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Inhibitory activities of Thai medicinal plants against herpes simplex type 1, poliovirus type 1, and measles virus

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Abstract

Forty-eight ethanol- and 43 water-extracts of 49 traditional Thai medicines were evaluated for antiviral activities by a plaque reduction assay. For preliminary characterization of the mode of their antiviral action, poliovirus type 1, measles virus and herpes simplex virus type 1 (HSV-1) that are different in nucleic acid component and enveloped structure were used in this study. Fifty-two, 28 and 29 extracts exhibited inhibitory activities against poliovirus, measles virus and HSV-1, respectively. Of 29 extracts with anti-HSV-1 activities, the inhibitory activities of *Rhinacanthus nasutus* (leaf), *Terminalia citrina* (fruit) and *Thevetia peruviana* (leaf) were observed in both ethanol and water extracts. The ethanol extracts of *Derris scandens* (leaf) and *Plumbago indica* (leaf) and the water extract of *Capsicum frutescens* (fruit) were active against only HSV-1, suggesting the mechanism of their antiviral action likely unique to HSV-1 but neither poliovirus nor measles virus. Contrarily, 26 extracts displayed inhibitory activities against poliovirus and/or measles virus. These findings suggest that the 29 extracts from traditional Thai medicines are potential candidates for anti-HSV agents.

Key words antiviral agent, herpes simplex virus, poliovirus, measles virus, Thai medicinal plant.

Abbreviations ACV, acyclovir; HSV, herpes simplex virus; DMSO, dimethyl sulfoxide; IC_{50}, 50% inhibitory concentration; MEM, minimum essential medium; PFU, plaque forming unit.

Introduction

Herpes simplex virus (HSV) is a common human pathogen that afflicts the majority of the population worldwide.[1,2] It causes a variety of diseases ranging in severity from mild to debilitating and life threatening. After primary infection, HSV establishes latency in sensory and autonomic neurons innervating mucosal membranes.[3] This location can then serve as a site for recurrent infection that may be provoked by a number of stimuli such as sunlight, stress, febrile illnesses, and immunosuppression. Efficacious drugs approved for treating HSV infection include acyclovir (ACV) and valaciclovir, the most widely used.[4] However, a significant risk of drug resistance has been observed during therapy, especially for chronic infection and for immunocompromised patients. Therefore, there is a need to develop new anti-HSV agents that substitute for or complement acyclovir.

Traditional medicines utilizing herbal plants have been shown to contain antiviral activities *in vitro*.[5-7] Active antiviral compounds isolated from herbal plants were purified and identified, including polysaccharides, triterpenes, polyphenols and fatty alcohols.[8-14] For instance, moronic acid and eugeniin were purified from

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extracts of *Rhus javanica* and *Genum japonicum*, respectively, that have been used traditionally for treatment of human diseases.[13,14] Both compounds displayed inhibitory activities against HSV-1 using a plaque reduction assay and showed anti-HSV-1 therapeutic activity in an animal infection model. Kurokawa *et al.*[14] demonstrated that eugeniin inhibited HSV DNA synthesis by interfering noncompetitively with HSV DNA polymerase activity and possessed different anti-HSV activity from ACV. Additionally, it was previously reported that some Thai medicinal plants such as *Centella asiatica* and *Mangifera indica* were active against HSV.[15] These results suggest traditional medicines as useful sources for the search of new anti-HSV agents. Hence, this study was carried out to search potentially anti-HSV candidates from Thai traditional medicines. Forty-nine Thai medicinal plants that were widely used in folk medicine to treat various diseases, including gastrointestinal and skin diseases as well as inflammation were selected. Ethanol and water extracts prepared from these plants were first examined for anti-HSV-1, anti-poliovirus and anti-measles virus activities *in vitro* using a plaque reduction assay. Concentrations of extracts that inhibited 50% anti-HSV-1 plaque formation (IC_{50}) were then determined. Finally, the preliminary results of anti-HSV therapeutic efficacy in mice were also included.

Materials and Methods

Cells and viruses: Vero cells (E6 strain) were grown in Eagle's minimum essential medium (MEM) supplemented with 5 % (v/v) calf serum in a humidified 5% CO_2 incubator at 37℃. The wild-type HSV-1 (7401H strain), poliovirus type 1 (Sabin strain), and measles virus (Tanabe strain) were propagated in Vero cells using a low multiplicity of infection as previously described.[5]

Preparation of extracts: Dried plants collected from Songkhla, Thailand were extracted with water and ethanol separately as described previously.[16] Briefly, five grams of each dried plant were extracted twice with 100 ml of water or ethanol under reflux for 3 h. The solvents were removed under reduced pressure to give the respective dry extracts. For a plaque reduction assay, the dried extracts were dissolved in dimethyl sulfoxide (DMSO) to give a concentration of 10 mg/ml and diluted with culture medium to make its various final concentrations as described below. The concentration of DMSO in each culture medium was equal to or less than 1.0 %. One percent DMSO solution was used as a control. For administration to mice, the extracts were dissolved in 0.4 ml of DMSO and then pyrogen-free distilled water was added to make a final volume of 40 ml for oral administration.[13] One percent DMSO solution was used as a control for oral administration in the animal experiments.

ACV: ACV was purchased as tablets from Glaxo Smith Kline K.K., Tokyo, Japan. A tablet (200 mg) was powdered and suspended in distilled water.

Plaque reduction assay: Duplicate cultures of Vero cells in 60 mm dishes were infected with 100 plaque forming units (PFU) of HSV-1, poliovirus, or measles virus for 1 h at room temperature. Subsequently, cells were overlaid with 5 ml of nutrient media (MEM, 2% calf serum and 0.8% (w/v) carboxymethylcellulose) in either the absence or presence of extracts. Cell cultures infected with HSV-1, poliovirus, and measles virus were then incubated for 2, 3 and 5 d at 37℃, respectively. The infected cells were fixed with a 5% formaldehyde solution and stained with 0.03% methylene blue. Numbers of plaques were counted under a binocular microscope. IC_{50} values were determined from a curve relating the plaque number to the concentration of samples.[13] Additionally, cytotoxicity of extracts was determined as visible cytotoxicity by comparing the cell monolayer of treatment with that of the control and scored as follows: (++) more than 50% of cell detachment as strong cytotoxicity; (+) 50-10% of cell detachment as intermediate cytotoxicity; (±) less than 10% of cell detachment as weak cytotoxicity; (-) no detachment of cells as no cytotoxicity.[5] In the plaque reduction assay to determine IC_{50} values, cytotoxicity was also evaluated at concentrations near IC_{50} values using the visible cytotoxicity. When strong cytotoxicity was observed, plaques were not clear. However, they were countable in intermediate and weak cytotoxicities as well as no cytotoxicity.

Cutaneous HSV-1 infection in mice: Female BALB/c mice (6-week-old, 17-19 g) were purchased from Sankyo Labo Service Co., Ltd. (Tokyo, Japan). Mice were cutaneously infected with wild type HSV-1 (1×10^6 PFU/mouse) after scarification of the shaved right midflank with 27-gauge needles as described previously.[5,13] The development of skin lesions and death

were observed three times daily with the severity of the lesions being scored as described previously:[5, 13] 0, no lesion; 2, vesicles in local region; 4, erosion and/or ulceration in local region; 6, mild zosteriform lesion; 8, moderate zosteriform lesion; 10, severe zosteriform lesion; 12, death. The infected mice were fed and observed for at least 20 d to determine their mortality.

Results and Discussion

Forty-nine traditional Thai medicines were evaluated regarding their antiviral activities using a plaque reduction assay at two concentrations of 50 and 100 μg/ml (Table I). As reported, the maximal serum concentration of D-glucose, a typical chemical with rapid absorption from alimentary tracts in human, is approximately 300 μg/ml after 10 gram oral administration in human with diabetes mellitus.[17] In addition, some of the low molecular weight components in extracts may be absorbed as rapidly and efficiently as D-glucose. Therefore, a 100 μg/ml dose in a plaque reduction assay was used to examine antiviral activity at putative concentrations in serum after oral administration. We also used the lower concentrations (50 μg/ml) to select extracts that could reduce the plaque formation to less than 50%.

To examine the mode of antiviral action of these Thai medicines preliminarily, three viruses with differences in nucleic acid component and structure were used

Table I. Antiviral activities of Thai medicinal plants by a plaque reduction assay.

Botanical name	Family	Part used	Extract	Plaque formation (%)[a]						Cytotoxicity[b]		HSV-1	Cytotoxicity[b,c] at IC_{50}
				50 (μg/ml)			100 (μg/ml)			50 μg/ml	100 μg/ml	IC_{50} μg/ml	
				HSV-1	Polio	Measle	HSV-1	Polio	Measle				
Acacia consinna DC.	Mimosaceae	Leaf	Ethanol	110.2	90.4	90.0	83.2	84.8	76.2	-	-		
			Water	110.8	99.3	109.1	109.8	71.3	89.5	-	-		
Adhatoda vasica Nees	Acanthaceae	Leaf	Ethanol	94.5	100.0	90.7	91.4	100.0	76.9	-	-		
			Water	71.4	89.0	110.5	66.7	76.6	100.7	±	±		
Alpinia galanga Sw.	Zingiberaceae	Rhizo	Ethanol	0.03[d]	0.0	2.2	0.0	0.0	0.0	+	++	7.0	±
			Water	114.4	104.4	117.3	112.1	81.5	100.9	-	-		
Alyxia nitens Kerr	Apocynaceae	Leaf	Ethanol	60.5	74.7	0.0	55.9	64.7	0.0	±	±		
			Water	111.6	83.4	104.5	109.8	59.2	90.9	-	-		
Andrographis Wall ex. Nees	Acanthaceae	Leaf	Ethanol	78.9	70.9	6.0	64.5	23.2	0.0	+	+		
			Water	98.2	81.5	123.2	90.0	56.0	87.3	-	-		
Baleria lupulina Lindl.	Acanthaceae	Leaf	Ethanol	100.5	91.2	90.3	88.6	48.8	80.0	±	±		
			Water	97.0	78.6	104.5	88.7	53.4	100.9	-	-		
Bixa orellana L.	Bixaceae	Leaf	Ethanol	98.5	80.0	81.5	85.2	36.7	63.2	-	-		
			Water	121.2	101.1	112.7	118.7	83.1	105.9	-	-		
Bixa orellana L.	Bixaceae	Seed	Ethanol	18.8	46.3	19.8	0.0	0.4	0.0	±	+	42.0	±
			Water	86.2	78.1	85.7	56.9	55.6	64.8	-	-		
Calophyllum	Guttiferae	Leaf	Ethanol	32.0	72.5	2.2	10.4	17.9	0.0	±	+	28.0	-
			Water	113.4	108.9	111.8	110.7	68.8	96.8	-	-		
Capsicum frutescens	Solanaceae	Fruit	Ethanol	73.3	86.6	87.0	62.6	57.1	83.9	-	±		
			Water	97.3	86.3	106.8	46.3	66.6	76.4	-	-	93.0	-
Cassia angustifolia	Caesalpiniace	Leaf	Ethanol	54.7	89.1	71.6	43.3	33.3	61.2	-	±	71.0	±
			Water	106.7	83.4	101.8	104.7	65.4	81.4	-	-		
Cassia fistula L.	Caesalpiniace	Fruit	Ethanol	67.2	88.4	88.1	60.0	53.6	76.9	±	±		
			Water	n.d.[e]	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.		

Clinacanthus nutans Lindau	Acanthaceae	Leaf	Ethanol	71.6	76.0	86.3	55.9	45.7	76.2	±	±			
			Water	90.7	102.8	104.1	81.0	65.6	98.6	-	-			
Coleus parvifolius Benth.	Labiatae	Aerial part	Ethanol	60.9	48.8	89.2	57.9	38.3	78.2	±	+			
			Water	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.			
Croton sublyratus Kurz	Euphorbiacea	Leaf	Ethanol	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.			
			Water	87.3	88.2	105.9	82.7	63.2	92.7	-	-			
Curcuma longa L.	Zingiberaceae	Rhizo	Ethanol	0.0	26.2	0.0	0.0	0.0	0.0	+	++	38.0	±	
			Water	98.7	81.1	98.2	93.3	64.5	81.8	-	-			
Curcuma zedoaria Roscoe	Zingiberaceae	Rhizo	Ethanol	6.6	33.1	0.0	0.0	0.0	0.0	+	++	40.0	±	
			Water	105.2	81.8	82.7	104.0	65.0	64.4	-	-			
Derris scandens Benth.	Papilionaceae	Leaf	Ethanol	64.8	107.7	68.0	0.0	51.8	56.2	±	±	60.0	±	
			Water	106.7	64.9	88.6	105.3	45.9	85.5	-	-			
Garcinia atroviridis Griff.	Guttiferae	Fruit	Ethanol	72.9	80.6	90.3	71.0	105.5	59.7	-	-			
			Water	101.6	92.9	110.0	100.2	72.7	87.7	-	-			
Glycyrrhiza glabra L.	Papilionaceae	Root	Ethanol	3.5	2.3	0.0	0.0	0.0	0.0	-	±	37.0	-	
			Water	92.6	96.9	93.6	83.0	51.9	77.3	-	-			
Hibiscus sabdariffa L.	Malvaceae	Flower	Ethanol	100.8	135.6	89.1	100.1	87.1	69.0	-	-			
			Water	104.0	100.0	93.2	101.2	100.0	90.9	-	-			
Kaempferia galanga L.	Zingiberaceae	Rhizo	Ethanol	71.4	6.0	23.6	4.3	0.0	0.0	-	±	63.0	-	
			Water	102.0	52.8	98.2	100.3	39.9	85.9	-	-			
Lawsonia inermis L.	Lythraceae	Leaf	Ethanol	69.4	121.1	72.0	19.6	10.9	0.0	±	±	80.0	±	
			Water	103.3	43.8	94.1	100.0	29.3	74.5	-	-			
Morinda citrifolia L.	Rubiaceae	Leaf	Ethanol	92.3	140.5	77.4	86.5	120.1	44.8	-	-			
			Water	101.4	45.5	105.5	100.7	37.6	87.7	-	-			
Myristica fragrans L.	Myristicaceae	Leaf	Ethanol	99.8	63.1	90.4	92.1	12.2	67.1	-	-			
			Water	100.9	52.0	91.8	94.0	35.7	71.4	-	-			
Ocimum basilicum L.	Labiatae	Leaf	Ethanol	57.9	63.0	77.8	48.8	44.3	61.9	-	-	93.0	-	
			Water	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.			
Ocimum canum Sims	Labiatae	Leaf	Ethanol	70.5	57.8	82.4	45.2	33.2	56.1	±	±	90.0	±	
			Water	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.			
Piper betle L.	Piperaceae	Leaf	Ethanol	52.0	77.6	33.8	36.4	7.7	1.6	±	+	61.0	±	
			Water	110.3	45.9	100.2	100	33.2	93.6	-	-			
Piper chaba Vahl	Piperaceae	Fruit	Ethanol	0.0	0.0	0.0	0.0	0.0	0.0	±	+	28.0	-	
			Water	97.3	48.3	100.4	75.3	41.3	83.6	-	-			
Piper nigrum L.	Piperaceae	Fruit	Ethanol	0.0	0.8	0.0	0.0	0.0	0.0	+	++	25.0	±	
			Water	107.7	49.0	99.5	100.0	41.0	91.4	-	-			
Piper ribesoides Wall[f]	Piperaceae	Stem	Ethanol	64.8	82.8	8.7	15.8	13.5	0.0	±	+	62.0	±	
			Water	101.3	58.4	87.3	100.7	45.5	75.9	-	-			
Piper ribesoides Wall[f]	Piperaceae	Leaf	Ethanol	96.0	114.4	63.6	74.0	91.6	47.7	-	±			
			Water	96.7	59.8	95.0	79.3	49.3	85.9	-	-			

Piper ribesoides Wall[g]	Piperaceae	Stem	Ethanol	25.1	101.1	7.3	6.0	5.6	0.0	-	±	34.0	-
			Water	103.3	53.1	98.2	100.0	47.5	70.0	-	-		
Piper ribesoides Wall[g]	Piperaceae	Leaf	Ethanol	58.3	135.2	98.2	41.6	35.6	54.1	-	±	80.0	±
			Water	105.2	55.7	89.5	103.5	42.1	87.7	-	-		
Piper sarmentosum	Piperaceae	Leaf	Ethanol	98.3	100.5	36.8	78.5	93.8	0.0	-	-		
Roxb.			Water	109.5	92.8	108.2	100.4	87.8	88.2	-	-		
Plumbago indica L.	Plumbaginace	Leaf	Ethanol	0.0	88.3	107.7	0.0	60.7	62.7	±	±	42.0	±
			Water	100.4	58.4	90.7	100.0	35.6	80.3	-	-		
Plumbago indica L.	Plumbaginace	Root	Ethanol	95.6	99.5	102.3	68.7	62.9	93.2	-	±		
			Water	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.		
Psidium guajava L.	Myrtaceae	Leaf	Ethanol	102.4	38.3	92.7	70.2	2.4	50.0	±	+		
			Water	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.		
Quisqualis indica L.	Combretaceae	Leaf	Ethanol	92.9	100.0	84.1	89.6	96.9	74.5	-	±		
			Water	100.1	72.2	99.7	95.2	37.1	92.7	-	-		
Rhinacanthus nasutus	Acanthaceae	Leaf	Ethanol	0.0	34.4	4.9	0.0	0.0	0.0	±	+	30.0	-
Kurz			Water	0.0	8.0	0.0	0.0	0.0	0.0	-	-	17.0	-
Terminalia citrina	Combretaceae	Fruit	Ethanol	58.6	72.2	80.2	2.6	7.7	50.4	±	+	68.0	±
Flemming			Water	68.9	59.1	54.3	5.6	42.6	53.0	-	±	70.0	-
Theobroma cacao L.	Sterculiaceae	Leaf	Ethanol	96.6	116.0	102.7	83.5	107.4	98.6	-	-		
			Water	100.0	58.6	101.4	97.6	42.4	72.3	-	-		
Thevetia peruviana	Apocynaceae	Leaf	Ethanol	0.0	0.0	0.0	0.0	0.0	0.0	+	+	18.0	-
Schum.			Water	0.2	0.0	4.7	0.0	0.0	0.0	±	+	12.0	-
Thunbergia laurifolia	Thunbergiace	Leaf	Ethanol	101.1	73.5	66.8	100.5	65.4	49.1	±	±		
L.			Water	100.7	73.3	105.5	95.0	41.2	91.4	-	-		
Tribulus terristris L.	Zygophyllace	Aerial	Ethanol	109.4	106.1	96.8	88.6	93.4	42.3	-	±		
		part	Water	100.0	70.0	118.6	97.6	65.8	101.8	-	±		
Trichosanthes anguina	Cucurbitaceae	Fruit	Ethanol	95.3	81.9	98.2	84.5	71.2	95.0	-	-		
L.			Water	98.8	78.4	118.2	86.7	72.0	104.1	-	-		
Zingiber cassumunar	Zingiberaceae	Rhizo	Ethanol	1.6	0.0	0.0	0.0	0.0	0.0	±	+	21.0	-
Roxb.			Water	83.3	49.7	88.4	61.9	23.4	75.1	-	-		
Zingiber officinale	Zingiberaceae	Rhizo	Ethanol	31.4	100.8	11.8	0.0	0.0	0.0	±	+	48.0	±
Roscoe			Water	85.7	66.9	103.2	60.7	45.3	99.9	-	-		
Zingiber zerumbet	Zingiberaceae	Rhizo	Ethanol	0.0	0.0	0.0	0.0	0.0	0.0	+	++	16.0	-
Smith			Water	98.8	69.1	106.7	92.8	46.5	102.6	-	-		

[a] Plaque formation represents the percentage compared to untreated controls. Percent plaque formation of HSV-1 was 0% at 5 μg/ml of ACV.
[b] ++, +, ± and - indicate strong, intermediate, weak, and no cytotoxicity, respectively, as described in text.
[c] Cytotoxicity observed at the range of IC_{50} values in the plaque reduction assay in the presence of various concentrations of extracts.
[d] Underlines represent extracts that reduced plaque formation to>50%
[e] Not determined.
[f] Lanocelate shaped leaf
[g] Cordate shaped leaf

in this study; poliovirus (a non-enveloped RNA virus), measles virus (an enveloped RNA virus), and HSV-1 (an enveloped DNA virus). In this study, the extracts were defined as antiviral active extracts if they reduced the plaque formation of each virus to less than 50%. Fifty-two extracts were effective against poliovirus; 14 ethanol- and 8 water-extracts were active at 50 μg/ml, and 30 ethanol- and 22 water-extracts exhibited activities at 100 μg/ml. For measles virus, 29 extracts were active; 20 ethanol- and 2 water-extracts displayed activities at 50 μg/ml, and 27 ethanol- and 2 water-extracts were active at 100 μg/ml. Twenty-nine extracts were active against HSV-1; 15 ethanol- and 2 water-extracts showed activities at 50 μg/ml, and 25 ethanol- and 4 water-extracts were active at 100 μg/ml. Of all extracts with anti-HSV-1 activities, *Rhinacanthus nasutus* (leaf), *Terminalia citrina* (fruit) and *Thevetia peruviana* (leaf) exhibited inhibitory activities in both ethanol- and water-extracts. The ethanol extracts of *Cassia angustifolia* Vahl (leaf), *Ocimum basilicum* (leaf), *Ocimum canum* (leaf), *Piper ribesoides* (leaf) and *Terminalia citrina* (fruit) as well as the water extract of *Terminalia citrina* (fruit) were also active against poliovirus. Moreover, 20 extracts exhibited inhibitory activities against not only HSV-1, but also both the poliovirus and measles virus; 11 ethanol- and 2 water-extracts at 50 μg/ml, and 18 ethanol- and 2 water-extracts at 100 μg/ml. These results suggest that some active compounds in extracts may be common in the mode of action of each virus. However, since these viruses are different in structure and replication, it is probable that extracts contain different active substances whose mechanism of action may be specific to each virus. Furthermore, it was found that the ethanol extracts of *Derris scandens* (leaf) and *Plumbago indica* (leaf), and the water-extract of *Capsicum frutescens* (fruit) showed appreciable inhibitory activities against only HSV-1, suggesting that the mechanism of their antiviral action is likely unique to HSV-1 but neither poliovirus nor measles virus. Although there were some extracts with intermediate visible cytotoxicity, all of the 29 extracts were also effective to reduce the plaque formation of HSV-1. We selected the 29 extracts as candidates of anti-HSV extracts.

Anti-HSV-1 activities of the 29 extracts were then evaluated by the determination of their IC_{50} values. The values of IC_{50} ranged from 7 to 93 μg/ml. Of these 29 extracts, the ethanol extract of *Alpinia galanga* (rhizome) showed the lowest IC_{50} values (7.0 μg/ml). Additional six medicinal plants of Zingiberaceae prepared from ethanol extracts also exhibited moderately HSV-1 inhibitory activities. In contrast, the water extracts of these seven plants were inactive against HSV-1, consistent with the results of Zingiberaceae plants including *Alpinia officinale, Curcuma aeruginosa*, and *Curcuma xanthorrhiza* previously reported by Kurokawa *et al.*[5] Besides the ethanol extract of *Alpinia galanga*, the water and ethanol extracts of *Thevetia peruviana* as well as the water extract of *Rhinacanthus nasutus* displayed appreciable anti-HSV-1 activities. At the all ranges of IC_{50} values of 29 extracts, strong or intermediate visible cytotoxicity was not observed (Table 1). Thus, all 29 extracts selected exhibited anti-HSV activity.

Therapeutic anti-HSV-1 efficacies *in vivo* of the 29 herbal extracts with anti-HSV-1 activities were preliminarily evaluated. Interestingly, it was found that some extracts significantly delayed the development of HSV-1 skin lesion. Although the actual cytotoxicity of these extracts in cell culture model had not been performed in this study, no toxic effects were observed after *in vivo* administration (data not shown). This suggested that some of antiviral compounds in extracts were selectively absorbed from alimentary tracts and exerted an antiviral effect which was not associated with toxicity. Therefore, the results reported in this study suggest 29 herbal extracts as possible candidates for anti-HSV agents. Further determination of their HSV-1 selective index and detailed investigation of their therapeutic anti-HSV-1 efficacies *in vivo* will be valuable to substantiate these findings. In addition, purification, isolation and identification of anti-HSV compounds from these medicinal extracts as well as characterization of their anti-HSV action should be performed.

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和文抄録

49種のタイ伝統薬物から作製した48のエタノールエキス，43の水エキスに対する抗ウイルス活性をプラーク減

少法で検索した。本研究では，核酸やエンベロープ構造の異なるポリオウイルス１型，麻疹ウイルス，単純ヘルペスウイルス１型に対して検討を行なった。その結果ポリオウイルス１型に対しては52種，麻疹ウイルスには28種，単純ヘルペスウイルス１型には29種のエキスが阻害活性を示した。単純ヘルペス１型に有効であった29種の中では，*Rhinacanthus nasutus*（葉），*Terminalia citrina*（果実），*Thevetia peruviana*（葉）はエタノール，水の両エキスで阻害作用を示した。*Derris scandens*（葉），*Plumbago indica*（葉）および*Capsicum frutescens*（果実）のエタノールエキスは単純ヘルペスウイルス１型にのみ活性があった。このことはポリオウイルスや麻疹ウイルスには無効で，単純ヘルペス１型のみに特異的に有効であることを示唆している。一方，26種のエキスはポリオウイルスあるいは麻疹ウイルス，また両方に有効であった。これらの結果はタイ伝統薬物からの29のエキスが抗ヘルペス剤として有力な候補となることを示している。

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