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## RESEARCH ARTICLE

# A Clinical Comparison of Imported or Locally Acquired COVID-19 Suggests Declining Severity after Viral Passages at the Early Stage of the Pandemic in China

Feifei Su<sup>1,\*</sup>, Shoufeng Yang<sup>1</sup>, Liang Hong<sup>2</sup>, Yao Sun<sup>3</sup> and Yue Qu<sup>3,4</sup>

<sup>1</sup>Department of Infectious Diseases, Wenzhou Central Hospital, Wenzhou, 325000, China

<sup>2</sup>Department of Infectious Diseases, Ryan People's Hospital, Rui'an, 325200, China

<sup>3</sup>Monash Biomedicine Discovery Institute Alliance in Clinical and Experimental Biomedicine, Wenzhou Medical University, Wenzhou, 325000, China

<sup>4</sup>Department of Microbiology, Monash Biomedicine Discovery Institute, Monash University, Melbourne, 3800, Australia

### Abstract:

#### Objective:

This retrospective, two-center study aimed to provide solid clinical evidence to support the viral attenuation theory after passages of SARS-CoV-2 during the early stages of the pandemic.

#### Methods:

Clinical characteristics and outcomes of 150 COVID-19 patients hospitalized in Wenzhou, China between January and April 2020 were compared. These patients had similar demographic characteristics. Infections of 77 patients who visited Wuhan within 14 days before symptom onset were categorized as imported cases and that of the remaining 73 patients who had no recent travel history were categorized as locally transmitted cases.

#### Results:

COVID-19 cases in Wenzhou appeared to be mostly mild at the early stages of the pandemic. There were no differences in clinical manifestations, laboratory testing results, and radiographic presentation between imported and locally transmitted cases in Wenzhou, except that a higher proportion of lymphopenia was found in the imported case group. Assessment of infection severity showed that severe conditions were observed in 10.7% of the patients, with the imported case group having a significantly higher rate (15.6%) than the locally transmitted case group (5.5%,  $\chi^2 = 4.016$ ,  $p = 0.045$ ).

#### Conclusion:

Although, the clinical manifestations of locally acquired infections were indistinguishable from those imported from Wuhan, they were less likely to develop into severe medical conditions, suggesting the possibility of virulence attenuation after viral passages during the early stages of the pandemic.

**Key words:** COVID-19, Imported cases, Locally transmitted infections, Clinical comparison, Multivariable analysis and severity, Pandemic.

### Article History

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## 1. INTRODUCTION AND BACKGROUND

The Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) emerged in the city of Wuhan (Hubei Province, China) in December 2019 as a novel member of the Coronavirus group and causes the highly infectious coronavirus disease 2019 (COVID-19) [1 - 3]. The initial

cluster of this disease was first reported in people associated with the Huanan Seafood Wholesale Market [1]. Numerous studies have since been published to extend our understanding of this disease. Clinical presentations of COVID-19 are variable, ranging from asymptomatic to mild and moderate manifestations such as fever, cough, and breathing difficulties, to more severe conditions in sensitive populations [1, 4 - 7]. Some patients developed Acute Respiratory Distress Syndrome (ARDS), acute respiratory failure and multiple organ failure, requiring mechanical ventilation, intensive care and/or

\* Address correspondence to this author at Department of Infectious Diseases, Wenzhou Central Hospital, 325000, Wenzhou, Zhejiang, China; Email: feifeisuzs@163.com

Extracorporeal Membrane Oxygenation (ECMO) therapy [1]. The gene sequence of SARS-CoV-2 is highly similar to that of Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) [8], with both viruses harboring genes encoding spike glycoproteins that specifically bind to Angiotensin Converting Enzyme 2 (ACE2) on the surface of Type II alveolar epithelial cells and/or hepatocytes, causing severe damage to the respiratory system and the liver [9, 10]. Post-mortem analysis of COVID-19 patients found pathological changes including diffuse alveolar damage, formation of hyaline membrane and pneumocyte atypical hyperplasia [11, 12].

Following the initial outbreak of COVID-19 in Wuhan, infections rapidly spread to other parts of mainland China [13]. Wenzhou was among the areas most severely affected by COVID-19 outside the Hubei province. The epidemic in Wenzhou was a mixture of case importations and local transmission, mainly associated with a large number of business persons fleeing Wuhan prior to the pandemic lockdown on the 23<sup>rd</sup> of January, 2020. The epidemiological features and transmission dynamics of COVID-19 beyond Wuhan and the Hubei province have been well described [13]. Human-to-human transmission of SARS-CoV-2 is well established, affecting individuals of all ages, including children and infants, with an incubation period of 3-14 days after infection [14, 15]. The asymptomatic incubation period also pose a high risk for viral transmission [16]. Multiple strategies have been implemented nationwide in China to combat the infection, including early detection, contact tracing, and strict containment for infected individuals [13]. Early detection and contact tracing are extremely important strategies for the successful management of the COVID-19 pandemic [17]. New diagnostic assays based on nanomaterial technologies, such as the plasmonic meta sensor technology that targets SARS-CoV-2 spike proteins, are currently under development for rapid and precise screening of SARS-CoV-2 virus carriers [18, 19].

Coronaviruses are able to undergo genetic recombination and rapid mutation that may significantly impact viral virulence and disease severity [14]. A less virulent SARS-CoV-2 variant with a 382-nucleotide deletion within its genome has recently been identified in Singapore and other countries [20]. Our study retrospectively collected and compared clinical data from COVID-19 patients with similar demographic characteristics and infected by different generations of SARS-CoV-2, aiming to provide more solid clinical evidence to support the viral attenuation theory after passages of SARS-CoV-2 during the early stages of the pandemic.

## 2. METHODS

### 2.1. Patients and Classification of COVID-19

We retrospectively collected epidemiological, clinical, laboratory, and radiological data from electronic medical records of 150 COVID-19 patients admitted to the Ryan People's Hospital (46 patients) and the Wenzhou Central Hospital (104 patients) between January 17<sup>th</sup>, 2020 and February 4<sup>th</sup>, 2020. All patients were consecutively selected for

this study. Both hospitals are designated tertiary hospitals for major infectious diseases in the Zhejiang province. These cases were divided into four generations according to the epidemic sources of the infection spread [9]: Generation one included patients that returned from Wuhan to Wenzhou within 14 days before symptom onset and were exposed to Huanan Seafood Wholesale Market; generation two were cases found in patients who returned from Wuhan within 14 days before symptom onset, but without exposure to Huanan Seafood Wholesale Market; generation three was comprised of patients who did not visit Wuhan or other cities between December, 2019 to February, 2020 but had known exposure to patients returned from Wuhan; generation four was for patients who did not visit Wuhan and had no suspected contact with patients returned from Wuhan. Cases of generation one and two were further categorized as imported cases and generation three and four as locally transmitted. This study was approved by the Ethics Review Boards of Wenzhou Central Hospital and Ryan People's hospital. Consent to participate was explained to the patient and written consent was received.

### 2.2. Clinical Management of COVID-19

Clinical management of COVID-19 followed the Guidelines on the Diagnosis and Treatment of COVID-19 Patients (Trial Version 4) of the National Health Commission of China [3]. All patients underwent Reverse Transcription Polymerase Chain Reaction (RT-PCR) tests, and chest CT scans (1.25mm in thickness, continuous scans, lung and mediastinum windows respectively). For RT-PCR tests, nasopharyngeal swabs were obtained and sent to Wenzhou Center for Disease Control for testing for SARS-CoV-2, influenza A and B, avian influenza, adenoviruses, swine-flu viruses, SARS-CoV and Middle East Respiratory Syndrome Coronavirus (MERS-CoV). Abnormal presentations on CT scans were categorized as ground-glass changes or pulmonary consolidation according to the density of pulmonary lesions, or as central or peripheral pulmonary diseases based on their distances to the hilum. Routine laboratory tests included complete blood counts and serum biochemical tests for kidney and liver function, levels of creatine kinase, lactic acid dehydrogenase and electrolytes. Patients with positive RT-PCR test results were isolated in negative pressure isolation wards. Antiviral (interferon alpha-2b, aerosol inhalation 5 million U, bid; lopinavir tablet, 500mg bid, arbidol tablet, 0.2 tid) and other supportive therapies were given. Nasal oxygen support and noninvasive ventilator were given to patients who became hypoxemic.

### 2.3. Criteria for COVID-19 Diagnosis, Classification of Disease Severity, and Patient Discharge

COVID-19 was diagnosed in both hospitals by following the same guidelines from the National Health Commission of China [3]. Diagnostic criteria for COVID-19 included 1) at least two of the following clinical manifestations, fever and respiratory tract symptoms, radiographic presentations suggesting pneumonia, normal or reduced white blood cell counts or reduced lymphocyte counts at the early stage of illness, and an epidemiological link to confirmed COVID-19 cases and, 2) positive RT-PCR result for SARS-CoV-2.

The severity of COVID-19 cases was classified according to the same guidelines, as follows; 1) mild infections, with mild clinical symptoms and no abnormal changes on chest CT scans; 2) moderate infections, with fever, respiratory tract and other symptoms and a typical pneumonia presentation on CT images; 3) severe infections, if any of the following indicators presented, respiratory distress with a Respiratory Rate (RR) > 30 times/min, an oxygen saturation of 93% or below, and a PaO<sub>2</sub>/ FiO<sub>2</sub> ratio (the ratio of arterial oxygen partial pressure to fractional inspired oxygen) < 300 mmHg; 4) critical infections, including respiratory failure requiring a mechanical ventilator, shock, other organ failures that required intensive care.

The discharge criteria for COVID-19 used by both hospitals between January and April, 2020 included a normal temperature for longer than 72h, significant improvement of respiratory tract symptoms including cough, sputum production, chest tightness and shortness of breath, and two consecutive negative SARS-CoV-2 RT-PCR tests at least 24 h apart [3].

#### 2.4. Data Processing and Statistical Analysis

Data were entered into a standardized collection form. Median values representing continuous variables were compared using the non-parametric Mann-Whitney method. Categorical variables for clinical manifestations and severity of patients in different groups were compared using the Chi-squared (X<sup>2</sup>) test. To more accurately predict the contribution of different patient related risk factors to disease severity or its associated clinical presentations, a multivariable analysis was further conducted, using infection severity and lowered leucocyte counts as response variables respectively and selected risk factors as explanatory variables. Risk factors that might differ between the imported infection group and locally acquired infection group were selected, including gender, age, cough, length of hospitalization, and pulmonary consolidation. Mechanical ventilation was excluded as this treatment had an immediate link to disease severity. SPSS 24.0 Software was used for statistical analysis. A *P*-value less than 0.05 was considered to be statistically significant.

### 3. RESULTS

#### 3.1. COVID-19 Arrived in Wenzhou

The first COVID-19 case in Wenzhou was diagnosed on January 16, 2020 and the patient was immediately admitted to the Wenzhou Central Hospital. Between January 17 and February 4, 2020, further 149 COVID-19 patients were hospitalized at the Ryan People's Hospital (46 patients) and the Wenzhou Central Hospital (104 patients in total). All 150 patients were permanent residents of Wenzhou. Among these patients, 77 visited Wuhan between December, 2019 and February, 2020 for business purposes; three had visited the Huanan Wholesale Seafood Market. Seventy-three patients had no travel history to Wuhan or other cities in the Hubei province during the same period; 37 had confirmed exposure to patients who recently returned from Wuhan. All patients were successfully treated and discharged before April 2, 2020.

#### 3.2. Similar Demographic Characteristics of Patients in the Imported and Locally Transmitted Groups

There was no significant difference between the imported case group and the local transmission group, in the birth origin of patients, gender (*P*=0.193), age (*P*=0.091), age distribution using 45 and 65 years of age as cutoff values (*P*=0.349), prevalence of underlying diseases (*P*=0.193), and the interval between symptom onset to hospitalization (*P*=0.702) (Table 1). Of 150 patients, 50.7% were male and 49.3% were female (Table 1). The median age of all patients was 45 years old (Table 1). Only 12% of patients had underlying diseases, with diabetes mellitus being the most prevalent (10/18). The median time from symptom onset to hospitalization for all patients was 5 days.

Epidemiological investigation of patients' travel and residence history as well as contact tracing revealed a clear link between individual patients with the second generation of COVID-19 and several small clusters with the third generation of infections, suggesting possible viral passages from imported cases to locally transmitted cases.

#### 3.3. Clinical Presentations of Imported and Locally Transmitted COVID-19

There was no significant difference in the clinical manifestations, radiographic characteristics and laboratory test results between the locally transmitted and imported infection groups, with the exception of a slightly lower lymphocyte count in the latter (Table 1). Common symptoms of symptomatic COVID-19 cases in Wenzhou included fever (129 cases, 86.0%), cough (99 cases, 66.0%), chest tightness and/or chest pain (21 cases, 14.0%) and diarrhea (25 cases, 16.7%) (Table 1). At the time of admission 136 patients (90.7%) had normal total white blood cell counts while 53 patients (35.3%) had a lowered lymphocyte count (<1 × 10<sup>9</sup>/L). Leukopenia was more common in the imported infection group in comparison to the locally infected group (*P* = 0.02) (Table 1). An increase in the CRP level was observed in 97 patients (64.7%). One hundred forty patients (93.3%) had abnormal chest CT images, with 134 patients (89.3%) showing ground-glass opacities and 107 patients (71.3%) showing pulmonary consolidation. Such CT changes often involved multiple lobes in both lungs, with a median number of four lobes affected per patient. Thirty-seven locally infected patients who had a confirmed exposure to imported COVID-19 (the 3<sup>rd</sup> generation) and 36 who had no known exposure (the 4<sup>th</sup> generation) were further compared. No differences in the clinical presentations, radiographic characteristics, and laboratory test results were identified.

#### 3.4. Management and Clinical Outcomes of Imported and Locally Transmitted COVID-19

No specific treatments were allocated to patients in the imported and locally-acquired infection groups, with the exception of noninvasive mechanical ventilation that was given to two patients of 3<sup>rd</sup> generation due to hypoxemia. Among 150 patients, 146 received interferon, 132 received Lopinavir/Ritonavir, and 77 patients received Umifenovir. Thirty-one patients also received empiric broad-spectrum antibiotics. Systemic steroids were not used in these patients.

There was no difference in the discharge rates and length of hospital stay between the two groups (Table 1). All patients were discharged before 2<sup>nd</sup> of April, 2020 with a median hospital stay of 21 days. There were no deaths in this cohort of COVID-19 patients.

### 3.5. Patients with Imported COVID-19 Developed more Severe Infections than Locally Infected Patients

The main focus of this study was to compare the severity of COVID-19 in patients who were infected in Wuhan prior to returning to Wenzhou and those who were infected locally, *via* a known or unknown exposure. There were 13 cases of severe infections and three cases of critical infections among the 150 COVID-19 cases (Table 2). A majority of severe and critical patients were noted as being in the imported case group (12/16 cases, 75.0%). All three critical infections occurred in patients with imported COVID-19. A Chi-square test suggested that patients who returned from Wuhan were more likely to develop severe or critical conditions compared to those who acquired infections locally (15.6% *versus* 5.5%,  $X^2=4.016$ ,  $P=0.045$ ).

Regression analysis indicated that there was a strong association between the severe condition of COVID-19 and the leukopenia status of patients ( $P=0.006$ ). Multivariable analysis using disease severity as the single outcome suggested that gender and age were significant contributors. Importation from Wuhan also appeared to be a substantial risk factor, with a  $P$  value (0.074) reaching a significant level (Table 3). A parallel multivariable analysis using leukopenia as the single outcome suggested that importation from Wuhan, age, length of hospitalization, and pulmonary consolidation were all significant predictors of lowered lymphocyte counts (Table 4).

## 4. DISCUSSION

This is a two-center retrospective study comparing clinical features and the severity of COVID-19 locally acquired in Wenzhou or imported from Wuhan at the very early stage of the pandemic. Our key finding is that although the clinical manifestations of COVID-19 remained the same after migration from Wuhan to Wenzhou, the disease severity dropped significantly, suggesting a possible virulence attenuation following viral passages.

**Table 1. Demographic and clinical characteristics of COVID-19 patients.**

Demographic Characteristics		All Patients (n=150)	Imported (n=77)	Locally Infected (n=73)	P- value
Gender	Male	76 (50.7%)	43 (55.8%)	33 (45.2%)	0.193
	Female	74 (49.3%)	34 (44.2%)	40 (54.8%)	
Age, Years	Ages*	45 (35, 54)	46 (39, 55)	43 (31, 52)	0.091
	< 45	69 (46.0%)	31 (40.3%)	38 (52.1%)	
	45--64	72 (48.0%)	41 (53.2%)	31 (42.5%)	
	≥ 65	9 (6.0%)	5 (6.5%)	4 (5.5%)	0.349
Underlying Diseases	Patient numbers(percentage)	18 (12.0%)	10 (13.0%)	8 (11.0%)	0.702
	Diabetes	10	8	2	
	Simple diabetes	1	0	1	
	Diabetes/HBP	8	7	1	
	Diabetes/Others	1	1	0	
	Other chronic wasting diseases including cancers	8	2	6	
Symptom Onset to Hospitalization	Median days (range)	5 (1, 30)	5 (1, 30)	5 (1, 23)	0.836
<b>Clinical characteristics</b>					
Signs and Symptoms	Fever	129 (86.0%)	68 (88.3%)	61 (83.6%)	0.402
	Cough	99 (66.0%)	56 (72.7%)	43 (58.9%)	0.074
	Chest pain/tightness	21 (14.0%)	11 (14.3%)	10 (13.7%)	0.945
	Diarrhea	25 (16.7%)	12 (15.6%)	13(17.8%)	0.715
WBC Counts	High, > 10 X 10 <sup>9</sup> L <sup>-1</sup>	0	0	0	/
	Low, < 4 X 10 <sup>9</sup> L <sup>-1</sup>	14 (9.3%)	8 (10.4%)	6 (8.2%)	0.648
Lymphocyte Counts	Lowered, < 1 X 10 <sup>9</sup> L <sup>-1</sup>	53 (35.3%)	34 (44.2%)	19 (26.0%)	0.020**
CRP	Increased, > 5.0 mg L <sup>-1</sup>	97 (64.7%)	52 (67.5%)	45 (61.6%)	0.451
Mechanical Ventilation	Patient number (percentage)	3 (2%)	3 (3.9%)	0 (0%)	0.088
CT Imaging	Ground-glass opacity	134 (89.3%)	71 (92.2%)	63 (86.3%)	0.241
	Pulmonary consolidation	107 (71.3%)	60 (77.9%)	47 (64.4%)	0.067
	Infected lobes (median, range)	4 (0, 5)	4 (0, 5)	4 (0, 5)	0.211
Discharged	Within two weeks (number, percentage)	41 (27.3%)	26(33.8%)	15 (20.5%)	0.069

(Table 1) contd.....

Demographic Characteristics	All Patients (n=150)	Imported (n=77)	Locally Infected (n=73)	P- value
Length of hospitalization (median days, range)	21 (3, 55)	18 (3, 55)	23 (7, 40)	0.093

HBP: High Blood Pressure; WBC: White Blood Cell; CRP: C-Reactive Protein; \* ages were presented as median (25<sup>th</sup>-75<sup>th</sup> percentile); \*\* statistically significant.

**Table 2. Comparative analysis of COVID-19 severity of imported and locally infected groups.**

Disease Severity (Patient Number)	All Patients (n=150)	Imported (n=77)	Locally Infected (n=73)	Chi-Square Tests
Severe	13 (8.7%)	9 (11.7%)	4 (5.5%)	-
Critical	3 (2.0%)	3 (3.9%)	0 (0%)	-
Combined	16 (10.7%)	12 (15.6%)	4 (5.5%)	$\chi^2=4.016, P=0.045$

**Table 3. Multivariable analysis of predictors of infection severity.**

-	Multivariable OR (95%CI)	P-value
Importation from Wuhan	3.852(0.877-16.925)	0.074
Gender	4.374(1.053-18.163)	0.042
Age	1.098(1.032-1.169)	0.003
Cough	2.499(0.489-12.763)	0.271
Length of hospitalization	1.059(0.995-1.128)	0.072
Pulmonary consolidation	1.144(0.262-4.992)	0.858
Constant	0.000(0.000-0.000)	0.000

**Table 4. Multivariable analysis of predictors of lowered lymphocyte counts.**

-	Multivariable OR (95%CI)	P-value
Importation from Wuhan	2.513(1.080-5.849)	0.033
Gender	2.076(0.904-4.767)	0.085
Age	1.047(1.011-1.085)	0.010
Cough	0.454(0.194-1.061)	0.068
Length of hospitalization	1.056(1.011-1.103)	0.015
Pulmonary consolidation	3.046(1.138-8.158)	0.027
Constant	0.005(0.005-0.005)	0.000

Very few clinical studies have explored the changes in disease severity following infection migration or transmission [16, 20]. Conclusions from these studies were also compromised by comparing patient groups with unparallel demographic characteristics [16, 20]. The unique COVID-19 patient cohort hospitalized in Wenzhou allowed us to avoid demographic differences across different patient groups that may have affected the conclusions of other studies; such as the difference in age [20], time between symptom onset to hospitalization [13], patients' residence status [21], and underlying diseases. A fair comparison of imported or locally acquired COVID-19 by our group provided solid clinical evidence to support the viral attenuation theory after passages of SARS-CoV-2 during the early stages of the pandemic. The declined clinical severity after viral passages was also supported by our laboratory-based finding that a significantly higher proportion of lymphopenia cases presented in the imported COVID-19 group in comparison with the locally acquired COVID-19 group; lymphopenia has been associated with poor outcome of COVID-19, as found in this study and also by others [22]. In addition to viral passage, age and gender

were found to have a significant influence on the severity of COVID-19. Others have reported age and gender as independent risk factors for severe respiratory failure and high mortality rate of COVID-19 [23].

The rapidly changing epidemiology and transmission dynamics of the COVID-19 outbreak beyond Wuhan during the early stages of the pandemic has been reported [13]. COVID-19 diagnosed in Wenzhou shared similar characteristics with infections reported elsewhere in China with the exception of Wuhan. Infections remain highly communicable and relatively mild, with significantly lower mortality rates relative to those initially reported in Wuhan [7, 13, 24]. COVID-19 death rates in Wuhan also varied, with the first clinical study reporting 15% of mortality [1], and later studies reporting 11%, 4.3%, and 1.4% mortality respectively [5, 7, 25]. It was suspected that viral attenuation may have occurred during the early stage of the epidemic in Wuhan. Among the 150 COVID-19 cases in Wenzhou, 10.7% were classified as severe or critical infections, lower than 25.5% of the first 41 published cases linked to the Huanan Seafood

Wholesale Market [1] and 15.7-18.5% of Wuhan local patients reported by a large-scale retrospective study [8]. It is not surprising that SARS-CoV-2 may tune its infectiousness and virulence, as a part of its natural evolutionary and survival strategies. Others using *in vitro* cell-culture assays and *in vivo* animal models have demonstrated viral attenuation of SARS-CoV-2 that may lead to increased transmissibility and persistence in the host [2, 26 - 28]. Young and colleague analyzed PCR-confirmed COVID-19 clinical samples from Singapore and identified SARS-CoV-2 variants with a 382-nucleotide deletion in its genome [20]. Such genetic variation has been linked to lowered viral virulence and less severe infections, possibly by up-regulating T-cell activation-associated cytokine and down-regulating lung injury-associated growth factors [20].

The treatment of COVID-19 patients in Wenzhou during the early stages of the pandemic was empirical and experimental. We, like other teams in the world, combined antiviral treatment with supporting therapies. We followed the Chinese Center for Disease Control and Prevention's guidelines, and used broad-spectrum antibiotics for COVID patients with an immunodeficient status to prevent opportunistic bacterial infections.

No COVID-19 related death was reported at either of the two study centers in Wenzhou. In addition to an attenuation of COVID-19 severity, this may also have been due to early detection of the infections, adequate medical support in place, and few old patients (> 65 years old) admitted to both hospitals [29, 30]. Lack of death-related COVID cases in this study suggested that the spectrum of the severity of COVID-19 in Wenzhou at the early stage of the pandemic might not fully reflect cases reported elsewhere including the initial epicenter Wuhan, where COVID-19 related deaths were frequently reported [7, 13, 24]. Nevertheless, the selection bias possibly stemming from recruiting few old patients may only have minimum impact on our findings as two classified groups of parallel demographic characteristics, including age and age distributions, were compared in this study.

We cannot deny that this study is subject to limitations associated with the nature of retrospective studies, as well as a relatively small sample size. Another evident limitation is that RT-PCR rather than viral genome sequencing was used for the diagnosis of COVID-19; bioinformatic analysis of viral genome sequences may allow a direct computational comparison of SARS-CoV-2 strains causing infections in Wuhan and Wenzhou. The analysis otherwise will assist the confirmation of our clinically proposed classification of COVID-19, as either locally acquired or imported infections.

## CONCLUSION

The spread of COVID-19 from Wuhan to Wenzhou appeared to be mostly mild at the early stage of the pandemic. Although locally acquired COVID-19 was similar to infections imported from Wuhan in the context of clinical manifestations, they were less likely to progress to severe conditions, suggesting a possibility of viral attenuation following transmission.

## LIST OF ABBREVIATIONS

<b>COVID-19</b>	= Coronavirus Disease 2019
<b>CT</b>	= Computed Tomography
<b>SARS-CoV-2</b>	= The Severe Acute Respiratory Syndrome Coronavirus 2
<b>ARDS</b>	= Acute Respiratory Distress Syndrome
<b>ECMO</b>	= Extracorporeal Membrane Oxygenation
<b>ACE2</b>	= Angiotensin Converting Enzyme 2
<b>RT-PCR</b>	= Reverse Transcription Polymerase Chain Reaction
<b>RR</b>	= Respiratory Rate
<b>X2 test</b>	= Chi-squared test
<b>SARS-CoV</b>	= Severe Acute Respiratory Syndrome Coronavirus
<b>MERS-CoV</b>	= Middle East Respiratory Syndrome Coronavirus.

## AUTHORS' CONTRIBUTIONS:

FS, XK and YQ designed the study. SF, YS and LH collected and analyzed clinical data. CW, FC, Y L, CH carried out statistical analysis. YQ and FS wrote the manuscript. All authors read and approved the final manuscript.

## ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This study was approved by the Ethics Review Boards of Wenzhou Central Hospital (Approval No. 202002232214000162765) and Ryan People's Hospital (Approval No. YJ2020013).

## HUMAN AND ANIMAL RIGHTS

No animals were used in this research. All human research procedures followed were in accordance with the ethical standards of the committee responsible for human experimentation (institutional and national), and with the Helsinki Declaration of 1975, as revised in 2013.

## CONSENT FOR PUBLICATION

The studied participants were informed about the present research. A written consent for publication was obtained from patient.

## AVAILABILITY OF DATA AND MATERIALS

The data supporting the findings of the article is available from corresponding author [F.S] upon reasonable request.

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## CONFLICT OF INTERESTS

The authors declare no conflict of interest, financial or otherwise.

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## REFERENCES

- [1] Huang C, Wang Y, Li X, *et al.* Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* 2020; 395(10223): 497-506. [http://dx.doi.org/10.1016/S0140-6736(20)30183-5] [PMID: 31986264]
- [2] Wang C, Horby PW, Hayden FG, Gao GF. A novel coronavirus outbreak of global health concern. *Lancet* 2020; 395(10223): 470-3. [http://dx.doi.org/10.1016/S0140-6736(20)30185-9] [PMID: 31986257]
- [3] Lancet T. Emerging understandings of 2019-nCoV. *Lancet* 2020; 395(10221): 311. [http://dx.doi.org/10.1016/S0140-6736(20)30186-0] [PMID: 31986259]
- [4] Zhang MQ, Wang XH, Chen YL, *et al.* Clinical features of 2019 novel coronavirus pneumonia in the early stage from a fever clinic in Beijing. *Zhonghua jie he he hu xi za zhi= Zhonghua jiehe he huxi zazhi= Chinese journal of tuberculosis and respiratory diseases* 2020; 43(3): 215-8.
- [5] Wang D, Hu B, Hu C, *et al.* Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus-infected pneumonia in Wuhan, China. *JAMA* 2020; 323(11): 1061-9. [http://dx.doi.org/10.1001/jama.2020.1585] [PMID: 32031570]
- [6] Heymann DL. Data sharing and outbreaks: Best practice exemplified. *Lancet* 2020; 395(10223): 469-70. [http://dx.doi.org/10.1016/S0140-6736(20)30184-7] [PMID: 31986258]
- [7] Guan WJ, Ni ZY, Hu Y, *et al.* Clinical characteristics of coronavirus disease 2019 in China. *N Engl J Med* 2020; 382(18): 1708-20. [http://dx.doi.org/10.1056/NEJMoa2002032] [PMID: 32109013]
- [8] Zhu N, Zhang D, Wang W, *et al.* A novel coronavirus from patients with pneumonia in China, 2019. *New Eng Medicine* 2020. [http://dx.doi.org/10.1056/NEJMoa2001017]
- [9] Hu B, Qiu J, Chen H, Tao V, Wang J, Lin H. First, second and potential third generation spreads of the COVID-19 epidemic in mainland China: An early exploratory study incorporating location-based service data of mobile devices. *Int J Infect Dis* 2020; 96: 489-95. [http://dx.doi.org/10.1016/j.ijid.2020.05.048] [PMID: 32425632]
- [10] Lan J, Ge J, Yu J, *et al.* Structure of the SARS-CoV-2 spike receptor-binding domain bound to the ACE2 receptor. *Nature* 2020; 581(7807): 215-20. [http://dx.doi.org/10.1038/s41586-020-2180-5] [PMID: 32225176]
- [11] Xu Z, Shi L, Wang Y, *et al.* Pathological findings of COVID-19 associated with acute respiratory distress syndrome. *Lancet Respir Med* 2020; 8(4): 420-2. [http://dx.doi.org/10.1016/S2213-2600(20)30076-X] [PMID: 32085846]
- [12] Carsana L, Sonzogni A, Nasr A, *et al.* Pulmonary post-mortem findings in a series of COVID-19 cases from northern Italy: A two-centre descriptive study. *Lancet Infect Dis* 2020; 20(10): 1135-40. [http://dx.doi.org/10.1016/S1473-3099(20)30434-5] [PMID: 32526193]
- [13] Zhang J, Litvinova M, Wang W, *et al.* Evolving epidemiology and transmission dynamics of coronavirus disease 2019 outside Hubei province, China: A descriptive and modelling study. *Lancet Infect Dis* 2020; 20(7): 793-802. [http://dx.doi.org/10.1016/S1473-3099(20)30230-9] [PMID: 32247326]
- [14] Chan JF, Yuan S, Kok KH, *et al.* A familial cluster of pneumonia associated with the 2019 novel coronavirus indicating person-to-person transmission: a study of a family cluster. *Lancet* 2020; 395(10223): 514-23. [http://dx.doi.org/10.1016/S0140-6736(20)30154-9] [PMID: 31986261]
- [15] Wei M, Yuan J, Liu Y, Fu T, Yu X, Zhang ZJ. Novel coronavirus infection in hospitalized infants under 1 year of age in China. *JAMA* 2020; 323(13): 1313-4. [http://dx.doi.org/10.1001/jama.2020.2131] [PMID: 32058570]
- [16] Gao Y, Shi C, Chen Y, *et al.* A cluster of the Corona Virus Disease 2019 caused by incubation period transmission in Wuxi, China. *J Infect* 2020; 80(6): 666-70. [http://dx.doi.org/10.1016/j.jinf.2020.03.042] [PMID: 32283165]
- [17] Gaeta G. Social distancing versus early detection and contacts tracing in epidemic management. *Chaos Solitons Fractals* 2020; 140110074 [http://dx.doi.org/10.1016/j.chaos.2020.110074] [PMID: 32834628]
- [18] Ahmadiwand A, Gerislioglu B, Ramezani Z, Kaushik A, Manickam P, Ghoreishi SA. Functionalized terahertz plasmonic metasensors: Femtomolar-level detection of SARS-CoV-2 spike proteins. *BiosenBioelect* 2021; 177112971 [PMID: 33434777]
- [19] Kevadiya BD, Machhi J, Herskovitz J, *et al.* Diagnostics for SARS-CoV-2 infections. *Nat Mater* 2021; 20(5): 593-605. [http://dx.doi.org/10.1038/s41563-020-00906-z] [PMID: 33589798]
- [20] Young BE, Fong SW, Chan YH, *et al.* Effects of a major deletion in the SARS-CoV-2 genome on the severity of infection and the inflammatory response: An observational cohort study. *Lancet* 2020; 396(10251): 603-11. [http://dx.doi.org/10.1016/S0140-6736(20)31757-8] [PMID: 32822564]
- [21] Price-Haywood EG, Burton J, Fort D, Seoane L. Hospitalization and mortality among black patients and white patients with COVID-19. *N Engl J Med* 2020; 382(26): 2534-43. [http://dx.doi.org/10.1056/NEJMsa2011686] [PMID: 32459916]
- [22] Tavakolpour S, Rakhshandehroo T, Wei EX, Rashidian M. Lymphopenia during the COVID-19 infection: What it shows and what can be learned. *Immunol Lett* 2020; 225: 31-2. [http://dx.doi.org/10.1016/j.imlet.2020.06.013] [PMID: 32569607]
- [23] Novelli L, Raimondi F, Ghirardi A, *et al.* At the peak of COVID-19 age and disease severity but not comorbidities are predictors of mortality. *COVID-19 burden in Bergamo, Italy Panminerva Med* 2020.
- [24] Tian S, Hu N, Lou J, *et al.* Characteristics of COVID-19 infection in Beijing. *J Infect* 2020; 80(4): 401-6. [http://dx.doi.org/10.1016/j.jinf.2020.02.018] [PMID: 32112886]
- [25] Wu JT, Leung K, Bushman M, *et al.* Estimating clinical severity of COVID-19 from the transmission dynamics in Wuhan, China. *Nat Med* 2020; 26(4): 506-10. [http://dx.doi.org/10.1038/s41591-020-0822-7] [PMID: 32284616]
- [26] Caccuri F, Zani A, Messali S, *et al.* A persistently replicating SARS-CoV-2 variant derived from an asymptomatic individual. *J Transl Med* 2020; 18(1): 362. [http://dx.doi.org/10.1186/s12967-020-02535-1] [PMID: 32967693]
- [27] Lau SY, Wang P, Mok BW, *et al.* Attenuated SARS-CoV-2 variants with deletions at the S1/S2 junction. *Emerg Microbes Infect* 2020; 9(1): 837-42. [http://dx.doi.org/10.1080/22221751.2020.1756700] [PMID: 32301390]
- [28] Armengaud J, Delaunay-Moisan A, Thuret JY, *et al.* The importance of naturally attenuated SARS-CoV-2 in the fight against COVID-19. *Environ Microbiol* 2020; 22(6): 1997-2000. [http://dx.doi.org/10.1111/1462-2920.15039] [PMID: 32342578]
- [29] Mody L, Cinti S. Pandemic influenza planning in nursing homes: Are we prepared? *J Am Geriatr Soc* 2007; 55(9): 1431-7. [http://dx.doi.org/10.1111/j.1532-5415.2007.01299.x] [PMID: 17767687]
- [30] Keita M, Camara AY, Traoré F, *et al.* Impact of infection prevention and control training on health facilities during the Ebola virus disease outbreak in Guinea. *BMC Public Health* 2018; 18(1): 547. [http://dx.doi.org/10.1186/s12889-018-5444-3] [PMID: 29699538]