

Supplementary Figure 5. Stopped-flow machine used for time resolved potassium permanganate footprinting studies. The instrument consists of four Hamilton syringes driven by three independent stepping motors. The stepping motors are controlled by computer based electronics that generate the necessary pulses with the appropriate timing. The outlets of the syringes are connected to a mixing device which is enclosed in a circulating water bath providing the temperature control. The mixing device of the machine comprises a system of reagent loops, mixing chambers and reaction loops which is set up according to the desired kinetic experiments. The first mixing chamber is a cross mixer designed to mix two reagents, RNAP and DNA promoter fragment, while the second mixing chamber is a T mixer designed to mix the reacted solution with the KMnO4 solution. The end of reaction loop 2 is connected to a short piece of capillary tubing (exit tube) that feeds into an Eppendorf tube containing the quenching reagent. The design of the stopped flow machine and the sample collector at the end of the machine allow the

sequential mixing of up to 11 samples.