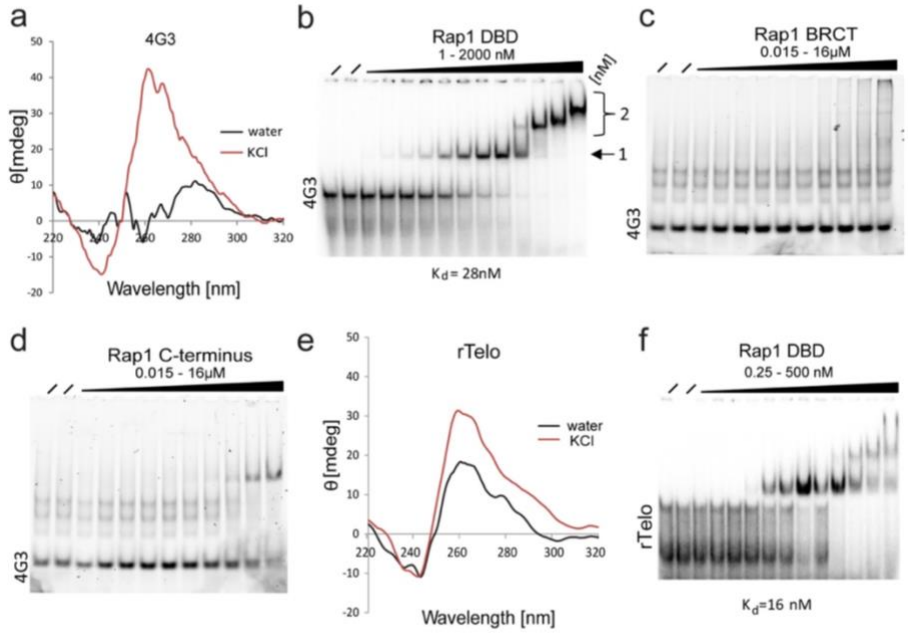
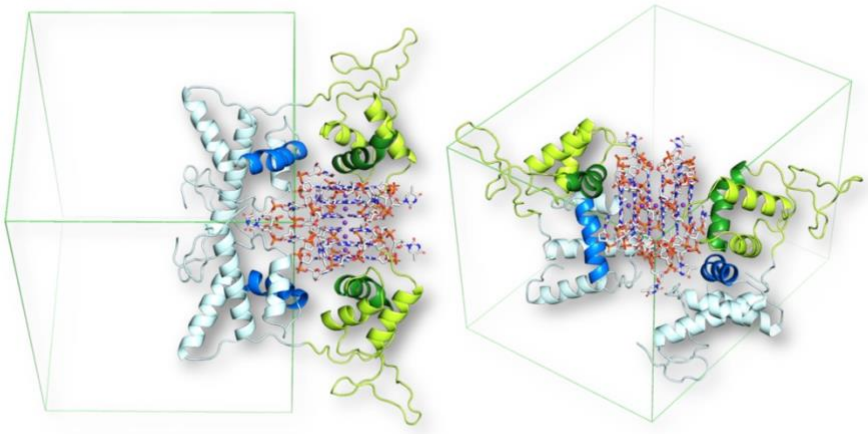
Supplementary Figure 1



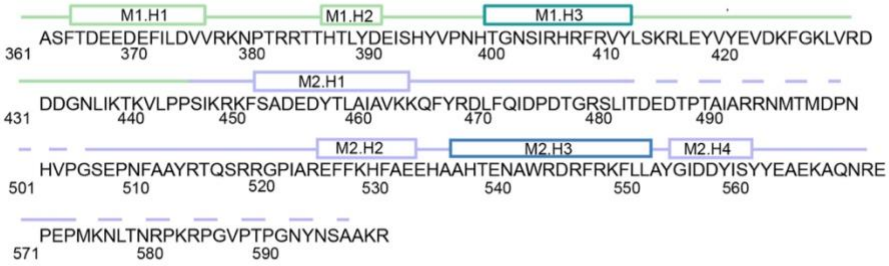
Supp. Figure 1. Binding activity of Rap1-DBD for different G-quadruplexes. a) CD spectrum of the 4G3 oligonucleotide folded in KCl. The spectrum confirms it forms a parallel G-quadruplex. **b)** EMSA of the 4G3 G-quadruplex incubated with increasing concentrations of Rap1-DBD. **c)** and **d)** EMSA of the 4G3 G-quadruplex incubated with increasing concentrations of the Rap1 BRCT and RAP1 C-Terminal RCT domain respectively. **e)** and **f)** CD spectrum of the rTelo oligonucleotide and EMSA of the parallel rG-quadruplex incubated with increasing concentrations of Rap1-DBD. The apparent Kd= 16nM.

Supplementary Figure 2



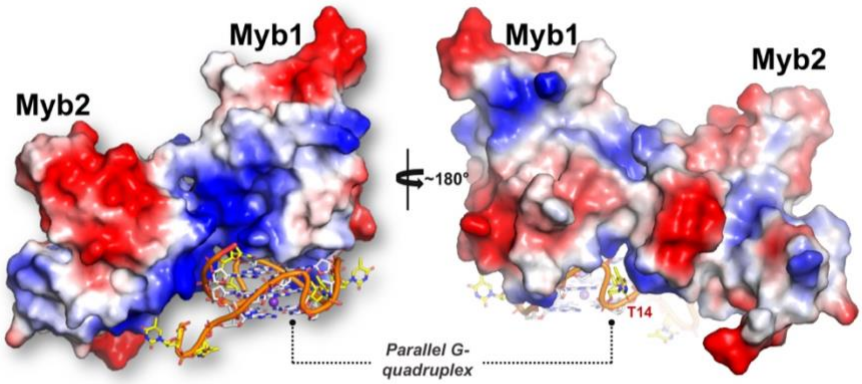
Supp. Figure 2. Crystal packing. Two views of the stacking of two G-quadruplex-Rap1-DBD complexes in the crystal forming a dimer. The box represents the unit cell.

Supplementary Figure 3



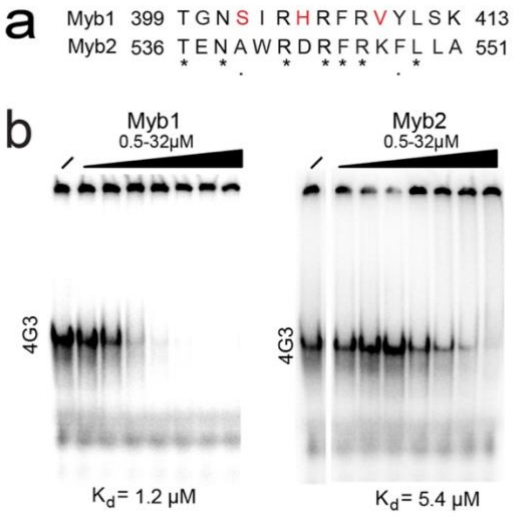
Supp. Figure 3. Amino acid sequence and structure of the S. cerevisiae Rap1-DBD. The structural domains of the protein are shown above the Rap1-DBD amino acid sequence and colour-coded as in the crystal structure: Myb1 domain in green; Myb2 domain in blue. Open rectangles indicate the positions of **α**-helices, and broken lines mark regions of structural disorder.

Supplementary Figure 4



Supp. Figure 4. Electrostatic surface representation of the Rap1-DBD structure. The two views are related by a 180° rotation.

Supplementary Figure 5



Supp. Figure 5. Binding analysis of the Myb1 and Myb2 DNA-binding helix peptides to G-quadruplex DNA. a) Sequence alignment of the two peptides representing the DNA-recognition α-helix of Myb1 and Myb2 domains. Identical residues are pinpointed by an asterisk and similar ones by a dot. Hydrophobic residues used in G-tetrad recognition by Myb1 are highlighted in red. B) EMSA comparing the binding affinity of the peptide from Myb1 spanning the DNA-recognition α-helix of Myb1 domain with that of the Myb2 domain for the 4G3 G-quadruplex. The [$_{32}$P] labelled DNA at 100pM and incubated with increasing protein concentrations (from 0.5 to 32μM).

Supplementary Table 1:

Data collection	
X-ray source	**X06DA (SLS)**
Wavelength (Å)	1.072
Space group	C121
Unit-cell dimensions	
a, b, c (Å)	84.68, 70.20, 64.85
α, β, γ (°)	90.0, 97.2, 90.0
Resolution range[a] (Å)	35.09 – 2.40 (2.53 – 2.40)
R_{merge}[a]	0.055 (0.406)
$\langle I \rangle / \langle \sigma(I) \rangle$ [a]	8.7 (2.0)
$CC_{1/2}$[a]	0.99 (0.91)
Rpim	0.04 (0.312)
Unique reflections	14498 (2078)
Completeness (%)	99.1 (99.5)
Structure Refinement	
Refinement (R_{work}/R_{free})	0.233 / 0.203
r.m.s.d distances (Å$_2$)	0.01
r.m.s.d bond angles (°)	0.89
Estimated coordinate error (Å)	0.34
Ramachandran plot	
Favoured regions (%)	94.12
Allowed regions (%)	5.88
Disallowed regions (%)	0.00

[a] Values in parentheses are for the highest-resolution shell.