Patients must meet the following criteria for study entry:

1. Patient/legally authorized representative has signed the Informed Consent Form
2. Age ≥18 years
3. AIS symptom onset within 4.5 to 24 hours

   Stroke onset is defined as the time the patient was last known to be at their neurologic baseline. (Wake-up strokes are eligible if they present within the 4.5- to 24-hour time limits of last known well.)

   Note: All study-related treatment needs to be initiated within 24 hours.
4. Signs and symptoms consistent with the diagnosis of an acute anterior circulation ischemic stroke involving occlusion of the ICA, M1, or M2 vessels
5. Functionally independent (mRS 0–2) prior to stroke onset
6. Baseline NIHSS ≥5 and that remains ≥5 immediately prior to randomization
7. **Neuroimaging:** ICA or M1, M2 occlusion (carotid occlusions can be cervical or intracranial, with or without tandem MCA lesions) by *magnetic resonance angiography* (MRA) or CTA **AND** target mismatch profile on *CTP* or *MR perfusion (MRP)* (ischemic core volume <70 mL, mismatch ratio is ≥ 1.8 and mismatch volume is ≥ 15 mL)
   - The mismatch volume is determined by FDA-approved imaging software in real time based on the difference between the ischemic core lesion volume and the *time to maximum of the residue function* (T_{max}) >6s lesion volume. If both a *CTP* and a multimodal MRI scan are performed prior to enrollment, the later of the 2 scans is assessed to determine eligibility. For patients screened with MRA, only an intracranial MRA is required (cervical MRA is not required). Cervical and intracranial CTA are typically obtained simultaneously in patients screened with CTA, but only the intracranial CTA is required for enrollment.
   - *Enrollment of patients with an ICA (including proximal and tandem ICA occlusions) will be capped at no more than 15% of the target study population.*

   Alternative neuroimaging:
   - If CTA (or MRA) is technically inadequate: T_{max} >6s perfusion deficit consistent with an ICA or M1, M2 occlusion **AND** target mismatch profile (ischemic core volume <70 mL, mismatch ratio ≥1.8 and mismatch volume >15 mL as determined by RAPID software)
   - If MRP is technically inadequate: ICA or M1, M2 occlusion by MRA **AND** diffusion-weighted imaging (DWI) lesion volume ≤ 25 mL for an M1 or ICA occlusion and ≤ 15 mL for an M2 occlusion. If MRA is technically inadequate, a CTA can be used if performed within 60 minutes prior to the MRI. Carotid occlusions can be cervical or intracranial; with or without tandem MCA lesions.
   - If CTP is technically inadequate: Patient can be screened with MRI and randomized if neuroimaging criteria are met.
8. Ability to comply with the study protocol, in the investigator's judgment

Patients who meet any of the following criteria will be excluded from study entry:

General

1. Current participation in another investigational drug or device study
2. Known hypersensitivity or allergy to any ingredients of tenecteplase
3. Active internal bleeding
4. Known bleeding diathesis
5. Known hereditary or acquired hemorrhagic diathesis, coagulation factor deficiency; recent oral anticoagulant therapy with INR >1.7
6. Use of one of the new oral anticoagulants within the last 48 hours (dabigatran, rivaroxaban, apixaban, edoxaban)
7. Treatment with a thrombolytic within the last 3 months prior to randomization
8. Intracranial neoplasm (except small meningioma), arteriovenous malformation, or aneurysm

   Any patient with an aneurysm located anywhere where a catheter or wire may be used in the thrombectomy procedure should be excluded. Additionally, patients with proximally thrombosed aneurysms suspected to be the mechanism of thromboembolism of the affected vascular territory should not be considered for enrollment due to the need for additional surgical or endovascular treatment that may require additional antithrombotic therapy.

   Aneurysms located elsewhere in the intracranial vasculature do not constitute an exclusion criterion per se, subject to the standards of care and the discretion of the treating physician. Additionally, previously treated intracranial aneurysms are not a criterion for exclusion, although, patients who underwent intracranial surgery within 2 months of screening are excluded.
9. Seizures at stroke onset if it precludes obtaining an accurate baseline NIHSS
10. Pre-existing medical, neurological, or psychiatric disease that would confound the neurological or functional evaluation

    COVID-19 positive and/or suspected (i.e., symptomatic) patients are not eligible unless previously tested positive for COVID-19 AND have been asymptomatic at a minimum 10 days from time of screening.
11. Severe, uncontrolled hypertension (systolic blood pressure >180 mmHg or diastolic blood pressure >110 mmHg)
12. For patients with suspected coagulopathy, platelet count must be checked prior to randomization and patient is excluded if baseline platelet count <100,000/uL
13. Baseline blood glucose >400 mg/dL (22.20 mmol/L)
14. Baseline blood glucose <50 mg/dL (2.78 mmol/L) needs to be normalized prior to randomization
15. Clot retrieval attempted using a neurothrombectomy device prior to randomization
16. Intracranial or intraspinal surgery or trauma within 2 months

17. Other serious, advanced, or terminal illness with life expectancy less than 6 months (investigator judgment)
18. History of acute ischemic stroke in the last 90 days
19. History of hemorrhagic stroke
20. Presumed septic embolus; suspicion of bacterial endocarditis
21. Any other condition that, in the opinion of the investigator, precludes an endovascular procedure or poses a significant hazard to the patient if an endovascular procedure was to be performed
22. Pregnant

Imaging

23. Unable to undergo a contrast brain perfusion scan with either MRI or CT
24. Extensive early ischemic change (hypodensity) on non-contrast CT estimated to be >1/3 MCA territory, or significant hypodensity outside the T_{max}>6s perfusion lesion that invalidates mismatch criteria (if patient is enrolled based on CT perfusion criteria)
25. Significant mass effect
26. Acute symptomatic arterial occlusions in more than one vascular territory confirmed on CTA/MRA (e.g., bilateral MCA occlusions, or an MCA and a basilar artery occlusion)
27. Evidence of intracranial tumor (except small meningioma) acute intracranial hemorrhage, neoplasm, or arteriovenous malformation

ML40787
Data Collection Checklist

TIMELESS
A study of imaging-eligible stroke treatment

➢ ***The following assessments are required during Screening/Randomization visit:***

- **Informed consent**
  - ☐ Informed Consent

  Demographics
  - ☐ Veteran Status
  - ☐ Race/Ethnicity
- **Hospital Arrival**
  - ☐ Date and Time of Qualifying Stroke
  - ☐ Randomization Hospital
  - ☐ Was subject transferred from nECC hospital?
  - ☐ Is mechanical thrombectomy planned?
- **Vital Signs**
  - ☐ Temperature
  - ☐ Pulse
  - ☐ Respiratory Rate
  - ☐ Systolic and Diastolic blood pressure
  - ☐ Actual Weight (whole number to the nearest tenth)
- **Acute Stroke Imaging**

  a. RAPID Imaging Software Results
  - ☐ Ischemic Core Volume
  - ☐ Absolute Mismatch Volume
  - ☐ Mismatch Ratio
  - ☐ Tmax >6s Lesion Volume

  b. MRI Scan
  - ☐ Type of Occlusion

    DWI Lesion Volume

  OR

  c. CT-Scan
  - ☐ Type of Occlusion
  - ☐ CTP Infarct Volume
  - ☐ ASPECTs Score
- **Questionnaires**
  - ☐ NIHSS (closest to randomization)
  - ☐ mRS
- **Lab Results**
  - ☐ Glucose
  - ☐ Coagulation (only if patient is taking an anticoagulant)
  - ☐ Platelets (Results must be available prior to treatment if suspected coagulopathy)
  - ☐ Pregnancy Test (if applicable – WOCBP)
- **Study Drug Administration**
  - ☐ Kit ID Number Administered
  - ☐ Dose Administered
  - ☐ Dose Volume Administered

  Note: Use a double verification system for both drug kit number and drug dose per your local hospital guidelines
- **Targeted Medical History**
  - ☐ Myocardial Infarction
  - ☐ Hypertension
  - ☐ Atrial Fibrillation
  - ☐ Hypercholesterolemia
  - ☐ Diabetes
  - ☐ Prior Stroke (excluding qualifying stroke)

Potential Eligible Patient Criteria

- ☐ **Age** 18 years or older
- ☐ **4.5-24 hour onset** window from last known well
- ☐ Signs and symptoms consistent with LVO **M1/M2**
- ☐ **NO** signs or symptoms of a potential **hemorrhage**
- ☐ **mRS** is 0-2 prior to onset
- ☐ **NIHSS** is 5 or greater
- ☐ **NO** known **contraindications** to thrombolytic treatment

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TIMELESS Procedural Checklist

TIMELESS
study of imaging-eligible stroke treatment

Prior to Treatment	Treatment
• **Baseline** ○ Pre-Stroke mRS Baseline (must be 0-2) ○ NIHSS Assessment (closest to randomization, must be ≥ 5) • **Qualified RAPID Imaging Results** ○ Ischemic Core Volume < 70 mL ○ Mismatch Ration ≥ 1.8 ○ Mismatch Volume ≥ 15 mL • **Signed Informed Consent by patient or LAR** • **Required Assessments** ○ **Glucose (finger or blood draw)**—*exclude if >400 mg/dL (22.20 mmol/L). If under <50mg/dL then will need to normalize prior to randomization.* ○ **Coagulation Tests**: INR, aPTT, PT (only if patient is taking an anticoagulant). *INR must be < 1.7* ○ **Platelet Count**—*exclude if < 100,000/*uL (Results must be available prior to treatment if suspected coagulopathy) ○ **Pregnancy Test (urine or serum test)**: If applicable ○ **Blood Pressure**—exclude if systolic blood pressure > 180 mmHg or diastolic blood pressure > 110 mmHg **Note:** All baseline blood and urine samples must be collected prior to randomization. **Prohibited:** Intra-arterial thrombolytic agents or intracranial stenting	• **IxRS Entry** • If the infarct volume = 0, the mismatch ratio will be infinity. Use "100.0" instead. • Patient Actual Weight (rounded to nearest whole number). Note: IxRS will require entry to the nearest tenths. Enter ".0" as a workaround (i.e. 57.0 kg) • **Kit Dispensation** • **Study Drug Administration** (Dose Within 90 Minutes of Qualified RAPID Imaging Results): The study drug should be administered prior to groin puncture and **must** be administered prior to manipulation of the clot.

Post-Treatment

- **Image Submission**
  - **Medical Quality Review Email** ASAP at timeless_rapid-d@gene.com Send over the RAPID Summary Maps for both Baseline and 24-Hour Images. Remember to archive your RAPID emails with patient medical records
  - **UCLA Central Core Imaging Lab** with *Core Lab Cover Page*
    - as a batch 2 weeks after last patient brain imaging
  - or within 5 business days for suspected sICH
  - Archive your raw data perfusion source images before they're purged from your servers

- **NIHSS Assessment and Imaging for sICH**
  (within 36 hours of drug admin)

- **Discharge Activities**
  (may be performed if discharge is prior to 72 hours)
  Ensure that caregiver, LAR or alternate contacts are collected for follow up visits

- **Follow-Up Visits**
  (24 hours +/- 6 hours; 72-96 Hours; Day 30 +/- 7 days and Day 90 +/- 14 days): In-Person, or Phone Call (at least verbal mRS; AE/AESI/SAE assessment)

- **Prohibited Medication After Treatment With Study Drug:**
  Within 24 Hours:

    Oral or parenteral anticoagulants excluding 1) prophylactic doses, if administered per institutional practice and 2) **low doses of IV heparin** during the thrombectomy procedure, if this practice is considered standard of care for patients who have recently received an intravenous thrombolytic at the treating institution.

  First 90 Days After Treatment:

    Any additional thrombolytic with the **exception of Cathflo Activase**, if needed for Central Venous Catheter Occlusion management.

TIMELESS IMAGING GUIDANCE CHECKLIST
(v. 28Sep2021)

IMAGING TO DETERMINE ELIGIBILITY FOR TIMELESS TRIAL	
Multimodal CT	**MRI**
To determine LVO (CTA) and Penumbra (CTP): - 8 cm of brain coverage required (for scanners unable to cover 8 cm of brain, 2 slabs may be required) - 2 contrast injections required if 2 slabs are needed for 8 cm of brain image - Occlusion can be cervical or intracranial and with/without tandem MCA lesions AND Target Mismatch required CTP can precede CTA *Used in multiple previous trials – no evidence of renal impairment or harm to patients*	**To determine LVO (MRA) and Penumbra (MRP/DWI):** -Occlusion can be cervical or intracranial and with or without tandem MCA lesions AND Target Mismatch required Cervical MRA is not required

IMAGE TYPE	REQUIRED IMAGES	DETAILS
Screening / Baseline	☐ **RAPID Summary Maps** ☐ **All Qualifying Images** ☐ Baseline Non-Contrast CT ☐ DICOM Raw/Source Images pushed to RAPID ☐ All CT or MR Perfusion Images ☐ CTA or MRA Images as Vessel Occlusion Evidence ☐ DSA Image (includes all pre-thrombectomy, mechanical thrombectomy, and post thrombectomy images obtained during angiogram) *NOTE: If images are repeated, only send the latest version over (i.e. we do not need the pre-baseline images)*	• **Non-Contrast CT** to rule out bleed. • **DICOM Images** are used as baseline criteria to help with final calculations of infarct volumes, and infarct growth. DICOM images are only available on your servers for a limited amount of time, so it is vital to ensure these are captured and included in the submission packet. • **CTA and/or MRA** is to verify the location of the vessel occlusion. • **RAPID Perfusion Maps** taken at Baseline are used to determine eligibility • **DSA Images** determine TICI scores and recanalization values
24 Hour	☐ **RAPID Summary Maps** ☐ **MRI/MRA/MRP** as the preferred imaging, but a CT perfusion package (**CT/CTA/CTP**) will also be sufficient. Include their associated **DICOM/Source Images** ☐ Any **additional contrast images** should also be included	All of the DICOM source images and RAPID post-processing maps need to be uploaded for the central reader. *Note: It's acceptable (though not preferred) if your site mix/match your imaging modality, as long as we have a complete "set" of perfusion, angiography, and DICOM/Source Images*
72 HR	☐ **MRI** with **Flair**, **DWI** and **GRE** image. ☐ If MR is not available a **Non-Contrast CT** will suffice ☐ Include **any CT or MRI scans** that are obtained as part of usual care or obtained if the patient has had clinical deterioration	These images are important for comparison to baseline and providing a final infarct volume *Note: Flair, DWI, and GRE images are required for the 72 HR Visit*
Unscheduled	If (for any reason) patients has an additional angiogram procedure, please collect: ☐ **Any additional CT or MRI images** obtained due to patient clinical changes or obtained to monitor swelling or hemorrhage.	These images are essential for providing information regarding patient status and adverse events. They help the trial team to evaluate the patients' full story from hospital admission to discharge

BRAIN IMAGING SUBMISSION GUIDANCE

(v. 09Sep2021)

Timeframe	When	What	Where	Why
Patient Dosing	ASAP	Forward the RAPID Summary Map Emails for Baseline and 24-Hour visits	Medical Review Team Email: timeless_rapid-d@gene.com Archive with patient medical records	For quality review and reference for the 24/7 On-Call Team
36 Hours of Patient Dosing	Within 5 Business Days of suspected ICH imaging	• RAPID Summary Map • ALL Brain Imaging Discussed Above	UCLA Central Core Imaging Portal Images must be sent all at once in a single batch (unless they're collected due to suspected ICH, for these images, they need to be sent within 5 business days for safety assessment) All images must include an emailed cover page sent to: timeless_imagingcoverpage-d@gene.com and please CC' your CRA	For Image Analysis
72-96 Hours of Patient Dosing	Within 2 Weeks after the patient's last brain scan			

Part 1: Study Drug Preparation: Reconstitution. To prepare each of the two study drug kits, you'll need:

- Assigned study drug vials
- Two 10 milliliter vial of sterile water for injection, USP (SWFI)—Do not use bacteriostatic water for injection
- Two 10 milliliter and a 5 milliliter syringe
- Two alcohol wipes
- A patent IV line, saline containing solution
- One to two syringes for flushing the line as needed

Please note, Genentech will only provide the study drug. All other items need to be sourced locally by your pharmacy.

Steps

1. Withdraw 10 milliliters of Sterile Water for Injection from the vial, with the 10-milliliter syringe.
2. Inject the Sterile Water for Injection into the study drug vial.
3. Gently swirl the vial until the contents are completely dissolved. The solution should look colorless or pale yellow and transparent. The reconstituted volume will be 10 milliliters, and contain 5 milligrams of study drug per milliliter.
4. Repeat for the second vial. Upon reconstitution, the study drug will remain stable for a maximum of 4 hours up to 30C (86F) in the vial.

Part 2: Prior to Administration:

- Validate the kit you are going to administer is the same kit as assigned in IWRS (see the number on the vial)
- Use a double verification system for both drug kit number and drug dose.

Part 3: Drug Administration:

1. Clean the study drug vial with an alcohol wipe.
2. Withdraw the calculated volume/dose of study drug, as assigned by IWRS, with the 5-milliliter syringe.
3. If the IV line contains dextrose, flush it with the saline-containing solution before and after the study drug administration.
4. Clean the hub of the IV line with an alcohol wipe and administer the study drug through the patient's IV line with the 5-milliliter syringe, as a single bolus over 5 seconds.
5. The study drug needs to be administered within 90 minutes of qualifying brain imaging.
6. Document the time of administration, and the dose.
7. Documentation of both vials assigned to the patient at screening should be recorded. For the vial to be administered, secondary review and verification is required and documentation is needed.
8. Once the study drug has been administered, discard the non-selected vial, syringes, and all other materials appropriately, according to local guidelines. Any leftover drug should be returned to Genentech, unless prior approval has been given to destroy it on site.

Contact Information & Instructions

24/7 Medical Emergency Hotline
Call: +1.888.316.3335

RAPID Quality Review
Submit RAPID maps **ASAP** following patient randomization to: timeless_rapid-d@gene.com

Core Imaging Lab
Send to: https://strokedrop.neurology.ucla.edu **Upload Timelines for all imaging:** • As a batch 2 weeks after last patient brain imaging (i.e. post 72-96 hours follow-up) • Within **5 business days** if imaging is related to sICH

IschemaView RAPID 24/7 Support
Call: +1 650.388.9767 (ext 2) This is an answering service, response to messages is immediate.

endpoint (IxRS) 24/7 Tech Help Desk
Call: +1 877.810.4786

TCS e-CONSENT Support
Call: +1 866.768.2875