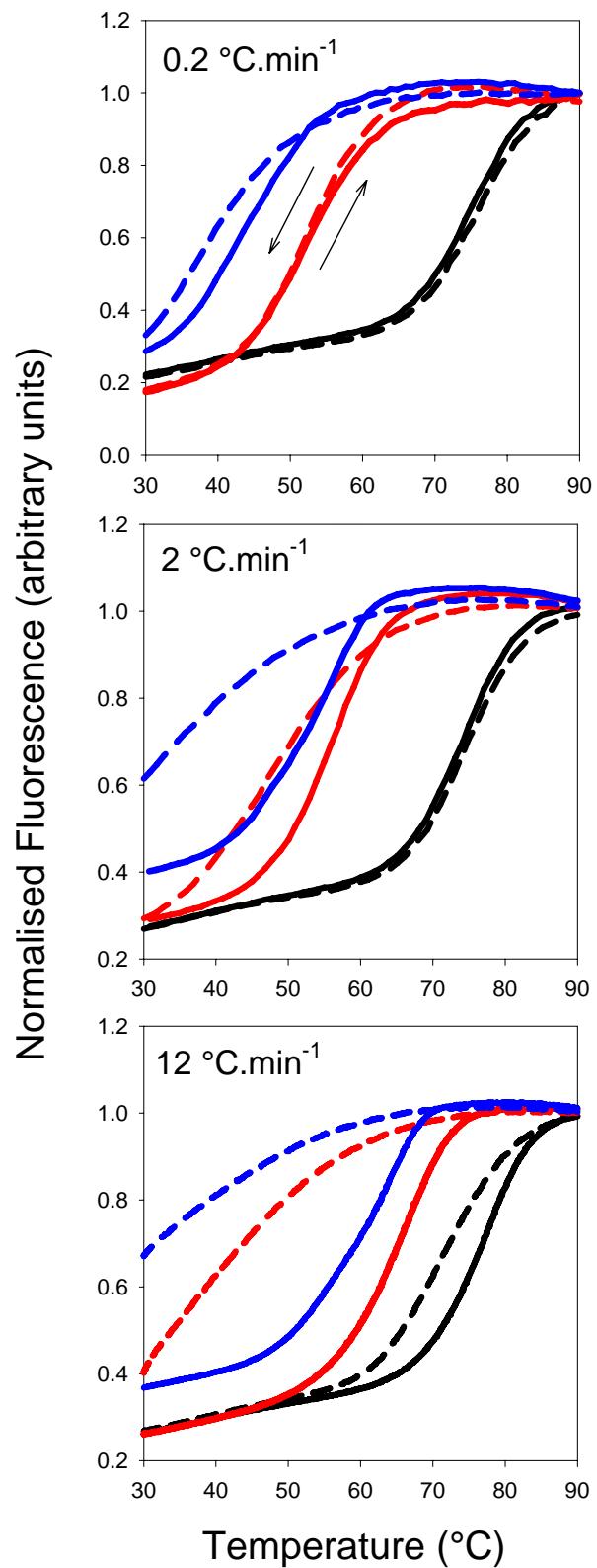
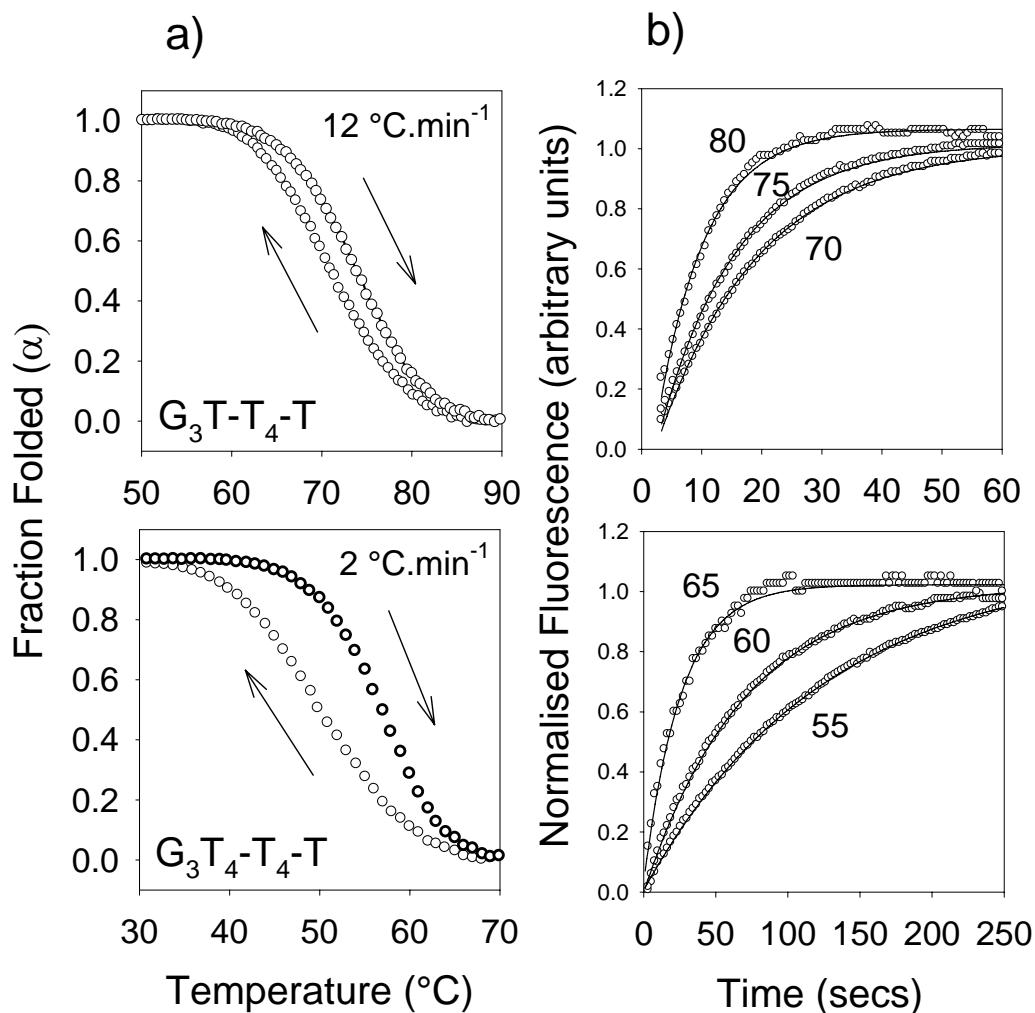


Supplementary Figure 1. Effect of oligonucleotide concentration on the melting temperatures (T_m) of the quadruplex-forming oligonucleotides, determined in 10 mM Lithium phosphate pH 7.4 containing 20 mM KCl or 200 mM NaCl. Samples were heated at $0.2 \text{ } ^\circ\text{C}.\text{min}^{-1}$.



Supplementary Figure 2. Fluorescence melting (solid lines) and annealing (dashed lines) profiles for $\text{G}_3\text{T-T}_4\text{-T}$ (black), $\text{G}_3\text{T}_4\text{-T-T}_4$ (red) and G_3T_4 (blue) determined at different rates of heating and cooling in the presence of 10 mM lithium phosphate containing 10 mM KCl.



Supplementary Figure 3. a) Hysteresis between the melting and annealing profiles for $\text{G}_3\text{T-T}_4\text{-T}$ (upper panel, with a temperature change of $12 \text{ }^{\circ}\text{C}.\text{min}^{-1}$) and $\text{G}_3\text{T}_4\text{-T}_4\text{-T}$ (lower panel, with a temperature change of $2 \text{ }^{\circ}\text{C}.\text{min}^{-1}$ in the presence of 10 mM lithium phosphate pH 7.4 containing 20 mM KCl. b) temperature-jump relaxation profiles for $\text{G}_3\text{T}_4\text{-T-T}$ (upper panel) and $\text{G}_3\text{T}_4\text{-T-T}_4$ (lower panel. The traces show the rate of approach to a new equilibrium following a rapid $5 \text{ }^{\circ}\text{C}$ increase in temperature to the value shown. The profiles have been normalised to show the fractional change in fluorescence with time.

[NaCl] mM	T_m °C					
	G ₃ T	G ₃ T-T ₄ -T	G ₃ T ₄ -T-T	G ₃ T ₄ -T-T ₄	G ₃ T ₄ -T ₄ -T	G ₃ T ₄
10	55.6					
50	63.4	43.4	39.4			
100	68.0	49.5	46.3			
200	73.3	58.6	54.9	43.4	46.7	41.5
500	78.4	63.7	61.1	50.6	54.1	51.9
1000				58.8	62.2	61.6

Supplementary Table 1. T_m and ΔH values for the fluorescently labelled quadruplex-forming oligonucleotides, determined in the presence of 10 mM lithium phosphate containing different concentrations of NaCl. The samples were heated and cooled at a rate of 0.2 °C·min⁻¹.

	20 mM KCl						200 mM NaCl	
	0.2 °C.min ⁻¹		2 °C.min ⁻¹		12 °C.min ⁻¹		12 °C.min ⁻¹	
Sequence	melt	anneal	melt	anneal	melt	anneal	melt	anneal
G ₃ T	> 95	> 95	-	-	-	-	75.4	75.7
G ₃ T-T ₄ -T	75.2	75.0	74.1	73.8	78.3	70.9	58.6	58.5
G ₃ T ₄ -T-T	72.9	73.2	72.1	71.7	76.0	67.3	55.0	54.8
G ₃ T ₄ -T-T ₄	52.7	52.3	56.2	46.6	66.6	< 30	43.7	43.1
G ₃ T ₄ -T ₄ -T	53.9	54.2	56.8	49.2	66.1	< 30	46.5	46.9
G ₃ G ₃ T ₄	44.5*	37.6	46.7	< 30			41.8	41.3

Supplementary Table 2. T_m values showing the hysteresis between the fluorescence melting and annealing profiles at different rates of temperature change. Samples were prepared in 10 mM lithium phosphate pH 7.4 containing either 20 mM potassium chloride or 200mM sodium chloride. * indicates a biphasic melting profile.