The Role of Excision DNA Repair in

Chloroacetaldehyde (CAA) Induced Neurotoxicity

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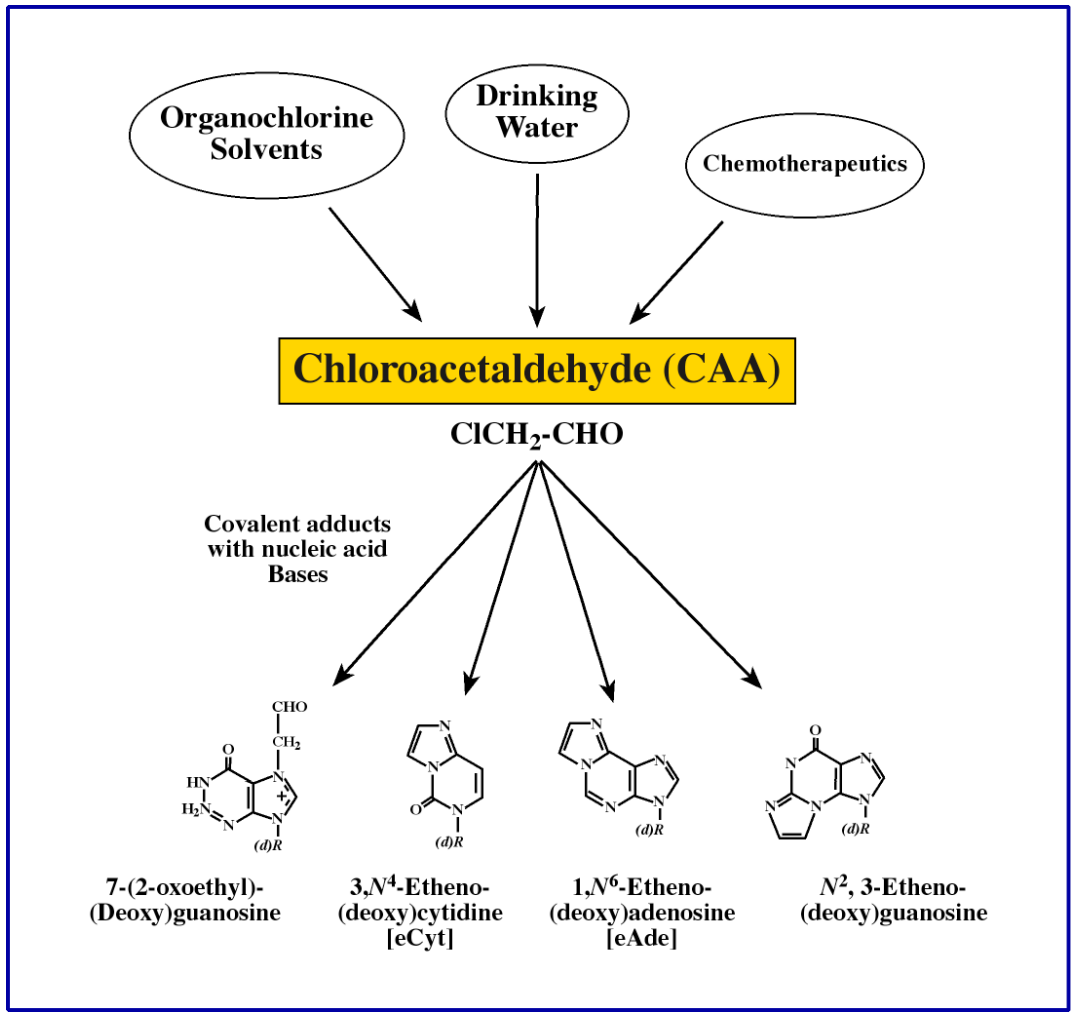
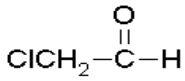

Organochlorine Solvents
Drinking Water
Chemotherapeutics
Chloroacetaldehyde (CAA)
ClCH2-CHO
Covalent adducts with nucleic acid Bases
7-(2-oxoethyl)-(Deoxy)guanosine
3,N4-Etheno-(deoxy)cytidine [eCyt]
1,N6-Etheno-(deoxy)adenosine [eAde]
N2, 3-Etheno-(deoxy)guanosine



2-Chloroacetaldehyde (CAA)

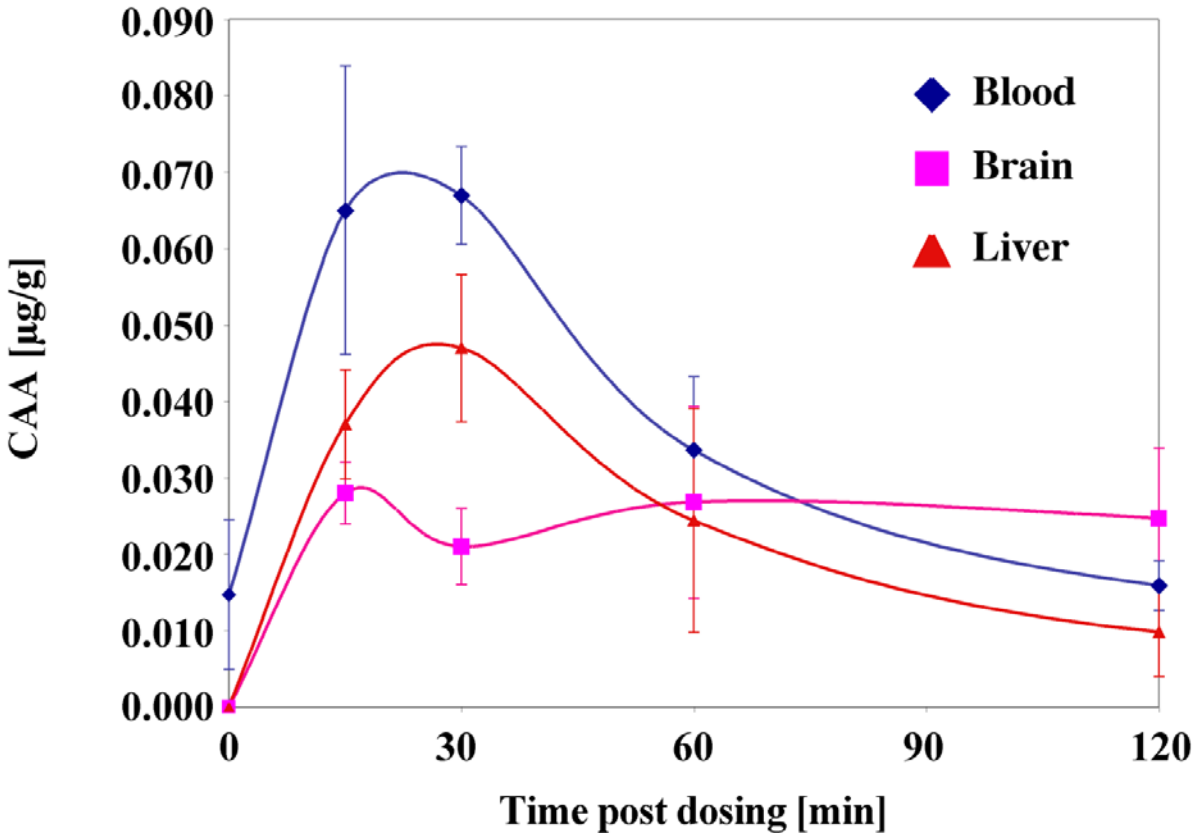
Postnatal day 6

Time intervals
[0h to 8h]

Blood **Liver** **Brain**

Solvent extraction
+ internal standard

Blood **Liver** **Brain**


0.090
0.080
0.070
0.060
0.050
0.040
0.030
0.020
0.010
0.000
CAA [µg/g]
0
30
60
90
120
Time post dosing [min]
Blood
Brain
Liver

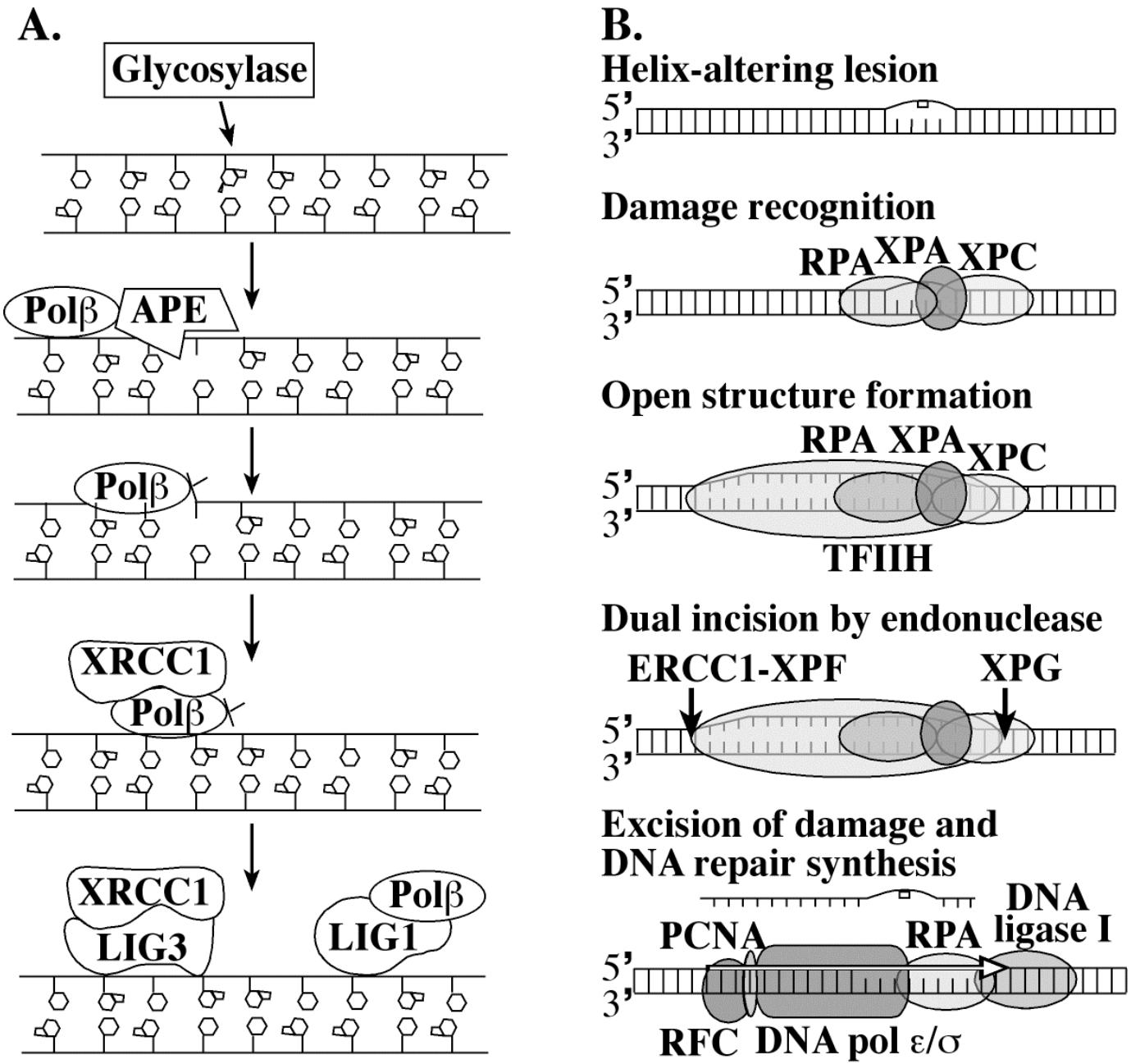
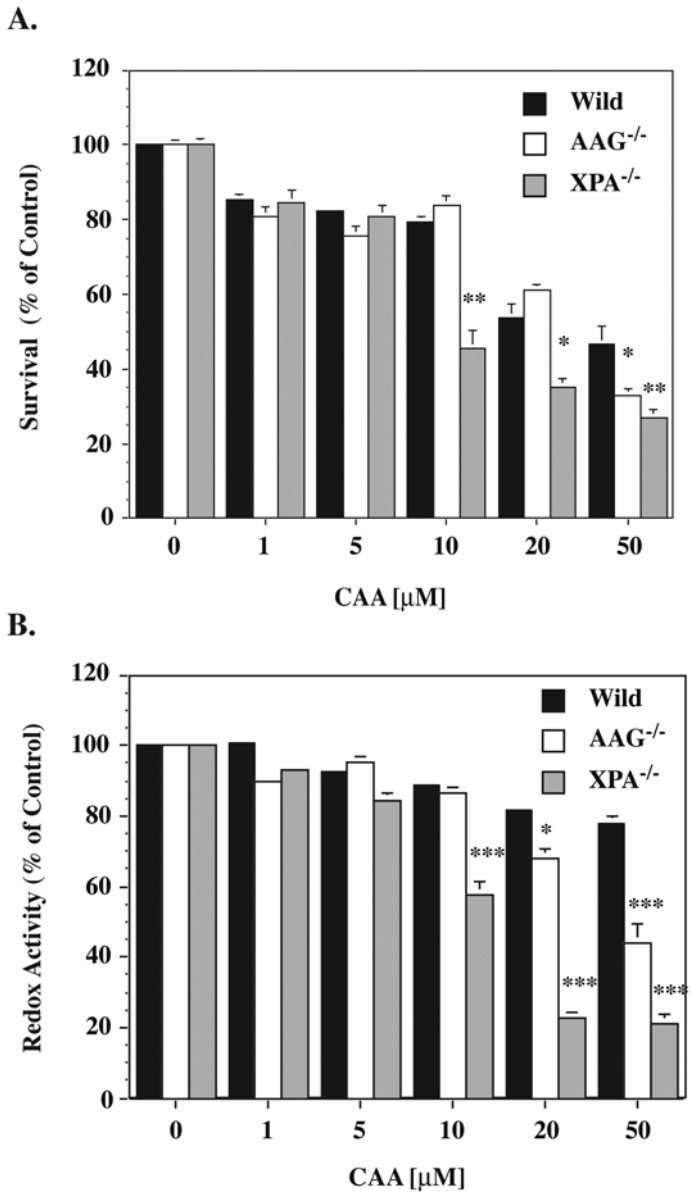
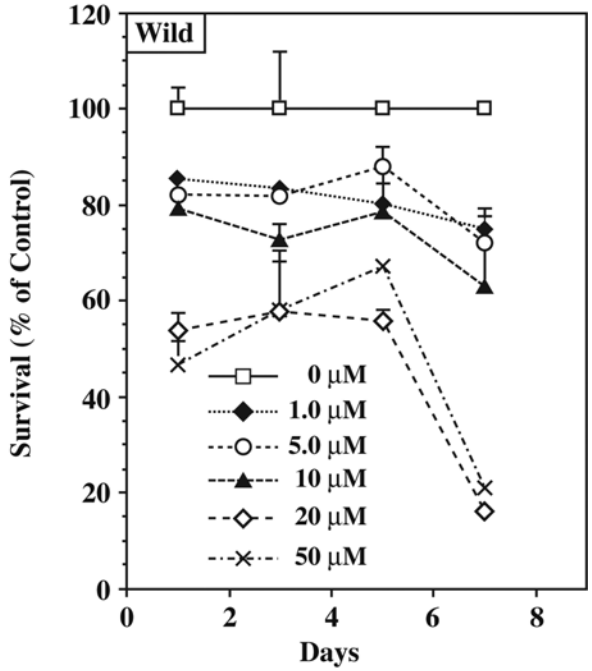
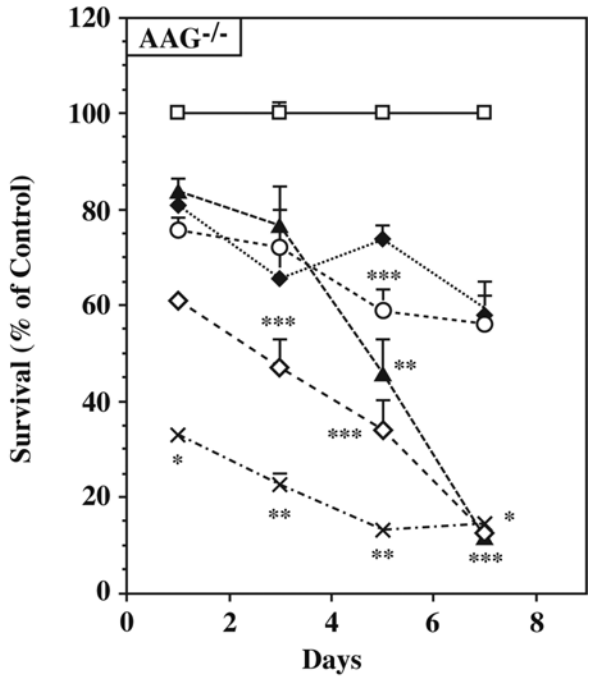
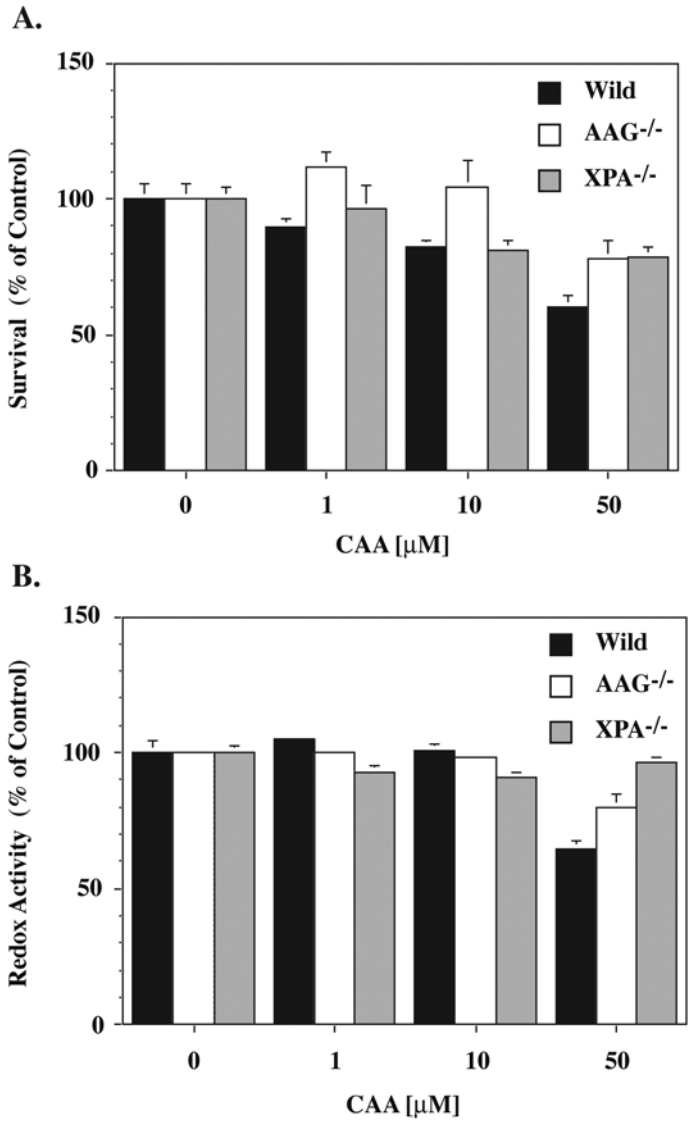


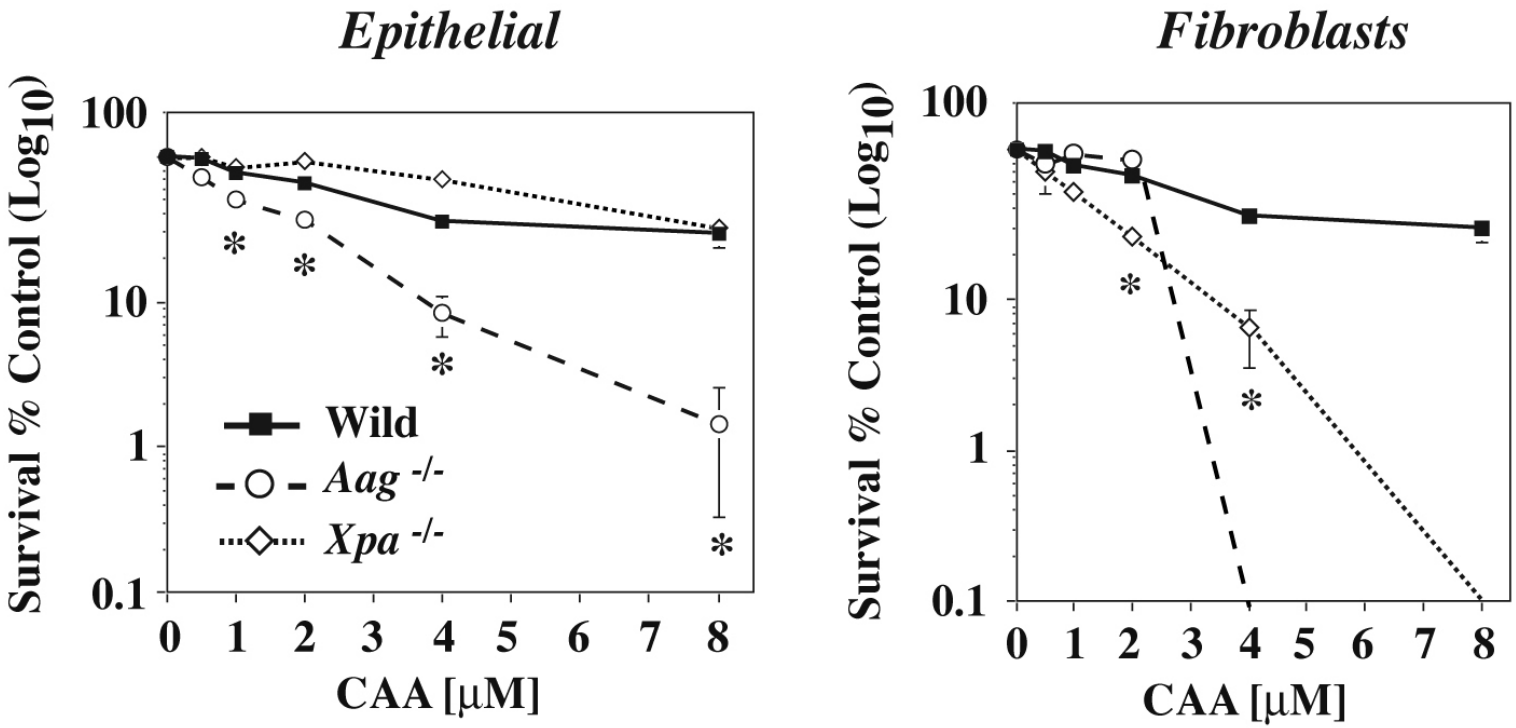
Figure 1. Enzymatic Steps of BER (A) or NER pathways (B).

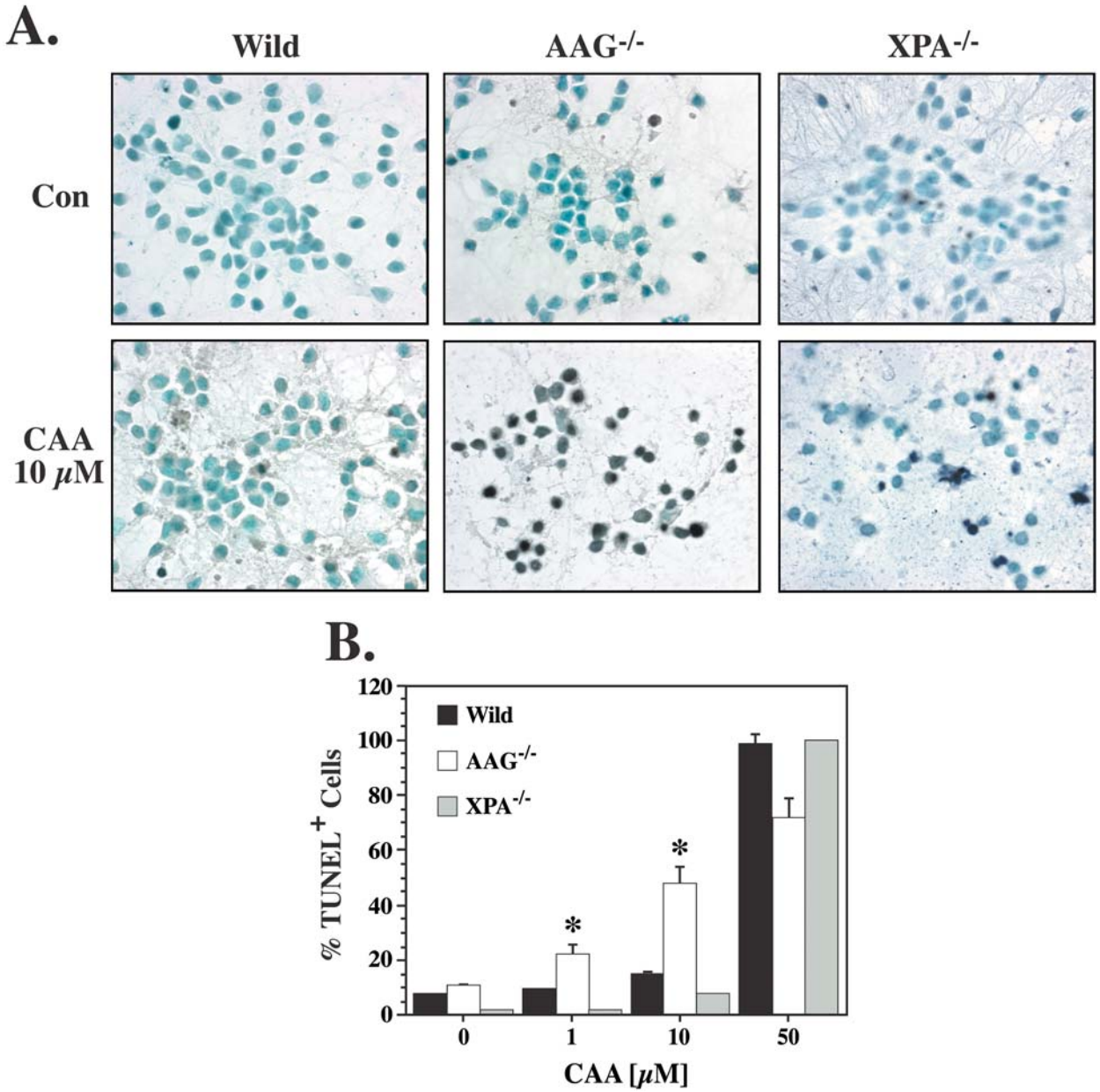

A.
Survival (% of Control)
120
100
80
60
40
20
0
Wild
AAG-/-
XPA-/-
**
*
*
**
0
1
5
10
20
50
CAA [μM]
B.
Redox Activity (% of Control)
120
100
80
60
40
20
0
Wild
AAG-/-
XPA-/-
***
*
***
***
***
0
1
5
10
20
50
CAA [μM]

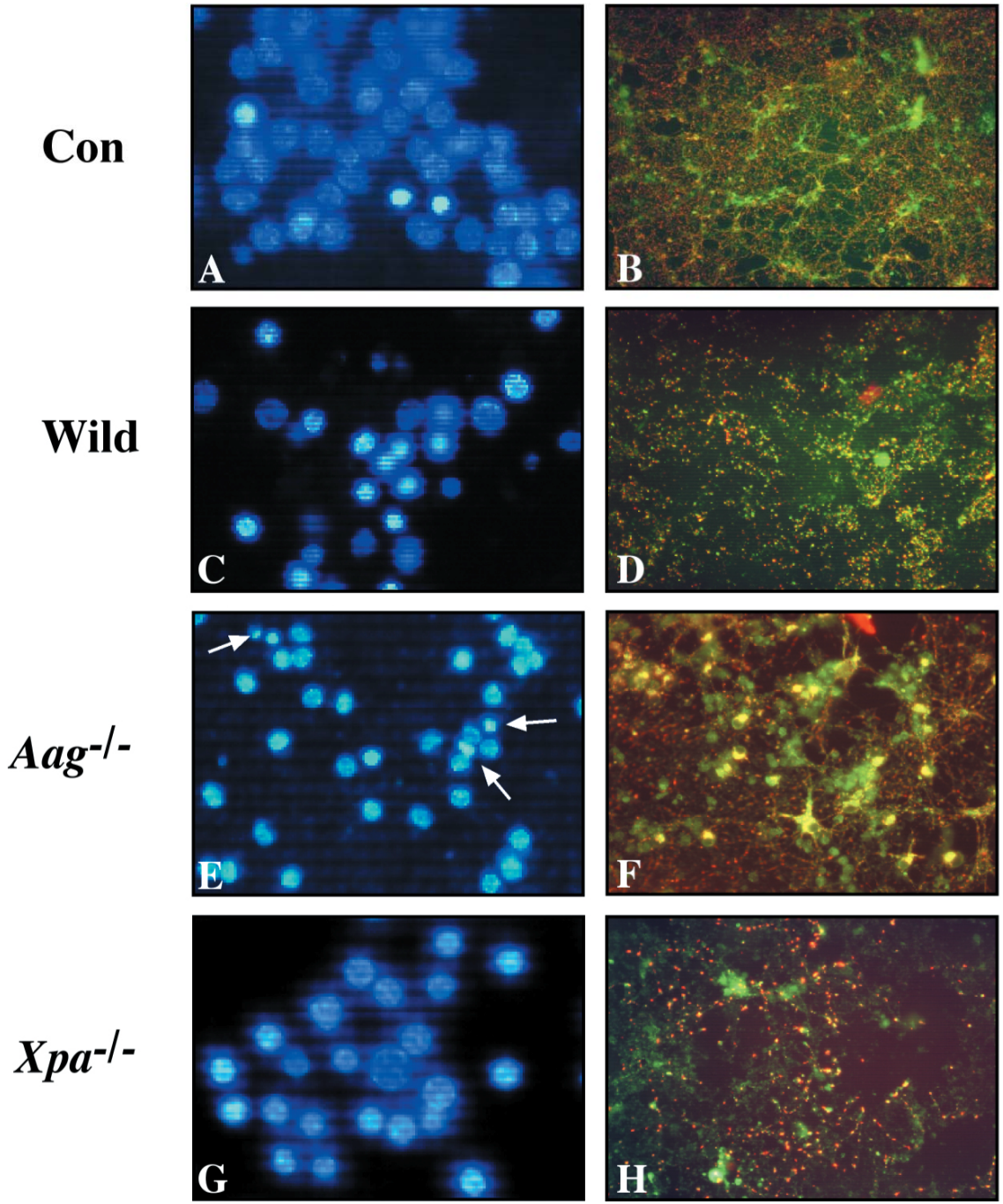

Wild
Survival (% of Control)
Days
0 μM
1.0 μM
5.0 μM
10 μM
20 μM
50 μM
0
20
40
60
80
100
120
0
2
4
6
8

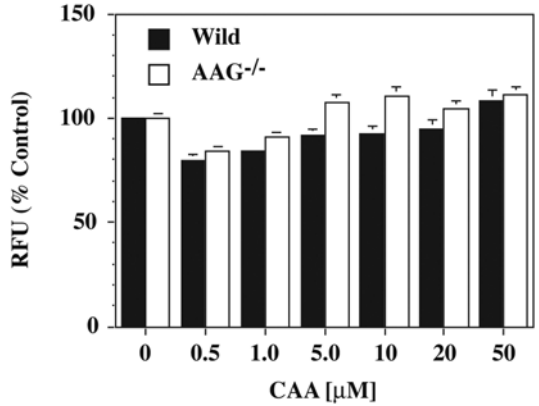

AAG-/-
Survival (% of Control)
Days
0
20
40
60
80
100
120
0
2
4
6
8
*
**
***

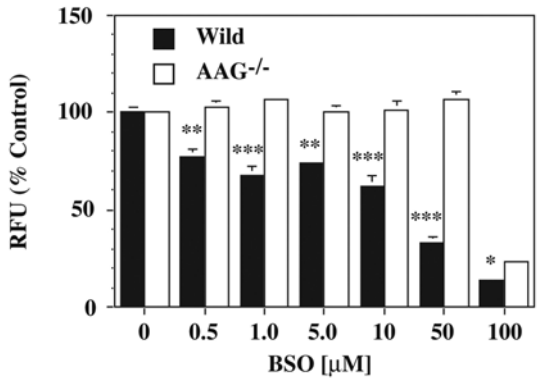

A.
150
100
50
0
Survival (% of Control)
Wild
AAG-/-
XPA-/-
0
1
10
50
CAA [μM]
B.
150
100
50
0
Redox Activity (% of Control)
Wild
AAG-/-
XPA-/-
0
1
10
50
CAA [μM]

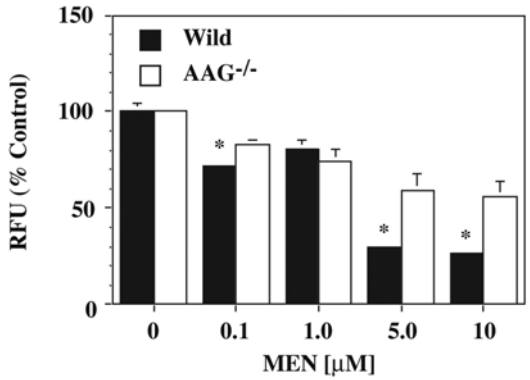

Epithelial
Fibroblasts
Survival % Control (Log10)
100
10
1
0.1
0 1 2 3 4 5 6 7 8
CAA [μM]
Wild
Aag -/-
Xpa -/-
*

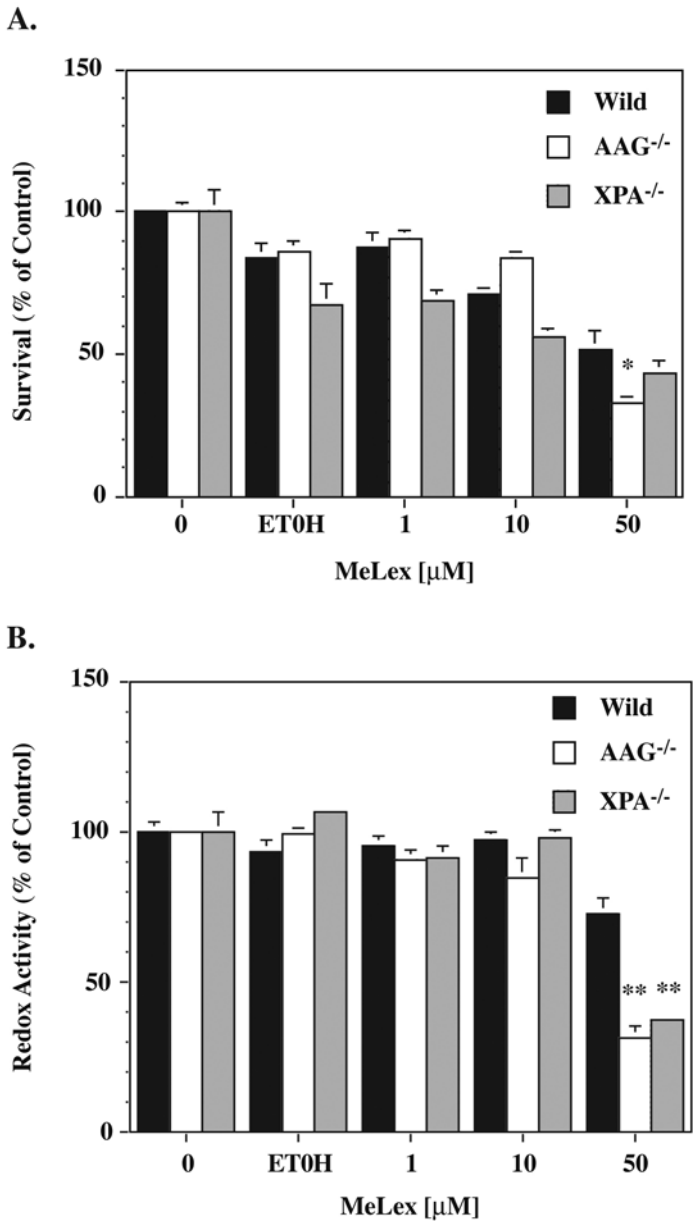

A.
Wild
AAG-/-
XPA-/-
Con
CAA
10 μM
B.
Wild
AAG-/-
XPA-/-
% TUNEL+ Cells
120
100
80
60
40
20
0
0
1
10
50
CAA [μM]
*
*


Con
Wild
Aag-/-
Xpa-/-
A
B
C
D
E
F
G
H


150
100
50
0
RFU (% Control)
Wild
AAG-/-
0
0.5
1.0
5.0
10
20
50
CAA [μM]


150
100
50
0
RFU (% Control)
Wild
AAG-/-
**
***
**
***
***
*
0
0.5
1.0
5.0
10
50
100
BSO [μM]


150
100
50
0
RFU (% Control)
Wild
AAG-/-
*
*
*
0
0.1
1.0
5.0
10
MEN [μM]


A.
150
100
50
0
Survival (% of Control)
Wild
AAG-/-
XPA-/-
*
0
ET0H
1
10
50
MeLex [μM]
B.
150
100
50
0
Redox Activity (% of Control)
Wild
AAG-/-
XPA-/-
** **
0
ET0H
1
10
50
MeLex [μM]

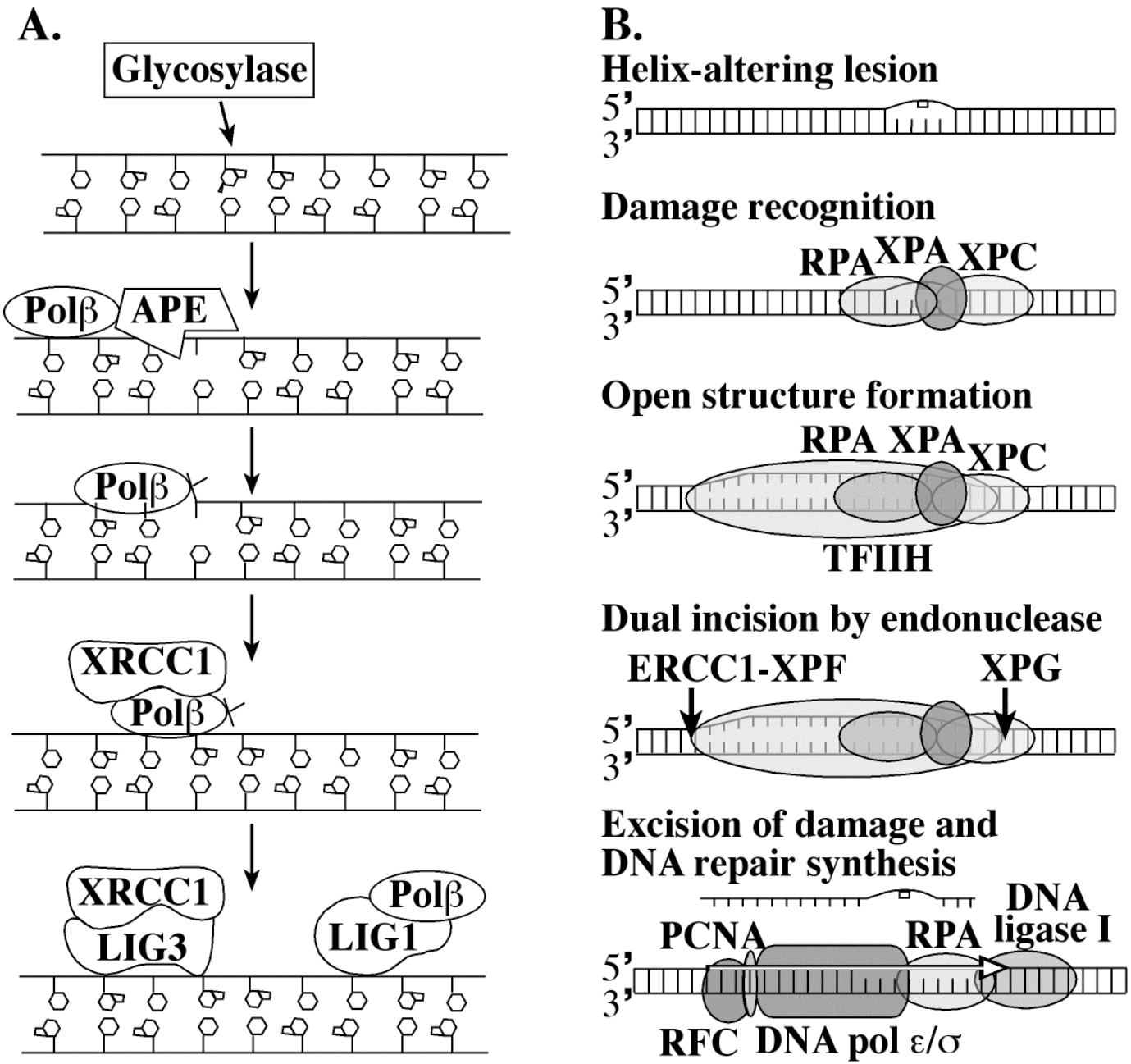
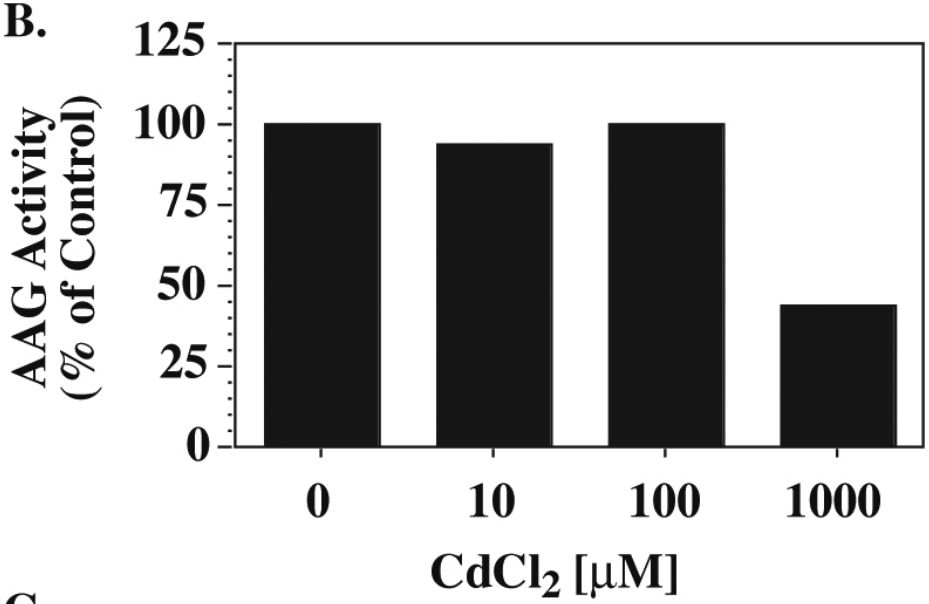


Figure 1. Enzymatic Steps of BER (A) or NER pathways (B).

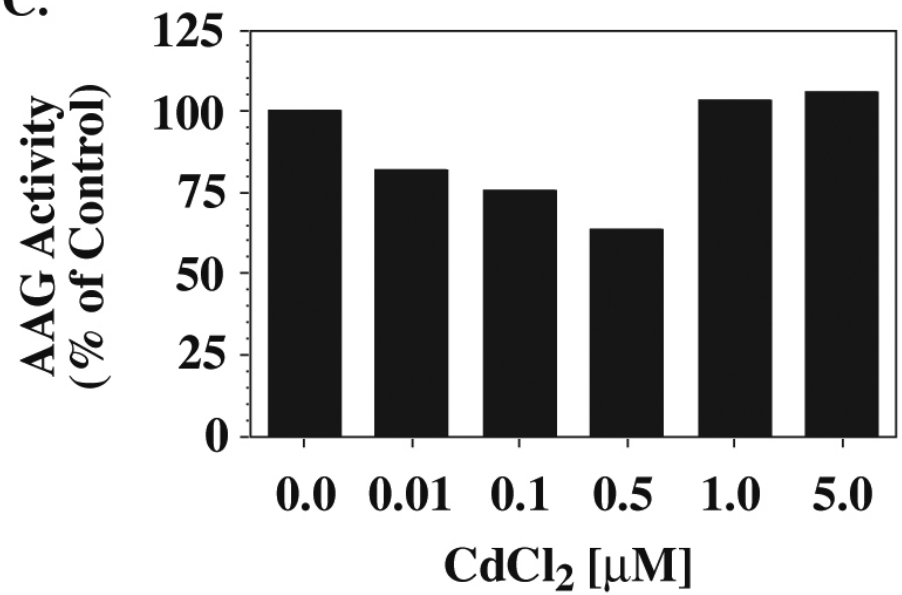
A. 5' CCTGCCCTGAGC A_ε GCTGTGGG 3'

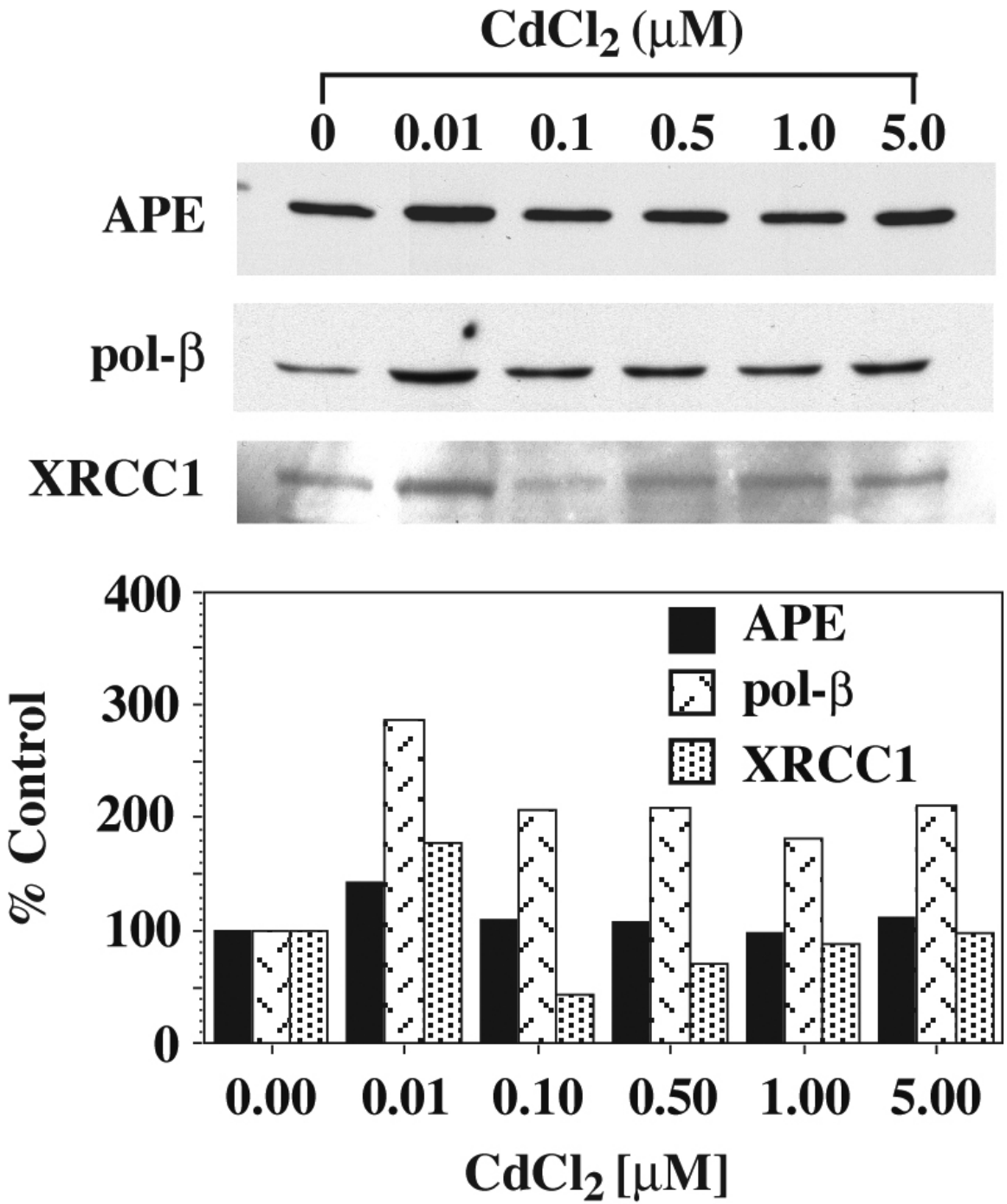
3' GGACGGGACTCG T CGACACCC 5'

B.



C.




CdCl2 (μM)
0
0.01
0.1
0.5
1.0
5.0
APE
pol-β
XRCC1
400
300
200
100
0
% Control
APE
pol-β
XRCC1
0.00
0.01
0.10
0.50
1.00
5.00
CdCl2 [μM]

SUMMARY

1. **Kinetic studies indicate that the uptake of 2-chloroacetaldehyde (CAA) into the liver and brain of rats is rapid (~30 min) following an oral dose. Conversely, clearance of CAA was rapid for the liver (paralleled blood), but substantially delayed in the brain with near steady-state levels observed >4h post dosing.**

2. **Mature neurons that are deficient in base-excision (BER) or nucleotide-excision (NER) repair were more sensitive to the acute and long-term effects of CAA than DNA repair-proficient neurons. However, NER-deficient neurons appeared to be more sensitive to CAA than BER-deficient neurons. The relative insensitivity of astrocytes to CAA (or Me-Lex, MAM) suggests that CAA specifically targets neurons.**

3. **Like neurons, epithelial cells or fibroblasts derived from the kidney or skin of BER- and NER-deficient mice were also similarly sensitive to CAA.**

4. **Low concentrations of metals (e.g., Cd) influence neuronal BER, which may significantly enhance CAA-induced neurotoxicity.**

5. **These studies demonstrate that BER and NER play a vital role in protecting neurons from CAA-induced toxicity by removing DNA lesions (i.e., ethenobase). The inability of CAA to significantly influence neuronal GSH metabolism or the viability of astrocytes is strong evidence in support of this hypothesis.**

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