NATIONAL INSTITUTE FOR HEALTH AND CARE EXCELLENCE

Diagnostics Assessment Programme

Therapeutic monitoring of TNF-alpha inhibitors in rheumatoid arthritis

Final scope

July 2018

1 Introduction

The Promonitor assays are manufactured by Grifols. The medical technologies topic oversight group selected and routed Promonitor for guidance development by the Diagnostics Assessment Programme on the basis of a briefing. The final scope was informed by discussions at the scoping workshop held on 6 June 2018 and the assessment subgroup meeting held on 19 June 2018. A glossary of terms and a list of abbreviations are provided in appendices A and B.

2 Description of the technologies

This section describes the properties of the diagnostic technologies based on information provided to NICE from the manufacturers and on information available in the public domain. NICE has not carried out an independent evaluation of this description.

2.1 Purpose of the medical technologies

Therapeutic drug monitoring ELISAs (enzyme-linked immunosorbent assays) for rheumatoid arthritis are intended for measuring the levels of biological disease-modifying anti-rheumatic drugs (DMARDS) and the levels of antibodies against the drug in blood samples. The drug level concentration and the anti-drug antibody levels can help to inform treatment decisions for people with rheumatoid arthritis when interpreted together along with other clinical signs and symptoms.

Biological DMARDS include, but are not limited to, TNF-α inhibitors. TNF-α inhibitors are used to inhibit the activity of the cell signalling protein, TNF-α, which promotes inflammatory responses. When the production of TNF-α is dysregulated it can lead to various inflammatory diseases, such as rheumatoid arthritis. TNF-α inhibitors, such as infliximab and adalimumab, are

therefore given to patients to inhibit TNF-α production and suppress the inflammatory response. Although TNF-α inhibitors can bring benefits to many patients with rheumatoid arthritis, there are some patients whose disease does not respond to treatment (primary non-responders). Furthermore, some patients who have disease that initially responds to treatment find their disease stops responding over time (secondary non-response). This loss of response can be caused by:

- changes in disease characteristics over time
- the presence of antibodies to TNF-α inhibitors
- fluctuations in circulating drug levels.

Presence of antibodies to TNF-α inhibitors

Treatment with biologics can lead to an immune response with anti-drug antibodies being produced by the immune system. Patients whose immune system produces anti-drug antibodies to a TNF-α inhibitor may have disease that is less likely to respond to further treatment with the drug. This is likely to be because the anti-drug antibodies bind to the TNF-α inhibitor molecules and decrease the availability of the drug in the body. In addition, the presence of anti-drug antibodies can lead to adverse events in some people.

Fluctuations in circulating drug levels

The concentration of TNF-α inhibitor in the blood of patients immediately before their next dose of TNF-α inhibitor (trough level) can vary widely between patients even though they have received the same initial dose. These variations can be caused by:

- Individual differences in drug pharmacokinetics
- Presence of anti-drug antibodies
- Concomitant administration of some immunosuppressive treatments, such as methotrexate.

The clinical scenarios in which these assays may be used include:

Remission / low disease activity	Test for drug levels and anti-drug antibodies 6 to 12 months after achieving treatment target (remission or low disease activity) to check whether continued treatment at the same dose is appropriate.
Non-response / moderate to high disease activity	**Primary non-response** Primary non-response is defined as little to no improvement in clinical signs and symptoms initially and as treatment continues.

	Secondary non-response Secondary non-response is defined as an initial response to a TNF-α inhibitor, followed by loss of efficacy. Testing could help clinicians and patients to understand the reasons why there is a non-response or loss of response. It could also indicate whether non-response could be because treatment is not being taken. Advice sought from clinical experts during scoping suggested that testing for people with primary non-response would be done no later than 3 months after starting treatment. People with secondary non-response would be tested when clinical indicators such as DAS28 suggested a loss of efficacy.

Currently, treatment decisions for rheumatoid arthritis are based on assessment of clinical indicators such as erythrocyte sedimentation rate, c-reactive protein (CRP), number of tender joints, number of swollen joints and patient global health scores, such as the disease activity score 28 (DAS28). Patients whose disease responds well to treatment with TNF-α inhibitor may continue to receive the same level of treatment even though a decrease in dose or withdrawal from the TNF-α inhibitor may be possible without any detrimental impact on clinical outcomes. This continued treatment may lead to patients experiencing side-effects of the treatment unnecessarily such as upper respiratory infections, bronchitis, bladder infections, skin infections, allergic reactions, development of autoantibodies, itching, and fever. Patients who experience a primary or secondary non-response typically switch to a biological treatment with a different mechanism of action, for example an IL-6 inhibitor such as tocilizumab.

2.2 Product properties

2.2.1 Promonitor (Grifols)

Promonitor (Grifols) is a portfolio of 8 CE marked enzyme linked immunosorbent assays (ELISAs) for the quantitative measurement of drug levels and their correlating anti-drug antibodies. The tests are done in a laboratory and can be performed manually or on an automated ELISA platform.

Assays that measure the levels of TNF-α inhibitors, adalimumab, etanercept, infliximab or golimumab, and their correlating drug-antibodies are currently available for use (table 1) and are relevant to this assessment. The assay for infliximab is validated for use with its biosimilars, CT-P13 (Remsima, Inflectra)

and SB2 (Flixabi, Renflexis). Assays for etanercept and adalimumab are being validated for use with biosimilars. In addition, assays are in development for the TNF-α inhibitor, certolizumab pegol, and its correlating antibodies.

Table 1 Promonitor ELISA kits

Name (code)	Detects	Microplate pre-coat	Secondary reagent
Promonitor-ADL-1DV (50802300DV)	Free[1] adalimumab	Anti-adalimumab human monoclonal antibody	Peroxidase labelled anti-adalimumab monoclonal antibody
Promonitor-IFX-1DV (50602300DV)	Free[1] infliximab (Remicade, and biosimilars)	Anti-TNF-α human monoclonal antibody bound to human recombinant TNF-α	Peroxidase labelled anti-infliximab monoclonal antibody
Promonitor-ETN-1DV (51102300DV)	Free[1] etanercept	Anti-etanercept human monoclonal antibody	Peroxidase labelled anti-etanercept monoclonal antibody
Promonitor-GLM-1DV (52002300DV)	Free[1] golimumab	Anti-TNF-α human monoclonal antibody bound to human recombinant TNF-α	Peroxidase-labelled anti-golimumab monoclonal antibody
Promonitor-ANTI-ADL-1DV (50902300DV)	Free[1] anti-adalimumab antibodies	Adalimumab	Peroxidase labelled adalimumab
Promonitor-ANTI-IFX-1DV (50702300DV)	Free[1] anti-infliximab antibodies	Infliximab	Peroxidase labelled infliximab
Promonitor-ANTI-ETN-1DV (51202300DV)	Free[1] anti-etanercept antibodies	Etanercept	Peroxidase labelled etanercept
Promonitor-ANTI-GLM (5210230000)	Free[1] anti-golimumab antibodies	Golimumab	Peroxidase labelled golimumab
[1] Free TNF-α inhibitor is drug that is unbound to antibody, and free anti-drug antibodies are those that are unbound to drug			

Each Promonitor kit consists of strips of pre-coated microtitre plate (96 wells), reagents, buffers, standards, controls and ELISA cover films. Additional equipment required to run the test includes; calibrated micropipettes and micropipette tips, spectrophotometer for ELISA plates and ethanol solution. It is recommended that each full run of the 96 well microtitre plate should include at least 6 calibrators and 2 controls therefore each run can contain up to 88 patients samples. It takes approximately 30 minutes to prepare the

reagents and samples plus around 3 hours to run the assay with a full plate of samples. Assays measure free drug levels or free anti-drug antibody levels.

The instructions for use recommend samples are collected before drug administration (trough levels), to minimise drug interference and to interpret results in line with published algorithms. To detect drug levels a sandwich ELISA is used and to measure anti-drug antibodies a bridging ELISA is used. In both assays pre-diluted samples, calibrators and controls are added to separate wells and any drug/anti-drug antibody present binds to the pre-coat on the plate. After the first incubation unbound sample is washed away and a peroxidase-labelled secondary reagent is added to each well. This binds to the immobilised drug/anti-drug antibodies during the second incubation. Unbound peroxidase is washed off the plates and a chromogenic substrate is added. The intensity of the colour that develops is measured using a spectrophotometer. The signal obtained is proportional to the amount of drug/anti-drug antibodies in the patient sample. Details on the interpretation of results, limits of detection and assay measurement ranges are presented in table 2.

Table 2 Interpretation of results, limits of detection and assay ranges for the Promonitor assays

Name	Results interpretation	Limit of quantification	Assay range
Promonitor-ADL-1DV	Quantitative through generation of standard curve	2.9 ng/ml	0.4 to 12 µg/ml
Promonitor-IFX-1DV		1.7 ng/ml	0.2 to 14.4 µg/ml
Promonitor-ETN-1DV		3.1 ng/ml	0.175 to 10 µg/ml
Promonitor-GLM-1DV		0.036 µg/ml	0.36 to 6.4 µg/ml
Promonitor-ANTI-ADL-1DV	Quantitative through generation of standard curve	3.7 AU/ml	10 to 400 AU/ml
Promonitor-ANTI-IFX-1DV		2 AU/ml	5 to 288 AU/ml
Promonitor-ANTI-ETN-1DV		61 AU/ml	142 to 900 AU/ml
Promonitor-ANTI-GLM		56 AU/ml	28 to 1008 AU/ml

2.2.2 IDKmonitor ELISA kits

There are 10 CE marked IDKmonitor ELISA kits (Immundiagnostik) that are relevant to this assessment, which are distributed in the UK by BioHit Healthcare Ltd (table 3): 4 kits measure the levels of free TNF-α inhibitor; 4 kits measure the levels of free anti-drug antibodies; and 2 kits measure the levels of total anti-drug antibodies (free antibodies and antibodies bound to the drug). All tests are performed in a laboratory.

Table 3 IDKmonitor ELISA kits

Name (code)	Detects	Microplate pre-coat	Secondary reagent
IDKmonitor infliximab drug level ELISA (K9655)	Free[1] infliximab (Remicade, Remsima, Inflectra)	Monoclonal anti-infliximab antibody	Peroxidase labelled antibody
IDKmonitor adalimumab drug level ELISA (K9657)	Free[1] adalimumab	Monoclonal anti-adalimumab antibody	Peroxidase labelled antibody
IDKmonitor etanercept drug level ELISA (K9646)	Free[1] etanercept	Monoclonal anti-etanercept antibody	Peroxidase labelled antibody
IDKmonitor golimumab drug level ELISA (K9656)	Free[1] golimumab	Monoclonal anti-golimumab antibody	Peroxidase labelled antibody
IDKmonitor infliximab free ADA ELISA (K9650)	Free[1] anti-infliximab antibodies	Infliximab $F(ab)_2$ fragments	Peroxidase labelled infliximab
IDKmonitor adalimumab free ADA ELISA (K9652)	Free[1] anti-adalimumab antibodies	Adalimumab $F(ab)_2$ fragments	Peroxidase labelled adalimumab
IDKmonitor etanercept free ADA ELISA (K9653)	Free[1] anti-etanercept antibodies	Etanercept $F(ab)_2$ fragments	Peroxidase labelled etanercept
IDKmonitor golimumab free ADA ELISA (K9649)	Free[1] anti-golimumab antibodies	Golimumab $F(ab)_2$ fragments	Peroxidase labelled golimumab
IDKmonitor infliximab total ADA ELISA (K9654)	Total[2] anti-infliximab antibodies	Streptavidin	N/A
IDKmonitor adalimumab total ADA ELISA (K9651)	Total[2] anti-adalimumab antibodies	Streptavidin	N/A

[1] Free TNF-α inhibitor is drug that is unbound to antibody, and free anti-drug antibodies are those that are unbound to drug

[2] Total anti-drug antibodies include both unbound (free) antibodies and those bound to TNF-α inhibitor

The kits consist of strips of pre-coated microtitre plate (96 wells), reagents, buffers, standards (drug level ELISAs only) and controls. The ELISAs can be performed manually or run on an automated ELISA processor.

The ELISAs that measure free drug levels and the ELISAs that measure free anti-drug antibodies follow a standard ELISA procedure. Firstly the samples are added to the wells and any drug/anti-drug antibodies bind to the pre-coat

on the plate. After incubation, a wash step is carried out to remove unbound sample, and a peroxidase-labelled secondary reagent is added. This binds to the immobilised drug/anti-drug antibodies during the second incubation. Unbound peroxidase is washed off the plates and a chromogenic substrate is added. The intensity of the colour that develops is measured using a spectrophotometer. The signal obtained is proportional to the amount of drug/anti-drug antibodies in the patient sample.

The drug level ELISA takes approximately 3 hours to run, and the TNF-α inhibitor is measured quantitatively using a dose response curve. The free anti-drug antibody ELISA requires an overnight incubation (16-20 hours), and the anti-drug antibodies are measured semi-quantitatively using a cut-off control. Details on the interpretation of results, limits of detection and assay measurement ranges are presented in table 4.

The 2 total anti-drug antibody ELISA kits enable the measurement of bound and unbound anti-drug antibodies. The assay take approximately 3 hours to run. During sample preparation complexes between anti-drug antibodies and TNF-α inhibitor are dissociated using an acidic buffer. Biotinylated and peroxidase-labelled drug are added to the sample and form complexes with the anti-drug antibodies. The complexes bind via biotin to the streptavidin coated plate. Following a wash step a chromogenic substrate is added, the colour change reaction is stopped by the addition of an acid solution and the colour intensity is read by a spectrophotometer. Anti-drug antibodies are measured semi-quantitatively using a cut-off control.

Table 4 Interpretation of results, limits of detection and assay ranges for the IDKmonitor ELISAs

Name (code)	Results interpretation	Limit of blank	Assay range
IDKmonitor infliximab drug level (K9655)	Quantitative. Generation of standard curve and determination of drug level in µg/ml	2.0 ng/ml	0.4 to 45 µg/ml
IDKmonitor adalimumab drug level (K9657)		2.3 ng/ml	0.4 to 45 µg/ml
IDKmonitor etanercept drug level (K9646)		1.65 ng/ml	0.2 to 11.25 µg/ml
IDKmonitor golimumab drug level (K9656)		3.56 ng/ml	0.4 to 22.50 µg/ml
IDKmonitor infliximab free ADA ELISA (K9650)	Semi-quantitative. Evaluated by a cut-off control (10 AU/ml) to	5.787 AU/ml	10 AU/ml to 353.83 AU/ml
IDKmonitor adalimumab free ADA ELISA (K9652)		t.b.d	10 AU/ml to t.b.d.

IDKmonitor etanercept free ADA ELISA (K9653)	give a positive or negative result	4.549 AU/ml	10 AU/ml to 186.52 AU/ml
IDKmonitor golimumab free ADA ELISA (K9649)		2.56 AU/ml	10 AU/ml to 132.93 AU/ml
IDKmonitor infliximab total ADA ELISA (K9654)	Semi quantitative. Evaluated by a cut-off control (10 AU/ml) to give a positive or negative result	2.653 AU/ml	10 AU/ml to 497.00 AU/ml
IDKmonitor adalimumab total ADA ELISA (K9651)		2.765 AU/ml	10 AU/ml to 164.81 AU/ml
t.b.d, to be determined			

2.2.3 LISA-TRACKER

There are 10 CE marked LISA-TRACKER ELISA kits (Theradiag) that are relevant to this assessment (table 5). All tests are laboratory based assays. Five of these kits measure the levels of free anti-drug antibodies and 5 kits measure the levels of free TNF-α inhibitor. LISA-TRACKER Duo kits are also available that include assays to measure the levels of both free anti-drug antibodies and TNF-α inhibitor.

Table 5 LISA-TRACKER ELISA kits

Name (code)	Detects	Microplate pre-coat	Secondary reagent
LISA-TRACKER Adalimumab (LTA002)	Free[1] adalimumab	TNF-α	Biotinylated anti-human IgG antibody
LISA-TRACKER Certolizumab (LTC002)	Free[1] certolizumab	TNF-α	
LISA-TRACKER Etanercept (LTE002)	Free[1] etanercept	TNF-α	
LISA-TRACKER Infliximab (LTI002)	Free[1] infliximab (Remicade, Flixabi, Inflectra and Remsima)	TNF-α	
LISA-TRACKER Golimumab (LTG002)	Free[1] golimumab	TNF-α	
LISA-TRACKER anti-Adalimumab (LTA003)	Free[1] anti-adalimumab antibodies	Adalimumab	Biotinylated adalimumab
LISA-TRACKER anti-Certolizumab (LTC003)	Free[1] anti-certolizumab antibodies	Certolizumab	Biotinylated certolizumab
LISA-TRACKER anti-Infliximab (LTI003)	Free[1] anti-infliximab antibodies	Infliximab	Biotinylated infliximab
LISA-TRACKER anti-Etanercept (LTE003)	Free[1] anti-etanercept antibodies	Etanercept	Biotinylated etanercept

LISA-TRACKER anti-Golimumab (LTG003)	Free[1] anti-golimumab antibodies	Golimumab	Biotinylated golimumab
[1] Free TNF-α inhibitor is drug that is unbound to antibody, and free anti-drug antibodies are those that are unbound to drug			

The LISA-TRACKER ELISA kits consist of pre-coated strips of microtitre plate (96 wells), reagents, wash buffer, standards and controls. The assays can be run simultaneously or individually on any manual or automated standard ELISA based processor platform, and take approximately 3 hours to run.

In all assays, samples, standards and controls are added to the pre-coated microtitre plate. Any drug/anti-drug antibody present binds to the pre-coated wells during the first incubation step and any unbound substances are removed in a subsequent washing step. A biotinylated secondary reagent is then added which binds to the immobilised drug/anti-drug antibody. Any unbound reagent is removed by a second wash step before peroxidase labelled streptavidin is added to the plate. Streptavidin binds to the biotin-labelled complex and any unbound streptavidin is removed by a final wash step. Finally, a chromogenic substrate solution is added and colour develops in proportion to the amount of drug/anti-drug antibody present in the sample. The colour change reaction is stopped by the addition of an acid solution and the colour intensity is read by a spectrophotometer. TNF-α inhibitor and anti-drug antibodies are measured quantitatively using dose response curves. The limits of detection and assay ranges are presented in table 6.

Table 6 Interpretation of results, limits of detection and assay ranges for LISA-TRACKER assays

Name (code)	Results interpretation	Limit of detection	Assay range
LISA-TRACKER Adalimumab (LTA002)	Quantitative. Generation of standard curve and determination of drug level in µg/ml	0.3 µg/ml	0.3 to 16 µg/ml
LISA-TRACKER Infliximab (LTI002)		0.3 µg/ml	0.3 to 16 µg/ml
LISA-TRACKER Etanercept (LTE002)		0.2 µg/ml	0.2 to 5 µg/ml
LISA-TRACKER Certolizumab (LTC002)		3 µg/ml	3 to 84 µg/ml
LISA-TRACKER Golimumab (LTG002)		0.1 µg/ml	0.1 to 8 µg/ml
LISA-TRACKER anti-Adalimumab (LTA003)	Quantitative. Generation of standard curve and determination of anti-drug antibody level	10 ng/ml	10 to 160 ng/ml
LISA-TRACKER anti-Infliximab (LTI003)		10 ng/ml	10 to 200 ng/ml

LISA-TRACKER anti-Etanercept (LTE003)		10 ng/ml	10 to 100 ng/ml
LISA-TRACKER anti-Certolizumab (LTC003)		5 AU/ml	5 to 160 AU/ml
LISA-TRACKER anti-Golimumab (LTG003)		2.5 ng/ml	2.5 to 80 ng/ml

2.2.4 RIDASCREEN ELISA kits

There are 4 CE marked RIDASCREEN ELISAs (R-Biopharm) that are relevant to this assessment. All are laboratory based assays. Two of the kits measure levels of free TNF-α inhibitor and 2 kits measure the levels of free anti-drug antibodies (table 7). The RIDASCREEN ELISAs are commercialised versions of the KU Leuven in-house ELISAs, and are marketed as apDia ELISA kits in the Benelux area of Europe.

Table 7 RIDASCREEN ELISA kits

Name	Detects	Microplate pre-coat	Secondary reagent
RIDASCREEN ADM Monitoring	Free[1] adalimumab	TNF-α	Peroxidase conjugated monoclonal antibody
RIDASCREEN IFX Monitoring	Free[1] infliximab (Remicade, Remsima, Inflectra)	TNF-α	Peroxidase conjugated monoclonal antibody
RIDASCREEN Anti-ADM Antibodies	Free[1] antibodies to adalimumab	Adalimumab	(1) Biotin conjugated infliximab. (2) Peroxidase conjugated streptavidin
RIDASCREEN Anti-IFX Antibodies	Free[1] antibodies to infliximab	Infliximab	(1) Biotin conjugated infliximab. (2) Peroxidase conjugated streptavidin
[1] Free TNF-α inhibitor is drug that is unbound to antibody, and free anti-drug antibodies are those that are unbound to drug			

Each RIDASCREEN kit includes a 96-well microtitre plate, 12 single‑use 8-microwell strips, and all necessary reagents for up to 40 duplicate or 80 singlicate tests on serum or plasma samples. For every test, 16 microwells are used for calibration and quality control using 6 standards and 2 control samples in duplicate. Other reagents provided with the kit include diluent, conjugates, substrate, wash fluid and a stop reagent. The ELISAs can be performed manually or run on an automated ELISA processor, and take just under 2 hours to run.

In all assays, samples, standards and controls are added to the pre-coated microtitre plate. Any drug/anti-drug antibody present binds to the pre-coated wells during the first incubation step and any unbound substances are removed in a subsequent washing step. In the drug level assays the secondary reagent (peroxidase labelled monoclonal antibody) is then added which binds to the immobilised drug. In the anti-drug antibody assays the secondary reagent is biotin-conjugated TNF-α inhibitor, which binds to the drug-antibody complex immobilised in the wells. After removal of the unbound biotin reagent, peroxidase-conjugated streptavidin is added. Finally, in both assays, any unbound peroxidase reagent is removed by a wash step, and a chromogenic substrate solution is added. Colour develops in proportion to the amount of drug/anti-drug antibody present in the sample. The colour change reaction is stopped and the colour intensity is read by a spectrophotometer. TNF-α inhibitor and anti-drug antibodies are measured quantitatively using dose response curves. The limits of detection and assay ranges are presented in table 8.

Table 8 Interpretation of results, limits of detection and assay ranges for the RIDASCREEN ELISAs

Name (code)	Results interpretation	Limit of detection	Assay range
RIDASCREEN ADM Monitoring	Quantitative. Generation of standard curve and determination of drug level in µg/ml	1 ng/ml	0.5 to 48 µg/ml
RIDASCREEN IFX Monitoring		1 ng/ml	0.5 to 48 µg/ml
RIDASCREEN Anti-ADM Antibodies	Quantitative. Generation of standard curve and determination of anti-drug antibody level in ng/ml	0.06 ng/ml	5 to 1000 ng/ml
RIDASCREEN Anti-IFX Antibodies		0.06 ng/ml	5 to 1000 ng/ml

2.2.5 MabTrack ELISA kits and Sanquin Diagnostic Services

Sanquin is a laboratory in the Netherlands and it provides laboratory test services including testing for TNF-α inhibitors using ELISA based assays. It also provides CE marked ELISA kits for local laboratory testing for adalimumab and infliximab levels and their correlating anti-drug antibodies. The kits available to purchase are called MabTrack ELISA kits. There are 4 CE marked ELISA kits available that are relevant to the assessment: 2 for testing free drug levels and 2 for their correlating free anti-drug antibodies (table 9). In addition, a testing service using validated ELISAs is available for etanercept and its correlating anti-drug antibodies, golimumab drug levels and certolizumab drug levels. Testing is performed at the Sanquin laboratories in

the Netherlands. Radioimmunoassays that measure drug levels or anti-drug antibodies are outside of the scope of this assessment.

Table 9: MabTrack ELISA kits

Name (code)	Detects	Microplate pre-coat	Secondary reagent
MabTrack level adalimumab M2910	Free[1] adalimumab	TNF-α	Peroxidase-labeled monoclonal anti-adalimumab antibody
MabTrack level infliximab M2920	Free[1] infliximab (Remicade, Remsima, Inflectra)	TNF-α	Peroxidase-labeled monoclonal anti-infliximab antibody
MabTrack ADA adalimumab M2950	Free[1] antibodies to adalimumab	Adalimumab	Peroxidase-labelled adalimumab
MabTrack ADA infliximab M2960	Free[1] antibodies to infliximab	Infliximab	Peroxidase-labelled infliximab
[1] Free TNF-α inhibitor is drug that is unbound to antibody, and free anti-drug antibodies are those that are unbound to drug			

The MabTrack ELISA kits consist of pre-coated strips of microtitre plate (96 wells), reagents, wash buffer, standards or calibrators, controls and ELISA cover films. The assays take approximately 3 hours to run. Sandwich-type ELISA methodology is used for all drug level and anti-drug antibody assays. One kit can test 44 drug level samples in duplicate and 22 antidrug antibody samples in duplicate. The samples, controls and standards are added to separate wells and any drug/anti-drug antibody present binds to the pre-coat on the plate. After the first incubation unbound sample is washed away and a peroxidase-labelled secondary reagent is added to each well. This binds to the immobilised drug/anti-drug antibodies during the second incubation. Unbound peroxidase is washed off the plates and a chromogenic substrate is added. The intensity of the colour that develops is measured using a spectrophotometer. The signal obtained is proportional to the amount of drug/anti-drug antibodies in the patient sample.

The drug level ELISA results are measured quantitatively using dose response curves and the anti-drug antibody ELISA results are measured semi-quantitatively using 2 cut-off levels. Details on the interpretation of results, limits of detection and assay measurement ranges are presented in table 10.

Table 10: Interpretation of results, limits of detection and assay ranges for the MabTrack ELISAs

Name (code)	Results interpretation	Limit of quantification	Assay range
MabTrack level adalimumab M2910	Quantitative. Generation of standard curve and determination of drug level in µg/ml	0.06 µg/ml (1:200 dilution)	1–30 µg/ml
MabTrack level infliximab M2920		0.08 µg/ml (1:200 dilution)	0.22–39.7 µg/ml
MabTrack ADA adalimumab M2950	Semi-quantitative. Evaluated by 2 cut-off controls to give a strong positive, positive or negative result	Not given in instructions for use	
MabTrack ADA infliximab M2960		Not given in instructions for use	

3 Target conditions

3.1 Rheumatoid arthritis

Rheumatoid arthritis is an inflammatory autoimmune disease that typically affects the synovial tissue (tissue lining the inner surface of joints surrounded by synovial fluid) of the small joints of the hands and feet but can affect any joint. It causes swelling, stiffness, pain and progressive joint destruction, and can also cause more general symptoms, such as tiredness, fever and weight loss. The inflammation associated with rheumatoid arthritis can sometimes lead to complications affecting other areas of the body, including the heart, lungs and eyes.

Rheumatoid arthritis is usually a chronic relapsing condition which has a pattern of flare-ups followed by periods of lower disease activity; however, for some people, the disease is consistently progressive. Rheumatoid arthritis can have a severe impact on quality of life and it is estimated that approximately one-third of people stop work within 2 years of onset because of the disease.

Rheumatoid arthritis affects approximately 0.8% of the population, or approximately 580,000 people in England. Of these, approximately 15% have severe disease. It is about 2- to 4-times more prevalent in women than in men and can develop at any age, but the peak age of onset in the UK is about 40–60 years (National Rheumatoid Arthritis Society 2018). There is no cure for rheumatoid arthritis and treatment aims to improve quality of life and to prevent or reduce joint damage.

3.2 Diagnostic and care pathway

3.2.1 Pharmacological management

The main aim of pharmacological management in newly diagnosed and recent-onset rheumatoid arthritis is to suppress disease activity and induce disease remission, prevent loss of function, control joint damage, maintain pain control and enhance self-management.

In people with newly diagnosed active rheumatoid arthritis, the existing NICE guideline on rheumatoid arthritis in adults recommends a combination of conventional disease-modifying anti-rheumatic drugs (DMARDs, for example, methotrexate, leflunomide and sulfasalazine), plus short-term glucocorticoids as first-line treatment as soon as possible. When combination therapy is not appropriate (for example, comorbidities or pregnancy) DMARD monotherapy is recommended.

If the disease does not respond to intensive therapy with a combination of conventional DMARDs and disease is severe (DAS28 greater than 5.1), NICE technology appraisals guidance recommend: TNF-α inhibitors (adalimumab, etanercept, certolizumab pegol, infliximab, golimumab); IL6 inhibitors (tocilizumab, sarilumab); T-cell inhibitors (abatacept); JAK inhibitors (tofacitinib, baricitinib), and B-cell antibodies (rituximab; only after failure of a TNF inhibitor).

The NICE technology appraisal guidance on adalimumab, etanercept, infliximab, certolizumab pegol, golimumab, tocilizumab and abatacept for rheumatoid arthritis recommends to start treatment with the least expensive drug (taking into account administration costs, dose needed and product price per dose).

In established disease, management should address complications and associated comorbidity, and the impact of the condition on the patient's quality of life. NICE's guideline on rheumatoid arthritis in adults recommends short-term treatment with glucocorticoids for managing flares in people with recent-onset or established disease to rapidly decrease inflammation. The guideline also recommends analgesics (for example, paracetamol, codeine or compound analgesics) to people whose pain control is not adequate, and notes that oral non-steroidal anti-inflammatory drugs/COX-2 inhibitors for symptom control should be used at the lowest effective dose for the shortest possible period of time.

The NICE guideline on rheumatoid arthritis in adults makes the following recommendations on withdrawing conventional DMARDs:

- In people with recent-onset rheumatoid arthritis receiving combination DMARD therapy in whom sustained and satisfactory levels of disease control have been achieved, cautiously try to reduce drug doses to levels that still maintain disease control.
- In people with established rheumatoid arthritis whose disease is stable, cautiously reduce dosages of disease-modifying or biological drugs. Return promptly to disease-controlling dosages at the first sign of a flare.
- When introducing new drugs to improve disease control into the treatment regimen of a person with established rheumatoid arthritis, consider decreasing or stopping their pre-existing rheumatological drugs once the disease is controlled.
- In any person with established rheumatoid arthritis in whom disease-modifying or biological drug doses are being decreased or stopped, arrangements should be in place for prompt review.

The NICE technology appraisal guidance on adalimumab, etanercept, infliximab, certolizumab pegol, golimumab, tocilizumab and abatacept for rheumatoid arthritis recommends to continue treatment only if there is a moderate response measured using European League Against Rheumatism (EULAR) criteria at 6 months after starting therapy. It also states that after the initial response within 6 months, treatment should be withdrawn if a moderate EULAR response is not maintained.

3.2.2 Monitoring

The current NICE guideline for rheumatoid arthritis recommends measuring c-reactive protein (CRP) and key components of disease activity (using a composite score such as DAS28) to monitor the disease response to treatment in people with rheumatoid arthritis. The monitoring can inform decision-making about increasing treatment to control disease and cautiously decreasing treatment when disease is controlled. Monitoring is recommended at monthly intervals in the case of recent-onset rheumatoid arthritis (less than 2 years) until the disease is controlled to a level agreed with the patient. The guideline also recommends that people with rheumatoid arthritis are offered an annual review, and that if disease is established and satisfactorily controlled review appointments are offered at a frequency suitable to the person's needs.

3.2.3 Surgery

Complications of rheumatoid arthritis may lead to the need for surgery. The main expected benefits of surgery are, pain relief, improvement of joint function and prevention of deformity. The NICE guideline for rheumatoid arthritis recommends offering a specialist surgical opinion for people with rheumatoid arthritis if there is; persistent pain, worsening joint function, progressive deformity and persistent localised synovitis. Surgery is also recommended for complications such as imminent or actual tendon rupture, nerve compression or stress fracture. The guideline recommends that urgent medical and surgical management should be offered to people with suspected or proven septic arthritis.

3.2.4 Draft updated rheumatoid arthritis guideline

NICE's guideline on rheumatoid arthritis is currently being updated and is due to be published in July 2018. The draft recommendations on monitoring state that a review appointment should be considered 6 months after remission or low disease treatment target is reached to ensure that the target has been maintained. It also recommends that all adults with rheumatoid arthritis are offered an annual review to:

- assess disease activity and damage
- measure functional ability
- check for development of comorbidities
- assess the need for referral to surgery
- assess the effect the disease is having on a person's life.

In adults who have maintained the treatment target (remission or low disease activity) for at least 1 year without glucocorticoids, the draft guideline states that step-down strategies should be considered, in which drug doses are cautiously reduced or stopped. It notes that if the treatment target is no longer met following step-down, the previous DMARD regimen should be restarted promptly. It should be noted that these recommendation apply to the use of conventional DMARDs rather than biologic DMARDs. Clinical experts noted that it is not standard practice to reduce the dose of biologic DMARDs, but that it is increasingly starting to happen and may become more common in future.

3.2.5 Other guidelines

European League Against Rheumatism (EULAR) has a guideline on the management of rheumatoid arthritis with synthetic and biological DMARDs (2016) and the British Society for Rheumatology also has guidelines on the

management of rheumatoid arthritis. Many NHS Trust have local guidelines which provided additional detail to the NICE guidelines, for example, Greater Manchester high cost drugs pathway for rheumatoid arthritis.

3.3 Patient issues and preferences

It is anticipated that testing could be done as part of the routine monitoring that already takes place when patients with rheumatoid arthritis have DMARDs, and therefore it is not expected that there would be a substantial difference to patients in terms of the frequency of testing. Testing could be done in advance of a scheduled consultation so that results are available for the appointment, or alternatively, the consultant could phone the patient with the results following the appointment. However there is the possibility that there would be an increased need for hospital or primary care appointments for blood tests if serum trough levels are to be taken.

The tests could help to personalise treatment for patients with rheumatoid arthritis and provide more confidence in treatment decisions. For people whose disease is in remission, high serum trough levels may help to reassure them that a reduction in treatment dose could be beneficial. Patients may welcome a lower dose, as this could lead to a fewer side effects and reduced anxiety about the risk of getting an infection because of a weakened immune system. Tests could help identify if adherence to treatment is poor, which could lead to additional support for patients or a switch to an alternative treatment that is more preferable for patients.

4 Comparator

Current practice for monitoring response in rheumatoid arthritis includes assessing clinical features alongside a composite score such as the DAS28. The DAS28 is calculated using inputs for erythrocyte sedimentation rate, number of tender joints, number of swollen joints and a patient global health score, there is also an option to include CRP levels. The DAS28 provides a number between 0 and 10 which indicates how active the rheumatoid arthritis is. A score of greater than 5.1 is classed as high disease activity. Low disease activity is defined as a score of less than 3.2 and remission is a score of less than 2.6.

The EULAR response criteria may also be used. These criteria have been developed based on the DAS28 score and categorise disease as good response, moderate response and non-response. The EULAR response criteria take in to account the present DAS28 score and improvement in DAS28 score (table 12).

Table 11 EULAR response criteria

		DAS28 improvement		
		>1.2	**>0.6 and ≤1.2**	**≤0.6**
Present DAS28	**≤3.2**	Good response	Moderate response	No response
	>3.2 and ≤5.1	Moderate response	Moderate response	No response
	>5.1	Moderate response	No response	No response

5 Scope of the assessment

Table 12 Scope of the assessment

Decision question	What is the clinical and cost-effectiveness of tests for monitoring TNF-α inhibitor drug serum levels and anti-drug antibodies in people with rheumatoid arthritis?
Populations	People with rheumatoid arthritis who are being treated with a TNF-α inhibitor (adalimumab, etanercept, infliximab, certolizumab pegol, golimumab), and: - have achieved treatment target (remission or low disease activity) or, - experience a primary non-response or, - experience a secondary non-response
Interventions	Promonitor ELISA kits (Grifols - Progenika) • Promonitor-ADL-1DV • Promonitor-ANTI-ADL-1DV • Promonitor-ETN-1DV • Promonitor-ANTI-ETN-1DV • Promonitor-GLM-1DV • Promonitor-ANTI-GLM • Promonitor- IFX-1DV • Promonitor-ANTI-IFX-1DV IDKmonitor ELISA kits (Immundiagnostik/BioHit Healthcare): • IDKmonitor adalimumab drug level • IDKmonitor adalimumab free ADA • IDKmonitor adalimumab total ADA • IDKmonitor etanercept drug level • IDKmonitor etanercept free ADA • IDKmonitor golimumab • IDKmonitor golimumab free ADA • IDKmonitor infliximab drug level • IDKmonitor infliximab free ADA

	• IDKmonitor infliximab total ADA LISA-TRACKER ELISA kits (Theradiag): • LISA-TRACKER Adalimumab (LTA002) • LISA-TRACKER anti-Adalimumab (LTA003) • LISA-TRACKER Duo Adalimumab (LTA005) • LISA-TRACKER Certolizumab (LTC002) • LISA-TRACKER anti-Certolizumab (LTC003) • LISA-TRACKER Duo Certolizumab (LTC005) • LISA-TRACKER Etanercept (LTE002) • LISA-TRACKER anti-Etanercept (LTE003) • LISA-TRACKER Duo Etanercept (LTE005) • LISA-TRACKER Golimumab (LTG002) • LISA-TRACKER anti-Golimumab (LTG003) • LISA-TRACKER Duo Gloimumab (LTG005) • LISA-TRACKER Infliximab (LTI002) • LISA-TRACKER anti-Infliximab (LTI003) • LISA-TRACKER Duo Infliximab (LTI005) RIDASCREEN ELISA kits (r-biopharm) • RIDASCREEN ADM monitoring • RIDASCREEN anti-ADM antibodies • RIDASCREEN IFX monitoring • RIDASCREEN anti-IFX antibodies MabTrack ELISA kits (Sanquin) • MabTrack level adalimumab M2910 • MabTrack ADA adalimumab M2950 • MabTrack level infliximab M2920 • MabTrack ADA infliximab M2960 Sanquin Diagnostic Services (testing service using validated ELISAs) • Adalimumab drug levels • Certolizumab drug levels • Etanercept drug levels • Etanercept anti-drug antibodies • Golimumab drug lelves • Infliximab drug levels Evidence permitting, the use of both free and total anti-drug antibody assays will be assessed.

	The intervention tests will be used in addition to current clinical practice (clinical assessment and monitoring using a composite score such as DAS28).
Comparator	Treatment decisions made using clinical judgement and regular monitoring using a composite score such as DAS28
Healthcare setting	Secondary and tertiary care
Outcomes	Intermediate measures for consideration may include: • Time to result • Number of inconclusive results • Impact on clinical management decisions
	Clinical outcomes for consideration may include: • Measures of disease activity • Rates of response, relapse and remission • Duration of response, relapse and remission • Rates of hospitalisation • Rates of surgical intervention • Adverse effects of treatment, such as infections
	Patient-reported outcomes for consideration may include: • Health-related quality of life
	Costs will be considered from an NHS and Personal Social Services perspective. Costs for consideration may include: • Cost of the testing, including sample transport where relevant • Cost of staff and associated training • Medical costs arising from testing including ongoing care, outpatient appointments, surgery and treatment • Medical costs arising from adverse effects of treatment
	The cost-effectiveness of interventions should be expressed in terms of incremental cost per quality-adjusted life year.
Time horizon	The time horizon for estimating clinical and cost effectiveness should be sufficiently long to reflect any differences in costs or outcomes between the technologies being compared.

6 Other issues for consideration

6.1 Reflex and concurrent testing

Tests for the therapeutic monitoring of TNF-α inhibitors may be performed in one of 2 ways. Firstly, testing for TNF-α inhibitor drug levels and testing for antibodies to TNF-α inhibitor could be performed concurrently. Alternatively,

reflex testing could be implemented; testing for TNF-α inhibitor drug levels is performed first and the result from this initial test is used to guide follow-up testing by the laboratory without a further request from the treating clinician. If the drug trough level is undetectable, testing for antibodies to TNF-α inhibitor would be performed. If TNF-α inhibitor is present in the sample, testing for antibodies would not be performed. Advice sought from clinical experts during scoping suggested that reflex testing would be preferred to concurrent testing. The economic model may include scenarios for both methods of testing.

6.2 Free and total anti-drug antibody levels

Anti-drug antibody assays can measure either free anti-drug antibody levels (anti-drug antibodies unbound to drug, free in systemic circulation) or total antidrug antibody levels (includes anti-drug antibodies bound to drug and free in systemic circulation). It is antibodies that are unbound to the drug that have the potential to inhibit the mode of action of the treatment. However, measuring total antidrug antibody levels might be beneficial for detecting early changes over time in levels of antibody and potentially could provide an early indication of a secondary non-response to treatment. There is no consensus on which type of test should be used for monitoring anti-drug antibodies in rheumatoid arthritis. Advice from clinical experts during scoping suggested that measurement of free anti-drug antibodies would be preferred, because the clinical relevance of total anti-drug antibody levels is uncertain.

6.3 Timing of testing

Testing could be carried out as part of routine monitoring, which in clinical practice is reported to be every 3 months in people with recent-onset rheumatoid arthritis and every 6 to 12 months after that. The draft updated NICE guideline on rheumatoid arthritis recommends all adults have an annual review. Advice from clinical experts during scoping suggested that testing for people newly started on a TNF-α inhibitor should occur about 3 months after starting treatment. For people who are responding well to treatment testing could be carried out every 6 to 12 months, but may be more frequent following a reduction in TNF-α inhibitor dose, for example 3 and 6 months after each dose reduction. For secondary non-response testing should be done based on clinical indicators of a loss of efficacy, for example DAS28.

6.4 Algorithms for interpreting test results

There are existing algorithms for interpreting the results of the assays. NHS Greater Glasgow and Clyde have guidance published on their website on rheumatology biologic drug monitoring, it recommends testing to:

- guide drug dose or interval changes after starting therapy and where drug tapering or escalation is being considered
- help determine if primary or secondary failure is because of immunogenicity.

Greater Manchester medicines management group published guidance on their website on high cost drugs pathway for rheumatoid arthritis, it suggests measuring anti-drug antibodies and trough drug levels for people who initially responded to treatment and are experiencing a loss of response (secondary non-response). Further details of the algorithms can be seen in table 14.

If reflex testing is used, or for drugs that have low immunogenicity such as etancercept, drug levels alone may be used for making treatment decisions.

Table 13 Algorithm for interpreting results of drug level and anti-drug antibody tests for people with rheumatoid arthritis on treatment with biological DMARDs

Response	Drug levels	Free anti-drug antibody present?	Outcome
Good response – low disease activity / remission	Low	Yes	Consider early review of treatment [a]; consider stopping treatment [b]
		No	Continue monitoring; consider stopping treatment; check adherence to TNF-α inhibitor [b]
		Not measured[1]	Check adherence to TNF-α inhibitor; continue monitoring [b]
	High	Yes	Scenario unlikely to occur
		No	Consider tapering treatment by increasing dosing interval [a]
		Not measured	
Loss of response/non-response – disease activity moderate/high	Low	Yes	Consider switching to less immunogenic drug [a, c]
		No	Assess adherence to TNF-α inhibitor and consider whether the dose is weight adjusted [c]
		Not measured[1]	Assess adherence to TNF-α inhibitor and consider switching to a different TNF-α inhibitor [b]
	High / normal	Yes	Scenario unlikely to occur
		No	Switch to a treatment with a different mechanism of action [a, c]
		Not measured[1]	Switch to a treatment with a different mechanism of action [b]
[a] Source: NHS Glasgow and Clyde guidance on rheumatology biologic drug monitoring			

[b] Informed by advice from clinical expert sought during scoping

[c] Source: Greater Manchester medicines management group high cost drugs pathway for rheumatoid arthritis (advice on assays is for secondary non-response only)

[1] Etanercept only

7 Potential equality issues

NICE is committed to promoting equality of opportunity, eliminating unlawful discrimination and fostering good relations between people with particular protected characteristics and others.

Rheumatoid arthritis can have a substantial and long-term adverse effect on a person's ability to carry out normal day-to-day activities. Therefore, people with rheumatoid arthritis may be covered under the disability provision of the Equality Act (2010).

Therapeutic monitoring may also be helpful in people who have paused treatment of rheumatoid arthritis due to pregnancy or treatment for other co-morbidities, and subsequently have re-started treatment with TNF-α inhibitors.

8 Potential implementation issues

The key considerations for adoption highlighted through discussions with expert contributors are:

Clinical confidence

Clinical experts noted that more understanding and knowledge is required about:

- What the test results mean for clinical practice and the treatment plan - clinicians will need training and education.
- Whether reflex or concurrent testing is the most cost effective testing strategy, and if certain biological therapies only need anti-drug antibodies testing.
- How the total and free anti-drug antibodies levels impact on decisions to change the treatment plan.

Obtaining a sample

The group of patients in whom biological therapies for rheumatoid arthritis are used receive regular reviews and blood tests, therefore blood samples may be available for therapeutic drug monitoring tests without the need for additional appointments. However, in some cases, if a trough level (taken just before a

drug dose is due) is required, an additional hospital appointment may be needed to take the sample or it could be taken at a GP surgery. This would be an additional cost to the service.

Laboratory
Results vary with different tests, therefore the test kit that was used to do the test should be recorded alongside the result.

Equipment and staff training
Adopting the tests would require increased technician time and a senior laboratory scientist to review and sign off the results. However, they are reasonably easy tests to run and minimal training would be needed.

Frequency and centralisation
Centralised testing, either nationally or regionally, would mean that tests could be run frequently and at full capacity, giving timely availability of results and optimum use of resources.

Costs
Costs of testing could be a barrier to adoption, but savings on biological therapies could be made if drug doses are reduced without impacting on clinical outcomes, and by guiding the most suitable and effective treatment for each patient. However, the cost of testing and the savings on drugs are likely to come from different budgets.

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Appendix A Glossary of terms

Anti-drug antibodies
Antibodies produced by the body in an immune response against a therapeutic antigen, for example a monoclonal antibody, which may inactivate the drug and modify the pharmacokinetic characteristics of the drug

Immunosuppressants
A class of drugs used to supress of prevent an immune response

Pharmacokinetics
The process by which a drug is absorbed, distributed, metabolized, and eliminated by the body

Primary non-response
A lack of improvement of clinical signs and symptoms during induction therapy

Recent onset rheumatoid arthritis
Rheumatoid arthritis with a duration of up to 2 years. Within recent-onset RA, categories of suspected persistent synovitis or suspected rheumatoid arthritis refer to patients in whom a diagnosis is not yet clear, but in whom referral to specialist care or further investigation is required.

Secondary non-response
Loss of clinical response to therapy in patients whose disease had initially demonstrated clinical response

TNF-α
An inflammatory cytokine which helps to regulate the immune system, but when present in high concentrations it is responsible for the destructive inflammatory processes that occur in inflammatory bowel disease

TNF-α inhibitors
Biological therapies which target the TNF-α protein with the aim of modifying the inflammatory disease process

Trough levels
In a medicine administered periodically the trough level is the lowest level of drug reached in the body before the next dose is administered.

Appendix B Abbreviations

ADA	Anti-drug antibodies
AU	Absorbance units
CRP	c-reactive protein
DAS-28	Disease activity score 28
DMARDs	Disease-modifying antirheumatic drugs
ELISA	Enzyme linked immunosorbent assays
EULAR	European League Against Rheumatism
TNF-α	Tumour necrosis factor alpha