(19)

(12)

(10)
(45)

(21) 201810968867 .4

(22) 2018 .08 .23

(65)

1. C
C 1:50 800
2. 1 C
1:50
3. 1
4. 1 3

5. 4

6. 4

7. 4

8. 4

[0001]

[0002] (onconase ,ONC)
104 ONC
RNA
ONC
ONC

[0003] (HCC))

(TACE)
3

[0004]

[0005] C

[0006]

[0007] C

[0008] (onconase ,ONC)

[0009] C(vitamin C ,VC)
VC ONC VC

[0010] RH 35

[0011] ONC VC RH 35 ONC VC

ONC VC

[0012] CI ONC VC
ONC
RNA(tRNA) DNA
RNA VC

[0013] ONC VC
RH 35
[0014] C 1:50 800 1:50
[0015] C 1:50 1:100 1:200 1:
300 1:400 1:500 1:600 1:700 1:800
[0016]
[0017]

[0018]

[0019]

[0020]
[0021]

[0022]
[0023]
[0024]
[0025]
[0026]
[0027] /

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[0031]

[0032] 1 1 ONC

[0033] 2 1 24

[0034] 3 1 24 Hoechst

[0035] 4 1 ONC VC RH 35

[0036] 5 1 ONC VC

[0037] 6 2

[0038]

[0039] 1

[0040] 1

[0041] 1.1

[0042] ONC (95 90 1 1 1 2 3 4 5 6 7 M protein ladder) VC ( ) RH 35 ( ) DMEM FBS(Gibico) B(Sulforhodamine B,SRB,Sigm) PE Annexin V Apoptosis Detection Kit I(BD Pharmingent)

[0043] 1.2

[0044] 1.2.1 RH 35 10 FBS 100U/mL (Invitrogen,USA) DMEM 37 5 CO_2 0.25 (0.02 EDTA) 2 3d 1

[0045] 1.1.2ONC VC 0.25 96 (5× 10^3 / /90μL) 37 5 CO_2 24h 10μL VC(800 1200 1600 2000 2400μmol/L) ONC(2 4 8 12 16μmol/L) 10μL PBS 5 3 24h ( ) SRB (inhibition rate,RI) (A A)/ A ×100 IC_{50}(inhibitory concentration 50,IC_{50})

[0046] 1.1.3ONC VC (IC_{50}) 4:1 3:2 2:3 1:4 4 PBS 6 1.1.2 96 37 5 CO_2 24h 10μL

10μL PBS 5 3 24h ( ) SRB

2 IC_{50} (ONC VC)

[0047] 1.1.4Hoechst 1.1.2 1.1.3 24h 4 15min PBS 3 5min/ hoechst33258 10min PBS 3 5min/

[0048] 1.1.5 (combination index,CI) CI $C_{50A}/IC_{50A}+C_{50B}/IC_{50B}$ C_{50A} C_{50B} A B 50 IC_{50A} IC_{50B} 50 CI 1 CI 1 CI 1

[0049] 1.1.6 (fractional inhibitory concentration,FIC) FIC_{ONC} 50 ONC (C_{50ONC})/ ONC 50 (IC_{50ONC}) FIC_{VC} 50 VC (C_{50VC})/ VC 50 (IC_{50VC}) FIC_{ONC} FIC_{VC} FIC 1 ( ) ONC FIC VC FIC

[0050] 2

[0051] 2.1 24h 2 2 NC PBS ONC (8μmol/L) VC (1600μmol/L) ONC VC (0:V 1:4 0:V 2:3 0:V 3:2 0:V 4:1) ONC VC

[0052] 2 24h ( )

[0053] 2.2 Hoechst 3 3 NC PBS ONC 8μmol/L VC 1600μmol/L ONC/VC (0:V 1:4 0:V 2:3 0:V 3:2 0:V 4:1) 3 (Fig 3)

[0054] 2.3ONC VC RH 35

[0055] SRB ONC(2 16μmol/L) VC(800 2400μmol/L) RH 35 24h (P 0.01) ONC VC IC_{50} 8μmol/L 1600μmol/L(Fig 4)

[0056] 2.4

SRB ONC VC

IC_{50} 1

[0058] 1ONC VC IC_{50}

[0059]

Cell line	V_{ONC}: V_{VC}	$IC_{50\,ONC}$ (μmol/L)	$IC_{50\,VC}$ (μmol/L)
RH-35	1:4	0.94	1059
	2:3	1.79	976
	3:2	2.42	883
	4:1	3.92	763

[0060] 1 4 IC_{50} CI CI 0 .78 0 .83 0 .85 0 .97 1 2

[0061] 2ONC VC RH 35

[0062]

Cell line	V_{ONC}: V_{VC}			
	1:4	2:3	3:2	4:1
RH-35	0.78	0.83	0.85	0.97

[0063] (5) ONC VC RH 35 ONC VC 4:1 IC_{500NC} IC_{50VC} 3 .92μmol/L 763μmol/L

[0064] 2

[0065]

[0066] 2×10^5/mL 6 24h 4 (1) PBS (2) ONC 8μmol/L (3) VC 1600μmol/L (4) ONC 8μmol/L+VC1600μmol/L 24h PBS (BD ,USA)

[0067] ± SPSS 13 .0 P 0 .01 P 0 .001

[0068] 6 PBS (ONC VC O+V) 4 .24 ± 0 .36 15 .73 ± 3 .08 61 .53 ± 2 .98 73 .65 ± 3 .38 ONC VC (P<0 .01) (P<0 .001)

[0069] ( ) ( ) ONC VC RH 35

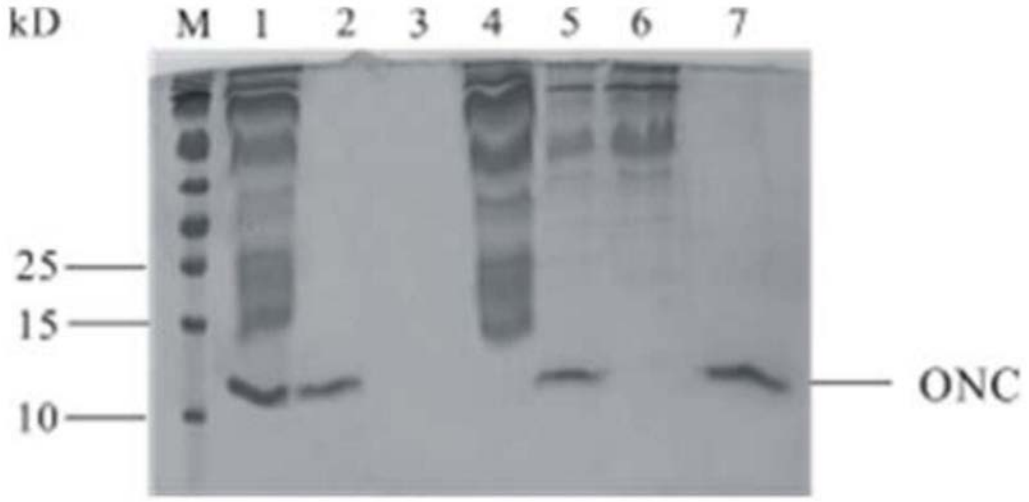
FCM (P 0 .01)

(P 0 .001) VC ONC RH 35

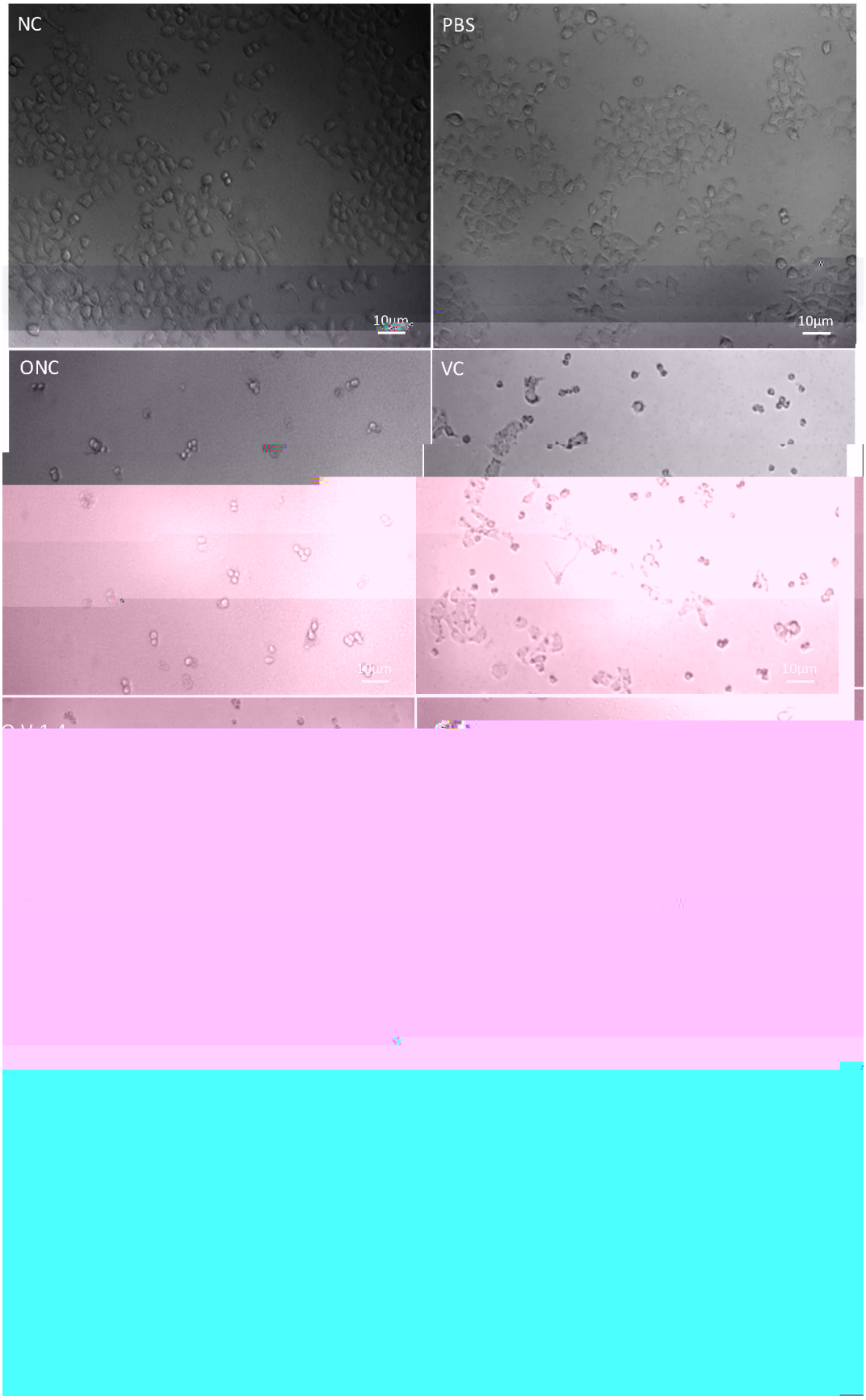
[0070] RH 35 SRB FCM ONC VC RH 35

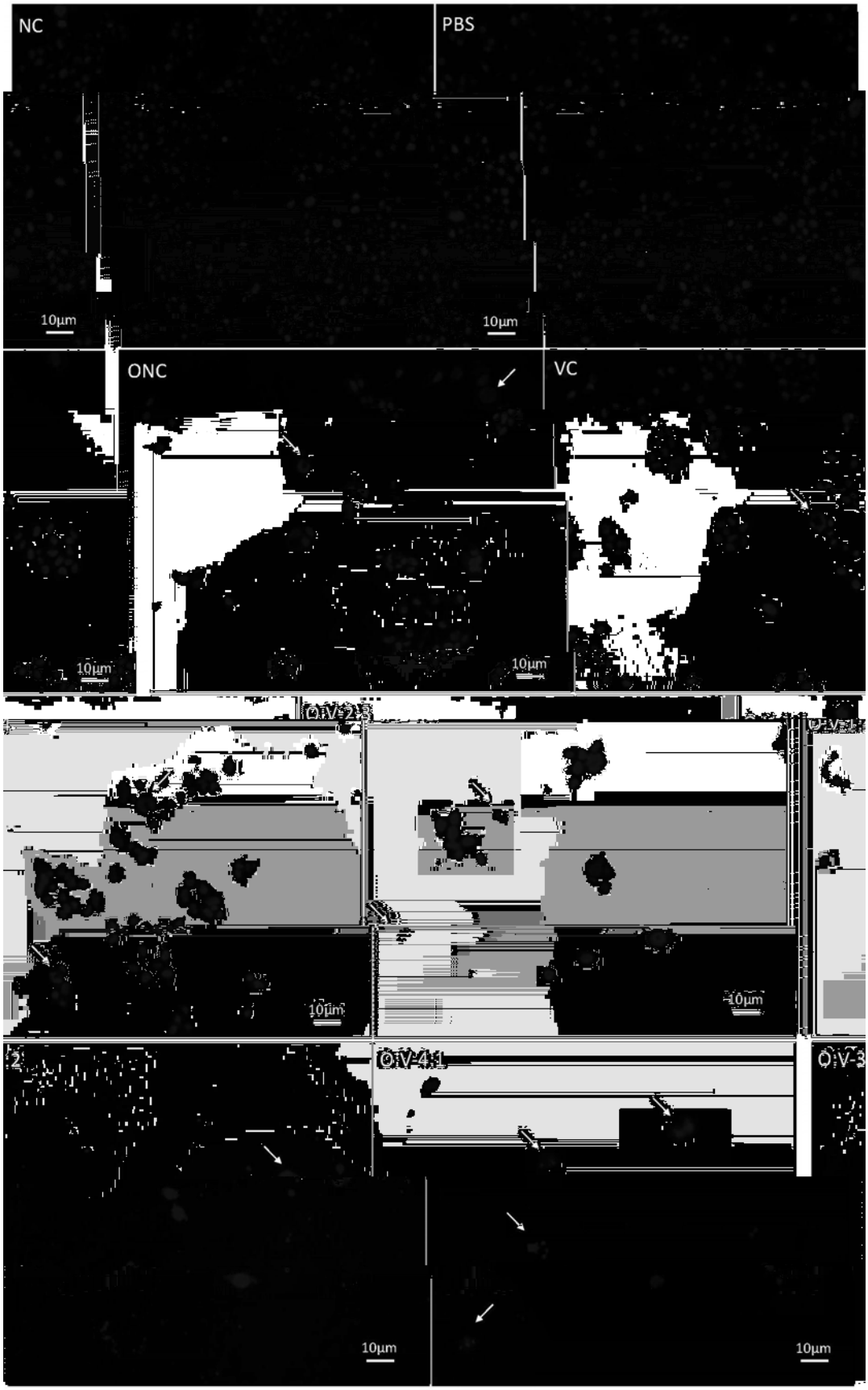
RH 35

[0071]

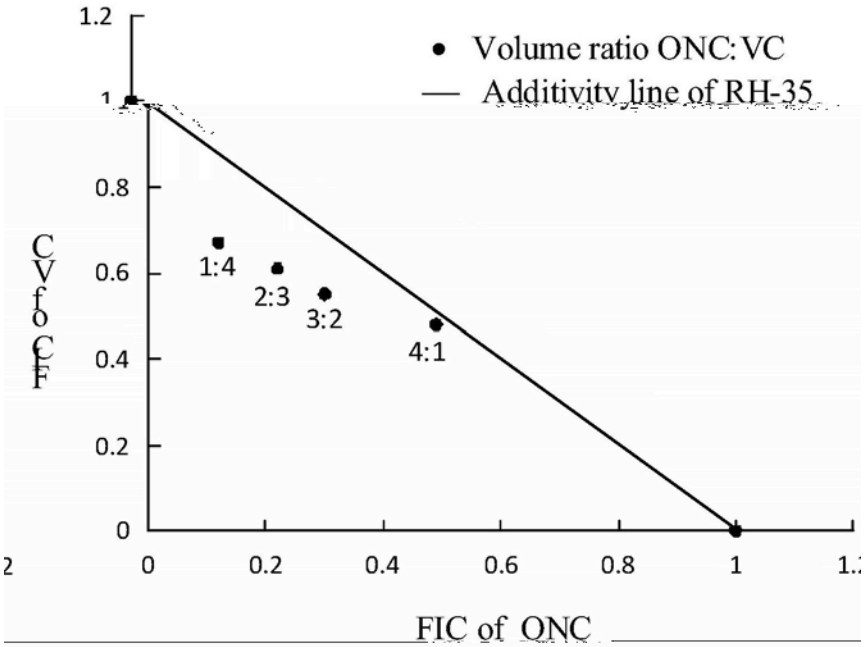


1

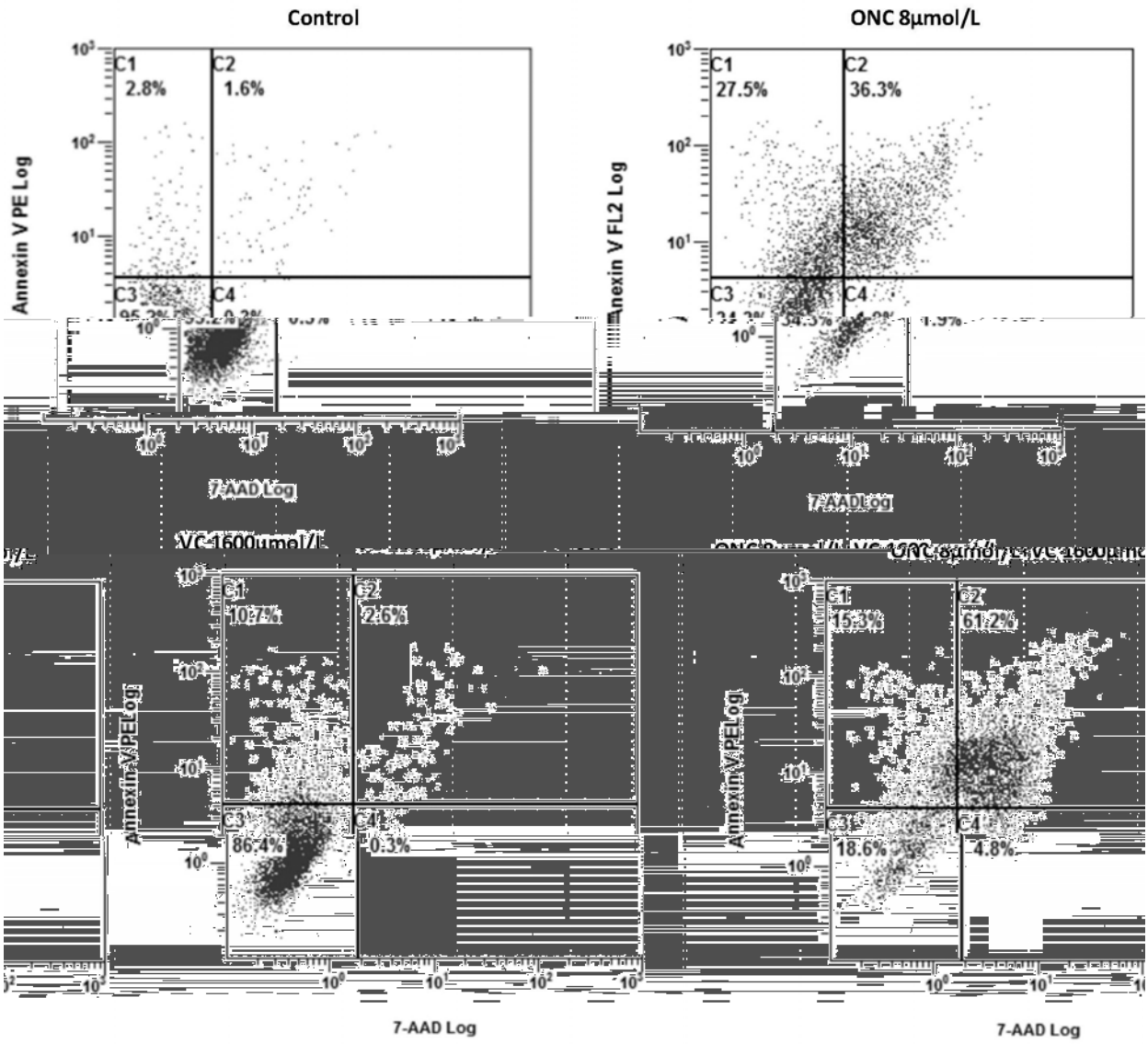

NC
10μm
PBS
10μm
ONC
VC
10μm
10μm

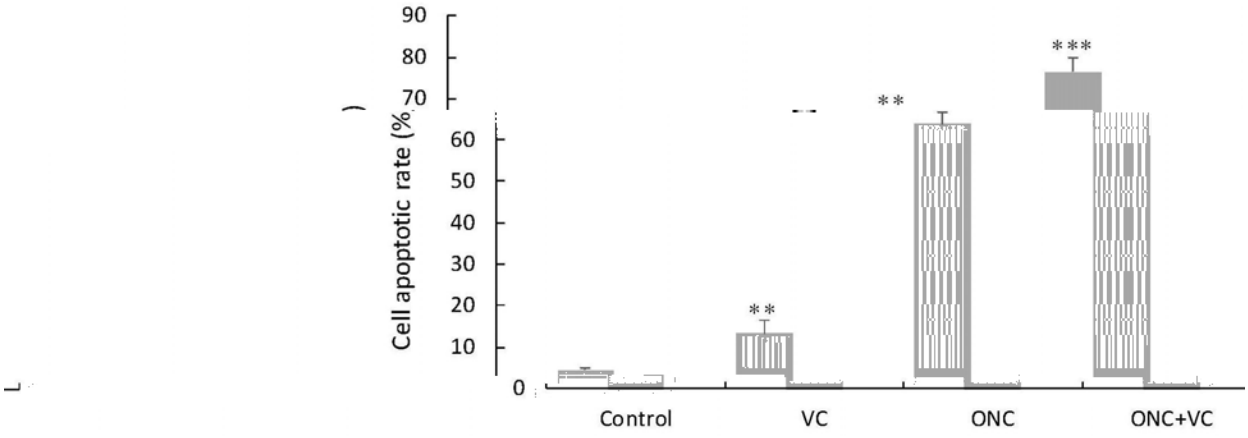

NC
PBS
10μm
10μm
ONC
VC
10μm
10μm
10μm
10μm
10μm
10μm

4


Volume ratio ONC:VC
Additivity line of RH-35
1:4
2:3
3:2
4:1
1.2
1
0.8
0.6
0.4
0.2
0
0
0.2
0.4
0.6
0.8
1
FIC of ONC

5


Control
ONC 8μmol/L
C1
2.8%
C2
1.6%
C3
C4
C1
27.5%
C2
36.3%
C3
C4
Annexin V PE Log
Annexin V FL2 Log
7-AAD Log
VC 1600umol/L
C1
10.7%
C2
2.6%
C3
86.4%
C4
0.3%
C1
15.3%
C2
61.2%
C3
18.6%
C4
4.8%
7-AAD Log
7-AAD Log


Cell apoptotic rate (%
90
80
70
60
50
40
30
20
10
0
**
**
***
Control
VC
ONC
ONC+VC