Synergistic Effects of Acyclovir and Andrographolide Derivative on Drug-Resistant Herpes Simplex Virus Type 1

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Acyclovir (ACV) is the common drug for therapy of herpes simplex virus (HSV) infections but ACV-resistant HSVs are frequently isolated from immunosuppressed patients. Therefore, the novel antiviral agents are still needed. In our previous report, andrographolide derivative or 3,19-isopropylideneandrographolide (IPAD) has inhibitory effect on HSV replication of both wild type and drug-resistant strains.

Objective

This study aimed to determine synergistic effects of ACV combining with IPAD on drug-resistant HSV-1.

Materials and Methods

Wild type HSV-1 (strain KOS 1-003) and drug-resistant HSV-1 consisting of dxpIII (phosphonoacetate- and phosphonoformate-resistant), ACGr4 (ACV-resistant with thymidine kinase (TK)-deficient), and dlsptk (ACV-resistant with TK deletion) were used.

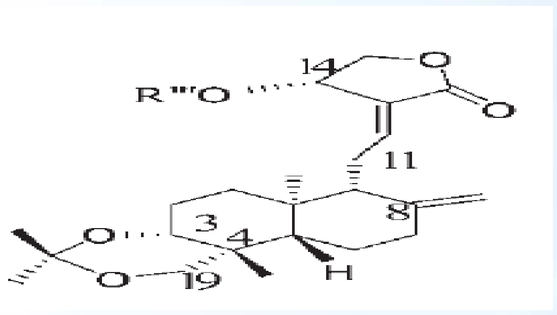
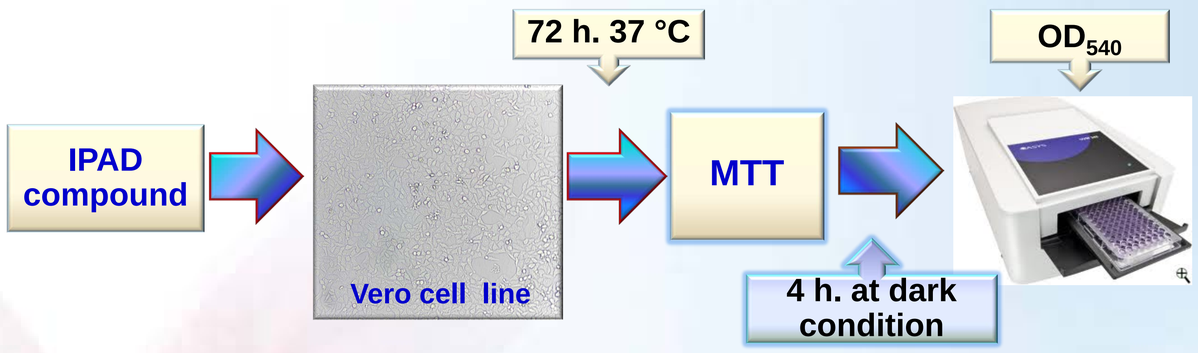
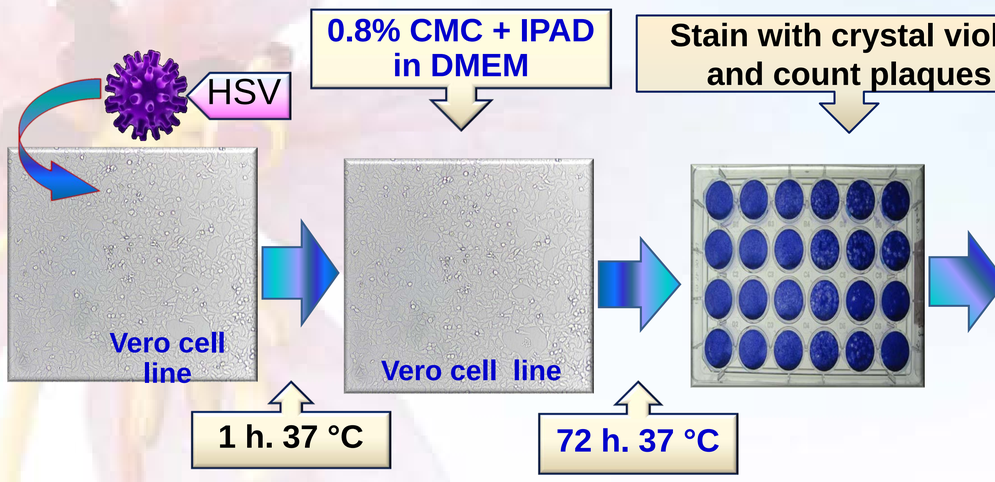


Figure 1. Chemical structure of 3,9-isopropylideneandrographolide (IPAD)

1. Cytotoxicity (MTT assay)



2. Synergistic effect in combination with acyclovir



$$CI = \frac{(D)1}{(D)12} + \frac{(D)2}{(D)12}$$

(D)1 = dose x % inhibition of compound 1
(D)2 = dose x % inhibition of compound 2
(D)12 = [dose compound 1 x % inhibition of compound 1+2] + [dose compound 2 x % inhibition of compound 1+2]

Results

Table 1. Cytotoxicity, antiviral activity and selective index of IPAD

Compound	CC50 (μM)	HSV-1 KOS		HSV-1 ACGr4		HSV-1 dlsptk		HSV-1 dxpIII	
		IC_{50} (μM)	SI	IC_{50} (μM)	SI	IC_{50} (μM)	SI	IC_{50} (μM)	SI
IPAD	39.71	16.96	2.34	17.12	2.32	17.89	2.22	16.86	2.36
ACV	>1000	0.49	>2040	161.45	>6.19	575.91	>1.74	450.25	>2.22

Table 2. Synergistic effects of combination of IPAD with acyclovir (ACV) on anti-HSV activity.

Compounds combination ratio	Compound concentration (μM)		Mean percentage of inhibition (%) IPAD+ ACV	Experimental CI values	Description
	IPAD	ACV			
			HSV-1 (KOS)		
	20.50	-	100		
	-	22.20	100		
2 X IC50	20.50	22.20	100	1	additive
1 X IC50	17.94	0.44	100	0.52	synergism
0.2 X IC50	12.81	0.04	100	0.04	strong syn.
			HSV-1 ACGr4		
	20.50	-	100		
	-	2220.2	100		
2 X IC50	20.50	2220.2	100	1	additive
1 X IC50	17.94	444.03	100	0.45	synergism
0.2 X IC50	12.81	22.20	88.5	0.01	strong syn.
			HSV-1 dlsptk		
	20.50	-	100		
	-	2220.2	100		
2 X IC50	20.50	2220.2	100	1	additive
1 X IC50	17.94	444.03	100	0.59	synergism
0.2 X IC50	12.81	22.20	88.41	0.01	strong syn.
			HSV-1 dxpIII		
	20.50	-	100		
	-	2220.2	100		
2 X IC50	20.50	2220.2	100	1	additive
1 X IC50	17.94	444.03	100	0.62	synergism
0.2 X IC50	12.81	22.20	100	0.06	strong syn.

CI, combination index, a quantitative measure calculated by Calcusyn Software. This index quantifies the interaction between the tested compounds as described by Chou (2006). In detail, CI > 1 means antagonism, 1 means additive effect 0.10 to 0.30 means strong synergism, 0.30–0.70 means synergism, 0.70–0.85 means moderate synergism, and 0.85–0.90 means slight synergism.

Conclusion

This result suggested that IPAD might be a candidate drug for HSV wild type and drug-resistant HSV-1 therapy.

Acknowledgement

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