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Tenecteplase thrombolytic therapy for acute ischaemic stroke in China: a real-world, multicentre, retrospective, controlled study

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ABSTRACT

Background and aims Tenecteplase (TNK) offers logistical advantages in stroke thrombolytic therapy with its single bolus administration compared with alteplase. We aim to investigate the real-world evidence regarding its safety and effectiveness in China.

Methods We conducted a retrospective study on patients receiving alteplase or TNK for acute ischaemic stroke (AIS) within 4.5 hours of onset between 1 March 2019 and 1 October 2023, from 18 stroke centres in China. Using propensity score matching (PSM), TNK-treated patients were matched 1:1 with alteplase-treated patients. The primary outcome was the rate of symptomatic intracranial haemorrhage (sICH) within 72 hours post-thrombolysis. Secondary outcomes comprised the rate of parenchymal haemorrhage type 2, any intracranial haemorrhage, any systematic bleeding and mortality at 90 days, as well as 24-hour National Institutes of Health Stroke Scale (NIHSS), early neurological improvement at 24 hours, modified Rankin Scale (mRS) shift, percentage of mRS 0–1 and mRS 0–2 at 90 days.

Results We identified 1113 patients with AIS who received TNK and 2360 patients who received alteplase. Following PSM, 1113 TNK-treated patients with AIS were matched to 1113 patients treated with alteplase. No significant differences were observed in rates of sICH (1.8% vs 1.98%, p=0.864) or other safety outcomes. Moreover, TNK-treated patients demonstrated a lower rate of any intracranial haemorrhage (OR: 0.51, 95% CI: 0.31 to 0.86, p=0.012). A higher proportion of patients achieving early neurological improvement at 24 hours (OR: 1.76, 95% CI: 1.48 to 2.09, p=0.000), better 90-day mRS (OR: 0.67, 95% CI: 0.57 to 0.79, p=0.000) as well as higher percentages of 90-day mRS 0–1 (OR: 1.27, 95% CI: 1.05 to 1.54, p=0.012) and mRS 0–2 (OR: 1.41, 95% CI: 1.14 to 1.75, p=0.001) compared with alteplase.

Conclusions Thrombolysis with TNK is not associated with an increased risk of sICH, and may result in better early neurological improvement and 90-day functional outcomes compared with alteplase in patients with AIS.

WHAT IS ALREADY KNOWN ON THIS TOPIC

⇒ Tenecteplase is non-inferior to alteplase in patients who had acute ischaemic stroke within 4.5 hours from symptom onset. Nevertheless, no real-world evidence concerning its safety and effectiveness in China exists.

WHAT THIS STUDY ADDS

⇒ This study confirms the safety of tenecteplase in Chinese patients with acute ischaemic stroke, as there is no increase in symptomatic intracranial haemorrhage or mortality within 90 days. Tenecteplase also shows potential for enhanced early neurological improvement at 24 hours and favourable functional outcomes at 90 days compared with alteplase in treating patients who had acute ischaemic stroke within 4.5 hours of onset.

HOW THIS STUDY MIGHT AFFECT RESEARCH, PRACTICE OR POLICY

⇒ Real-world data add confidence in the use of tenecteplase for eligible patients who had acute ischaemic stroke within 4.5 hours of onset.

INTRODUCTION

Tenecteplase (TNK) is a genetically engineered medication derived from alteplase (rt-PA).[1] Its prolonged half-life enables rapid administration through bolus infusion, offering logistic advantages over rt-PA. Moreover, TNK possesses increased resistance to inactivation by plasminogen activator inhibitor-1, resulting in greater potency in lysing platelet-rich clots.[2] Its heightened specificity for fibrin may contribute to a reduction in haemorrhagic compilations, particularly in systemic bleeding.[3 4] These features are evidenced both in studies on myocardial infarction[5] and acute ischaemic stroke (AIS).[6]

Recent phase III randomised controlled clinical trials (RCT)[7–9] have shown that TNK at 0.25 mg/kg is non-inferior to rt-PA in efficacy and exhibits similar safety profiles for patients with AIS within 4.5 hours of symptom onset. Meta-analyses of multiple trials suggest potential additional benefits for patients with large vessel occlusion (LVO) with

higher recanalisation rates and smaller hypoperfusion lesions.[10 11] Consequently, guidelines from the European Stroke Organization (ESO)[12] and the American Heart Association (AHA)[13] recommend considering TNK at a dose of 0.25 mg/kg for eligible patients with AIS–LVO instead of rt-PA within 4.5 hours of symptom onset. Furthermore, real-world data from Europe and the USA indicate comparable or lower rates of symptomatic intracranial haemorrhage (sICH) with TNK thrombolysis, along with significantly improved functional outcomes for AIS.[14 15]

Given these circumstances, TNK has seen extensive off-label use in patients with AIS at stroke centres in China under various conditions in recent years. Hence, this study aims to evaluate the safety and efficacy of TNK in patients with AIS within 4.5 hours of symptom onset in a real-world clinical setting in China by analysing propensity score-matched (PSM) data.

METHODS

Study design and participants

Our study was a retrospective, observational, comparative study involving 18 thrombolysis-capable stroke centres (online supplemental table 1) across various regions in China, including Shanghai, Inner Mongolia, Liaoning Province, Jilin Province, Shandong Province, Henan Province and Zhejiang Province. Patients included in our study consisted of consecutive individuals over 18 years old, diagnosed with ischaemic stroke within 4.5 hours of symptom onset. These patients were subjected to intravenous thrombolysis using either TNK or rt-PA between 1 March 2019 and 1 October 2023. The decision of administering rt-PA (Actilyse; Boehringer Ingelheim, Germany) or TNK (CSPC Recomgen Pharmaceutical (Guangzhou) Co, Ltd) was made by local clinicians in accordance with local protocols. The majority of sites used the standard rt-PA dosage (0.9 mg/kg), while few cases used a reduced dosage (0.6 mg/kg). On the other hand, TNK was administered at 0.25 mg/kg or 16 mg per individual patient. Due to financial considerations, local clinicians often choose to administer 16 mg (one vial) for patients with weights slightly over 64 kg. Thrombolysis was administered within 4.5 hours of symptom onset or the last known-well time, with non-contrast CT being the standard imaging modality. Patients with contraindications to thrombolysis, those who lacked post-thrombolysis CT imaging follow-up, those with unclear onset-to-treatment times and those who participated in other clinical trials were excluded from the study. The modified Rankin scale (mRS) score was evaluated by local investigators via structured telephone interviews, which were standard follow-up procedures for thrombolytic patients at 90±7 days post-symptoms onset.

Patient and public involvement

Since this is a retrospective study, patients are not enrolled as research participants at the time they received thrombolytic treatment. Patients are only contacted if consent is required for the use of their data in the study.

Baseline characteristics

Standardised and prespecified variables were documented by local investigators at participating centres using electronic case report forms. These variables included demographic characteristics (sex, age, prestroke disability measured by the mRS), risk factors (smoking, hypertension, diabetes, dyslipidaemia, atrial fibrillation), concomitant medications (antiplatelet agents, oral anticoagulants) and medical history (prior heart failure, coronary heart disease and previous ischaemic stroke/TIA). Clinical variables on admission included the National Institutes of Health Stroke Scale (NIHSS) score, systolic blood pressure (SBP), blood glucose levels and thrombectomy performance. Time parameters such as door-to-needle time (DNT), door-to-puncture (DTP) time and onset-to-treatment time (OTT) were also documented. A plausibility check was conducted, and missing data were addressed (online supplemental table 2).

Outcomes

The primary outcome of the study was the rate of sICH, defined as clinical deterioration or neurological decline resulting in a ≥4-point increase in NIHSS score due to intracranial haemorrhage evidenced on CT imaging within 72 hours post-thrombolysis compared with baseline (ECASSIII criteria).[16] This outcome was reported and reviewed centrally to confirm the accuracy. Other safety and efficacy endpoints were evaluated by local investigators without blinding. Secondary outcomes comprised additional safety and efficacy endpoints. Safety endpoints included the rates of parenchymal haemorrhage type 2 (PH2) according to the ECASS morphological definitions,[17] any intracranial bleeding, any systematic bleeding requiring blood transfusion and mortality at 90 days. The efficacy endpoints included the 24-hour NIHSS score and the proportion of patients showing early neurological improvement, defined as a reduction of at least four points or achieving a score of no more than 1 on the NIHSS score at 24 hours.[7 18] Additionally, a shift analysis of the 90-day mRS score was conducted, along with the proportion of patients achieving excellent (mRS=0–1) and good (mRS=0–2) functional outcomes at 90 days.

Statistical analysis

Missing values were handled using the Multiple Imputation Chained Equations method in Rstudio (V.12.0). The imputation model included all baseline variables with missing data using logistic regression for categorical variables and predictive mean matching for continuous variables. Five imputed datasets were generated to address uncertainty. Variables with a missing rate>20% or lacking primary outcomes were excluded.

Ordinal and categorical variables were presented as absolute numbers (No.) and percentages (%), while normally distributed continuous variables were expressed

as means (SD), and non-normally distributed variables were expressed as medians (IQR).

To minimise confounding effects from baseline characteristics, we employed PSM to match patients in the TNK group with those receiving rt-PA. Propensity scores were estimated using logistic regression, incorporating a range of covariates that were statistically significant in the univariate analysis and variables known to predict outcome (ie, age, sex, NIHSS score) even if p>0.05. Patients in the TNK group were matched to rt-PA recipients in a 1-to-1 ratio, with a calliper of 0.2 SD using the nearest neighbour matching approach. A non-significant univariable analysis (p>0.05) and a standardised mean difference<0.2 indicated a successful balance between the groups (online supplemental table 3).

Statistical comparisons were first conducted in unmatched and then propensity score matched groups using the Mann-Whitney U test for continuous and ordinal variables and the χ^2 test for categorical variables. Observed differences were considered significant if p<0.05. Logistic regression was used to evaluate the association between treatment and outcomes, with ORs and 95% CIs calculated. Different hospitals were treated as random effects in the model. Additionally, quantile and ordinal logistic regression were used to analyse the 24-hour NIHSS score and 90-day mRS shift. The DNT of TNK thrombolysis was previously shown to be shorter than that of alteplase,[19] primarily due to the single-bolus injection administration. Additionally, DNT was considered as an intrinsic feature of the thrombolytic drug rather than a differentiating factor between the two patient groups. Therefore, in the PSM cohort, if a significant difference in DNT between the two groups is identified, it will be appropriately adjusted for in the analysis.

A sensitivity analysis was conducted post hoc to examine the robustness of results, using multivariable logistic regression to adjust for additional factors. Model 1 adjusted the same variables adopted for PSM, including demographic variables (age, sex), pre-mRS, risk factors (smoking, diabetes, hyperlipidaemia, atrial fibrillation), clinical presentation (baseline NIHSS, blood glucose level at emergency, SBP at emergency, thrombectomy performance), previous medical history (heart-failure, ischaemic stroke/TIA) and prior medication (antiplatelet drugs). Model 2 incorporated the same variables from model 1 as well as onset-to-treatment time. Model 3 retained the variables from model 1 and excluded data from one of the largest centres (Inner Mongolia), which contributed 40% of the total TNK data.

Subgroup analysis was also performed post hoc, stratifying factors such as sex (female or male), age (≤80 years old or >80 years old), stroke severity (baseline NIHSS score ≤10 or baseline NIHSS score >10), thrombectomy performance (yes or no) and centre level (tertiary A or non-tertiary A). Despite DNT not being accounted for in the baseline variables, its substantive influence on outcomes warranted investigation. Therefore, we conducted a mediation analysis to address any potential impact of DNT on the primary outcome.

PSM analyses were conducted using RStudio (R, 2023.12.0) with the 'MatchIt' package and other statistical analyses were performed using Stata Statistical Software Release 13 (StatCorp, College Station, Texas, USA).

RESULTS

We identified 1113 patients with AIS who received intravenous TNK (median dose: 0.25 mg/kg, IQR=0.23–0.25) and 2360 patients who received intravenous rt-PA (median dose: 0.9 mg/kg, IQR=0.9–0.9) (table 1, online supplemental figure 2) between 1 March 2019 and 1 October 2023, at 18 stroke centres across China.

Patients treated with intravenous TNK were significantly younger (66 years vs 68 years, p=0.000), with lower prevalence of smoking (18.33% vs 34.32%, p=0.000), diabetes mellitus (18.06% vs 29.94%, p=0.000), dyslipidaemia (5.12% vs 16.02%, p=0.000), atrial fibrillation (8.09% vs 14.49%, p=0.000), heart failure (1.08% vs 2.29%, p=0.015), previous ischaemic stroke/TIA (14.3% vs 19.58%, p=0.000) and antiplatelet medication usage (9.52% vs 18.09%, p=0.000) compared with patients treated with intravenous rt-PA. The TNK-treated patients also had a significantly lower rate of prestroke disability, as indicated by the mRS before the stroke event (0 vs 0, p=0.006), while the neurological symptoms presented at the hospital, quantified by the NIHSS scores at baseline (5 vs 5, p=0.334), were comparable with the rt-PA group. The rate of subsequent bridging with endovascular thrombectomy was lower in the TNK group (8.36% vs 11.27%, p=0.008) compared with the rt-PA group. Patients treated with TNK had faster DNT (29 min vs 38 min, p=0.000) and DTP time (110 min vs 154 min, p=0.001) despite a longer OTT time (180 min vs 140 min, p=0.000) (table 1).

No significant differences were observed in rates of sICH (1.8% vs 2.33%, OR: 0.77, 95% CI: 0.46 to 1.29, p=0.313). Moreover, patients treated with TNK had a significantly lower occurrence of any intracranial bleeding (2.07% vs 4.62%, OR: 0.44, 95% CI: 0.27 to 0.69, p=0.000), and all-cause mortality at 90 days (6.22% vs 8.20%, OR: 0.74, 95% CI: 0.56 to 0.99, p=0.042) compared with the rt-PA group. However, there were no differences of PH2 (1.35% vs 1.74%, OR: 0.77, 95% CI: 0.43 to 1.40, p=0.396) or any systemic bleeding (0.45% vs 0.13%, OR: 3.55, 95% CI: 0.85 to 14.86, p=0.065) (table 2, figure 1).

Patients treated with TNK showed superior early neurological improvement with reduced NIHSS scores (2 vs 3, p=0.000) and a higher proportion achieving an NIHSS score reduction of more than 4 or an NIHSS score less than 1 (57.49% vs 41.01%, OR: 1.95, 95% CI: 1.68 to 2.25, p=0.000) within the first 24 hours. At 90 days, ordinal logistic regression analysis demonstrated that those treated with TNK had better 90-day mRS (OR: 0.60, 95% CI: 0.52 to 0.68, p=0.000) and were more likely to achieve mRS 0–2 (80.90% vs 73.60%, OR: 1.52, 95% CI: 1.27 to 1.82, p=0.000) and mRS 0–1 (72.03% vs 64.18%,

Table 1 Baseline characteristics of unmatched and propensity score matched TNK and alteplase group

Characteristic	Unmatched groups			Propensity score matched group		
	TNK n=1113	Alteplase n=2360	P value	TNK n=1113	Alteplase n=1113	P value
Demographics						
Age, years, median (IQR)	66 (58.5, 73)	68 (60, 76)	0.000	66 (58.5, 73)	67 (58, 74)	0.214
Male, n, (%)	713 (64.06%)	1567 (66.4%)	0.176	713 (64.06%)	698 (62.71%)	0.509
Pre-mRS, median (IQR)	0 (0, 0)	0 (0, 0)	0.006	0 (0–0)	0 (0–0)	0.595
mRS=0, %	1002 (90.03%)	2041 (86.48%)		1002 (90.03%)	1008 (90.57%)	
mRS=1, %	66 (5.93%)	194 (8.22%)		66 (5.93%)	64 (5.75%)	
mRS=2, %	32 (2.88%)	76 (3.22%)		32 (2.88%)	35 (3.14%)	
mRS=3, %	7 (0.63%)	26 (1.1%)		7 (0.63%)	3 (0.27%)	
mRS=4, %	6 (0.54%)	21 (0.89%)		6 (0.54%)	3 (0.27%)	
mRS score=5, %	0 (0%)	2 (0.08%)		0 (0%)	0 (0%)	
Pre-mRS=0–1, n (%)	1068 (95.96%)	2235 (94.7%)	0.110	1068 (95.96%)	1072 (96.32%)	0.66
Risk factors, No. (%)						
Smoking	204 (18.33%)	810 (34.32%)	0.000	204 (18.33%)	201 (18.06%)	0.869
Hypertension	702 (63.07%)	1538 (65.17%)	0.228	702 (63.07%)	675 (60.65%)	0.239
Diabetes mellitus	201 (18.06%)	683 (28.94%)	0.000	201 (18.06%)	212 (19.05%)	0.549
Dyslipidaemia	57 (5.12%)	378 (16.02%)	0.000	57 (5.12%)	74 (6.65%)	0.126
Atrial fibrillation	90 (8.09%)	342 (14.49%)	0.000	90 (8.09%)	87 (7.82%)	0.814
Previous history, No. (%)						
Coronal heart disease	118 (10.60%)	284 (12.03%)	0.218	118 (10.60%)	107 (9.61%)	0.439
Heart failure	12 (1.08%)	54 (2.29%)	0.015	12 (1.08%)	19 (1.71%)	0.205
Ischaemic stroke/TIA	159 (14.3%)	462 (19.58%)	0.000	159 (14.3%)	170 (15.27%)	0.511
Concomitant medication, No. (%)						
Antiplatelet drugs	106 (9.52%)	427 (18.09%)	0.000	106 (9.52%)	100 (8.98%)	0.611
Anticoagulant drugs	39 (3.50%)	69 (2.92%)	0.358	39 (3.50%)	19 (1.71%)	0.008
Clinical presentation at emergency						
NIHSS score on admission, median (IQR)	5 (3, 8)	5 (3, 9)	0.334	5 (3, 8)	5 (3, 9)	0.427
Glucose, mmol/L, median (IQR)	6.53 (5.6, 8.2)	6.8 (5.6, 8.8)	0.007	6.53 (5.6, 8.2)	6.7 (5.6, 8.35)	0.423
Systolic blood pressure, median (IQR)	146 (130, 160)	146 (133, 162)	0.023	146 (130, 160)	143 (130,160)	0.599
Thrombolytic drug dose (mg/kg), median (IQR)	0.25 (0.23, 0.25)	0.9 (0.9, 0.9)	NA	0.25 (0.23, 0.25)	0.9 (0.9, 0.9)	NA
Thrombectomy, n (%)	93 (8.36%)	266 (11.27%)	0.008	93 (8.36%)	109 (9.79%)	0.238
Door to needle time, DNT, minutes, median (IQR)	29 (21, 42)	38 (27, 55)	0.000	29 (21, 42)	36 (25, 53)	0.000
Door to puncture time, DTP, minutes, median (IQR)	110 (100, 167.5)	154 (104.5, 210)	0.001	110 (100, 167.5)	132 (95, 207)	0.0667
Onset to treatment time, OTT, minutes, median (IQR)	180 (123, 224.5)	140 (102, 185)	0.000	180 (123, 224.5)	142 (100, 186)	0.000

*Univariate comparison between TNK and alteplase.
BP, blood pressure; DNT, door to needle time; DTP, door to puncture; mRS, modified Rankin Scale; NIHSS, National Institutes of Health Stroke Scale; OTT, onset to treatment time; TNK, tenecteplase.

Table 2 Safety and efficacy outcomes of unmatched and PSM groups

Outcome	Unmatched groups			PSM groups		
	TNK	Alteplase	P value	TNK	Alteplase	*Adj. P* value
Primary outcomes						
sICH, n/N (%)	20/1113 (1.8%)	55/2360 (2.33%)	0.313	20/1113 (1.8%)	22/1113 (1.98%)	0.864
Secondary outcomes						
Safety outcomes, n/N (%)						
Any intracranial bleeding	23/1113 (2.07%)	109/2360 (4.62%)	0.000	23/1113 (2.07%)	45/1113 (4.04%)	0.012
PH2	15/1113 (1.35%)	41/2360 (1.74%)	0.396	15/1113 (1.35%)	16/1113 (1.44%)	0.979
Any systemic bleeding	5/1113 (0.45%)	3/2360 (0.13%)	0.065	5/1113 (0.45%)	2/1113 (0.18%)	0.379
Mortality at 90 days	68/1094 (6.22%)	181/2208 (8.2%)	0.042	68/1094 (6.22%)	77/1034 (7.45%)	0.231
Efficacy outcomes						
24-hour NIHSS score* median (IQR)	2 (1, 5), n=1108	3 (1, 7), n=2324	0.000	2 (1, 5), n=1108	3 (1, 7), n=1110	0.000
24-hour NIHSS score improvement ≥4 OR ≤1	637/1108 (57.49%)	953/2324 (41.01%)	0.000	637/1108 (57.49%)	478/1100 (43.45%)	0.000
mRS at 90 days, median (IQR)†	0 (0–2) n=1094	1 (0–3) n=2208	0.000	0 (0–2), n=1094	1 (0–3), n=1034	0.000
mRS 0–1 at 90 days, n (%)	788 (72.03%)	1417 (64.18%)	0.000	788/1094 (72.03%)	690/1034 (66.73%)	0.012
mRS 0–2 at 90 days, n (%)	885 (80.90%)	1625 (73.6%)	0.000	885/1094 (80.9%)	773/1034 (74.76%)	0.001

p value in the PSM groups were adjusted for oral anticoagulant use and DNT.
*Quantile regression.
†Ordinal logistic regression.
DNT, door-to-needle time; NIHSS, National Institutes of Health Stroke Scale; PH2, parenchymal haematoma type 2; PSM, propensity score matching; sICH, symptomatic intracranial haemorrhage; TNK, tenecteplase.

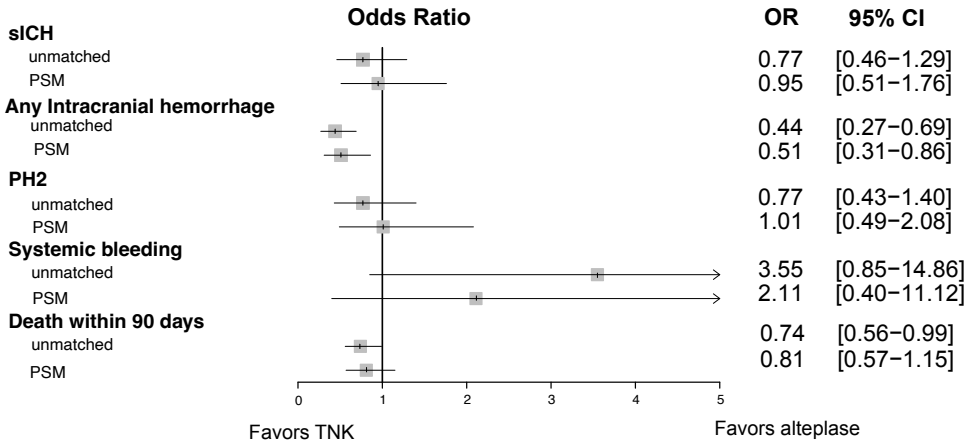

Figure 1 Safety profile of intravenous TNK compared with alteplase in the unmatched groups and after propensity score matching (PSM). The safety outcomes are presented with ORs and 95% CIs, including symptomatic intracranial haemorrhage (sICH), parenchymal haematoma type 2 (PH2), any systemic bleeding and mortality within 90 days. PSM groups were adjusted for oral anticoagulant use and DNT. No significant differences were found between the TNK and alteplase groups for these outcomes in both unmatched and PSM groups. However, the TNK group demonstrated significantly lower odds of any intracranial haemorrhage in both unmatched and PSM groups.

OR: 1.44, 95% CI: 1.23 to 1.68, p=0.000) at 90 days (table 2, figure 2).

After PSM, 1113 patients with AIS treated with intravenous TNK were matched to an equivalent number of patients treated with intravenous rt-PA. Baseline characteristics between the two groups were well balanced except for anticoagulant use, which was not included in the matching variables (online supplemental table 3). In the PSM analysis, no differences were found in the rates of sICH (1.8% vs 1.98%, OR: 0.95, 95% CI: 0.51 to 1.76, p=0.864), PH2 (1.35% vs 1.44%, OR: 1.01, 95% CI: 0.49 to 2.08, p=0.979) any systemic bleeding (0.45% vs 0.18%, OR: 2.11, 95% CI: 0.4 to 11.12, p=0.379) or 90-day mortality (6.22% vs 7.45%, OR: 0.81, 95% CI: 0.571 to 1.15, p=0.231). Notably, patients treated with TNK exhibited significantly lower rates of any intracranial bleeding (2.07% vs 4.04%, OR: 0.51, 95% CI: 0.31 to 0.86, p=0.012) than those treated with rt-PA (table 3, figure 1). Furthermore, TNK-treated patients demonstrated enhanced early neurological improvement at 24 hours. This was evidenced by significantly lower 24-hour NIHSS scores (2 vs 3, p=0.000) and a higher proportion achieving major NIHSS improvement compared with the rt-PA group (57.49% vs 43.45%, OR: 1.76, 95% CI: 1.48 to 2.09, p=0.000). Additionally, TNK-treated patients exhibited better 90-day mRS by ordinal logistic regression analysis. This included overall favourable 90-day mRS (OR: 0.67, 95% CI: 0.57 to 0.79, p=0.000) and increased odds of achieving good (80.9% vs 74.76%, OR: 1.41, 95% CI: 1.14 to 1.75, p=0.001) and excellent functional outcomes (72.03% vs 66.73%, OR: 1.27, 95% CI: 1.05 to 1.54, p=0.012) (table 3, figure 2).

Sensitivity analysis yielded similar results for primary and secondary outcomes when adjusting for prespecified confounders (model 1 and model 2). However, in model 3, the significant reductions in any intracranial bleeding,

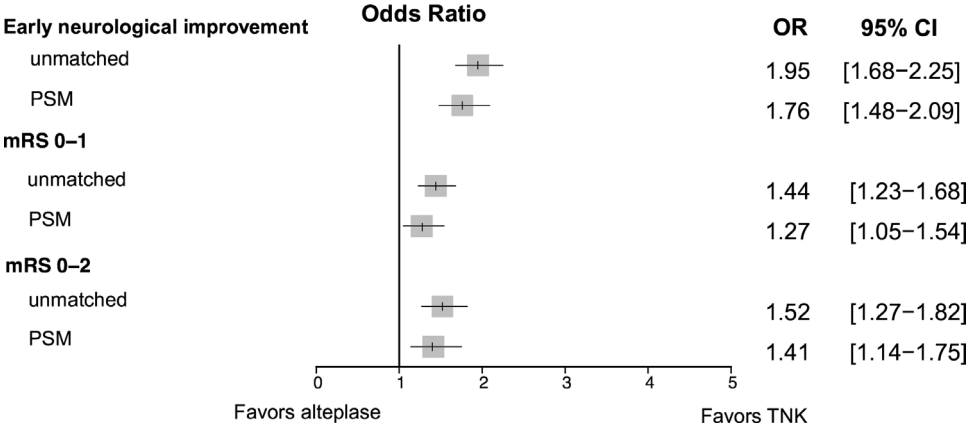

Figure 2 Efficacy profile of intravenous TNK compared with alteplase in the unmatched groups and after PSM. PSM groups were adjusted for oral anticoagulant use and DNT. The figure illustrates favourable tendency towards TNK group compared with the alteplase group, showing higher odds of better early neurological improvement and a higher percentage of achieving 90-day mRS scores 0–1 and 0–2. mRS, modified Rankin Scale; propensity score matching; TNK, tenecteplase.

Table 3 Outcomes of adjusted models in unmatched groups and PSM groups (sensitivity analysis)

	Unmatched groups			Model 1	Model 2	Model 3
Outcome	**TNK**	**Alteplase**	**P value**	**aOR (95% CI), *Adj. P* value**	**aOR (95% CI), *Adj. P* value**	**aOR (95% CI), *Adj. P* value**
Primary outcome						
sICH	20/1113 (1.8%)	55/2360 (2.33%)	0.313	0.98 (0.57–1.71), 0.964	0.96 (0.55, 1.66), 0.875	1.09 (0.61–1.95), 0.766
Secondary outcome						
Safety profile						
Any intracranial bleeding	23/1113 (2.07%)	109/2360 (4.62%)	0.000	0.49 (0.31– 0.79), 0.003	0.47 (0.29–0.75), 0.002	0.65 (0.40–1.06), 0.085
PH2	15/1113 (1.35%)	41/2360 (1.74%)	0.396	0.99 (0.52–1.86), 0.966	0.99 (0.52–1.86), 0.971	1.21 (0.64–2.28), 0.561
Any systemic bleeding	5/1113 (0.45%)	3/2360 (0.13%)	0.065	2.48 (0.51–12.07), 0.259	2.47 (0.52–11.86), 0.258	4.13 (0.85–20.09), 0.079
Death within 90 days	68/1094 (6.22%)	181/2208 (8.2%)	0.042	0.92 (0.66–1.29), 0.636	0.94 (0.67–1.31), 0.703	1.06 (0.74–1.52), 0.75
Efficacy						
24-hour NIHSS score improvement ≥4 OR ≤1	637/1108 (57.49%)	953/2324 (41.01%)	0.000	1.72 (1.48–2.01), 0.000	1.81 (1.54–2.11), 0.000	1.12 (0.93–1.35), 0.22
mRS at 90 days	0 (0-2) N=1094	1 (0-3) N=2208	0.000	0.64 (0.55, 0.74), 0.000	0.65 (0.56–0.76), 0.000	1.11 (0.94–1.32), 0.216
mRS=0–1 at 90 days	788 (72.03%)	1417 (64.18%)	0.000	1.31 (1.09–1.59), 0.005	1.30 (1.07–1.58), 0.008	0.82 (0.66–1.02), 0.073
mRS=0–2 at 90 days	885 (80.90%)	1625 (73.6%)	0.000	1.47 (1.19–1.81), 0.000	1.47 (1.18–1.82), 0.001	0.96 (0.76–1.21), 0.726

Model 1: Unmatched cohort adjusts for age, sex, smoking, pre-mRS, NIHSS, glucose at emergency, SBP at emergency, bridging with thrombectomy, diabetes, dyslipidemia, atrial fibrillation, heart failure, ischaemic stroke/TIA, taking antiplatelet drugs.
Model 2: Unmatched cohort adjusts for variables in model 1 combined with onset-to-treatment (OTT) time.
Model 3: Unmatched cohort excludes data from the Inner Mongolia centre, and adjusts for variables in model 1.
mRS, modified Rankin Scale; NIHSS, National Institutes of Health Stroke Scale; PH2, parenchymal haemorrhage type 2; PSM, propensity score matching; SBP, systolic blood pressure.

improvements in a shift analysis of mRS score at 90 days, as well as good and excellent functional outcomes were attenuated (table 3). Further analysis of baseline characteristics, comparing the Inner Mongolia centre with other participating centres, revealed a relatively balanced population within the rt-PA group. However, significant differences were found in the TNK group at the Inner Mongolia centre, characterised by a younger patient demographic with fewer comorbidities. These findings, together with the absence of thrombectomy procedures, may help in understanding the exceptional positive outcomes observed at the Inner Mongolia centre (online supplemental table 4).

No significant interactions were found between different subgroups in post hoc analyses (p interaction>0.05) regarding the probability of sICH in PSM-matched groups treated with intravenous TNK versus rt-PA. Subgroups were stratified by age, sex, stroke severity, thrombectomy performance and the centre level (online supplemental figure 1).

Mediation analysis showed no significant mediation of DNT on the rate of sICH (35% of the total effect size; β=−0.001, 95% CI: −0.002 to 0, p=0.64) (online supplemental table 5).

DISCUSSION

Our study found that thrombolysis with TNK is not associated with an increased risk of sICH, and may lead to better early neurological improvement and 90-day functional outcomes compared with alteplase in patients with AIS.

Our real-world study revealed that patients receiving TNK under off-label conditions tended to be younger, with fewer cerebrovascular risk factors and comorbidities, and generally experienced milder strokes. However, after matching, we observed similar characteristics and thrombectomy rates between the two groups. Moreover, despite a shorter DNT, the longer OTT for TNK recipients indicated they may present at a later thrombolytic time window compared with rt-PA recipients.

Our PSM analysis showed that stroke thrombolysis with TNK in off-label settings demonstrated a safe profile comparable to rt-PA, with a lower overall incidence of intracranial bleeding. These findings remained consistent when we stratified the data based on age, sex, baseline NIHSS score, thrombectomy performance and the level of the medical centre. The sICH rate observed in our study for TNK (1.8%) closely resembles the rates reported in the TRACE-II (2%)[7] and ORIGINAL (1.2%),[20] both of which included Chinese stroke populations with similar age ranges, baseline NIHSS scores and percentages of patients undergoing bridging thrombectomy. The elevated sICH rates observed in studies like Act (3.4% at 24 hours)[8] and ATTEST2 (3%)[9] could be due to factors such as higher NIHSS score and a higher percentage of patients undergoing thrombectomy. Nevertheless, these findings generally aligned with RCTs comparing TNK and rt-PA. A recent meta-analysis including 16 studies indicated that the risk of sICH was similar between TNK and rt-PA groups (RR: 0.89, 95% CI: 0.65 to 1.23). Notably, observational studies (RR: 0.56, 95% CI: 0.31 to 0.99) reported lower incidence rates compared with RCT (RR: 1.11, 95% CI: 0.75 to 1.64).[21] The higher rate of sICH in RCTs was possibly influenced by the inclusion of NOR-TEST, which used a dose of 0.4 mg/kg.[22 23] The real-world study CERTAIN[14] and another PSM study from SITS-ISTR,[15] which recruited patients with more severe strokes and higher NIHSS scores, reported similar sICH rates of 1.8% and 1%, respectively. Notably, CERTAIN reported a high bridging percentage of 38%, yet the sICH remained low. A subanalysis within CERTAIN further demonstrated that regardless of the total, non-thrombectomy, or thrombectomy groups, the TNK group consistently showed lower sICH rates than the alteplase group. These along with other off-label studies,[24–29] provide reassurance regarding the safety of the 0.25 mg/kg dose of TNK in patients with AIS .

Our study revealed that the TNK-treated patients had lower NIHSS scores and greater odds of achieving a significant improvement in NIHSS score at 24 hours compared with patients treated with rt-PA. These distinctions persisted even after conducting a sensitivity analysis, but not when data from the Inner Mongolia centre were excluded. Early neurological improvements had been linked to favourable functional outcomes in previous studies,[18 30] and a similar improving trend had also been reported in the meta-analyses of both non-RCT[31] and RCT studies.[32]

Furthermore, at the 90-day mark, a higher proportion of TNK-treated patients achieved either excellent (72.03% vs 66.73%, OR: 1.27, 95% CI: 1.05 to 1.54) or good functional outcomes (80.9% vs 74.76%, OR: 1.41, 95% CI: 1.14 to 1.75) compared with those treated with rt-PA. The generally favourable functional outcomes in both groups may be attributed to the comparatively milder strokes observed (NIHSS median=5), with percentages exceeding those reported in the majority of studies.[31 32] Although RCT studies involving all patients with AIS did not demonstrate this superiority,[7 8 22] various real-world studies have indicated better functional outcomes in the TNK group compared with the rt-PA group[14 15] as well as meta-analysis of non-randomised studies.[31] Despite the consistent favourable functional outcomes observed in the sensitivity analysis, the exclusion of data from the Inner Mongolia centre led to the disappearance of this favourable trend suggesting potential bias of patient selection in certain centres. This observation can be attributed to several factors. First, patients from the Inner Mongolia centre are generally younger and have a significantly lower percentage of smoking and comorbidities, contributing to a milder stroke profile, as indicated by a lower IQR of stroke severity. These baseline differences may lead to better neurological outcomes both early and at 90 days. Additionally, the Inner Mongolia centre lacks the capability to perform thrombectomy, potentially resulting

in the admission of patients who had less severe stroke from the local emergency system. The milder patient population and the absence of thrombectomy capability could contribute to the observed better outcomes with TNK in this centre, potentially skewing the overall study results. Future studies should consider stratifying results by centre or controlling for center-specific characteristics to better understand the generalisability of the findings.

Strengths and limitations

This is the largest real-world study to report safety and efficacy in TNK thrombolysis compared with rt-PA with a low rate of loss to follow-up (TNK: 1.7%, rt-PA: 6.4%) in China. Furthermore, it serves as a valuable supplement to TRACE-II for the drug manufactured in China. Moreover, the inclusion of a broad spectrum of stroke populations enhances its applicability to clinical practice and external generalisability compared with RCT studies. While the retrospective nature and the lack of detailed stroke subtype data are limitations, the diverse patient population provides valuable insights that are more reflective of real-world clinical scenarios.

Nonetheless, it is important to acknowledge several limitations in the study. First, its non-randomised and retrospective design may introduce biases related to patient and centre selection. Clinicians might have favoured 'safer' patients for TNK treatment, mainly concerning its off-label use and the bleeding risk. On the other hand, tertiary A centres contribute a relatively small proportion of TNK data (15%) due to tighter regulation of off-label drug use while 40% of TNK data are from a single centre in Inner Mongolia due to a shortage of rt-PA supply in the area. Sensitivity analyses are conducted to exclude data from this centre, and univariable analyses between the Inner Mongolia centre and other participating centres of TNK and rt-PA groups are performed to assess center-related bias. Second, although efforts are made to minimise biases through population matching and sensitivity analyses, confounding factors at different centre levels may influence functional outcomes. However, these factors are less likely to affect the rate of sICH, given that 80% of such events occur within 24 hours after thrombolysis.[33] Center-level is not included as a variable in our PSM due to a potential loss of 30% of TNK data, which could introduce significant bias. Nevertheless, post hoc analyses are performed within centre classifications to assess safety outcomes at different levels. Third, efficacy outcomes, such as NIHSS at 24 hours and mRS at 90 days, are collected and reported by local investigators unblinded to the treatment, potentially introducing variations. However, the primary outcome, imaging of sICH and corresponding history are centrally reviewed and confirmed, resulting in greater objectivity. Lastly, the absence of imaging data prevents further analysis of specific subgroups, such as those with LVO or the population achieving early recanalisation. Future studies involving a broader range of stroke types including more of those undergoing thrombectomy, are necessary for a more comprehensive understanding.

CONCLUSION

Our TTT-AIS study has provided compelling evidence that the administration of TNK thrombolysis within 4.5 hours of symptoms onset in patients with AIS is safe. Also, it may be associated with potential improvements in functional outcomes and early neurological improvement. These findings support the clinical safety of using TNK thrombolysis in routine ischaemic stroke management.

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