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Efficacy and safety of Reyanning mixture in patients infected with SARS-CoV-2 Omicron variant: A prospective, open-label, randomized controlled trial

Xiangru Xu, Shuang Zhou, Caiyu Chen, Jinhua Li, Hongze Wu, Guoqiang Jin, Jing Zhou, Gang Wang, Min Cao, Ding Sun, Wen Zhang, Wei Peng, Yuting Pu, Yuting Sun, Bangjiang Fang, Jianguang Xu

Department of Emergency, Longhua Hospital, Shanghai University of Traditional Chinese Medicine, Shanghai, China
Acupuncture and Massage College, Shanghai University of Traditional Chinese Medicine, Shanghai, China
Jiangxi Provincial Traditional Chinese Medicine Nephropathy Clinical Research Center, Jiujiang Hospital of Traditional Chinese Medicine, Jiujiang, Jiangxi, China
Department of Health Management, The First Affiliated Hospital of Nanchang University, Nanchang, Jiangxi, China
Department of Respiratory and Critical Care Medicine, The First Affiliated Hospital of Nanchang University, Nanchang, Jiangxi, China
Department of Critical Care Medicine, The Second Affiliated Hospital of Xi’an Jiaotong University, Xi’an, Shaanxi, China
Institute of Critical Care, Shanghai University of Traditional Chinese Medicine, Shanghai, China
Acupuncture and Massage College, Shanghai University of Traditional Chinese Medicine, Shanghai, China

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ABSTRACT

Background: A wave of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) Omicron variant rapidly resulted in a steep increase in the infected population and an overloaded healthcare system. Effective medications for Omicron are currently limited. The previous observational study supports the efficacy and safety of Reyanning (RYN) mixture in the treatment of coronavirus disease 2019 (COVID-19).

Purpose: To evaluate the efficacy of RYN in asymptomatic and mildly infected patients with SARS-CoV-2 infection.

Study Design and methods: This study was a prospective, open-label, randomized controlled trial. We consecutively recruited 2830 patients from Shanghai New International Expo Center mobile cabin hospital and randomized them in a 1:1 ratio to receive RYN plus standard care or receive standard care alone. The primary outcomes were the negative conversion of nucleic acid. Secondary outcomes included the hospital duration, new-onset symptoms, proportion of disease progression, and the viral load measured by the cycle threshold (Ct) value.

Result: A total of 1393 patients in the intervention group and 1407 patients in the control group completed the study. The negative conversion time of nucleic acid was significantly shortened in the intervention group (median: 6 d vs. 7 d, Hazard ratio: 0.768, 95CI %: 0.713-0.828, p < 0.0001). The negative conversion rate of nucleic acid was significantly higher in the intervention group (Day 3: 32.4% vs. 18.3%; Day7: 65.3% vs. 55.2%, p < 0.001). The hospitalization duration was significantly shortened in the intervention group (median: 8 d vs. 9 d, Hazard ratio: 0.759, 95% CI: 0.704-0.818, p < 0.0001). The proportion of new-onset fever (2.4% vs. 4.1%, p = 0.046), coughing (12.2% vs. 14.8%, Hazard ratio: 0.759, 95% CI: 0.704-0.818, p < 0.0001). The proportion of new-onset fever (2.4% vs. 4.1%, p = 0.012), coughing (12.2% vs. 14.8%, p = 0.046), and expectoration (6.0% vs. 8.0%, p = 0.032) in the intervention group was significantly lower. RYN treatment increased Ct values and reduced the viral load. No disease progression and serious adverse events were reported during the study.

Conclusion: RYN is a safe and effective treatment that can accelerate virus clearance and promote disease recovery in asymptomatic and mild Omicron infections.

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The coronavirus disease 2019 (COVID-19) has been spreading globally for nearly three years. As of August 2022, there have been over 579 million confirmed cases of COVID-19, including 6.4 million deaths worldwide (World Health Organization, 2022b). Numerous severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) variants, including alpha, beta, gamma, and delta, have emerged as a constant threat to global health. The emerging Omicron variant has rapidly swept throughout the world since it was first reported in South Africa in November 2021 (Gao et al., 2022). At present, the Omicron variant has become the main epidemic strain throughout the world (Brüssow, 2022). In late February 2022, a significant epidemic of SARS-CoV-2 broke out in Shanghai, China. The scale of this epidemic was more significant than the Wuhan epidemic in 2019, with about 650 thousand people infected in a short period of 4 months (Shanghai Municipal Health Commission, 2022). Gene sequencing analysis in the Shanghai Municipal Center of Disease Control and Prevention laboratory showed that the new viral genomes were clustered into the SARS-CoV-2 BA.2.2 sub-lineage (Zhang et al., 2022b). The Omicron’s R0 could be as high as 10, which was significantly higher than the value of 2.5 of the original strain (Burki, 2022). Compared with the original strain of SARS-CoV-2, Omicron variants displayed reduced pathogenicity but increased infectivity, resulting in a steep increase in the infected population and an overloaded healthcare system (Chen et al., 2022c).

Several antiviral drugs, remdesivir, nirmatrelvir, and molnupiravir, retain their activity against current variants including Omicron in vitro experiment (Vangeel et al., 2022), but further studies are needed to determine whether these drugs are indeed effective in the clinical situation. Effective treatment options for SARS-CoV-2 are currently limited. For more than two years, traditional Chinese medicine (TCM) has been involved in the prevention and control of the COVID-19 epidemic. In March 2022, the World Health Organization (WHO) released the “WHO Expert Meeting on Evaluation of Traditional Chinese Medicine in the Treatment of COVID-19”, which affirmed the safety and effectiveness of TCM in the treatment of COVID-19 (World Health Organization, 2022c). The report stated that TCM might play a role in all stages of viral infection. For mild and moderate diseases, the previous multicenter outbreak, RYN was applied in the clinical observation period and initial treatment of COVID-19 in Shaanxi Province and Hubei Province. Subsequently, RYN was recommended by “Shaanxi Province Novel Coronavirus Pneumonia Traditional Chinese Medicine Treatment Plan (Trial 2nd version)” to treat mild and severe cases (Ma et al., 2020). Preliminary results of the multicenter observational study showed that RYN could effectively improve clinical symptoms, shorten the duration of fever, improve chest CT images, and promote the positive-to-negative conversion of nucleic acid (Yang et al., 2020b). In coronavirus-infected mice, RYN could reduce lung viral loads and severe pulmonary pathology and inhibit the production of pro-inflammatory cytokines, such as TNF-α, IFN-γ, IL-6, and IL-10 (Bao et al., 2020).

Though there is evidence regarding the efficacy of RYN treatment for COVID-19, large-scale randomized controlled clinical trials assessing the efficacy and safety of RYN are still lacking. Besides, the effectiveness of RYN against Omicron remains unclear. For all these reasons, we performed a prospective, open-label, randomized controlled trial to evaluate the efficacy of RYN on virus clearance and disease improvement in asymptomatic and mildly infected patients with SARS-CoV-2 Omicron infection in Shanghai.

Methods

Study oversight

In this prospective, open-label, randomized controlled trial, we recruited COVID-19 patients with asymptomatic/mild infection from Shanghai New International Expo Center mobile cabin hospital, Shanghai, China. The Shanghai New International Expo Center mobile cabin was the first large-scale cabin hospital in Shanghai to be rebuilt in an emergency. The cabin hospital had ten cabins and more than 14,000 beds. The general ward of the cabin hospital mainly treated asymptomatic and mild infected patients who could take care of themselves. The cabin hospital was equipped with a certain proportion of medical staff, medical testing equipment, rescue medicines, and supplemental oxygen, which could ensure daily monitoring of clinical conditions and symptomatic treatment, as well as timely referrals when the disease condition aggravated. The protocol was designed in accordance with the Good Clinical Practice and the Declaration of Helsinki. The clinical study protocol was approved by the Medical Ethics Committee of Jiujiang Hospital of Traditional Chinese Medicine (No. JJSZYYY20220403). The protocol has been registered on the China Clinical Trial Registry website (www.chictr.org.cn/, ChiCTR2200060292). Participation was voluntary, and written informed consent was obtained from all patients.

Study design, randomization, and procedures

This study was a prospective, open-label, randomized controlled trial. Subjects were consecutively enrolled by the investigators. Random permutation sequence and patient random assignment numbers were generated using SPSS software (version 26) by a statistician who was not involved in the trial. Eligible patients were randomly assigned (1:1) into the intervention group (RYN plus standard care) or the control group (standard care alone) according to the simple unrestricted randomization list by researchers. Treatment assignments were concealed from the patients and investigators. Researchers and patients were not masked to treatment assignment and the study medication. All participants received standard care when necessary (e.g., physical condition monitoring, antiviral, antibacterial, symptomatic treatment, and underlying disease treatment) following the Diagnosis and Treatment Protocol for Novel Coronavirus Pneumonia (Triial 9th version) (National Health Commission & State Administration of Traditional Chinese Medicine, 2022). Patients in the intervention group received additional oral RYN (20 ml, 4 times a day) from the first day of enrollment for 7 consecutive days. After 7-day treatment, standard care was maintained for the remaining days until hospital discharge. Other concomitant medications should be documented in detail in the study. Patients hospitalized were...
followed up during their hospital stay until discharge. Patients discharged within 7 days were followed up by telephone during the 7-day treatment course to monitor any adverse events. If patients required hospital transfer for further treatment due to condition deterioration or were discharged from the hospital, they were deemed to have completed the study. The demographic, clinical, and epidemiologic characteristics were investigated on the first day of enrollment. Eligible patients were studied by collecting data about clinical symptoms, comorbidities, concomitant medications, vaccination history, clinical outcomes, and adverse events. The throat swab samples were collected every day for real-time polymerase chain reaction (RT-PCR) analysis of SARS-CoV-2 until discharge. The cycle threshold (Ct) value (as semiquantitative viral load) in the RT-PCR test for SARS-CoV-2 open reading frame (ORF) 1ab or nucleocapsid (N) genes was also included in the analysis.

Patients

We recruited patients with asymptomatic and mild infection at the Shanghai New International Expo Center mobile cabin hospital between April 15 and May 12, 2022. The inclusion criteria were as follows: 1) compliance with the diagnostic criteria for asymptomatic/mild COVID-19 infections, and within 48 hours of admission; 2) patients aged 18-80 years of either sex; 3) voluntarily providing informed consent.

The exclusion criteria included: 1) clinical classification as common type, severe type, or critical type; 2) patients with other severe primary respiratory diseases, such as lung cancer, severe bronchiectasis, and interstitial lung disease; 3) severe systemic diseases (e.g., malignancy, autoimmune diseases, and blood, metabolic, or endocrine diseases) and diseases involving vital organs (heart, brain, liver, and kidney) that could affect the assessment of efficacy; 4) pregnant or lactating women; 5) mental disorders; 6) participating in other clinical trials within 3 months; 7) allergic constitution or history of allergy to the investigational medications.

According to the New Coronavirus Pneumonia Prevention and Control Program (8th edition) (Joint Prevention and Control Mechanism of the State Council, 2022), asymptomatic infection was defined as a positive nucleic acid test result (N gene and ORF gene Ct values <35) without clinical manifestations. The mild type of COVID-19 was defined as slight clinical symptoms with no radiographic evidence of pneumonia based on the Diagnosis and Treatment Protocol for Novel Coronavirus Pneumonia (Trial 9th version) (National Health Commission & State Administration of Traditional Chinese Medicine, 2022).

Study medications

The trial product RYN was composed of *Taraxacum*, *Polygonum cuspidatum*, *Patrinia villosa*, and *Scutellaria barbata*. The RYN (batch No. 220222) was produced by Tsinghua Deren Xi’an Happiness Pharmaceutical Co., Ltd., China. The product was launched in 2005 and approved by the National Medical Product Administration (China) (Approval number: Z20050493). According to the Chinese Pharmacopoeia (2020 edition), 1000 ml of RYN is from 372 g *Taraxacum*, 372 g *Polygonum cuspidatum*, 372 g *Patrinia villosa*, and 186 g *Scutellaria barbata*. According to the fingerprint analysis, the main components of RYN were chlorogenic acid, caffeic acid, polydatin, luteolin, and emodin (Su et al., 2021). The quality control and ingredient identification analyzed by fingerprints were consistent with the Chinese pharmacopoeia reference standards.

Outcomes and measurements

The primary outcome of this trial was the negative conversion time...
of nucleic acid and the negative conversion rate of nucleic acid on day 3 and day 7. Secondary outcomes included the hospital duration, new-onset symptoms of asymptomatic infected patients, and the proportion of disease progression. It also included the SARS-CoV-2 viral load measured by the Ct value of RT-PCR. The Ct value is inversely proportional to the viral load, meaning that a higher Ct value corresponds to a lower SARS-CoV-2 mRNA expression. In samples with undetectable Ct values, the Ct value was manually set to 45 for statistical purposes. All adverse events assessed as possibly or unlikely related to the study medications were recorded.

Negative conversion of nucleic acid was defined as the time from hospitalization to the first day of at least two consecutive nucleic acid tests with N gene and ORF gene Ct values ≥ 35 occurring at least 24 hours apart. The discharge criteria were as follows: (1) body temperature returned to normal for more than 3 days; (2) significantly improved respiratory symptoms; (3) negative conversion of nucleic acid.

Table 1

| Variable                                      | Intervention group (n = 1411) | Control group (n = 1407) | p     |
|-----------------------------------------------|------------------------------|--------------------------|-------|
| Age, years (Median, IQR)                      | 47 (33-55)                   | 45 (32-56)               | 0.159 |
| Age Category                                  |                              |                          |       |
| 18-44 yr (No., %)                             | 638 (45.2%)                  | 688 (48.9%)              |       |
| 45-59 yr (No., %)                             | 565 (40.0%)                  | 486 (34.5%)              |       |
| ≥60 yr (No., %)                               | 208 (14.7%)                  | 233 (16.6%)              | 0.482 |
| Sex                                           |                              |                          |       |
| Males (No., %)                                | 823 (58.3%)                  | 839 (59.6%)              |       |
| Female (No., %)                               | 588 (41.7%)                  | 568 (40.4%)              |       |
| Diseases classification                       |                              |                          | 0.534 |
| Asymptomatic infection (No., %)               | 1123 (79.6%)                 | 1133 (80.5%)             |       |
| Mild infection (No., %)                       | 288 (20.4%)                  | 274 (19.5%)              |       |
| Symptoms                                      |                              |                          |       |
| Fever (No., %)                                | 73 (5.2%)                    | 67 (4.8%)                | 0.615 |
| Fatigue (No., %)                              | 117 (8.3%)                   | 97 (6.9%)                | 0.161 |
| Coughing (No., %)                             | 255 (18.1%)                  | 247 (17.6%)              | 0.72  |
| Expectoration (No., %)                        | 153 (10.8%)                  | 161 (11.4%)              | 0.613 |
| Muscular soreness (No., %)                    | 73 (5.2%)                    | 64 (4.5%)                | 0.441 |
| Concomitant medications                       |                              |                          |       |
| Antiviral (No., %)                            | 11 (0.8%)                    | 24 (1.7%)                | 0.026 |
| Antibiotics (No., %)                          | 87 (6.2%)                    | 69 (4.9%)                | 0.143 |
| Oral corticosteroids (No., %)                 | 0                            | 2 (0.1%)                 | 0.249 |
| Antipyretic analgesics (No., %)               | 44 (3.1%)                    | 50 (3.6%)                | 0.52  |
| Antihistamines (No., %)                       | 27 (1.9%)                    | 30 (2.1%)                | 0.68  |
| Antitussive (No., %)                          | 82 (5.8%)                    | 89 (6.3%)                | 0.568 |
| Expectorant (No., %)                          | 101 (7.2%)                   | 119 (8.5%)               | 0.199 |
| Nasal decongestant (No., %)                   | 15 (1.0%)                    | 10 (0.7%)                | 0.319 |
| Antitussive (No., %)                          | 5 (0.4%)                     | 9 (0.6%)                 | 0.281 |
| Comorbidities                                  |                              |                          |       |
| Hypertension (No., %)                         | 168 (11.9%)                  | 182 (12.9%)              | 0.408 |
| Diabetes mellitus (No., %)                    | 48 (3.4%)                    | 56 (4.0%)                | 0.416 |
| Cerebrovascular disease (No., %)              | 10 (0.7%)                    | 7 (0.5%)                 | 0.469 |
| Cardiovascular disease (No., %)               | 41 (2.9%)                    | 35 (2.5%)                | 0.493 |
| Chronic pulmonary disease (No., %)            | 9 (0.6%)                     | 20 (1.4%)                | 0.039 |
| Chronic liver disease (No., %)                | 4 (0.3%)                     | 1 (0.1%)                 | 0.375 |
| Chronic kidney disease (No., %)               | 2 (0.1%)                     | 2 (0.1%)                 | >0.99 |
| Vaccination history                           |                              |                          |       |
| 0 doses (No., %)                              | 303 (21.5%)                  | 323 (23.0%)              | 0.344 |
| 1 dose (No., %)                               | 45 (3.2%)                    | 48 (3.4%)                | 0.741 |
| 2 doses (No., %)                              | 400 (28.3%)                  | 410 (29.1%)              | 0.643 |
| ≥3 doses (No., %)                             | 663 (47.0%)                  | 626 (44.5%)              | 0.184 |

Statistical analysis

The sample size calculation was determined by the time window of this study, and no sample size calculation was made. For the continuous variables, normal and non-normal distribution data were analyzed using the independent-sample t-test and the Mann-Whitney test, respectively. Normal distribution data were expressed as mean ± standard deviation (mean ± SD), and non-normal distribution data were expressed as median with interquartile range (IQR). The categorical variables were summarized by presenting the frequency and proportion (%) and analyzed by the chi-square test or Fisher’s exact test. The time to events was presented as median with a 95% confidence interval (CI) and analyzed by Kaplan-Meier analysis. Changes in SARS-CoV-2 viral loads during the observation were normalized using the local regression (loess) method. Statistical analyses were performed using SPSS 26.0 (IBM Corp, USA), with statistical significance defined as p < 0.05 (two-sided test).

Results

Baseline characteristics

A total of 5759 patients were assessed for eligibility between April 14 and May 12, 2022, and 2929 were excluded for not meeting eligibility criteria or declining to participate. Therefore, 2830 patients were enrolled and randomly assigned (1:1) into the intervention group (treated by RYN plus standard care) and the control group (treated by standard care). Twelve patients were excluded due to protocol violations after randomization. During this trial, 18 patients in the intervention group dropped out. Finally, 1393 patients in the intervention group and 1407 patients in the control group were analyzed. The study flowchart was shown in Fig. 1.

Baseline demographic and clinical characteristics were summarized in Table 1. Of the 2818 patients, 2256 patients (80.1%) were asymptomatic infections and 562 (19.9%) were mild infections. Among the included patients, 59.0% of those were male, and 41.0% were female. The median age of enrolled patients was 46 years (IQR: 33-56 years). The proportion of patients was highest in adults aged 18-44 years. The most common symptoms on admission were coughing (17.8%), expectoration (11.1%), fatigue (7.6%), fever (5.0%), and muscular soreness (4.9%). Hypertension (12.4%) and diabetes (3.7%) were the most common comorbidities at baseline. Overall, 156 (5.5%) patients received antibiotics, 35 (1.2%) received antiviral treatment, and 2 (0.1%) corticosteroids. Symptomatic therapies included antipyretic analgesics (3.3%), antihistamines (2.0%), antitussive (6.1%), expectorant (7.8%), nasal decongestant (0.9%), and antiasthmatic (0.5%). Most patients were fully vaccinated, and 2099 (74.5%) cases had received two or more doses of vaccines. Baseline demographic and clinical characteristics had no statistically significant difference between the two groups (p > 0.05), except for antiviral treatment and underlying chronic pulmonary disease. (p < 0.05).

Primary outcome

The median negative conversion time of nucleic acid was 6 days (IQR: 3, 9) in the intervention group and 7 days (IQR: 5, 9) in the control group. The Kaplan-Meier analysis showed a higher negative conversion rate of nucleic acid in the patients receiving RYN (Hazard ratio: 0.768, 95% CI: 0.713-0.828, p < 0.0001) (Fig. 2). By day 3, 452 (32.4%) patients in the intervention group and 258 (18.3%) in the control group were tested negative for nucleic acid (p < 0.001). Asymptomatic infection was associated with a higher negative conversion rate of 29.4% vs. 16.1% (p < 0.001). By day 7, 910 (65.3%) patients in the intervention group tested negative for nucleic acid versus 776 (55.2%) in the control group (p < 0.001). The negative conversion rate of asymptomatic infection patients in the intervention group was significantly higher than
that in the control group (55.1% vs. 46.2%, \( p < 0.001 \)) (Fig. 3).

**Secondary outcome**

There were no cases of disease progression during the observation. The median hospitalization duration was 8 days (IQR: 5, 11) in the intervention group and 9 days (IQR: 7, 11) in the control group. The Kaplan-Meier analysis showed a higher discharge rate in the patients receiving RYN (Hazard ratio: 0.759, 95% CI: 0.704-0.818, \( p < 0.0001 \)) (Fig. 4).

Among the 2256 patients who were initially asymptomatic on admission, 183 cases in the intervention group and 245 in the control group developed symptomatic COVID-19 during observation. The proportion of new-onset fever (2.4% vs. 4.1%, \( p = 0.012 \)), coughing (12.2% vs. 14.8%, \( p = 0.046 \)), and expectoration (6.0% vs. 8.0%, \( p = 0.032 \)) in the intervention group was significantly lower than that in the control group (Fig. 5).

For quantitative PCR assays, a Ct value of < 40 was used as the cut-off value for determining positivity for most organisms. However, the Ct value of 35 or more was set as the cut-off value for defining as non-detected according to the latest guideline in China (Trial 9th version) (National Health Commission & State Administration of Traditional Chinese Medicine, 2022). The viral load Ct values of the SARS-CoV-2 ORF gene and N gene at each time point were plotted to make the trend line using a loess method. It was observed that in Fig. 6, the overall trend towards the Ct values of the ORF/N gene in the intervention group was higher than those of the control group. During the treatment period, the Ct values continued to increase, and the smoothed curve flattened after the Ct value reached 40. Fig. 7 showed that asymptomatic infections exhibited higher Ct values than mild symptomatic infections. RYN treatment promoted a faster increasement in the ORF/N gene Ct values in asymptomatic patients (Fig. 7A, B). From Fig. 7C, D, it could be
seen that the Ct values of the first nucleic acid test were higher in the vaccinated group. Besides, RYN treatment promoted the negative conversion of nucleic acid, especially for unvaccinated patients.

Safety

During the observation, 323 single adverse events (AEs), with 162 in the intervention group and 161 in the control group, were observed and assessed as mild. The most common adverse event was diarrhea (2.2%), gastralgia (2.6%), insomnia (1.8%), and constipation (1.1%). Of all the AEs, diarrhea was assessed as possibly related to RYN treatment, while others were assessed as unrelated to the medication. All AEs were improved with symptomatic therapy. No statistically significant difference between groups was observed in the incidence of AEs (Table 2).

Discussion

The Omicron variant has rapidly spread around the world and become the dominant variant circulating globally. As of August 7, 2022, 201 countries have discovered and shared 5,117,867 Omicron genome
sequences on the GISAID website (GISAID, 2022). Omicron variants, including BA.1, BA.2, BA.3, BA.4, BA.5, and descendent lineages, have continued to evolve, resulting in humoral immune escape potential and higher transmissibility (World Health Organization, 2022c). As a result, Omicron was registered by the WHO as a variant of concern (VOC) that requires prompt action (World Health Organization, 2022a). We previously performed a study and preliminarily analyzed the clinical characteristics of Omicron-infected patients in Shanghai (Xu et al., 2022). The results showed that the Omicron variant mainly caused asymptomatic and mild infections, with upper respiratory symptoms such as cough, expectoration, and fever. The males and middle-aged population have a relatively high infection rate. These findings were consistent with the demographic and clinical characteristics of this study. In this prospective, open-label, randomized controlled trial, we
provide clinical evidence of RYN in the treatment of SARS-CoV-2 Omicron infection, demonstrating that RYN is a safe treatment associated with rapid viral clearance and disease recovery. Specifically, compared with standard care, RYN improved the negative conversion rate of nucleic acid, shortened the negative conversion time and hospitalization duration, reduced new-onset symptoms in asymptomatic patients, and reduced the viral load. For patients with the asymptomatic infection on admission, RYN inhibited the progression from asymptomatic infection to mild disease. Furthermore, RYN had a favorable safety profile in COVID-19 patients, as no serious side effects were reported. These findings all suggested the utility of RYN as a promising treatment against the SARS-CoV-2 Omicron variant.

From the data of this study, the overall vaccination coverage rate of infected patients reached 78%, and nearly half of the patients (45.7%) received booster doses. Combined with previous studies of blood samples and emerging real-world investigations, all the evidence suggests that Omicron developed escaped immunity, whether from previous infections or vaccination (Chen et al., 2022b; Cohen, 2021). However, no vaccine against the Omicron variant has yet entered clinical trials. In addition, the choice of antiviral drugs is quite limited. Antivirals against SARS-CoV-2 can be divided into two classes: monoclonal antibodies against the Spike protein and small molecules that interfere with the viral replication. Most of the existing approved monoclonal antibodies have lost neutralizing activity against SARS-CoV-2 Omicron due to the hypermutability of the spike protein (VanBlargan et al., 2022). Several small-molecule antiviral drugs targeting conserved SARS-CoV-2 RNA-dependent RNA polymerase (RdRp) or the main protease (Mpro) have received approval or emergency use authorization (EUA), and are progressing in clinical trials. Remdesivir and Molnupiravir are RdRp inhibitors of SARS-CoV-2, and nirmatrelvir/ritonavir (Paxlovid) targets the Mpro. Previous clinical studies have shown that among mild-to-moderate, unvaccinated, nonhospitalized adults at high risk for severe COVID-19 progression, early use of the above antiviral drugs can cause a lower risk of hospitalization or death (Gottlieb et al., 2022; Hammond et al., 2022; Jayk Bernal et al., 2022). However, these trials excluded patients who had received SARS-CoV-2 vaccines. Thus, it remains unknown whether these drugs also apply to areas with high vaccination coverage.

Since the outbreak of COVID-19, substantial clinical evidence supports TCM as an add-on therapy for COVID-19 and has shown clinical benefits in promoting symptom improvement and disease recovery. For patients with mild-to-moderate COVID-19 pneumonia, Lianhua Qingwen capsules can promote the symptom recovery of fever, fatigue, and cough and increase the clinical cure rate (Hu et al., 2021); Xiyanping injection significantly reduced cough, fever, and virus clearance time.

Table 2

| Adverse events | Intervention group (n = 1411) | Control group (n = 1407) | P-value |
|----------------|-------------------------------|-------------------------|---------|
| Diarrhea       | 30 (2.1%)                     | 33 (2.3%)              | 0.694   |
| Gastralgia     | 37 (2.6%)                     | 35 (2.5%)              | 0.821   |
| Insomnia       | 20 (1.4%)                     | 30 (2.1%)              | 0.151   |
| Constipation   | 15 (1.1%)                     | 17 (1.2%)              | 0.716   |
| Chest tightness| 11 (0.8%)                     | 11 (0.8%)              | 0.995   |
| Dyspepsia      | 5 (0.4%)                      | 3 (0.2%)               | 0.726   |
| Flatulence     | 7 (0.5%)                      | 6 (0.4%)               | 0.785   |
| Diarrhea       | 7 (0.5%)                      | 1 (0.1%)               | 0.07    |
| Headache       | 12 (0.9%)                     | 7 (0.5%)               | 0.252   |
| Limb pain      | 7 (0.5%)                      | 7 (0.5%)               | 0.996   |
| Conjunctivitis | 5 (0.4%)                      | 4 (0.3%)               | >0.99   |
| Rash           | 6 (0.4%)                      | 7 (0.5%)               | 0.777   |

Fig. 7. Variation of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) viral load Cycle Threshold (Ct) values in subgroups. (A-B) Smoothed loess-curves of ORF/N gene Ct values in asymptomatic and mild infection subgroups. (C-D) Smoothed loess-curves of ORF/N gene Ct values in vaccinated and unvaccinated subgroup.
P53, C-reactive protein, peroxisome proliferator-activated receptor—targets (such as tumor necrosis factor, interferon-gamma, tumor protein) modulates immune function, and reduces lung injury by modulating has anti-inflammatory and antiviral effects and is widely used in the clinical treatment of respiratory tract infections and pneumonia. The network pharmacology study of RYN showed it inhibits inflammation, modulates immune function, and reduces lung injury by modulating targets (such as tumor necrosis factor, interferon-gamma, tumor protein P53, C-reactive protein, peroxisome proliferator-activated receptor—gamma) to achieve the purpose of treating SARS-CoV-2 (Yang et al., 2020a). Here, our results provide additional evidence that RYN is also an effective measure for COVID-19 treatment and provides novel therapeutic strategies to treat Omicron variants.

However, the study also has several limitations. First, this was an open-label study design which might introduce bias in outcome assessment. And since the initial positive nucleic acid test results was not obtained before admission, the viral shedding duration could not be assessed in this study. Therefore, future explorations, especially randomized, double-blind, placebo-controlled clinical trials, are needed to further confirm the efficacy of RYN. Second, this study only included patients with mild or asymptomatic COVID-19. The safety and efficacy of RYN in patients with moderate, severe, and critical diseases require further investigation. Finally, TCM formulations have multiple components and targets, and designing it difficult to elucidate their mechanisms in depth.

Conclusion

RYN is a safe and effective treatment that can accelerate virus clearance and promote disease recovery in asymptomatic and mild Omicron infections. Further randomized, double-blind, placebo-controlled clinical trials are needed to assess the efficacy of RYN in patients of different ages and with varying disease severity.

CRediT authorship contribution statement

Xiangru Xu: Methodology, Visualization, Writing – original draft. Shuang Zhou: Methodology, Supervision, Writing – review & editing. Caiyu Chen: Formal analysis, Writing – original draft. Jinhua Li: Investigation, Data curation. Hongze Wu: Investigation, Supervision. Guoqiang Jin: Investigation, Supervision. Jing Zhou: Investigation, Data curation. Gang Wang: Formal analysis, Writing – review & editing. Min Cao: Formal analysis, Data curation. Ding Sun: Formal analysis, Data curation. Wen Zhang: Data curation. Wei Peng: Data curation. Yuting Pu: Data curation. Yuting Sun: Data curation. Bangliang Fang: Conceptualization, Project administration, Funding acquisition, Writing – review & editing. Jianguang Xu: Project administration, Supervision, Writing – review & editing.

Declaration of Competing Interest

The trial product Reyanning mixture (batch No. 220222) was provided by Tsinghua Deren Xi’an Happiness Pharmaceutical Co., Ltd., China. The company played no role in the study design, data collection, analysis, and manuscript preparation. The authors declare that they have no competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Trial protocol

The full trial protocol can be accessed at http://www.chictr.org.cn/showproj.aspx?proj=170609 or by contacting the corresponding author.

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