Solutions used in the isolation of mononucleosomal DNA from Haloferax volcanii

MNase Buffer: 10 mM Tris (pH 8)
1 mM $CaCl_2$

MNase add 850 microliters of dH_20 to vial

RNAse A (DNAse free) Dissolve contents of tube in .01 M Na Acetate, pH 5.2
Heat at 100 degrees C for 15 minutes.
Allow to cool slowly to room temperature.
Adjust pH with .1 volumes of 1M Tris Hcl, pH 7.5
Aliquot and freeze.

10% SDS 10% weight: volume Sodium dodecyl sulfate in dH_20

.5 M EDTA

Stop Buffer Mix together equal volumes of 10% SDS and .5 M EDTA

5 M NaCl

Ethidium Bromide 10 mg/ml

(Caution: ethidium bromide is highly mutagenic and must be handled with very carefully. Be sure to wear gloves and change them when you are done handling this reagent.)

Tris / Glycine 5X Stock Buffer 288 g glycine
60 g Tris
dH_20 to 1 liter

Note: this buffer will be diluted to **1/2 X** concentration as needed as follows:

100 mls 5X Tris/Glycine Stock Buffer
1900 mls dH_20

4% Agarose gel Add 4g Nuseive GTG Agarose
per 100 mls 1/2 X Tris / Glycine Running Buffer
(use about 80 mls per gel)

example: for 3 gels, dissolve 9.6 g agarose in 240 mls
of 1/2 X Running Buffer. Microwave 3-4 minutes till clear.
Pour while still quite hot. **Do not add Ethidium Bromide.**

3 M Na Acetate, pH 5.2

7.5 M Ammonium Acetate (when doing this procedure using yeast instead of haloferax)

70% Ethanol

Chloroform / Isoamyl alcohol mixture 96 mls chloroform
4 mls isoamyl alcohol

(need a 24 : 1 ratio)

Note: These are toxic so handle with care in the fume hood and store there.

Phenol / Chloroform / Isoamyl alcohol mixture

Add 25 mls Phenol to 25 mls Chlorform/Isoamyl Alcohol mixture
in the fume hood. Dispose of pipettes in Hazardous waste trash.

Note that phenol is also very toxic and please use this mixture with care.