Minocycline in Acute Cerebral Hemorrhage
An Early Phase Randomized Trial

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Background and Purpose—Minocycline is under investigation as a neurovascular protective agent for stroke. This study evaluated the pharmacokinetic, anti-inflammatory, and safety profile of minocycline after intracerebral hemorrhage.

Methods—This study was a single-site, randomized controlled trial of minocycline conducted from 2013 to 2016. Adults ≥18 years with primary intracerebral hemorrhage who could have study drug administered within 24 hours of onset were included. Patients received 400 mg of intravenous minocycline, followed by 400 mg minocycline oral daily for 4 days. Serum concentrations of minocycline after the last oral dose and biomarkers were sampled to determine the peak concentration, half-life, and anti-inflammatory profile.

Results—A total of 16 consecutive eligible patients were enrolled, with 8 randomized to minocycline. Although the literature supports a time to peak concentration (T_{max}) of 1 hour for oral minocycline, the T_{max} was estimated to be at least 6 hours in this cohort. The elimination half-life (available on 7 patients) was 17.5 hours (SD±3.5). No differences were observed in inflammatory biomarkers, hematoma volume, or perihematomal edema. Concentrations remained at neuroprotective levels (>3 mg/L) throughout the dosing interval in 5 of 7 patients.

Conclusions—In intracerebral hemorrhage, a 400 mg dose of minocycline was safe and achieved neuroprotective serum concentrations. However, oral administration led to delayed absorption in these critically ill patients and should not be used when rapid, high concentrations are desired. Given the safety and pharmacokinetic profile of minocycline in intracerebral hemorrhage and promising data in the treatment of ischemic stroke, intravenous minocycline is an excellent candidate for a prehospital treatment trial.

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Intracerebral hemorrhage (ICH) is the most severe form of stroke, and despite the high rates of morbidity and mortality, no effective therapies exist.[1] After ICH, a secondary inflammatory response is triggered by blood constituents and their degradation products.[2] Key mediators to this poststroke inflammatory response include matrix metalloproteinases (MMPs), iron, and activated microglia and macrophages. MMP inhibition, iron sequestration, and inflammation suppression have been associated with reduced cell death and improved behavioral outcomes in preclinical models of ICH.[3,4]

Minocycline is a broad-spectrum tetracycline antibiotic with a well-documented safety profile previously studied for neuroprotective activity in acute ischemic stroke.[5,6] Minocycline is a potent MMP inhibitor and iron chelator demonstrating unique anti-inflammatory properties.[7,8] Lampl et al[6] observed that a 5-day course of oral minocycline significantly reduced neurological impairment in acute ischemic stroke.

The MACH trial (Minocycline in Acute Cerebral Hemorrhage) evaluated the pharmacokinetic, anti-inflammatory, and safety profile of minocycline in ICH.

Materials and Methods

Trial Design

This study was a single-site, randomized controlled trial of minocycline conducted from February 2013 to January 2016. The Augusta University Institutional Review Board approved the study protocol. Adult patients with computerized tomography-documented primary ICH who could have study drug administered within 24 hours of symptom onset were eligible. Patients were excluded for known allergy to tetracycline antibiotics, pregnancy, liver function tests >3× the upper limit of normal,

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serum creatinine >2 mg/dL, National Institutes of Health Stroke Scale score of ≤4, Glasgow Coma Scale score of ≤5, planned surgical evacuation of hematoma with 24 hours, secondary ICH, platelet count <75 000/mm^3, international normalized ratio >1.4, prestroke modified Rankin Scale (mRS) score of >2, or preexisting do not resuscitate orders.

Drug Administration

Patients received an initial intravenous administration of open-label minocycline 400 mg followed by 400 mg oral (per oral) daily for 4 days. A fixed dose of minocycline was chosen based on its wide safety range and to facilitate accurate admixing making it a potentially feasible drug in rural hospital and prehospital settings. The 400 mg dose is within the minocycline safe dose range shown to inhibit MMP-9 activity in the MINOS trial.[5,7]

Outcome Assessments

All patients underwent a noncontrast head computerized tomography for confirmation of ICH. Follow-up computerized tomography scans were performed at 24 hours and 7 days (or discharge). Hematoma location and volume, perihematomal edema (PHE), and the presence of intraventricular extension were assessed. Research-related clinical examinations were conducted at screening, baseline, 24 hours, 7 days, and 90 days. Severity of neurological deficit was determined using National Institutes of Health Stroke Scale and Glasgow Coma Scale. Functional outcome was assessed using the mRS at 90 days. The Hemphill ICH score was determined based on Glasgow Coma Scale, ICH volume, ICH location, presence of intraventricular hemorrhage, and age.[9] Minocycline-associated adverse events were assessed for 90 days.

Blood Sampling

Plasma samples for biomarker evaluation were obtained at baseline (pre-minocycline), and at 1 and 24 hours, days 3, 7 (or discharge), and 90. These samples were analyzed for MMP-9 activity, iron, and interleukin-6. Additional serum samples were collected 1 hour before the final per oral dose (day 5), and at 1, 6, 12, 48, and 72 hours after the same dose to calculate minocycline approximate peak concentration (C_{peak}) and half-life ($t_{1/2}$) using targeted liquid chromatography tandem-mass spectrometry.

Evaluation of hemorrhage growth and PHE, plasma biomarkers, and 90-day mRS was blinded to treatment assignment.

Statistical Analysis

All statistical analysis, except for the pharmacokinetic analysis, was performed using SAS 9.4. Statistical significance was assessed using an α level of 0.05. χ^2 or *t* tests were used to examine differences in demographic characteristics. A 2-sample *t* test was used to examine differences in mRS between groups. To examine differences between minocycline over time for MMP-9, interleukin-6, iron, ferritin, total iron binding capacity, ICH volume, and PHE-repeated measures, mixed models were used. The statistical test of interest was the interaction between minocycline and day. Post hoc pairwise comparisons were examined using a Bonferroni correction to the overall α level.

Results

Sixteen consecutive subjects were enrolled, with 8 randomized to minocycline treatment and pharmacokinetic sampling. Demographics and baseline characteristics of the population are shown in Table I in the online-only Data Supplement. At baseline, mean ICH volume was 20.1 mL and ICH score was 1.3. The cause of the hemorrhages was hypertensive in 12 and possible or probable cerebral amyloid angiopathy in 4 patients. There were no statistically significant differences between those given minocycline and controls for any baseline variable.

Adverse Events

Minocycline was well tolerated. One subject in the minocycline arm suffered bihemispheric embolic strokes at 1 week resulting in herniation and death that was deemed unrelated to the study medication.

Biomarkers and Clinical Outcomes

There was no statistically significant difference in mRS at 90 days between groups. No statistically significant difference between minocycline and controls was detected for MMP-9, interleukin-6, iron, ferritin, total iron binding capacity, ICH volume, or PHE (Table II in the online-only Data Supplement).

Pharmacokinetic Analysis

Pharmacokinetic analyses were available for 8 patients (Figure). The mean concentration at 1 hour after the final oral dose was 6.20±2.81 mg/L. T_{max} was estimated to be at least 6 hours in 4 patients. Concentrations remained >3 mg/L throughout the dosing interval in 5 patients, and 7 patients achieved this concentration at 1 hour. A mean half-life of 17.5 hours (SD±3.5) was observed based on complete data from 7 patients. On the basis of the weight of patients in the MACH trial, the dose was ≈4 mg/kg.

Discussion

In a randomized clinical trial of minocycline in ICH, a fixed dose of 400 mg IV followed by 400 mg daily oral for 4 days was safe with sufficient serum concentrations and a half-life conducive to once daily dosing. Our trial is the first to investigate minocycline in ICH and determine its pharmacokinetics in this critically ill population.

Minocycline was eliminated with a half-life of 17.5±3.5 hours. This half-life is consistent with a previously conducted dose escalation trial (MINOS) of intravenous minocycline in ischemic stroke.[5] In healthy individuals, minocycline achieves rapid, complete absorption after oral administration, peaking at 1 hour.[10] Doses in MACH were large enough to achieve peak concentrations of 3 mg/L, which have been shown to be neuroprotective.[11] In addition, a majority of patients maintained concentrations >3 mg/L throughout the dosing interval.

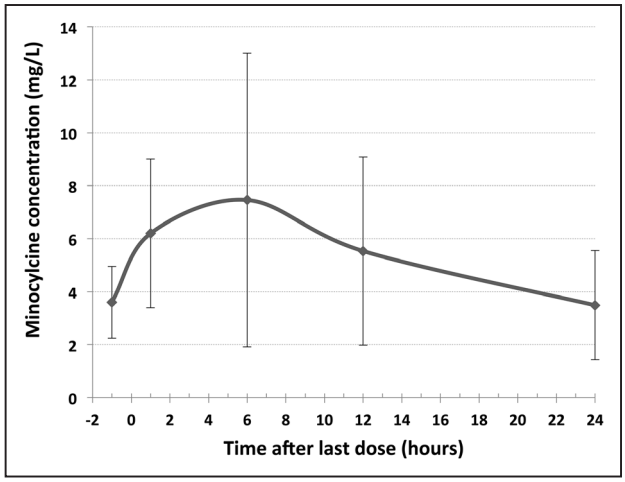

Figure. Minocycline concentrations after the last oral dose.

We hypothesized that oral minocycline would achieve C_{max} in the plasma of ICH patients who were within 20% of those previously achieved with similar intravenous doses in ischemic stroke. In MINOS, the concentration at 1 hour after intravenous administration was 14.32 mg/L, compared with 6.2±2.81 mg/L in the MACH trial, indicating delayed oral absorption in this critically ill population. The wide range of C_{max} indicates erratic absorption, thus making oral administration of minocycline an unreliable dosing route in critical illness. Therefore, intravenous minocycline would be the preferable route of administration for future acute stroke studies.

We hypothesized that minocycline would reduce plasma MMP-9 and interleukin-6 and PHE in ICH; however, we were unable to detect differences between the groups. These results are in contrast to our findings in ischemic stroke and in preclinical studies of minocycline in ICH. Our inability to find a significant difference may reflect the limitations of our trial including its small sample size, a prolonged (24 hour) window for enrollment, and the heterogeneity of our population in terms of ICH volume, location, and cause, in addition to the apparent erratic absorption after oral minocycline dosing. Nevertheless, minocycline seemed to be safe in ICH patients, and, therefore, an excellent candidate for a prehospital stroke trial with administration via the intravenous route.

Conclusions

Given the apparent safety and pharmacokinetics of a fixed dose of minocycline in ICH and promising data in the treatment of ischemic stroke, minocycline is an excellent candidate for a prehospital treatment trial (before neuroimaging and the differentiation of ischemic stroke from ICH).[12]

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Disclosures

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