

1 SUPPLEMENTARY DATA SHEETS

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6 **Attenuation of UVB-induced Photoaging by polyphenolic rich Spatholobus**
7 **suberectus stem extract via modulation of MAPK/AP-1/MMPs signaling in**
8 **human keratinocytes**

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Table S1: List of the primer sets used in this study

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<i>Gene name</i>		<i>Sequences</i>
<i>MMP-1</i>	<i>forward</i>	<i>CCTAGCTACACCTTCAGTGG</i>
	<i>reverse</i>	<i>GCCCAGTACTTATTCCCTTT</i>
<i>MMP-2</i>	<i>forward</i>	<i>GATACCCCTTTGACGGTAAGGA</i>
	<i>reverse</i>	<i>CCTTCTCCCAAGGTCCATAGC</i>
<i>MMP-7</i>	<i>forward</i>	<i>GTATGGGACATTCTCTGATCC</i>
	<i>reverse</i>	<i>CCAATGAATGAATGAATGGATG</i>
<i>MMP-9</i>	<i>forward</i>	<i>TGTACCGCTATGGTTACTCTCG</i>
	<i>reverse</i>	<i>GGCAGGGACAGTTGCTTCT</i>
<i>MMP-12</i>	<i>forward</i>	<i>CATGAACCGTGAGGATGTTGA</i>
	<i>reverse</i>	<i>GCATGGGCTAGGATTCCACC</i>
<i>TIMP-1</i>	<i>forward</i>	<i>TGGTGGGTGGATGAGTAATG</i>
	<i>reverse</i>	<i>CGCTGGTATAAGGTGGTCTG</i>
<i>TIMP-2</i>	<i>forward</i>	<i>ATGCACATCACCTCTGTGA</i>
	<i>reverse</i>	<i>CTCTGTGACCCAGTCCATCC</i>
<i>COL1A1</i>	<i>forward</i>	<i>GAGGGCCAAGACGAAGACATC</i>
	<i>reverse</i>	<i>CAGATCACGTCATCGCACAAAC</i>
<i>ELN</i>	<i>forward</i>	<i>CAAGGCTGGTTACCCAACAG</i>
	<i>reverse</i>	<i>CACCTGGGACAACCTGGAATC</i>
<i>HAS2</i>	<i>forward</i>	<i>GAGCACCAAGGTTCTGCTTC</i>
	<i>reverse</i>	<i>CTCTCCATACGGCGAGAGTC</i>
<i>Gapdh</i>	<i>Forward</i>	<i>TTGTGATGGGTGTGAACCAC</i>
	<i>reverse</i>	<i>ACACATTGGGGGTAGGAACA</i>

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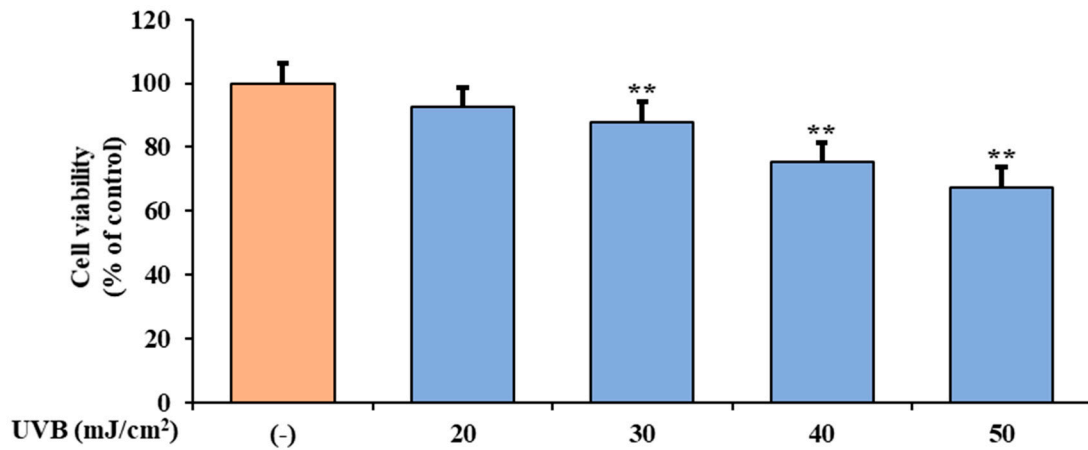
Table S2: List of antibodies used in this study

Name	Catalog number	Company	Antigen	Host
Anti-MMP1	sc-21731	Santa Cruz Biotechnology, Inc.	MMP-1	Mouse
Anti-MMP-2	BS90870	Bioworld Technology, Inc.	MMP-2	Rabbit
Anti-TIMP-1	BS1697	Bioworld Technology, Inc.	TIMP-1	Rabbit
Anti-type I procollagen	sc-376350	Santa Cruz Biotechnology, Inc.	COL1A1	Mouse
Anti-ELN	BS7427	Bioworld Technology, Inc.	ELN (Elastin)	Rabbit
Anti-HAS2	sc-365263	Santa Cruz Biotechnology, Inc.	HAS2	Mouse
Anti-p-p38	sc-166182	Santa Cruz Biotechnology, Inc.	p38	Mouse
Anti-p38	BS3567	Bioworld Technology, Inc.	p38	Rabbit
Anti-p-ERK1/2	sc-7383	Santa Cruz Biotechnology, Inc.	ERK	Mouse
Anti-ERK1/2	BS 6472	Bioworld Technology, Inc.	ERK	Rabbit
Anti-p-JNK	BS 4322	Bioworld Technology, Inc.	JNK	Rabbit
Anti-JNK	sc-7345	Santa Cruz Biotechnology, Inc.	JNK	Mouse
Anti-NF- B (p65)	BS1254	Bioworld Technology, Inc.	NF- B (p65)	Rabbit
Anti-p-c-Jun	BS4050	Bioworld Technology, Inc.	c-Jun (Y-170)	Rabbit
Anti-c-jun	MB0004	Bioworld Technology, Inc.	c-Jun	Rabbit
Anti Lamin B	Sc-6217	Santa Cruz Biotechnology, Inc.	Lamin B	Goat
Anti- actin	Sc-47778	Santa Cruz Biotechnology, Inc.	-actin	Mouse

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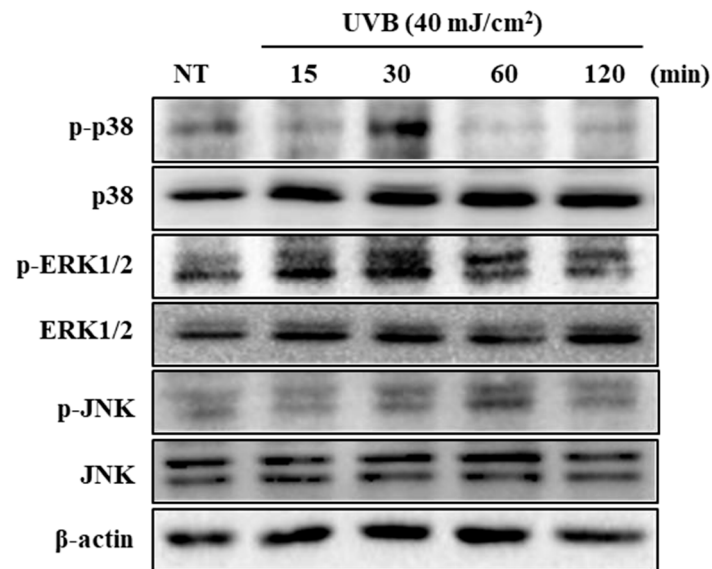


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60 **Figure S1.** Cell viability UVB-irradiation. HaCaT cells (1×10^5 cells/mL) were seeded in a 96-well
61 plate and treated with UVB-irradiation at 20,30,40 and 50 mJ/cm². The cell viability was determined
62 using the MTT assay.

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66 **Figure S2.** The effects of UVB-irradiation on the phosphorylation of MAP kinase in HaCaT
67 cells. HaCaT cells (1×10^5 cells/mL) were seeded in a 6-well plate and treated with
68 UVB irradiation at 40 mJ/cm² and the protein was extracted after 15, 30, 60 and 120 min Protein
69 expression was confirmed by western blot.

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CompuSyn Report

Experiment Name: combination effect

Date:

File Name: E:\koori\combination report.cse

Description

Drug: syringic acid (SA) [ug/mL]

Drug: Epicatechin (EP) [ug/mL]

Drug: Vanilic acid (VA) [ug/mL]

Drug Combo: combination 1 (SAEP) (SA+EP)

Drug Combo: combination 2 (EPVA) (EP+VA)

Drug Combo: combination 3 (SAVA) (SA+VA)

Drug Combo: combination 4 (SEV) (SA+EP+VA)

88

89 Data for Drug: SA [ug/mL] **Dose Effect**

0.25 0.18

0.5 0.26

1.0 0.31

90 3 data points entered.

X-int: 0.64112

Y-int: -0.3312 +/- 0.03634

m: 0.51665 +/- 0.09351

Dm: 4.37645 **r:**
0.98401

91

92 Data for Drug: EP [ug/mL] **Dose Effect**

0.25 0.2

0.5 0.24

1.0 0.3

93 3 data points entered.

X-int: 0.95979

Y-int: -0.3732 +/- 0.01162

m: 0.38880 +/- 0.02989

Dm: 9.11578 **r:**
0.99706

94

95 Data for Drug: VA [ug/mL] **Dose Effect**

0.25 0.15

0.5 0.2

Dose Effect

1.0 0.28

96 3 data points entered.

X-int: 0.73153
Y-int: -0.4169 +/- 0.01514
m: 0.56997 +/- 0.03895
Dm: 5.38922 **r:**
 0.99767

97
 98 Data for Non-Constant Combo: SAEP (SA+EP)

Dose SA Dose EP Effect

1.0 9.0 0.51 1.0
 4.0 0.45 1.0
 2.0 0.39
 1.0 1.0 0.38 99

4 data points entered.

100
 101 Data for Non-Constant Combo: EPVA (EP+VA)

Dose EP Dose VA Effect

9.0 1.0 0.49 4.0
 1.0 0.45 2.0
 1.0 0.41
 1.0 1.0 0.36 102

4 data points entered.

103
 104 Data for Non-Constant Combo: SAVA (SA+VA)

Dose SA Dose VA Effect

1.0 9.0 0.52
 1.0 4.0 0.5
 1.0 2.0 0.55 1.0
 1.0 0.58 2.0
 1.0 0.57 4.0
 1.0 0.59
 9.0 1.0 0.62 105

7 data points entered.

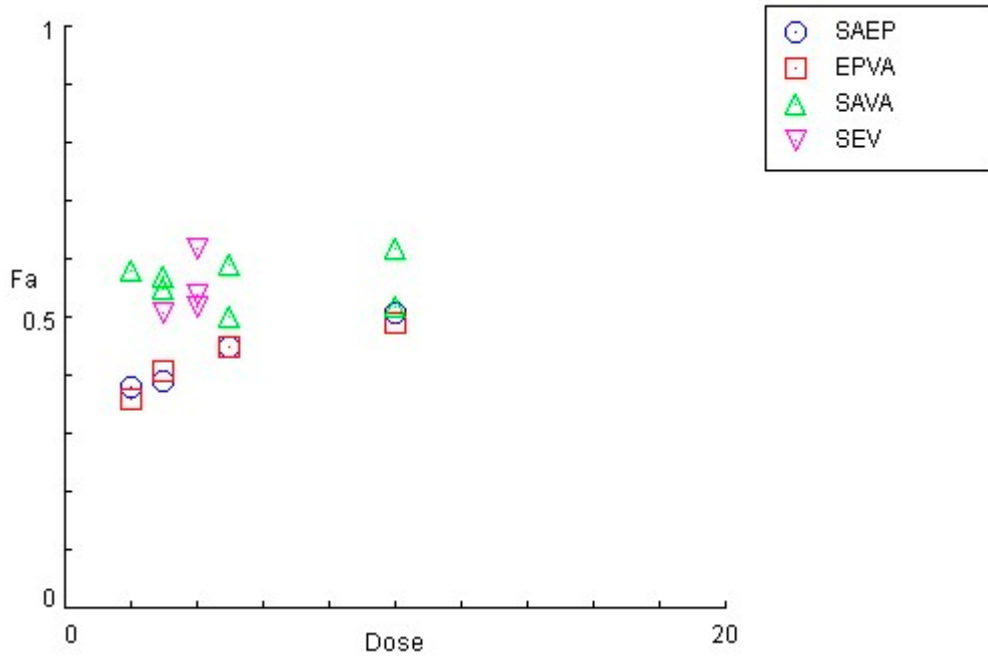
106
 107 Data for Non-Constant Combo: SEV (SA+EP+VA)

Dose SA Dose EP Dose VA Effect

1.0 1.0 1.0 0.51 2.0
 1.0 1.0 0.62 1.0
 1.0 2.0 0.54
 1.0 2.0 1.0 0.52 108 4

data points entered.

Dose-Effect Curve for Drug Combos



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112 CI Data for Non-Constant Combo: SAEP (SA+EP)

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Dose SA	Dose EP	Effect	CI
1.0	9.0	0.51	1.10223
1.0	4.0	0.45	1.07216
1.0	2.0	0.39	1.23635
1.0	1.0	0.38	0.97577

114

115 CI Data for Non-Constant Combo: EPVA (EP+VA)

116

Dose EP	Dose VA	Effect	CI
9.0	1.0	0.49	1.29334
4.0	1.0	0.45	0.99908
2.0	1.0	0.41	0.91088
1.0	1.0	0.36	0.99102

117

118 CI Data for Non-Constant Combo: SAVA (SA+VA)

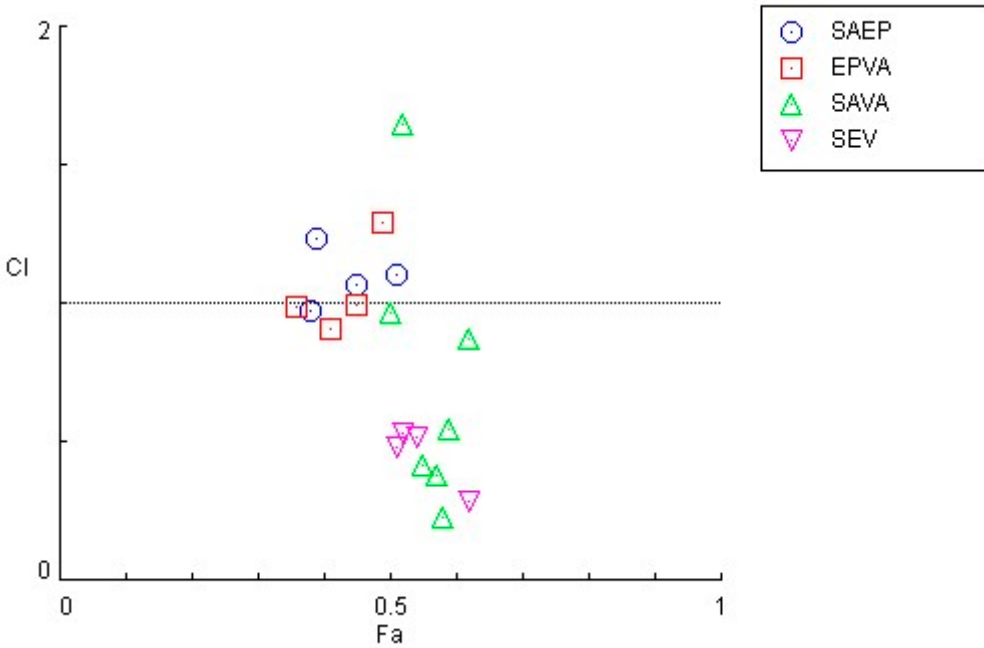
119

Dose SA	Dose VA	Effect	CI
1.0	9.0	0.52	1.64690
1.0	4.0	0.5	0.97072 1.0
2.0	0.55	0.41593	1.0
1.0	0.58	0.22766	2.0
1.0	0.57	0.37801	4.0
1.0	0.59	0.54983	
9.0	1.0	0.62	0.87588

119
 120 CI Data for Non-Constant Combo: SEV (SA+EP+VA)

Dose SA	Dose EP	Dose VA	Effect	CI
1.0	1.0	1.0	0.51	0.48342
	1.0	1.0	0.62	0.28692
	1.0	2.0	0.54	0.52027
1.0	2.0	1.0	0.52	0.53552

121
 122 Combination Index Plot



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 124
 125 DRI Data for Non-Constant Combo: SAEP (SA+EP) Fa Dose SA

Dose EP	DRI SA	DRI EP	Fa	Dose SA
0.51	4.72880	10.1037	4.72880	1.12263
2.96783	5.44057	2.96783	1.36014	0.39
1.84125	2.88500	1.84125	1.44250	0.38
1.69671	2.58802	1.69671	2.58802	

126
 127 DRI Data for Non-Constant Combo: EPVA (EP+VA) Fa Dose EP

Dose VA	DRI EP	DRI VA	Fa	Dose EP
0.49	8.22447	5.02392	0.91383	5.02392
5.44057	3.78984	1.36014	3.78984	0.41
3.57473	2.84575	1.78737	2.84575	0.36
2.07544	1.96389	2.07544	1.96389	

128
 129 DRI Data for Non-Constant Combo: SAVA (SA+VA) Fa Dose SA

Dose VA	DRI SA	DRI VA
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0.52	5.10982	6.20177	5.10982	0.68909	
Fa	Dose SA	Dose VA	DRI SA	DRI VA	
0.5	4.37645	5.38922	4.37645	1.34730	
0.55	6.45365	7.66355	6.45365	3.83178	0.58
8.17419	9.49442	8.17419	9.49442	0.57	
7.55172	8.83663	3.77586	8.83663	0.59	
8.85260	10.2060	2.21315	10.2060		
0.62	11.2885	12.7217	1.25428	12.7217	

130

131 DRI Data for Non-Constant Combo: SEV (SA+EP+VA)

Fa	Dose SA	Dose EP	Dose VA	DRI SA	DRI EP	DRI VA
0.51	4.72880	10.1037	5.78107	4.72880	10.1037	5.78107
	0.62	11.2885	32.1085	12.7217	5.64424	32.1085
	12.7217	0.54	5.96906	13.7688	7.14005	5.96906
	13.7688	3.57003				
0.52	5.10982	11.1996	6.20177	5.10982	5.59979	6.20177

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Summary Table

Experiment Name: combination effect

Date:

File Name: E:\koori\combination report.cse

Description

Drug: syringic acid (SA) [ug/mL]

Drug: Epicatechin (EP) [ug/mL]

Drug: Vanilic acid (VA) [ug/mL]

Drug Combo: combination 1 (SAEP) (SA+EP)

Drug Combo: combination 2 (EPVA) (EP+VA)

Drug Combo: combination 3 (SAVA) (SA+VA)

Drug Combo: combination 4 (SEV) (SA+EP+VA)

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Drug/Combo	Dm	m	r
SA	4.37645	0.51665	0.98401
EP	9.11578	0.38880	0.99706
VA	5.38922	0.56997	0.99767

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CI values at:
Combo ED50 ED75 ED90 ED95

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Data for Fa = 0.5
Drug/Combo CI value Dose SA Dose EP Dose VA

SA	4.37645	
EP	9.11578	
VA		5.38922

140

141 Data for Fa = 0.75

Drug/Combo CI value Dose SA Dose EP Dose VA

SA	36.6956	
EP	153.796	
VA		37.0366

142

143 Data for Fa = 0.9

Drug/Combo CI value Dose SA Dose EP Dose VA

SA	307.684	
EP	2594.76	
VA		254.529

144

145 Data for Fa = 0.95

Drug/Combo CI value Dose SA Dose EP Dose VA

SA	1306.81	
EP	17731.2	
VA		944.257

146

147 Data for Fa = 0.97

Drug/Combo CI value Dose SA Dose EP Dose VA

SA	3656.99	
EP	69598.8	
VA		2399.92

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