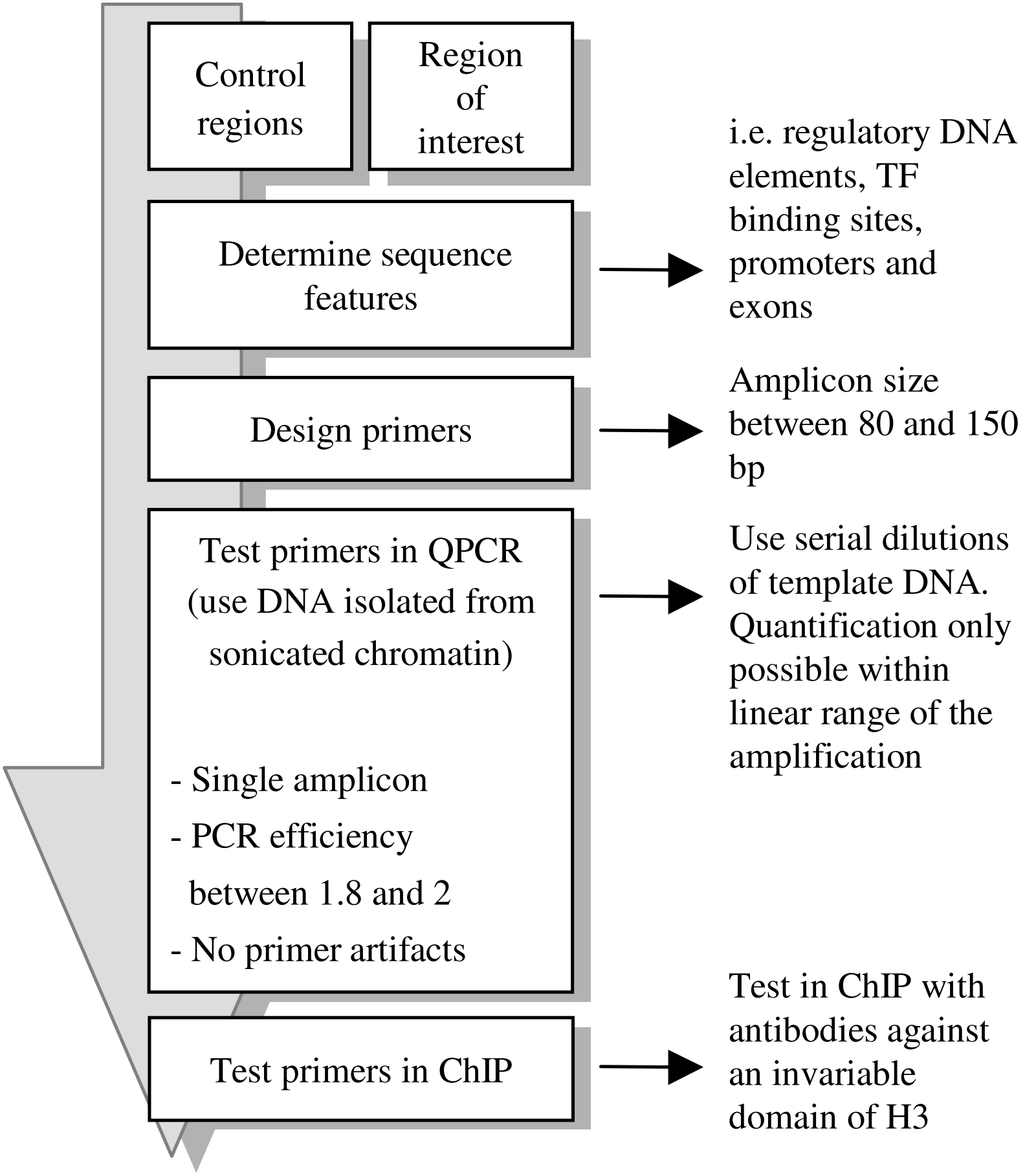
Control regions
Region of interest
Determine sequence features
i.e. regulatory DNA elements, TF binding sites, promoters and exons
Design primers
Amplicon size between 80 and 150 bp
Test primers in QPCR (use DNA isolated from sonicated chromatin)
- Single amplicon
- PCR efficiency between 1.8 and 2
- No primer artifacts
Use serial dilutions of template DNA. Quantification only possible within linear range of the amplification
Test primers in ChIP
Test in ChIP with antibodies against an invariable domain of H3