# 14C Respiration/Production Methods

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### Overview

The simultaneous measurement of bacterial respiration and production will give the best results for estimating growth efficiency. Here, we have constructed custom bacterial respiration/production vials (Fig 1). We will use these vials with 14C-resources. However, first we must optimize the methods.

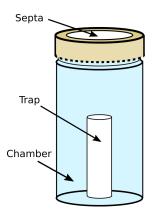


Figure 1: Bacterial Respiration and Production Vial

## **Optimizations**

As we start there are a few things we need to optimize. We are going to start by determining the efficiency of our apparatus. We are doing this using 14C-Bicarbonate. But we need to check a few things first:

- 1. How much Bicarbonate should we add
- 2. What is the efficiency of the trap

### 1) Bicarbonate

I did a simple experiment where I added known volumes of the bicarbonate stock to 1500  $\mu$ L of scintillation cocktail (1, 5, and 10  $\mu$ L). Based on this I discovered that the LSC could only measure 1  $\mu$ L max. The CPM for this voucher was ~2,300,000 cpm.

#### 2) Trap Efficiency

```
# Import Data
trap.data <- read.csv("../notes/20150921_BRBP.csv", header=T)
# Raw Data For Intra Vial Test</pre>
```

```
in1 <- trap.data[trap.data$Sample == "Tube1_in" & trap.data$Time == "1", 3]</pre>
out1 <- trap.data[trap.data$Sample == "Tube1_out" & trap.data$Time == "1", 3]</pre>
# Define Theoretical Value
voucher <- trap.data[trap.data$Sample == "Voucher" & trap.data$Time == "1", 3]</pre>
# Correct For Vol
in1.c <- in1 * 5 # used 100ul of 500ul
out1.c <- out1 * 50 # used 100ul of 5000ul
# Percent of Theoretical
in1.p <- in1.c / voucher</pre>
out1.p <- out1.c / voucher</pre>
# Calculate mean and sem
means <- c("Trap" = mean(in1.p), "Residual" = mean(out1.p))</pre>
ses <- c("Trap" = se(in1.p), "Residual" = se(out1.p))</pre>
# Plot
png(filename="../figures/Recovery1.png",
    width = 800, height = 800, res = 96*2)
par(mar=c(3,5,0.5,0.5), oma=c(1,1,1,1)+0.1, lwd=2)
bp_plot <- barplot(means, ylab = "Percent Recovery",</pre>
                   ylim = c(0, 1), lwd=3, yaxt="n", col="gray",
                   cex.lab=1.5, cex.names = 1.25)
arrows(x0 = bp_plot, y0 = means, y1 = means - ses, angle = 90,
       length=0.1, lwd = 2)
arrows(x0 = bp_plot, y0 = means, y1 = means + ses, angle = 90,
       length=0.1, lwd = 2)
axis(side = 2, labels=T, lwd.ticks=2, las=2, lwd=2)
dev.off() # this writes plot to folder
graphics.off() # shuts down open devices
```

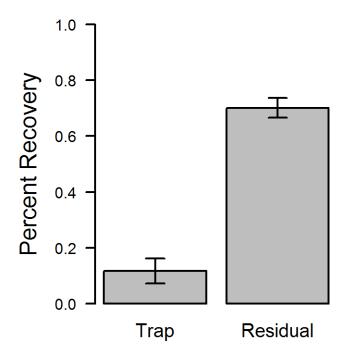


Figure 2: 14C-Bicarbonate Recovery