Dipartimento Area Medica
SC Medicina – Emostasi e Trombosi - Direttore: Prof.ssa Flora Peyvandi
Tel. 02 5503.5422
E-mail: emostasietrombosi@policlinico.mi.it

"Daratumumab in immune-mediated thrombotic thrombocytopenic purpura (iTTP)"

Acronym: DarTTP

Promoter: Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico,
Via Sforza 28, 20122 Milano, Italia

Coordinating Center: SC Medicina - Emostasi e Trombosi – Centro Emofilia e Trombosi "Angelo Bianchi Bonomi"
Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico,
Via Pace 9, 20122 Milano, Italia

Principal Investigator: Juri Alessandro Giannotta

Type of document: non-interventional study

Version: v.1.0

Date: 16/07/2025

TABLE OF CONTENTS

2. Abbreviations

iTTP: immune-mediated thrombotic thrombocytopenic purpura

ADAMTS13: a disintegrin and metalloproteinase with a thrombospondin type 1 motif member 13

CTCAE: Common Terminology Criteria for Adverse Events

REDCap: Research Electronic Data Capture

IRCCS: Istituto di Ricerca e Cura a Carattere Scientifico (Scientific Institute for Research, Hospitalization and Health Care)

3. Study responsibilities and collaborations

The study promoter is the Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico of Milan. The principal investigator (PI) is Dr. Juri Alessandro Giannotta of Medicina – Emostasi e Trombosi Complex Structure of Fondazione. Dr. Giannotta will be responsible for the dissemination of the results of the study. He will coordinate the data analysis of patients recruited at other Italian and foreign collaborating centers.

The local working group is composed by:

-Andrea Artoni (patients' enrollment),

-Addolorata Truma (patients' enrollment),

-Syna Miri (creation of the electronic Case Report Form),

-Ilaria Mancini (data analysis).

4. Abstract

Title: "DarTTP: an observational, international, multicentric study on daratumumab in immune-mediated thrombotic thrombocytopenic purpura (iTTP)" – v.1.0, 16-JUL-2025 – Juri Alessandro Giannotta (PI) - Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Angelo Bianchi Bonomi Hemophilia and Thrombosis Center, Milan, Italy

Backgroud and rationale: iTTP in an autoimmune disease caused by autoantibodies directed against the metalloproteinase ADAMTS13. Rituximab is the standard immune suppressive treatment suggested from international guidelines. However, 10-15% of patients do not achieve a

sustained ADAMTS13 remission with rituximab, and a significant portion of responders eventually need re-treatment after 12 months or less. Other therapeutic options are scarce and based on old immunosuppressive agents or splenectomy, all burdened by relevant toxicity and lack of solid efficacy data. Recently, targeting CD20-negative long-lived plasma cells appears to be a promising strategy in refractory iTTP. Bortezomib allows an ADAMTS13 response in 60-70% of rituximab-refractory iTTP patients, although adverse events (particularly, neurotoxicity) are reported in 15-50% of cases. In the last two years, the less toxic anti-CD38 monoclonal antibody daratumumab has been employed in selected iTTP patients with good results. However, evidence stems only from isolated case reports and a French nation-wide study including 9 patients.

Aim of the study: to collect evidence on a larger number of patients about the efficacy and safety of daratumumab in iTTP subjects who are refractory or intolerant to previous immunosuppressive treatments. The primary endpoint is the proportion of patients with ADAMTS13 activity levels above 20% of normal at 6 months from the first daratumumab administration.

Study design: international, multicentric, non-interventional, retrospective observational study.
Study population: adult iTTP patients treated with daratumumab in the acute or remission phase of disease.
Variables: patient's demographics, date of iTTP diagnosis, immunosuppressive treatments before daratumumab, comorbidities, ADAMTS13 activity and anti-ADAMTS13 antibody titer at daratumumab treatment and during follow-up, immunoglobulin A, G and M levels before daratumumab treatment and 3-6 months after, date of daratumumab infusions, date of clinical and ADAMTS13 remission, daratumumab-related adverse events (specifying grade), dates of clinical and/or ADAMTS13 relapses after daratumumab treatment.
Source document: medical records.
Sample size: 40 patients (minimum 16 for the calculation of the sample size).
Data analysis: Data were expressed as counts and percentages, indicating medians and interquartile range (IQR). The duration of ADAMTS13 remission after daratumumab treatment was expressed as ADAMTS13 relapse-free survival, estimated by the Kaplan–Meier analysis.

Timelines:

Start of data collection: OCT-2025

End of data collection: JAN-2026

Final report: APR-2026

5. Protocol amendments

Not applicable.

6. Timelines

Step of the study	***Planned date***
Start of the study and data collection	OCT-2025
End of the study and data collection	JAN-2026
Final report of the study	APR-2026

7. Background

Immune-mediated TTP (iTTP) in an autoimmune disease caused by autoantibodies directed against ADAMTS13. The current standard of treatment, including plasma exchanges, steroids +/- rituximab, and caplacizumab, warrants a significant reduction of mortality to less than 5% [1-3]. Moreover, pre-emptive treatment with rituximab during clinical remission in case of ADAMTS13 relapse (i.e., a decline in ADAMTS13 activity levels below 20% [4]) is recommended by the International TTP guidelines to prevent clinical iTTP relapse [3]. However, 10-15% of patients do not achieve a sustained ADAMTS13 remission with rituximab after the acute phase or when rituximab is used pre-emptively [5, 6]. Additionally, about half of rituximab-responsive patients eventually need re-treatment after 12 months or less, and long-term safety of frequent rituximab cycles are still unknown in iTTP [6, 7]. For this selected category of patients, therapeutic options are scarce, and rely on old immunosuppressive agents (e.g., cyclosporine, azathioprine, mycophenolate mofetil, cyclophosphamide, vincristine) or splenectomy, all burdened by relevant toxicity and lack of solid efficacy data [8, 9].

Recently, targeting CD20-negative long-lived plasma cells appears to be a promising strategy in

refractory iTTP. Anti-proteasome bortezomib allows an ADAMTS13 response in 60-70% of rituximab- and often multi-refractory iTTP patients [10, 11], although adverse events (particularly, neurotoxicity) are reported in 15-50% of cases. Considering the generally young age of iTTP patients, the toxicity mitigation along with the achievement of ADAMTS13 remission represents a clinically significant composite outcome. In the last two years, the less toxic monoclonal antibody daratumumab, targeting CD38-positive plasma cells, has been employed in selected iTTP patients with good results. However, evidence stems only from isolated case reports [12-15] and a French nation-wide study including 9 patients [16].

8. Aim of the study

This study aims to collect retrospective evidence on a larger number of iTTP patients treated with daratumumab because of refractoriness or intolerance to other immunosuppressive drugs. In detail, the primary objective of the study is to evaluate the efficacy of daratumumab in iTTP patients.

The secondary objectives are:

- efficacy of daratumumab in patients treated for refractory iTTP;
- efficacy of daratumumab in iTTP patients who were responding to other immunosuppressive therapies but experienced adverse reactions, leading to a change in therapeutic strategy;
- safety of daratumumab;
- time to response to daratumumab;
- duration of response to daratumumab;
- frequency of clinical relapses following daratumumab treatment.

9. Methods

9.1 Study design

No profit, multicentric, pharmacological, observational, retrospective, cohort study.

9.1.1 Primary endpoint

- Proportion of responders to daratumumab (i.e., number of patients responding to daratumumab / total number of patients treated with daratumumab); response is defined as the achievement of

ADAMTS13 activity levels above 20% of normal at 6 months from the first daratumumab administration, without new additional immunosuppressants.

9.1.2. Secondary endpoints

- Proportion of patients achieving ADAMTS13 complete (> lower limit of the normal, LLN, range of ADAMTS13 activity test used) or partial (>20% of normal) remission, if daratumumab used in refractory patients (i.e., while ADAMTS13 activity is <20% of normal);
- proportion of patients maintaining ADAMTS13 complete or partial remission, if daratumumab used in patients responding to the ongoing immune suppressive treatment (i.e., ADAMTS13 activity >20%) but experiencing adverse reactions (including unsatisfactory duration of response to rituximab) leading the clinician to change the therapeutic strategy;
- number of adverse events related to daratumumab, using CTCAE v5.0;
- median time to ADAMTS13 partial and complete remission after daratumumab treatment;
- median duration of ADAMTS13 remission after daratumumab treatment;
- proportion of patients experiencing clinical relapses after daratumumab treatment.

9.2 Setting

The study includes both inpatients and outpatients treated with daratumumab for iTTP from January 2010 to 6 months before the time of data collection. The medical records will be used as source documents. The estimated time for enrollment is four months.

9.2.1 Study population

The study includes adult iTTP patients who were treated with daratumumab in the acute phase of disease (i.e., with plasma exchanges and/or caplacizumab ongoing) or during remission (i.e., due to ADAMTS13 relapse or ADAMTS13 activity plasma levels persistently <20%) from January 2010 to 6 months before the time of data collection. It includes iTTP patients receiving daratumumab either for refractory disease or in ADAMTS13 remission but intolerant to other immune suppressive agents.

9.2.2 Inclusion criteria

- patients with a confirmed diagnosis of iTTP (i.e., ADAMTS13 activity <10% with anti-ADAMTS13 antibodies detected);
- aged ≥ 18 years;
- male and female patients;
- treated with daratumumab for iTTP.

9.2.3 Exclusion criteria

- patients unwilling or unable to provide their informed consent;
- follow-up < 6 months after daratumumab administration.

9.2.4 Study drug administration and schedule

Daratumumab is a human anti-CD38 monoclonal antibody of the IgG1κ isotype, produced in a mammalian cell line (Chinese Hamster Ovary [CHO]) using recombinant DNA technology. The recommended dose of daratumumab is 16 mg/kg of body weight administered as an intravenous infusion or 1800 mg administered as a subcutaneous infusion. Daratumumab is not licensed for the treatment of iTTP, therefore clinicians used it at variable schedules and doses as per physician's choice and based on published available evidence in iTTP [11-15].

9.2.5 Planned visits and evaluations

No additional visits/evaluations are requested than those planned as per current clinical practice.

9.3 Variables

a) iTTP history before daratumumab administration

- patient's date of birth
- date of iTTP diagnosis (defined as non-immune microangiopathic anemia and thrombocytopenia with severe ADAMTS13 activity deficiency in presence of anti-ADAMTS13 autoantibodies)
- ADAMTS13 activity and anti-ADAMTS13 antibody titer at diagnosis
- immunosuppressive treatments used before daratumumab

- dates of clinical and/or ADAMTS13 relapses

b) Variables at daratumumab treatment
- patient's weight and height
- tobacco use
- comorbidities, with date of diagnosis
- setting of daratumumab treatment (acute phase of disease or remission phase); if acute phase, date of TTP event and treatments (start and stop date of plasma exchanges and/or caplacizumab)
- ADAMTS13 activity and anti-ADAMTS13 antibody titer at daratumumab treatment and during follow-up
- immunoglobulin A, G and M levels before daratumumab treatment and 3-6 months after
- immunosuppressive treatments ongoing at daratumumab treatment
- date of daratumumab infusions
- ancillary drugs used as premedication and/or anti-infective prophylaxis
- date of clinical and ADAMTS13 remission
- daratumumab-related adverse events, specifying grade (according to CTCAE v.5.0 criteria)
- dates of clinical and/or ADAMTS13 relapses after daratumumab treatment, with respective treatments employed

9.4 Source document

All data will be obtained from clinical records collected at the enrolling centers.

9.5 Sample size

This is a descriptive, single-arm study designed to explore the efficacy of daratumumab in a subpopulation of patients with iTTP, a rare and life-threatening autoimmune condition. Due to the rarity of the disease, all patients who meet the inclusion criteria and are followed up at the participating centers during the study period will be enrolled, with no formal upper limit to recruitment.

Given this context, the aim of the study is not only exploratory but also intended to consolidate and

support the existing evidence in the literature, which suggests that daratumumab is effective in approximately 89% of treated patients in achieving and maintaining a clinically relevant increase in ADAMTS13 activity [16].

Assuming an actual treatment efficacy of 89%, and comparing it to the historical reference value of 48% observed with conventional immunosuppressive agents such as azathioprine [9], a formal power analysis indicated that a minimum of 16 patients would be required to statistically confirm the treatment effect. This target is considered feasible within the framework of this multicenter collaboration on a rare disease.

The sample size was calculated using a one-sided exact binomial test, performed in G*Power version 3.1.9.7, with the following settings:

- Test family: Exact
- Statistical test: Proportions: Inequality, one sample, binomial
- Tail(s): One (right-tailed)
- Null proportion: 0.48 (assumed as the minimum clinically relevant response threshold, based on our previous data in iTTP patients treated with azathioprine)
- Proportion under the alternative hypothesis: 0.89 (treatment efficacy of daratumumab according to existing literature)
- Alpha error probability: 0.05 (one-sided)
- Power (1 – β): 0.80

9.6 Data management

At enrollment, each participant will be assigned a unique code. Data de-identification will be performed in such a way that individuals accessing the database will not be able to trace the identity of the subjects in any manner. Only local investigators will have access to the identities of the enrolled subjects. The data necessary for the study will be recorded in a dedicated electronic Case Report Form (eCRF) within a Data Management System validated according to national regulations, provided by the Scientific Directorate of the Foundation. The platform used will be REDCap (Research Electronic Data Capture). The REDCap Consortium consists of over 1,000 institutional partners worldwide (research institutions, universities, ministries, etc.). The consortium

supports a secure web application (REDCap) designed exclusively to facilitate data acquisition for research studies. The REDCap application allows users to create and manage online databases quickly and securely, and is currently in use for more than 110,000 projects with approximately 150,000 users covering numerous research interest areas across the consortium. Through REDCap, the following measures will be implemented for this study: a) user-level identification with specific restrictions based on role in the study, b) real-time data validation and integrity checks, c) patient de-identification prior to data export, d) centralized data storage with daily backups on a secure server within the Foundation's IT infrastructure.

9.7 Data analysis

Data were expressed as counts and percentages, indicating medians and interquartile range (IQR). The duration of ADAMTS13 remission after daratumumab treatment was expressed as ADAMTS13 relapse-free survival, estimated by the Kaplan–Meier analysis.

9.8 Quality control

To ensure adequate quality control of the study, the investigator will allow, upon request, direct access to all relevant documentation and will dedicate part of their time to discuss the study results. Additionally, Regulatory Authorities may conduct inspections. In this case, the investigator must grant the inspector direct access to all pertinent documentation and allocate part of their time and staff to the inspector to discuss the monitoring results and any other aspects of the study. The presence of two operators is required for all manual data entries to prevent potential errors.

9.9 Study limitations

There are some limitations for this study:

- its retrospective nature, with an enrollment period spanning presumably >3 years and including different treatment schedules of daratumumab and variable ADAMTS13 testing timepoints after daratumumab administration in different enrolling Centers;
- the small sample size, limited by the rarity of iTTP;

- a possible underestimation of the prevalence of adverse events, since they were captured retrospectively neither in a standardized way nor at standard timepoints.

10. Protection of patients included in the study

The study will be conducted in accordance with Good Clinical Practice guidelines, the ethical principles derived from the Declaration of Helsinki, and the current regulations regarding observational studies. The observational study and its related documentation will be submitted to the appropriate Ethics Committee. The study will commence only after receiving the necessary approvals according to the internal procedures of the institution. The Ethics Committee must also approve any modifications to the protocol and the advertising used to recruit subjects for the study, in accordance with local regulations.

10.1 Subject information note and consent form for the processing of personal data

It is the responsibility of the physicians to obtain consent to the information note through the privacy consent form with date and signature from each patient before the start of data collection. The signature attests to the understanding of the consents and the information contained therein. In addition, the investigator must sign and date the informed consent form. The signed documents must be archived by the investigator, while a copy must be given to the patient.

Given the nature of Scientific Institute for Research, Hospitalization and Health Care (IRCCS) of the Policlinico, promoter of the study, in compliance with the current legislation on the protection of personal data (EU Reg. 679/2016) and Legislative Decree 196/2003 (Privacy Code) as amended by Legislative Decree 101/2018, for the data collected and processed by the Promoter and other participating centers having the nature of IRCCS, it will not be mandatory to ask the participants in the study for consent to the use of their personal data for the conduct of the study itself. In compliance with the provisions of art. 110 bis paragraph IV of the amended Legislative Decree 196/2003, due to the instrumental nature that the healthcare activity assumes with respect to research at IRCCS, the processing for research purposes of personal data already collected for

clinical activity, including the anonymization of the same, does not in fact constitute further processing.
As stipulated by the Data Protection Authority, the aforementioned provision applies to all types of medical research (both prospective and retrospective) promoted by IRCCS, including multicentric studies involving entities without the same institutional status.

Therefore, the abovementioned legal basis also extends to all other participating centers in the study that do not hold the status of IRCCS.
The IRCCS, as the Study Sponsor, will be responsible for drafting a Data Protection Impact Assessment to be published on its official website and for notifying the Data Protection Authority accordingly.

10.2 Insurance coverage

Given the observational nature of the present study, no additional insurance policies are necessary other than those already provided for normal clinical practice.

11. Management and notification of adverse events/reactions

All adverse events occurring during the treatment and throughout the study observation period have been reported in accordance with the applicable post-marketing pharmacovigilance regulations. Any adverse events that have occurred will also be documented in the study Case Report Form (CRF).

11.1 Definitions of adverse event and adverse reaction

Adverse event: any unwanted medical event that occurs in a patient included in a clinical trial to whom a medicinal product is administered and that does not necessarily have a causal relationship with the treatment.
Adverse reaction: harmful and unwanted effect resulting from the use of a medicinal product, including effects resulting from therapeutic error, abuse, misuse, off-label use, overdose and

professional exposure. The reaction, unlike the event, presupposes at least a reasonable possibility of correlation between the effect and the administered drug.

A serious adverse reaction is defined as one that:

- endangers the patient's life
- requires hospitalization of the patient or prolongs a hospitalization that has already occurred
- causes persistent or significant disability or incapacity
- causes death
- involves a congenital anomaly or birth defect.

12. Study results dissemination

The principal investigator will write a final report for the dissemination of the results at the end of the study. Data will be made public anonymously and presented as requested in aggregate form.

13. Publication and intellectual property rights of the study results

Publications

The Foundation, as Promoter, will guarantee the dissemination and publication of the study results, even in the event of negative results, without any constraints and guaranteeing the collaborating Centers visibility proportional to the actual participation. Each scientific journal or publication containing the results and data of the study must indicate the role and participation of the Centers and the Foundation, in a manner proportional to the actual contribution made to the study and the role covered by each party. The data will be published in aggregate form or in any case anonymized, so as not to allow in any way the identification of the interested party to whom the data refers**.**

Intellectual property rights

The Parties acknowledge that for the conduct of the collaboration within the scope of the study, data, information, know-how, inventions (patentable or not) owned by each party may be used and shared, which remains the exclusive owner even if it grants the other a right of access and use, non-exclusive and free of charge, for the sole purpose of carrying out the activities covered by the study

and limited to the duration of the study. It is understood that this right of use does not include the right to sublicense to third parties. In accordance with current legislation, the data and results generated within the scope of the Study will be owned by the Promoter, except for specific agreements between the Promoter and the Centers.

14. Funding

None.

15. References

1. Scully M, McDonald V, Cavenagh J, Hunt BJ, Longair I, Cohen H, et al. A phase 2 study of the safety and efficacy of rituximab with plasma exchange in acute acquired thrombotic thrombocytopenic purpura. Blood. 2011;118(7):1746-53.

2. Scully M, Cataland SR, Peyvandi F, Coppo P, Knöbl P, Kremer Hovinga JA, et al. Caplacizumab Treatment for Acquired Thrombotic Thrombocytopenic Purpura. N Engl J Med. 2019; 380(4):335-46.

3. Zheng XL, Vesely SK, Cataland SR, Coppo P, Geldziler B, Iorio A, et al. ISTH guidelines for treatment of thrombotic thrombocytopenic purpura. J Thromb Haemost. 2020; 18: 2496-502.

4. Cuker A, Cataland SR, Coppo P, de la Rubia J, Friedman KD, George JN, et al. Redefining outcomes in immune TTP: an international working group consensus report. Blood. 2021; 137(14):1855-61.

5. Hie M, Gay J, Galicier L, et al. Preemptive rituximab infusions after remission efficiently prevent relapses in acquired thrombotic thrombocytopenic purpura: experience of the French Thrombotic Microangiopathies Reference Center. Blood; 2014; 124(2):204–10.

6. Coppo P, Cuker A, George JN. Thrombotic thrombocytopenic purpura: Toward targeted therapy and precision medicine. Res Pract Thromb Haemost. 2018; 3(1):26-37.

7. Tieu J, Smith RM, Gopaluni S, Kumararatne DS, McClure M, Manson A, et al. Rituximab Associated Hypogammaglobulinemia in Autoimmune Disease. Front Immunol. 2021; 12: 671503.

8. Westwood JP, Scully M. Management of acquired, immune thrombocytopenic purpura (iTTP): beyond the acute phase. Ther Adv Hematol. 2022; 13:20406207221112217.

9. Bichard C, Mancini I, Agosti P, Capecchi M, De Leo P, Arcudi S, et al. Efficacy and safety of azathioprine during remission of immune-mediated thrombotic thrombocytopenic purpura. Blood Adv. 2022; 6:5463-5466.

10. Lee NCJ, Yates S, Rambally S, Sarode R, Ibrahim IF, Shen YM, et al. Bortezomib in relapsed/refractory immune thrombotic thrombocytopenic purpura: A single-centre retrospective cohort and systematic literature review. Br J Haematol. 2024; 204: 638-43.

11. Giannotta JA, Artoni A, Mancini I, Agosti P, Carpenedo M, Truma A, Miri S, Ferrari B, De Leo P, Salutari P, Mancini G, Molteni A, Rinaldi E, Bocchia M, Napolitano M, Prezioso L, Cuccaro A, Scarpa E, Condorelli A, Grimaldi D, Massaia M, Peyvandi F. Bortezomib for rituximab-refractory immune-mediated thrombotic thrombocytopenic purpura in the caplacizumab era: an Italian multicenter study. J Thromb Haemost. 2024; S1538-7836(24)00686-X.

12. van den Berg J, Kremer Hovinga JA, Pfleger C, Hegemann I, Stehle G, Holbro A, et al. Daratumumab for immune thrombotic thrombocytopenic purpura. Blood Adv. 2022; 6: 993-7.

13. Aggarwal A, White D, Pavord S, Thomas W, Desborough MJR. Daratumumab for refractory immune-mediated thrombotic thrombocytopenic purpura. Br J Haematol. 2023; 202: 429-33.

14. Xie XT, Xiao YY, Zhang Y, Luo ZM, Luo Y. Combination regimens containing daratumumab for initial diagnosed acquired thrombotic thrombocytopenic purpura. J Thromb Thrombolysis. 2023; 55: 399-405.

15. Jansen AJG, Rab MAE, Te Boekhorst PAW, Leebeek FWG. Successful treatment with daratumumab for splenectomy refractory immune-mediated thrombotic thrombocytopenic purpura. Br J Haematol. 2024 Oct 20. doi: 10.1111/bjh.19856. Epub ahead of print.

16. Weisinger J, Bouzid R, Ranta D, Woaye-Hune P, Cohen-Aubart F, Gaible C, Marjanovic Z, Corre E, Joly AC, Baylatry MT, Joly BS, Veyradier A, Coppo P. Efficacy and safety of daratumumab in multiresistant immune-mediated thrombotic thrombocytopenic purpura. Br J Haematol. 2024 Nov;205(5):1951-1958.