Figure S1: The assay for ApeXPF activity was carried out in 20  $\mu$ l reactions which contained: 30 mM HEPES (pH 7.6), 5 % glycerol, 40 mM KCl, 1 mM EDTA, 0.1 mg/ml BSA, 0.1 mg/ml calf thymus DNA and 80nM of 3' flap substrate. The substrate was incubated with 1  $\mu$ M PCNA at room temperature for 5 min. After adding 7  $\mu$ M protein the relevant samples were incubated for 2 minutes at 55 °C and each reaction was initiated by adding MgCl<sub>2</sub> in a final concentration of 10 mM and further incubated at 55 °C for 1 minute, 3 minute and 10 minute time points respectively. The assay point without PCNA was left for 10 minutes. The control experiment using SsoXPF and PCNA was incubated for 10 seconds. Aliquots (5  $\mu$ l) were taken at selected time points and added to 90  $\mu$ l chilled stop solution (10 mM Tris, pH8; 5 mM EDTA) to terminate the reaction. The DNA was ethanol precipitated and resuspended in formamide loading buffer. Samples were heated at 95 °C for 5 minutes.

