

Measuring telomere length with qPCR

Primers for telomeres

Telo1fwd: CGGTTTGTGGTTGGGTTTGGGTTTGGGTTTGGGTTTGGGTT

Telo1rev: GGCTTGCCTTACCCTTACCCTTACCCTTACCCTTACCCT

see Callicott et al. 2006 and O'Callaghan et al. 2008

Primers for 36B4 (human single copy gene)

36B4 fwd: CAGCAAGTGGGAAGGTGTAATCC

36B4 rev: CCCATTCTATCATCAACGGGTACAA

see Cawthon et al. 2002

Prepare DNA standard:

1. 20 ng/μl
2. 10 ng/μl
3. 5 ng/μl
4. 2.5 ng/μl
5. 1.25 ng/μl

Use 5 ng DNA

2 μl DNA (2.5 ng/μl)

1 μl Primer-Mix (5 μM each fwd and rev primer)

2 μl nuclease-free H₂O

5 μl SYBR Green I Master (Roche)

RTq PCR Program:

95°C – 10 min

95°C – 15 s

60°C – 1 min | 40 cycles

Determine relative quantities for telo and 36B4 with standard curves and calculate the telo/36B4 ratio for each sample.