Supplementary Material

To: Development of APE1 enzymatic DNA repair assays: Low APE1 activity is associated with increase lung cancer risk

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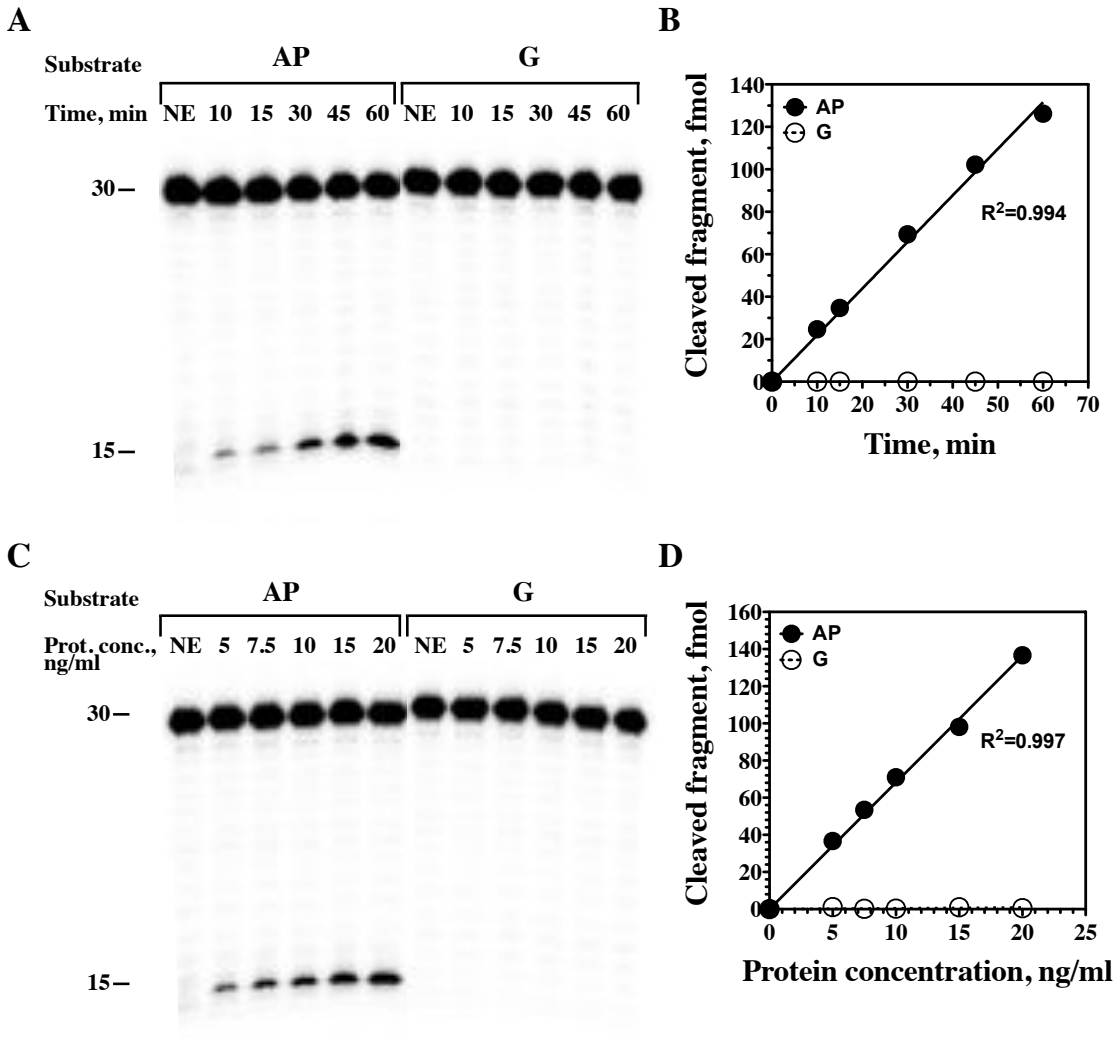
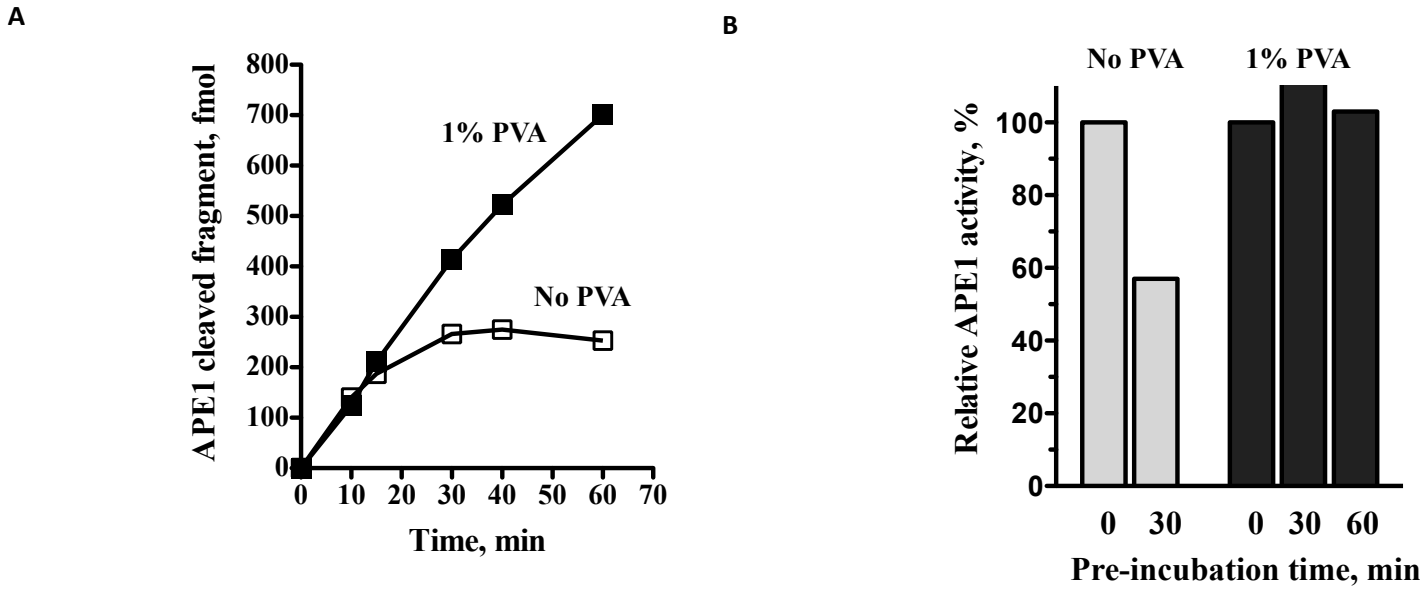


Supplementary Fig. 1. Gel electrophoresis images showing fractionation of the products of radioactivity-based APE1 enzyme activity reactions. (**A**) Gel image of the kinetics of APE1 activity under optimized conditions. **(B)** Quantification of the gel presented in A was done by phosphorimaging (**C**) Protein titration of APE1 activity under optimal reaction conditions with a protein extract prepared from PBMC. **(D)** Quantification of the gel presented in C was done by phosphorimaging. In both panels (**A**) and (**C**) the substrate containing the abasic site (AP) was cleaved yielding the 15-mer, whereas the control DNA "G" without the damage was not cleaved. Closed circles, substrates with the abasic site (AP); Open circles, control DNAs without the lesion.



Supplementary Fig. 2. Effect of the stabilizing additive polyvinyl alcohol (PVA) on APE1 reaction. **(A)** Time course of the APE1 reaction in the presence (closed squares) or absence (open square) of 1% PVA. **(B)** Effect of pre-incubation of a diluted protein extract prior to its addition to the reaction mixture in the presence (black) or absence (gray) of 1% PVA. APE1 enzyme activity is presented relative to the activity without pre-incubation of the extract (set as 100%).

Supplementary Table 1. Analysis of the relationships between APE1 Asp148Glu, APE1 activity (fluorescence-based) and lung cancer risk

A. Association of APE1 Asp148Glu with lung cancer, and with APE1 activity (fluorescence -based)

Allele		Controls	Cases	Overall	χ^2 for association with lung cancer*	F-test for association with APE1 activity§
All	n	99	99	198 (100%)		
	Mean APE1 (SE)	896 (24)	787 (18)	842 (15)		
T/T	n	42	34	76 (38.4%)		
	Mean APE1 (SE)	843 (31)	763 (35)	807 (24)		
T/G	n	46	50	96 (48.5%)	0.51 (2df)	2.62 (2, 97df)
	Mean APE1 (SE)	947 (40)	810 (25)	876 (24)	P=0.77	P=0.08
G/G	n	11	15	26 (13.1%)		
	Mean APE1 (SE)	887 (57)	770 (33)	819 (32)		
Trend					0.51 (1df) P=0.48	0.82 (1, 98df) P=0.37

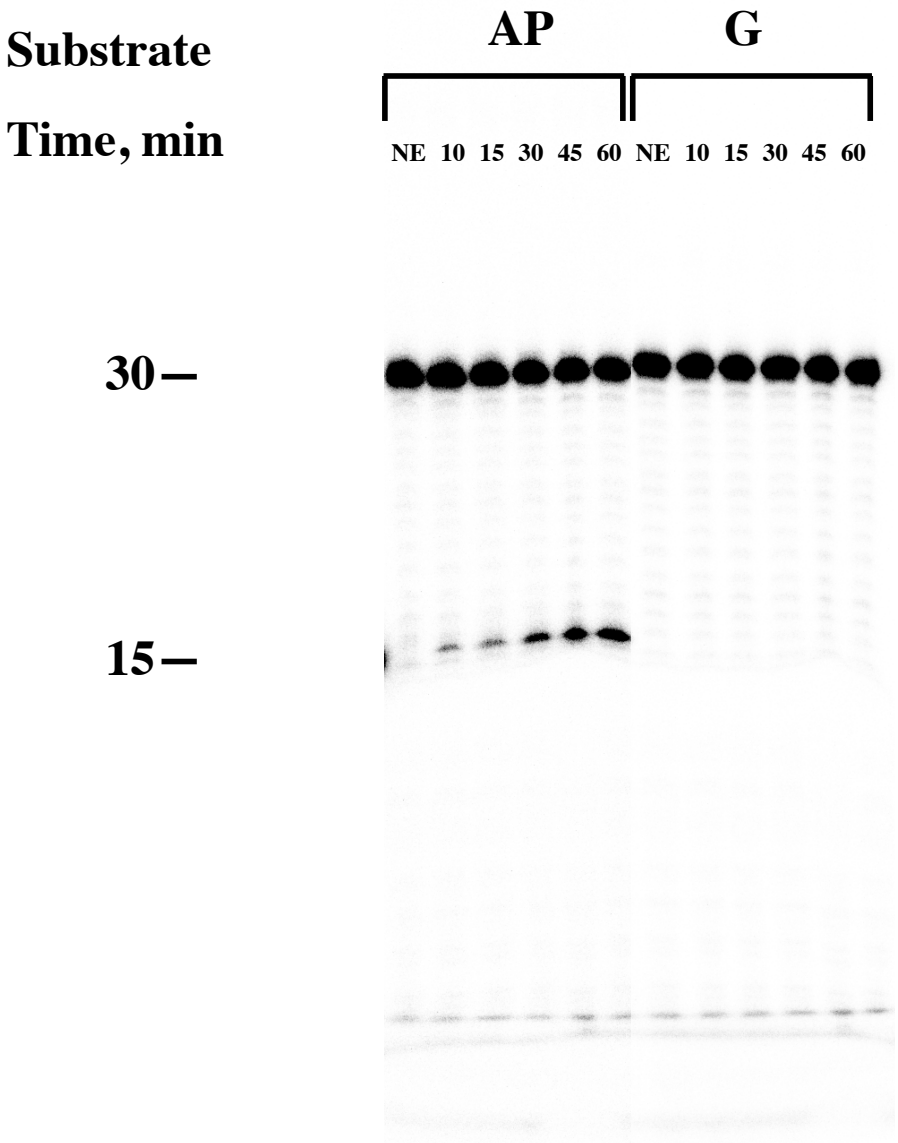
B. Logistic regression analysis of APE1 Asp148Glu and APE1 activity (fluorescence-based) in lung cancer

Model	OR¶ APE1 (95%CI)	OR SNP1 (95%CI)	OR SNP2 (95%CI)	OR SNP trend (95%CI)
Smoking + APE1	1.34 (1.11, 1.61) P=0.002	-	-	-
Smoking + APE1 + SNP1 (wt v htrz) + SNP2 (wt v homz)	1.38 (1.14, 1.69) P=0.001	1.74 (0.83, 3.65) P=0.32	1.54 (0.52, 4.55) P=0.32	-
Smoking + APE1 + SNP trend	1.36 (1.12, 1.65) P=0.002	-	-	1.36 (0.82, 2.27) P=0.24

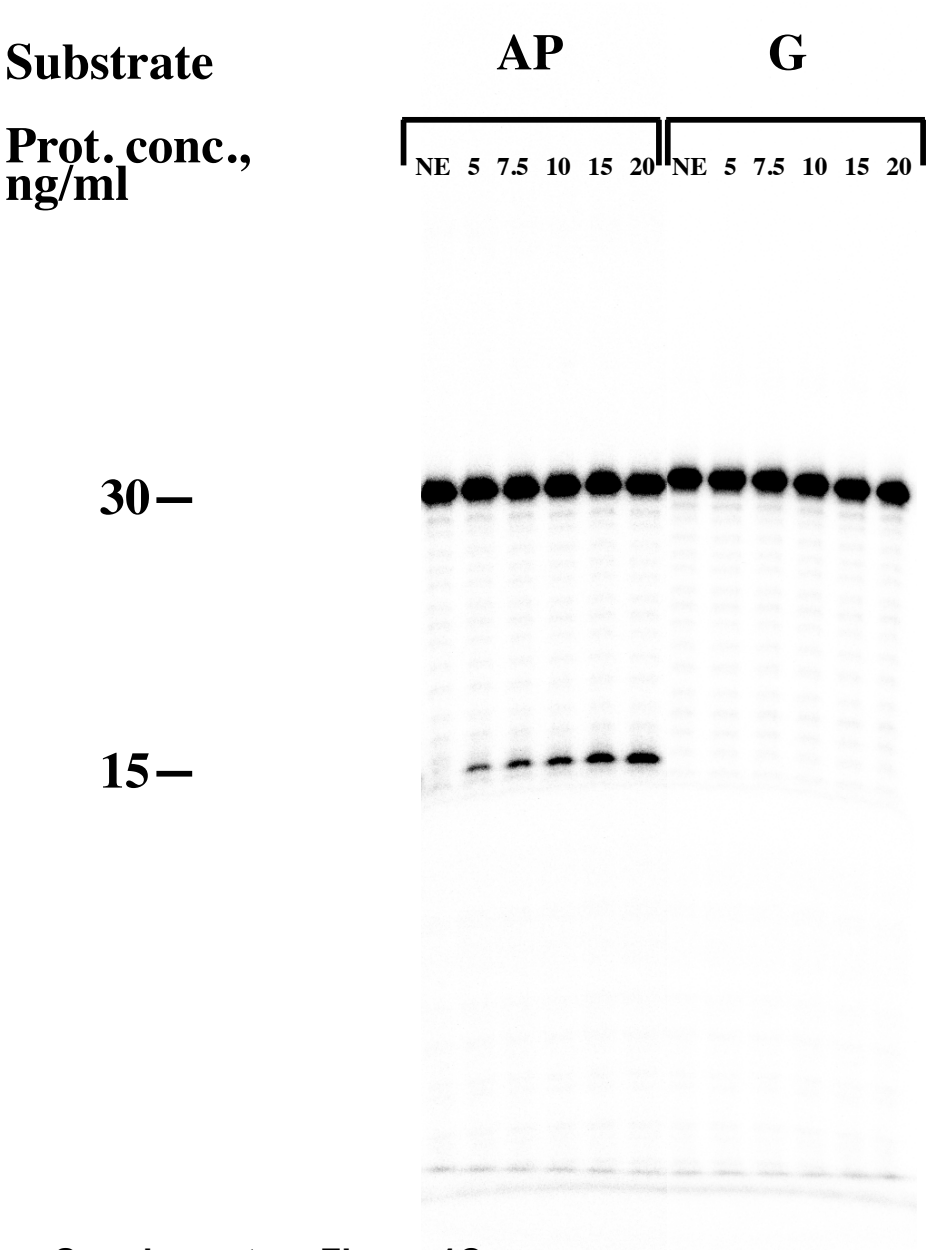
* Test for association between SNP and lung cancer using conditional logistic regression adjusted for smoking: 2 degree of freedom test and test for trend

§ Test for association between SNP and APE1 activity (fluorescence-based assay) using multiple linear regression, controlling for smoking and matched pairs: 2 degree of freedom test and test for trend

¶ Odds ratios are expressed per 100 APE1 units



Full gel image of Supplementary Figure 1A



Full gel image of Supplementary Figure 1C