REFERENCES

ประเสริฐ ทองเจริญ, 2528. เริม พิมพ์ครั้งที่ 1. กรุงเทพมหานคร: บริษัทเมดาร์ท.

Abad, M. J., Bermejo, P., Gonzales, E., et al. 1999a. Antiviral activity of Bolivian plant extracts. Gen Pharmacol 32: 499-503.

Abad, M. J., Bermejo, P., Sanchez Palomino, S., et al. 1999b. Antiviral activity of some South American medicinal plants. Phytother Res 13: 142-146.

Abou-Karam M., and Shier W. T. 1990. A simplified plaque reduction assay for antiviral activity agents from plants, demonstration of frequent occurrence of antiviral activity in higher plants. J Nat Prod 53(2):340-44.

Afolayan, A. J.and Meyer, J. J. M. 1995. Morphology and ultrastructure of secreting and nonsecreting foliar trichomes of *Helichrysum aureonitens* (Asteraceae). Int J Plant Sci 156: 481-487. [Abstract]

Almeida, A. P. , Miranda, M. M. , Simoni, I. C., et al. 1998. Flavonoid monoglycosides isolated from the antiviral fractions of *Persea americana* (Lauraceae) leaf infusion. Phytother Res 12: 562-567. [Abstract]

Anani, K., Hudson, J.B., Souza, C. , et al. 2000. Investigation of medicinal plants of Togo for antiviral and antimicrobial activities. Pharm Biol ;38:40-45. [Abstract]

Andrei, G., Snoeck, R., and De Clercq, E. 1995. Susceptibilities of several drug-resistant herpes simplex virus type 1 strains to alternative compounds. Antimicrob Agents Chemother 39(7): 1632-1635.

Backer, C.A. , Bakhuizen V.D., ank Brink, R.C. 1965 Flora of Java 2 vols. The Netherlands Rijksherbarium, Leyden.

Balzarini, J., and De Clercq. E. 1991. 5-Phosphoribosyl 1-pyrophosphate synthetase converts the acyclic nucleoside phosphonates 9-(3-hydroxy-2-phosphonylmethoxypropyl)adenine and 9-(2-phosphonylmethoxyethyl)adenine directly to their antivirally active diphosphate derivatives. J Biol Chem 266:8686–8689.

Batista, O., Simoes, M. F., Duarte, A., et al. 1995. An antimicrobial abietane from the root of *Plectranthus hereroensis*. Phytochemistry 38: 167-169. [Abstract]

Berghe, V, Vlietinck, D.A.and Vanhoof, A.J. 1986. Plant products as potential antiviral agent. Bulletin de I;Institut Pasteur, 84,101

Beswick, T.S. 1962. The origin and use of DAO herpes. Med Hist 6:214.

Birch, C.J., Tachedjian, G., Doherty, R.R., Hayes, K., Gust, I.D. 1990. Altered sensitivity to antiviral drugs of herpes simplex virus isolates from a patient with the acquired immunodeficiency syndrome. J Infect Dis 162:731-734.

Brooks, G.F., Butel, J.S., Morse, S.A. 2001. Jawetz, Melnick and Adelberg's Medical Microbiology 22nd ed. New York. McGraw-Hill.

Bryson, Y.J., Dillon, M., Lovett, M., et al. 1983. Treatment of first episodes of genital herpes simplex virus infection with oral acyclovir: a randomized double-blind controlled trial in normal subjects. N Engl J Med 308:916-921.

Buchman, T.G., Roizman, B., Adams, G., and Stover, B.H. 1978. Restriction endonuclease fingerprinting of herpes simplex DNA: a novel epidemiological tool applied to a nosocomial outbreak. J Infect Dis 138:488-498.

Bunyapraphatsara, N., Dechsree, S. and Yoosook, C., et al. 2000. Anti-herpes simplex virus component isolated from *Maclura cochinchinensis*. Phytomedicine 6:421-424. [Abstract]

Cassady, K. A. and Whitley, R. J. 1997. New therapeutic approaches to the alphaherpesvirus infections. J Antimicrob Chemother 39: 119-128.

Claus, J. D., Coto, C. E., and Torres, R. A. de. 1998. Natural resistance of BALB/c mice to herpes simplex virus-type 1 intraperitoneal infection is abrogated by a plant extract with *in vitro* anti-herpes activity. Phytother Res 12: 250-254.

Corey, L., Benedetti, J.,and Crichlow, C. et al. 1983 Treatment of primary episodes of genital herpes virus infections with acyclovir : results of tropical, intravenous, and oral therapy. J Antimicrob Chemother12:79.

Corey, L. and Spear, P.G. 1986. Infections with herpes simplex viruses, part I. N Eng J Med 314(11): 686-691.

De Clercq, E., 1993 Antiviral of the treatment of herpesvirus infections. J Antimicrob Chemother 32, supplA:121-123.

Ellis, M. N., Keller, P. M., Fyfe, J. A., et al. 1987. Clinical isolate of herpes simplex virus type 2 that induces a thymidine kinase with altered substrate specificity. Antimicrob Agent Chemother 31: 1117-1125.

Gadler, H. 1983. Nucleic acid hybridization for measurement of effects on antiviral compounds on human cytomegalovirus DNA replication. Antimicrob Agent Chemother 24: 370-374.

Ginsberg, H. S. 1980. Herpesviruses. In H. S. Ginsberg and R. Dulbecco (eds.), Virology, 3rd ed., pp. 161-168. Philadelphia: Harper & Row Press.

Greger, H., Zechner, G., Hofer, O., Hadacek, F. and Wurz, G. 1993 Sulphur-containing amides from Glycosmis species with different antifungal activity. Phytochemistry 34(1):175-179.

Harmenberg, J., Wahren, B., and Oberg, B. 1980. Influence of cells and virus multiplicity on the inhibition of herpesvirus with acyclovir guanosine. Intervirology 14: 23-224.

Harmenberg, J., Abele, G., and Wahren, B. 1985. Nucleoside pools of acyclovir-treated herpes simplex type 1 infected cells. Antiviral Res 5:75-81.

Hasegawa, T. and Kaeagushi, T. 1994. Future of prodrugs in antiviral therapy. Clin Pharmacokinet; 27: 331-336.

Hayashi K., Hayashi T., Morita N., et al. 1990. Antiviral activity of an extract *of Cordia salicifolia* on HSV type 1. Planta Med 56:439-43.

Hayashi, K., Kamiya, M., and Hayashi, T. 1995. Virucidal effects of the steam distillate from *Houttuynia cordata* and its components on HSV-1, influenza virus, and HIV. Planta Med 61: 237-241.

Hayashi, K., Niwayama, S., and Hayashi, T. *In vitro* and *in vivo* antiviral activity of scopadulcic acid B from *Scoparia dulcis*, Scrophulariaceae, against herpes simplex virus type 1. Antiviral Res 9(6): 345-54.

Hayashi, K., Hayashi, T., Ujita, K. and Takaishi, Y. 1996. Characterization of antiviral activity of a sesquiterpene, triptofordin C-2. J. of Antimicrob Chemotherapy 37, 759-768.

Honess, R.W. and Roizman, B. 1974. Regulation of herpesvirus macromolecular synthesis. I. Cascade regulation of the synthesis of the three groups of viral proteins. J Virol 14: 8-19.

Jassim, S.A.A. and Naji, M.A. 2003 Novel antiviral agents: a medicinal plant perspective. J of Applied Microbiology 95(3): 412-427.

Jayavasu C., Balachandra K., Sankitjaporn S., et al. 1992. Clinical trial in the treatment of genital herpes patient with *Clinacanthus nutans* extract. Com Dis J 18(3):152-61.

Jayawasu, C., Dechatiwongse, T., Balachandra, K. et al 1992 The virucidal activity of *Clinacanthus nutans* Lindau. Extracts against herpes simplex virus type2: An invitro study. Bull Dept Med Sci 34(4):153-158.

Kott, V., Barbini, L., Cruanes, M., et al. 1999. Antiviral activity in Argentine medicinal plants. J Ethnopharmacol 64: 79-84.

Kuo, Y. C., Chen, C. C., Tsai, W. J., et al. 2001. Regulation of herpes simplex virus type 1 replication in Vero cells by *Psychotria serpens*: relationship to gene expression, DNA replication, and protein synthesis. Antiviral Res 51: 95-109.

Kurokawa M, Ohyama H, Hozumi T, et al. 1996. Assay for antiviral activity of herbal extracts using their absorbed sera. Chem Pharm Bull 44(6):1270-2.

Latba, C. and Joseph, A.1999 Evaluation of the larvicidal potential of Glycosmis against four important mosquito species of Kerala India International Pest Control41 (2):50-51.

Lee, J.B., hayashi, K., Hayashi, T., Sankawa, U. and Maida, M. 1999 Antiviral activity against HSV-1 and HIV-1 of Rhamnan sulfate from *Monostroma latissimum*. Planta Med 65:439-441.

Quader, M.A., Metan, M.T.H. et al. 1999 Antitumor alkaloid from *Glycosmis pentaphylla* Fitoterapia 70(3):305-397.

Leung, D. T. and Sacks, S. L. 2000. Current recommendations for the treatment of genital herpes. Drugs 60(6): 1329-1352.

Lowenstein, A. 1919. Aetiologische untersuchgen uber den fieberhaften, herpes. Munch Med Wochenschr 66: 769-770.

Marchetti, M., Pisani, S., Pietropaolo, V., et al. 1996. Antiviral effect of a polysaccharide from *Sclerotium glucanicum* toward herpes simplex virus type 1 infection. Planta Med 62: 303-307.

Mattison, H. R., Eisenberg, R. J., and Reichman, R. C. 1991. Herpes simplex virus. In: Belshe, R.B., editor. Textbook of Human Virology. 2nd ed. pp. 822-41. Mosby, MO, St Louis.

McClain, D.S., and Puller, A. O. 1994 Cell-surface kinetics and efficiency of herpes simplex virus type1 etry are determined by two distinct phase of attachment. Virology 198:690-702..

Mertz, G.J., Critchlow, C.W., Benedetti, J. et al. 1984 Double-blind placebo-controlled trial of oral acyclovir in first episode genital herpes simplex virus infection. JAMA:251, 2103

Meyer, J. J. M., Afolayan, A. J., Taylor, M. B., et al.1996. Inhibition of herpes simplex virus type 1 by aqueous extracts from shoots of *Helichrysum aureonitens* (Asteraceae). J Ethnopharmacol 52: 41-43.

Naesens, L. and De Clercq, E. 2001. Recent developments in herpesvirus therapy. Herpes 8(1): 12-16.

Nahmias, A. J. and Dowdle, W. R. 1968. Antigenic and biologic differences in herpersvirus hominis. Prog Med Virol 10:110-159.

Neyts, J., Andrei, G., Snoeck, R., Jähne, G., Winkler, I., Helsberg, M., Balzarini, J., and De Clercq, E. 1994. The N-7-substituted acyclic nucleoside analog 2-amino-7-[(1,3-dihydroxy-2-propoxy)methyl]purine is a potent and selective inhibitor of herpesvirus replication. Antimicrob Agents Chemother 38:2710–2716.

O'Brien, J.J. and Campoli-Richards, D.M. 1989. Acyclovir: an updated review of its antiviral activity, pharmacokinetic properties and therapeutic efficacy. Drugs 37:233-309.

Peacock, J.E., Kaplowitz, L.G., Sparling, P.F. et al 1988. Intravenous acyclovir therapy of first episodes of genital herpes: a multicenter double-blind, placebo-controlled trial. Ame J Med:85, 301.

Phrutivorapongkul, A., Lipipun, V., Ruangrungsi, N., Watanabe, T., and Jshikawa, T. 2002 Studies on the constituents of seeds of *Pachyrrhizus erosus* and their anti-herpes simplex virus (HSV) activities. Chem Pharm Bull 50:534-537.

Piret, J. , Roy, S., Gagnon, M., Landry, S., Desormeaux, A., Omar, R., and Begeron, M.G. 2002. Mechanisms of herpes simplex virus inactivation by sodium lauryl sulfate and n-lauroylsarcosine were studied. Antimicrob and Chemother 46(9):2933-2942.

Plummer, G. 1964. Serological comparison of the herpesviruses. Br J Exp Pathol 45: 135.

Pongboonrod, S. 1979 Medicinal plants in Thailand Bangkok, Kungthon Press.

Pottage, J.C., and Kessler, H.A. 1995. Herpes simplex virus resistance to acyclovir: clinical relavance. Infectious Agent & Disease; 4(3): 115-124.

Prichard, M. N., Turk, S. R., Coleman, L. A., et al. 1990. A microtiter virus yield reduction assay for the evaluation of antiviral compounds against human cytomegalovirus and herpes simplex virus. J Virol Methods 28: 101-106.

Rabalais, G. P., Levin, M. J., and Berkowitz, F. C. 1987. Rapid herpes simplex susceptibility testing using an enzyme-linked immunosorbent assay performed *in situ* on fixed virus-infected monolayers. Antimicrob Agent Chemcther 31: 946-948.

Reed, J.L. and Muench, H. 1938. A simple method of estimating fifty percent endpoints. Am J Hyg ; 27: 493-497.

Richards, D.M., Carmine, A.A., Brogden, R.N., Heel, R.C.ม Speight, T.M., et al. 1983. Acyclovir: a review of its pharmacodynamic properties and therapeutic efficacy. Drugs 26: 378-438.

Roizman, B. 1996. *Herpesviridae.* In B. N. Fields, D. M. Knipe, P. M. Howley, et al. (eds.), Fields Virology, pp.2221-2230ม Lippincott-Raven Publisher, Philadelphia:.

Roizman, B., Carmichael, L.E., Deinhardt, F., de The, G., Nahmias, A. J., Plowright, W., Rapp, F. Sheldrick, P., Takahashi, M., and Wolf, K. 1981. Herpesviridae: definition, provisional nomenclature and taxonomy. Intervirology 16:201-17.

Roizman, B. Hayword, G., Jacob, R., et al. 1975. Human herpesvirus 1: A model for molecular organization and regulation of herpesvirus. A review. In G. de The , M.A. Epstein, and H. Zur Hausen (eds.), Oncogenes and herpes viruses III, part 1, No 11, pp.3-38. Lyon: IARC.

Roizman, B., Jenkins, F.J. 1985. Genetic engineering of novel genomes of large DNA viruses. Science 229:1208-1214.

Roizman, B., Norrild, B., Chan, C., Pereira, L. 1984. Identification and preliminary mapping with monoclonal antibodies of a herpes simplex virus 2 glycoprotein lacking a known type 1 counterpart. Virology 133:242-247.

Roizman, B. and Sears, A.E. 1993. Herpes simplex viruses and their replication. In: Roizman, B., Whitley, R.J., Lopez, C. (eds). The human herpesviruses, pp. 11-68.Raven, New York:

Roizman, B. and Sears, A.E. 1996. Herpes simplex virus and their replication. In B.N. Fields, D.M., Knipe, P.M. Howley, et al. (eds), Fields of virology. pp. 2231-2240. Phiadelphia: Lippincott-Raven.

Roizman, B. and Whitley, R. 2001. The nine ages of herpes simplex virus.Herpes 8 (1):23-27.

Saifah, E., Suttisri, R., Shamsub, S., Pengsuparp, T. and Lipipun, V. 1999 Bisamides from *Aglaia edulis*. Phytochemistry52:1085-1088.

Sangiporn, S., Polihan, K., Thawatsupa, P., Bunchok, M. and Chawalit thumrong, P. 1993 Treatment of recurrent genital herpes simplex virus infections with *Clinacanthus nutans* extract. Bull Dept. Med. Serv 18:226-231.

Sangiporn, S.,, Chiwat, S., Balachandra, K., Dechotiwongse Nu-Ayudhaya, T., Bunchok, M., and Jawasu, C. 1995 Treatment of herpes zoster with *Clinacanthus nutans* (BiPhya Yaw) extracts. J. Med Assoc. Thai. 78:624-627.

Safrin, S., Crumpacker, C., Chalis, P. et al. 1991 A controlled trial comparing foscarnet with vidarabine for acyclovir-resistant mucocutaneous herpes simplex in the acquired immunodeficiency syndrome. N Engl J Med 325:551-555.

Scholar, E. M. and Pratt, W. B., eds. 2000. The Antimicrobial Drugs. 2nd ed University Press., New York.

Serkedjieva, J. and Manolava, N. 1992 Plant polyphenolic complex inhibits the reproduction of influenza and herpes simplex virus. Basic Life Sci. 59:705-715.

Shin, Y.K., Cai, G.Y. Weinberg, A., Leary, J.J., Levin, M.J. 2001. Frequency of acyclovir-resistant herpes simplex virus in clinical specimens and laboratory isolates. J Clin Microbiol 39(3): 913-7.

Silver, S. and Roizman, B. 1985. Thymidine kinase chimeras are identically transcribed but regulated as $\gamma 2$ genes in herpes simplex virus genomes and as β genes in cell genomes. Mol Cell Biol 5: 518-528.

Smithnand, T. 1980 Thai plant names (Botanical names-Vernacular names)2nd ed Bangkok, Royal Forest Department , Ministry of Agriculture and Cooperation.

Sotanaphun, U., Lipipun, V., Suttisri, R., and *Pavovada, R. 1999 A new antiviral and antimicrobial sesquiterpenes from Glyptopetalum sclerocarpum.* Planta Med 65:257-258.

Stanberry, L.R. 1990 Capsaicin interferes with the centrifugal spread of virus in primary and current genital herpes simplex virus infections. J.Infect Dis. 162:29-34.

Stevens, J., Wagner, E., Devi-Rao, O., Godi, M., Seldman, L. 1987. RNA complementary to a herpesvirus alpha mRNA is prominent in latently infected neurons. Science 235: 1056-1059.

Sydiskis, R.J., Owen, D.G., Lohr, J.L. et al 1991 Inactivation of enveloped viruses by anthraquinones extracted from plants J Antimicrob Chemother35:2463-2466.

Tebas, P., Stabell, E. C., and Olivo, P. D. 1995. Antiviral susceptibility testing with a cell line which express beta-galactosidase after infection with herpes simplex virus. Antimicrob Agent Chemother 39: 1287-1291.

Thompson, K.D. 1998. Antiviral activity of viracea against acyclovir susceptible and resistant strains of herpes simplex virus. Antiviral Research 39: 55-61.

Unna, P.G.1886. The Histopathology of the Diseases of the Skin. : MacMillian.,New York:

Vermani, K. and Garg, S. 2002. Herbal medicines for sexually transmitted diseases and AIDS. J. of Ethnopharmacology 80(1): 49-66.

Vlietinck, A. J. and Vanden Berghe, D. A. 1991. Can ethnopharmacology contribute to the development of antiviral drugs?. J Ethnopharmacol 32: 141-153.

Whitley, R. J. and Roizman, B. 2001. Herpes simplex virus infections. Lancet 357 (9267): 1513-1518.

Whitley, R.J. Soong, S.J, Dolin, R., Galasso, G.J. Chien, L.T., Alford, C.A.,Jr,.1977. NIAID Collaborative Antiviral Study Group. Adenine arabinoside therapy of biopsy-proved herpes simplex encephalitis: National Institute of Allergy and Infectious Diseases Collaborative Antiviral Study. N Engl J Med 297:289-294.

Wolbling, R.H., Leonhardt, K. 1994 Local therapy of herpes simplex with dried extract of *Melissa officinalis* Phytomedicine 1:25-31.

Yip, L. Pei, S. Hudson, J. B., and Towers, G. H. N. 1991. Screening of medicinal plants from Yunnan province in southwest China for antiviral activity. J. Ethnopharmacol 34: 1-6.

Yoosook, C., Bunyapraphatsara, N., Boonyakiat, Y., Kantasuk, C. 2000 Antiherpes simplex virus activities of crude water extracts of Thai medicinal plants. Phytomedicines 6:411-419.

APPENDIX

1. Chemical agents and instruments

Chemical agents

Absolute methanol GR (E. Merck, Darmstadt, Germany)

Citric acid (E. Merck, Darmstadt, Germany)

Chlroform (Sigma, MO, U.S.A.)

Crystal violet (Fluka, Switzerland)

DMSO (dimethyl sulfoxide) (Sigma, MO, U.S.A.)

Formaldehyde 40% w/v AR (Carlo Erba, Milano, Italy)

Hexane (E. Merck, Darmstadt, Germany)

Potassium chloride (KCl) (M&B, England)

Potassium dihydrogen phosphate (KH_2PO_4) (E. Merck, Darmstadt, Germany)

Dipotassium hydrogen phosphate (K_2HPO_4) (E. Merck, Darmstadt, Germany)

Sodium bicarbonate($NaHCO_3$) (E. Merck, Darmstadt, Germany)

Disod hydrogen phosphate(Na_2HPO_4 anh) (E. Merck, Darmstadt, Germany)

Sulfuric acid) (Sigma, MO, U.S.A.)

Tragacanth (Pharmaceutical chemicals, Denmark)

Trypsin-EDTA(10x0.5/0.2%w/v)(Seromed, Germany)

Instruments

Analytical balance (Sartorius, Germany)

Automatic pipet, P10-100/p50-200(Socorex, Switzerland)

Centrifuge (Sigma, Germany)

Hemocytometer(Spencer, U.S.A.)

Laminar Air Flow (Holten, U.S.A.)

Mixer Vortex(Scientific, NY, USA.)

Multichannel automatic pipette , 8 channel(Socorex, Switzerland)

pH meter (Beckman, USA.)

Refrigerator4°C (Sharp, Thailand)

Refrigerator20°C (Ariston , USA.)

Refrigerator80°C (Forma Scientific, Ohio, USA)

Water bath (Thelco, USA)

Laboratory supplies

Cryotubes (Nunc, Denmark)

Glassware (Pyrex, USA)

Microcentrifuge tubes

Millipore filters 0.2 um (Gelman Sciences, USA)

Pipette tips (Nunc, Denmark)

Tissue culture flask(Nunc, Denmark)

Tisssue culture plate(Nunc, Denmark)

2. Medium and reagents

Growth Medium

Dissolved 9.5 g MEM powder with Earle's balance salts (Hyclone, Lot No. AJJ10788D, USA.), with L-glutamine (Sigma, Lot. No. 69H0740, USA.) without sodium bicarbonate in deionized distilled water and added 2.2g/l sodium bicarbonate (Sigma, USA). The solution was mixed well and adjusted pH to 7.2 with 6N HCl. Then, the solution was adjusted volume to 1,000 ml. This solution was sterilized by filtration (0.22 μm millipore filter membrane). Before use, this solution was supplemented with 10% fetal bovine serum (FBS, Biochrom AG, Lot No. 230B, Berlin, Germany) and 1% antibiotic-antimycotic agents (Gibco BRL,Lot. No. 1106811, USA.) which contained 10,000 units/ml penicillin G sodium, 10,000 μg/ml streptomycin sulfate and 25 μg/ml fungizone.

Maintenance Medium

This is similar to growth medium except the amount of FBS is reduced to 5%.

Phosphate Buffer Saline Solution (PBS)

NaCl (Merck, Lot No. K27736104021, Germany)	8.00	g
KCl (May&Baker, England)	0.20	g
KH_2PO_4 (Merk, Lot No. 547A17873, Germany)	0.20	g
Na_2HPO_4 (May&Baker, Lot No. 50028, England)	1.15	g

Deionized distilled water to	1,000	ml

This solution is sterilized by autoclave (121^{0}C, 15 psi for 15 min).

Trypsin-EDTA (1X)

Trypsin-EDTA (10X) (Biochrome AG, Lot. No. 521B, Germany)	10	ml
PBS	90	ml

Plaque Overlay medium

Solution A

2x MEM with 20% FBS and 2% antibiotic

This solution was sterilized by filtration (0.22 μm millipore filter membrane).

Solution B

Tragacanth (BP, 1973, Lot. No. TEC12 ศรีจันทร์สหโอสถ, กรุงเทพฯ)	2	g
Dionized distilled water	100	ml

This solution is sterilized by autoclave (121^{0}C, 15 psi for 15 min).

The solution A and B are mixed at a ratio of 1 : 1 before use.

Low-pH Citrate buffer, pH 3.0

Sodium citrate	40	mM
Potassium chloride	10	mM
Sodium chloride	135	mM

pH is adjusted to 3.0

0.05% Methylene blue in distilled water

methylene blue	5	g
distilled water	100	ml

Calculation of median cytotoxic concentration(CC_{50}), effective concentration(EC_{50}) and inhibitory concentration(IC_{50}) by regression formula.

CC_{50} is the concentration of plant extract which exhibit 50% cytotoxicity. CC_{50} is calculated from the regression formula Y = a+b X or Y = a+b log X; whenY = optical density, X = concentration of plant extract in μg/ml, a = intercept or the distance between X axis and the point where the regression line comed across Y axis, and b = slope.

EC_{50} is the concentration of plant extract which can inhibit 50% of plaque formation. EC_{50} is calculated from the regression formula Y = a+b X or Y = a+b log X; whenY = number of plaque, X = concentration of plant extract in μg/ml, a = intercept or the distance between X axis and the point where the regression line comed across Y axis, and b = slope.

IC_{50} is the concentration of an plant extract which can inhibit 50% of virus yield. IC_{50} is calculated from the regression formula Y = a+b X or Y = a+b log X; whenY = number of plaque, X = concentration of plant extract in μg/ml, a = intercept or the distance between X axis and the point where the regression line comed across Y axis, and b = slope.

Determination of correlation coefficient

Correlation coefficient between concentration and percent inhibition or incubation time and percent inhibition was determined using SPSS program as following example.

Concentration(ug/ml)	% inhibition	Correlation coefficient
100	10.58	R= 0.95
200	27.05	
400	48.77	
800	71.01	
1600	92.92	