**Data Descriptor**

# Testing the Diagnostic Utility of Recombinant *Toxoplasma Gondii* Chimeric Antigens – Generated Datasets

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## Abstract

The datasets titled *Toxoplasma gondii* recombinant chimeric antigens – IgM and IgG ELISAs – mouse serum samples and *Toxoplasma gondii* recombinant chimeric antigens – IgG and IgM ELISAs – human serum samples contain absorbance measurements obtained during serological tests using mouse and human sera in enzyme-linked immunosorbent assay (ELISA) tests based on recombinant chimeric antigens. The datasets allows a comparison of absorbance values obtained for individual recombinant chimeric antigens in relation to the whole *Toxoplasma* lysate antigens (TLA) used in commercial tests.

**Keywords:** *Toxoplasma gondii*, recombinant chimeric antigen, diagnostic utility, toxoplasmosis

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### Specification table (data records)

| | |
|---|---|
| Subject area | Immunology; Parasitology; Diagnostics |
| More specific subject area | Serological diagnostics |
| Type of data | Text |
| How the data was acquired | The data was collected using a commercially available Multiscan FC (Thermo Scientific) microplate photometer |
| Data format | Mixed (raw and processed) |

| | |
|---|---|
| Experimental factors | The obtained measurement data were obtained for:<br>healthy animals and within a specified period of time after infestation with the parasite<br>patients in various stages of the disease and healthy people |
| Experimental features | The data was obtained directly in the laboratory and the averaged measurement values were used to perform the data analysis |
| Data source location | MOST Wiedzy Open Research Data Catalog, Gdańsk University of Technology, Gdańsk, Poland |
| Data accessibility | The dataset is accessible and is publicly and freely available for any research or educational purposes |

## Background

Toxoplasmosis is a zoonotic infection with the protozoan parasite *Toxoplasma gondii*; which can affect most warm-blooded animals from mammals to birds all over the world. Human infection of *T. gondii* is generally asymptomatic, however, diagnosis of *T. gondii* infection is of great medical importance, especially for pregnant women and immunocompromised patients (e.g. AIDS and cancer patients) (Tenter, Heckeroth and Weiss, 2000). Primary infection of a pregnant woman is often associated with foetal infection, which can lead to miscarriage, stillbirth or severe neonatal malformation (Dunn et al., 1999; Fatoohi et al., 2002).

Routine diagnosis of T. gondii infection in humans is based on the detection of specific anti-*T. gondii* antibodies in serum samples. Serological techniques are suitable for the analysis of a large number of samples and play a major role in the diagnosis of toxoplasmosis. Among the available serological tests, ELISA has been adapted for the detection of IgM, IgG and IgA class antibodies. The serological methods for detection of specific *T. gondii* antibodies have usually involved the preparation of TLA, the production of which is expensive and laborious, moreover, a quantity of the antigen mixture is difficult to standardise. In recent years, the utility of recombinant antigens has been demonstrated in the diagnosis of toxoplasmosis in humans (Holec-Gąsior, 2013). The use of recombinant antigens for the diagnosis of *T. gondii* infection has proven highly beneficial for improving the standardisation of the method since the antigen composition of the test is precisely known. Furthermore, the advantages of these proteins are that the production cost of antigens is reduced and more than one defined antigen can be used for the detection of specific antibodies. A relatively new approach is the construction of a new generation of recombinant products so-called chimeric antigens which can replace native antigens from a lysed whole parasite.

The datasets titled *Toxoplasma gondii* recombinant chimeric antigens – IgG and IgM ELISAs – mouse serum samples and *Toxoplasma gondii* recombinant chimeric antigens – IgM and IgG ELISAs – human serum samples have been used to assess the diagnostic usefulness of recombinant chimeric antigens composed of immunodominant fragments of parasite antigens such as apical membrane antigen 1 (AMA1), surface antigen 2 (SAG2),

dense granule antigen 1 (GRA1), and rhoptry antigen 1 (ROP1). The first dataset was used to determine the humoral immune response dynamics during murine experimental toxoplasmosis using tetravalent recombinant chimeric proteins. The second dataset was used to determine the diagnostic usefulness of recombinant chimeric antigens in indirect IgM and IgG ELISAs with human serum samples. These datasets were used to develop the figures and tables included in the publication under the title The first study on the usefulness of recombinant tetravalent chimeric proteins containing fragments of SAG2, GRA1, ROP1 and AMA1 antigens in the detection of specific anti-*Toxoplasma gondii* antibodies in mouse and human sera (Ferra et al., 2019).

## Methods

### Ethical statements

The data contained in the datasets required obtaining appropriate approvals from ethical committees. Mouse serum samples were obtained according to experimental procedure guidelines provided by the Polish Local Ethics Commission for Experiments on Animals No. 9 in Lodz (Agreement 75/ŁB639/2012). The human serum samples were collected as part of the project entitled "Toxoplasmosis—facts and myths. Educational initiative raising social awareness about the infection with protozoan *Toxoplasma gondii*", the "Our Children" foundation, funded from the Civil Initiatives Fund of the Ministry of Labour and Social Policy (FIO 2008, contract No. 813). To ensure the anonymity of the subjects, only the date of serum collection and the individual's immune status in regard to anti-T. gondii specific antibodies and IgG avidity were disclosed.

ELISAs tests with mouse and human serum samples

To assess the reactivity of anti-T. gondii IgM and IgG antibodies from mouse sera with the native (TLA) and recombinant antigens, an immunoenzymatic assay was used, as described previously (Ferra et al., 2019). The diagnostic utility of recombinant antigens in detecting T. gondii infection in human sera was determined according to the IgM and IgG ELISA procedures described previously (Ferra et al., 2019).

### Data records

All of the obtained data contained in the datasets are the values of the measured absorbance. The data was collected using a commercially available Multiscan FC (ThermoFischer Scientific) microplate photometer. Data obtained from the microplate reader were saved to external memory in the form of text files which were then imported into Microsoft Excel spreadsheets. The obtained measurement values for individual serum samples were then segregated into appropriate groups depending on the phase of the disease, as well as depending on the antibody titre. Finally, the obtained datasets, containing measurement results for large pools of serum samples, ranged in size from several dozen to several hundred kB. The datasets were subjected to statistical analysis using the SigmaPlot 14.0 software (Systat Software), as described previously (Ferra et al., 2019). The

results of the performed statistical analyses will be added to the datasets soon. A sample graph (Fig. 9.1) and a table (Tab. 9.1) of the performed statistical analysis are presented below.

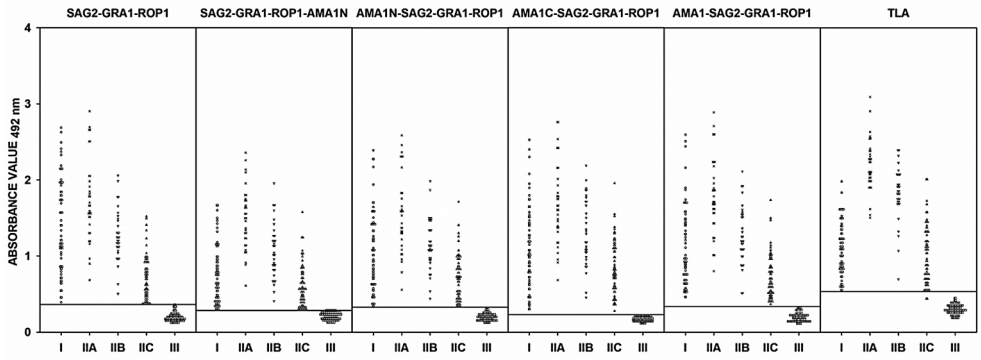


**Fig. 9.1.** Comparison of immunoreactivity, in IgG ELISA using SAG2-GRA1-ROP1, SAG2-GRA1-ROP1-AMA1N, AMA1N-SAG2-GRA1-ROP1, AMA1C-SAG2-GRA1-ROP1, and AMA1-SAG2-GRA1-ROP1 chimeric proteins, as well as TLA, with sera from patients with suspected acute (I) and chronic T. gondii infection (IIA–IgG titre>300 IU/ml, IIB–IgG titre between 101–300 IU/ml, and IIC IgG titre≤100 IU/ml), and from seronegative individuals (III). The horizontal lines represent the cut-off values (Ferra et al., 2019)

**Tab. 9.1**

IgG ELISA tests of recombinant antigens and TLA performances to discriminate among samples from suspected acute (I) and chronic (II) phase of T. gondii infection patient groups vs. control (III) group (Ferra et al., 2019)

| ANTIGEN | calculated cut-off | ROC cut-off | AUC | Sensitivity, % | Specificity, % |
|---|---|---|---|---|---|
| SAG2-GRA1-ROP1 | 0.3641 | 0.3560 | 0.9980 | 100 | 97.8 |
| SAG2-GRA1-ROP1-AMA1N | 0.2882 | 0.2875 | 1.0000 | 100 | 100 |
| AMA1N-SAG2-GRA1-ROP1 | 0.3286 | 0.3240 | 1.0000 | 100 | 100 |
| AMA1C-SAG2-GRA1-ROP1 | 0.2294 | 0.2508 | 1.0000 | 100 | 100 |
| AMA1-SAG2-GRA1-ROP1 | 0.3389 | 0.3478 | 1.0000 | 100 | 100 |
| TLA | 0.5344 | 0.4523 | 0.9999 | 99.5 | 100 |

Fig. 9.1 shows the plots obtained for each of the recombinant chimeric antigens. For comparative purposes, the previously tested trivalent recombinant antigen SAG2-GRA1-

ROP1 and the commercially used TLA antigen preparation were used (Ferra, Holec-Gąsior and Kur, 2015). The figure shows the values of the measured absorbance for each of the tested serum groups. Each single dot corresponds to the mean absorbance value obtained for a single serum sample. The size of a single plot obtained for one antigen preparation is several MB.

**Serum groups:**

I – suspected acute T. gondii infection (IgM +; IgG +; low avidity), n=64
II – chronic T. gondii infection (IgM –; IgG +; high avidity), n=128
III – control group (IgM –; IgG –), n=92

Sensitivity and specificity were determined using the cut-off value obtained by ROC analysis for the best discrimination.

Tab. 9.1 contains one of the most important statistical parameters calculated for antigen preparations tested in diagnostic tests. The table compares the cut-off values calculated by the standard method (based on a separate pool of seronegative serum samples) with the cut-off value obtained using the Receiver Operating Curve (ROC) analysis, which is based on all sera in relation to the results of the commercial test. The ROC analysis was performed to obtain the Area Under the Curve (AUC), the sensitivity, and the specificity percentages for the different groups of the compared sera. ROC analysis is currently one of the best statistical tools for determining and comparing the diagnostic utility of antigen preparations.

## Data quality and availability

All serum samples were previously stored at –20°C. The serum samples were thawed immediately before testing, thoroughly mixed, and then their appropriate dilution was prepared. All of the tested serum samples were clear and did not contain any sediments. Quality-certified microplates (Nunc) with an appropriate sorption surface were used for the tests. The tests were performed on the same antigen preparations, different fractions of purified proteins were not used. All buffers used at various stages of the tests were prepared on an ongoing basis from certified reagents and then autoclaved. The components were weighed on an analytical balance, the liquid volumes were measured using serological pipettes or automatic pipettes with the appropriate volume range. All tests were performed with the use of multichannel pipettes (Eppendorf) and a Wellwash microplate washer (ThermoFischer Scientific). Test microplates were incubated at each stage at a constant temperature of 37°C in a StabiliTherm incubator (ThermoFischer Scientific). In order for the data to be of adequate quality and reliable, each sample of sera was tested twice. If the measured absorbance values were close to each other, the measurement was considered correct. The mean was calculated for the obtained duplicates. Moreover, for the test pool of serum samples used on a given day, tests were performed for all antigen preparations on the same day. In addition, an internal negative and positive control was used for the same serum samples on each test plate.

**Datasets DOI**
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