Anti-Dengue Virus Drug Screening and Mechanistic Study

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Abstract

We choose three pure compounds provided from Dr. Gopinath S. Khasole (drug code: PGN-G-1, PGN-G-10, PGN-G-11) to conduct anti-DENV drug screening via western blot, the result reveals that PGN-G-11 has the potential to become an antiviral drug. Based on the result, we will further perform experiments to find out the molecular mechanism regarding PGN-G-11 against DENV.

Background

Dengue virus is a mosquito-borne Flavivirus which causes the dengue fever, prevalence in global range, the annual incidence close to 390 million, mainly transmit via Aedes aegypti and Aedes albopictus, there are four serotypes of the virus. In this study, we use type 2 dengue virus as our screening target.

Methodology

Western Blot

Take western blot as preliminary screening strategy to test the DENV expression compare between control group and different drug group in protein level.

RT-qPCR

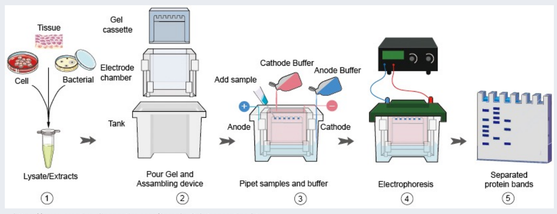
Via western blot, we can have a preliminary result regarding the effective drug. Then take advanced step in RT-qPCR to check on RNA level.

MTS Assay

MTS assay help us to determine the cytotoxicity of drug, ranging cell survival rate with higher drug level.

Reporter Assay

Have a drug with most effective concentration to conduct reporter assay. The exist of DENV in cell will inhibit HO-1 and induce COX-2 expression, after treat cell with drug, the level of HO-1 and COX-2 should recovery.



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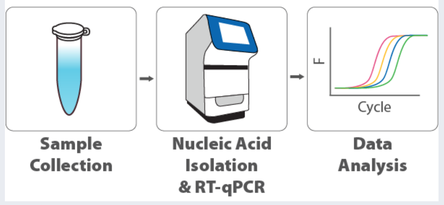

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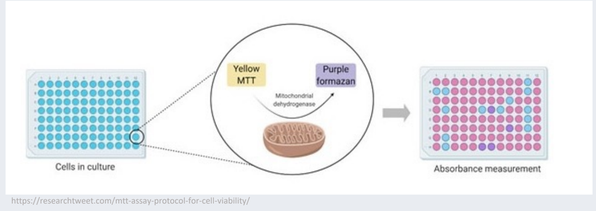

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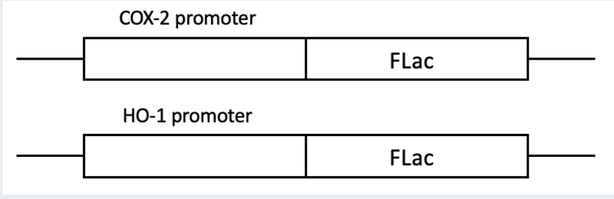
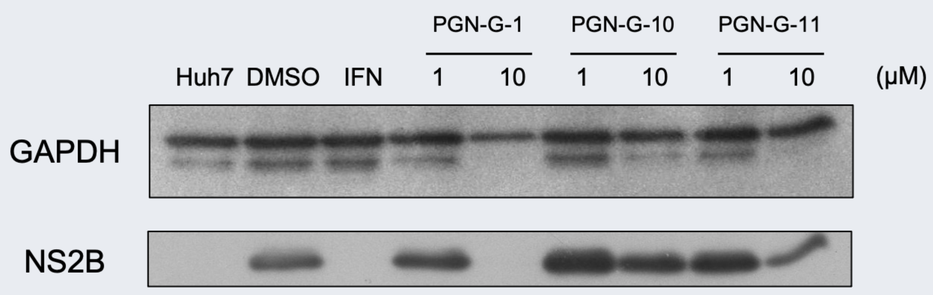


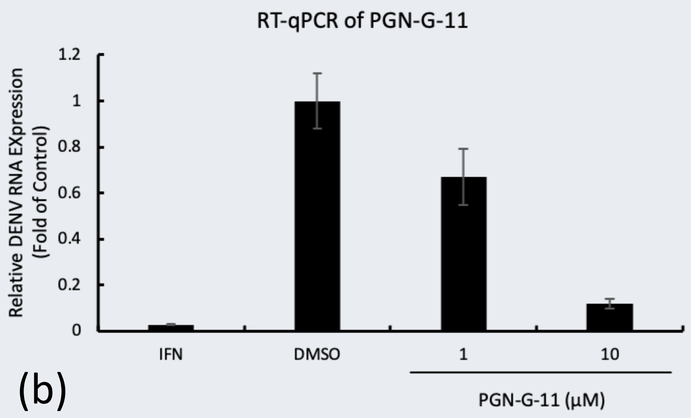
DENV ⊣ HO-1 ↓ —DRUG→ HO-1 ↑

DENV → COX-2 ↑ —DRUG→ COX-2 ↓

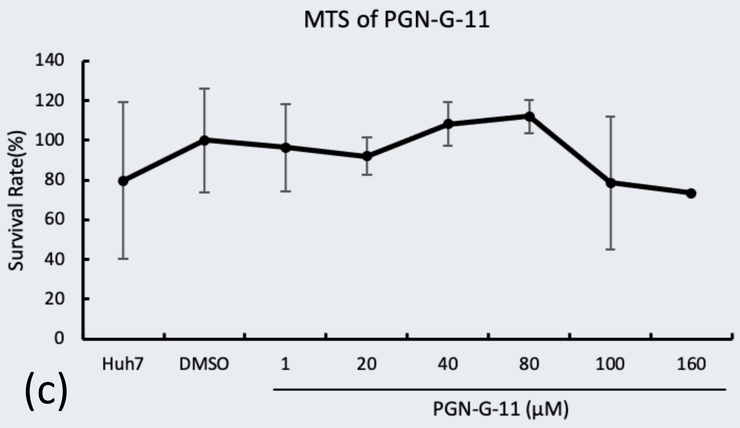
Result



(a)



(b)



(c)

(a)Western Blot of PGN-G-1, PGN-G-10, PGN-G-11. (b)RT-qPCR of PGN-G-11. (c)MTS Assay PGN-G-11.

Conclusion

The result of western blot and RT-qPCR reveal that PGN-G-11 is the most effective compound against virus among three candidates from the analysis of protein and RNA levels. MTS result also indicates the PGN-G-11 with low cytotoxicity, even reach in the high concentration. Promoter-based reporter assay will be the next step to study anti-oxidant and anti-inflammatory properties of PGN-G-11. In conclusion, PGN-G-11 is a potential anti-DENV drug, since the structure of PGN-G-11 is derive from PGN-G-1, it can develop more derivative compounds through structure–activity relationship (SAR).

Future Work

1. Using DENV-infected ICR suckling mouse model for *in vivo* study.
2. The underlying mechanism of drug.

Reference

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