Pulmonary Delivery of Favipiravir Formulation Using Soft-Mist Inhaler For COVID-19

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INTRODUCTION

Favipiravir (RNA-dependent RNA polymerase (RdRp) inhibitor) was shown to be insufficient in many clinical trials in oral form[1,2]. In this study, we have proved «Favi» to be effective against SARS-CoV-2 virus, if applied via inhalation. Our study entails:

- ✓ A soluble and stable solution of favipiravir.
- ✓ Good aerodynamic particle characteristics for lung targeting.
- ✓ Antiviral efficiency of the predetermined inhalation dosage in Vero6 cells.
- ✓ Preclinical study of possible toxic effects of favipiravir's repetitive delivery via soft mist spray on lung tissue of healthy rats.

INHALED FAVIPIRAVIR HYPOTHESIS

ORAL 1600 mg

- High number/dosage tablet intake
- Low lung localisation
- Low efficiancy in lungs
- Low patient adherence

INHALATION 2 mg

Soft Mist Inhaler

- Low dosage intake
- High lung localisation
- High efficiancy in lungs
- High patient adharence
- Minimized contamination risk
- Personalized dose adjustment

SOLUBILITY STUDY
ANTIVIRAL ACTIVITY
STABILITY STUDY → **PATENT APPLICATION** → **PRECLINICAL STUDY** → **PHASE II CLINICAL STUDY**

FORMULATION DEVELOPMENT

- ✓ **Challenges of Favipiravir:** Poor solubility and low stability
- ✓ **Favipiravir solution, prepared in PBS and stored protected from light, showed superior stability over that was prepared in normal saline.**

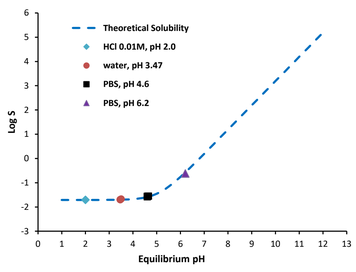


Fig 1. Theoretical pH - Solubility profile of favipiravir, and the solubility values obtained practically in water, HCl 0.01 M, PBS.

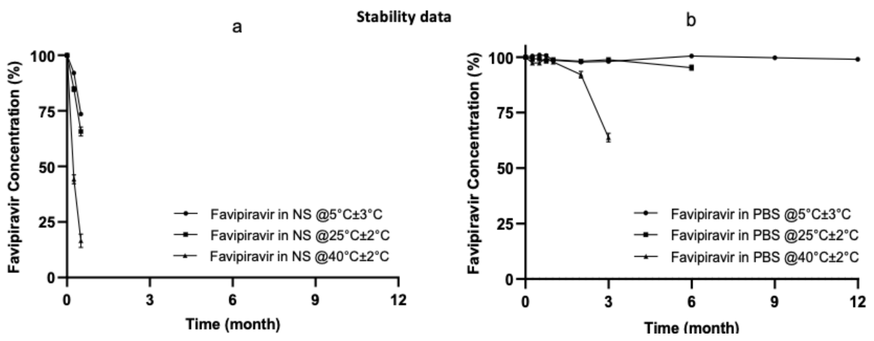


Fig 2. Stability profiles of favipiravir in (a) normal saline and (b) PBS, stored protected from light at 5C ± 3 °C, 25 ± 2 °C, and 40 ± 2 °C [values are expressed as mean ± SD (n = 3), note that the error bars at some points are of the size of the symbols].

- ✓ The MMAD, FPF, GSD values for various favipiravir solutions of NS and PBS were evaluated under NGI.

Table 1. Particle size distribution obtained for favipiravir solutions.

Parameter	Favipiravir in NS	Favipiravir in PBS
MMAD[a] (µm)	5.43 ± 0.32	4.83 ± 0.11
FPF 5 µm[b] (%)	45.63 ± 2.43	52.66 ± 1.67
GSD[c]	1.63 ± 0.07	1.70 ± 0.01

[a] Mean mass aerodynamic diameter.
[b] Fine particle fraction.
c Geometric standard deviation (mean ± SD; n= 3).

IN VITRO ANTIVIRAL ACTIVITY OF FAVIPIRAVIR

- ✓ **Proposition:** Dose estimation for the inhaled «Favi» formulation was calculated as ~2 mg/mL.
- ✓ **Outcome:** The maximum favipiravir activity performed in Vero-6 cells with **xCELLigence Real Time Cell Analyzer RTCA MP** was found at 2 mg/mL and 3 mg/mL.
- ✓ **«Favi» was found to 'antivirally-active' between 2 and 3 mg/mL.**

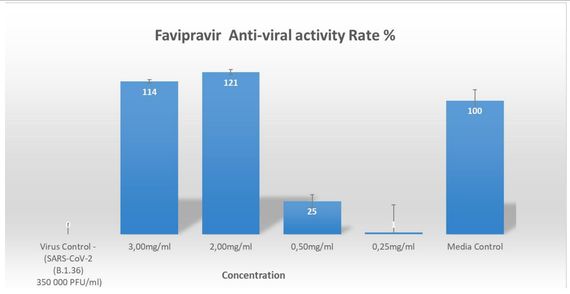


Fig 3. Vero E6 cells, infected with 3.5x 105 PFU mL^{-1} of SARS-CoV-2 (B1.36), were pre-incubated with different concentrations of Favipiravir.

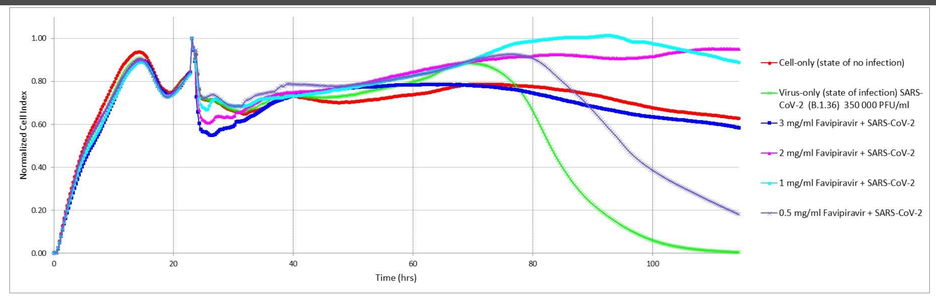
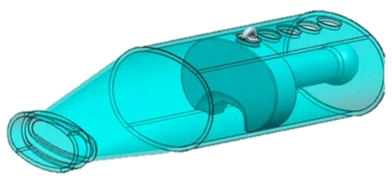


Fig 4. Cell-only (red line) were non-infected. Virus-only Vero E6 cells (green line) were infected that was not pre-exposed to Favipiravir molecule.

PRECLINICAL

STUDY DESIGN

- ✓ First preclinical study* using Pulmospray
- ✓ Design of a **3D cage system** by the team
- ✓ **Favipiravir quantification method**: Development of sensitive HPLC method for plasma and lung tissue.
- ✓ Treatments were given twice a day for 5 consecutive days, and application time was 2 minutes.
- ✓ Wistar albino male and female rats of 8-12 weeks were distributed.



- ✓ **Delivery of favipiravir by soft-mist inhaler did not result in oxidative injury of lung tissue.**
- ✓ **Lung localization achieved a 45-fold higher value compared to plasma.**

*Ethical approval (No: 42.2020.marmara university).

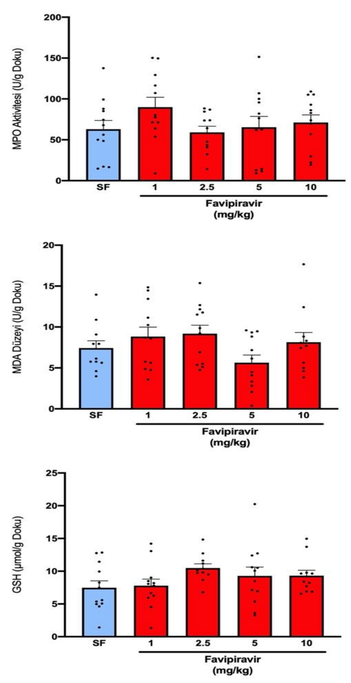


Fig 5. Neutrophil infiltration (MPO activity), lipid peroxidation (MDA) and antioxidant glutathione (GSH) levels in lung tissue of normal saline or favipiravir (1, 2.5, 5 and 10 mg/kg) applied with soft mist inhaler.

RESULTS

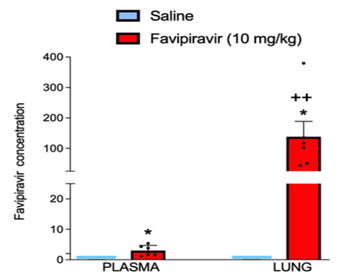


Fig 6. The concentration measured was 140 µg/g lung tissue, which is more than 45-folds compared w/ plasma, showing that the local administration of favipiravir resulted in a higher accumulation in the lung tissue.

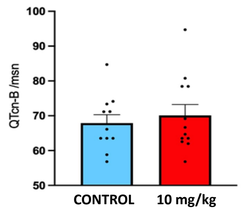


Fig 7. At the 10 mg/kg of favipiravir, calculated QT intervals were not significantly different than those of the saline-treated rats, showing that the highest dose has not resulted in the prolongation of QT interval.

CONCLUSION

Challenges of Favipiravir Resolved

Soluble form of favipiravir → **1 Year stability data** → **Effective at 2 mg/mL** → **High lung localization** → **Safe inhalation application** → **Inhalable favipiravir solution**

This study is currently under revision for Phase II clinical trial.

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