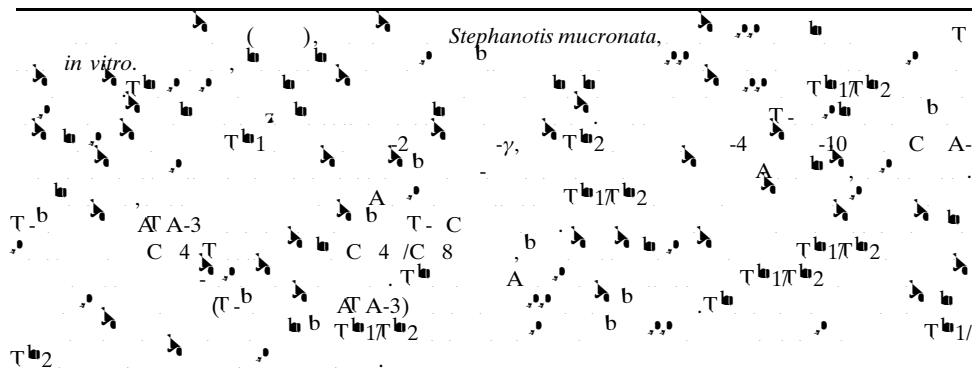


Stemucronoside L, a Pregnane Glycoside from the Roots of *Stephanotis mucronata*, Inhibits Th1/Th2 Immune Responses *in vitro*

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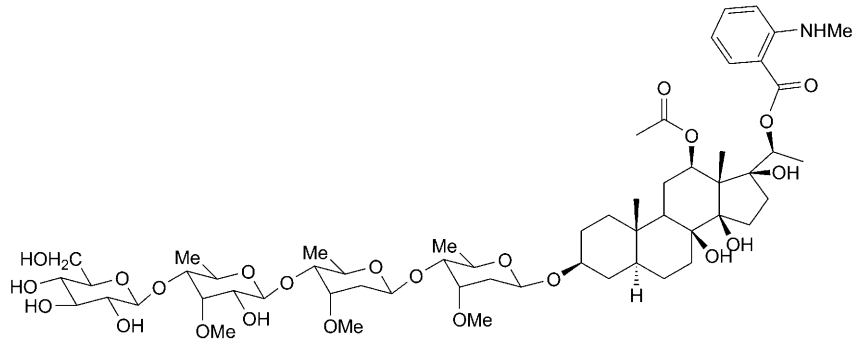
Introduction. *Stephanotis mucronata* (BLANCO) ERR. (A) (*Fig. 1*)

in vitro 2, 6, (;

A (C A)-

in vitro 6 .

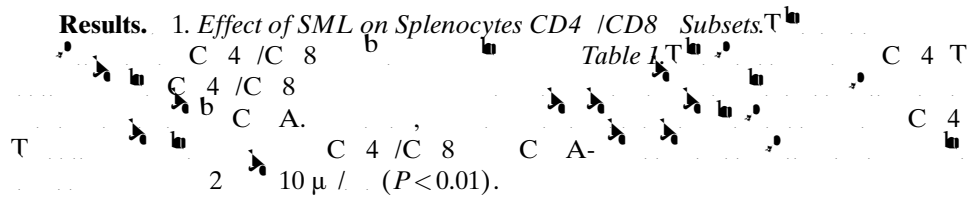
Th1- C 4 /C 8 b , Th1- Th2- (-γ, -2,



Formula: $C_{58}H_{91}NO_{23}$, M_r : 1192.5914

1. Chemical structure of stemucronatoside L ()

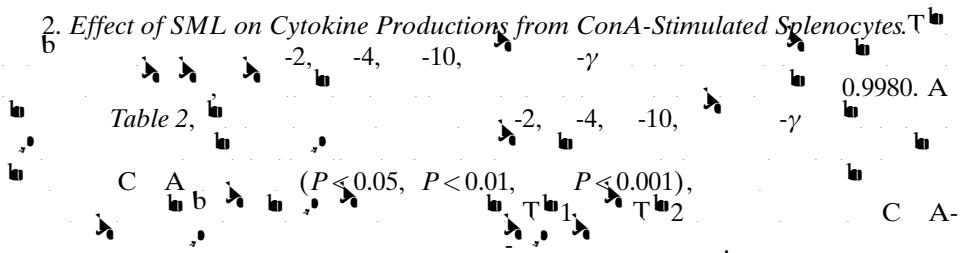
Results. 1. Effect of SML on Splenocytes CD4 /CD8 Subsets.



1. Effects of Stemucronatoside L () on Splenocytes CD4 /CD8 Subsets.

5900 % 42912195.0899486.4818 (/ 3 7.9 06664. 43257.222483.7605 (/ 5 2.283.95939.24.9969 (5900 % 425750254.83

2. Effect of SML on Cytokine Productions from ConA-Stimulated Splenocytes.



2. Effects of Stemucronatoside L () on Cytokine Production from Con A-Stimulated Mice Splenocytes.

(3 μ /) 24 T b A.T ± (-2, -4, -10, -γ (n 3).

	C	3 μ /	10 μ /	24 μ /
C	20±10	2.33±0.33	20±1	936±197
C A	636±15	9.13±0.11	204±18	3366±265
C A± (0.08 μ /)	495±58	6.35±0.57 ^b	165±5	1931±216 ^b
C A± (0.4 μ /)	488±51 ^b	5.92±0.66 ^b	146±15	1266±127
C A± (2.0 μ /)	452±41 ^b	4.83±0.88 ^b	135±8 ^b	1241±91
C A± (10 μ /)	301±56	2.60±0.28	76±15	1011±63

) P<0.05, ^b) P<0.01, ^c) P<0.001.

3. Effect of SML on Expression of Cytokines and Transcription Factor mRNAs in ConA-Stimulated Splenocytes.

Fig. 2 Table 3. (P<0.05, P<0.01, P<0.001), A A-3 A (P<0.05, P<0.01, P<0.001)

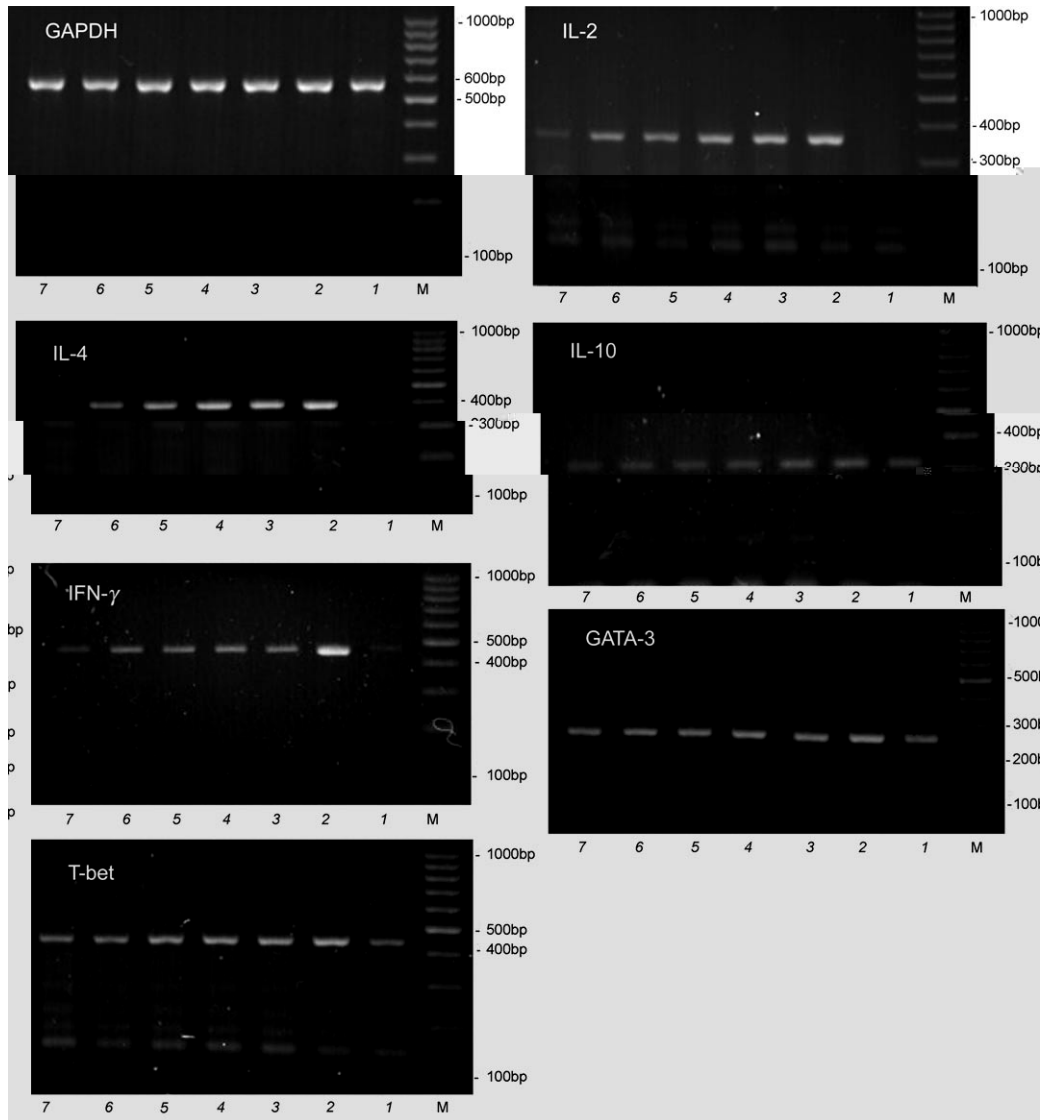
Discussion.

3. The mRNA Expression Level of Cytokines and Transcription Factors in Mice Splenocytes Treated with Stemucronatoside L () and Con A.

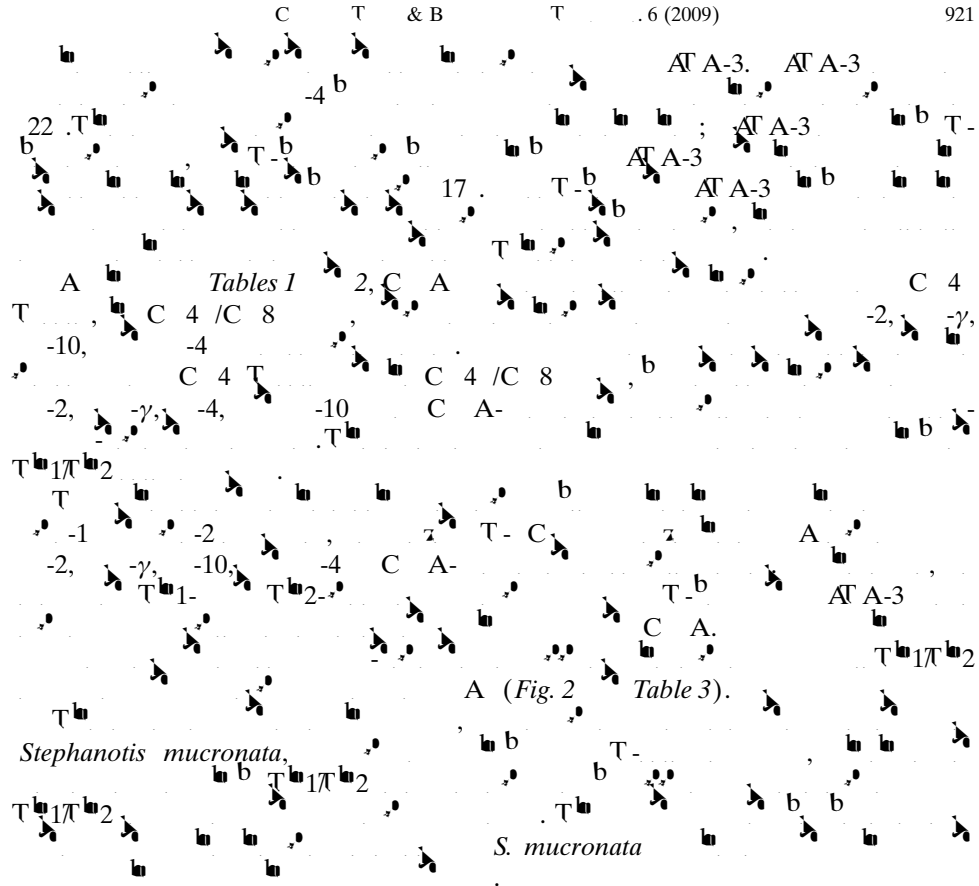
() 0 10 μ / C A (3 μ /) 16 T b A A-3 A ± (n 3).

	C	μ /					
	0	0.016	0.08	0.4	2	10	
-2	0.49±0.01	0.42±0.02	0.41±0.02 ^b	0.30±0.03	0.28±0.04	0.09±0.01	
-γ	0.45±0.03	0.21±0.03	0.19±0.03	0.18±0.03	0.15±0.03	0.09±0.02	
T b	0.39±0.03	0.33±0.01	0.33±0.01	0.33±0.01	0.27±0.02 ^b	0.22±0.02	
-4	0.52±0.04	0.42±0.04	0.41±0.01	0.30±0.01	0.28±0.01	0.05±0.01	
-10	0.39±0.02	0.34±0.02	0.26±0.01	0.22±0.01	0.21±0.01	0.13±0.01	
A A-3	0.51±0.07	0.38±0.03	0.37±0.01	0.37±0.05	0.37±0.02	0.27±0.01 ^b	

) P<0.05, ^b) P<0.01, ^c) P<0.001.



2. The mRNA expression level of GAPDH, cytokines and transcription factors in mice splenocytes treated with stemucronatoside L () and ConA. Lane M: A, Lane 1: A, Lane 2: C A, Lane 3: C A (0.016 μ /), Lane 4: C A (0.08 μ /), Lane 5: C A (0.4 μ /), Lane 6: C A (2 μ /), Lane 7: C A (10 μ /).



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 Zhejiang Provincial Medicinal Health Program of China (2006-2009) (004)

Experimental Part

General. C (TT) A, (C A) 3-(4,5-)-2,5-)-2H- Sigma Chemical Co., A; RPMI 1640 Gibco BRL, A; (TC)- C 4 (3T4, 129.19) ()- C 8 (-2, 53-6.7) BD Biosciences Pharmingen, CA, A; (-2, -4, -10) A Wuhan Boster Biological Technology, Ltd., Trizol Invitrogen, CA, A; C Shanghai Sangon Biological Engineering Technology & Services Co., Ltd., (B) Hangzhou Sijiqing Corp.,

Extraction, Isolation, and Identification of SML.

1192.5914) *Stephanotis mucronata* (; C₅H₉O₁ 23; M : 6, 1, 1, 1-C, C, BC).T b >99% b C

RPMI-1640 0.1%

Experimental Animals. Zhejiang Experimental Animal Center (C 2003-0001, ad libitum, 24±1%, 50±10%, 12/12)

Institute for Experimental Animals, Committee for Animal Experiments.

Preparation of Splenocytes. Hank's B (B; Sigma), (0.8% (w/v)). A (1500×g 4°

(RPMI 1640 100 μ / 10% C). C b 95%.

Flow Cytometry. 3524) 5×10⁶ / 1 (RPMI 1640 0.016, 0.08, 0.4, 2, 10 μ /) 5% C₂ 37°

CellQuest 3.0f (BD Biosciences Pharmingen, CA, A)

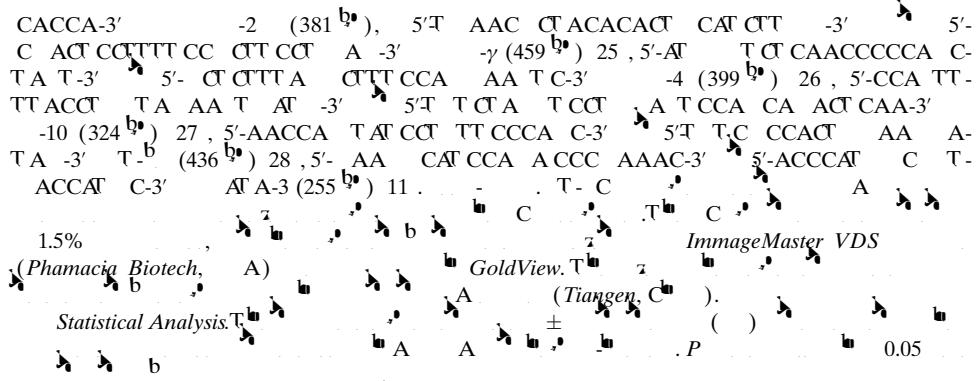
Measurement of Cytokines. 1400×g 5, 96-1.5 (b γ 2¹⁰). 1 b 37° (ABC). A 15 T 30 100 μ T B (b b) 37°

RT-PCR for Cytokine and Transcription-Factor Gene Expression. 5×10⁶ / 4 (RPMI 1640 0.016, 0.08, 0.4, 2, 10 μ /) 5% C₂ 37°

M-MuLV Reverse-Transcriptase (Fermentas, C 0441). A 1 T A, 10 M

PTC-200 Thermal Cycler (MJ Research, A) 58° (-4, -10, 94° 2, 55° (-2, A A-3, T-b), 57°

CCCACA T AAATT CAAC CAC-3' 5'-CATT TTA AACAC A-3' A (70 bp) 24, 5'-CTCTACA C AA CACA C-3' 5'-CATCTCTCA AAA TC-



C

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