PAR Assay

PAR Stock Solution:

8.5mL 6M Urea 100μL 1M KOH 1ml 0.25mM PAR

To make Urea: RMM = 60.06 gmol^{-1}

 $n = 6M \times 25mL / 1000 = 0.15$ moles

 $m = 0.15 \text{ moles } x \ 60.06 \text{ gmol}^{-1} = 9.009 \text{ g}$

- Weigh 9g of Urea
- Dissolve in 25mL pure (Milli-Q) water
- Transfer to labelled bottle

To make the PAR: RMM = 215.21 gmol^{-1}

 $n = 0.5 \times 10^{-3} \times 25 \text{ mL} / 1000 = 1.25 \times 10^{-5} \text{ moles}$

$$m = 1.25 \times 10^{-5} \times 215.21 = 0.0027g$$

- Make sure that the 0.5mM stock solution is well mixed
- Sonicate if necessary
- Pipette 0.5mL PAR into an Eppendorf tube and make up to 1mL with water. This is now 1mL of 0.25mM PAR.

To make KOH: RMM = 54.11 gmol^{-1}

 $n = 1 \times 5 \text{ mL} / 1000 = 5 \times 10^{-3} \text{ moles}$

$$m = 5x10^{-3} x 54.11 = 0.271g$$

Assay:

- Mix the urea, KOH and PAR and leave for $\frac{1}{2}$ hour.
- Take 450uL of PAR in an Eppendorf and add 50uL of sample
- Leave the samples/assay for 1/2 hour
- Zinc presence is indicated by a pink colour