**IZ Extraction**

*Modified from Jardon et al. Euk. Cell 2008*

Dilute saturated culture back 1:100 and in 10ml (2x5ml) and grow to OD 0.6-0.8.

Spin cells out of media at 4°C 5 min at 1,500 rpm.

Remove media, resuspend in 1ml H2O and move to 1.5ml tube. Spin out of water at 4°C 5 min at 1,500 rpm.

Wash 2x with 1ml 20mM IZ buffer. Pellet with same conditions as above.

Resuspend in 500ul IZ buffer. Add full scoop of glass beads.

Vortex at max speed 1 minute. Incubate on ice 1 minute. Repeat total of 5 times.

Centrifuge homogenate at 4°C 15 minutes at 13,000 rpm.

Pull off supernatant. Use immediately or store at -80°C.

**Reagents**

IZ Buffer

*20mM imidazole*

*HCl to pH 7*

**KCl extraction**

*Modified from Zelle et al. AEM 2008*

Dilute saturated culture back 1:100 and in 10ml (2x5ml) and grow to OD 0.6-0.8.

Spin cells out of media at 4°C 5 min at 1,500 rpm.

Remove media, resuspend in 1ml H2O and move to 1.5ml tube. Spin out of water at 4°C 5 min at 1,500 rpm.

Wash 2x with 1ml 20mM KHPO buffer. Pellet with same conditions as above.

Resuspend in 500ul KHPO buffer. Add a full scoop of beads.

Vortex at max speed 1 minute. Incubate on ice 1 minute. Repeat total of 5 times.

Centrifuge homogenate at 4°C 15 minutes at 13,000 rpm.

Pull off supernatant. Use immediately or store at -80°C.

**Reagents**

KHPO buffer

*100mM potassium phosphate*

*2mM MgCl2*

*1mM dithiothreitol*

*HCl to pH 7.5*