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Therapeutic Outcomes of an Herbal Remedy in Treating and Preventing African Swine Fever: A Real-World Evidence Report

Zhenghua Ma[1], Jiangnan Feng[1*], Chengsheng Yi[1], Xia Liu[1], Dengbin Zhu[1], Minghai Ma[1], Xue Feng[1], Yu Yan[1] and Meiying Gao[2]

[1]Tongrun Tang TCM Clinic, Nanchong City, Sichuan, China

[2]Department of Medicine, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, Hubei, China

ABSTRACT

Backgound: African swine fever (ASF) is a highly contagious viral disease characterized by a 100% case fatality rate in domestic pigs, for which there is currently no effective treatment. Recent studies suggest that Marecipe AV, an herbal therapeutic may provide significant therapeutic benefits against severe viral infections.

Methods: This study involved treating domestic pigs diagnosed with African swine fever (ASF) or exposed to the African swine fever virus (ASFV) with Marecipe AV herbal medicine. Mortality rates were recorded in both the treated group and an untreated control group to evaluate efficacy.

Results: Oral administration of Marecipe AV herbal medicine significantly reduced mortality in infected domestic pigs, with rates declining from 100% in the control group to between 0% and 28.6% in treated animals ($P < 0.001$). All survivors exhibited full clinical recovery and viral clearance. Furthermore, prophylactic treatment afforded 100% protection against ASF challenge, resulting in 0% mortality and 0% disease incidence, compared to 100% in the untreated control group.

Conclusion: Marecipe AV herbal therapeutics has shown potent effectiveness in both treating and preventing African swine fever in domestic pigs, achieving complete containment of the disease.

***Corresponding author**

Jiangnan Feng, Tongrun Tang TCM Clinic, Nanchong City, Sichuan, China.

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Introduction

African swine fever (ASF) is an acute, virulent, common infectious disease of domestic and wild pigs caused by the African swine fever virus (ASFV), with morbidity and mortality rates reaching 100 percent. Due to the lack of safe and effective vaccines and antiviral drugs, culling is the most effective method to control the epidemic [1-4].

Feng and colleagues recently described a potent therapeutic strategy involving an herbal remedy for viral infections. It has been demonstrated that Marecipe AV therapeutics can reduce mortality rates from nearly 100% to nearly 0% for several high-mortality diseases in animals, including avian influenza, canine distemper, canine parvovirus, porcine reproductive and respiratory syndrome. The herbal therapy, known as Marecipe AV, originated from an ancient herbal prescription used to treat venomous snake bites [5,6]. Marecipe AV is formulated from powdered extracts of four botanical species and a rare freshwater fish. It has identified by Mass spectrometry analysis the principal components of the Marecipe AV compound as betaine (content: 22.34%), rosmarinic acid (content: 17.41%), isoorientin (content: 11.41%), linolenic acid (content: 8.827%), caffeic acid (content: 4.54%), cichoric acid (content: 4.34%), sucrose (content: 4.08%) , Vizenin III (content: 2.30%) , Citric acid (content: 2.02%) and Azelaic Acid (content: 1.82%) [7-9]. A search of the scientific literature revealed no correlation between these components and antiviral activity. The potent therapeutic activity and curative-level efficacy demonstrated by the Marecipe AV herbal therapy suggest a distinct antiviral mechanism of action. This study reports real-world outcomes of the Marecipe AV therapy for the treatment and prevention of African swine fever (ASF).

Methods

Study Design: This study documented evidence of Marecipe AV herbal therapy for the treatment and prevention of African Swine Fever (ASF) in a real-world setting. Since mortality is nearly 100% in swine infected with ASF and is the only non-modifiable outcome, it serves as a definitive measure of efficacy, eliminating the need for a control group treated with other drugs. Nevertheless, to enhance the reliability of the findings, a control group was still established. Mortality rates in both the treated and untreated control groups were recorded for efficacy analysis.

Subject's: Domestic pigs from an ASF epidemic area were selected for this study. Based on prior breeding experience, it was anticipated that these pigs would inevitably contract ASF. In compliance with relevant management regulations, all domestic pigs used in the experiments were euthanized and buried after the study was completed.

Interventions and Procedures

The confirmation of ASF was achieved by detecting ASFV in nasopharyngeal swabs using quantitative polymerase chain reaction (qPCR). ASFV infection was confirmed by detecting serum antibodies against ASFV through enzyme-linked immunosorbent assay (ELISA). The primary endpoint and evaluation indicator was mortality. A comparison of mortality rates, such as incidence rates, between the treatment and control groups allows for the assessment of therapeutic efficacy.

Therapeutic and Preventive Intervention Protocols: Domestic pigs received an oral loading dose of Marecipe AV at 0.2 grams per kilogram of body weight, administered twice daily for 5 to 7 days. Administration methods included artificial gastric instillation and self-drinking water mixed with the Marecipe AV soaking solution. The Procedure of Trial 1: In the first trial, domestic pigs diagnosed with ASF and housed in the same pigpens were randomly divided into two equal groups. The treatment group received Marecipe AV herbal medicine via artificial oral gavage, while the placebo group did not receive any treatment. The mortality of domestic pigs was recorded from 0 to 15 days post-administration.

The Procedure of Trial 2: ASFV-infected domestic pigs in the early stages of disease progression (characterized by their ability to self-drink water independently) were randomly allocated into two experimental groups. ASF confirmation was achieved by detecting ASFV. In this trial, the treatment group was administered Marecipe AV herbal medicine, prepared as an immersion extract and incorporated into the drinking water, allowing the subjects to consume it through self-drinking. Conversely, the control group received no intervention. The treatment was carried out over a period of 15 days. Mortality was monitored for a period of 0 to 60 days post-infection (PID).

The Procedure of Trial 3: The dosing regimen in this trial was identical to that in Trial 2; however, the treatment duration was reduced from 15 days to 9 days. Administration occurred twice daily from days 1 to 6 and once daily from days 7 to 9. Virus detection and serum antibody assessments were conducted before and after administration.

The Procedure of Trial 4: The subjects consisted of healthy domestic pigs sourced from ASF-affected areas that exhibited no symptoms of ASF. For the treatment group, the intervention included daily oral administration of Marecipe AV herbal medicine and daily disinfection with a Marecipe AV soaking solution. This intervention lasted for 30 days. In contrast, the control group received no intervention. Throughout the 60-day observation period, the presence of clinical signs of African swine fever (ASF), the virus itself (ASFV), and specific serum antibodies against ASFV was monitored and recorded in the animals.

The Procedure of Trial 5: This trial comprised two parts. First, the soaking solution of the herbal preparation was used as a disinfectant. Second, in addition to its use as a disinfectant spray, the preparation was also administered orally. The trial was conducted on piglets housed in sties considered at high risk for ASFV infection, as these facilities had recently housed pigs that succumbed to ASF and had been emptied only days prior. Starting three days before piglet introduction and continuing through day 20, the intervention group's pigsties were disinfected once daily with Marecipe AV soaking solution. Nasopharyngeal swabs (n=10 per sty) were collected for ASFV detection on days 0 and 20, alongside close monitoring and recording of clinical signs and mortality to determine ASF incidence.

During days 20–40, all piglets received daily oral Marecipe AV treatment alongside continued environmental disinfection. Throughout the following 60 days, clinical signs and body weight were monitored, and samples were tested for ASFV and specific serum antibodies.

Statistical analysis: Variance analysis was conducted using the Statistical Package for the Social Sciences (SPSS) version 27 for Windows.

Ethics: In this study, the treatment protocol was designed and implemented in accordance with standard clinical practices of Traditional Chinese Medicine (TCM). Prescriptions were formulated by licensed TCM practitioners and subsequently dispensed by authorized pharmacies. The protocol adhered to relevant TCM regulations and ethical guidelines. Although the methods and preparations complied with established TCM norms, the study protocol was submitted for ethical review and approved by the TCM Ethics Committee of Tongren Tang Clinic (Approval No. 202101). As the study did not involve experimental animals, approval from an institutional animal care and use committee was not required.

Results

A total of five trials were conducted to Increase the reliability of the evidence and to evaluate the efficacy of Marecipe AV in the treatment and prevention of ASF.

In the first trial, a cohort of 30 ASFV-infected domestic pigs exhibiting severe clinical symptoms, including cessation of active eating, was established. Fourteen randomly selected subjects received Mareciep AV via oral gavage, while the remaining 16 served as untreated controls within the same facility. Additionally, 60 swine housed in nearby backyard pigpens within the same epidemiological unit were monitored as an external control group. Viral detection was performed using qPCR on a subset of randomly selected swine that displayed clinical signs consistent with ASF. All specimens (100%, n=3) tested positive for the ASFV. On day 15 after the initial dose of Mareciep AV, the mortality rates in the treatment and internal control groups were 28.57% (4/14) and 100% (16/16), respectively, while all 60 swine (100%) as an external control group succumbed to ASF. The Fisher exact test analysis revealed a significant difference in mortality rates between the treated and untreated groups (28.57% [4/14] vs. 100% [16/16], $P < 0.001$).

Administering drugs to domestic pigs via gavage presents significant challenges, even in slow-moving pigs exhibiting severe symptoms of ASF. Experimental operators have estimated that the success rate of gavage in these domestic pigs is less than 50%. Consequently, a substantial number of pigs involved in these studies did not receive an adequate dosage of Marecipe AV. This situation resulted in a significant deviation in the treatment outcome.

In the secondary trial, 63 ASFV-infected domestic pigs in the early stages of disease progression (characterized by their ability to self-drinking) were randomly allocated into a treatment group

and a control group. By post-infection day (PID) 15, the treated group exhibited 0% mortality (0/30) compared to 100% lethality (33/33) in the control group. Longitudinal monitoring via PID 60 confirmed complete clinical recovery in all survivors, with sustained virological remission maintained, as confirmed by qPCR. The Fisher exact test revealed a significant difference in mortality rates between the treatment and control groups. The administration of Marecipe AV herbal therapy reduced the mortality rate of ASF in domestic pigs from 100% to 0%. This result is astonishing. The finding of zero fatalities attributed to African swine fever suggests that it may even challenge current scientific experimentation standards.

In Trial 3, a total of 3,000 domestic pigs affected by the ASF epidemic were included. The treatment group comprised 300 domestic pigs, approximately four months old, housed in a single pigpen, while 200 domestic pigs were closely observed as a control group. The remaining 2,500 pigs in the same village were also monitored as a control group. Prior to the intervention, all 50 nasopharyngeal swab samples obtained from 500 domestic pigs were detected for ASFV by qPCR. During the seven-day treatment period, three pigs in the treatment group died. This was because they developed severe symptoms and were unable to drink water independently. As a result, they were unable to receive drug treatment. Consequently, these pigs were not included in the mortality count. No additional deaths occurred in the treatment group during the subsequent 60-day observation period. In contrast, all 200 domestic pigs in the control group and all 2,500 pigs in the village succumbed to ASF within the same timeframe□Among an estimated 1,500 pigs were subsequently culled.) Thus, the mortality rate was 0% in the treatment group and 100% in the control group.

Fever subsided within two days following the initial administration of Marecipe AV in all treated pigs. Feed intake returned to normal in the majority of domestic pigs by the fifth to seventh day post-treatment. Most pigs in the treatment group achieved full recovery by the ninth day after the initial dose. Each time, 10 samples from the treatment group were tested for ASFV using qPCR on days 15, 45, and 60 following the initial treatment, and none were found to contain ASFV. Antibody testing for ASFV was conducted 60 days post-treatment, with all serum samples testing positive, indicating that the surviving domestic pigs had been infected with ASFV.

In Trial 4, a total of 300 three-month-old domestic pigs were housed in pigpen A in a village experiencing an outbreak of ASF, while 200 domestic pigs were housed in pigpen B as a control group. During the observation period from days 15 to 60, all pigs in pigpen B exhibited ASF-like symptoms, and all 200 pigs succumbed. In contrast, the 300 domestic pigs in pigpen A reported no fatalities or symptoms of ASF. The mortality rates for the preventive treatment group and the control group were 0% and 100%, respectively.

ASFV was not detected in any of the 30 samples collected from pigpen A after the 60-day intervention. Ten serum samples were collected for ASFV antibody detection; two samples tested positive, while eight were negative. Exposure to African swine fever virus resulted in a 20% infection rate in the domestic pigs.

In Trial 5, a total of 2,000 piglets were allocated to distinct pigpens. Three hundred 60-day-old piglets were randomly assigned to pigpens C and D, while the remaining 1,700 piglets were distributed across different pens within the same village. Before the piglets were placed in the pigsty, three surface samples from each pigpen in the village were collected and tested for ASFV, with all samples returning positive results. Nasopharyngeal swabs from 10 randomly selected piglets from each pigpen were tested for ASFV, and all results were negative. After being housed in pigpens for 15 to 20 days, more than half of the piglets in pens C and D displayed clinical signs consistent with ASF. Seven piglets succumbed to the disease. Nasopharyngeal swabs were collected from 10 piglets in pens C and D, and all samples tested positive for ASFV. At the same time point, 7 out of the remaining 1700 piglets developed symptoms of ASF. Nasopharyngeal swabs were collected from these 7 piglets, resulting in 5 out of the 7 samples testing positive for ASFV. Twenty days after the introduction of the piglets into the pigsty, the incidence rate in the Marecipe AV herbal medicine disinfection group was 0.41%, compared to approximately 50% in the conventional disinfectant group, indicating a statistically significant difference between the two treatments. After the implementation of oral Marecipe AV prophylaxis, there were no new cases of ASF or associated deaths among the piglets, including those in pens C and D. During the four months after the piglets were placed in the pens, the weight gain of the 1,700 piglets remained within the normal range, with no instances of abnormal weight gain observed. Serum antibody testing for ASFV was conducted 60 days after the initiation of oral Marecipe AV treatment. All 10 samples from pigpens C and D tested positive for ASFV antibodies. In contrast, among 1,700 piglets from various pens, 195 tested negative and 5 tested positive. These results showed that all piglets in pens C and D were infected with ASFV, whereas most pigs receiving Marecipe AV prophylaxis either remained uninfected or cleared the virus before mounting a detectable antibody response. Based on this analysis, the incidence of ASF (i.e., the infection rate of ASFV) was approximately 2.5% in the Marecipe AV group, compared to 100% in the control group.

Discussion

In the case of ASF, death remains the only definitive outcome of infected swine. Given that no intervention can alter the nearly 100% fatality rate associated with this disease, utilizing changes in mortality as endpoints to assess therapeutic effects ensures reliable conclusions.

Marecipe AV herbal therapeutics has demonstrated significant efficacy in treating African Swine Fever, achieving a 100% recovery rate with no mortality reported when administered during the early stages of infection. The results indicate that no mortality was observed in two independent trials (Trial 2 and Trial 3) involving ASFV-infected swine that were still able to drink water voluntarily. In three independent large-scale trials (Trials 2, 3, and 4), the administration of Marecipe AV herbal medicine as a prophylactic measure resulted in zero mortality among swine exposed to ASFV. The mortality rate was 28.57% in the treatment of swine infected with ASFV that were unable to drink water voluntarily. Although this is already a quite good result for the treatment of ASF, the actual situation should have a much lower mortality rate. This higher mortality rate result may be attributed to the fact that none of the pigs in the treatment group received a sufficient dosage of the medication. Based on five independent therapeutic outcomes, Marecipe AV herbal medicine has demonstrated highly satisfactory efficacy in combating African Swine Fever, achieving complete containment of the disease.

Marecipe AV herbal therapy exhibits potent pharmacological activity against ASF. The results show that fever subsided within two days of the initial administration of Marecipe AV in all treated pigs. Following treatment, feed intake normalized within 5–7 days

for most pigs and by day 9 for all, even in those exhibiting severe clinical symptoms initially. The weight gain of piglets treated was no distinct deviations in the growth curve.

Marecipe AV prophylaxis has demonstrated significant efficacy in preventing African swine fever (ASF). In ASFV-challenged settings, most piglets receiving Marecipe AV prophylaxis showed no clinical symptoms of ASF, whereas all piglets in the control group were confirmed ASFV-infected. These results suggest that Marecipe AV prophylaxis can fully protect domestic pigs from ASFV infection in exposed environments. Moreover, the growth performance of treated pigs remained completely unaffected following Marecipe AV administration. Collectively, these findings indicate that Marecipe AV prophylaxis achieves complete containment of ASF.

In an evaluation of Marecipe AV herbal medicine as a disinfectant, clinical signs of African swine fever (ASF) appeared in more than half of the 300 piglets within 15–20 days of rearing. In comparison, only 7 out of 1,700 piglets treated with the Marecipe AV disinfectant developed the disease after 20 days. Serum antibody testing revealed that all domestic pigs in the untreated group were infected with ASFV, while only 2.5% of the treated group showed signs of infection. These data suggest that Marecipe AV, as a disinfectant, has a preventive effect on domestic pigs in an ASFV exposure environment. However, the Marecipe AV herbal medicine, which is used as a disinfectant, has no practical value against ASF. Regarding ASF, once infected pigs are present within a herd, the transmission of ASFV to other healthy pigs becomes unavoidable.

At least two studies have reported the results of long-term follow-up on convalescent pigs and their offspring. There were neither carriers of nor recurrence of disease in the convalescent pigs and their offspring following the outbreak of acute ASF. The infectious ASFV was not detected in any of the tissue samples from ASFV convalescent and in-contact pigs [10,11]. In the present study, no ASFV pathogens were detected in any of the convalescent pigs. From a biosafety perspective, cured domestic pigs should pose minimal risk of ASF transmission.

Limitations

This study has several limitations. It is important to note that these findings are derived from real-world records of Marecipe AV herbal medicine applications in the treatment of ASF, rather than from a prospective clinical trial. The experimental data in this study were obtained under unsupervised conditions. Furthermore, constraints imposed by ASF biosecurity protocols and limited funding have compromised the rigor of experimental design and data collection. However, this is not expected to substantially affect the interpretation of the results, given that African swine fever has a mortality rate approaching 100%. secondly, the specific active components of Marecipe AV and their precise mechanisms of action have not been fully elucidated. In addition, due to the lack of complete in vitro experimental data, the data of Marecipe AV herbal medicine as a disinfectant for ASF has limited reference value. Oral administration did not achieve adequate dosing in any experimental subject. This insufficient dosing directly confounded the outcomes and compromised the evaluation of therapeutic efficacy.

This report demonstrates the efficacy of an herbal therapy for both treating and preventing ASF in real-world settings, establishing a robust evidence base for the effectiveness of Marecipe AV in managing and controlling ASF, thereby bridging a critical gap in the field.

Conclusions

This report provides compelling evidence that Marecipe AV herbal therapy exhibits potent and satisfactory therapeutic efficacy in the treatment and prevention of African swine fever (ASF), achieving complete containment of the disease. Marecipe AV herbal therapy has the potential to be an effective approach to ending ASF.

Zhenghua Ma, Jiangnan Feng and Chengsheng Yi contributed equally to this work

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