VO2sim 0.1: Using Simulation to Understand Measurement Error in Indirect Calorimetry

by Matthew S Tenan
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by Matthew S Tenan

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# VO2sim 0.1: Using Simulation to Understand Measurement Error in Indirect Calorimetry

Indirect calorimetry is commonly used in military research to examine the energetic cost of exercise during various perturbations and as a risk factor for injury/illness. The Army has recognized the importance of understanding oxygen consumption in the field and is developing models to aid in operational decision making. A common study design within the Army and in other exercise applications is a test-retest design. Failure to control for expected variability in testing may lead researchers to conclude that an intervention is effective when their results are only a product of chance variability within the human or machine system. This report details the development of a simulation program that accounts for system and human variability to determine if 2 testing sessions are different.

## Subject Terms
- metabolic, indirect calorimetry, COSMED, VO\(_2\), VCO\(_2\), caloric expenditure, statistical simulation

## Abstract
Indirect calorimetry is commonly used in military research to examine the energetic cost of exercise during various perturbations and as a risk factor for injury/illness. The Army has recognized the importance of understanding oxygen consumption in the field and is developing models to aid in operational decision making. A common study design within the Army and in other exercise applications is a test-retest design. Failure to control for expected variability in testing may lead researchers to conclude that an intervention is effective when their results are only a product of chance variability within the human or machine system. This report details the development of a simulation program that accounts for system and human variability to determine if 2 testing sessions are different.
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1. Introduction

Indirect calorimetry is commonly used in military research to examine the energetic cost of exercise during various perturbations and as a risk factor for injury/illness. The US Army has used indirect calorimetry to determine the metabolic cost of using functional exoskeletons (Gregorczyk et al. 2006) and when comparing rucksack design (LaFiandra et al. 2003). Indirect calorimetry has also been used to understand how military personnel acclimatize to high altitude (Amann et al. 2013) and hypoxia (Self et al. 2013). The Army has recognized the importance of understanding oxygen consumption in the field and is developing models to aid in operational decision making (Weyand 2013). Decreasing the energetic cost, or caloric expenditure, of a task is important since it results in a decreased need to carry food as well as a lower likelihood of fatigue and injury (Mair et al. 1996; Gefen 2002; Borotikar et al. 2008).

Caloric expenditure was originally quantified via direct calorimetry where a subject exists within a room-sized module insulated by a water current. The heat given off by the body heats the water current and is measured to determine the energy expended by the body (Atwater and Benedict 1983). The cumbersome direct calorimetry method was later updated so that volumes of expired oxygen (VO₂) and carbon dioxide (VCO₂) can be measured to determine the caloric expenditure while subjects are engaged in activities outside a singular room (Weir 1949). These volumes are calculated by determining the total gas ventilation (VE), assuming a standard inspired gas ratio (20.95% O₂ and 0.03% CO₂) and assuming that gaseous nitrogen is inert (inspired N₂ = expired N₂); the expired fractions of O₂ and CO₂ determined by a metabolic system are then used to calculate VO₂ and VCO₂. This indirect calorimetry method has been further refined by advances in O₂ and CO₂ sensors and resulted in the present generation of mobile metabolic devices from a variety of manufacturers.

The most commonly used fully portable indirect calorimetry device is the Cosmed K4b². This is the device presently in use at the US Army Research Laboratory’s Human Research and Engineering Directorate. A common study design within the Army and in other exercise applications is a test–retest design. In this design, a Soldier will be tested under normal or control conditions and then tested again during experimental conditions. The difference observed between the control and experimental conditions is theoretically a direct result of the experimental perturbation. However, testing session variability at the individual physiologic level (e.g., dietary changes, diurnal hormonal oscillations) and at the level of the measuring device (e.g., O₂/CO₂ sensor variability, ambient air temperature,
barometric pressure) may obscure any minor changes due to the experimental intervention. Therefore, it is important to account for the variability expected between 2 identical “control” testing sessions when examining the result of an experimental trial. Failure to control for expected variability in testing may lead researchers to conclude that an intervention is effective when their results are only a product of chance variability within the human or machine system.

The author developed a computer program, VO2sim, to address the concern that nonuniform results in experimental trials may be a product of natural measurement variability. VO2sim 0.1 is a simulation tool to determine if the experimental calorimetry measurement is outside the range that would be expected for normal session-to-session variability (e.g., 100% chance experimental condition is higher than control condition). If the experimental measure is determined to be within session-to-session variability, a probability measure is returned, indicating the probability that the experimental value is higher or lower than the control measure (e.g., 60% chance that experimental value is lower than control value). The general purpose of VO2sim is to provide a probabilistic context for VE, VO2, and VCO2 measures that are commonly made within a test–retest study methodology.

2. Methodology

VO2sim 0.1 is written in R 3.1.2 (R Core Team 2014), which is a free programming language for statistics and data science. The present iteration of VO2sim is valid only for the Cosmed K4b² system and uses previously published reliability data to determine the distribution of measurement error (Duffield et al. 2004; Darter et al. 2013). VO2sim is an umbrella term for 3 functions: VO2sim.repeat, VCO2sim.repeat, and VEsim.repeat, which assess the probability of real differences between the control and experimental conditions for VO2, VCO2, and VE, respectively (raw code for functions is available in Appendix A). The operational flow for each function is the same; VO2sim.repeat will be used as an example.

The control and experimental VO2 measures (liters/minute) obtained at the same exercise workload are put into the function with the “cosmed_k4b2” indicated as the system that collected the data. The control volume and system are used as the means to create a random uniform distribution of 100,000 observations, where the maximum and minimum of the distribution are calculated based upon the intraclass correlation (ICC) for that volume. The formula for calculating the maximum and minimum of the distribution is:

\[
\text{Distribution max/min} = \text{volume} \pm [(1 - \text{ICC}) \times \text{volume}].
\]
For example, a VO₂ measurement of 1.5 L/min has a repeatability ICC of 0.85 (Darter et al. 2013); therefore, a random uniform distribution between 1.275 and 1.725 is created from a control measurement of 1.5 L/min (Fig. 1). ICCs for session-to-session reliability differ depending upon the total volume of gas flow; therefore, the ICC differs for each system depending upon on the volume of gas measured. The random uniform distribution was selected in lieu of other distributions because previous research has demonstrated that session-to-session agreement follows a random uniform distribution (Crouter et al. 2006).

Fig. 1 Random uniform distribution with a mean of 1.5 L/min
The simulated distribution of expected values for a second control condition is then compared against the measured experimental session VO₂. If the experimental value falls outside of the distribution on the high end, it will return an output that there is a 100% probability that the experimental value is higher than the control value (Fig. 2). If the experimental value falls within the expected control distribution, it is determined whether the greatest number of samples in the distribution are higher (Fig. 3), lower (Fig. 4), or identical to the experimental value. The highest number of samples is calculated in ratio with the total samples in the distribution to determine the probability that the experimental value is higher, lower, or identical to the control value.

Fig. 2 Comparison of random uniform distribution to an experimental value falling outside the control distribution
Fig. 3  Comparison of the random uniform distribution to a value that falls within the control distribution and where 60% of the control distribution is lower than the experimental value.
Fig. 4 Comparison of the random uniform distribution to a value that falls within the control distribution and where 75% of the control distribution is greater than the experimental value.

### 2.1 Calling VO2sim 0.1 Functions

`VO2sim.repeat` is called with up to 3 arguments:

```
VO2sim.repeat(a, b, system = 'cosmed_k4b2')
```

Variable “a” is the control value and variable “b” is the experimental value. The default system is “cosmed_k4b2”. In the current version of VO2sim, there is not presently support for other indirect calorimetry systems, so any other input will result in a system message indicating that the data are unable to be processed. Future versions of VO2sim will support other popular indirect calorimetry systems that have appropriate repeatability data available.
2.2 VO2sim 0.1 Results

The VO2sim.repeat function returns an object with 3 respective values:

- Probability that the measures are the same
- Probability that “b” is greater than “a”
- Probability that “b” is less than “a”

A string is also returned explaining which probability in the previous object is the largest. The string is designed to explain the primary findings of VO2sim for a single subject. The data in the object allow the researcher to aggregate VO2sim results from multiple subjects and return a “net probability difference” based upon the mean of the data from the column appropriate to their a priori hypothesis.

2.3 Calculating Difference Probability Curves

The benefit of having VO2sim functions contained within R allows for entire vectors of data to be analyzed with ease. Using simulated sequential data, curves can be constructed to demonstrate how a 3% (Fig. 5), 6% (Fig. 6), 10% (Fig. 7), and 15% (Fig. 8) measured increase in experimental VO2 result in the probability that it is actually greater than the control session. The implementation of these functions on simulated vectors for the creation of Figs. 5–8 is contained within Appendix B.
Fig. 5 VO2sim probabilities that experimental value is different from control when the experimental value is 3% higher than control
Fig. 6 VO2sim probabilities that experimental value is different from control when the experimental value is 6% higher than control
Fig. 7 VO2sim probabilities that experimental value is different from control when the experimental value is 10% higher than control
3. Discussion and Conclusions

VO2sim is designed to test the hypothesis that VO2, VCO2, and VE values from an experimental trial are either lower or higher than the control trial. To that end, it can be used in lieu of a standard statistical test for a single subject analysis. When data from more than one subject are obtained, best practice is to first use a repeated-measures t-test. If statistical significance of a 2-tailed test is obtained, a directional hypothesis should be made; the individual data for all subjects should be analyzed with VO2sim and a “net probability difference” can be calculated. Instances where statistical differences are achieved but the net probability of VO2sim difference is less than 85% should lead the researcher to interpret their results with heavy skepticism. The most conservative interpretation of the VO2sim output is that any difference probability less than 100% is not different between the 2 conditions. Proposed conventions for applying VO2sim are as follows:

- Experimental value definitely different from Control: 100%
- Experimental value probably different from Control: 85%–99%

Fig. 8 VO2sim probabilities that experimental value is different from control when the experimental value is 15% higher than control
• Experimental value unlikely to be different from Control: 70%–84%
• Experimental value not different from Control: Below 69%

The probability curves (Figs. 5–8) can also be used during study design. With the goal of maximizing group differences, investigators may want to consider a certain workload where measurements are less likely to be obscured by measurement error. Similarly, if there is a standard exercise protocol, they may want to purchase a system that has high test–retest reliability in their typical range of workloads.

The current iteration of VO2sim is only able to assess the measurement error for one device, the Cosmed K4b². The reliability data for the Cosmed K4b² are reported for discrete volume measurements (e.g., VE = 45 L/min) and not as a continuous regression line (Duffield et al. 2004; Darter et al. 2013). This accounts for the visual grouping of results based upon the volume of gas measured (see Figs. 5–8). Future versions will support the assessment of other indirect calorimetry systems and will incorporate a continuous estimation of test–retest agreement. Future versions will also implement a graphical user interface to facilitate the use of VO2sim for researchers/clinicians not familiar with the R language.
4. References


Weyand P. Locomotion with loads: practical techniques for predicting performance outcomes. Ft. Detrick (MD): Army Medical Research and Materiel Command (US); 2013 May.
Appendix A. VO2sim Functions

This appendix appears in its original form, without editorial change.
**VO2sim.repeat**

#'a' is the base condition, 'b' is experimental.

```r
VO2sim.repeat <- function(a, b, system='cosmed_k4b2'){
a.round <- round(a, digits=2)
b.round <- round(b, digits=2)
if (system=='cosmed_k4b2'){
  if (a.round <= 0.5){
    a.min <- a.round*0.43
    a.max <- a.round*1.57
    a.dist <- round(runif(100000, min= a.min, max= a.max), digits=2)
  } else if (a.round>0.5 & a.round<1.0){
    a.min <- a.round*0.90
    a.max <- a.round*1.10
    a.dist <- round(runif(100000, min= a.min, max= a.max), digits=2)
  } else if (a.round>=1.0 & a.round<3.5){
    a.min <- a.round*0.85
    a.max <- a.round*1.15
    a.dist <- round(runif(100000, min= a.min, max= a.max), digits=2)
  } else if (a.round>= 3.5){
    a.min <- a.round*0.87
    a.max <- a.round*1.13
    a.dist <- round(runif(100000, min= a.min, max= a.max), digits=2)
  } else{
    print('I can not process data from systems other than Cosmed k4b2 at this time')
  }
}
if (system =='cosmed_k4b2'){
  probsame <- (sum(a.dist == b.round)/100000)*100
  probhigh <- (sum(a.dist < b.round)/100000)*100
  problow <- (sum(a.dist > b.round)/100000)*100
  probs <- c(probsame, probhigh, problow)
  highest.prob <- probs[probs==max(probs)]
  if (highest.prob == probsame){
    print(paste0("There is a ", highest.prob, " probability that these measures are the same VO2"))
  } else if (highest.prob == probhigh){
    print(paste0("There is a ", highest.prob, " probability that VO2 b is greater than VO2 a"))
  } else if (highest.prob == problow){
    print(paste0("There is a ", highest.prob, " probability that VO2 b is less than VO2 a"))
  }
  return(probs)
}
```

16
VCO2sim.repeat  
# a is the base condition, 'b' is experimental.
VCO2sim.repeat<- function(a, b, system='cosmed_k4b2'){
  a.round <- round(a, digits=2)
  b.round <- round(b, digits=2)
  if (system=='cosmed_k4b2'){
    if (a.round < 3.25){
      a.min <- a.round*0.81
      a.max <- a.round*1.19
      a.dist <- round(runif(100000,min= a.min, max= a.max), digits=2)
    } else if (a.round>= 3.25){
      a.min <- a.round*0.72
      a.max <- a.round*1.28
      a.dist <- round(runif(100000,min= a.min, max= a.max), digits=2)
    }
  } else{
    print('I can not process data from systems other than Cosmed k4b2 at this time')
  }
  if (system =='cosmed_k4b2'){
    probsame <- (sum(a.dist == b.round)/100000)*100
    probhigh <- (sum(a.dist < b.round)/100000)*100
    problow <- (sum(a.dist > b.round)/100000)*100
    probs <- c(probsame, probhigh, problow)
    highest.prob <- probs[probs==max(probs)]
    if (highest.prob == probsame){
      print(paste0("There is a ", highest.prob, " probability that these measures are the same VCO2"))
    } else if (highest.prob == probhigh){
      print(paste0("There is a ", highest.prob, " probability that VO2 b is greater than VCO2 a"))
    } else if (highest.prob == problow){
      print(paste0("There is a ", highest.prob, " probability that VO2 b is less than VCO2 a"))
    }
    return(probs)
  }
}
VEsim.repeat
# a is the base condition, 'b' is experimental.
VEsim.repeat<- function(a, b, system='cosmed_k4b2'){
a.round <- round(a, digits=2)
b.round <- round(b, digits=2)
if (system=='cosmed_k4b2'){
if (a.round < 80.00){
a.min <- a.round*0.88
a.max <- a.round*1.12
a.dist <- round(runif(100000,min= a.min, max= a.max), digits=2)
}
else if (a.round>= 80.00){
a.min <- a.round*0.78
a.max <- a.round*1.22
a.dist <- round(runif(100000,min= a.min, max= a.max), digits=2)
}
else{
  print('I can not process data from systems other than Cosmed k4b2 at this time')
}
if (system =='cosmed_k4b2'){
  probsame <- (sum(a.dist == b.round)/100000)*100
  probhigh <- (sum(a.dist < b.round)/100000)*100
  problog <- (sum(a.dist > b.round)/100000)*100
  probs <- c(probsame, probhigh, problog)
  highest.prob <- probs[probs==max(probs)]
  if (highest.prob == probsame){
    print(paste0("There is a ", highest.prob, " probability that these measures are the same VE"))
  } else if (highest.prob == probhigh){
    print(paste0("There is a ", highest.prob, " probability that VO2 b is greater than VE a"))
  } else if (highest.prob == problog){
    print(paste0("There is a ", highest.prob, " probability that VO2 b is less than VE a"))
  }
}
return(probs)
}
This is the R Markdown document detailing the use of VO2sim.repeat and creating the figures for the technical note.

Install VO2sim package and place it in library + load ggplot2 for graphing

```r
setwd("L:/Biomechanics/VO2sim/VO2sim")
install.packages("devtools", repos= "http://cran.rstudio.com/")
## Installing package into 'C:/Users/matthew.s.tenan/Documents/R/win-library/3.1'
## (as 'lib' is unspecified)
## package 'devtools' successfully unpacked and MD5 sums checked
## The downloaded binary packages are in
## C:\Users\matthew.s.tenan\AppData\Local\Temp\RtmpKae3C6\downloaded_packages
install.packages( 'gridExtra', repos = "http://cran.rstudio.com/")
## Installing package into 'C:/Users/matthew.s.tenan/Documents/R/win-library/3.1'
## (as 'lib' is unspecified)
## package 'gridExtra' successfully unpacked and MD5 sums checked
## The downloaded binary packages are in
## C:\Users\matthew.s.tenan\AppData\Local\Temp\RtmpKae3C6\downloaded_packages
library(gridExtra)
## Loading required package: grid
library("devtools")
## WARNING: Rtools is required to build R packages, but is not currently installed.
```
## Please download and install Rtools 3.1 from http://cran.r-project.org/bin/windows/Rtools/ and then run find_rtools().

```r
install('VO2sim')
```

## Installing VO2sim
```r
"C:/PROGRA~1/R/R-31~1.2/bin/x64/R" --vanilla CMD INSTALL
"L:/Biomechanics/VO2sim/VO2sim"
--library="C:/Users/matthew.s.tenan/Documents/R/win-library/3.1"
--install-tests
```

```r
library('VO2sim')
library('ggplot2')
```

### Creation of example plots to demonstrate how VO2sim.repeat works

#### Simulate Data
```r
set.seed(71185)
dist.x <- as.data.frame(round(runif(100000, min= 1.275, max= 1.725), digits=2))
colnames(dist.x) <- 'sim_con'
```

#### Plots 1 & 2
```r
ggplot(dist.x, aes(x = sim_con)) +
  geom_histogram(colour = "black", fill = "darkgreen", binwidth = .01) +
  theme_bw() +
  xlab('Simulated VO2 for Second Control Session (L/min)') +
  ylab('Count') +
  ylim(0, 3000)
```
Plot 3

```r
ggplot(dist.x, aes(x = sim_con)) +
  geom_histogram(colour = "black", binwidth = .01, aes(fill = (sim_con==1.55))) +
  scale_fill_manual(values=c('FALSE'='darkgreen', 'TRUE'='firebrick')) +
  theme_bw() +
  xlab('Simulated VO2 for Second Control Session (L/min)') +
  ylab('Count') +
  ylim(0, 3000) +
  scale_color_identity() +
  theme(legend.position="none")
```
Plot 4

```r
ggplot(dist.x, aes(x = sim_con)) +
  geom_histogram(colour = "black", binwidth = .01, aes(fill = (sim_con==1.38)) +
  scale_fill_manual(values=c('FALSE'='darkgreen', 'TRUE'='firebrick')) +
  theme_bw() +
  xlab('Simulated VO2 for Second Control Session (L/min)') +
  ylab('Count') +
  ylim(0, 3000) +
  scale_color_identity() +
  theme(legend.position="none")
```
Creating Data and Plot 5

```r
###Creation of data for figure 5, 3% increase in VO2
control <- seq(from = 0.01, to = 4.5, by = 0.01)
exp <- control*1.03
#Run VO2sim
fig5dat <- Map(VO2sim.repeat, a=control, b=exp)

# There is a 87.718 probability that these measures are the same VO2
# There is a 43.869 probability that these measures are the same VO2
# There is a 35.604 probability that VO2 b is greater than VO2 a
# There is a 39.157 probability that VO2 b is greater than VO2 a
# There is a 41.284 probability that VO2 b is greater than VO2 a
# There is a 42.769 probability that VO2 b is less than VO2 a
# There is a 43.77 probability that VO2 b is less than VO2 a
# There is a 44.54 probability that VO2 b is greater than VO2 a
# There is a 45.339 probability that VO2 b is less than VO2 a
# There is a 45.77 probability that VO2 b is less than VO2 a
# There is a 46.125 probability that VO2 b is greater than VO2 a
# There is a 46.61 probability that VO2 b is less than VO2 a
# There is a 46.668 probability that VO2 b is less than VO2 a
# There is a 47.063 probability that VO2 b is greater than VO2 a
# There is a 47.326 probability that VO2 b is less than VO2 a
# There is a 52.852 probability that VO2 b is greater than VO2 a
# There is a 52.284 probability that VO2 b is greater than VO2 a
# There is a 52.094 probability that VO2 b is greater than VO2 a
```
There is a 52.229 probability that VO2 b is greater than VO2 a
There is a 52.037 probability that VO2 b is greater than VO2 a
There is a 51.892 probability that VO2 b is greater than VO2 a
There is a 52.179 probability that VO2 b is greater than VO2 a
There is a 51.585 probability that VO2 b is greater than VO2 a
There is a 51.691 probability that VO2 b is greater than VO2 a
There is a 51.594 probability that VO2 b is greater than VO2 a
There is a 51.447 probability that VO2 b is greater than VO2 a
There is a 51.34 probability that VO2 b is greater than VO2 a
There is a 51.177 probability that VO2 b is greater than VO2 a
There is a 51.271 probability that VO2 b is greater than VO2 a
There is a 51.502 probability that VO2 b is greater than VO2 a
There is a 51.335 probability that VO2 b is greater than VO2 a
There is a 51.271 probability that VO2 b is greater than VO2 a
There is a 50.992 probability that VO2 b is greater than VO2 a
There is a 51.039 probability that VO2 b is greater than VO2 a
There is a 51.196 probability that VO2 b is greater than VO2 a
There is a 51.534 probability that VO2 b is greater than VO2 a
There is a 50.967 probability that VO2 b is greater than VO2 a
There is a 50.98 probability that VO2 b is greater than VO2 a
There is a 50.994 probability that VO2 b is greater than VO2 a
There is a 50.97 probability that VO2 b is greater than VO2 a
There is a 50.69 probability that VO2 b is greater than VO2 a
There is a 50.827 probability that VO2 b is greater than VO2 a
There is a 50.743 probability that VO2 b is greater than VO2 a
There is a 50.926 probability that VO2 b is greater than VO2 a
There is a 52.648 probability that VO2 b is greater than VO2 a
There is a 64.697 probability that VO2 b is greater than VO2 a
There is a 64.599 probability that VO2 b is greater than VO2 a
There is a 64.186 probability that VO2 b is greater than VO2 a
There is a 63.862 probability that VO2 b is greater than VO2 a
There is a 63.996 probability that VO2 b is greater than VO2 a
There is a 63.633 probability that VO2 b is greater than VO2 a
There is a 62.914 probability that VO2 b is greater than VO2 a
There is a 62.785 probability that VO2 b is greater than VO2 a
There is a 62.827 probability that VO2 b is greater than VO2 a
There is a 62.508 probability that VO2 b is greater than VO2 a
There is a 62.216 probability that VO2 b is greater than VO2 a
There is a 61.929 probability that VO2 b is greater than VO2 a
There is a 62.008 probability that VO2 b is greater than VO2 a
There is a 61.702 probability that VO2 b is greater than VO2 a
There is a 61.732 probability that VO2 b is greater than VO2 a
There is a 61.277 probability that VO2 b is greater than VO2 a
There is a 61.293 probability that VO2 b is greater than VO2 a
There is a 61.016 probability that VO2 b is greater than VO2 a
There is a 60.91 probability that VO2 b is greater than VO2 a
There is a 60.858 probability that VO2 b is greater than VO2 a
There is a 60.275 probability that VO2 b is greater than VO2 a
There is a 60.711 probability that VO2 b is greater than VO2 a
There is a 60.22 probability that VO2 b is greater than VO2 a
There is a 60.118 probability that VO2 b is greater than VO2 a
There is a 60.006 probability that VO2 b is greater than VO2 a
There is a 59.9 probability that VO2 b is greater than VO2 a
There is a 59.582 probability that VO2 b is greater than VO2 a
There is a 59.892 probability that VO2 b is greater than VO2 a
There is a 59.463 probability that VO2 b is greater than VO2 a
There is a 59.203 probability that VO2 b is greater than VO2 a
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## Create plots for figure 5

### Same plot

defig5dat <- as.data.frame(do.call(rbind, fig5dat))
colnames(fig5dat) <- c('same', 'higher', 'lower')
fig5dat <- cbind(fig5dat, as.data.frame(exp))

# Same plot

same.3 <- ggplot(fig5dat, aes(x=exp, y=same)) + geom_point() + theme_bw() + ylab('Probability that Experimental Value is Identical to Control') + xlab('Experimental Value (L/min)') + ylim(0,100)

### Higher Plot

higher.3 <- ggplot(fig5dat, aes(x=exp, y=higher)) + geom_point() + theme_bw() + ylab('Probability that Experimental Value is Greater Than Contr
# Lower Plot
lower.3 <- ggplot(fig5dat, aes(x=exp, y=lower)) + geom_point() + theme_bw() + ylab('Probability that Experimental Value is Less Than Control') + xlab('Experimental Value (L/min)') + ylim(0,100)

# make figure 5
grid.arrange(same.3,higher.3,lower.3, ncol=3)