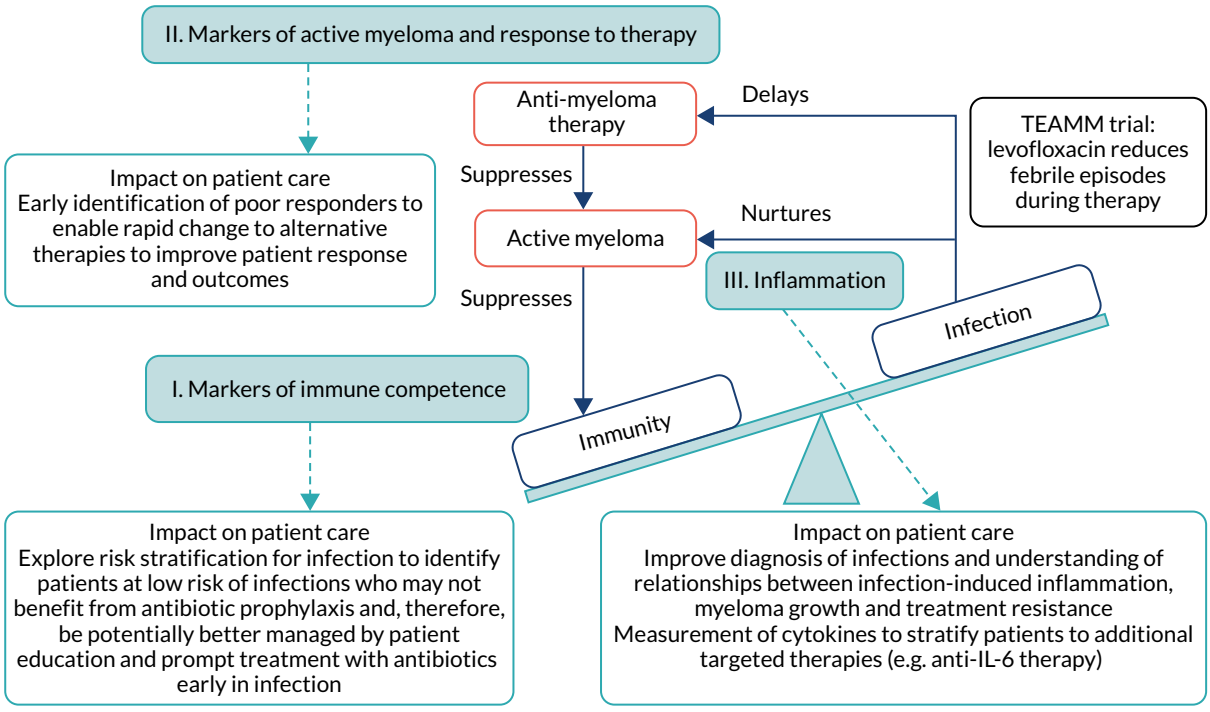

FIGURE a The cycle of immunity–infection–active disease in multiple myeloma. Active myeloma and anti-myeloma therapy suppress immunity. Infections delay administration of anti-myeloma therapy, and inflammation associated with infection is thought to nurture myeloma activity and resistance to anti-myeloma therapy. These interactions are central to the TEAMM trial, which aims to prevent infections during anti-myeloma therapy and help break the cycle of immunity–infection–myeloma activity that fosters the maintenance of active myeloma. The TEAMM trial measured three core aspects relating to this cycle (shown in the light blue boxes), with identifiable routes to patient impact. The objective was to understand the mechanisms underlying these interactions, and to provide further routes and refinement of interventions through stratification and identification strategies. IL-6, interleukin 6.

The TEAMM trial team collected five serum samples from 977 patients, from entry to 16 weeks post diagnosis. The aim of the present Efficacy and Mechanism Evaluation programme-funded study was to investigate the interaction between myeloma disease activity, immune competence and occurrence of infections in order to identify the patients most at risk of infections and, therefore, help inform future guidance for use of antibiotics for myeloma patients which, in turn, could improve their responses to anti-myeloma therapy and progression-free and overall survival.

Objectives

Objective 1

Objective 1 was to measure biomarkers of immune competence to develop risk stratification of patients for infection so that the decision to prescribe antibiotic prophylaxis can be personalised.

The objective 1 measures were:

- levels of serum immunoglobulins [specifically serum polyclonal immunoglobulin type G (IgG)] against –
  - bacterial antigens (such as pneumococcus, meningococcus, *Haemophilus influenza* type b, and tetanus and diphtheria toxoids)
  - viral antigens (such as influenza strain B, H1N1 and H3N2)
- serum biomarkers of neutrophil activity (such as neutrophil elastase, matrix metallopeptidase 9, calprotectin, lactoferrin and neutrophil extracellular traps)
- levels of the serum anti-inflammatory cytokine interleukin 10.

Objective 2

Objective 2 was to measure biomarkers of myeloma activity to sensitively measure the speed and the depth of the myeloma response.

The objective 2 measures were serum biomarkers:

- for measurement of malignant and uninvolved immunoglobulins
- of myeloma disease activity soluble CD138 (also known as syndecan-1).

Objective 3

Objective 3 was to measure biomarkers of inflammation to identify patients who may be at risk of poor treatment responses and patients who may benefit from additional targeted therapies.

The objective 3 measures were:

- levels of the serum proinflammatory cytokines interleukin 6 and interleukin 8
- levels of the serum biomarker of bacterial infections procalcitonin.

Methods

The TEAMM trial recruited 977 patients aged $\geq$ 21 years with a newly diagnosed symptomatic myeloma, and within 14 days of starting active myeloma treatment. Blood samples from patients were collected at randomisation, then every 4 weeks for 16 weeks and 1 year after randomisation. The primary outcome in TEAMM trial was time to first febrile episode or death from all causes within the first 12 weeks of trial treatment. For the purpose of the Efficacy and Mechanism Evaluation programme-funded project, two cohorts of healthy volunteers were used in the assays to identify normal ranges of biomarkers to

be investigated. Serum anti-bacterial antibodies were measured using an in-house multibeads Luminex® (R&D Systems, Minneapolis, MN, USA) assay to measure immunoglobulins against 19 antigens (12 pneumococcal serotypes, four meningococcal serotypes, *Haemophilus influenza* type b, tetanus and diphtheria toxoids). A commercially available multibeads Luminex assay was used for the quantification of cytokines interleukin 6, interleukin 8 and interleukin 10. Detection and measurement of serum levels of neutrophil biomarkers, soluble CD138 and procalcitonin in TEAMM trial patients was performed by sandwich enzyme-linked immunosorbent assay. Levels of anti-viral antibodies were measured in selected TEAMM trial patients by a haemagglutination assay.

Objective 1 key findings

At baseline, polyclonal immunoglobulins were below the normal range (i.e. IgG was 43%; IgA was 14% and IgM was 19% of the median in healthy donors) in > 75% of patients (IgG, 76%; IgA, 83%; and IgM, 90%). Levels of polyclonal IgG were further decreased during therapy (–22% at 16 weeks), but levels recovered at 1 year (+ 51% compared with baseline).

For 18 out of 19 bacterial antigen targets, levels of specific IgG were more suppressed substantially (an average of 19% of healthy median levels) than levels of total polyclonal IgG (43% of healthy median levels). A significantly lower proportion of myeloma patients (median 17%) demonstrated protective levels against all bacterial antigens than healthy donors (median 59%; $p < 0.05$). At disease presentation, < 6% of multiple myeloma patients had serum IgG antibodies above the World Health Organization's 35 µg/ml protective threshold for at least 8 of the 12 investigated pneumococcal serotypes. Additionally, before treatment, < 20%, 21%, 40% and 13% of patients exhibited protective levels of anti-meningococcal serotypes, *Haemophilus influenza* type b polysaccharide, tetanus and diphtheria antibodies, respectively. The median levels of anti-bacterial antibodies were below the threshold for 18 out of the 19 bacterial antigens tested.

A higher proportion of patients aged < 65 years were protected against meningococcal serotypes, *Haemophilus influenza* and tetanus than those patients aged ≥ 65 years. Conversely, a higher proportion of patients in the ≥ 65-year-old age group were protected against pneumococcal serotypes compared with those patients aged < 65 years, reflecting the pneumococcal polysaccharide vaccination (i.e. PPV23) programme in the UK for those people aged ≥ 65 years, that is people vaccinated with PPV23 have better protection (i.e. immune response) against pneumococcal serotypes. However, the majority of older patients still failed to meet protective levels (median 75%). Patients in the bottom tertile for pneumococcal serotype 19A (Pn19A; i.e. with levels < 0.06 µg/ml) had a higher incidence of febrile episodes. However, pneumococcal serotype 19A was not a significant factor for risk of infections (febrile and/or non-febrile episodes) within 12 weeks of starting the trial treatment (hazard ratio 0.95, 95% confidence interval 0.81 to 1.12). None of the anti-bacterial antibody levels was found to be important predictors for risk of infections. However, a significant benefit was observed in patients with higher levels of polyclonal immunoglobulin IgG and IgM ($p = 0.04$ and $p = 0.05$, respectively). This benefit was retained after adjusting for age of the patient for IgG ($p = 0.05$), IgM ($p = 0.02$) and was present for IgA ($p = 0.03$). The benefit for patients with higher levels of polyclonal immunoglobulins remained evident after adjusting for treatment pathways (IgG, $p = 0.05$; IgM, $p = 0.03$; and IgA, $p = 0.04$). There was an overall survival benefit up to 12 months for patients with pneumococcal serotype 7 (Pn7F) levels of ≥ 0.35µg/ml ($p = 0.03$), this benefit was no longer significant when adjusted for age of the patient, but patients with pneumococcal serotype 5 (Pn5) levels of ≥ 0.35µg/ml had better survival compared with patients with pneumococcal serotype 5 levels < 0.35µg/ml ($p = 0.03$). In addition, in age-adjusted analyses, polyclonal IgG levels ≥ 6g/l were associated with better survival at 12 months in both age groups ($p = 0.04$).

In a subcohort of patients who received influenza vaccine after diagnosis, anti-flu antibodies increased in response to vaccination in some but not all patients (< 60% of patients had protective levels of antibodies); at 1 year from diagnosis levels had diminished (14% of patients had protective levels of antibodies). Anti-flu antibodies post vaccination were correlated with some [in particular tetanus and pneumococcal serotype 4 (Pn4) antibodies], but not all anti-bacterial antibodies.

Biomarkers of neutrophil activity were elevated in myeloma patients compared with healthy controls. The majority of patients (i.e. 61–97%) had biomarker levels above the median levels of those of healthy controls (lactoferrin: myeloma patients 7.65 µg/ml vs. healthy controls 3.24 µg/ml; matrix metallopeptidase 9: myeloma patients 8.07 µg/ml vs. healthy controls 5.20 µg/ml; neutrophil elastase: myeloma patients 2.83 µg/ml vs. healthy controls 0.16 µg/ml; neutrophil extracellular traps: myeloma patients 0.53 OD vs. healthy controls 0.4 OD; and calprotectin: myeloma patients 0.26 µg/ml vs. healthy controls 0.11 µg/ml. A strong relationship was observed between two of the biomarkers: neutrophil elastase and calprotectin. No associations were found between levels of neutrophil biomarkers and C-reactive protein, nor prevalence of infection episodes, febrile episodes and deaths.

Myeloma patients had significantly higher levels of interleukin 10 compared with those of healthy controls. At diagnosis, 95% of the myeloma patients from the TEAMM trial had interleukin 10 levels above the median concentration of healthy controls (myeloma patients: median concentration 5.7 pg/ml, range 0.41–982.1 pg/ml; healthy controls: median concentration 1.74 pg/ml, range 1.54–1.94 pg/ml). At diagnosis, interleukin 10 levels were related to infection during the trial, that is patients with interleukin 10 levels $\geq$ 10 pg/ml had a greater risk of infections than patients with interleukin 10 levels < 10 pg/ml (hazard ratio 1.49, 95% confidence interval 1.10 to 2.02; $p = 0.01$). No associations were found between interleukin 10 levels and response to therapy during the trial or up to 1 year of follow-up.

Objective 2 key findings

Levels of soluble CD138 were elevated in 72% of TEAMM trial patients compared with the healthy cohort (myeloma patients: median level 120.4 International Unit (IU)/ml, range 1.41–256 IU/ml; healthy controls: median level 36.6 IU/ml, range 18.49–57.42 IU/ml), confirming its utility as a biomarker of active disease. During treatment levels of soluble CD138 decreased every 4 weeks, that is:

- by the end of week 4 of the TEAMM trial levels of soluble CD138 were 25% lower than baseline levels
- by the end of week 8 of the TEAMM trial levels of soluble CD138 were 25% lower than week 4 levels
- by the end of week 12 of the TEAMM trial levels of soluble CD138 were 12% lower than week 8 levels
- by the end of week 16 of the TEAMM trial levels of soluble CD138 were 7% lower than week 12 levels
- by the end of week 52 (i.e. 1 year of follow-up) of the TEAMM trial levels of soluble CD138 were 17% lower than week 16 levels.

A complete response for soluble CD138 was achieved by 21% of TEAMM trial patients by 4 weeks, 40% by 16 weeks and 44% by 1 year. In comparison, a complete response for levels of myeloma monoclonal protein (also known as m-protein) and free light chains was achieved by 1% and 19% of TEAMM trial patients, respectively, by 4 weeks, 12% and 39% by 8 weeks, and 26% and 52% by 1 year. This suggests that the speed and depth of response is more sensitive when measured by soluble CD138 and free light chain levels than when measured with whole m-protein.

The soluble CD138 response correlates poorly with the free light chain response. Of the 76 TEAMM trial patients achieving a complete response at 16 weeks, in terms of percentage free light chain change,

30% of patients had a soluble CD138 complete response, 30% of patients had a soluble CD138 response within the normal range at baseline, 9% of patients achieved a very good partial response, 16% of patients had a partial response and 11% of patients showed a reduction in the levels of soluble CD138 by < 50% (minimal response, stable disease and progressive disease).

Objective 3 key findings

Procalcitonin levels were analysed in TEAMM trial patients 1 day before, or up to 3 days after, experiencing a febrile or non-febrile episode. Levels of procalcitonin were elevated in only 50% of patients suggesting that it was not a sensitive biomarker of infection in TEAMM trial patients.

At presentation, levels of cytokines interleukin 6 and 8 were lower in TEAMM trial patients than in a healthy cohort (myeloma patients vs. healthy controls: interleukin 8 median concentrations 305.9 pg/ml vs. 455.54 pg/ml; interleukin 6 median concentrations 2.28 pg/ml, vs. 4.3 pg/ml), suggesting a relative incompetence of leucocytes in myeloma patients at diagnosis that was not improved with therapy at 12 weeks or at 1 year. At baseline, lower levels of interleukin 6 were identified in poor responders (median concentration 2.95 pg/ml, range 0.35–56.18 pg/ml) than in good responders (median concentration 2.18 pg/ml, range 0.47–785.1 pg/ml), and the difference was lost after therapy (poor responders: median concentration 2.73 pg/ml, range 0.47–83.55 pg/ml; good responders: median concentration 2.56 pg/ml, range 0.47–60.93 pg/ml). Levels of interleukin were also lower in patients who had febrile and non-febrile episodes during the trial than in patients who had only non-febrile episodes (patients who had febrile and non-febrile episodes: median concentration 3.78 pg/ml, range 0.75–1314 pg/ml; patients who had only non-febrile episodes: median concentration 2.24 pg/ml, range 0.47–900.5 pg/ml).

Discussion

This study provided new evidence that reveals that functional antibodies are severely compromised in a large cohort of myeloma patients at disease presentation. These data also provide evidence that low levels of certain antibodies are associated with an increased incidence of febrile episodes and deaths. The investigation based on age revealed valuable insights into the efficacy of the current UK pneumococcal vaccination programme in older adults, with over 65-year-old vaccinated patients demonstrating higher anti-pneumococcal antibody levels than their younger unvaccinated counterparts. However, the overwhelming majority of patients still failed to meet protective levels. The analysis demonstrated no correlation between low levels of some functional antibodies and the occurrence of febrile and non-febrile episodes. However, the analysis showed correlations between low levels of some functional antibodies and the occurrence of deaths, in particular for anti-pneumococcal serotype 5 antibodies and polyclonal IgG.

The analysis suggests that TEAMM trial patients had poor levels of anti-viral antibodies at presentation and a successful response to vaccination could be achieved in only half of the patients, potentially leaving them exposed and more susceptible to flu virus infections. Alarmingly, most of the patients achieving a response after vaccination lost the protective antibodies by 1 year, which may reflect a negative effect of anti-myeloma therapy on duration as well as immediate antibody response to vaccination.

Multiple myeloma patients are rarely neutropenic, but their infection profile is similar to neutropenic individuals. The results of this study indicate levels of neutrophil elastase and calprotectin were elevated compared with a healthy population of volunteers, but no correlation was identified with levels of C-reactive protein or with the occurrence of infections in TEAMM trial patients. Overall, these results suggest that serum biomarkers of neutrophil function are not impaired in TEAMM trial patients and do not provide additional associations with infections in this cohort and functional analysis of neutrophils from whole blood should be focused on in future studies.

The analysis of cytokine interleukin 10 found elevated levels in the TEAMM trial patients at presentation and that this biomarker represents an important factor for time to infection. The results of the study suggest that interleukin 10 may be implicated in the downregulation of the immune system and have a role in the increased susceptibility to infections of myeloma patients. Therefore, monitoring levels of interleukin 10 might have implications for the management of infections in multiple myeloma patients.

The investigation of soluble CD138 confirmed that the biomarker provides an indication of myeloma disease activity and response to therapy. However, the reduction in soluble CD138 levels was different from the reduction in levels of m-protein and free light chains, suggesting that soluble CD138 is independent and might provide additional information on therapy efficacy or myeloma disease processes.

The results indicate that levels of procalcitonin, and interleukins 6 and 8, well-known biomarkers of inflammation, do not provide additional information on the interaction between myeloma disease activity and the occurrence of infections in TEAMM trial patients.

Conclusions

The outcomes of the study have highlighted new strategies for monitoring disease activity and the immune competence of myeloma patients. Overall, these data demonstrate the need to protect patients against infections not just following diagnosis but also after anti-myeloma therapy.

Risk-stratifying patients for infection based on their immune competence and their interleukin 10 levels could introduce targeted prescription of antibiotics and channel patients who may not benefit towards alternative approaches. Rather than routine prescription, this stratification could enable responsible and evidence-based recommendation of prophylactic antibiotic treatment and, as such, help balance the risk of infection from disease with the risk of health-care-associated infections.

Identifying non-responders early through measuring the levels of free light chains and soluble CD138 could allow patients to be directed to alternative therapies to gain control over active disease and reduce the risk of infection. It could prevent the pursuit of treatment that may not have long-term benefits and avoid adverse events associated with ineffective therapies; early poor responders are unlikely to have an adequate response to a full schedule of that chosen therapy and have poor outcomes with resistant disease and early relapse. Furthermore, early control of free light chain secretions by successful anti-myeloma therapy prevents further renal damage and allows recovery of renal function.

Information from this Efficacy and Mechanism Evaluation project can help inform risk stratification and patient identification strategies enable clinical practice to be responsive to individual patient needs, and ultimately increase patient's chance of successful response to therapy and chance of survival.

Trial registration

This trial is registered as ISRCTN51731976.

Funding

This project was funded by the Efficacy and Mechanism Evaluation programme, a Medical Research Council and National Institute for Health Research (NIHR) partnership, and will be published in full in *Efficacy and Mechanism Evaluation*; Vol. 7, No. 10. See the NIHR Journals Library website for further project information.

Efficacy and Mechanism Evaluation

ISSN 2050-4365 (Print)

ISSN 2050-4373 (Online)

This journal is a member of and subscribes to the principles of the Committee on Publication Ethics (COPE) (www.publicationethics.org/).

Editorial contact: journals.library@nihr.ac.uk

The full EME archive is freely available to view online at www.journalslibrary.nihr.ac.uk/eme. Print-on-demand copies can be purchased from the report pages of the NIHR Journals Library website: www.journalslibrary.nihr.ac.uk

Criteria for inclusion in the *Efficacy and Mechanism Evaluation* journal

Reports are published in *Efficacy and Mechanism Evaluation* (EME) if (1) they have resulted from work for the EME programme, and (2) they are of a sufficiently high scientific quality as assessed by the reviewers and editors.

EME programme

The Efficacy and Mechanism Evaluation (EME) programme funds ambitious studies evaluating interventions that have the potential to make a step-change in the promotion of health, treatment of disease and improvement of rehabilitation or long-term care. Within these studies, EME supports research to improve the understanding of the mechanisms of both diseases and treatments.

The programme supports translational research into a wide range of new or repurposed interventions. These may include diagnostic or prognostic tests and decision-making tools, therapeutics or psychological treatments, medical devices, and public health initiatives delivered in the NHS.

The EME programme supports clinical trials and studies with other robust designs, which test the efficacy of interventions, and which may use clinical or well-validated surrogate outcomes. It only supports studies in man and where there is adequate proof of concept. The programme encourages hypothesis-driven mechanistic studies, integrated within the efficacy study, that explore the mechanisms of action of the intervention or the disease, the cause of differing responses, or improve the understanding of adverse effects. It funds similar mechanistic studies linked to studies funded by any NIHR programme.

The EME programme is funded by the Medical Research Council (MRC) and the National Institute for Health Research (NIHR), with contributions from the Chief Scientist Office (CSO) in Scotland and National Institute for Social Care and Health Research (NISCHR) in Wales and the Health and Social Care Research and Development (HSC R&D), Public Health Agency in Northern Ireland.

This report

The research reported in this issue of the journal was funded by the EME programme as project number 14/24/04. The contractual start date was in February 2017. The final report began editorial review in March 2019 and was accepted for publication in October 2019. The authors have been wholly responsible for all data collection, analysis and interpretation, and for writing up their work. The EME editors and production house have tried to ensure the accuracy of the authors' report and would like to thank the reviewers for their constructive comments on the final report document. However, they do not accept liability for damages or losses arising from material published in this report.

This report presents independent research. The views and opinions expressed by authors in this publication are those of the authors and do not necessarily reflect those of the NHS, the NIHR, the MRC, NETSCC, the EME programme or the Department of Health and Social Care. If there are verbatim quotations included in this publication the views and opinions expressed by the interviewees are those of the interviewees and do not necessarily reflect those of the authors, those of the NHS, the NIHR, NETSCC, the EME programme or the Department of Health and Social Care.

Published by the NIHR Journals Library (www.journalslibrary.nihr.ac.uk), produced by Prepress Projects Ltd, Perth, Scotland (www.prepress-projects.co.uk).