PCR Purification

Aim

To purify PCR products.

Procedure

Before use, ethanol must be added to the PE buffer (see instructions on the bottle) and 1:250 pH indicator must be added to the PB buffer. The yellow color of the PB buffer mixed with pH indicator indicates a pH $\leq$ 7.5 and is required to obtain an efficient binding of DNA to the membrane.

1. Add 5 volumes of PB buffer to the sample. Transfer the mixture to a clean the spin column and centrifuge for 30 sec, 13 000 rpm.
2. Pour the mixture into the spin column again, centrifuge for 30 sec, 13 000 rpm. Repeat this step 5 times
3. Add 750 μl of PE washing buffer to the spin column. Centrifuge for 30 sec, 13 000 rpm. Discard flow-through.
4. Centrifuge for 30 sec, 13 000 rpm
5. Place the spin column in a clean Eppendorf tube. Elute the DNA by adding 30 μl dH_2O. Let stand for 5 min and centrifuge for 1 min.

Note!

This protocol is originally distributed by QIAGEN and have been modified with the aim to achieve higher yield.

This protocol is for purification of up to 10 μg of PCR products, 100 bp-10 kb in size.

Sources

https://www.qiagen.com/ie/resources/resourcedetail?id=3987caa6-ef28-4abd-927e-d5759d986658&lang=en (retrieved 04.10.2016)