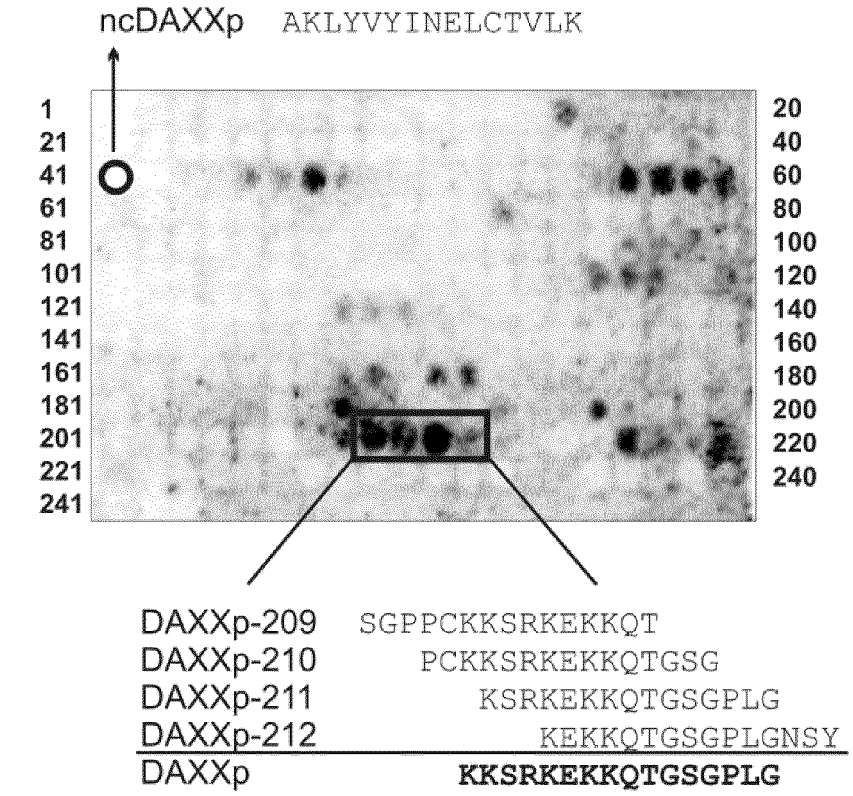
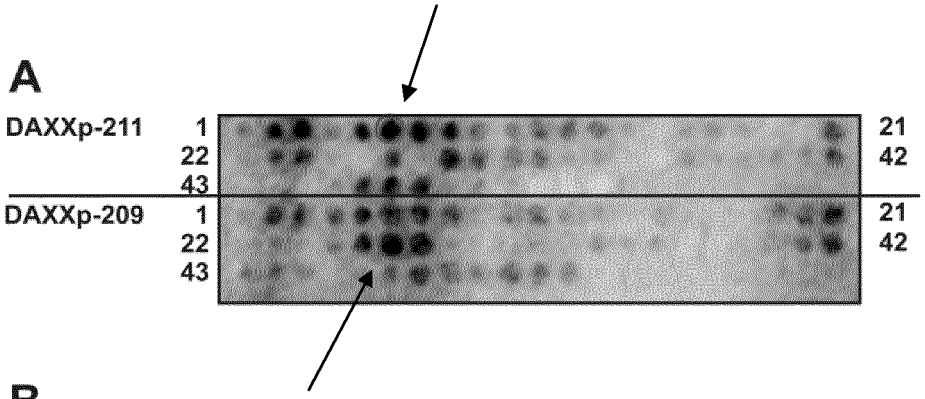
A



B

```
582  ISFFRKQSEEPFTTVLENGAGMVSSTSFNGGVSPHNWGDSGPPCKKSRKEKKQTGSGPLG  641  DAXX_CHLAE
586  ISSSRKQSEEPFTTVLENGAGMVSSTSFNGGVSPHNWGDSGPPCKKSRKEKKQTGSGPLG  645  DAXX_HUMAN
584  ISSSRKQSEEPLTTVLENGAAMVTSTSFNGGVSPHTWGDSCPPCKKSRKEK-ETGAEPLG  642  DAXX_CANFA
586  ISSPRKKSEDSLPTILENGAAVVTSTSVNGRVSSHTWRDASPPSKRFRKEKKQLGSGLLG  645  DAXX_MOUSE
577  ISSSRRKSDSSLPTILENGAAMVTSTSFNGRVSSHPCRDASPPSKRFRKEKKQLGPGPLG  636  DAXX_RAT
     **  *::*:..:.*:*****.:*:***.** **.*   *: **.*: **** : *.  **
```

Figure 1



B

DAXXp-211 (spot no. 6)	K S R K E K K Q T G S G P L G
DAXXp-209 (spot no. 27)	S G P P C K K S R K E K K Q T
DAXXp	K K S R K E K K Q T G S G L P

Figure 2

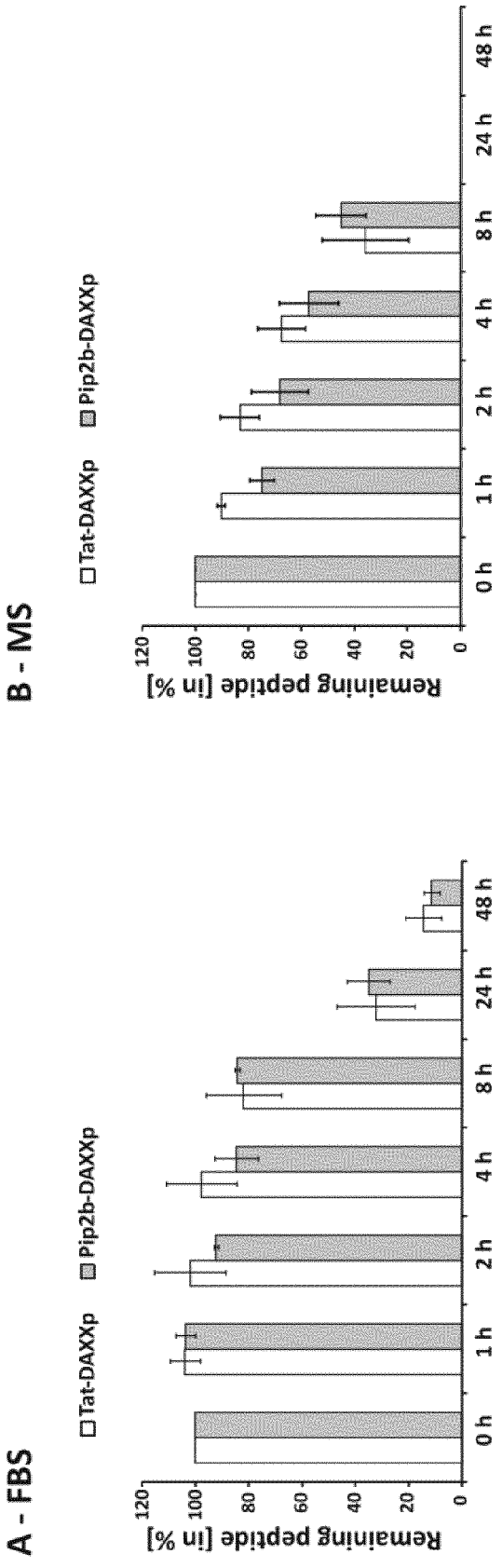

A - FBS
B - MS
Tat-DAXXp
Pip2b-DAXXp
Remaining peptide [in %]
0
20
40
60
80
100
120
0 h
1 h
2 h
4 h
8 h
24 h
48 h

Figure 3

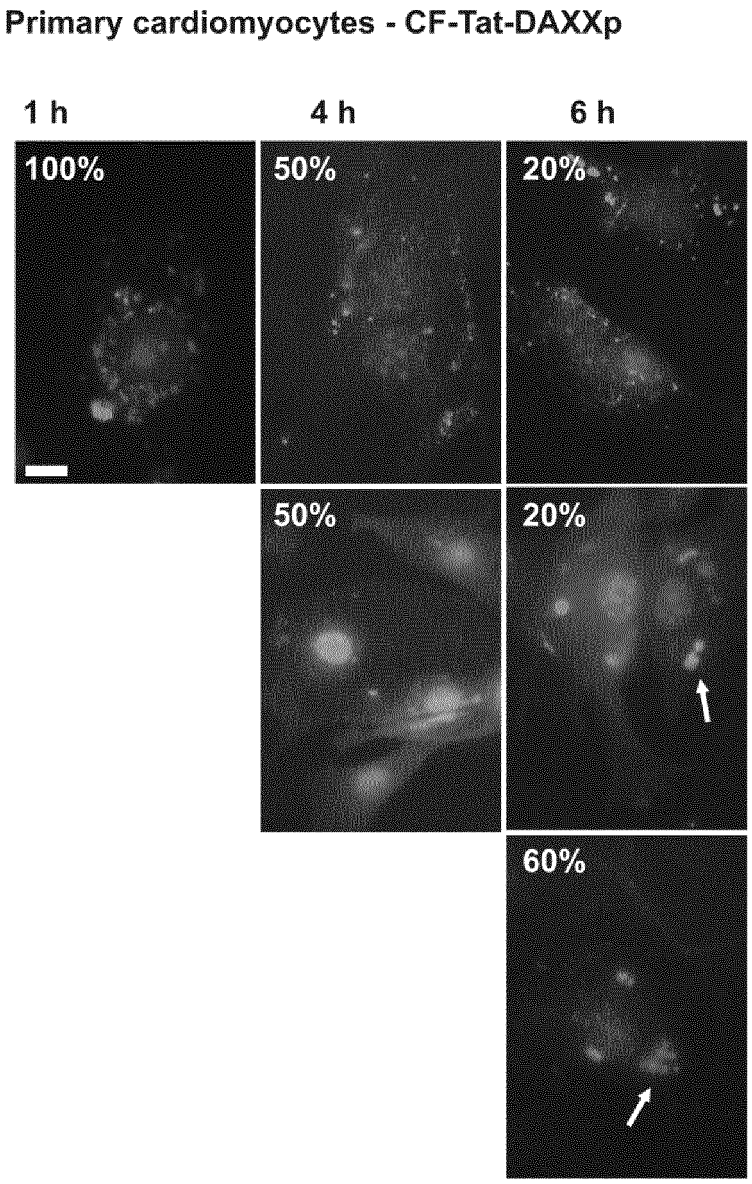

Primary cardiomyocytes - CF-Tat-DAXXp
1 h
4 h
6 h
100%
50%
20%
50%
20%
60%

Figure 4

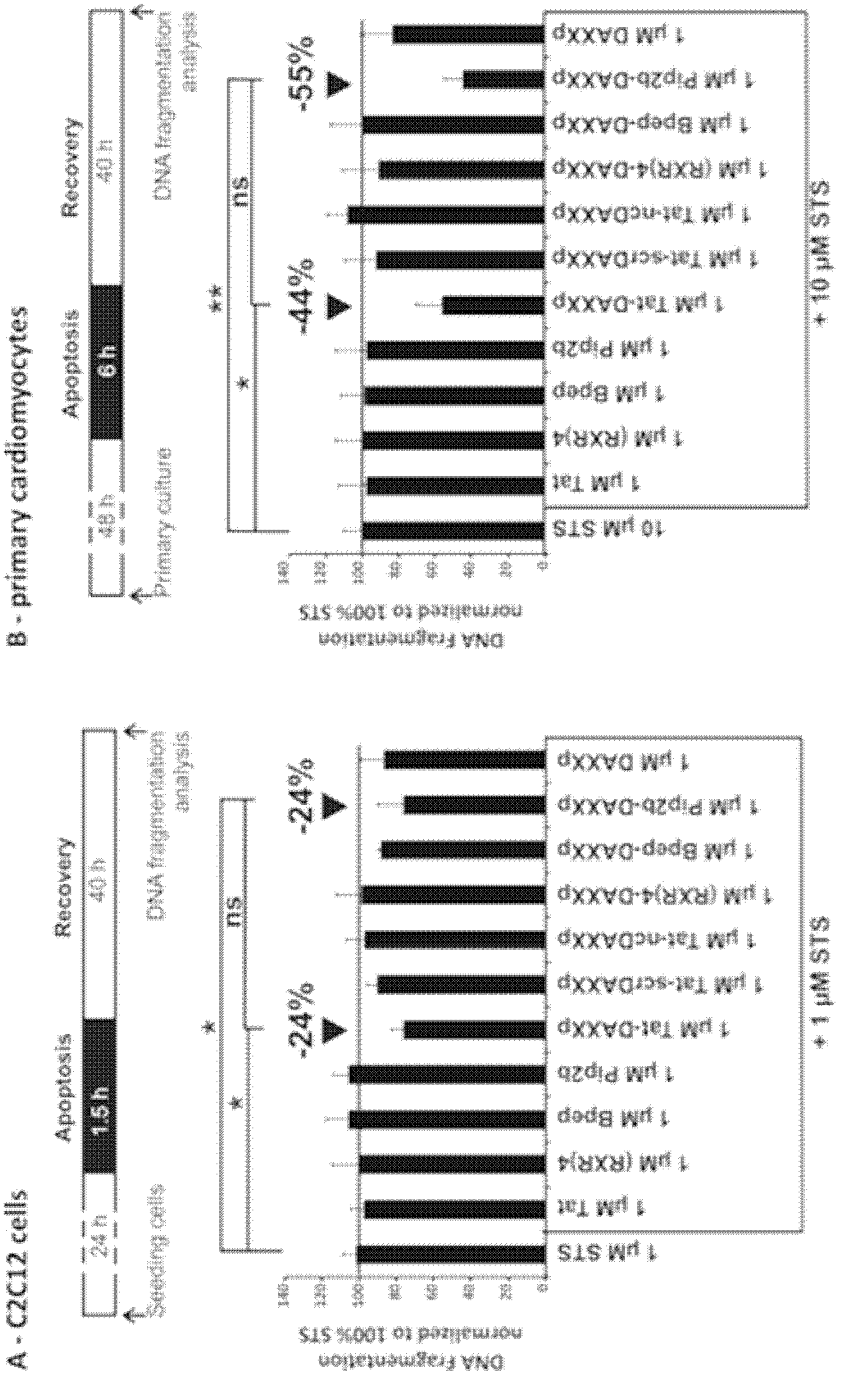

B - primary cardiomyocytes
Apoptosis
Recovery
6 h
40 h
48 h
Primary culture
DNA fragmentation analysis
-55%
ns
-44%
**
*
DNA fragmentation normalized to 100% STS
10 µM STS
1 µM Tat
1 µM (RXR)4
1 µM Bpep
1 µM PIpZb
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-ncDAXXp
1 µM (RXR)4-DAXXp
1 µM Bpep-DAXXp
1 µM PIpZb-DAXXp
1 µM DAXXp
+ 10 µM STS
A - C2C12 cells
Apoptosis
Recovery
15 h
40 h
24 h
Seeding cells
DNA fragmentation analysis
-24%
ns
-24%
*
*
DNA fragmentation normalized to 100% STS
1 µM STS
1 µM Tat
1 µM (RXR)4
1 µM Bpep
1 µM PIpZb
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-ncDAXXp
1 µM (RXR)4-DAXXp
1 µM Bpep-DAXXp
1 µM PIpZb-DAXXp
1 µM DAXXp
+ 1 µM STS

Figure 5(A) -(B)

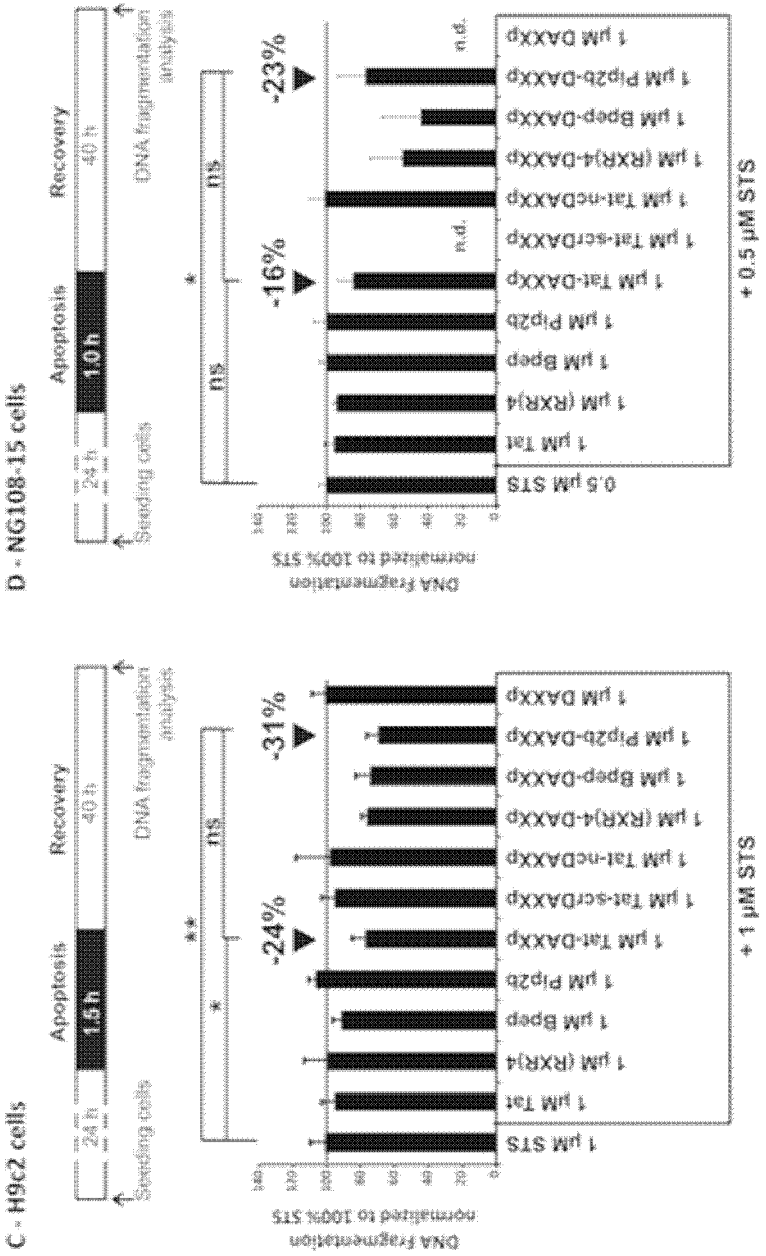

D - NG108-15 cells
Seeding cells
24 h
Apoptosis
10 h
Recovery
40 h
DNA fragmentation analysis
DNA fragmentation normalized to 100% STS
0,5 µM STS
1 µM Tat
1 µM (RXR)4
1 µM Bpep
1 µM PIpZb
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-ncDAXXp
1 µM (RXR)4-DAXXp
1 µM Bpep-DAXXp
1 µM PIpZb-DAXXp
1 µM DAXXp
+ 0.5 µM STS
ns
*
-16%
n.d.
-23%
C - H9c2 cells
15 h
1 µM STS
+ 1 µM STS
**
-24%
-31%

Figure 5(C) -(D)

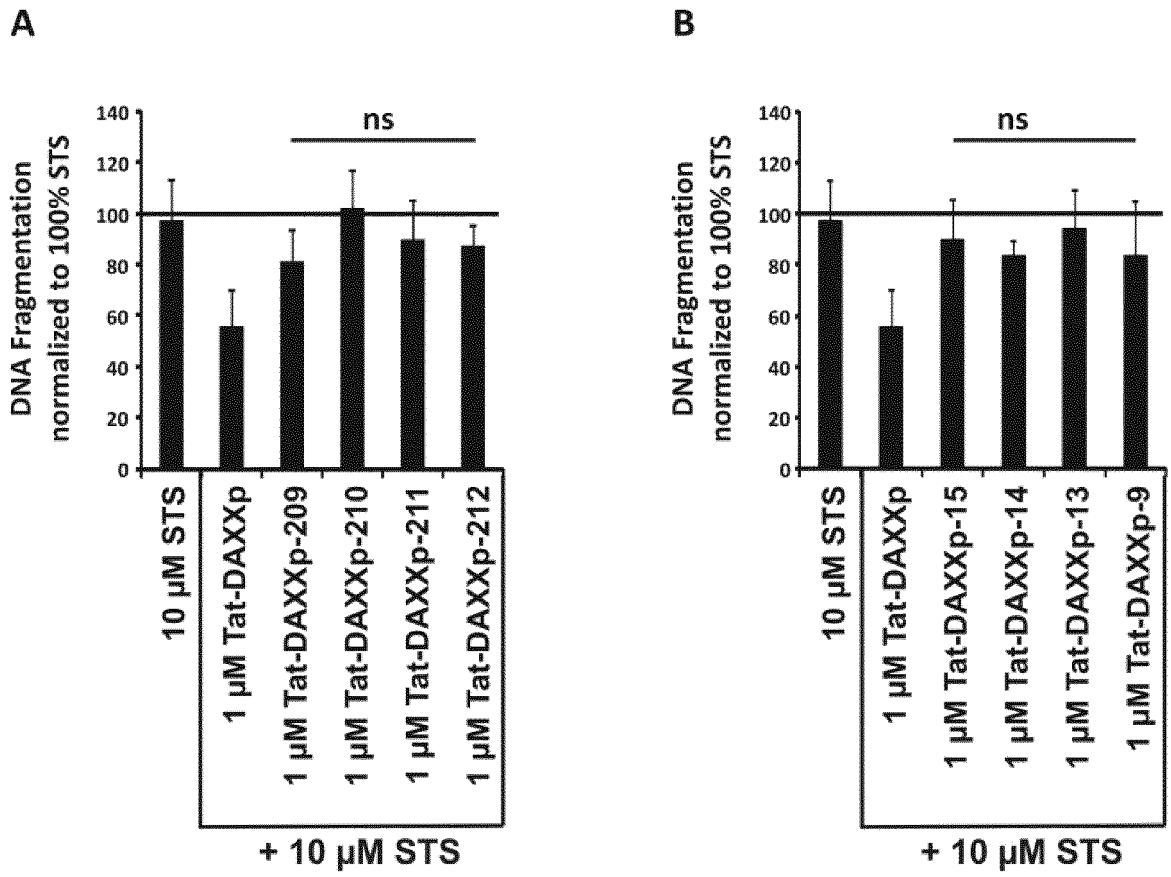

A
DNA Fragmentation normalized to 100% STS
140
120
100
80
60
40
20
0
ns
10 µM STS
1 µM Tat-DAXXp
1 µM Tat-DAXXp-209
1 µM Tat-DAXXp-210
1 µM Tat-DAXXp-211
1 µM Tat-DAXXp-212
+ 10 µM STS
B
DNA Fragmentation normalized to 100% STS
140
120
100
80
60
40
20
0
ns
10 µM STS
1 µM Tat-DAXXp
1 µM Tat-DAXXp-15
1 µM Tat-DAXXp-14
1 µM Tat-DAXXp-13
1 µM Tat-DAXXp-9
+ 10 µM STS

Figure 6

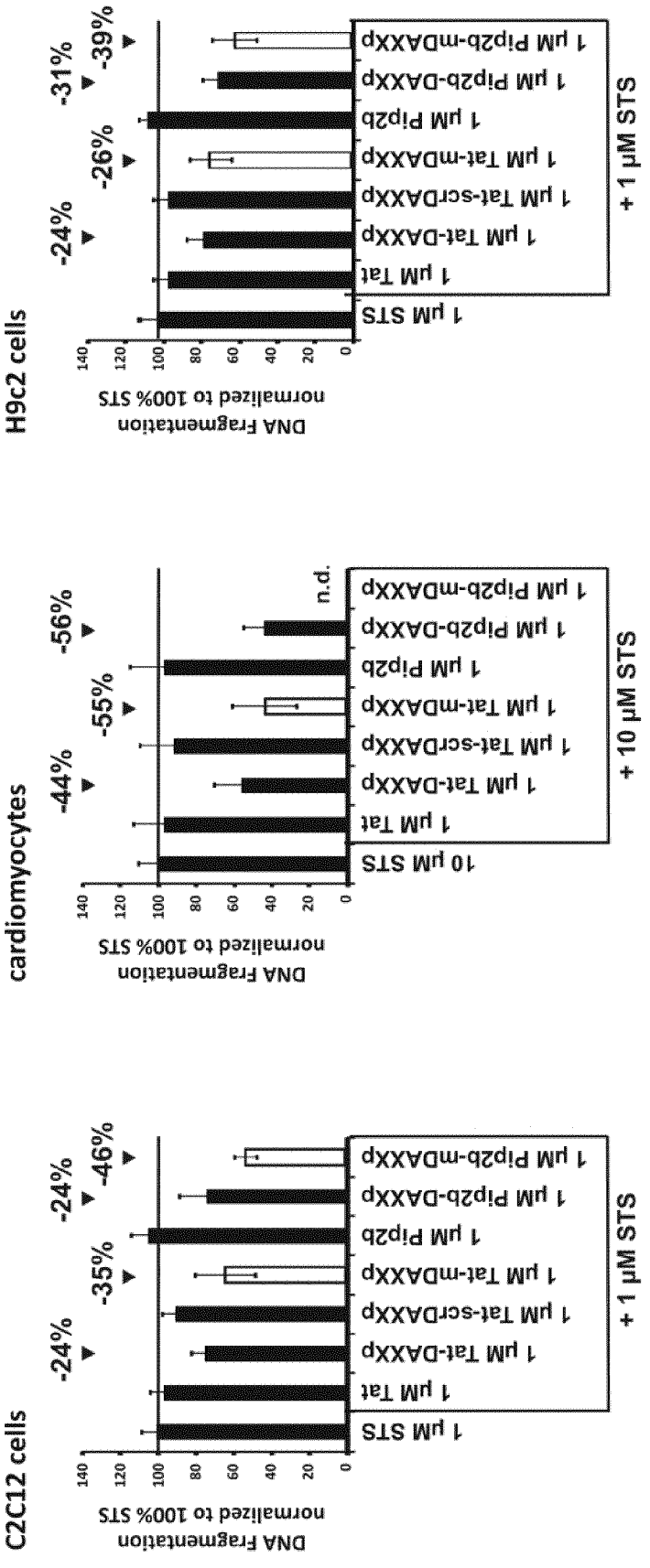

H9c2 cells
cardiomyocytes
C2C12 cells
DNA Fragmentation
normalized to 100% STS
0
20
40
60
80
100
120
140
-24%
-26%
-31%
-39%
-44%
-55%
-56%
-24%
-35%
-24%
-46%
n.d.
1 µM STS
10 µM STS
1 µM Tat
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-mDAXXp
1 µM Pip2b
1 µM Pip2b-DAXXp
1 µM Pip2b-mDAXXp
+ 1 µM STS
+ 10 µM STS

Figure 7

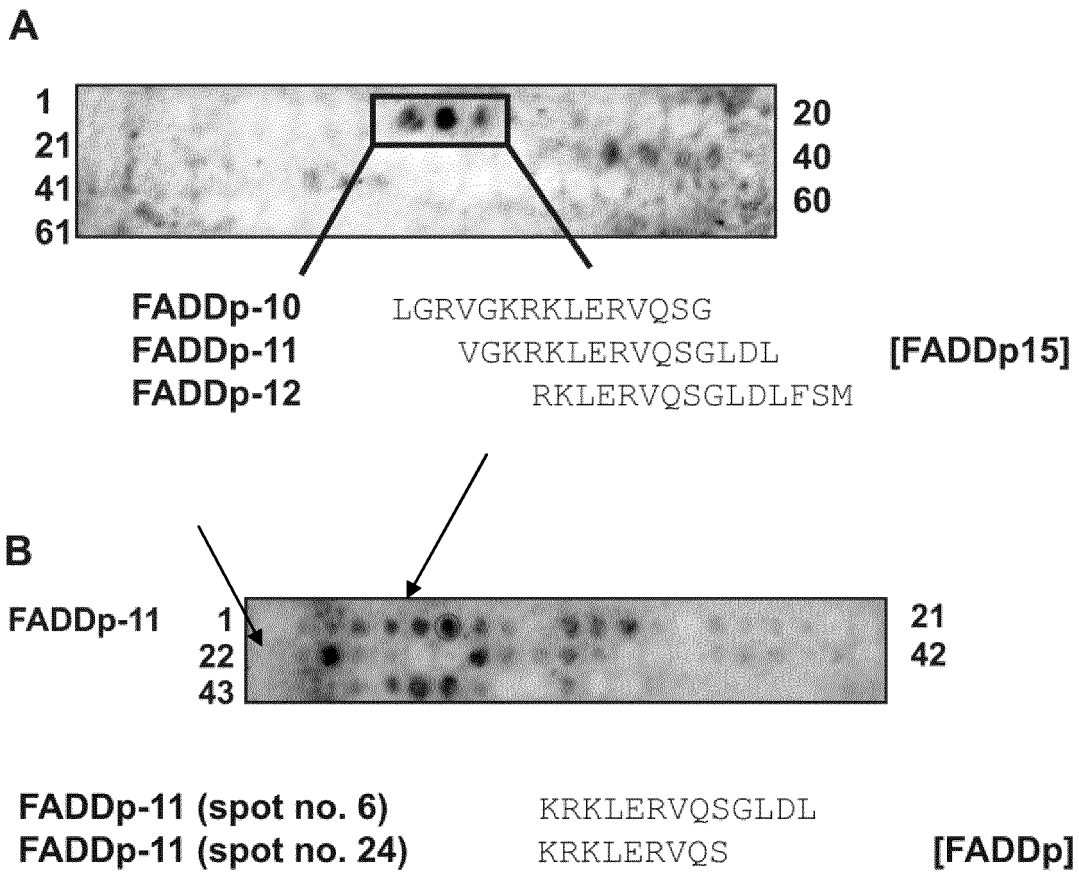

A
1
21
41
61
20
40
60
FADDp-10 LGRVGKRKLERVQSG
FADDp-11 VGKRKLERVQSGLDL [FADDp15]
FADDp-12 RKLERVQSGLDLFSM
B
FADDp-11
1
22
43
21
42
FADDp-11 (spot no. 6) KRKLERVQSGLDL
FADDp-11 (spot no. 24) KRKLERVQS [FADDp]

Figure 8

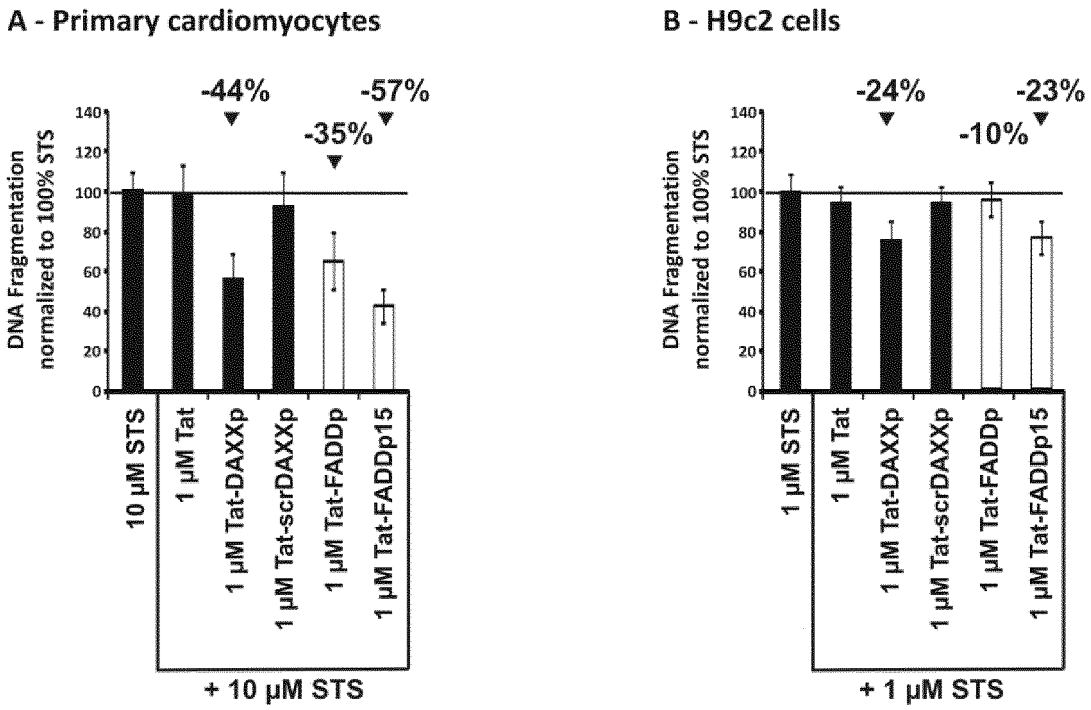

A - Primary cardiomyocytes
DNA Fragmentation normalized to 100% STS
-44%
-57%
-35%
10 µM STS
1 µM Tat
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-FADDp
1 µM Tat-FADDp15
+ 10 µM STS
B - H9c2 cells
DNA Fragmentation normalized to 100% STS
-24%
-23%
-10%
1 µM STS
1 µM Tat
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-FADDp
1 µM Tat-FADDp15
+ 1 µM STS

Figure 9

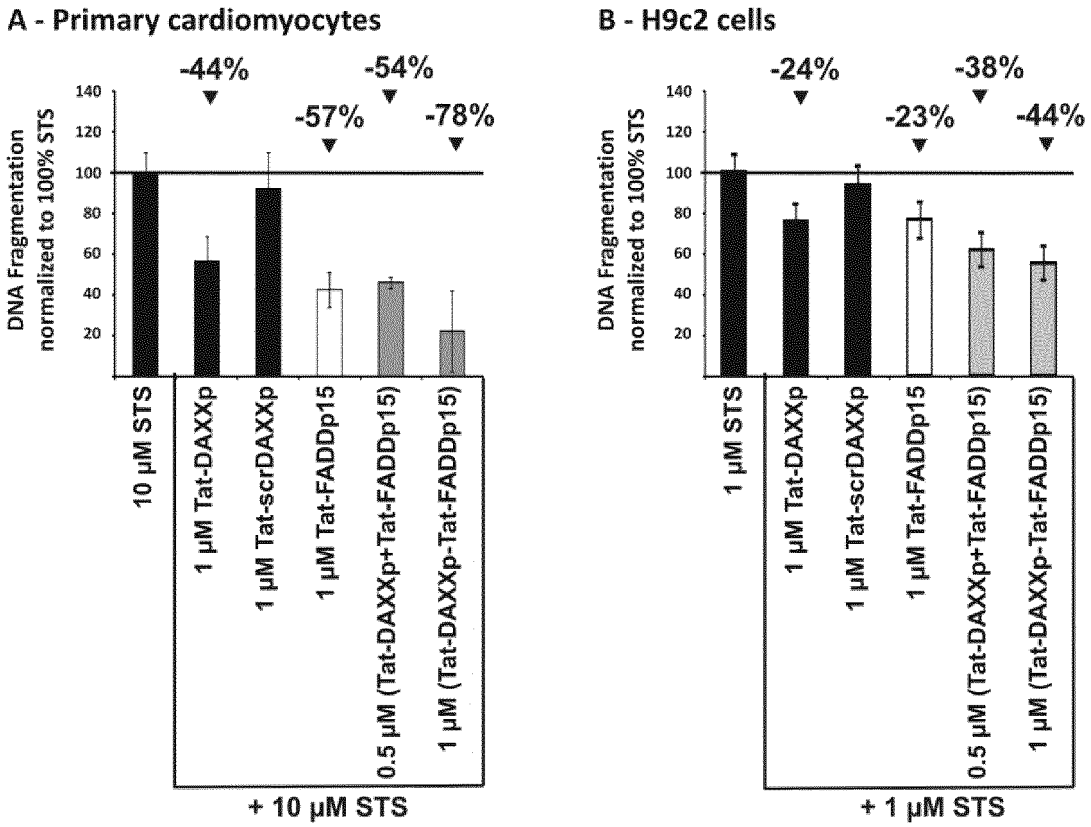

A - Primary cardiomyocytes
DNA Fragmentation normalized to 100% STS
-44%
-57%
-54%
-78%
10 µM STS
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-FADDp15
0.5 µM (Tat-DAXXp+Tat-FADDp15)
1 µM (Tat-DAXXp-Tat-FADDp15)
+ 10 µM STS
B - H9c2 cells
DNA Fragmentation normalized to 100% STS
-24%
-23%
-38%
-44%
1 µM STS
1 µM Tat-DAXXp
1 µM Tat-scrDAXXp
1 µM Tat-FADDp15
0.5 µM (Tat-DAXXp+Tat-FADDp15)
1 µM (Tat-DAXXp-Tat-FADDp15)
+ 1 µM STS

Figure 10

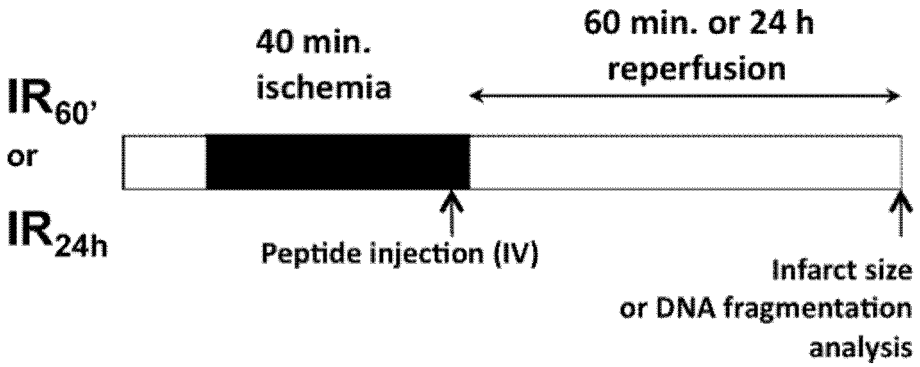

40 min.
ischemia
60 min. or 24 h
reperfusion
IR60'
or
IR24h
Peptide injection (IV)
Infarct size
or DNA fragmentation
analysis

Figure 11

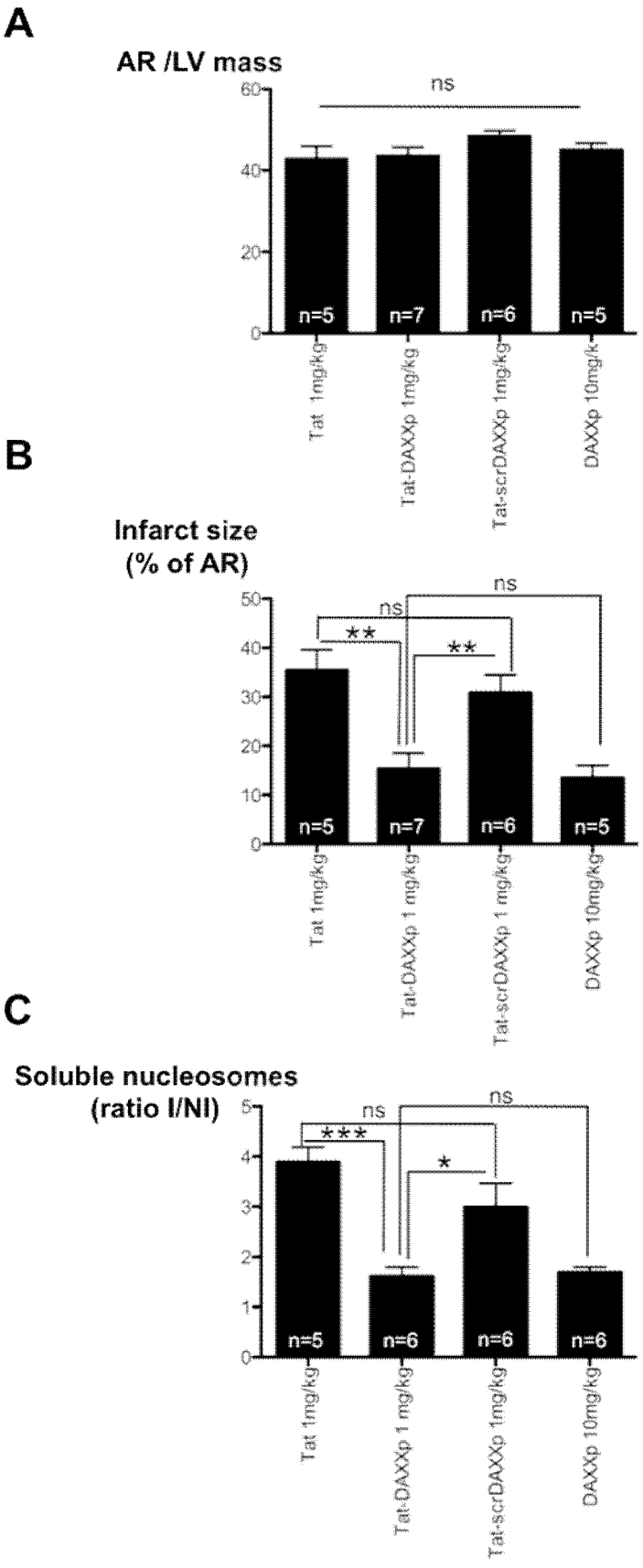

A
AR /LV mass
ns
60
40
20
0
n=5
n=7
n=6
n=5
Tat 1mg/kg
Tat-DAXXp 1mg/kg
Tat-scrDAXXp 1mg/kg
DAXXp 10mg/k
B
Infarct size
(% of AR)
ns
ns
**
**
50
40
30
20
10
0
n=5
n=7
n=6
n=5
Tat 1mg/kg
Tat-DAXXp 1 mg/kg
Tat-scrDAXXp 1 mg/kg
DAXXp 10mg/kg
C
Soluble nucleosomes
(ratio I/NI)
ns
ns
***
*
5
4
3
2
1
0
n=5
n=6
n=6
n=6
Tat 1mg/kg
Tat-DAXXp 1 mg/kg
Tat-scrDAXXp 1mg/kg
DAXXp 10mg/kg

Figure 12

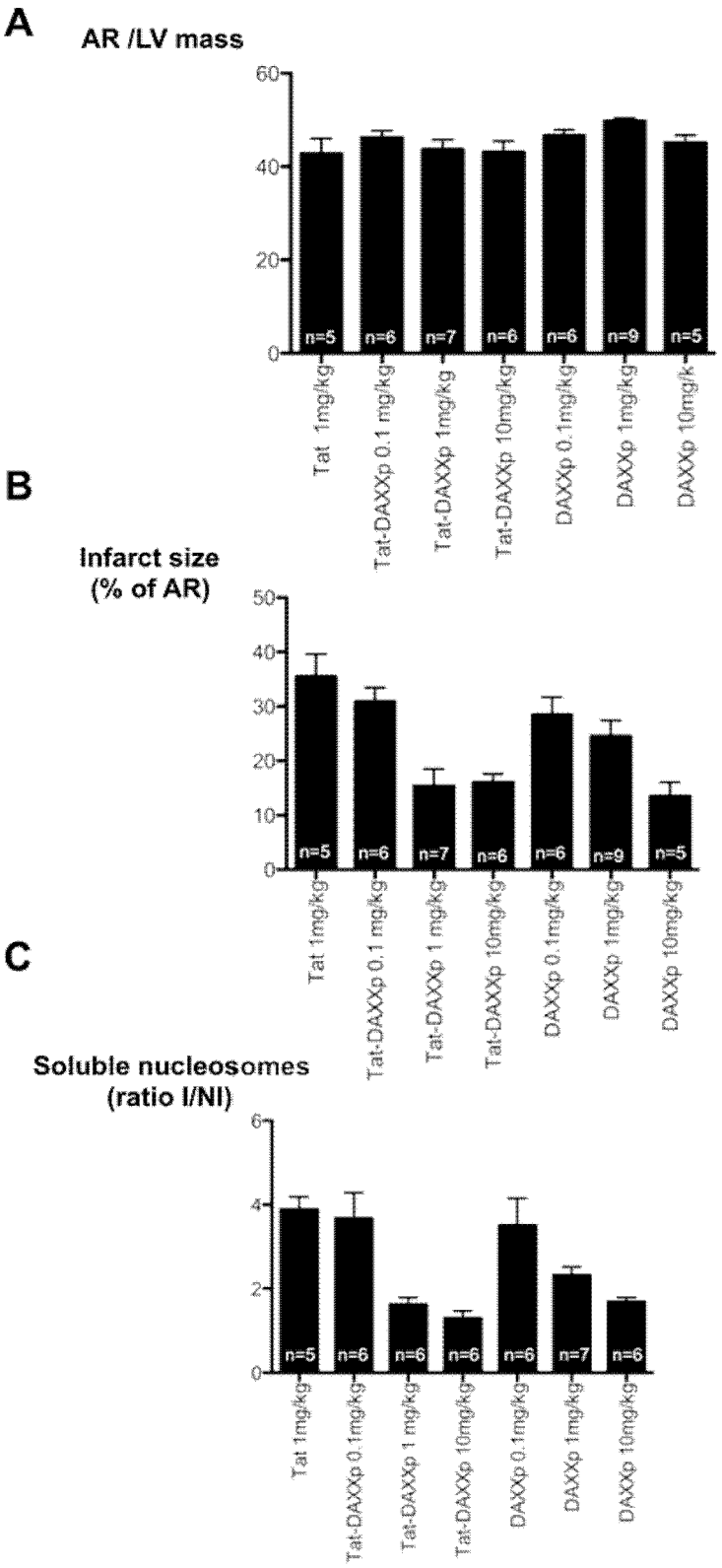

A
AR /LV mass
60
40
20
0
n=5
n=6
n=7
n=6
n=6
n=9
n=5
Tat 1mg/kg
Tat-DAXXp 0.1 mg/kg
Tat-DAXXp 1mg/kg
Tat-DAXXp 10mg/kg
DAXXp 0.1mg/kg
DAXXp 1mg/kg
DAXXp 10mg/k
B
Infarct size
(% of AR)
50
40
30
20
10
0
n=5
n=6
n=7
n=6
n=6
n=9
n=5
Tat 1mg/kg
Tat-DAXXp 0.1 mg/kg
Tat-DAXXp 1 mg/kg
Tat-DAXXp 10mg/kg
DAXXp 0.1mg/kg
DAXXp 1mg/kg
DAXXp 10mg/kg
C
Soluble nucleosomes
(ratio I/NI)
6
4
2
0
n=5
n=6
n=6
n=6
n=6
n=7
n=6
Tat 1mg/kg
Tat-DAXXp 0.1mg/kg
Tat-DAXXp 1 mg/kg
Tat-DAXXp 10mg/kg
DAXXp 0.1mg/kg
DAXXp 1mg/kg
DAXXp 10mg/kg

Figure 13

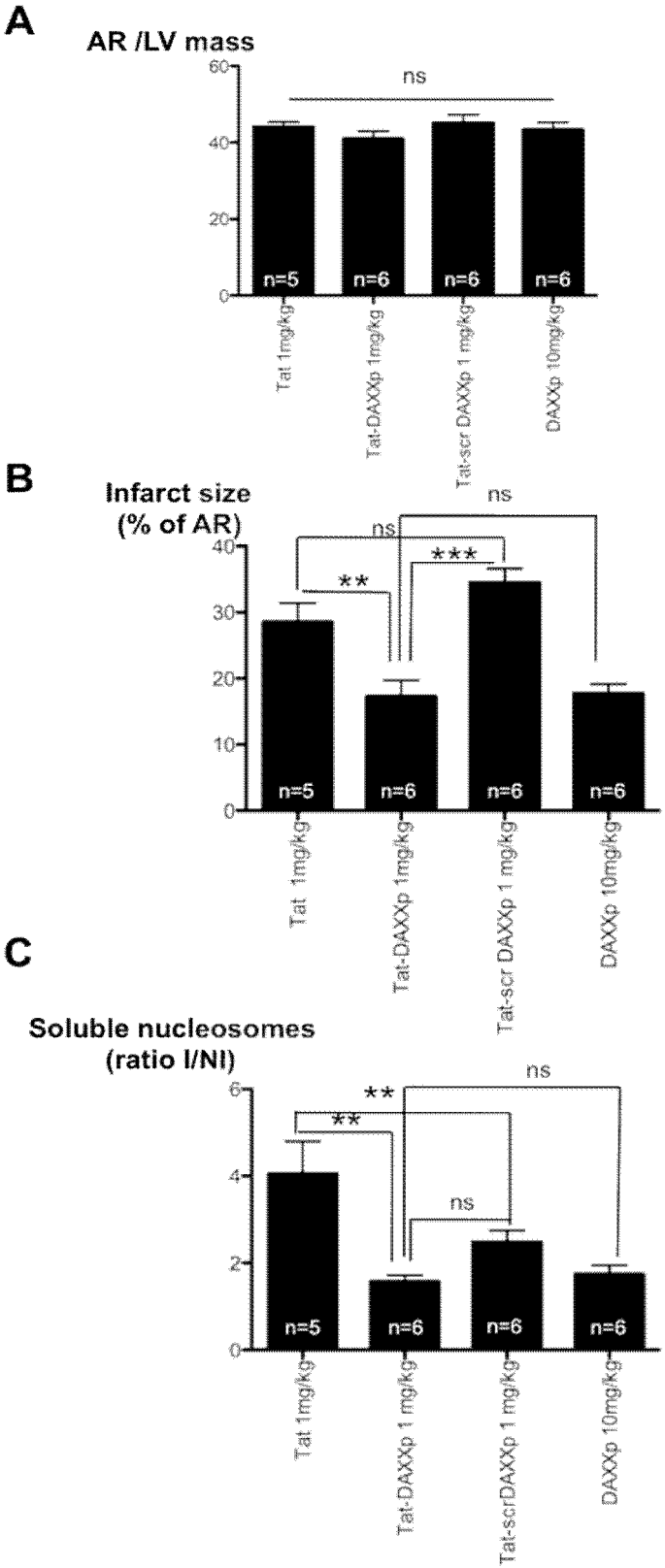

A
AR /LV mass
ns
60
40
20
0
n=5
n=6
n=6
n=6
Tat 1mg/kg
Tat-DAXXp 1mg/kg
Tat-scr DAXXp 1 mg/kg
DAXXp 10mg/kg
B
Infarct size
(% of AR)
ns
ns
***
**
40
30
20
10
0
n=5
n=6
n=6
n=6
Tat 1mg/kg
Tat-DAXXp 1mg/kg
Tat-scr DAXXp 1 mg/kg
DAXXp 10mg/kg
C
Soluble nucleosomes
(ratio I/NI)
ns
**
**
ns
6
4
2
0
n=5
n=6
n=6
n=6
Tat 1mg/kg
Tat-DAXXp 1 mg/kg
Tat-scrDAXXp 1 mg/kg
DAXXp 10mg/kg

Figure 14

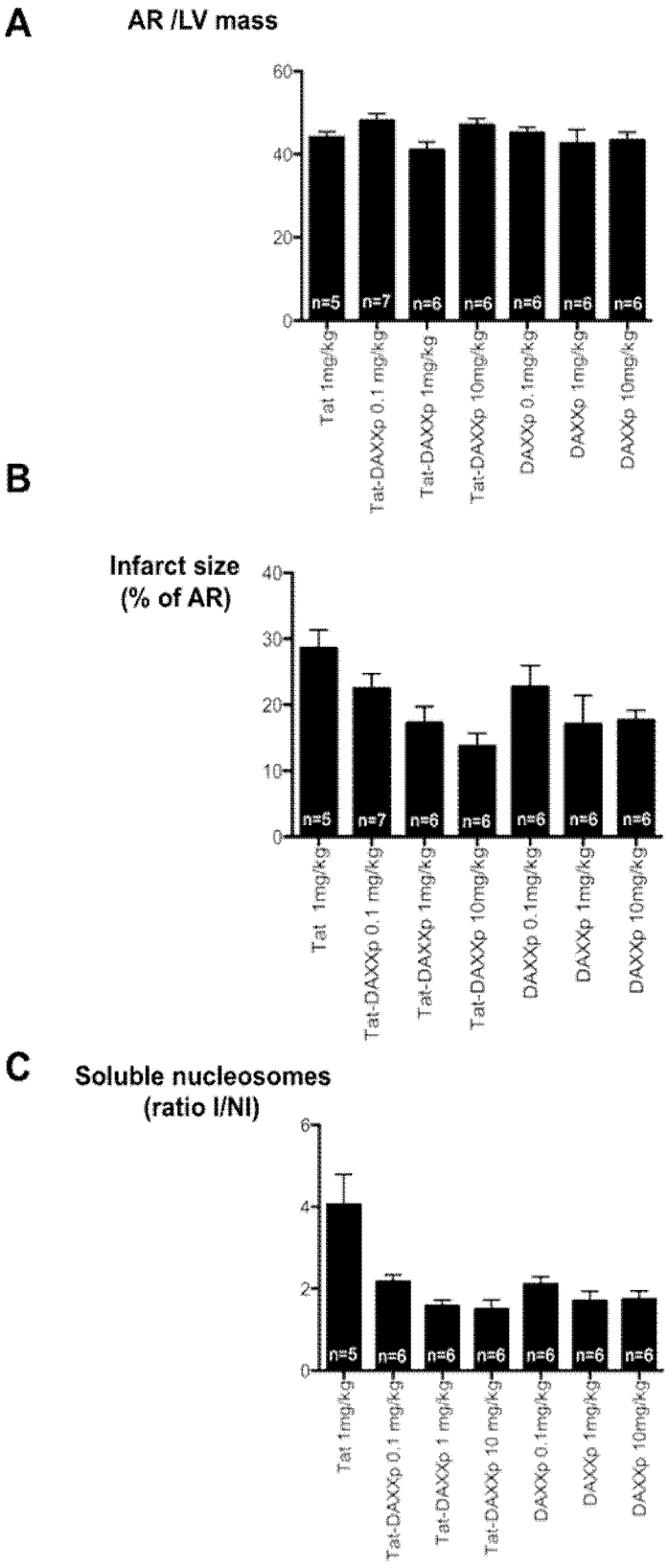

A
AR /LV mass
60
40
20
0
n=5
n=7
n=6
n=6
n=6
n=6
n=6
Tat 1mg/kg
Tat-DAXXp 0.1 mg/kg
Tat-DAXXp 1mg/kg
Tat-DAXXp 10mg/kg
DAXXp 0.1mg/kg
DAXXp 1mg/kg
DAXXp 10mg/kg
B
Infarct size
(% of AR)
40
30
20
10
0
n=5
n=7
n=6
n=6
n=6
n=6
n=6
Tat 1mg/kg
Tat-DAXXp 0.1 mg/kg
Tat-DAXXp 1mg/kg
Tat-DAXXp 10mg/kg
DAXXp 0.1mg/kg
DAXXp 1mg/kg
DAXXp 10mg/kg
C
Soluble nucleosomes
(ratio I/NI)
6
4
2
0
n=5
n=6
n=6
n=6
n=6
n=6
n=6
Tat 1mg/kg
Tat-DAXXp 0.1 mg/kg
Tat-DAXXp 1 mg/kg
Tat-DAXXp 10 mg/kg
DAXXp 0.1mg/kg
DAXXp 1mg/kg
DAXXp 10mg/kg

Figure 15

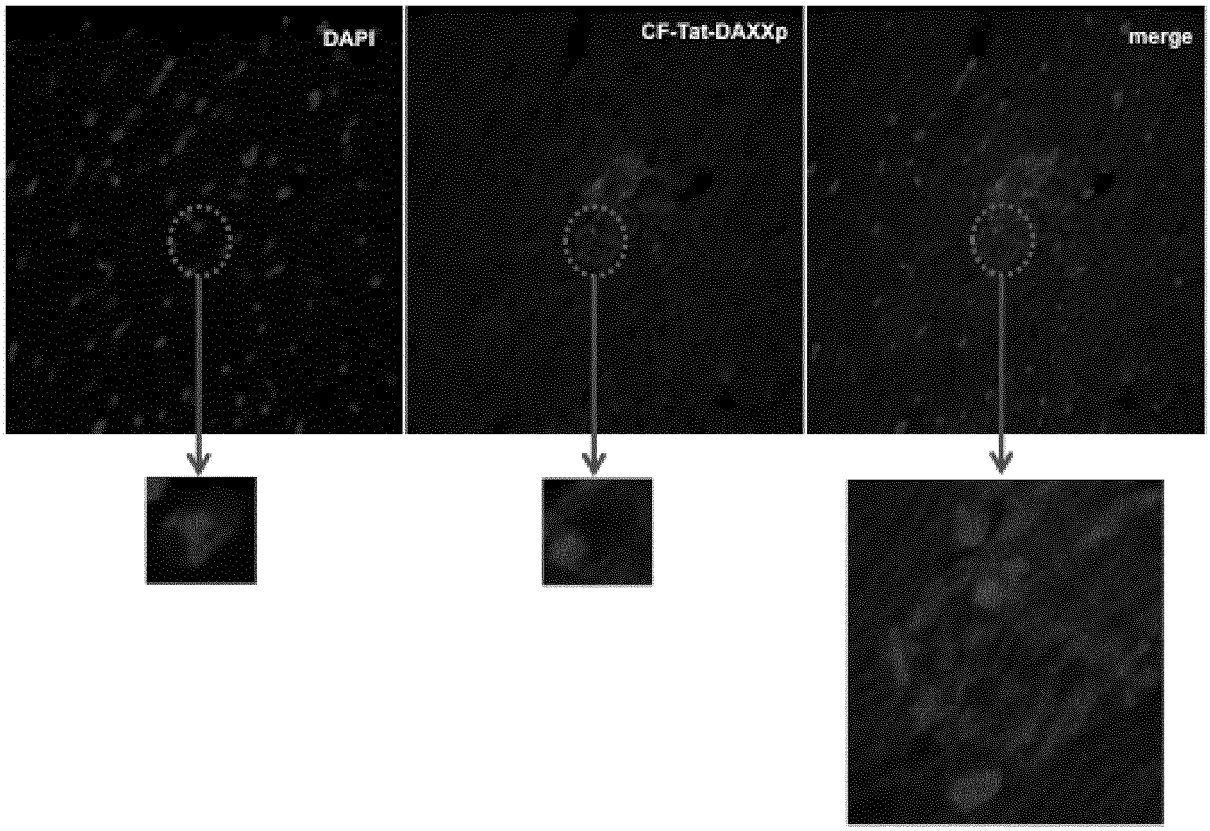
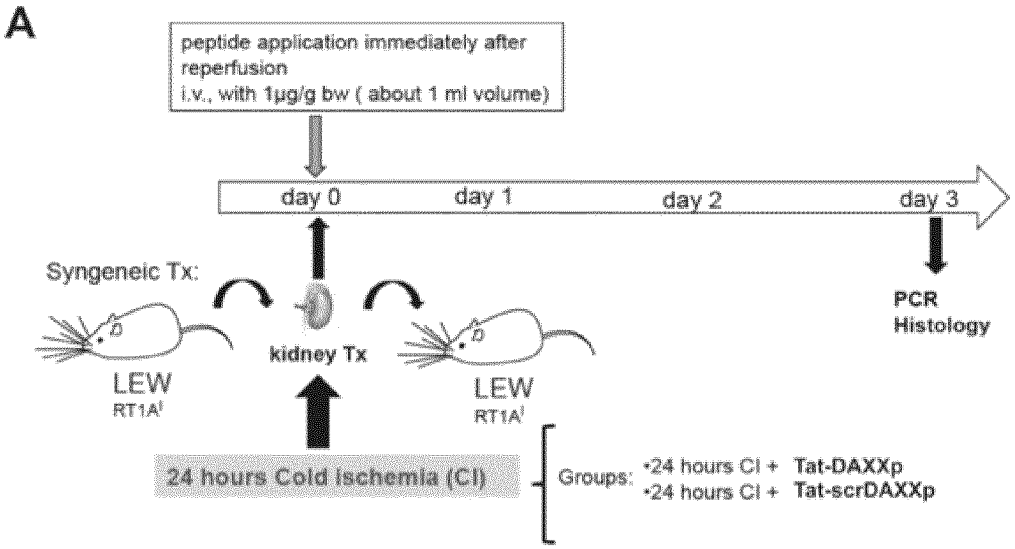


Figure 16


A
peptide application immediately after reperfusion
i.v., with 1µg/g bw (about 1 ml volume)
day 0
day 1
day 2
day 3
PCR
Histology
Syngeneic Tx:
kidney Tx
LEW
RT1A^l
LEW
RT1A^l
24 hours Cold Ischemia (CI)
Groups:
•24 hours CI + Tat-DAXXp
•24 hours CI + Tat-scrDAXXp

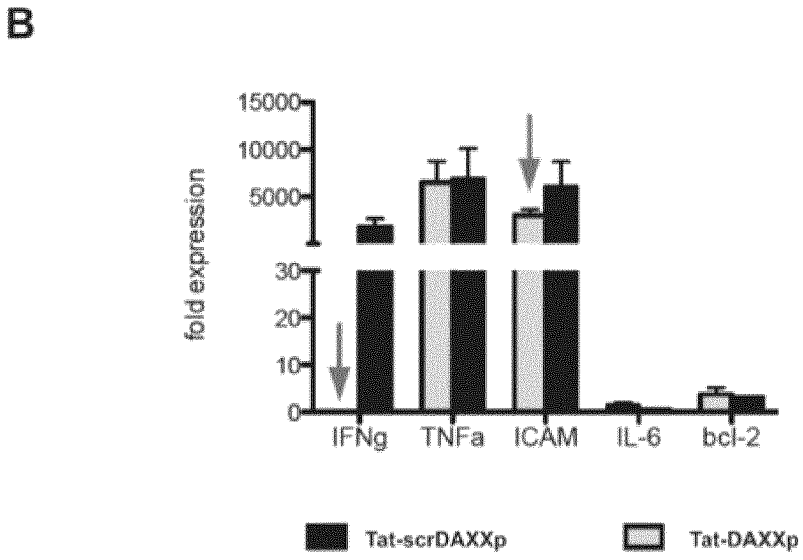

B
fold expression
15000
10000
5000
30
20
10
0
IFNg
TNFa
ICAM
IL-6
bcl-2
Tat-scrDAXXp
Tat-DAXXp

Figure 17