Antiviral activity against SARS-CoV-2

Molecule tested: Nelfinavir

Date: 15/10/2020 Version: 1

Report written by: Antoine NOUGAIREDE, Franck TOURET, Maxime COCHIN

Affiliation: Unité des Virus Émergents (Aix Marseille Univ, IRD 190, INSERM 1207, Marseille, France) headed by Xavier de Lamballerie (Xavier.De-Lamballerie@univ-amu.fr)

Contacts: antoine.nougairede@univ-amu.fr; Franck.touret@univ-amu.fr

Introduction

In response to the recent emergence of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of COVID-19, the Unité des Virus Émergents performed preclinical evaluation of drug candidates against SARS-COV-2. **This interim report has not been peer-reviewed.** The data of this report are available upon request.

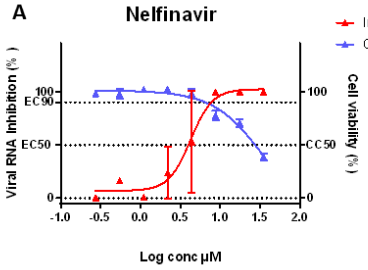
The molecule tested here is the Nelfinavir: a protease inhibitors (PIs) used in the treatment of the human immunodeficiency virus (HIV).

In vitro evaluation of efficacy of Nelfinavir

Nelfinavir was evaluated in our preclinical pipeline due to its possible activity against SARS-CoV-2 viral protease.

Then, the antiviral activity was further characterized from 40 µM to 0.6 µM in Vero-E6 cells using a viral RNA yield reduction assay as previously described (Touret et al., 2019, Touret et al., 2020). Dose-response curves are presented in figure 1A. Nelfinavir showed half maximal effective concentration (EC50) and 90% effective concentration (EC90) of 3.98µM and 7.72µM respectively. Nelfinavir has a half cytotoxic concentration (CC50) of 27.34 µM (Figure 1A). As Nelfinavir was found to be effective against SARS-CoV-2 with a selectivity index (SI) of 6.87, its antiviral activity was evaluated *in vivo*.

A



B

µM	Nelfinavir
EC50	3,98
EC90	7,72
CC50	27,34
SI	6,87

Figure 1: *In vitro* evaluation of Nelfinavir in VeroE6 cells. A: Dose response and cytotoxicity evaluation. B Nelfinavir interpolated antiviral activity values. All assay were performed as previously described (Touret et al., 2020).

In vivo evaluation of efficacy of Nelfinavir

Experiments were approved by the local ethical committee (C2EA—14) and French authorities (APAFIS#23975). Four-week-old female Syrian hamsters were intranasally infected with 10^4 TCID50 of the BavPat1 SARS-CoV2 strain as described previously (Driouich et al., 2020). To assess the efficacy of Nelfinavir, a group of 6 hamsters received the drug, orally, two times a day (BID). We used one dose of Nelfinavir 240mg/kg/day to that we added ritonavir, as a booster, at 84mg/kg/day QD. Treatment was initiated at day of infection, ended at 2 day post infection (dpi) and animals were sacrificed at 3 dpi. A control group of 6 infected/untreated animals received, orally, 0.9% Sodium Chloride solution. Viral replication was measured in lungs (viral RNA yields) and in plasma (viral RNA yields).

Results showed no significant reduction of viral replication in lungs and plasma compared to control group (**Figure 2**).

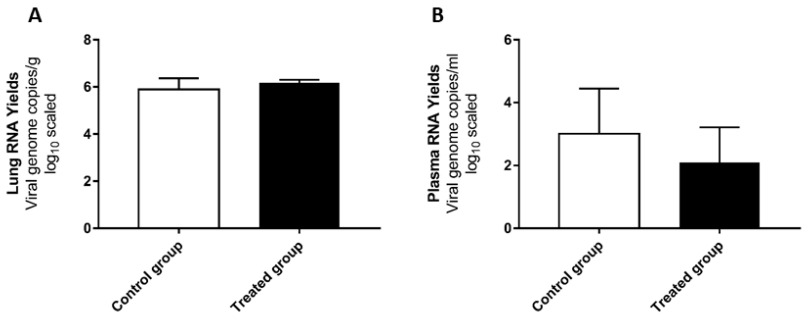


Figure 2: Viral replication in lungs (A) and in plasma (B). Lung and plasma viral load were measured using a viral RNA yields RT-qPCR assay as previously described (Driouich et al., 2020).

Conclusion

Nelfinavir was found to inhibit SARS-CoV-2 replication *in vitro* using Vero-E6 cells. However, results obtained using a hamster model suggest that this molecule is a poor inhibitor of SARS-CoV-2 replication *in vivo.*

References

DRIOUICH, J.-S., COCHIN, M., LINGAS, G., MOUREAU, G., TOURET, F., PETIT, P. R., PIORKOWSKI, G., BARTHÉLÉMY, K., COUTARD, B., GUEDJ, J., DE LAMBALLERIE, X., SOLAS, C. & NOUGAIRÈDE, A. 2020. Favipiravir antiviral efficacy against SARS-CoV-2 in a hamster model. *bioRxiv***,** 2020.07.07.191775.

TOURET, F., BARONTI, C., GOETHALS, O., VAN LOOCK, M., DE LAMBALLERIE, X. & QUERAT, G. 2019. Phylogenetically based establishment of a dengue virus panel, representing all available genotypes, as a tool in dengue drug discovery. *Antiviral Res,* 168**,** 109-113.

TOURET, F., GILLES, M., BARRAL, K., NOUGAIREDE, A., VAN HELDEN, J., DECROLY, E., DE LAMBALLERIE, X. & COUTARD, B. 2020. In vitro screening of a FDA approved chemical library reveals potential inhibitors of SARS-CoV-2 replication. *Sci Rep,* 10**,** 13093.