OneStep[™] PCR Inhibitor Removal Kit

Cat. No. D6030 (50 spin columns/purifications)

Protocol



Features:

- For high quality DNA or RNA that is free of enzymatic inhibitors including polyphenolics, humic/fulvic acids, tannins, melanin, etc.
- Fast, one-step procedure for "cleaning" impure samples prior to PCR, sequencing, RT, etc.

Description:

The OneStep[™] PCR Inhibitor Removal Kit contains all the components needed to efficiently remove contaminants from DNA/RNA preparations that can inhibit downstream enzymatic reactions such as PCR and RT. The column matrix has been specifically designed for efficient removal of polyphenolic compounds, humic/fulvic acids, tannins, melanin, etc. from most impure DNA and RNA preparations. Sample cleanup is as simple as: applying, spinning and recovering a sample from the column.



Performance Characteristics of the Zymo-Spin[™] III-HRC Column



Figures A & B (above) depict the performance characteristics of the Zymo-Spin" III-HRC Column. Figure A shows that some loss of DNA can occur with lower (50 µl) input volumes. However at higher input volumes, the recovery approaches 100%. Figure B shows that input volume recovery is complete after 1 minute for the input volumes tested. In all cases, data were plotted as the mean from experiments performed in triplicate.



DNA is efficiently amplified by PCR following humic acid removal with the OneStep[™] PCR Inhibitor Removal Kit. The figure shows amplification of a 200 bp product from DNA containing humic acid that was "treated" with the kit. Alternatively, PCR amplification was completely inhibited in the case of the "non-treated" sample. In each case, equal amounts of DNA were used for each PCR and equivalent amounts of the reaction were then analyzed in a 2.0% (w/v) agarose/TAE/EtBr gel. The ladder is a 100 bp DNA marker (Zymo Research). Hot start PCR was performed using Zymo Taq[™] PreMix (Zymo Research).

References:

Clancy, C.M.R. et al. (2001) *Biochemistry*, 40, 13353-13360. Stevenson, F.J. (1982) *Humus Chemistry*. Wiley-Interscience, New York.

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Column Preparation:

Zymo-Spin[™] III-HRC Columns need to be prepared prior to use:

 Insert column into a Collection Tube.
*Please note that matrix in the column may appear dehydrated, or powdery. This is normal.

 Open the cap, add 600 µl of Prep-Solution and centrifuge at 8,000 x g for 3 minutes.

Inhibitor Removal:

3. Transfer the prepared column to a clean 1.5 ml microcentrifuge tube. Add 50 - 200 μl DNA or RNA (in water, TE, or similar) to the Zymo-Spin™ III-HRC Column and centrifuge at 16,000 x g for 3 minutes.

The filtered DNA (or RNA) is suitable for PCR, (RT), and other downstream applications.

Kit Contents:

	Qty.	Storage Temp.
Zymo-Spin™ III-HRC Columns	50	Room Temp.
Collection Tubes	50	Room Temp.
Prep-Solution	30 ml	Room Temp.
Protocol	1	-

Also Available:







