

SI 09 - Meta-analysis outcome for inorganic and organic osmolyte ratios

Meta-analysis

The raw data from the systematic review was used to calculate the overall effect size, after filtering out influential studies. Standardized mean differences were used to calculate effect size. No variance data was available and could not be re-calculated, hence an unweighted fixed-effects meta-analysis was conducted.

This document shows the output from the meta-analysis for the total inorganic pool vs. total organic pool. Three ratios were analysed: 1) inorganic vs. organic pool 2) inorganic vs. total osmolyte pool 3) organic vs. total osmolyte pool

Analysis, where possible, was run for intracellular data (which accounted for extracellular space (ECS)) and whole tissue data (including ECS).

1) Meta-analysis outcome inorganic vs. organic osmolyte pool

Intracellular values We found no significant salinity effect on the inorganic/organic osmolyte pool ratio for intracellular values.

```
##
## Fixed-Effects Model (k = 3)
##
##   logLik  deviance      AIC      BIC      AICc
## -1.2985    0.1920    4.5970    3.6956    8.5970
##
## I2 (total heterogeneity / total variability):  0.00%
## H2 (total variability / sampling variability):  0.09
##
## Test for Heterogeneity:
## Q(df = 2) = 0.1788, p-val = 0.9145
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
## -0.2542    0.3504   -0.7254    0.4682   -0.9409    0.4325
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Whole tissue data We found no significant salinity effect on the inorganic/organic osmolyte pool ratio for tissues.

```
##
## Fixed-Effects Model (k = 5)
##
##   logLik  deviance      AIC      BIC      AICc
## -6.9306   16.9859   15.8613   15.4707   17.1946
##
```

```
## I^2 (total heterogeneity / total variability): 69.37%
## H^2 (total variability / sampling variability): 3.27
##
## Test for Heterogeneity:
## Q(df = 4) = 13.0610, p-val = 0.0110
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
## 0.2014 0.1500 1.3423 0.1795 -0.0927 0.4954
##
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

2) Meta-analysis outcome inorganic osmolyte pool vs. total osmolyte pool

Intracellular values We found an initial significant salinity effect on the inorganic/total osmolyte pool ratio for intracellular space, which was however not robust. To account for dependent data clusters and adjust for small sample size a robust test was used on the fixed-effects model.

```
##
## Fixed-Effects Model (k = 3)
##
## logLik deviance      AIC      BIC      AICc
## -59.5323 139.8353 121.0646 120.1632 125.0646
##
## I^2 (total heterogeneity / total variability): 98.42%
## H^2 (total variability / sampling variability): 63.49
##
## Test for Heterogeneity:
## Q(df = 2) = 126.9738, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
## -0.1064 0.0074 -14.3551 <.0001 -0.1210 -0.0919 ***
##
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Number of outcomes: 3
## Number of clusters: 2
## Outcomes per cluster: 1-2 (mean: 1.50, median: 1.5)
##
## Model Results:
##
## estimate      se      tval      pval      ci.lb      ci.ub
## -0.1064 0.0862 -1.2352 0.4333 -1.2012 0.9884
##
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Whole tissue data We found an initial significant salinity effect on the inorganic/total osmolyte pool ratio for tissues, which was however not robust. To account for dependent data clusters and adjust for small

sample size a robust test was used on the fixed-effects model.

```
##
## Fixed-Effects Model (k = 5)
##
##      logLik      deviance      AIC      BIC      AICc
## -1146.8748    2331.1345    2295.7496    2295.3590    2297.0829
##
## I2 (total heterogeneity / total variability): 99.82%
## H2 (total variability / sampling variability): 567.18
##
## Test for Heterogeneity:
## Q(df = 4) = 2268.7368, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
##    0.0799    0.0043   18.6595   <.0001    0.0715    0.0883   ***
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Number of outcomes: 5
## Number of clusters: 4
## Outcomes per cluster: 1-2 (mean: 1.25, median: 1)
##
## Model Results:
##
## estimate      se      tval      pval      ci.lb      ci.ub
##    0.0799    0.1260    0.6339    0.5712   -0.3211    0.4809
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

3) Meta-analysis outcome organic vs. total osmolyte pool

Intracellular values We found an initial significant salinity effect on the organic/total osmolyte pool ratio for intracellular space, which was however not robust. To account for dependent data clusters and adjust for small sample size a robust test was used on the fixed-effects model.

```
##
## Fixed-Effects Model (k = 3)
##
##      logLik  deviance      AIC      BIC      AICc
## -60.9061   142.0436   123.8121   122.9107   127.8121
##
## I2 (total heterogeneity / total variability): 98.47%
## H2 (total variability / sampling variability): 65.43
##
## Test for Heterogeneity:
## Q(df = 2) = 130.8533, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
```

```
## 0.1166 0.0081 14.3196 <.0001 0.1006 0.1326 ***
##
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Number of outcomes: 3
## Number of clusters: 2
## Outcomes per cluster: 1-2 (mean: 1.50, median: 1.5)
##
## Model Results:
##
## estimate      se      tval      pval      ci.lb      ci.ub
## 0.1166 0.0946 1.2324 0.4340 -1.0856 1.3188
##
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Whole tissue data We found an initial significant salinity effect on the organic/total osmolyte pool ratio for tissues, which was however not robust. To account for dependent data clusters and adjust for small sample size a robust test was used on the fixed-effects model.

```
##
## Fixed-Effects Model (k = 5)
##
## logLik deviance AIC BIC AICc
## -971.6342 1971.6337 1945.2684 1944.8779 1946.6018
##
## I^2 (total heterogeneity / total variability): 99.80%
## H^2 (total variability / sampling variability): 492.88
##
## Test for Heterogeneity:
## Q(df = 4) = 1971.5381, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
## -0.1216 0.0134 -9.0873 <.0001 -0.1479 -0.0954 ***
##
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Number of outcomes: 5
## Number of clusters: 4
## Outcomes per cluster: 1-2 (mean: 1.25, median: 1)
##
## Model Results:
##
## estimate      se      tval      pval      ci.lb      ci.ub
## -0.1216 0.2333 -0.5212 0.6383 -0.8642 0.6210
##
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Subgroup Analyses

No subgroup analyses were conducted since the number data points per subgroup (from independent clusters) was insufficient.

Testing for influential studies and outliers

1) Inorganic vs. organic osmolyte pool

Intracellular The analysis identified no outliers.

Whole tissue The analysis identified 4 of the 5 results as influential studies. Since this would exclude almost all studies, no studies were removed.

2) Inorganic vs. total osmolyte pool

Intracellular The analysis identified 3 of the 3 results as influential studies. Since this would exclude all studies, no studies were removed.

Whole tissue The analysis identified 5 of the 5 results as influential studies. Since this would exclude all studies, no studies were removed.

3) Organic vs. total osmolyte pool

Intracellular The analysis identified 3 of the 3 results as influential studies. Since this would exclude all studies, no studies were removed.

Whole tissue The analysis identified 5 of the 5 results as influential studies. Since this would exclude all studies, no studies were removed.

Sensitivity analysis

Funnel plots showed no publication bias for the 1) inorganic/organic osmolyte pool ratio. High asymmetry was found for the 2) inorganic/total osmolyte pool ratio and the 3) organic/total osmolyte pool ratio for both tissue and intracellular data.