Investigating the clinical and cost effectiveness of 4 ELISA kits for monitoring infliximab in patients with Crohn’s disease: Protocol for a validation study

Authors: Langford T, Arkir Z, Chalkidou A, Goddard K, Samaan MA, Irving PM

Abstract

Background: Currently, treatment decisions for people with Crohn's disease (CD) are based on clinical judgement and 'trial and error'. Consequently, people may continue to receive high drug dosages and experience unnecessary toxicity when it is possible to reduce or discontinue without a detrimental effect on clinical outcomes. Therapeutic drug monitoring (TDM) involves regularly testing blood samples for drug and antibody levels that could help clinicians identify the optimal treatment strategy and pre-empt treatment failure. However, heterogeneity in the assays can lead to discrepancy in results and difficulties in decision-making. Standardisation of the kits, and therefore results, would allow clinicians to optimise the use of biologics. Currently, there is also a lack of evidence for the cost-effectiveness of TDM using commercial test kits.

Objective: This study aims to analyse clinical and cost effectiveness of 4 commercial ELISA kits (LISA TRACKER, IDKmonitor, Promonitor and RIDASCREEN) to generate evidence which could support a recommendation for wider adoption in the NHS.

Methods: We propose to carry out a prospective-retrospective predictive biomarker validation study using the blood samples and clinical/utilization data collected during the ongoing SPARE trial (NCT02177071). 200 stored samples from people with CD whose disease responds to treatment with infliximab will be used along with clinical and cost data from the trial. We will investigate the relationship between drug and anti-drug antibody levels with the main clinical outcomes (relapse rate at 2 years and time spent in remission), as well as resource utilisation and quality of life.

Results: Funding is being sought to conduct this research.

Conclusions: This study will be the first study to compare the 4 ELISA kits for monitoring infliximab in patients with CD. It aims to address the uncertainties in the potential benefits of using the technologies for TDM.

Keywords: anti-drug antibodies; anti-TNF; Crohn's disease; ELISA; inflammatory bowel disease; infliximab; therapeutic drug monitoring;

Introduction

Background

Crohn's disease (CD) and ulcerative colitis are the main conditions described as inflammatory bowel disease (IBD). CD is a chronic, fluctuating inflammatory condition of the digestive tract that can affect both adults and children. The main symptoms include chronic or nocturnal diarrhoea, abdominal pain, rectal bleeding and weight loss. The disease follows an unpredictable relapse (active disease) and remission (no symptoms) course with significant variation in the pattern and complexity of symptoms. During relapse, patients often suffer substantial morbidity
and require intensive treatment, including invasive investigations, costly drugs and surgery. The prevalence of IBD in the UK is estimated to be 240,000 with CD affecting about 115,000 people [1]. In 2006, the cost of IBD to the National Health Service (NHS) was estimated at about £720 million, based on the prevalence and an average cost of £3,000 per patient per year [2]. The cost today is likely to be significantly higher with the availability of new biological therapies that have an average annual cost per patient estimated between £10,000 and £15,000 [3]. At Guy’s and St Thomas’ NHS Foundation Trust (GSTT), the IBD Service has approximately 600 patients receiving biological therapies incurring costs of £6 million per year.

Currently, there is no ‘cure’ for this lifelong condition. Drugs are used to suppress the overactive immune system in people with CD, with the intention of inducing and maintaining remission. However, 30% of patients fail to respond to first-line drugs and will then be considered for anti-tumour necrosis factor–alpha (anti-TNFα) biological therapies, such as infliximab (IFX) and adalimumab (ADAL). Anti-TNF treatment aims to induce remission and prevent relapse by targeting the inflammation-causing protein, TNFα, rather than suppressing the immune system as a whole. Despite this, loss of response (LOR) and relapse are common; the annual risk of LOR is estimated at about 13% per patient [4]. The typical response to LOR is dose intensification, however the underlying cause of the LOR is not fully understood. The main hypothesis is that some patients develop antibodies against the biologics preventing the concentrations of the drug in the patient’s bloodstream from reaching levels required to maintain remission. People whose disease responds to a TNFα inhibitor may continue receiving the same level of drug even when it may be possible (or even beneficial [5]) to reduce the dose or withdraw the drug entirely without any detrimental effect on clinical outcomes. This continued treatment may lead to people experiencing unnecessary side effects. Treatment decisions for people with CD are based on clinical judgement and ‘trial and error’. Measuring the levels of TNFα inhibitors and associated antibodies in the blood could help clinicians to identify the best treatment strategy for a person with CD; this is known as therapeutic drug monitoring (TDM).

Numerous commercial kits are available for TDM of biologics for CD. The literature is inconclusive on whether ELISA testing improves patient outcomes or is cost-effective [6-10], as they are not routinely used to optimise treatment. The optimal approach and frequency of delivering TDM is also uncertain. In a study of healthcare professionals’ routine practice, only 45% reported using TDM during maintenance therapy for patients in remission [11]. One reason for this being heterogeneity in the assays which can lead to discrepancy in results. Standardisation of the kits, and therefore results, would allow clinicians to optimise the use of biologics.

In 2016, the National Institute for Health and Care Excellence (NICE) published diagnostics guidance on TDM of TNFα inhibitors (IFX and ADAL) in CD (referred to as DG22) [12]. DG22 evaluated the clinical and cost-effectiveness of 3 enzyme-linked immunosorbent assay (ELISA) kits. The 3 commercial kits considered were LISA-TRACKER (Theradiag), Immundiagnostik (Immundiagnostik/BioHit Healthcare) and Promonitor (GRIFOLS) ELISA kits. They were considered for testing levels of TNFα inhibitors and anti-drug antibodies in two populations, people with CD whose disease responds to treatment with TNFα inhibitors and those who experience secondary LOR. DG22 found a number of limitations within the evidence identified. No studies were found to be assessing direct clinical outcomes for any of the commercially-available test kits and there was a paucity of evidence on cost-effectiveness in general. DG22
concluded that although the kits show promise, there is insufficient evidence to recommend routine adoption across the NHS.

In response to the uncertainties identified in DG22, NICE recommended future research focuses to address gaps in the current evidence and investigate potential benefits of using ELISA kits for CD treatment monitoring within the NHS. In May 2016, NICE requested King’s Technology Evaluation Centre (KiTEC), based at King’s College London (KCL), to plan and obtain funding for research that will address the uncertainties mentioned above. As part of this research, KITEC will carry out a prospective-retrospective predictive biomarker validation study to assess the clinical and cost-effectiveness of the ELISA kits. This study will use the stored samples and clinical and cost data from a multi-centre, international randomised controlled trial, the SPARE trial (NCT02177071). Results from this study would provide further impetus to carry out research on the remaining NICE research recommendations;

1. to assess analytical and clinical validity of the tests (developing standardised primary reference standards)
2. to prospectively evaluate the clinical utility of the ELISA kits in people with CD who are losing responsiveness to infliximab.

**SPARE Trial**

One of the treatment strategies used in the management of severe active CD that has not responded to conventional therapy is combination therapy, in which an immunosuppressant drug (also known as an antimetabolite) such as azathioprine, mercaptopurine or methotrexate and an anti-TNF agent called infliximab are used together. This combination is highly effective in inducing remission. The multi-national SONIC trial (Europe, Israel and North America) demonstrated that infliximab plus azathioprine combination therapy was superior to infliximab monotherapy and azathioprine monotherapy to achieve steroid-free remission and mucosal healing in anti-metabolites naïve steroid-dependent or steroid-refractory patients [13].

Despite this superiority, maintaining such combination therapy long term may generate cost and safety issues. The NICE and Scottish Medicines Consortium mandate reassessment of patients on combination therapy at 12 monthly intervals with a consideration of drug withdrawal where patients are in sustained deep remission.

However, there is currently insufficient data on relapse and recapture rates to inform such decision making [14-17]. In response to the lack of evidence, a prospective open-label, international 3-arm randomized controlled trial (RCT), the SPARE trial, was launched in October 2015 to assess the benefits of the continuation of combination therapy and the feasibility of infliximab or antimetabolites discontinuation in patients in sustained steroid free remission after prolonged treatment with a combination of infliximab and anti-metabolites. The purpose of the SPARE study, therefore, is to find the safest and most effective way for patients to discontinue their combination therapy by comparing three different withdrawal strategies:

- Continued combination therapy (immunosuppressant drug and infliximab)
- Immunosuppressant drug alone (so infliximab discontinued)
- Infliximab alone (so immunosuppressant discontinued)
This will help find out which strategy has the best chance of maintaining remission of CD and to determine the risk factors for disease flare, as well as side effects, quality of life and the impact on people's social and professional life. The aim is to be able to identify which patients for whom discontinuation of immunosuppressant drug or infliximab could be considered after one year of treatment and what would be the best treatment strategy. The SPARE trial has a planned duration of 2 years main study plus 2 years follow-up and the main co-primary outcomes are clinical relapse rate at 2 years and mean remission duration within 2 years. Secondary outcomes include time to relapse in each arm, treatment failure rate, time to treatment failure, tissue damage progression and others. The estimated completion date is January 2020.

Other Relevant Trials
There are a number of additional ongoing or recently completed trials which fulfill the requirements of our validation study and could be contacted to obtain samples. Three trials were identified, two taking place in Europe and one in America, all include populations with luminal CD treated with infliximab who have been in remission for at least 6 months.

The NOR-SWITCH trial (NCT02148640) was recently completed in 2017. 155 adult patients with CD were recruited in Norway. The blood samples collected are being stored in a biobank [18].

The Precision IFX trial (NCT02624037) is currently recruiting 800 adult and paediatric patients with CD or UC in the US who are in remission. This study is expected to be completed in December 2018.

The GIS-SUSANTI-TNF-2015 trial (NCT02994836) is currently recruiting 300 adult participants with UC or CD in Spain. The primary completion date for this trial is December 2020.

Study Objectives
The primary objective of this study is validate the kits by examining the relationship between infliximab and infliximab antibody levels as measured by the different kits in duplicate. These will be compared with the main clinical outcomes (relapse rate at 2 years and mean restricted time spent in remission).

The secondary objective is to evaluate the effect of monitoring infliximab and anti-drug antibody concentrations using serum samples on resource utilisation and health-related quality of life in patients with CD who respond to treatment with infliximab.

Methods
Interventions
Manufacturer laboratories have developed various assay procedures for TNF inhibitors and antibodies against TNF inhibitors. The LISA-TRACKER, IDKmonitor, Promonitor, and RIDASCREEN are particular examples of these essays classed as solid phase ELISAs and are intended to be used for measuring the levels of TNFα inhibitors and antibodies against TNF-alpha inhibitors in the blood of people having treatment with biologics for CD.
Study population
Patients who have been in steroid-free remission for at least 6 months and with scheduled infliximab/antimetabolites combination therapy for at least 1 year, with a scheduled infliximab treatment administered every 8 weeks for the last 6 months. More detailed inclusion and exclusion criteria are in the 'Inclusion and Exclusion Criteria' section below.

The overall SPARE target is 300 randomized patients (100 per arm) worldwide over 20 months from 70 centres. Currently, the plan of recruitment is as follows:
- France: 100 patients in 20 centres
- UK: 70 patients in 21 centres
- Sweden: 50 patients in 10 centres
- Germany: 45 patients in 10 centres
- Belgium: 35 patients in 9 centres.

The KiTEC study will focus on 2 of the 3 study arms in which participants are being treated with infliximab; participants from the other arm were excluded because they are not currently being treated with infliximab.

Study design
The retrospective-prospective study will focus on patients with luminal CD who have sustained remission and are being treated with infliximab. Assays will be prospectively performed in duplicate on 200 blood serum samples. The samples were retrospectively collected from the SPARE trial. Our primary collaborator from the SPARE trial is Dr Edouard Louis (Centre Hospitalier Universitaire de Liège, Belgium), who is also the Principal Investigator.

The SPARE trial is an open label, multi-centre trial with 3 parallel randomized arms comparing three strategies of maintenance therapy in patients in sustained clinical remission without steroids for at least 6 months and having been treated by a combination of anti-metabolites and infliximab for at least 1 year. Figure 1 illustrates the study design of the SPARE trial. Participants in study arms 1 and 3 who have sustained remission (referred to as ‘responders’) are the population focused on in this study. The SPARE trial is estimated to run for 5 years; 2 years of enrolment, 2 years of patient follow-up and 1 year of data analysis. The trial began on October 2015 and has an estimated study completion date on January 2020. The KiTEC study is planned to last 18 months and overlap with the SPARE trial.

The blood samples will be sent to Viapath Analytics, the provider of pathology services at St Thomas’ Hospital, for testing with the 4 ELISA kits under investigation. All ELISA kits will be automated on Dynex DS2 2-Plate ELISA processing system in accordance with the manufacturer’s instructions for use. Sample analysis will be completed sequentially to avoid further freeze thaw cycles. This is important to eliminate sources of variation that can be introduced due to repeated freeze thaw cycles. Pseudo-anonymised results from the testing will be compared with the results from the SPARE trial to validate the kits. Stored samples will be prepared, stored and shipped according to the SPARE lab manual. Serum samples will be centrifuged and frozen before shipping. All biological samples will be retained within the central labs (in Israel) at -80°C for at least 6 years. All SPARE trial data will be collected in an electronic case report form by staff at participating sites. The Trial Statistician, Professor Sylvie Chevet (Biostatistics and Medical
Information Department, Saint-Louis Hospital) will perform data collection and data quality controls.

**Inclusion and Exclusion Criteria**

The following inclusion criteria will be used:

- Diagnosis of luminal Crohn’s disease.
- Male or female, age > 18 years (to ensure data is comparable with other participating regions).
- Currently treated with a combination therapy with infliximab and anti-metabolites for luminal Crohn’s disease.
- Combined therapy with scheduled infliximab and anti-metabolites for at least 12 months.
- Scheduled administration of infliximab 5 mg/Kg every 8 weeks over the last 6 months.
- Anti-metabolites administered at a stable dosage for the last 6 months: at least 1 mg/Kg or 2 mg/Kg for mercaptopurine and azathioprine, respectively, or the highest tolerated dosage if intolerance to standard dose; at least 15 mg/week subcutaneously for methotrexate.
- Patients in steroid free clinical remission for at least 6 months according to retrospective assessment of the patients’ files.
- Crohn’s Disease Activity Index (CDAI) < 150 at baseline.
- Adequate contraceptive (as judged by the Principal Investigator) during the whole study for female participants of childbearing potential.
- Patients able to understand the information provided to them and to give written informed consent for the study.

*Figure 1 – flowchart of SPARE trial: three parallel arm-trial design and population of interest (denoted as ‘Responders’)*
The following exclusion criteria will be used:

- Patients who have presented a severe acute or delayed reaction to infliximab.
- Perianal fistulae as the main indication for infliximab treatment
- Active perianal/abdominal fistulae at time of inclusion, defined by active drainage
- Patients with ostomy or ileoanal pouch
- Pregnancy or planned pregnancy during the study or breastfeeding
- Inability to follow study procedures as judged by the investigator
- Non-compliant subjects.
- Participation in another therapeutic study
- Steroid use ≤6 months prior to screening
- Currently receiving steroids, immunosuppressive agents (other than purine, methotrexate), biologic treatment (other than infliximab) or thalidomide.

**Statistical Analysis**

All SPARE trial data are collected in an electronic case report form by staff at participating international sites. The data will be pseudo-anonymised. The Statistical Centre in France will perform data collection and data quality control.

The co-primary outcomes are clinical relapse rate at 2 years and mean remission duration within 2 years. The following outcomes will be also assessed: IBD Disability Index scores, CD Activity Index, adverse events, trough levels of infliximab and faecal calprotectin, direct and indirect costs associated with using or not using TDM and health-related quality of life (EQ-5D and Short Health Scale), work productivity and activity index (WPAI-CD).

**Ethics**

All serum samples analysed will be obtained during the SPARE trial. The SPARE trial was conducted in accordance with the principles of the International Conference on Harmonisation Tripartite Guideline for Good Clinical Practice. The SPARE trial was reviewed by a research ethics board at the respective site. All subject information used in this study was de-identified with respect to the subject identification number and investigational site. The informed consent process complied with the International Conference on Harmonisation Tripartite Guideline for Good Clinical Practice and all applicable regulatory requirement(s). The consent of subjects included the use of the collected data and serum for other medical purposes. Therefore, additional consent for the current study was not required.

**Results**

Funding has been sought to carry out this proposed research. This study is expected to take 18 months.

**Discussion**

The rationale behind conducting this study is to contribute to answering the questions identified by the research recommendations from DG22. If substantive evidence is generated, NICE will update its guidance to recommend the clinical use of one or more of the commercial ELISA kits for therapeutic monitoring and personalising anti-TNF alpha inhibitor treatment. NICE guidance
would encourage adoption both nationally and internationally, underpinning the potential value of this research. This technology has the potential to improve patient outcomes in terms of clinical outcomes and patient-reported heart-related quality of life, it also may be found to be cost effective. All of these are under investigation in our research.

Previous trials have found that varying infliximab in CD patients can have beneficial implications. One study, the STORI trial (NCT00571337) is an RCT in France and Belgium which suggested that steroid-free remission may be maintained after infliximab discontinuation, with more than half of the 115 patients having reached sustained steroid-free remission after infliximab treatment with antimetabolites combination therapy for one to two years [19]. This infliximab-free remission for a majority of the population will lead to substantial reductions in associated costs and side effects. This study also suggested that infliximab retreatment is safe and effective in relapsing patients. Further, in a 7 year follow-up, 21% of the population did not restart treatment with infliximab or another biologic [20]. However, the STORI trial did not have a control group of patients who were continuing infliximab treatment. Therefore, no results on when it is appropriate to recommend the withdrawal of infliximab from patients were discussed. The present studies differs in that the population in focus is those continuing to use and respond to infliximab. Validating the technologies may lead to them being used clinically for informing treatment decisions and then, further clinical studies into reduction or withdrawal of infliximab would be possible.

One advantage of utilising the SPARE trial data for our research is that a quarter of the study population will be recruited from UK-based centres. Meaning that the cost data collected as part of the health economic analysis portion of our study will be directly relevant to treatment on the NHS. However, the circumstance of this study being an add-on to an international RCT means we are highly dependent on the progress of that trial. Delays in the SPARE trial will impact on our own progress and factors affecting recruitment numbers or data quality will have a direct impact on our study. The samples obtained from the trial are to be retained for 6 years, it is highly likely we will be able to obtain the samples and conduct the validation study within this timeframe.

Conclusion and future direction
The proposed study will validate 4 commercially available ELISA test kits and potentially impact on a NICE recommendation for using the technology clinically. This research is directly answering one of the research recommendation from NICE to investigate clinical outcomes associated with using the ELISA kits for TDM in people with Crohn's disease whose disease responds to treatment with TNF-alpha inhibitors. Future studies into evaluating the clinical and cost effectiveness of the technologies prospectively in an NHS clinical environment will be a key aim of further research.

Author’s contributions
Kate Goddard, Zehra Arkir, Peter Irving and Mark Samaan led on designing the study protocol. Tom Langford led on writing the manuscript. All authors participated in critical review of the methods and read and approved the final manuscript.

Conflicts of Interest
None declared.
**Abbreviations**

ADAL: Adalimumab
CD: Crohn's disease
CDAI: Crohn's Disease Activity Index
ELISA: enzyme-linked immunosorbent assay
GSTT: Guy’s and St Thomas’ NHS Foundation Trust
IBD: inflammatory bowel disease
IFX: Infliximab
JMIR: Journal of Medical Internet Research
KCL: King’s College London
KHP: King’s Health Partners
KITEC: King’s Technology Evaluation Centre
LOR: loss of response
NHS: National Health Service
NICE: National Institute for Health and Care Excellence
RCT: randomised controlled trial
TDM: Therapeutic drug monitoring
TNF: Tumour necrosis factor
WPAI-CD: Work productivity and activity impairment questionnaire: Crohn’s disease

**References**


8. Steenholdt C, Bendtzen K, Brynskov J, Thomsen OØ, Munck LK, Christensen LA, et al. Changes in Serum Trough Levels of Infliximab During Treatment Intensification but not in


