Eμmyc PCR

Note, this protocol is very prone for contamination. So it is important for a water control and exercise caution when you see a faint band.

Use simple method for DNA extraction to minimize contamination

Material:

- 10x PCR buffer
- dNTP mix, 10mM each nucleotide
- Taq Polymerase [1U/ μl?]
- Eμmyc primers:
  - pUC-1 (22mer, 5'-CAG CTG GCG TAA TAG CGA AGA G-3')
  - pUC-2 (23mer, 5'-CTG TGA CTG GTG AGT ACT CAA CC-3')
- genomic DNA [100 - 500ng/ μl]
- 1kb DNA ladder

Method:

1. Prepare a mastermix (without DNA) with a volume of 20μl/ sample:
   2μl 10x buffer
   15.6μl dH_2O
   0.4μl dNTP
   0.4μl pUC-1
   0.4μl pUC-2
   0.2μl Taq polymerase
   add 1μl DNA

2. PCR conditions:
   94°C/ 5min
   [94°C/ 1min, 64°C/ 1min, 72°C/ 1.5min] x 32
   72°C/ 5min, 4°C/ hold

The primers detect the Eμmyc transgene and generate a **830**bp product. Run on a 1% agarose gel to visualize the product.