

Pharmacokinetics, safety, and immunogenicity of HLX03, an adalimumab biosimilar, compared with reference biologic in healthy Chinese male volunteers: Results of a randomized, double-blind, parallel-controlled, phase 1 study

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Abstract

The primary objective of this randomized, double-blind, parallel-controlled study (from December 2016 to October 2018) was to evaluate pharmacokinetic (PK) equivalence of adalimumab biosimilar HLX03 and reference adalimumab in healthy volunteers, and to assess safety, and immunogenicity of HLX03. The primary PK endpoints were maximum observed plasma concentration (C_{max}) and area under the concentration curve from time zero to the last quantifiable concentration (AUC_{0-t}). Equivalence was determined if the 90% confidence interval (CI) of geometric least square mean ratio between the two treatment groups were within the predefined range of 80%–125%. Safety and immunogenicity were monitored during the study. Healthy Chinese males ($N = 220$) were randomized 1:1 to receive a single subcutaneous 40 mg dose of HLX03 or China (CN)-sourced adalimumab. The ratios of the geometric mean of C_{max} and AUC_{0-t} were 102.2% and 105.7%, respectively, with corresponding 90% CIs falling in the predefined margins, which demonstrated PK equivalence between HLX03 and CN-adalimumab. The incidence of treatment-emergent adverse events (TEAEs) was similar in the two groups (73.8% and 66.0% in the HLX03 and CN-adalimumab groups, respectively). Grade 3–4 TEAEs were reported in 7.5% and 5.7% of participants, respectively. The incidences of participants with antidrug antibodies (HLX03: 96.2%; CN-adalimumab: 93.4%) or neutralizing antibodies (HLX03: 40.6%, CN-adalimumab: 41.4%) were comparable between groups. This study demonstrated PK bioequivalence between HLX03 and CN-adalimumab, with similar safety and immunogenicity

Abbreviations: %AUC_{ex}, area under the concentration curve extrapolated from time 0 to infinity; ADAs, antidrug antibodies; AEs, adverse events; ALT, alanine aminotransferase; AS, ankylosing spondylitis; AST, aspartate aminotransferase; AUC_{0-inf}, area under the concentration curve from time zero to infinity; AUC_{0-t}, area under the concentration curve from time zero to the last quantifiable concentration; BMI, body mass index; CD, Crohn's disease; CI, confidence interval; CL, clearance; C_{max} , maximum observed plasma concentration; CN, China; CV, co-efficient of variation; ECG, electrocardiography; ELISA, enzyme-linked immunosorbent assay; FAS, full analysis set; HBV, hepatitis B virus; LS, least square; NABs, neutralizing antibodies; PK, pharmacokinetic; PPS, per-protocol set; PT, preferred term; QC, quality control; RA, rheumatoid arthritis; SOC, system organ class; $t_{1/2}$, elimination half-life; TEAEs, treatment-emergent adverse events; Tmax, time to Cmax; TNF α , Tumor necrosis factor alpha; Vd/F, apparent volume of distribution; λ_z , terminal elimination rate constant.

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profiles. These data support further clinical development of HLX03 as an adalimumab biosimilar.

KEYWORDS

adalimumab, bioequivalence, biosimilar, pharmacokinetics

1 | INTRODUCTION

Tumor necrosis factor- α (TNF α), is a pleiotropic cytokine with important functions in homeostasis and disease pathogenesis. Although initially discovered as an anticancer agent, TNF α has been demonstrated to be a major mediator of inflammation and plays a pivotal role in autoimmune diseases.¹ Biologic agents targeting TNF α are available as treatment options for patients with inflammatory diseases.^{2,3}

Adalimumab was the first fully humanized anti-TNF α monoclonal antibody approved by the United States Food and Drug Administration in 2002.⁴ It is currently indicated for the treatment of nine indications, including rheumatoid arthritis (RA), ankylosing spondylitis (AS), plaque psoriasis, Crohn's disease (CD), in the United States,⁵ and five indications in Europe,⁶ and has been approved in China for the treatment of RA, AS, plaque psoriasis, and CD since August 2010.^{7,8} These chronic inflammatory conditions are common in China, with a prevalence of 0.47% for plaque psoriasis, 0.2%–0.93% for RA, and 0.2%–0.54% for ankylosing spondylitis^{9–11}; however, the high cost of biologics limits access to treatment for many eligible patients.¹²

Biosimilars, biologic medicines with no clinically meaningful differences in safety or efficacy versus approved reference products, can potentially reduce drug costs and increase patient access to treatment.¹² Similarity to the reference product in terms of quality, biologic activity, structure/physicochemical, and functional attributes, efficacy, immunogenicity, and safety must be established before the biosimilar product can be authorized in the United States, Europe, or other regulated countries, including China.^{7,13–16}

HLX03 has been approved by China National Medical Products Administration at December 2nd 2020 as a biosimilar to adalimumab.¹⁷ The active ingredient, recombinant anti-TNF α fully human monoclonal antibody, was obtained by designing the gene sequence based on the protein sequence of adalimumab and overexpressed in Chinese hamster ovary cells system through transfection of an expression vector.¹⁸ In an extensive analytical comparison, HLX03 demonstrated structural and functional similarity to the China (CN)-sourced adalimumab product.¹⁹ In vitro, the effective concentration of HLX03 was equivalent to that of adalimumab, with a similar in vitro neutralizing activity. In vivo, HLX03 and adalimumab showed equivalent efficacy in inhibiting the pathology of arthritis in mice: HLX03 and adalimumab (10 mg/kg) both inhibited histopathologic lesions, reduced the severity of the disease, and restored the state of potential early arthritis before treatment (data on file). In addition, a comparative, repeat-dose toxicity study in cynomolgus monkeys

found no adverse effects after repeated subcutaneous injections of HLX03 or adalimumab (data on file).

Here, we present the results of a phase 1 trial that compared the pharmacokinetics (PK), safety, and immunogenicity of HLX03 with the reference CN-adalimumab in healthy Chinese male volunteers.

2 | MATERIALS AND METHODS

2.1 | Study design

A randomized, double-blind, single-dose, two-arm, parallel-controlled clinical trial with a total post-dose follow-up time of 70 days was conducted at a single research center (First Hospital of Jilin University, Jilin, China) between December 2016 and October 2018. All participants gave written informed consent prior to participation.

The primary objective of this trial was to demonstrate PK equivalence between HLX03 and reference CN-adalimumab in healthy Chinese male volunteers. Secondary objectives included comparing the safety, tolerability, and immunogenicity of HLX03 and CN-adalimumab.

Eligible participants were randomized in a ratio of 1:1 to receive a single 40 mg (0.8 ml) dose of HLX03 (Shanghai Henlius Biotech, Inc., Shanghai, China; Batch number: M20161003) or CN-adalimumab (AbbVie Ltd., Pointe-Claire, Quebec, Canada; Batch number: 63169XH02 and 73247XH01) via subcutaneous injection into the lower abdomen. Block randomization was used to assign participants to treatment groups, with four subjects in each block, for a total of 115 blocks. Study participants and investigators were blinded to treatment allocation. Study drugs were prepared and administered by unblinded personnel, and PK, antidrug antibodies (ADAs), and neutralizing antibodies (NABs) were all determined by a third-party laboratory under blinded conditions.

2.2 | Study participants

Healthy adult men, aged 18–45 years, with a body weight of 55–80 kg, and a body mass index (BMI) of 19–28 kg/m² were eligible for inclusion. Potential participants were deemed healthy following a review of medical history and an examination that included a general physical examination, electrocardiography (ECG), and routine laboratory tests. Other key inclusion criteria were: no history of allergies to multiple drugs and food; no history of heart, liver, kidney, digestive tract, nervous system,

or metabolic abnormalities; no receipt of other drugs within two weeks of the trial start date; no smoking or smoking fewer than five cigarettes per day within three months before screening; and either no alcohol consumption or a weekly alcohol consumption of fewer than 14 units within six months before screening.

Individuals with a history and/or current presence of tuberculosis, heart disease, mental disease, malignancy, herpes zoster, or epilepsy were excluded from the trial. Individuals with a positive hepatitis C antibody test, negative hepatitis B virus (HBV) surface antigen test, positive core antibody test, and peripheral blood HBV deoxyribonucleic acid (DNA) levels >0 IU/ml were also excluded, along with those who tested positive for human immunodeficiency virus (HIV) or *treponema pallidum* at screening. Other key exclusion criteria were: known or suspected clinically relevant drug hypersensitivity; history of systemic or local infections; invasive systemic fungal infections or other opportunistic infections; serious infection (defined as an infection that required hospitalization and/or intravenous injection of antibiotics) within 2 months before screening; and previous treatment with adalimumab, its analogs, or drugs that target TNF α . Additional details of participants' eligibility criteria are provided in supplementary document.

2.3 | PK assessments and endpoints

The primary endpoints were maximum observed plasma concentration (C_{\max}) and area under the concentration curve from time zero to the last quantifiable concentration (AUC_{0-t}). Secondary endpoints were AUC from time zero to infinity (AUC_{0-inf}), time to C_{\max} (T_{\max}), elimination half-life ($t_{1/2}$), terminal elimination rate constant (λ_z), apparent volume of distribution (V_d/F), clearance (CL), and percentage of AUC extrapolated from time 0 to infinity (% AUC_{ex}).

Blood samples for PK evaluation were obtained pre-dose (within 30 minutes before dosing) and at 4, 8, 12, and 24 h post-dose. Additional samples were obtained every 24 h thereafter until day 9 and at 264 h (day 11), 336 h (day 14), then every 7 days thereafter until day 56. The last collection time point was at 1680 h (day 70). Serum samples were extracted and stored at -70°C to -90°C within 2 h after collection.

Serum concentrations of HLX03 and CN-adalimumab were determined by a central laboratory (United-Power Pharma Tech Co, Ltd) using a validated enzyme-linked immunosorbent assay (ELISA). The lower limit of quantification was 15.625 ng/ml and the linear range was 15.625 to 800.000 ng/ml. Inter-assay precision and accuracy were calculated from quality control (QC) in six validation runs (upper limit of quantification), high QC, medium QC, lower QC, lower limit of quantification.

2.4 | Safety and tolerability assessments

Safety assessments included adverse events (AEs), serious AEs, vital signs, routine laboratory (blood and urine) tests, 12-lead ECG, and

physical examination. AEs were observed throughout the study (from prior dose to last visit at 70 days after dosing). All AEs were coded according to the Medical Dictionary for Regulatory Activities version 20.0, and the severity of AEs was graded in accordance with the National Cancer Institute Common Terminology Criteria for Adverse Events, version 4.03. Vital signs were examined simultaneously with blood sample collection. Routine laboratory tests, 12-lead ECG, and physical examination were performed pre-dose, at 48 h (day 2), day 9, 35 and 70 after dosing.

2.5 | Immunogenicity testing

Immunogenicity of the administered drug was evaluated based on the incidence of ADAs and NABs (by a central laboratory [United-Power Pharma Tech Co., Ltd.]). Blood samples for immunogenicity testing were collected on days 7, 14, 28, 42, and 70 after dosing; participants with ADAs detected in the screening period were excluded from sampling. ADAs were measured using a bridging electrochemiluminescence assay with a sensitivity (lower limit of quantification) of 5.2 ng/ml. HLX03 and CN-adalimumab NABs were measured using a validated method based on an L-929 cell proliferation endpoint, with a sensitivity (lower limit of quantification) of 187.5 ng/ml.

2.6 | Statistical analysis

Based on previous bioavailability studies with adalimumab, an initial sample size of 148 participants was estimated to achieve 90% power to show bioequivalence of the primary endpoint (C_{\max} and AUC_{0-t}), assuming a co-efficient of variation (CV) for C_{\max} of 33%, a ratio for the primary endpoint of 95% between groups, the significance level α of 0.05 (two one-sided) and a 20% dropout rate. The planned number of participants was increased to 176 following a slightly higher than expected intersubject CV of 39.4% for AUC_{0-t} in a blinded prespecified interim analysis (α was not adjusted) of the PK data from 98 participants. Using a predetermined acceptance range for the PK parameters (the 90% confidence interval [CI] for the log-transformed values within the range of 80% to 125%), a CV of 40%, and a dropout rate of 20%, the re-estimated sample size was 220 participants (110 in each group).

PK parameters were derived using the non-compartmental model of WinNonlin software version 7.0 (Certara LP). The primary PK endpoints, AUC_{0-t} and C_{\max} , were log-transformed for analysis, and the 90% CI for the difference in mean parameters between the two groups was calculated and re-expressed on the original ratio scale to assess equivalence. PK equivalence between HLX03 and CN-adalimumab was declared if the 90% CI for the ratio of the geometric mean was within the prespecified equivalence range of 80% to 125%. Bioequivalence was demonstrated if the two PK parameters met the PK equivalence criteria. Analysis of AUC_{0-inf} , one of the secondary PK endpoints, was the same as that of C_{\max} and AUC_{0-t} (the ratio of geometric mean and its 90% CI were

calculated). The Wilcoxon rank-sum test was used to compare T_{\max} between the two groups. Other PK parameters, serum drug concentrations, and safety data were summarized using descriptive statistics.

The PK full analysis set (FAS) included all randomized participants who received the study drug and had at least one PK parameter (C_{\max} and AUC_{0-t}) assessment. The PK per-protocol set (PK-PPS) included all PK-FAS participants who had no serious protocol violations that affected PK. The safety set comprised all randomized participants who received the study drug, whereas the immunogenicity analysis set included all randomized participants who received the study drug and underwent immunogenicity evaluation.

All statistical analyses were performed using 9.2 or higher version of SAS software (SAS Institute).

2.7 | Nomenclature of targets and ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in <http://www.guidetopharmacology.org>, the common portal for data from the IUPHAR/BPS Guide to PHARMACOLOGY,²⁰ and are permanently archived in the Concise Guide to PHARMACOLOGY 2019/20.²¹

3 | RESULTS

3.1 | Participants disposition

A total of 1057 participants were screened between December 29, 2016 and October 09, 2018, and 220 were enrolled into the trial (the higher screen failure rate is associated with reasons: vital sign abnormality, outrange body weight, T-SPOT positivity, and so on). Among those enrolled, seven withdrew before dosing and 213 received one dose of the study drug (HLX03, $n = 107$; CN-adalimumab, $n = 106$) and comprised the safety set (Figure 1). After dosing, there were two withdrawals in the HLX03 group who were lost to follow-up for personal reasons. Altogether, 211 of 220 (95.9%) participants completed the study (HLX03, $n = 105$; CN-adalimumab, $n = 106$) and comprised the PK-FAS and immunogenicity set (Figure 1). As one

participant in the HLX03 group had missing blood sample collections, 210 participants were included in the PK-PPS (Figure 1).

Most participants were of Han ethnicity (97.2% in HLX03 group and 95.3% in CN-adalimumab group). Baseline demographics (e.g., age, weight, BMI) and clinical characteristics were well balanced between the two treatment groups (Table 1).

3.2 | PK results

In the PK-PPS analysis, the mean plasma concentration–time profiles (linear and semi-log) following a single subcutaneous injection of HLX03 or CN-adalimumab were similar (Figure 2). T_{\max} was observed after a median of 144 h in both groups. Unadjusted mean C_{\max} levels were 3.4 and 3.3 $\mu\text{g}/\text{ml}$ for HLX03 and CN-adalimumab, respectively; unadjusted mean AUC_{0-t} levels were 1938.7 and 1847.9 $\mu\text{g h}/\text{ml}$, respectively (Table 2). Levels of AUC_{0-inf} , $\%AUC_{ex}$, λ_z , $t_{1/2}$, V_d/F , and CL in HLX03 group were comparable with those in the CN-adalimumab group (Table 2). PK parameters for the PK-FAS population are shown in Table S1.

Based on the PK-PPS analysis, HLX03 demonstrated PK equivalence to CN-adalimumab for both primary PK endpoints (C_{\max} and AUC_{0-t}). The geometric mean ratios for C_{\max} and AUC_{0-t} after log transformation were 102.2% and 105.7%, respectively. And the 90%

TABLE 1 Baseline demographics and clinical characteristics (safety analysis set)

Baseline characteristics	HLX03 $n = 107$	CN-adalimumab $n = 106$
Age, mean (SD), years	33.0 (6.5)	31.9 (6.7)
Ethnicity, n (%)		
Han	104 (97.2)	101 (95.3)
Other	3 (2.8)	5 (4.7)
Height, mean (SD), cm	167.9 (5.8)	169.3 (5.5)
Weight, mean (SD), kg	66.1 (6.7)	66.5 (6.4)
BMI, mean (SD), kg/m^2	23.5 (2.1)	23.3 (2.5)

Abbreviations: BMI, body mass index; CN-adalimumab, China-sourced adalimumab; SD, standard deviation.

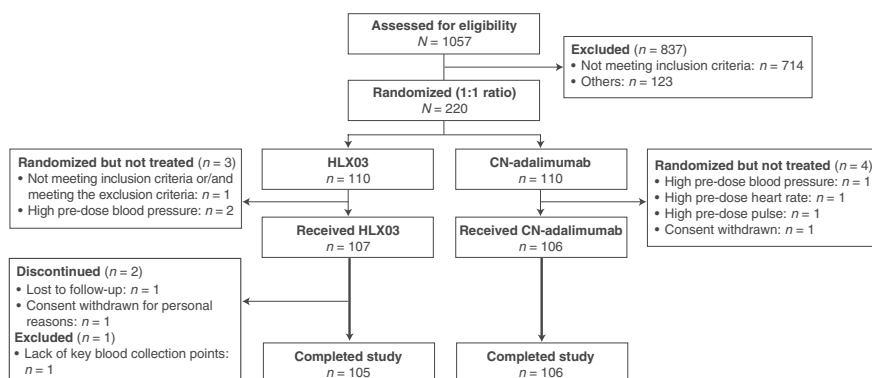


FIGURE 1 Participants disposition summary. CN-adalimumab, China-sourced adalimumab; PK-FAS, pharmacokinetic full analysis set; PK-PPS, pharmacokinetic per protocol set

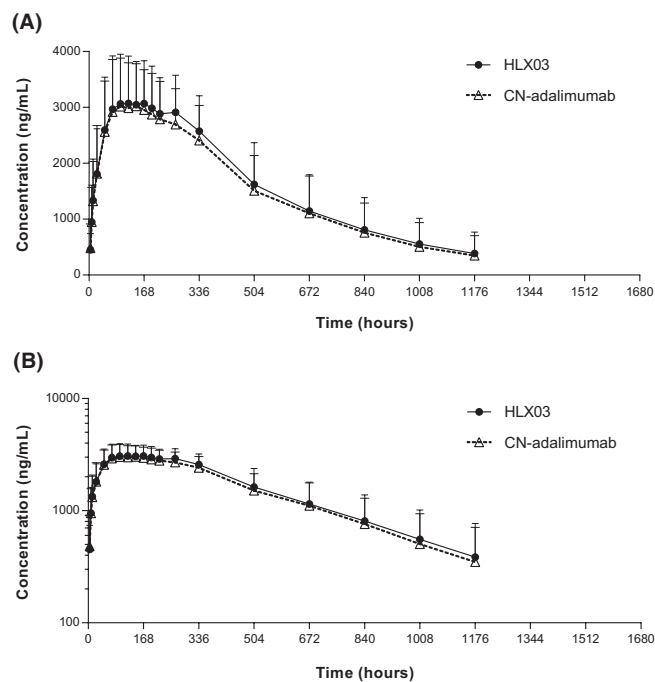


FIGURE 2 Mean plasma concentration–time curves (A) linear scale (B) semi-logarithmic scale following a single subcutaneous injection of 40 mg HLX03 and CN-adalimumab (PK-PPS). CN-adalimumab, China-sourced adalimumab; PK-PPS, pharmacokinetic per protocol set

TABLE 2 Summary of PK parameters (PK-PPS)

Parameter (units)	HLX03 <i>n</i> = 104 ^a	CN-adalimumab <i>n</i> = 106
T_{max} (h), median (min–max)	144.0 (8.0–504.5)	144.0 (24.0–264.8)
C_{max} (µg/ml), mean (CV%)	3.4 (25.8)	3.3 (25.3)
AUC_{0-t} (µg h/ml), mean (CV%)	1938.7 (34.9)	1847.9 (36.8)
AUC_{0-inf} (µg h/ml), mean (CV%)	2017.6 (37.8)	1936.5 (40.9)
% AUC_{ex} (%), mean (CV%)	2.9 (135.3)	3.3 (134.7)
λ_z (1/h), mean (CV%)	0.0046 (64.0)	0.0048 (66.6)
$t_{1/2}$ (h), mean (CV%)	226.8 (66.6)	231.9 (71.2)
V_d/F (L), mean (CV%)	6.2 (44.9)	6.7 (51.7)
CL (L/h), mean (CV%)	0.0229 (39.7)	0.0243 (42.2)

Abbreviations: AUC_{0-inf} , area under the concentration curve from time zero to infinity; AUC_{0-t} , area under the concentration curve from time zero to the last quantifiable concentration; % AUC_{ex} , percentage of area under the concentration curve extrapolated from time 0 to infinity; CL, clearance; C_{max} , maximum observed concentration; CN-adalimumab, China-sourced adalimumab; CV, co-efficient of variation; λ_z , elimination rate constant; PK, pharmacokinetic; PPS, per-protocol set; T_{max} , time to maximum observed concentration; $t_{1/2}$, elimination half-life; V_d/F , apparent volume of distribution.

^aData on AUC , λ_z , $t_{1/2}$, V_d/F , and CL parameters were excluded for five participants whose % AUC_{ex} exceeded 20%.

CI_s were within the prespecified equivalence range of 80% to 125% (Table 3).

PK equivalence was also observed between HLX03 and CN-adalimumab for the secondary PK endpoints AUC_{0-inf} and T_{max} ,

based on PK-PPS analyses. The geometric mean ratio of AUC_{0-inf} was 105.3% with the 90% CI of 96.1% to 115.4%, which fell in the 80%–125% range (Table 3). Furthermore, Wilcoxon rank-sum test showed no statistically significant difference in T_{max} between the two treatment groups.

The results of a sensitivity analysis based on the FAS were consistent with those from the PK-PPS analysis, confirming the similarity between HLX03 and CN-adalimumab for both primary and secondary PK endpoints (Table S2).

3.3 | Safety

Treatment-emergent AEs (TEAEs) were reported in 79 (73.8%) participants in the HLX03 group and 70 (66.0%) in the CN-adalimumab group. The most frequently reported TEAEs were hypertriglyceridemia, increased alanine aminotransferase (ALT), increased aspartate aminotransferase (AST), rhinorrhea, cough, and oropharyngeal pain. Most TEAEs were considered mild to moderate in severity, with grade 1 (mild) or 2 (moderate) TEAEs reported in 71 (66.4%) and 64 (60.4%) participants in the HLX03 and CN-adalimumab groups, respectively. Grade 3 (severe) TEAEs were reported in eight (7.5%) participants in the HLX03 group and five (4.7%) in the CN-adalimumab group. The most frequent grade 3 TEAEs were hypertriglyceridemia, hypertension, and increased blood creatine phosphokinase. Grade 3 TEAEs were resolved by the end of the study in 12 participants, except one participant with grade 3 increased ALT in the HLX03 group was lost to follow-up. One participant in the CN-adalimumab group had a grade 4 (life-threatening) TEAE of increased blood creatine phosphokinase (resolved) (Table 4). One participant in the HLX03 group reported a serious AE of pulmonary tuberculosis; this participant was lost to follow-up, determined by the investigator as possibly unrelated to the study drug. No serious AEs occurred in the CN-adalimumab group.

TEAEs that were assessed as possibly or probably related to the study drug (adverse reactions) were reported for 61.7% and 55.7% of participants in the HLX03 and CN-adalimumab groups, respectively. No unexpected adverse reactions occurred. The most frequently reported adverse reactions were increased ALT, increased AST, and hypertriglyceridemia in the HLX03 group, and rhinorrhea, cough, increased ALT, and hypertriglyceridemia in the CN-adalimumab group (Table 4). The majority of adverse reactions were considered mild to moderate in severity. Grade 3–4 adverse reactions occurred in four (3.7%) participants in the HLX03 group and six (5.7%) in the CN-adalimumab group. There were no clinically significant abnormalities in vital signs, and no AEs leading to early withdrawal or deaths in both groups.

3.4 | Immunogenicity

No pre-existing ADAs were detected at baseline; all ADAs detected during the study developed after dosing with HLX03 or

Parameter, units	Treatment	n	Geometric LS mean	Ratio (%)	90% CI of ratio
Primary PK parameters					
C_{\max} , $\mu\text{g/ml}$	HLX03	104	3.3	102.2	96.2–108.6
	CN-adalimumab	106	3.2		
AUC_{0-t} , $\mu\text{g h/ml}$	HLX03	99 ^a	1823.3	105.7	97.1–115.2
	CN-adalimumab	106	1724.6		
Secondary PK parameters					
$AUC_{0-\infty}$, $\mu\text{g h/ml}$	HLX03	99 ^a	1880.4	105.3	96.1–115.4
	CN-adalimumab	106	1785.6		

TABLE 3 Statistical comparison of PK endpoints between HLX03 and CN-adalimumab (PK-PPS)

Abbreviations: $AUC_{0-\infty}$, area under the concentration curve from time zero to infinity; AUC_{0-t} , area under the concentration curve from time zero to the last quantifiable concentration; CI, confidence interval; C_{\max} , maximum observed concentration; CN-adalimumab, China-sourced adalimumab; LS, least square; PK, pharmacokinetic; PPS, per-protocol set.

^a Data on AUC parameters were excluded for five participants whose % AUC_{ex} exceeded 20%.

CN-adalimumab. At day seven, a higher number of positive ADAs were recorded in the CN-adalimumab group, but there were no statistically significant differences in the incidence of positive ADAs and the incidence of positive NABs between the two groups at other time points (Figure 3A,B).

Over time, the number of ADA-positive participants gradually increased. A total of 101 (96.2%) and 99 (93.4%) participants in the HLX03 and CN-adalimumab groups, respectively, developed ADAs by the end of the trial (day 70 after dosing) (Figure 3A). The number of NAB-positive participants also gradually increased from 14 days post-dose, with NABs detected in 41 (40.6%) participants in the HLX03 group and 41 (41.4%) in the CN-adalimumab group, respectively, by the end of the study (day 70 after dosing). The development of NAB positivity was comparable between the two treatment groups (Figure 3B).

4 | DISCUSSION

This phase 1 clinical trial of HLX03 was designed in accordance with the National Medical Products Administration guidelines to evaluate the PK equivalence of a single dose of HLX03 and the CN-adalimumab reference product. As a secondary objective, this study evaluated the safety and tolerability of HLX03 and its immunogenicity profiles in healthy Chinese volunteers, without the effect of other confounding factors (such as prior exposure to biologics and concomitant medications). A dose level of 40 mg was selected as this is the recommended therapeutic dose of adalimumab for patients weighing ≥ 30 kg.^{5,6,8}

The results of the study demonstrated PK equivalence between HLX03 and CN-adalimumab in healthy Chinese men. After a single 40 mg subcutaneous injection of HLX03 or CN-adalimumab, the mean plasma concentration–time curves of the two drugs were mostly overlapping. The ratios of the geometric means for C_{\max} and AUC_{0-t} were 102.2% and 105.7%, respectively, with corresponding 90% CI that fell within the specified equivalence range of 80% to 125%, thus confirming that HLX03 and CN-adalimumab have

equivalent PK. Furthermore, there was no statistically significant difference in T_{\max} between the two study drugs.

The C_{\max} and T_{\max} values reported in this study were broadly consistent with those reported for adalimumab in healthy adults in the United States (4.7 ± 1.6 $\mu\text{g/ml}$ and 131 ± 56 h, respectively)⁵ and in a Chinese population with RA.⁸ The study showed that the blood concentration–time curve of adalimumab and other PK parameters of Chinese participants were similar to those of Caucasians, with a median T_{\max} of approximately 5 days.⁸

The safety analysis showed that a single dose of HLX03 or CN-adalimumab was generally safe and tolerable in healthy Chinese men, with no statistically significant differences in the incidences of TEAEs or adverse reactions between the two groups. Most TEAEs were grade 1 or 2; one participant had a grade 4 TEAE (increase in blood creatine phosphokinase [CN-adalimumab group]). There were no unexpected adverse reactions. Only one serious AE (pulmonary tuberculosis) was reported in a participant from HLX03 group, which was lost to follow-up without resolution and deemed possibly unrelated to the study drug. No AEs led to early withdrawal or death. Together, these data indicate that HLX03 has a favorable safety profile, with no new safety signals when compared with the reference product. Indeed, the data are broadly consistent with safety data reported in adalimumab-treated patients with plaque psoriasis.²² In that study, the rates of serious AEs, serious infection, and study drug discontinuation due to AEs were also low; however, two participants had tuberculosis that was deemed possibly related to study treatment. The occurrence of tuberculosis is noteworthy,²² as this has been associated with anti-TNF α therapy^{23,24} and occurs at a relatively high incidence in China.²⁵ This suggests that caution may be warranted—along with adequate screening and appropriate prophylaxis, if necessary—in Chinese patients receiving adalimumab or its biosimilars.²²

Anti-TNF α biologics can induce immunogenicity, an antibody-mediated immune response, and cause hypersensitivity reactions. The formation of ADAs can potentially affect PK, safety, and lead to diminished clinical efficacy of biologic drugs.²⁶ On day seven post-dose, the number of participants testing positive for ADAs was

TABLE 4 Summary of safety (safety analysis set)

	HLX03 n = 107	CN-adalimumab n = 106
TEAE, n (%)	79 (73.8)	70 (66.0)
Adverse reaction, n (%)	66 (61.7)	59 (55.7)
Serious AE, n (%)	1 (0.9)	0
TEAE grade 3 and above by PT		
Total	8 (7.5)	6 (5.7)
Hypertriglyceridemia	4 (3.7)	2 (1.9)
Increased ALT	1 (0.9)	0
Blood creatine phosphokinase increased	0	1 (0.9) ^a
Decreased neutrophil count	1 (0.9)	1 (0.9)
Hypertension	2 (1.9)	2 (1.9)
Adverse reaction ^b reported in >5% of participants in any treatment group, n (%) by SOC and PT		
Investigations	33 (30.8)	24 (22.6)
Increased ALT	16 (15.0)	7 (6.6)
Elevated AST	9 (8.4)	5 (4.7)
Blood creatine phosphokinase increased	6 (5.6)	2 (1.9)
Respiratory, thoracic, and mediastinal disorders	16 (15.0)	20 (18.9)
Rhinorrhea	6 (5.6)	12 (11.3)
Nasal obstruction	7 (6.5)	5 (4.7)
Cough	6 (5.6)	8 (7.5)
Oropharyngeal pain	8 (7.5)	6 (5.7)
Metabolism and nutrition disorders	13 (12.1)	9 (8.5)
Hypertriglyceridemia	9 (8.4)	7 (6.6)
Skin and subcutaneous tissue disorders	11 (10.3)	8 (7.5)
Rash	7 (6.5)	5 (4.7)
Gastrointestinal disorders	8 (7.5)	8 (7.5)
Infections and infestations	7 (6.5)	3 (2.8)
Musculoskeletal and connective tissue disorders	7 (6.5)	3 (2.8)
Arthralgia	6 (5.6)	2 (1.9)
General disorders and administration-site conditions	3 (2.8)	6 (5.7)

Abbreviations: AE, adverse events; ALT, alanine aminotransferase; AST, aspartate aminotransferase; CN-adalimumab, China-sourced adalimumab; PT, preferred term; SOC, system organ class; TEAE, treatment-emergent adverse event.

^a This event was reported as a grade 4 TEAE.

^b Adverse reactions are defined as TEAEs possibly or probably related to the study drug.

significantly higher in the CN-adalimumab group than in the HLX03 group; however, there were no statistically significant differences between the two groups in ADA or NAb positivity at any other time point.

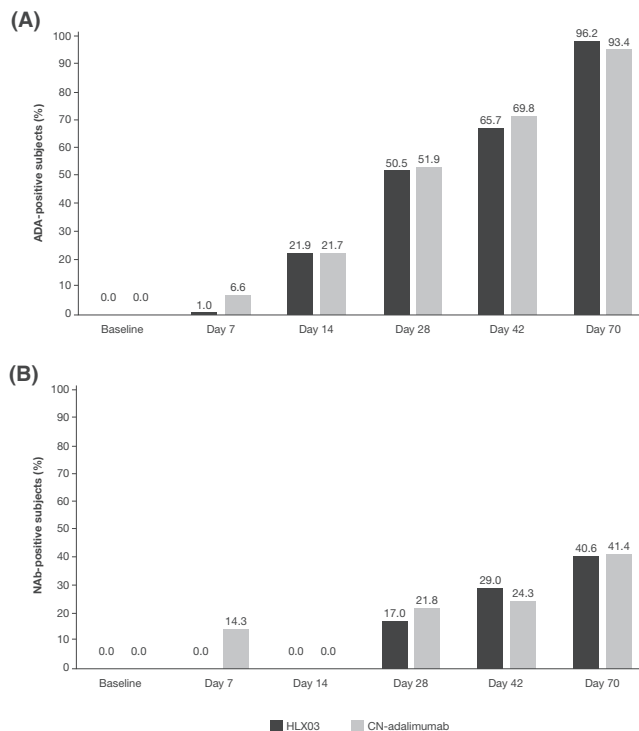


FIGURE 3 Development of (A) ADAs and (B) NABs in healthy participants after a single dose of HLX03 or CN-adalimumab. The positive rate of binding antibody assay was calculated with the number of participants in the analysis set as the denominator; the positive rate of neutralizing antibody was calculated with the number of ADA-positive participants as the denominator. ADA, antidrug antibody; CN-adalimumab, China-sourced adalimumab; NAb, neutralizing antibody

The highest proportions of ADA-positive (96.2% and 93.4%) and NAb-positive healthy participants (40.6% and 41.4%) were observed by day 70 post-dose in the HLX03 and CN-adalimumab groups, respectively. The prevalence of ADA and NAb varies within different populations. For HLX03 in our study, the highest incidence of ADA (96.2%) was similar to those reported in the phase 1 adalimumab biosimilar studies in USA or Europe for BI 695501 (92.5%)²⁷ and SB5 (96.8%),²⁸ but higher than ABP501 (43.3%),²⁹ MSB1102 (82.1%),²⁹ and FKB327 (69.5%).²⁹ In addition, the rate of NAb development in our study (40.6%) was lower than those reported for BI 695501 (59.8%)²⁷ and SB5 (80.3%),²⁸ but higher than ABP501 (17.9%).²⁹ The variations in ADA and NAb positivity were probably due to the different testing methods used and variable sensitivity rates, and thus a larger treatment population to further evaluate the immunogenicity profiles of HLX03 in a phase 3 setting is required.

In conclusion, this phase 1 study in healthy Chinese men demonstrated PK equivalence of HLX03 and CN-adalimumab, with similar safety and immunogenicity profiles. These results add to the totality of evidence supporting the clinical development of HLX03 as an adalimumab biosimilar for the treatment of patients with inflammatory conditions in China, including a phase 3 trial in patients with severe plaque psoriasis (NCT03316781).

ETHICS APPROVAL STATEMENT

The trial was conducted in accordance with the principles of both Good Clinical Practice from the International Conference on Harmonization and the Declaration of Helsinki. The biologic samples were tested in strict accordance with standard operating procedures, the analysis plan, and the principles of Good Laboratory Practice. The trial, which is reported in accordance with CONSORT guidelines, was approved by the Ethics Committee in the First Hospital of Jilin University.

PARTICIPANT CONSENT STATEMENT

All participants gave written informed consent prior to participation.

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CONFLICT OF INTEREST

Xiaodi Zhang and Katherine Chai are employees of Shanghai Henlius Biotech, Inc. All other authors have no conflict of interest.

AUTHOR CONTRIBUTIONS

Xiaoqiao Li and Yanhua Ding contributed to the study design and conception. Hong Zhang, Min Wu, Jixuan Sun, Xiaoxue Zhu, and Cuiyun Li contributed to the data collection and analysis. Xiaodi Zhang and Katherine Chai contributed to the data analysis, interpretation, and manuscript writing and editing. All authors reviewed and provided comments on the manuscript. All authors read and approved the final manuscript.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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