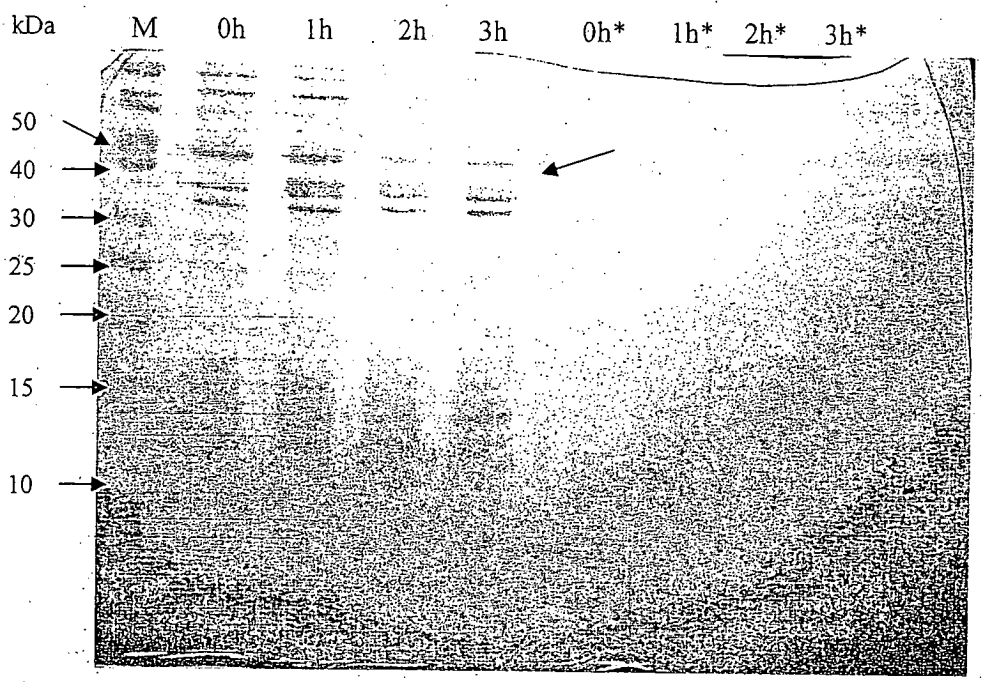
```
   1 ATGAAATACCTGCTGCCGACCGCTGCTGCTGGTCTGCTGCTCCTCGCTGCCCAGCCGGCG
   1  M  K  Y  L  L  P  T  A  A  A  G  L  L  L  L  A  A  Q  P  A
  61 ATGGCCATGGGGCGTATTTGCCGTTGCATTTGCGGCCGTGGCATTTGCCGCTGCATCTGT
  21  M  A  M  G  R  I  C  R  C  I  C  G  R  G  I  C  R  C  I  C
 121 GGCGTGCCGGGTGTTGGTGTTCCGGGTGTGGGTGGTGCGACCGGATCCGATGTGAACTTT
  41  G  V  P  G  V  G  V  P  G  V  G  G  A  T  G  S  D  V  N  F
 181 GATCTGAGCACCGCGACCGCGAAAACCTATACCAAATTCATCGAAGATTTTCGTGCGACC
  61  D  L  S  T  A  T  A  K  T  Y  T  K  F  I  E  D  F  R  A  T
 241 CTGCCGTTTAGCCATAAAGTGTATGATATCCCGCTGCTGTATAGCACCATTAGCGATAGC
  81  L  P  F  S  H  K  V  Y  D  I  P  L  L  Y  S  T  I  S  D  S
 301 CGTCGTTTTATTCTGCTGGATCTGACCAGCTATGCGTATGAAACCATTAGCGTGGCGATT
 101  R  R  F  I  L  L  D  L  T  S  Y  A  Y  E  T  I  S  V  A  I
 361 GATGTGACCAACGTGTATGTGGTGGCGTATCGTACCCGTGATGTGAGCTACTTTTTCAAA
 121  D  V  T  N  V  Y  V  V  A  Y  R  T  R  D  V  S  Y  F  F  K
 421 GAAAGCCCGCCGGAAGCGTACAACATTCTGTTTAAAGGCACCCGTAAAATTACCCTGCCG
 141  E  S  P  P  E  A  Y  N  I  L  F  K  G  T  R  K  I  T  L  P
 481 TATACCGGCAACTATGAAAACCTGCAGACCGCGGCGCATAAAATTCGTGAAAACATCGAT
 161  Y  T  G  N  Y  E  N  L  Q  T  A  A  H  K  I  R  E  N  I  D
 541 CTGGGCCTGCCGGCCCTGAGCAGCGCGATTACCACCCTGTTTTATTATAACGCGCAGAGC
 181  L  G  L  P  A  L  S  S  A  I  T  T  L  F  Y  Y  N  A  Q  S
 601 GCGCCGAGCGCGCTGCTGGTGCTGATTCAGACCACCGCGGAAGCGGCGCGTTTTAAATAT
 201  A  P  S  A  L  L  V  L  I  Q  T  T  A  E  A  A  R  F  K  Y
 661 ATTGAACGCCACGTGGCGAAATATGTGGCGACCAACTTTAAACCGAACCTGGCCATTATT
 221  I  E  R  H  V  A  K  Y  V  A  T  N  F  K  P  N  L  A  I  I
 721 AGCCTGGAAAACCAGTGGAGCGCCCTGAGCAAACAAATTTTTCTGGCCCAGAACCAGGGC
 241  S  L  E  N  Q  W  S  A  L  S  K  Q  I  F  L  A  Q  N  Q  G
 781 GGCAAATTTCGTAATCCGGTGGATCTGATTAAACCGACCGGCGAACGTTTTCAGGTGACC
 261  G  K  F  R  N  P  V  D  L  I  K  P  T  G  E  R  F  Q  V  T
 841 AATGTGGATAGCGATGTGGTGAAAGGCAACATTAAACTGCTGCTGAACAGCCGTGCGAGC
 281  N  V  D  S  D  V  V  K  G  N  I  K  L  L  L  N  S  R  A  S
 901 ACCGCGGATGAAAACTTTATTACCACCATGACCCTGCTGGGCGAAAGCGTGGTGGAATTC
 301  T  A  D  E  N  F  I  T  T  M  T  L  L  G  E  S  V  V  E  F
 961 CCGTGGGCGCTGTGGAAAACCATGCTGAAAGAACTGGGCACGATGGCGCTGCATGCGGGT
 321  P  W  A  L  W  K  T  M  L  K  E  L  G  T  M  A  L  H  A  G
1021 AAAGCGGCGCTGGGTGCGGCAGCGGATACCATTAGCCAGGGCACCCAGGTTCCGGGCGTG
 341  K  A  A  L  G  A  A  A  D  T  I  S  Q  G  T  Q  V  P  G  V
1081 GGCGTTCCGGGCGTTGGTAAGCTTGCGGCCGCACTCGAGCACCACCACCACCACCACTGA
 361  G  V  P  G  V  G  K  L  A  A  A  L  E  H  H  H  H  H  H  *
```

FIGURE 1

(A)


kDa
M
0h
1h
2h
3h
0h*
1h*
2h*
3h*
50
40
30
25
20
15
10

(B)

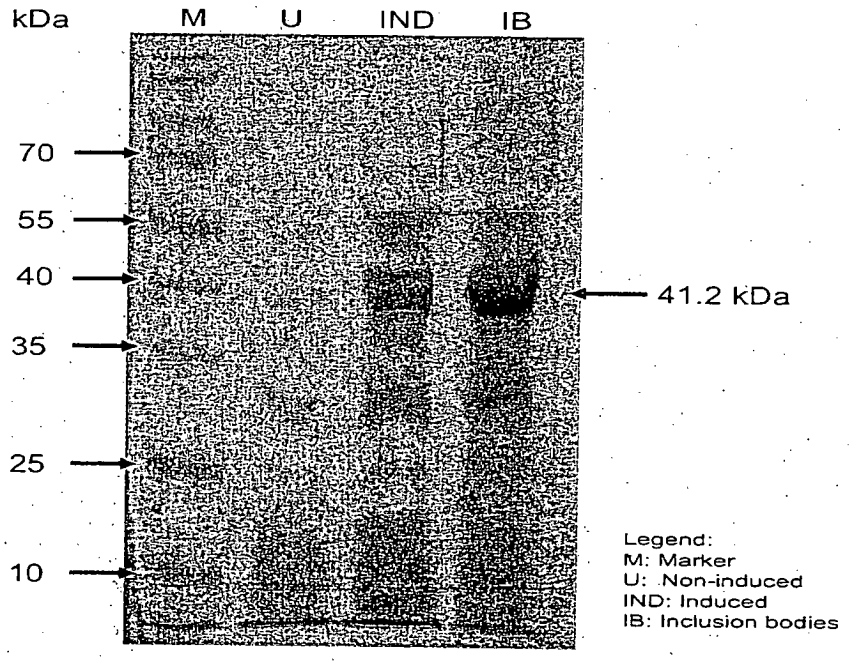

kDa
M
U
IND
IB
70
55
40
35
25
10
41.2 kDa
Legend:
M: Marker
U: Non-induced
IND: Induced
IB: Inclusion bodies

FIGURE 2

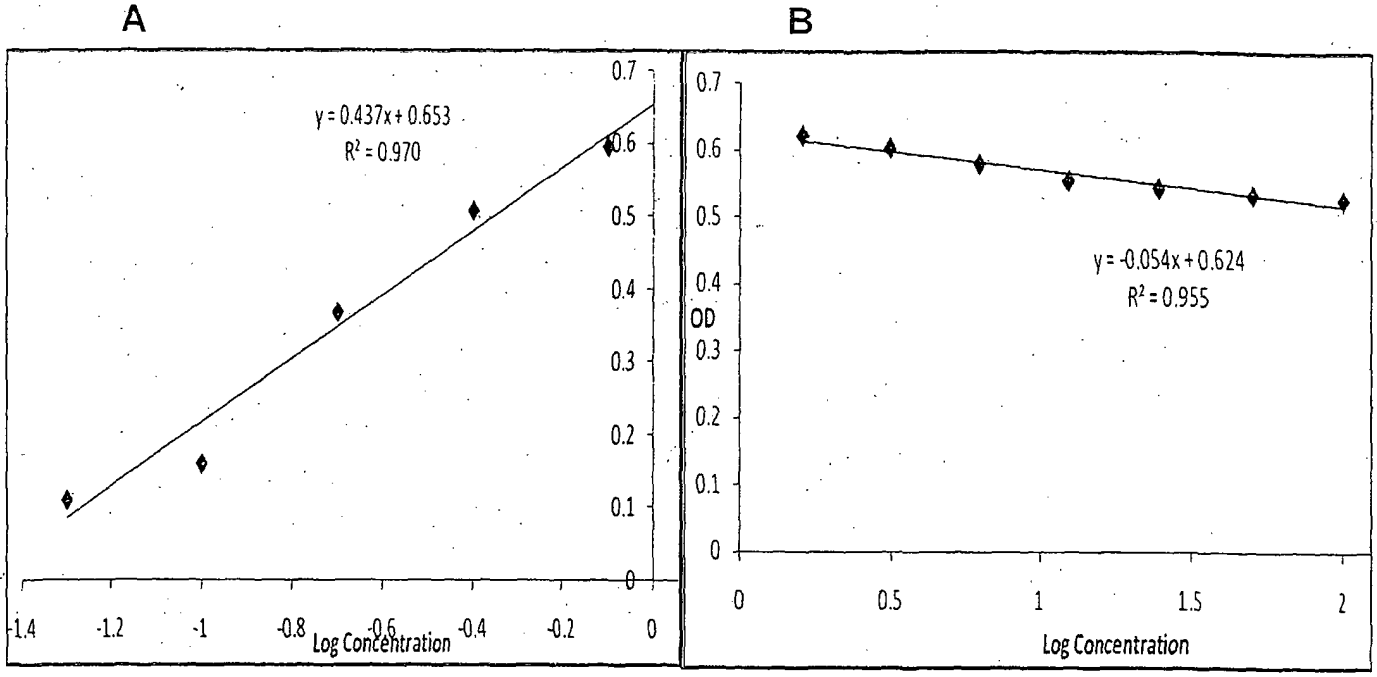


FIGURE 3

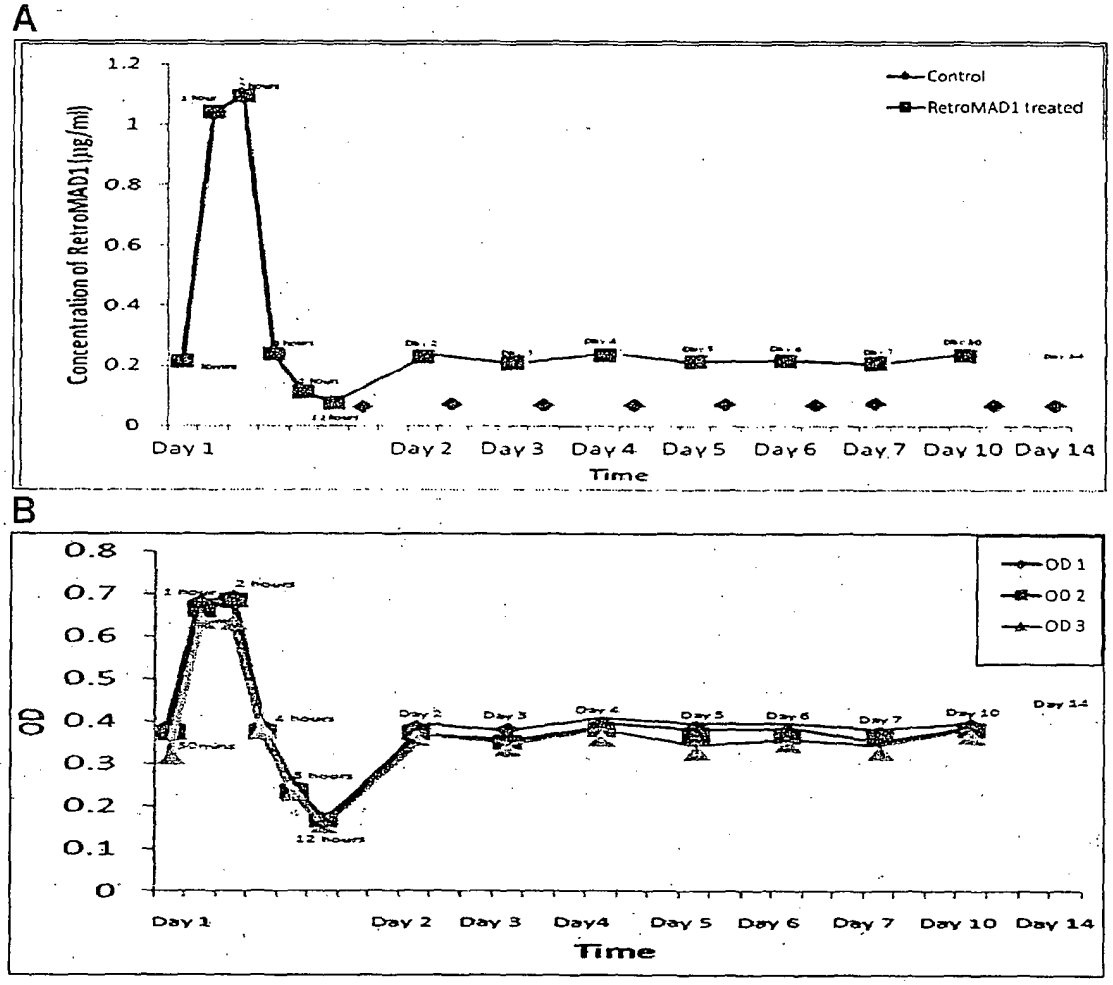
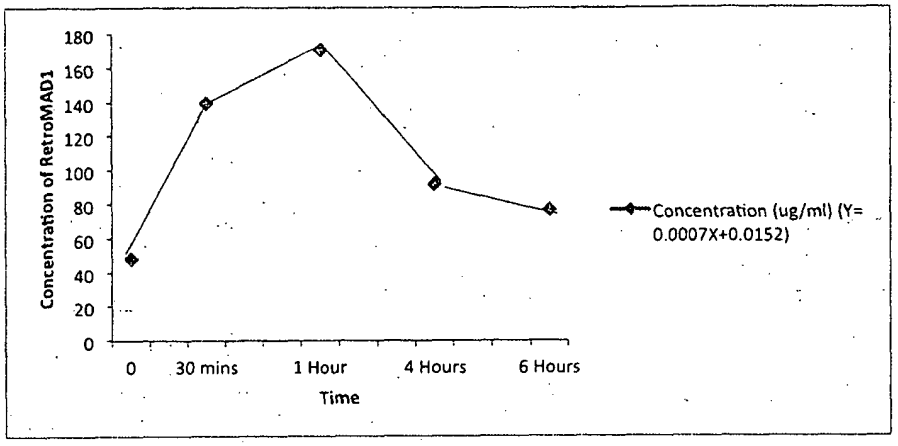
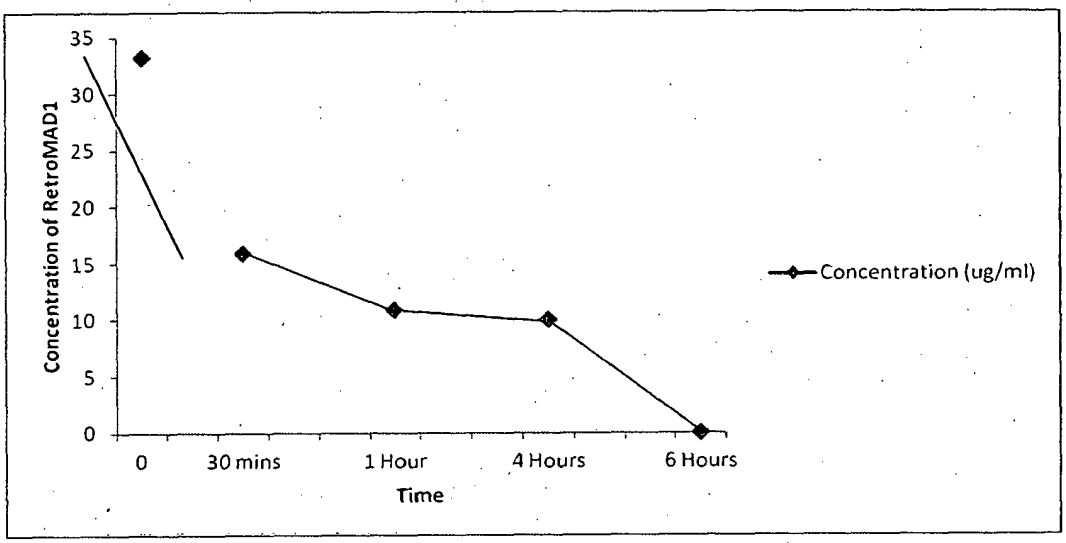


FIGURE 4


Concentration of RetroMAD1
180
160
140
120
100
80
60
40
20
0
0
30 mins
1 Hour
4 Hours
6 Hours
Time
Concentration (ug/ml) (Y= 0.0007X+0.0152)

B


Concentration of RetroMAD1
35
30
25
20
15
10
5
0
0
30 mins
1 Hour
4 Hours
6 Hours
Time
Concentration (ug/ml)

C

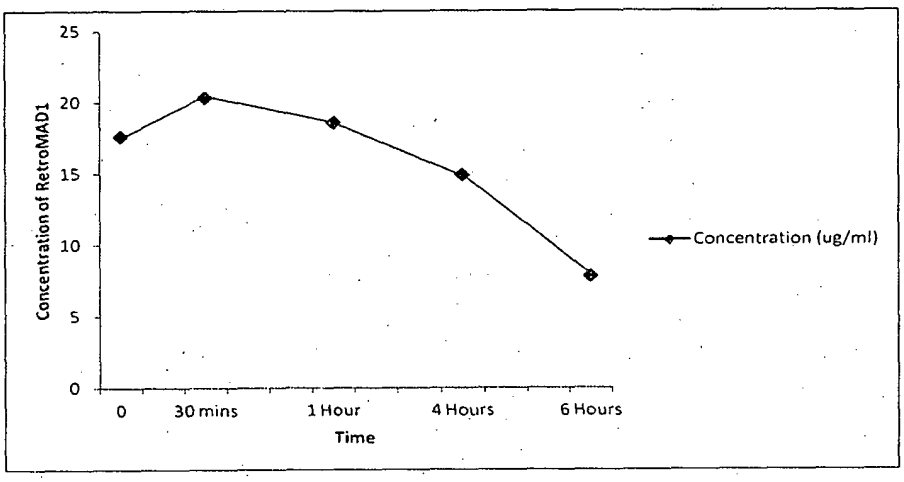

Concentration of RetroMAD1
25
20
15
10
5
0
0
30 mins
1 Hour
4 Hours
6 Hours
Time
Concentration (ug/ml)

FIGURE 5

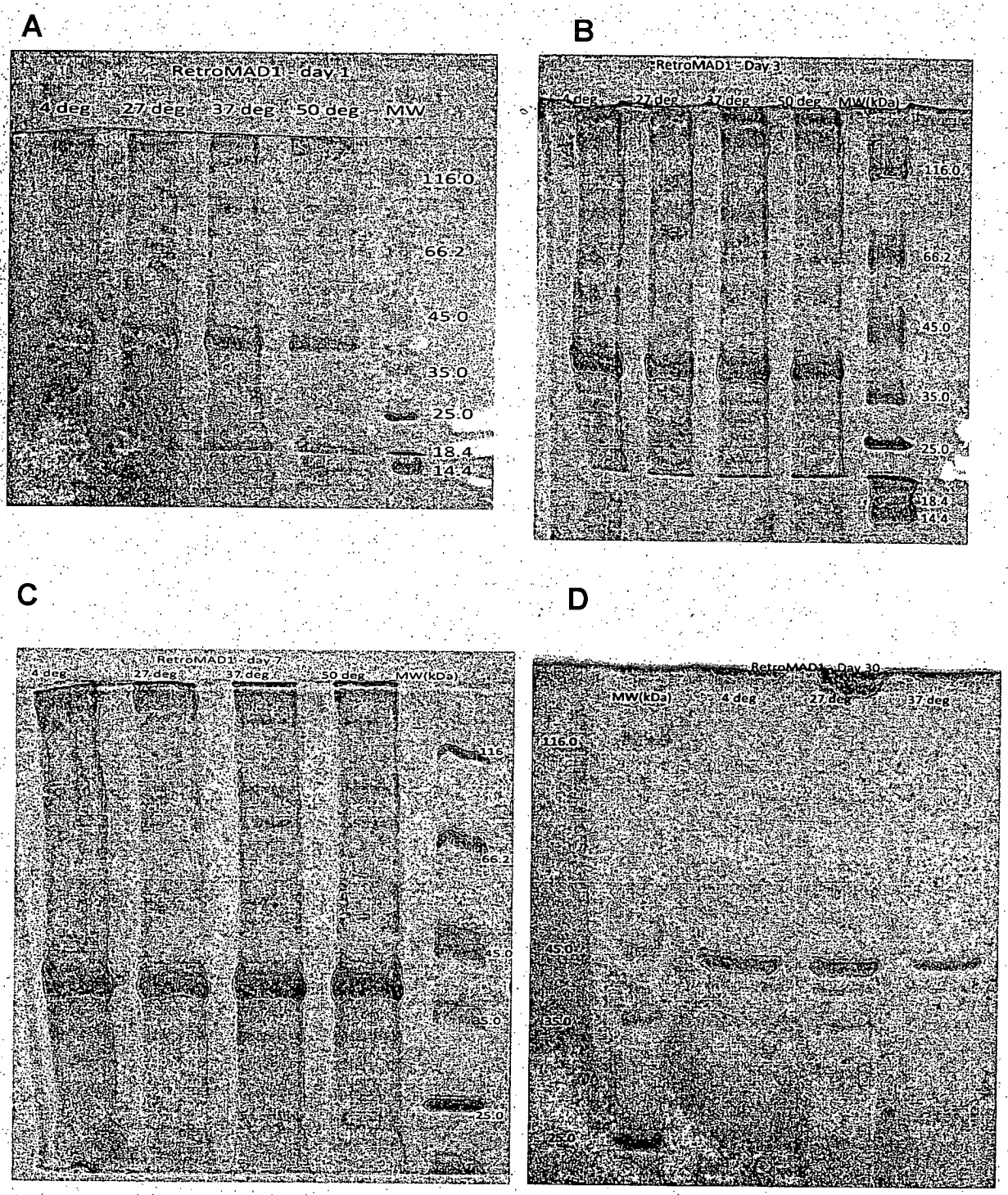
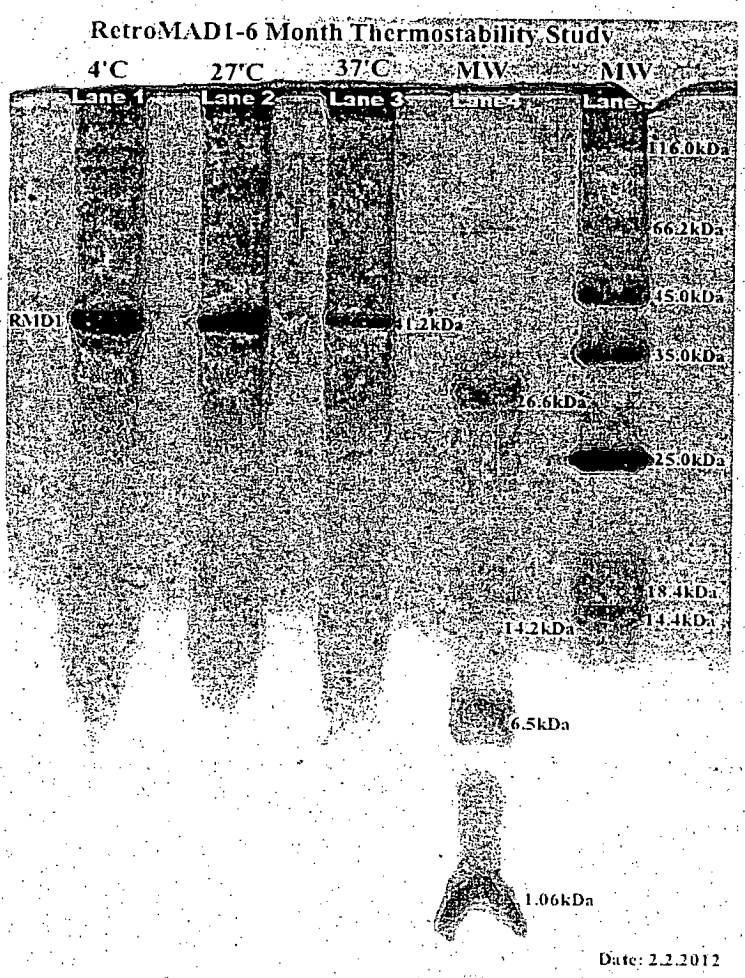

A
RetroMAD1 - day 1
4 deg
27 deg
37 deg
50 deg
MW
116.0
66.2
45.0
35.0
25.0
18.4
14.4
B
RetroMAD1 - Day 3
4 deg
27 deg
37 deg
50 deg
MW(kDa)
116.0
66.2
45.0
35.0
25.0
18.4
14.4
C
RetroMAD1 - day 7
4 deg
27 deg
37 deg
50 deg
MW(kDa)
116
66.2
45.0
35.0
25.0
D
RetroMAD1 - Day 30
MW(kDa)
4 deg
27 deg
37 deg
116.0
45.0
35.0
25.0

FIGURE 6

A


RetroMAD1-6 Month Thermostability Study
4'C
27'C
37'C
MW
MW
Lane 1
Lane 2
Lane 3
Lane 4
116.0kDa
66.2kDa
45.0kDa
RMD1
42kDa
35.0kDa
26.6kDa
25.0kDa
18.4kDa
14.4kDa
14.2kDa
6.5kDa
1.06kDa
Date: 2.2.2012

B

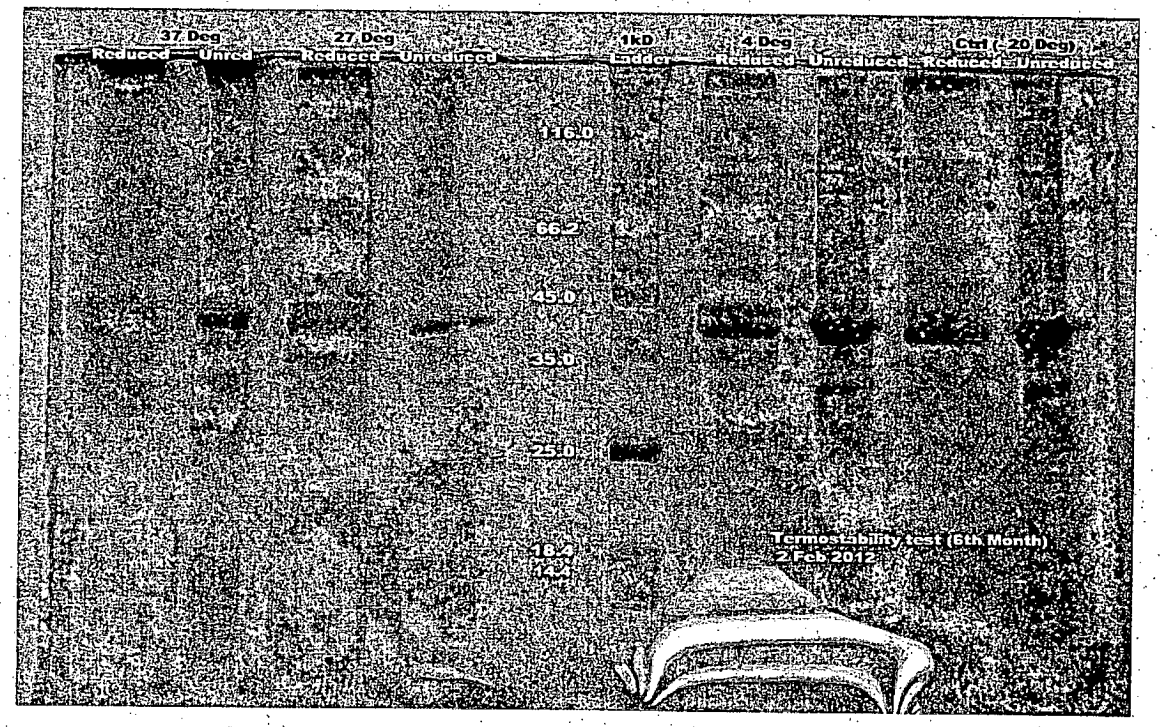

37 Deg
27 Deg
1kD
4 Deg
Ctrl (-20 Deg)
Reduced
Unred
Ladder
Unreduced
116.0
66.2
45.0
35.0
25.0
18.4
14.4
Temostability test (6th Month)
2 Feb 2012

FIGURE 7

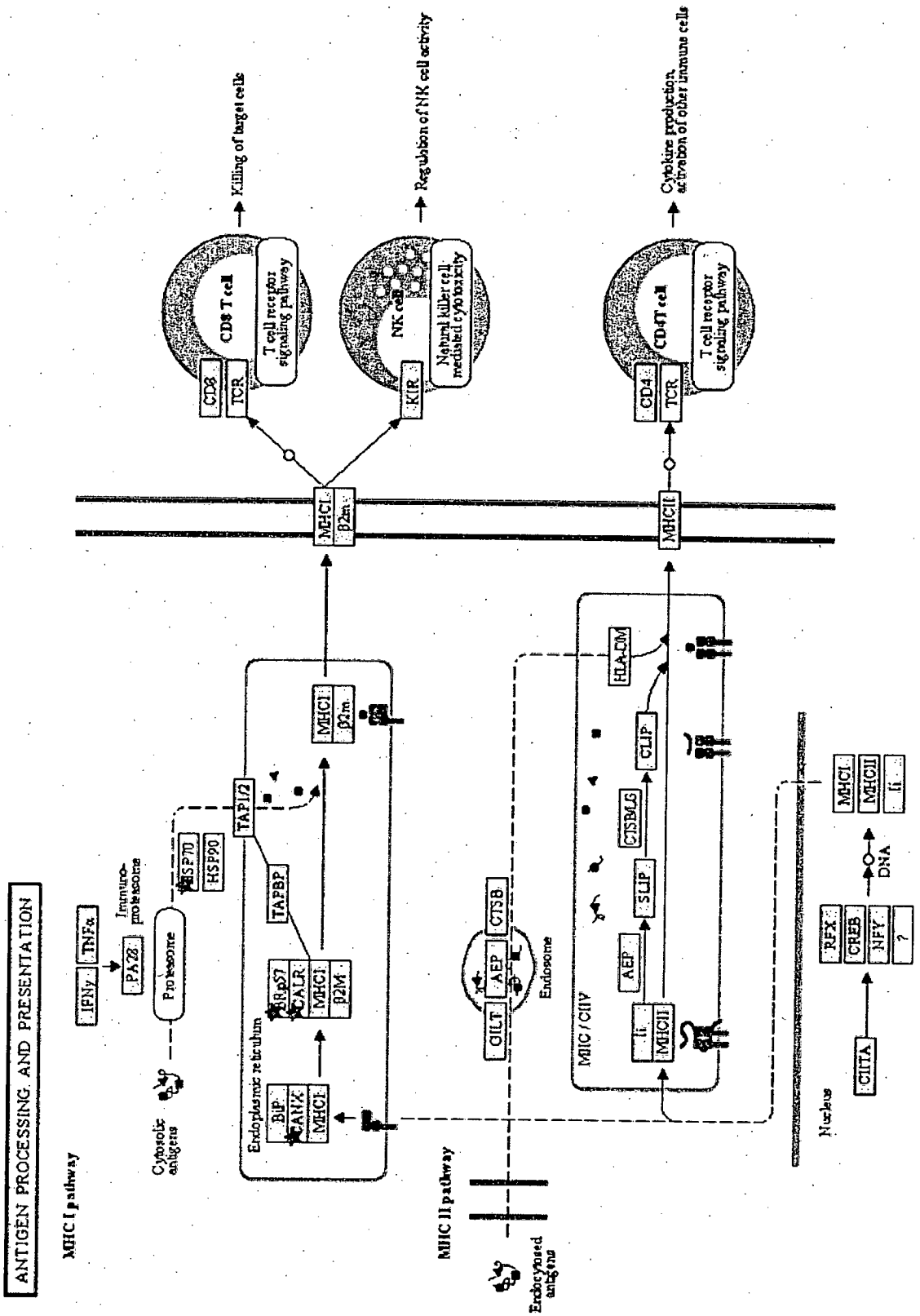
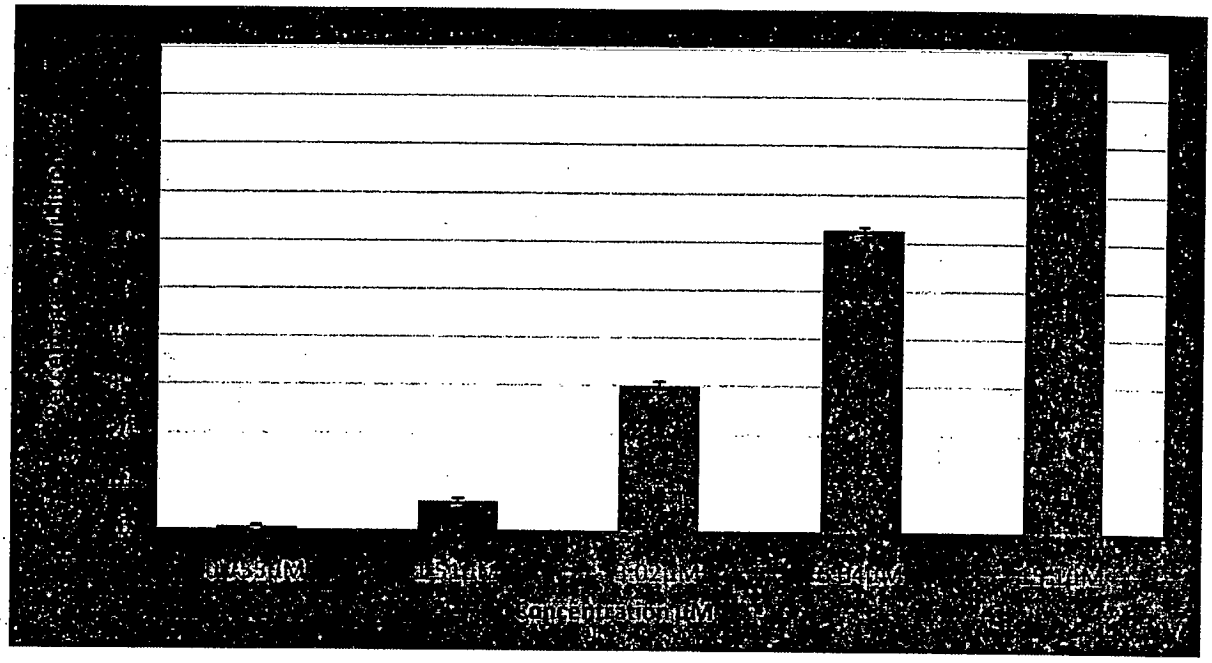

ANTIGEN PROCESSING AND PRESENTATION
MHC I pathway
IFNγ
TNFα
PA28
Immuno-proteasome
Proteasome
HSP70
HSP90
Cytosolic antigens
TAP1/2
TAPBP
ERp57
CALR
MHCI
B2M
BiP
ERAAP
MHCI
Endoplasmic reticulum
MHCI
β2m
MHC II pathway
Endocytosed antigens
CTSB
GILT
AEP
Endosome
HLA-DM
CLIP
CTSB/L/S
SLIP
AEP
Ii
MHCII
MHC / CIIV
MHCI
MHCII
Ii
RFX
CREB
NFY
?
DNA
CIITA
Nucleus
MHCI
β2m
MHCII
CD8 T cell
CD8
TCR
T cell receptor signaling pathway
Killing of target cells
NK cell
KIR
Natural killer cell mediated cytotoxicity
Regulation of NK cell activity
CD4 T cell
CD4
TCR
T cell receptor signaling pathway
Cytokine production, activation of other immune cells

FIGURE 8

A



B

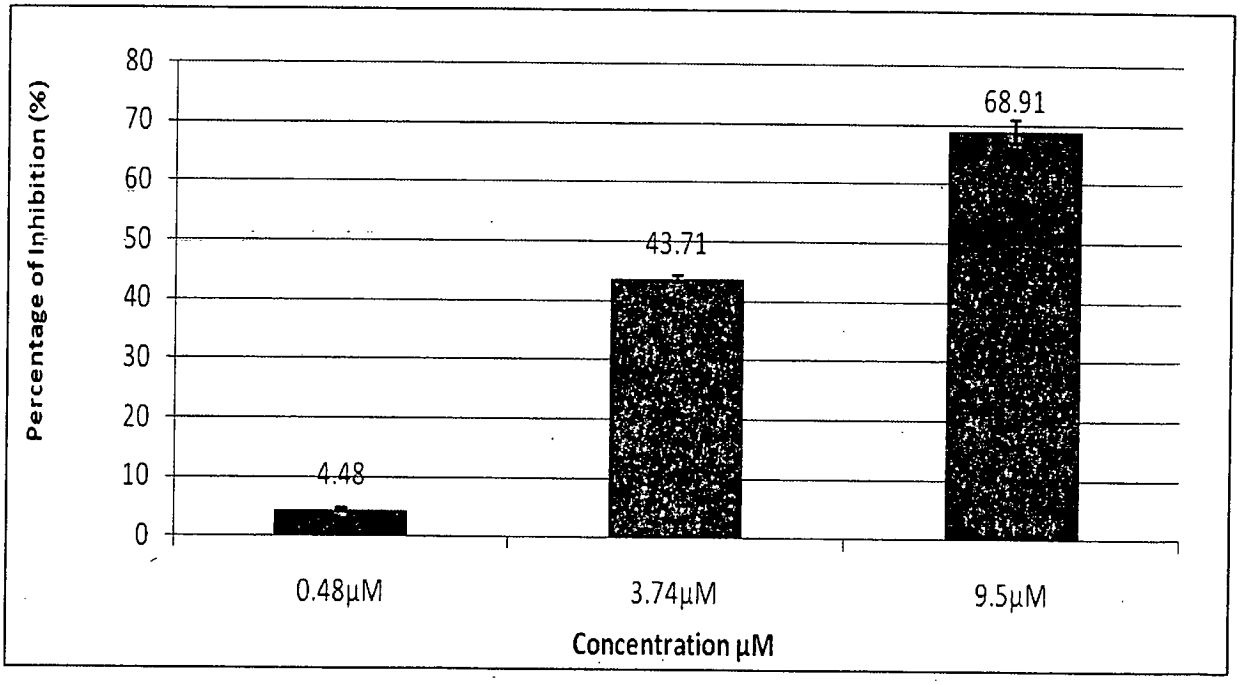

Percentage of Inhibition (%)
80
70
60
50
40
30
20
10
0
4.48
43.71
68.91
0.48µM
3.74µM
9.5µM
Concentration µM

FIGURE 9

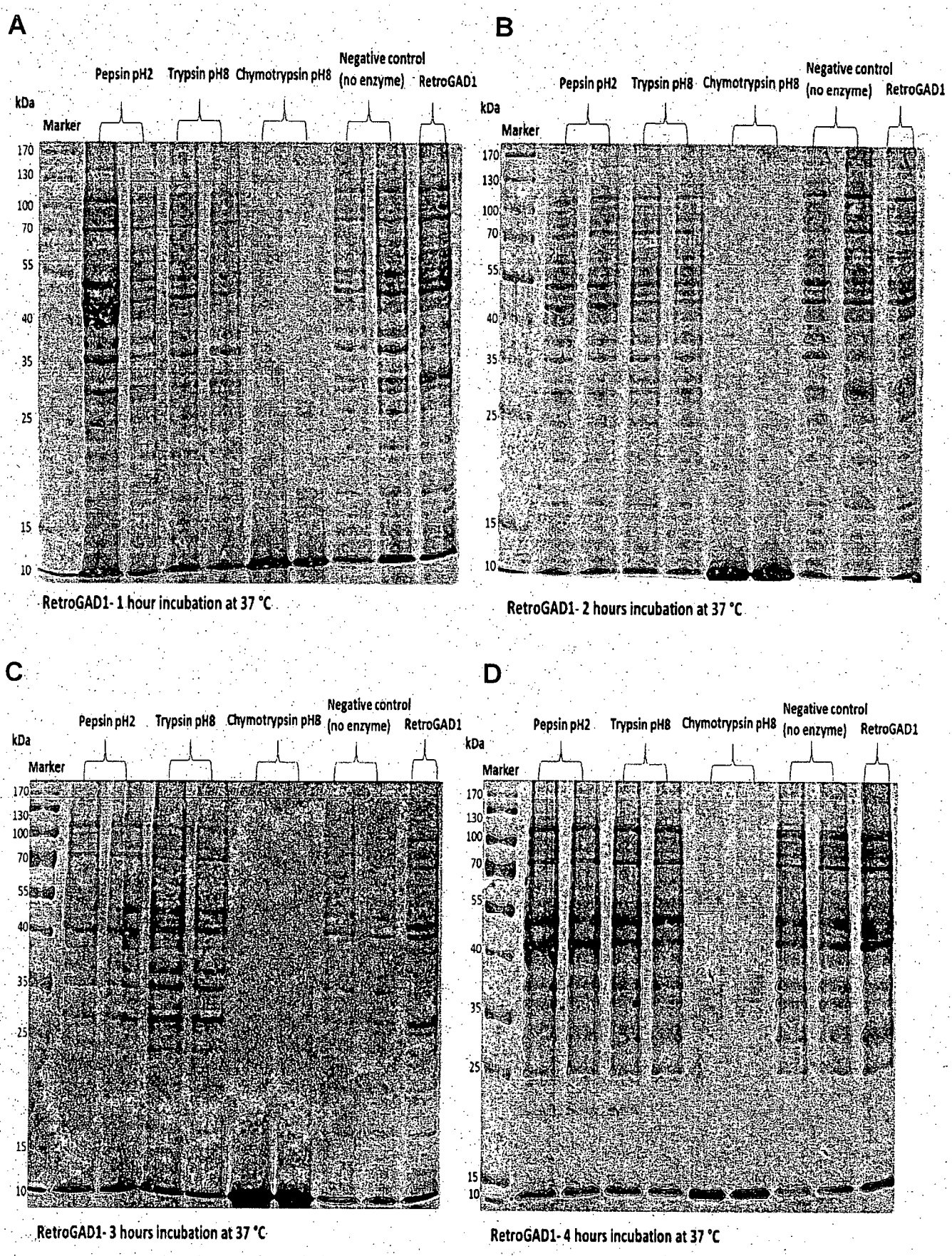

A
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
RetroGAD1
kDa
Marker
170
130
100
70
55
40
35
25
15
10
RetroGAD1- 1 hour incubation at 37 °C
B
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
RetroGAD1
kDa
Marker
170
130
100
70
55
40
35
25
15
10
RetroGAD1- 2 hours incubation at 37 °C
C
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
RetroGAD1
kDa
Marker
170
130
100
70
55
40
35
25
15
10
RetroGAD1- 3 hours incubation at 37 °C
D
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
RetroGAD1
kDa
Marker
170
130
100
70
55
40
35
25
15
10
RetroGAD1- 4 hours incubation at 37 °C

FIGURE 10

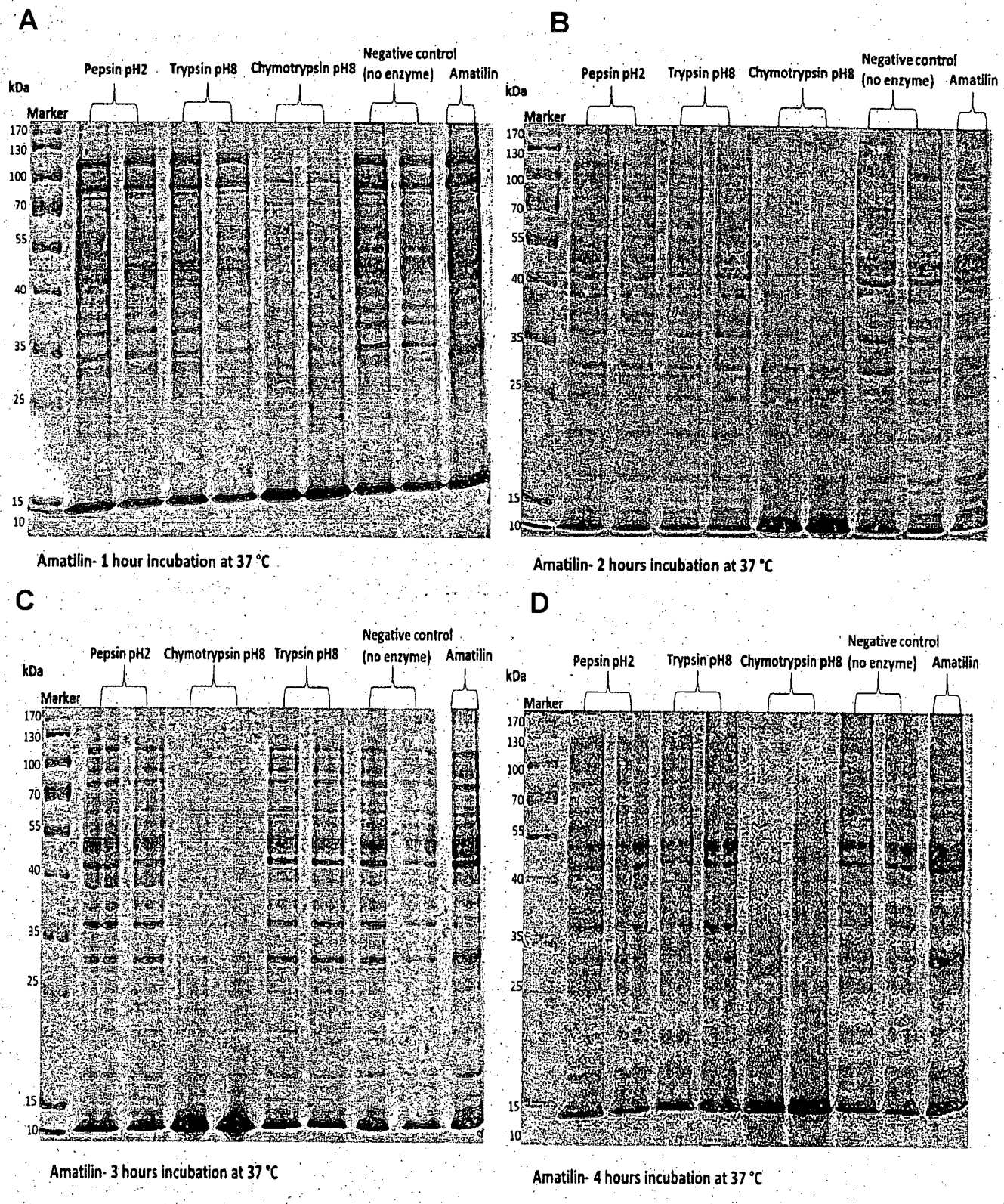

A
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
Amatilin
kDa
Marker
170
130
100
70
55
40
35
25
15
10
Amatilin- 1 hour incubation at 37 °C
B
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
Amatilin
kDa
Marker
Amatilin- 2 hours incubation at 37 °C
C
Pepsin pH2
Chymotrypsin pH8
Trypsin pH8
Negative control (no enzyme)
Amatilin
kDa
Marker
Amatilin- 3 hours incubation at 37 °C
D
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
Amatilin
kDa
Marker
Amatilin- 4 hours incubation at 37 °C

FIGURE 11

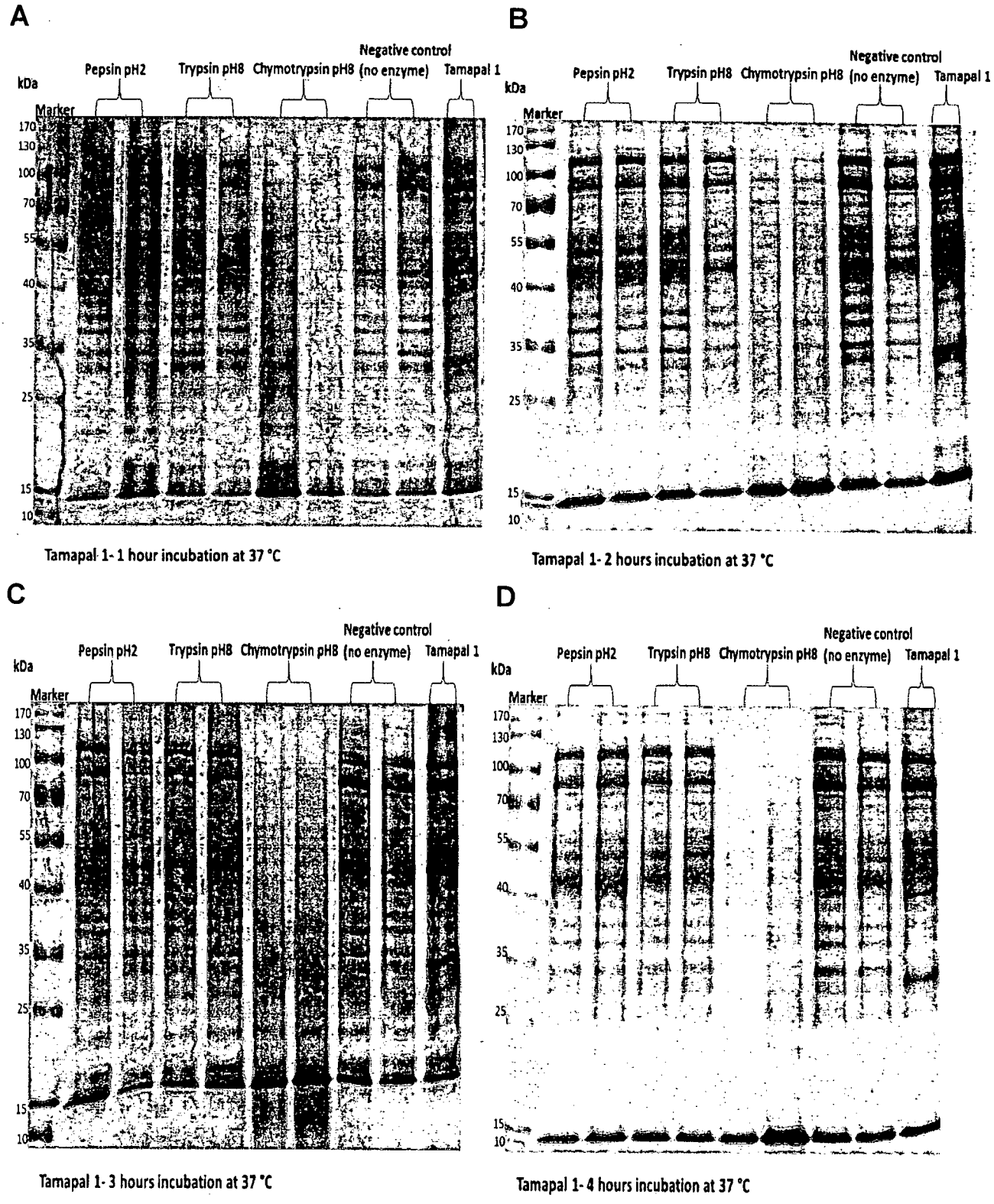

A
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
Tamapal 1
kDa
Marker
170
130
100
70
55
40
35
25
15
10
Tamapal 1- 1 hour incubation at 37 °C
B
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
Tamapal 1
kDa
Marker
Tamapal 1- 2 hours incubation at 37 °C
C
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
Tamapal 1
kDa
Marker
Tamapal 1- 3 hours incubation at 37 °C
D
Pepsin pH2
Trypsin pH8
Chymotrypsin pH8
Negative control (no enzyme)
Tamapal 1
kDa
Marker
Tamapal 1- 4 hours incubation at 37 °C

FIGURE 12

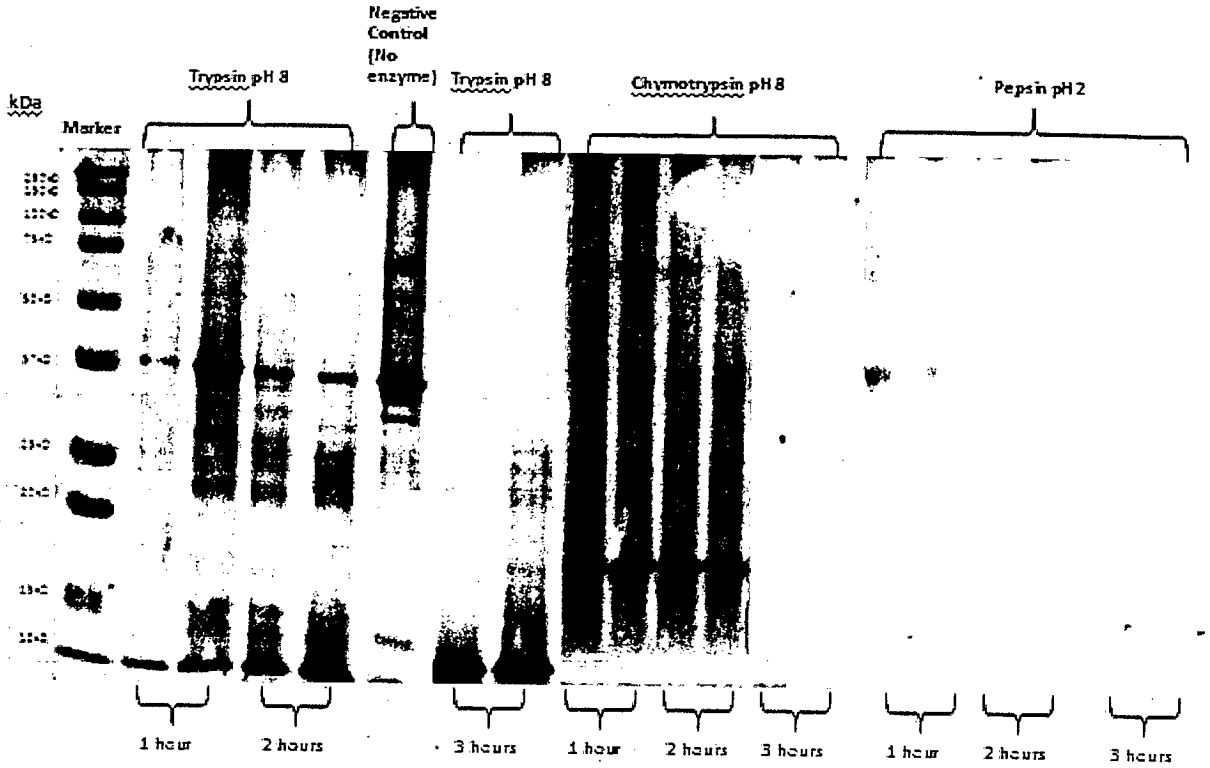


FIGURE 13

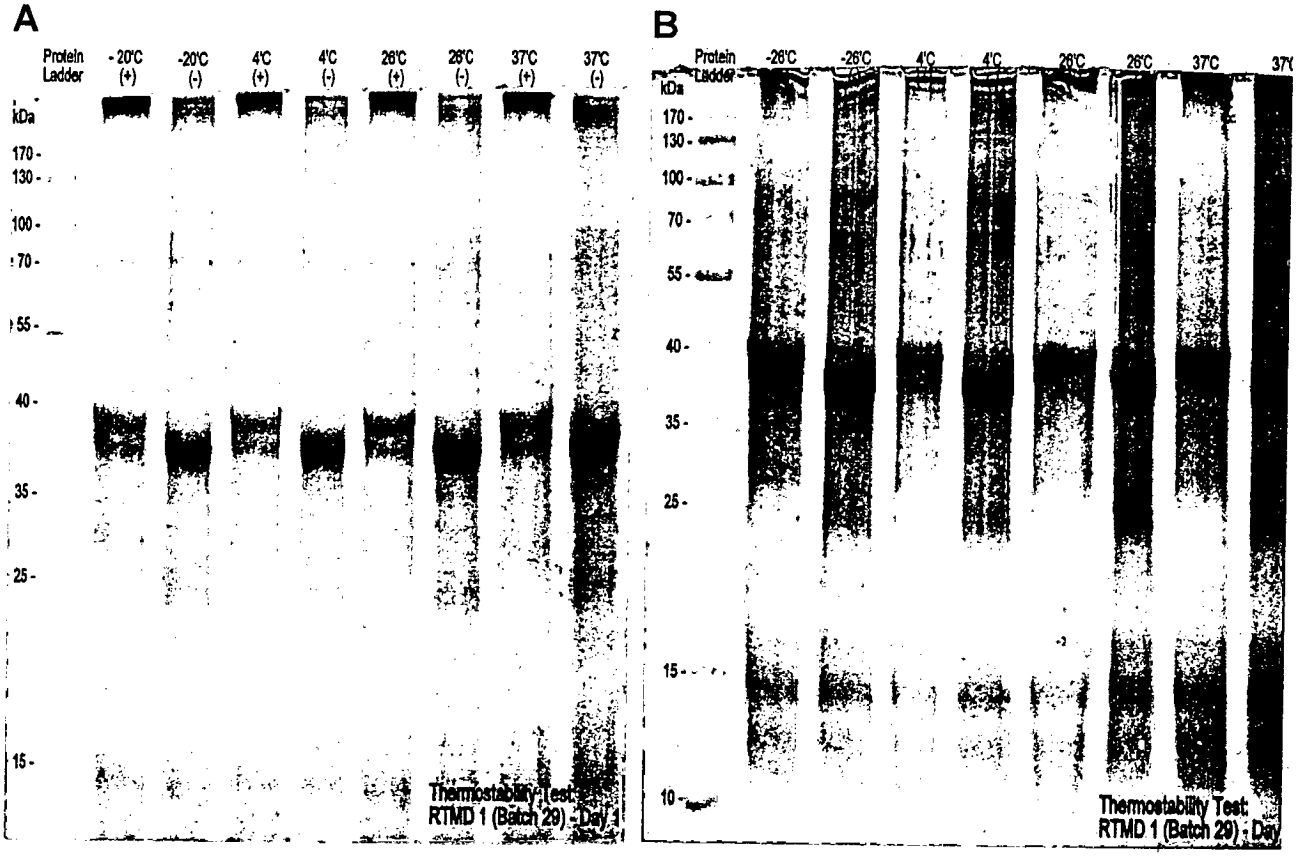


FIGURE 14 (CONTINUED)

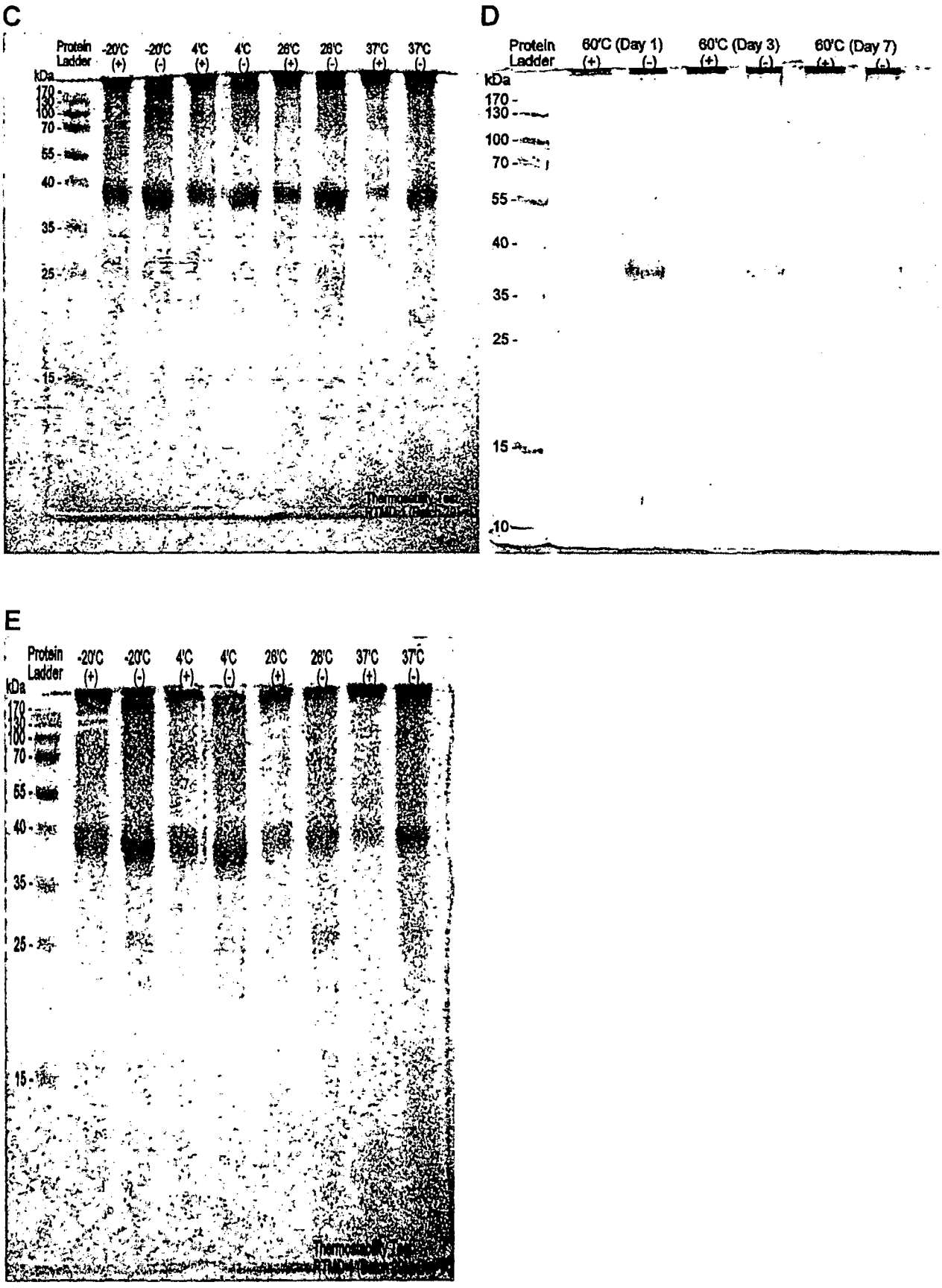


FIGURE 14(CONTINUED)

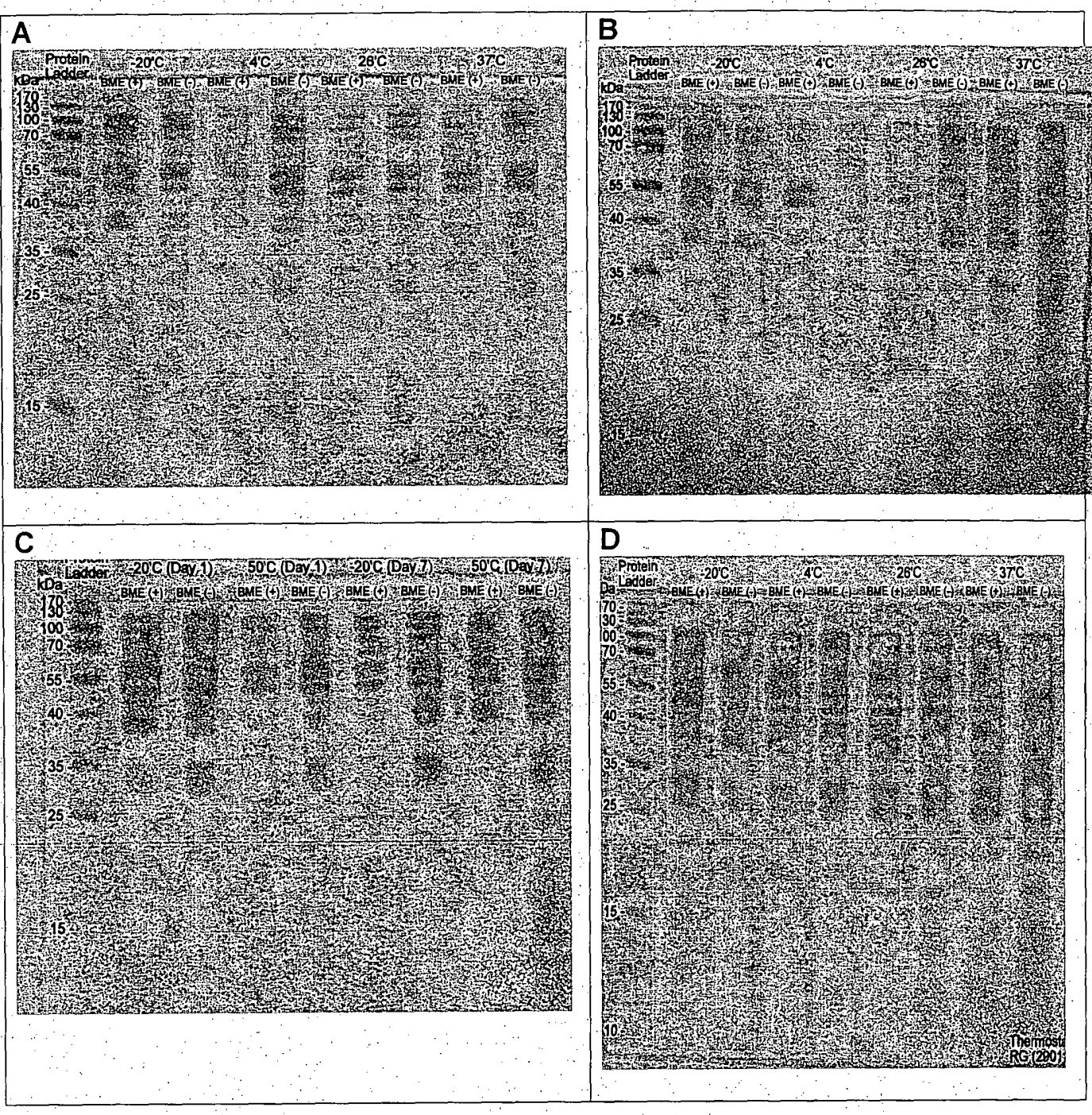

A
Protein Ladder
kDa
-20°C
4°C
26°C
37°C
BME (+)
BME (-)
170
130
100
70
55
40
35
25
15
B
Protein Ladder
kDa
-20°C
4°C
26°C
37°C
BME (+)
BME (-)
170
130
100
70
55
40
35
25
15
C
Ladder
kDa
-20°C (Day 1)
50°C (Day 1)
-20°C (Day 7)
50°C (Day 7)
BME (+)
BME (-)
170
130
100
70
55
40
35
25
15
D
Protein Ladder
kDa
20°C
4°C
26°C
37°C
BME (+)
BME (-)
70
30
00
70
55
40
35
25
15
10
Thermostu
RG (2901

FIGURE 15

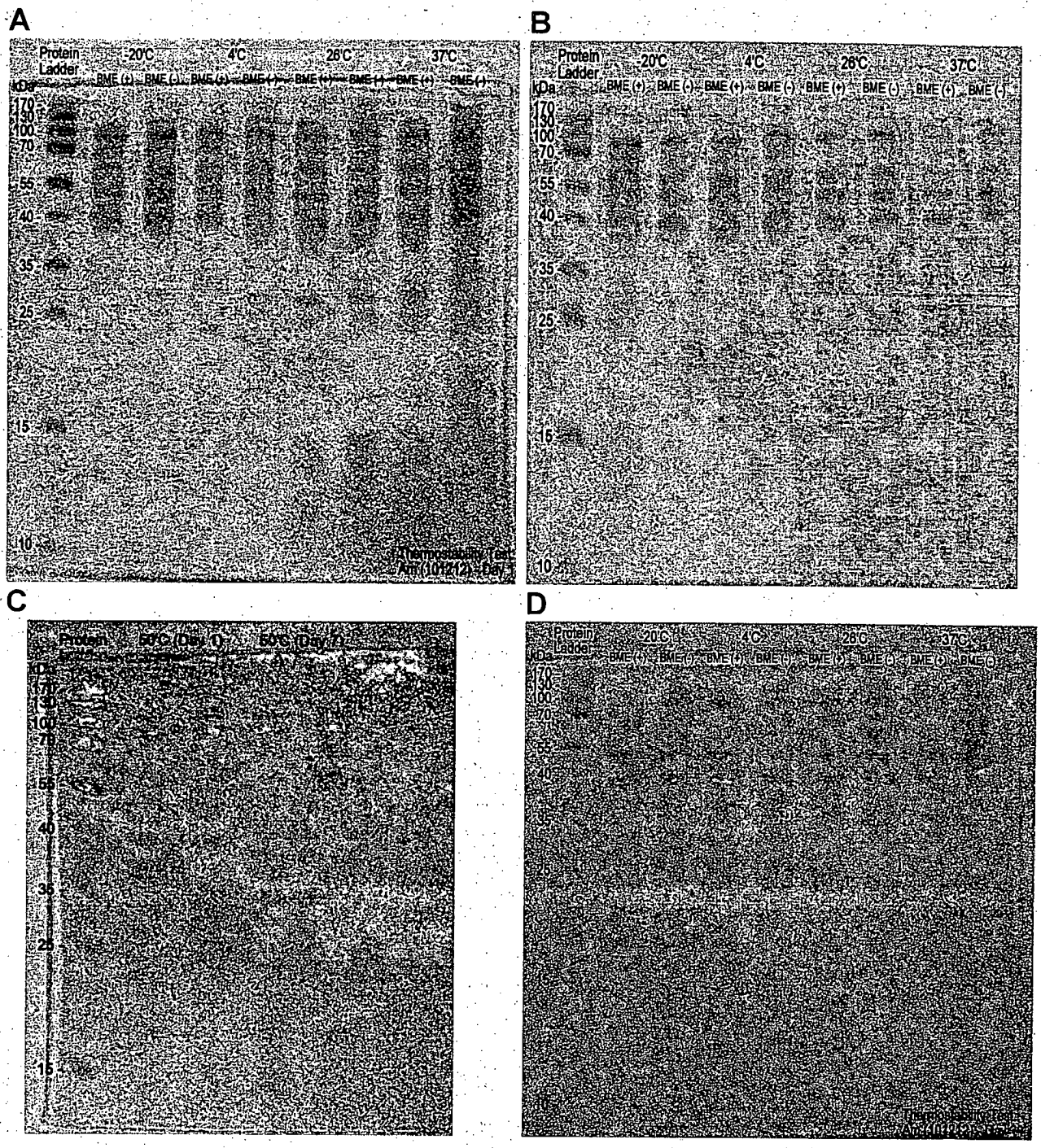
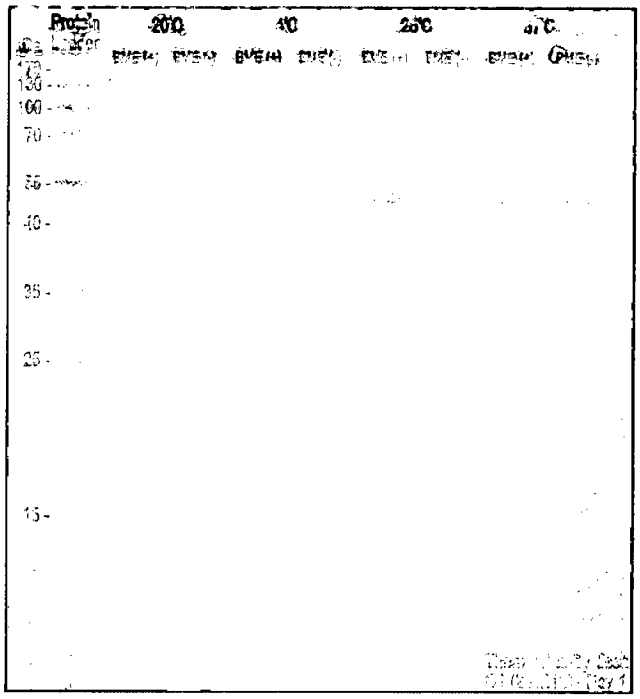
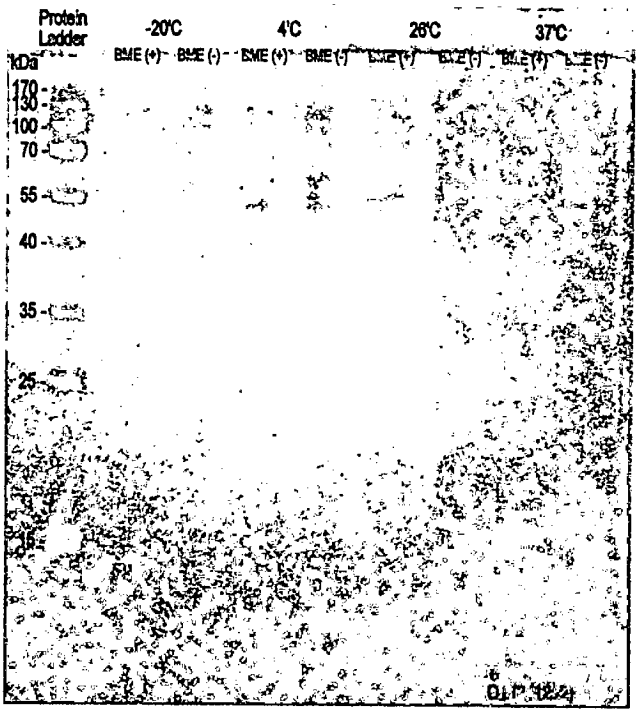

A
Protein Ladder
-20°C
4°C
26°C
37°C
BME (+)
BME (-)
kDa
170
130
100
70
55
40
35
25
15
10
B
C
50°C (Day 1)
50°C (Day 7)
D

FIGURE 16

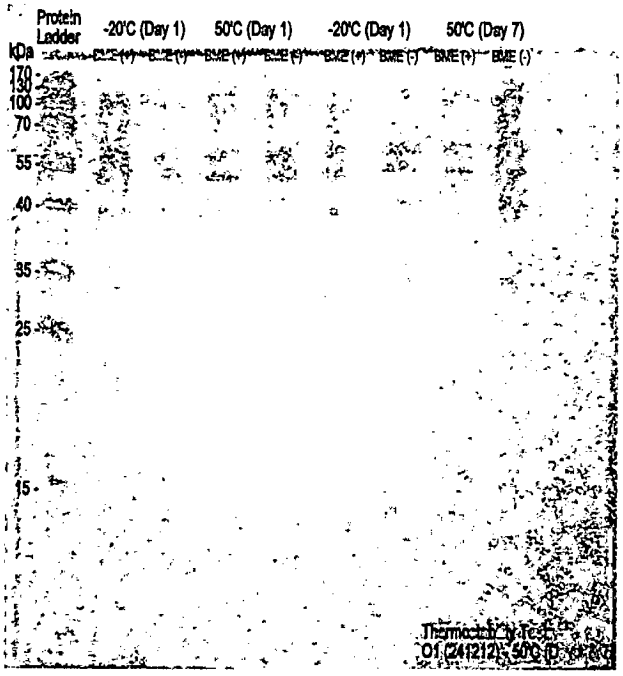
A



B



C



D

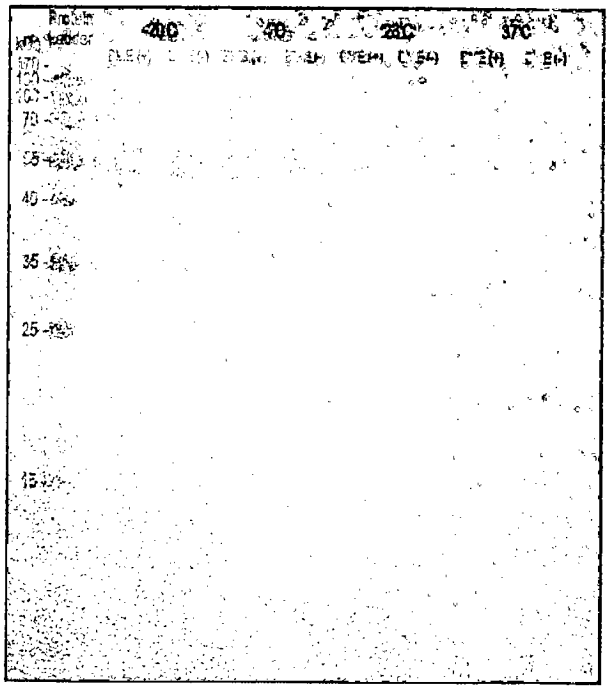
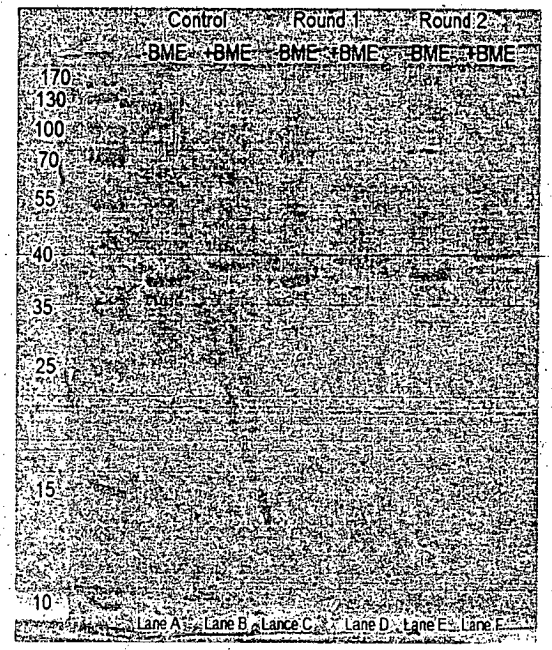
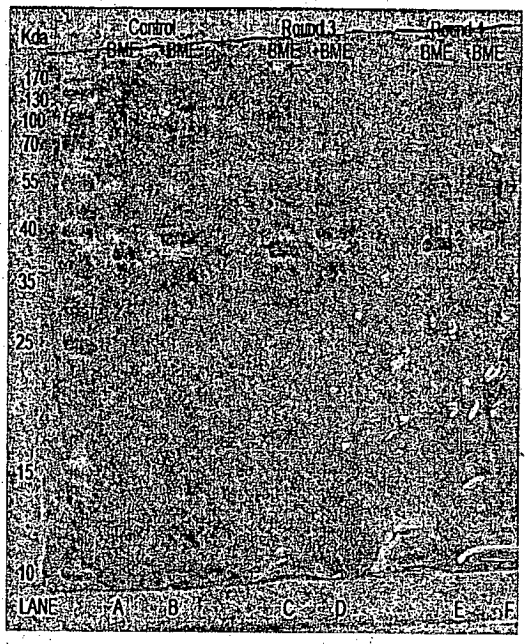


FIGURE 17

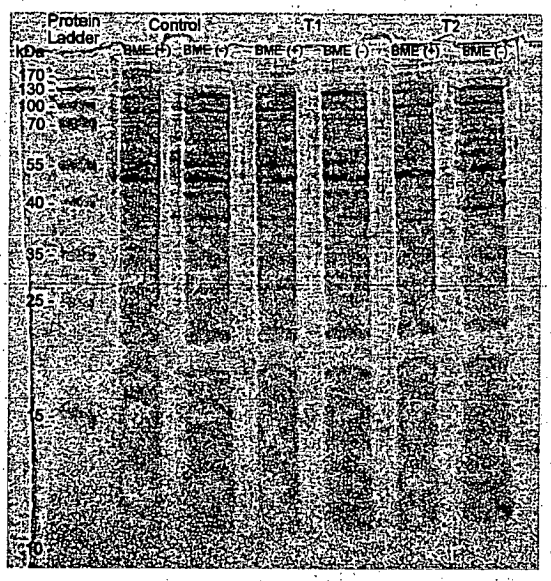
A1



A2



B1



B2

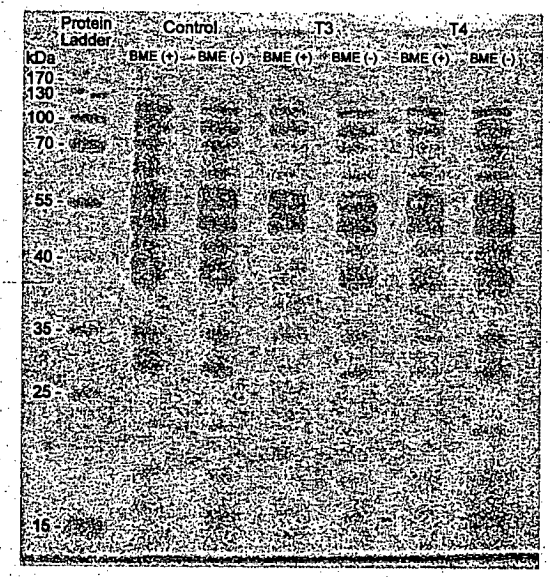
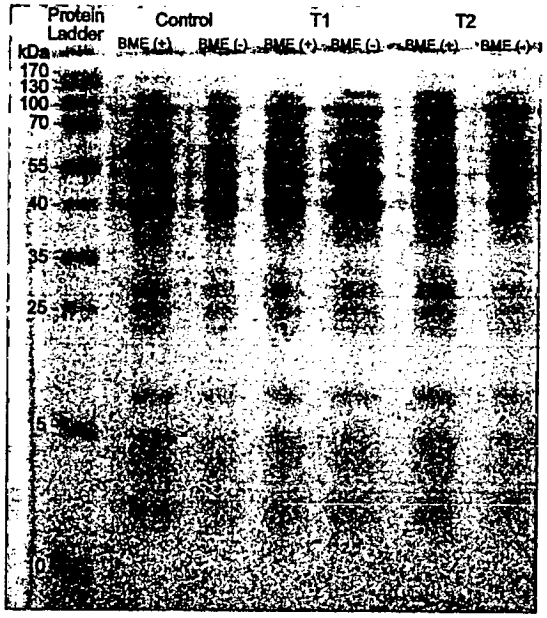
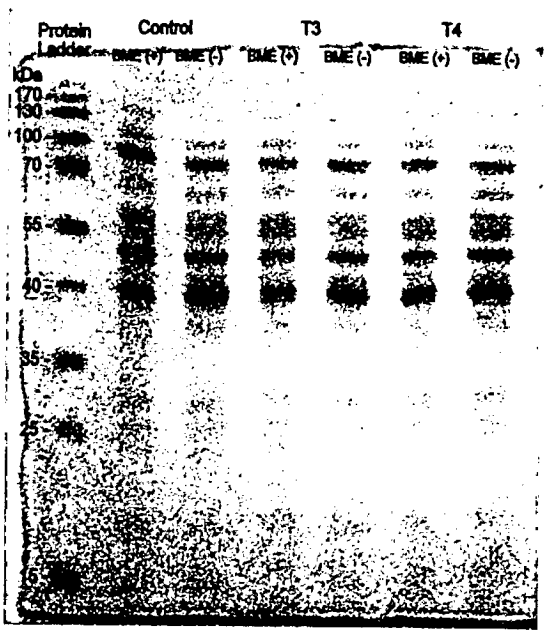


FIGURE 18

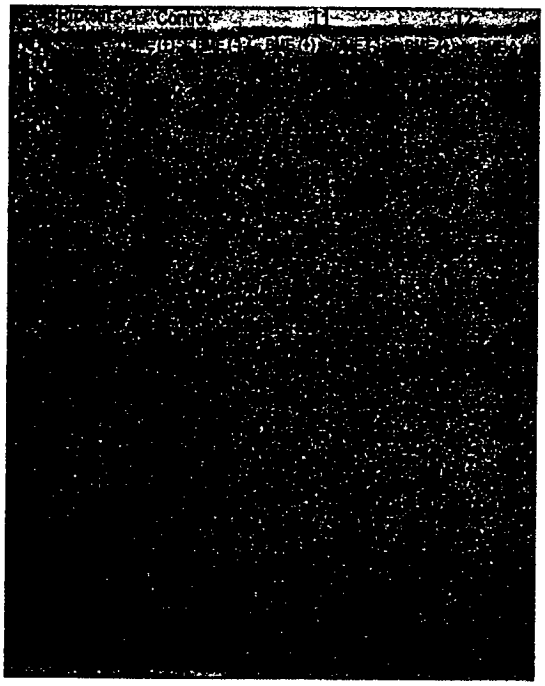
C1



C2



D1



D2

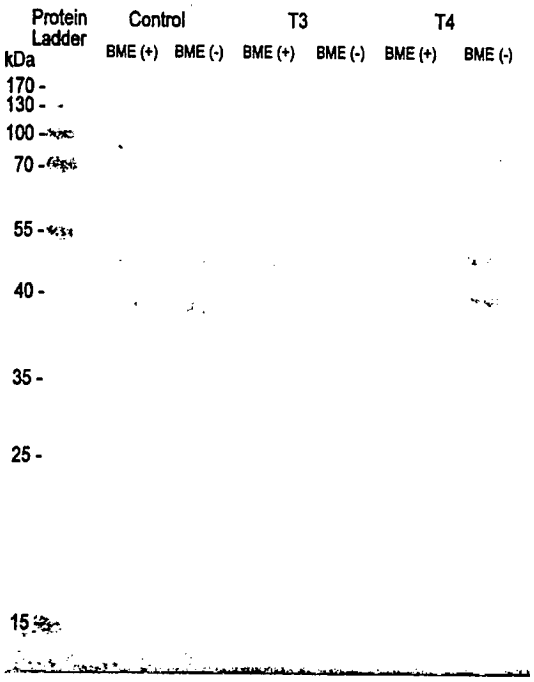
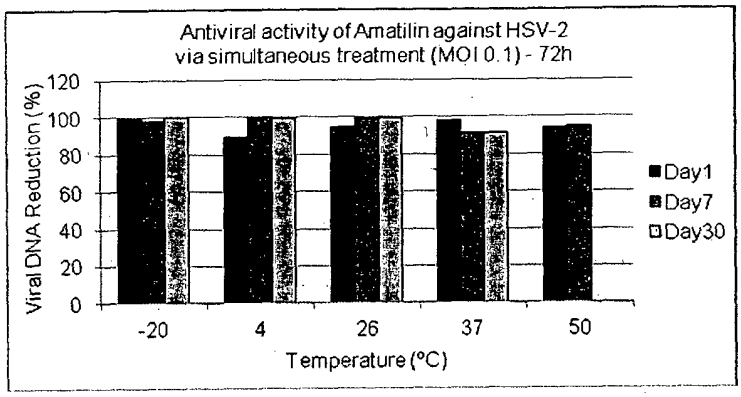
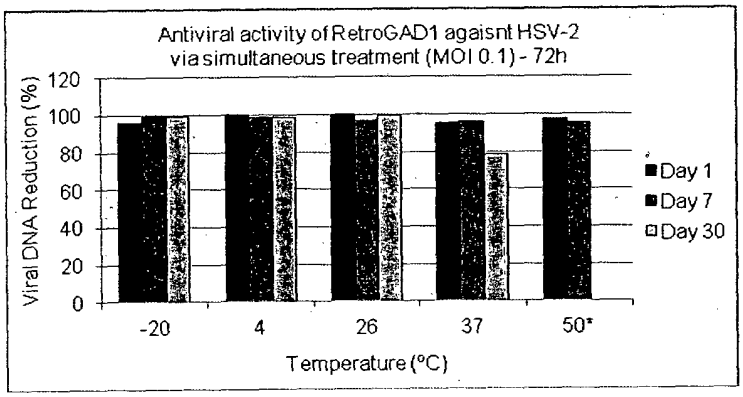


FIGURE 18 (CONTINUED)

A



B



C

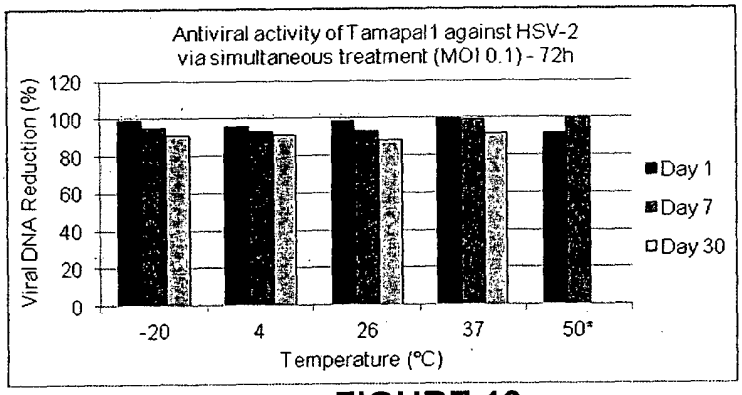


FIGURE 19

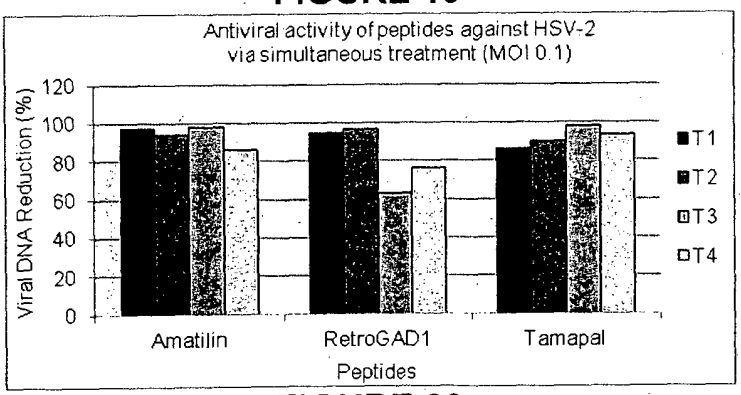
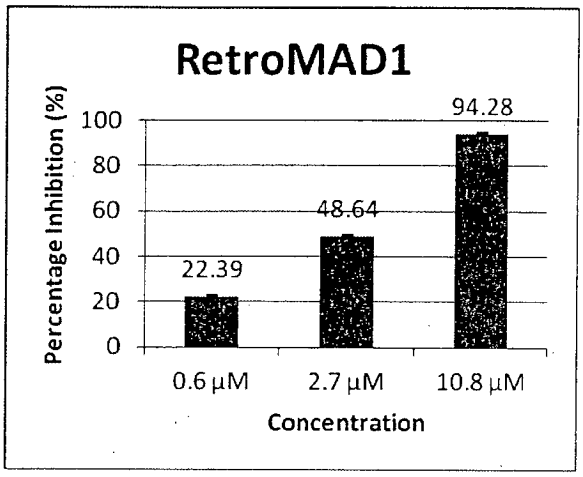
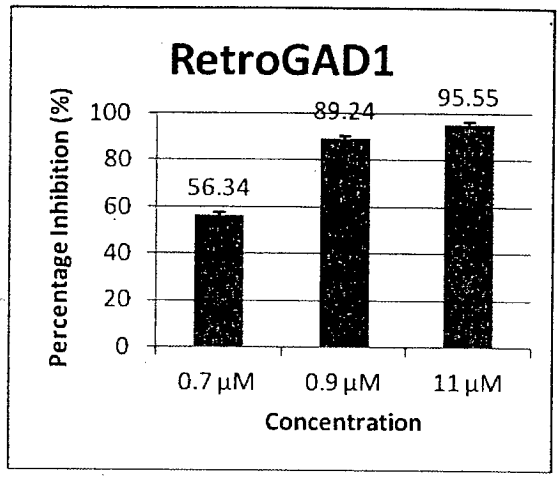


FIGURE 20

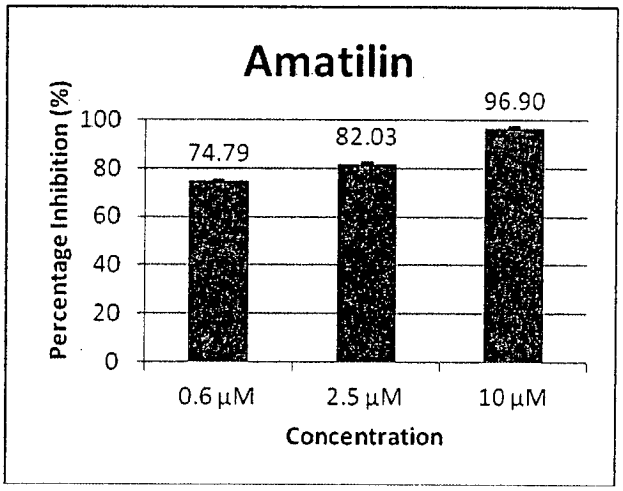
A



B



C



D

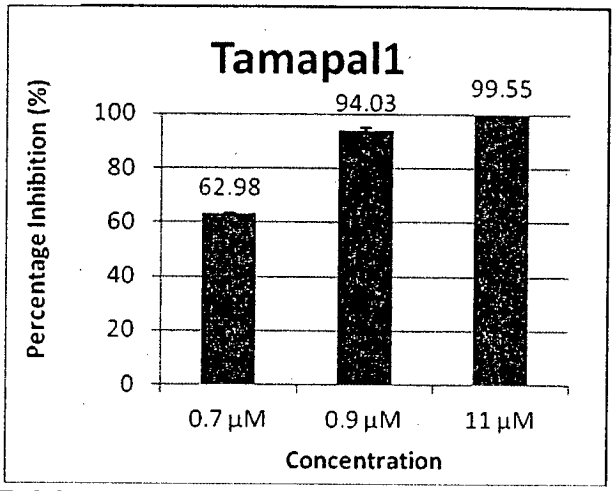


FIGURE 21

A

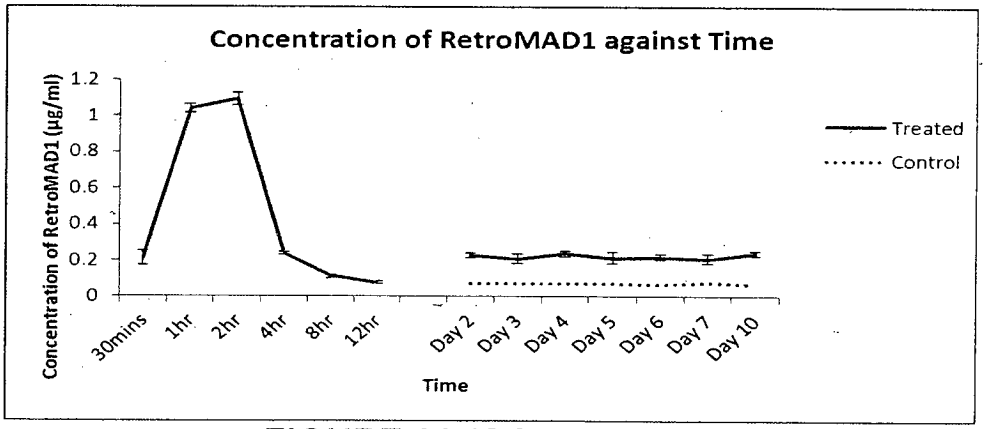
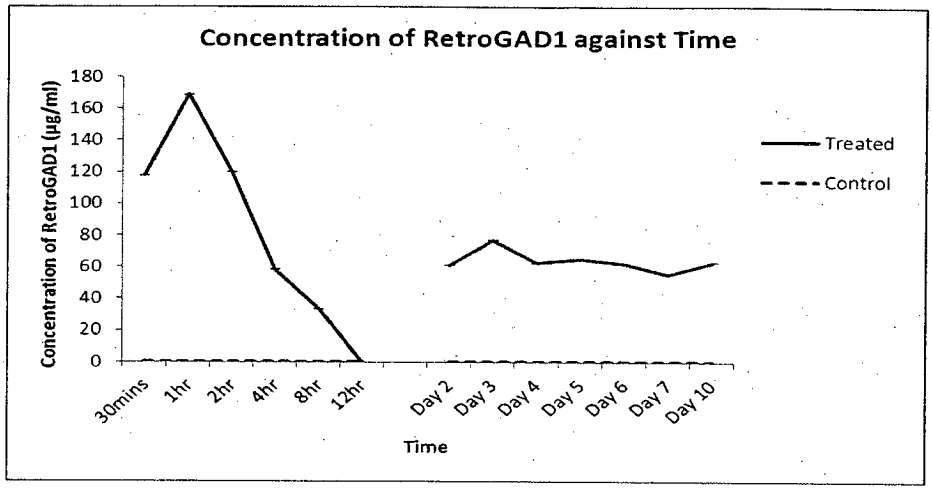
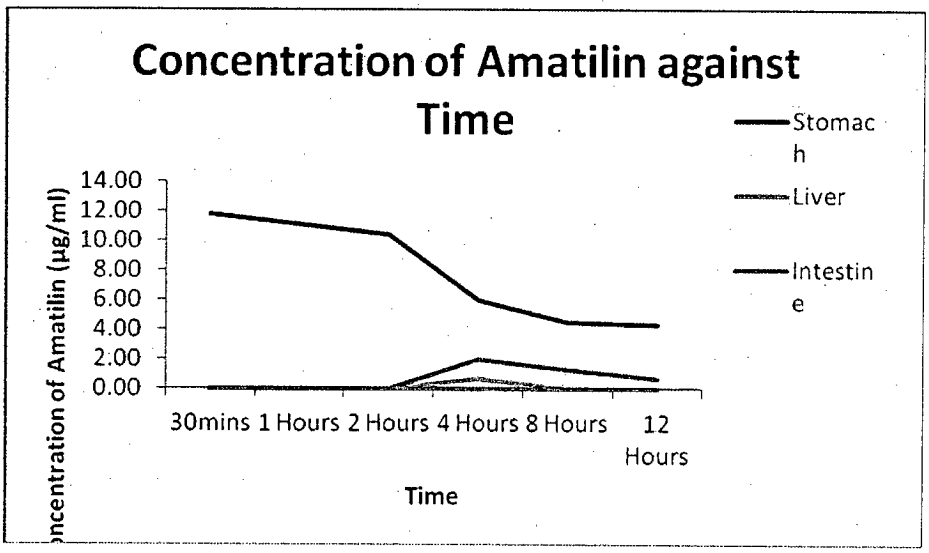


FIGURE 22 (CONTINUED)

B



C



D

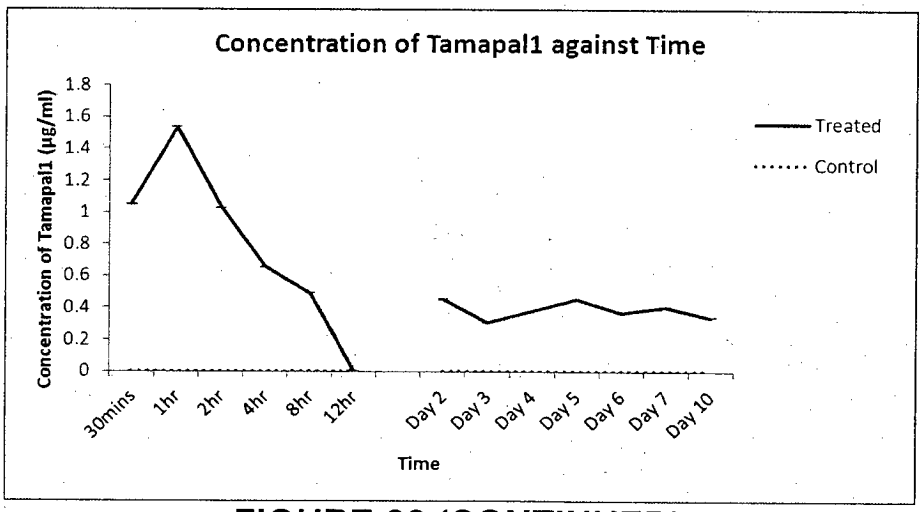
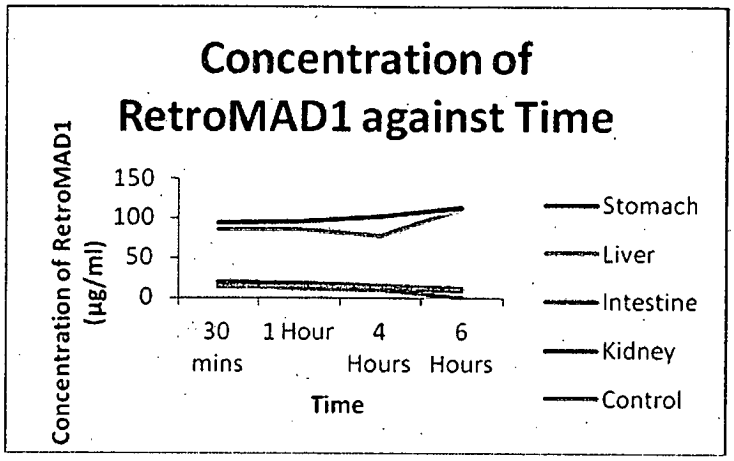
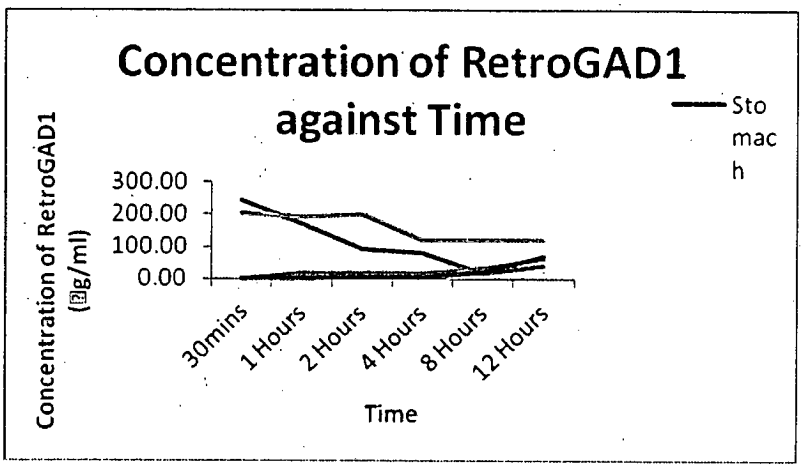


FIGURE 22 (CONTINUED)

A



B



C

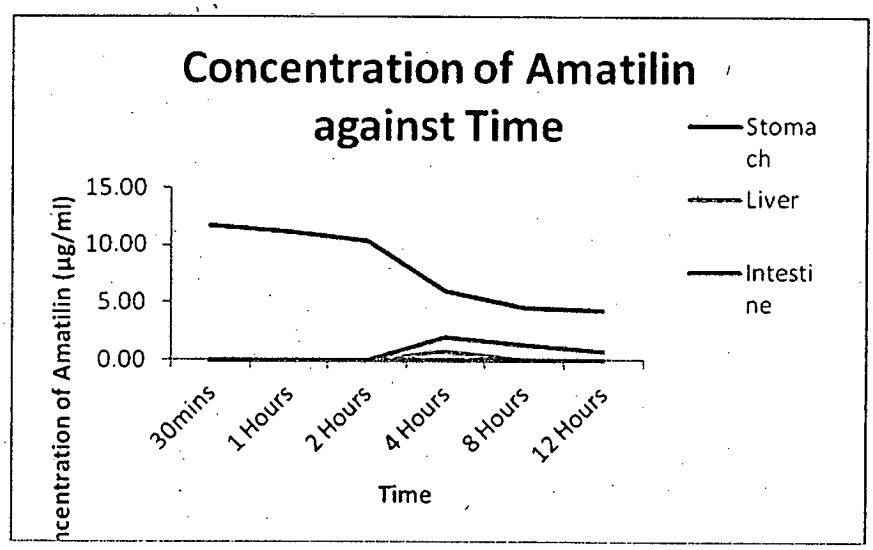


FIGURE 23 (CONTINUED)

D

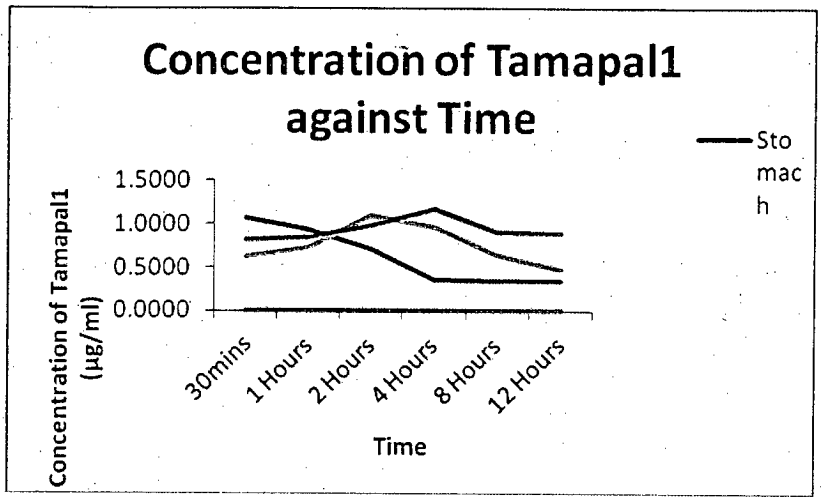
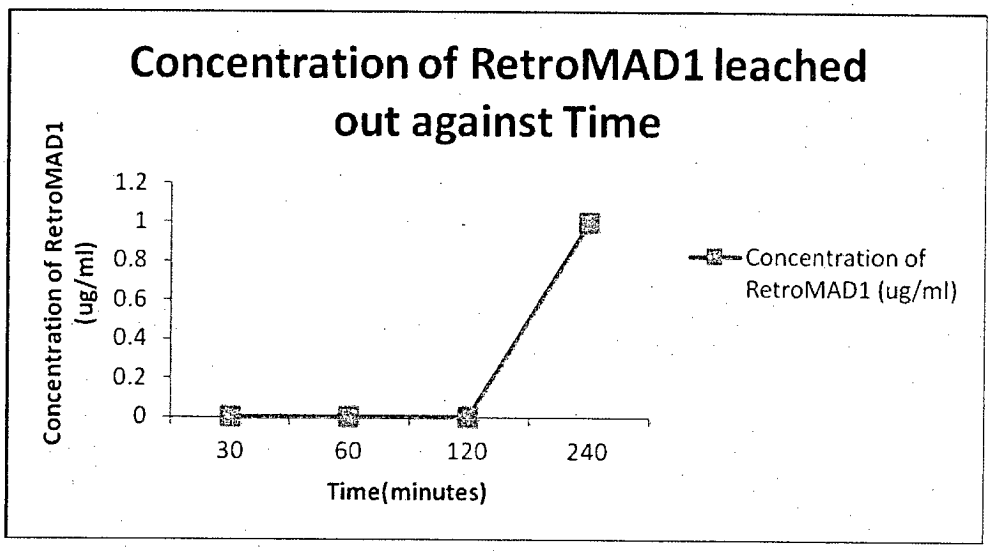


FIGURE 23 (CONTINUED)

A



B

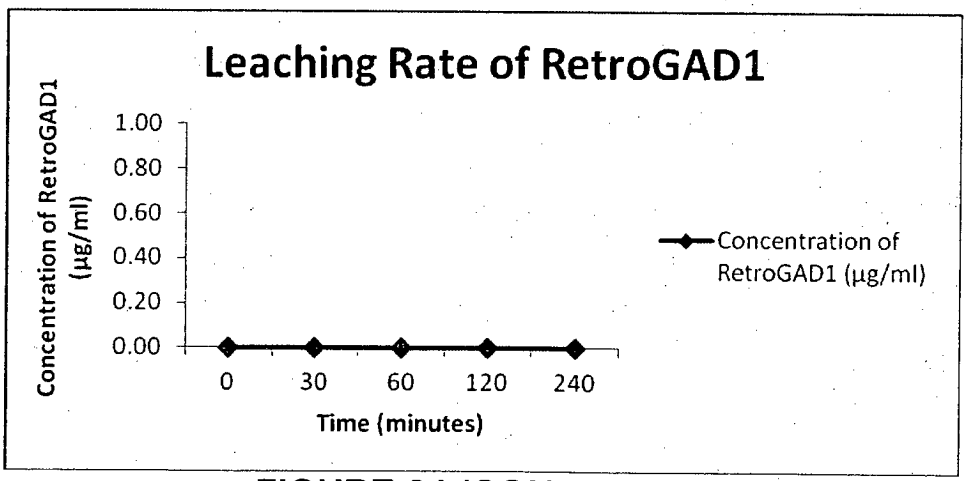
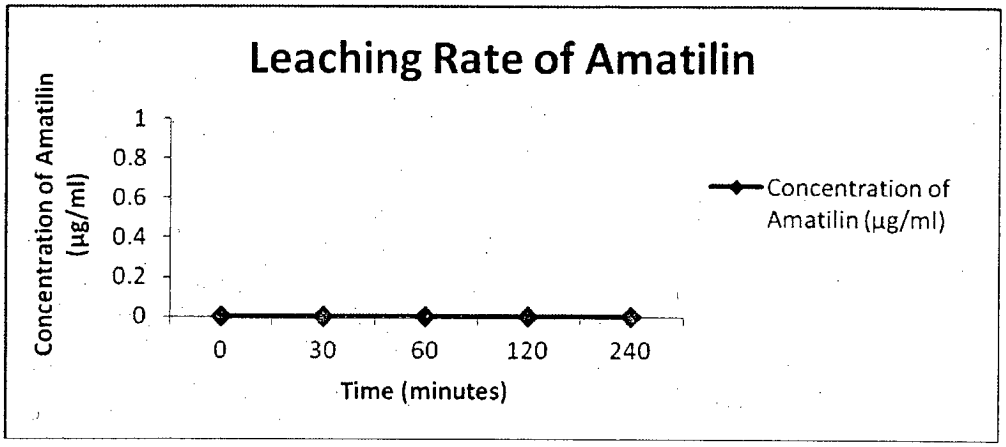


FIGURE 24 (CONTINUED)

C



D

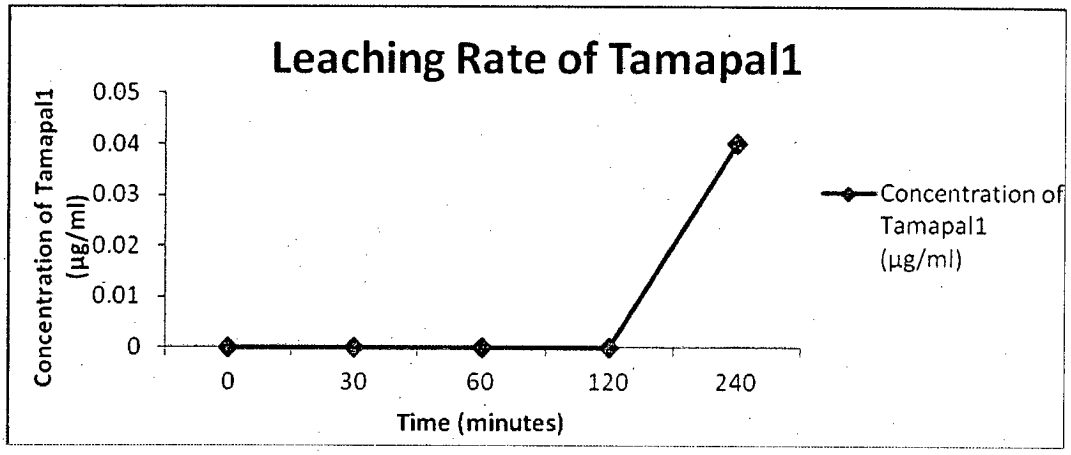


FIGURE 24 (CONTINUED)

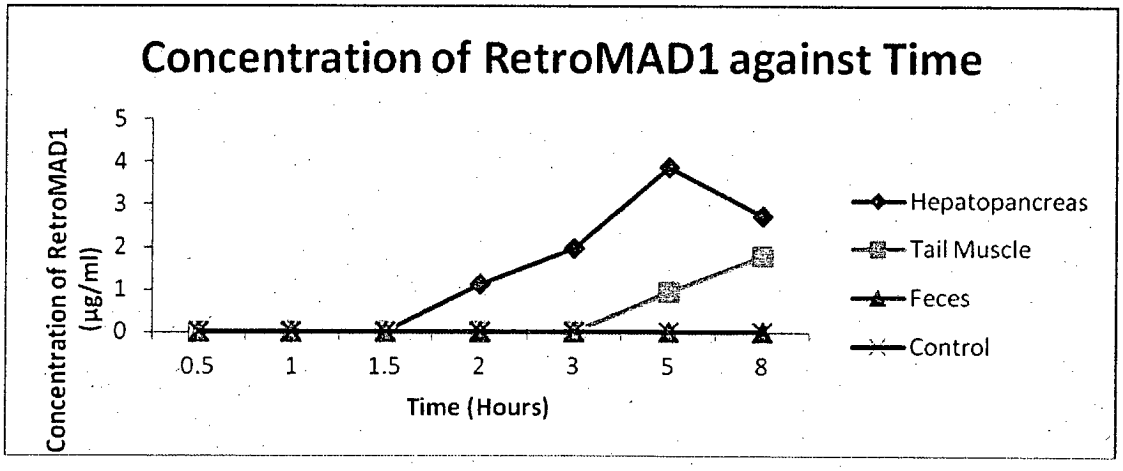


FIGURE 25

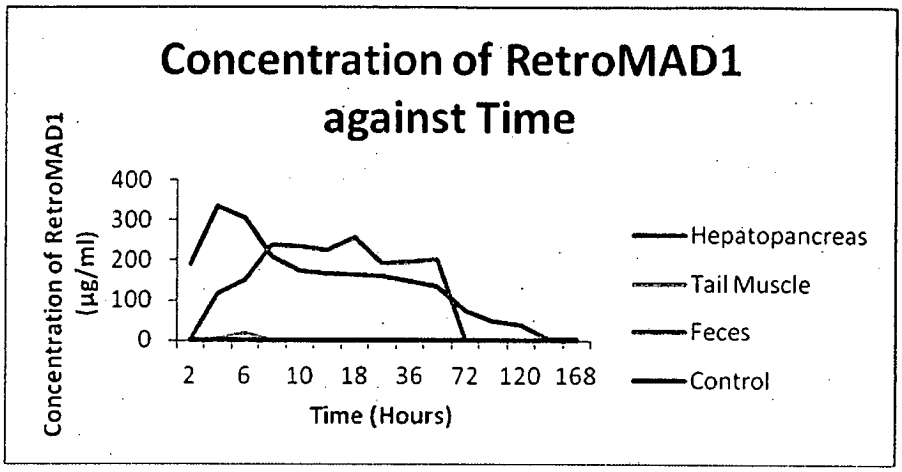


FIGURE 26

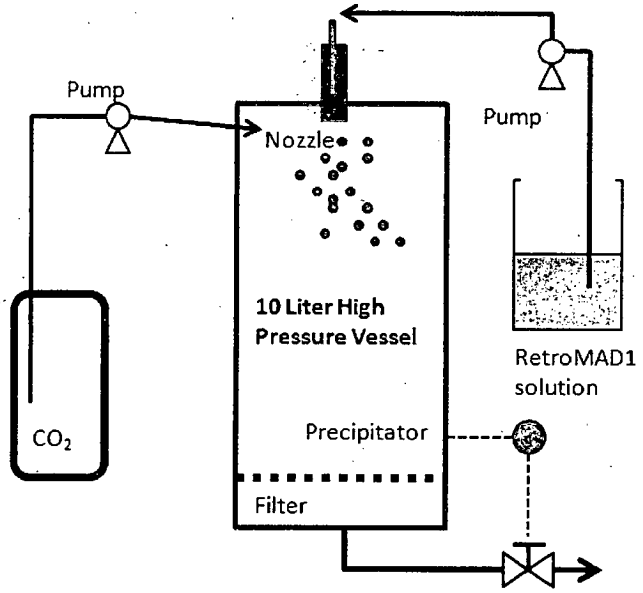


FIGURE 27

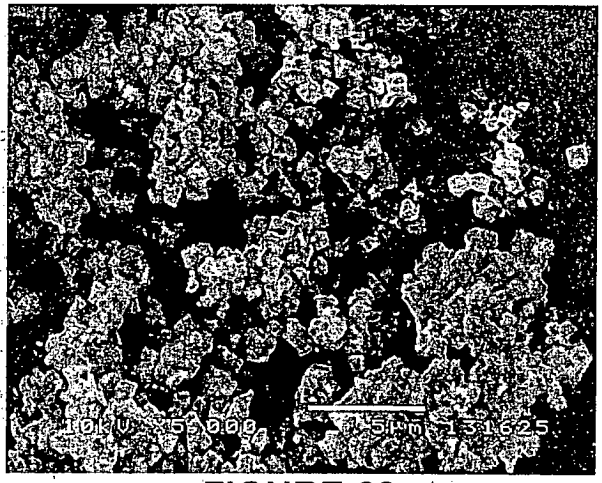


FIGURE 28

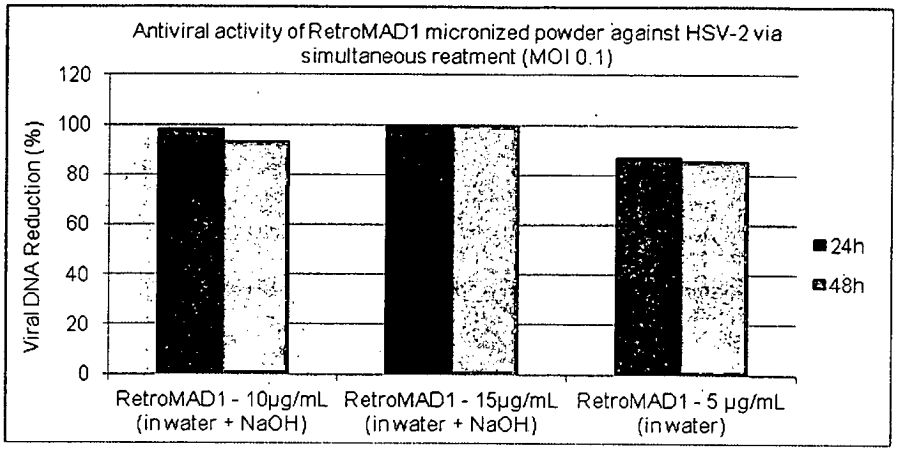


FIGURE 29

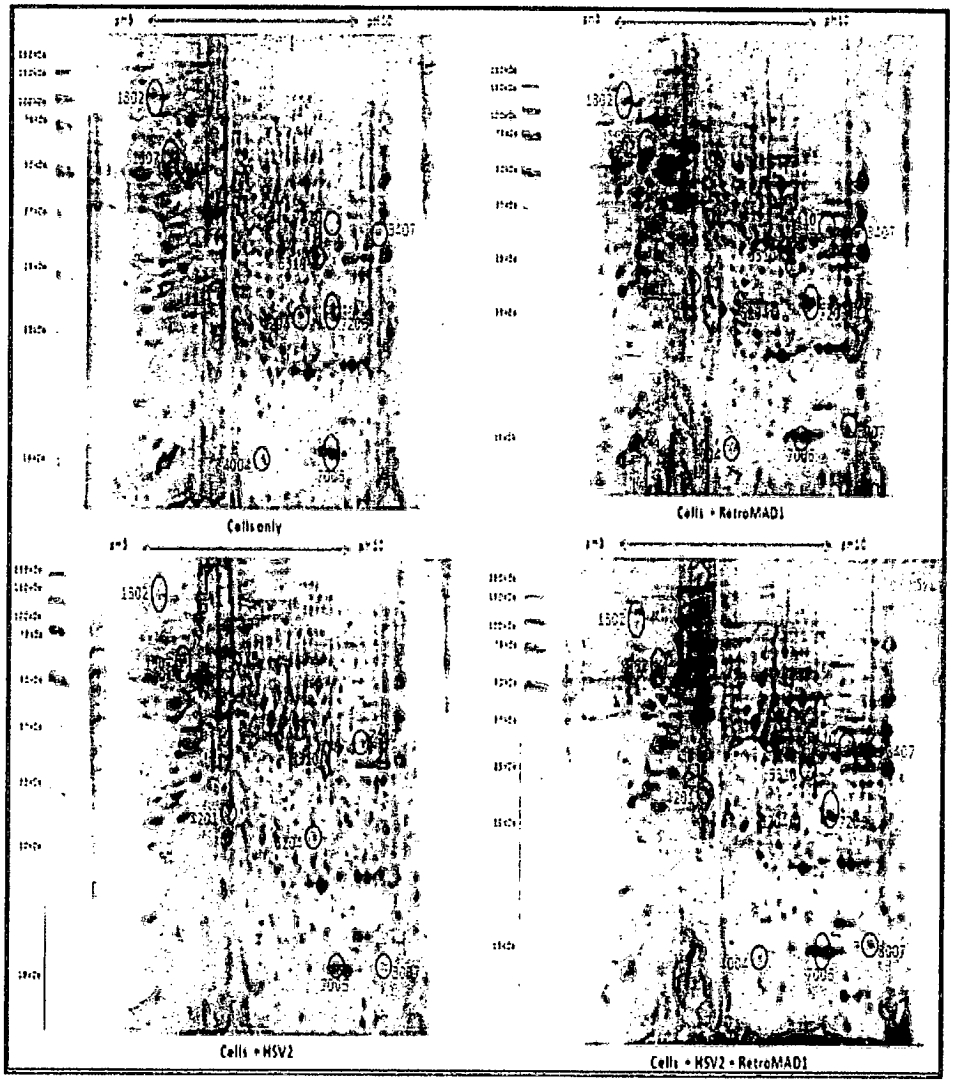


FIGURE 30

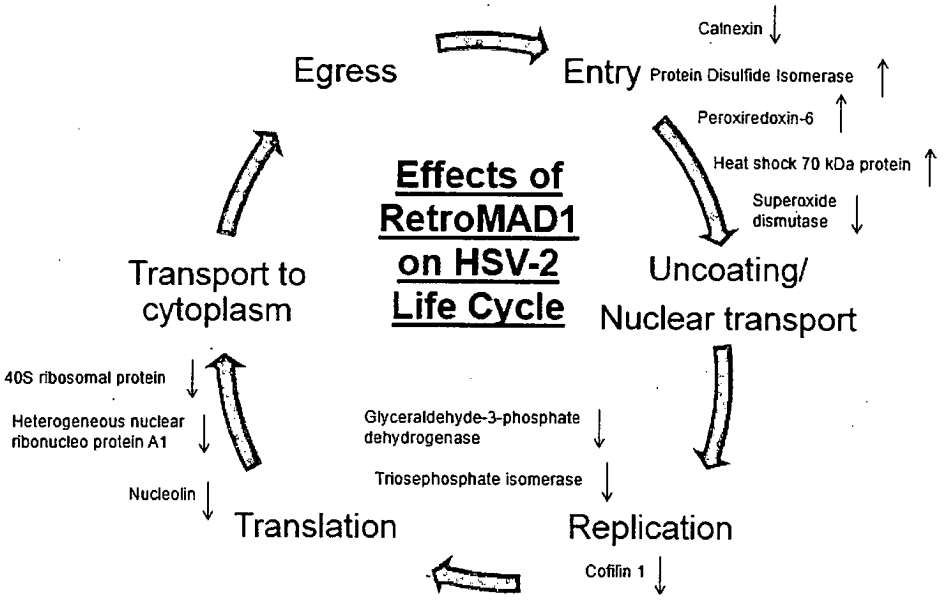


FIGURE 31

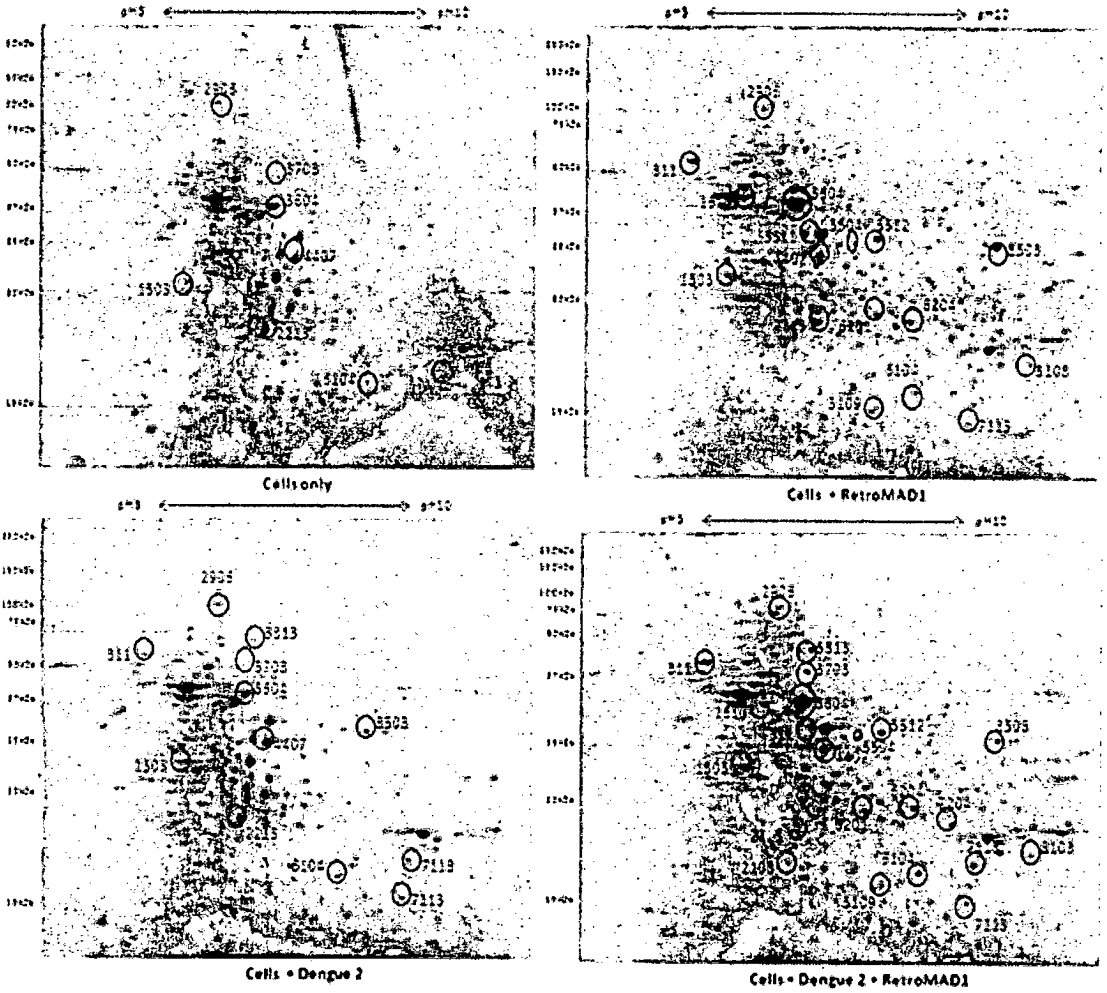


FIGURE 32

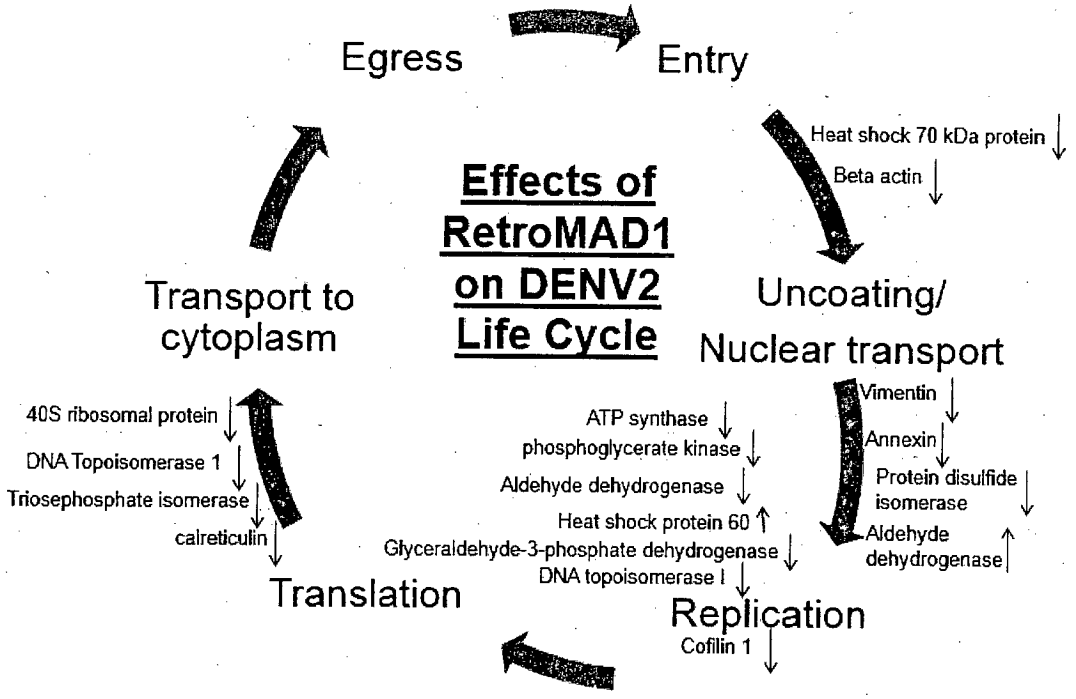

Egress
Entry
Heat shock 70 kDa protein
Beta actin
Effects of RetroMAD1 on DENV2 Life Cycle
Transport to cytoplasm
Uncoating/ Nuclear transport
40S ribosomal protein
DNA Topoisomerase 1
Triosephosphate isomerase
calreticulin
ATP synthase
phosphoglycerate kinase
Aldehyde dehydrogenase
Heat shock protein 60
Glyceraldehyde-3-phosphate dehydrogenase
DNA topoisomerase I
Vimentin
Annexin
Protein disulfide isomerase
Aldehyde dehydrogenase
Translation
Replication
Cofilin 1

FIGURE 33