# A fast procedure for yeast DNA purification

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The treatment of a cell suspension with 3% SDS, to cause cell lysis, has been reported effective as a simple method for the isolation of DNA from recalcitrant gram-positive bacteria (1). We have applied this procedure to isolate yeast DNA, overcoming the troublesome generation of protoplasts included in standard protocols (2).

Media and procedures for yeast cultivation were those described by Sherman *et al.* (3). The protocol used for DNA purification was as follows. *Saccharomyces cerevisiae* IVPX2−1C (a haploid strain derived from the standard wild type S288C) was grown overnight in liquid YPD medium. The cells from 1 ml aliquots of the culture were collected by centrifugation in 1.5 ml microfuge tubes, resuspended in 100 μl of TE buffer (10 mM Tris, 1 mM EDTA, pH 7.5) containing 3% SDS, and maintained at room temperature for 15 minutes with occasional agitation. The disrupted cells were diluted by adding 0.5 ml TE to each tube; this was done to allow phase separation after treatment with phenol in the following step. Deproteinization was carried out by treatment with 1 volume of liquid phenol equilibrated with TE. The nucleic acids in the aqueous phase were treated with ether to eliminate residual phenol, and recovered by isopropanol precipitation. RNA was eliminated by treatment with RNase A (DNase free), 2 μg/μl, for 1 h at 37°C. About 1 μg DNA, of a size higher than 20 kb, was reproducibly obtained from 1 ml of early stationary phase culture ($2\times10^8$ cells). The protocol could easily be scaled up.

Figure 1 shows agarose gel electrophoresis of nucleic acids purified by the described protocol. Lanes 2 and 3 show samples obtained before and after treatment with RNase, respectively. Two discrete bands observable in lane 2, which disappear after treatment with RNase, correspond to 4.2 kb, linear double-stranded RNA (killer RNA), and 18 S rRNA. Lanes 4 and 5 show DNA recovered after treatment with RNase, digested to completion with *Eco*RI and *Hin*dIII, respectively.

Bollet *et al.* (1) have shown that heating a cell suspension in 3% SDS for a few minutes in a micro-wave oven improves yield in the isolation of DNA from bacteria. We have concluded that heating is not advisable in the case of yeast. Incubation for 10−20 minutes at 70°C in a water bath or heating for 1−2 minutes at 900 W in a micro-wave oven increased by several times the amount of RNA recovered without substantial improvement in the yield of DNA.

The procedure for DNA purification has been adapted to recover plasmids from yeast. For this purpose, cell lysis was carried out in 3% SDS, 0.2 N NaOH. The cell suspension was diluted with TE, and the alkali neutralized with sodium acetate (0.3 N, final concentration), added prior to the treatment with phenol. For two different episomal plasmids tested, plasmid preparations obtained by this method or by the common procedure of making protoplasts (4), yielded an equivalent number of transformants.

## ACKNOWLEDGEMENT

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## REFERENCES

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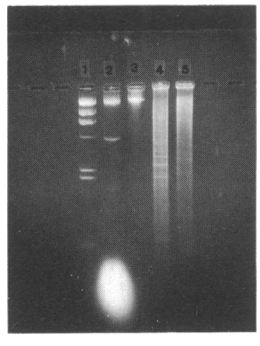

**Figure 1.** Lane 1, Lambda phage DNA digested with *Hin*dIII. Lane 2, yeast nucleic acids obtained by lysis with 3% SDS and deproteinization with phenol. Lane 3, DNA recovered after treatment with RNase. Lanes 4 and 5, samples of DNA digested with *Eco*RI and *Hin*dIII, respectively.