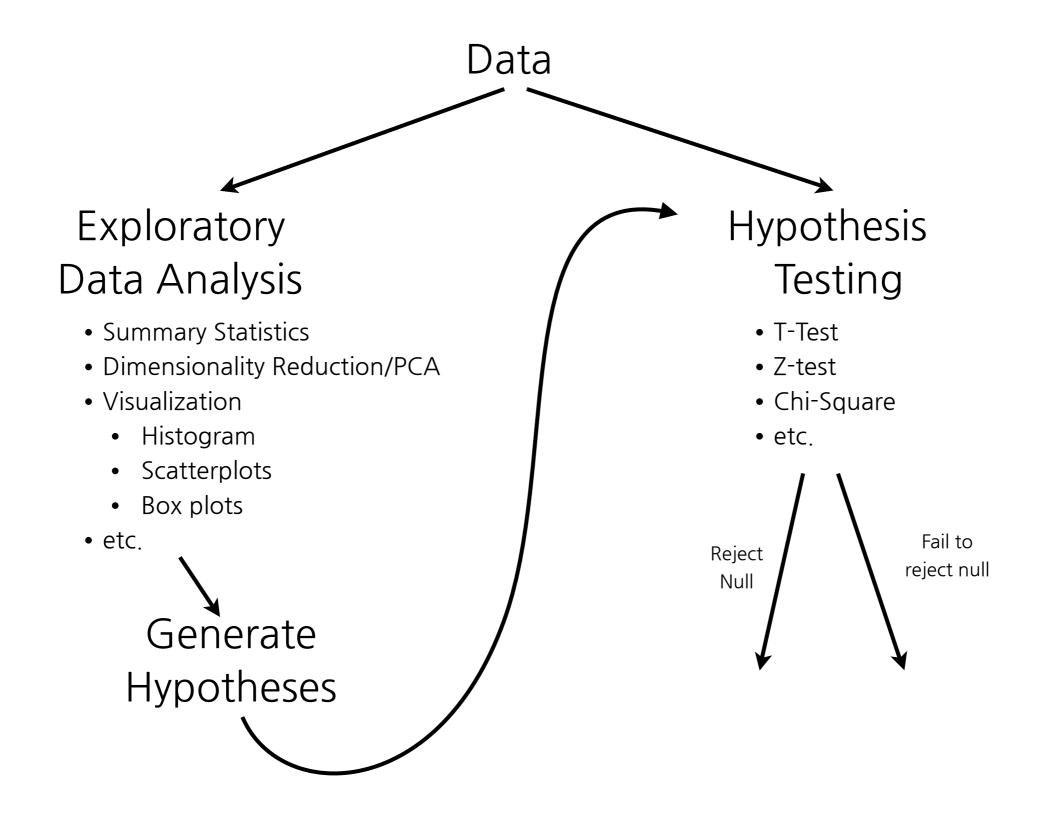
# Statistics and Hypothesis Testing

NENS 230: Data Analysis for the Biosciences using MATLAB

Eddy Albarran November 3, 2015

## Analysis Methodology



#### Outline

## Summary statistics functions Random Variables

- Random variables, PDF, CDFs
- Estimates of central tendency and dispersion
- Standard error of the mean, confidence intervals

#### **Statistical Hypothesis Testing**

- Tests and significance
- Student's t test walkthrough
- Other commonly used tests

#### Analysis of Variance Homework

## Summary Statistics

```
Commonly used functions:
  -mean()
```

- std()
- var()
- sum()
- min()
- $-\max()$

#### mean() function

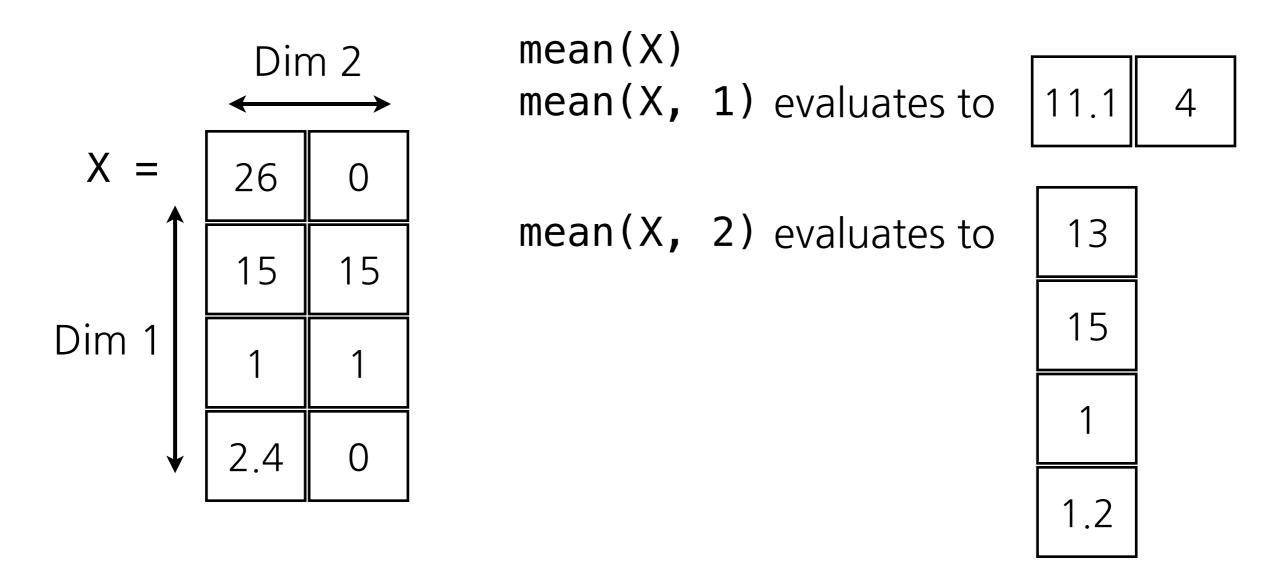
mean() computes the average (sample mean) of a vector. With matrices, you need to specify which dimension to average along.

mean (X, 1) means return the average row (average across the rows). This is the default if you only specify one argument.

mean(X, 2) means return the average column (average across the columns)

### mean() function

mean() computes the average (sample mean) of a vector. When dealing with matrices, you need to specify which dimension to average along.



## mean() function

mean() operates on its first argument. Be careful when averaging two things together that you pack them in a vector using [ ]

```
mean(1, 5) evaluates to 1
```

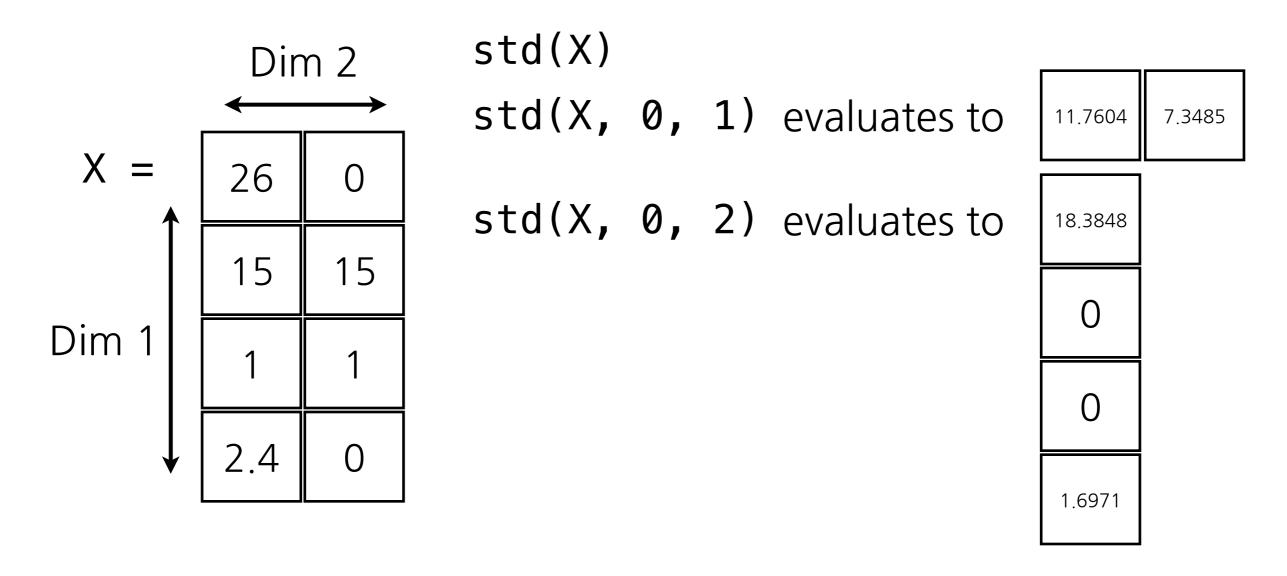
"Take the mean of [1] along the 5th dimension"

mean([1 5]) evaluates to 3

### std() function

std() computes the standard deviation of a list of numbers

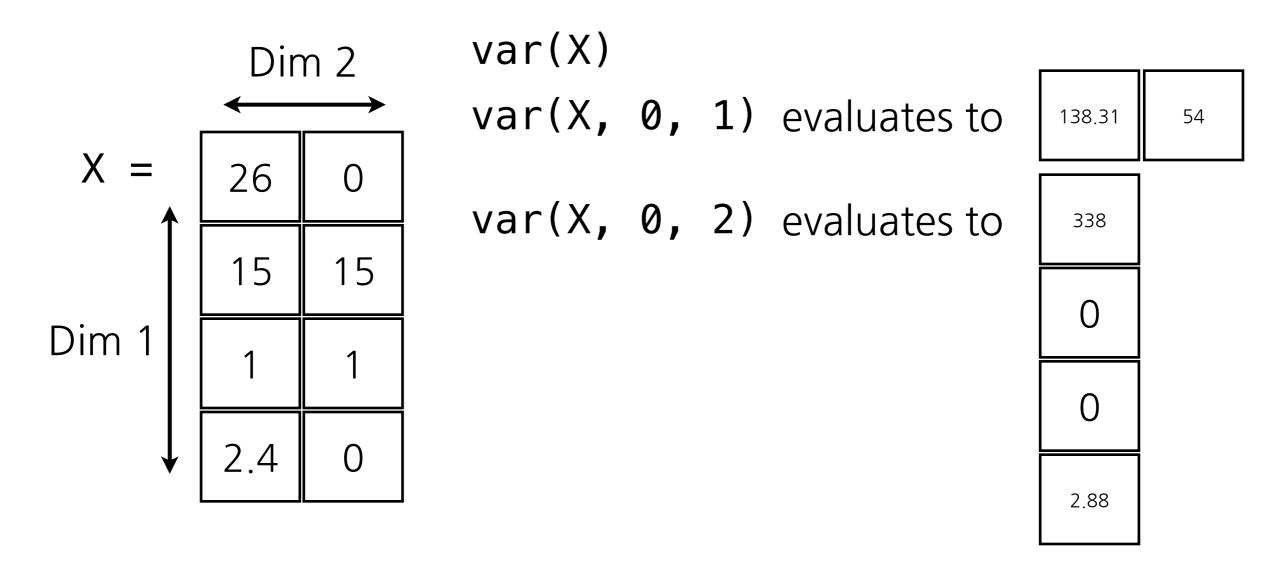
- When dealing with matrices, you need to specify which dimension to average along, as the third argument.
- The second argument should be 0 if you want the unbiased estimator that normalizes by n-1, where n is the number of samples



### var() function

var() computes the sample variance of a list of numbers

- When dealing with matrices, you need to specify which dimension to operate along, as the third argument.
- The second argument should be 0 if you want the unbiased estimator that normalizes by n-1, where n is the number of samples. (This is the default)



#### sum() function

**sum()** computes the sum of a vector. When dealing with matrices, you should specify which dimension to average along.

sum(X, 1) means return the sum over rows (sum over rows within each column). This is the default if you only specify one argument.

**sum(X, 2)** means return the sum over columns (sum over columns within each row)

#### min() function

min() computes the minimum of a vector. When dealing with matrices, you should specify which dimension to find the minimum along.

min(X, Y) means return an array the same size as X and Y consisting of the smaller of the elements in X and Y at each location.

min(X, [], 1) means return the minimum value in each column. This is the default if you only specify one argument.

min(X, [], 2) means return the minimum in each row.

#### max() function

max() computes the maximum of a vector. When dealing with matrices, you should specify which dimension to find the maximum along.

max(X, Y) means return an array the same size as X and Y consisting of the larger of the elements in X and Y at each location.

max(X, [], 1) means return the maximum value in each column. This is the default if you only specify one argument.

max(X, [], 2) means return the maximum in each row.

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Homework

#### Discrete random variables

Suppose we have a random variable X.

Discrete random variables take one value within a set of k possible values.

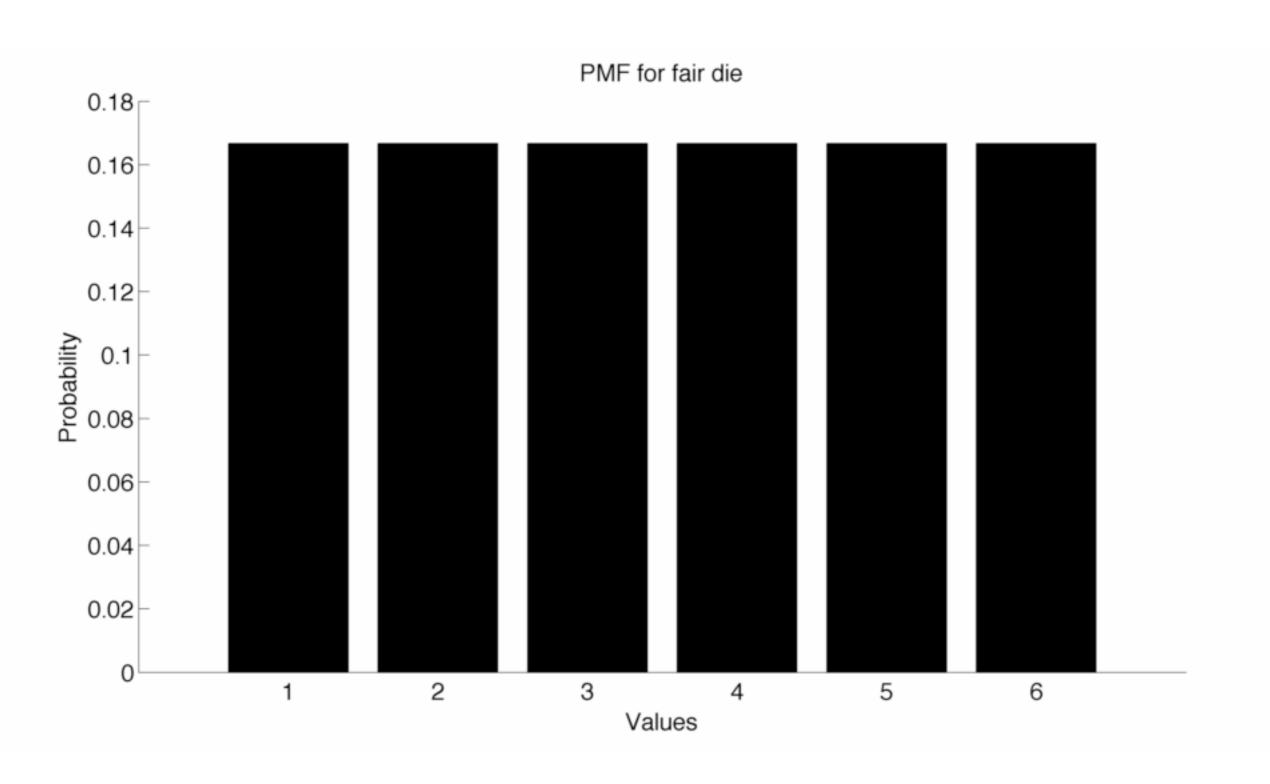
**Probability mass function:** For a given value x<sub>i</sub> returns the probability p<sub>i</sub> of X taking that value.

$$Pr[X = x_i] = p_i$$

Sum of these probabilities must be 1.

$$p_1 + p_2 + \dots + p_k = 1$$

## Probability Mass Function



#### Continuous random variables

Suppose we have a random variable X.

Continuous random variables take values within some continuous range of values.

**Probability density function (PDF):** integrating this function over some interval gives you the probability that X lies in that interval.

$$Pr[a \le X \le b] = \int_a^b f(x)dx$$

Therefore, the integral under this function is 1.

$$\int_{-\infty}^{\infty} f(x)dx = 1$$

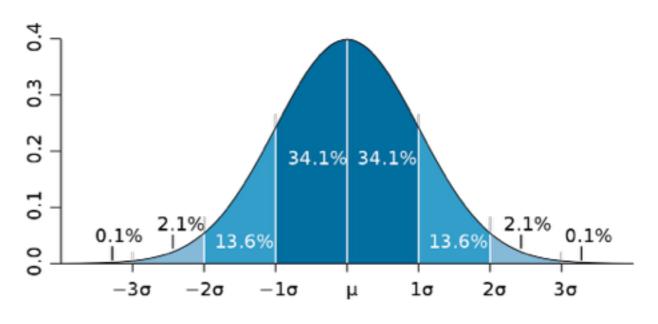
#### Normal distribution

Normal or Gaussian distributions describe many naturally occurring phenomena, due to the central limit theorem.

Specified by two parameters:

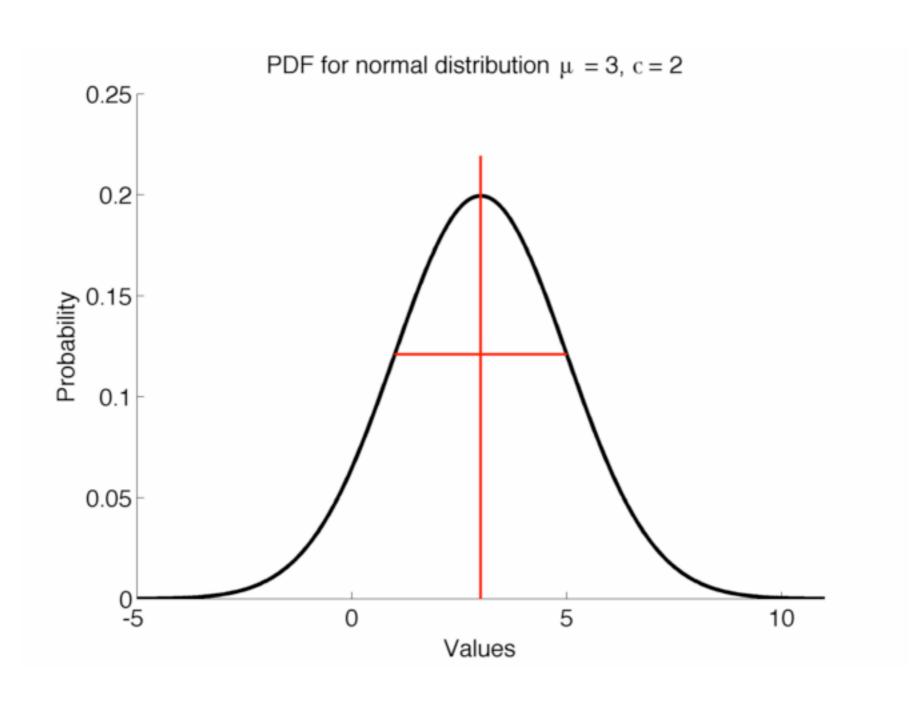
- **Location parameter:** the mean (µ)
- **Scale parameter:** the standard deviation (σ)

$$\frac{1}{\sigma\sqrt{(2\pi)}}e^{-\frac{(x-\mu)^2}{2\sigma^2}}$$



Source: wikipedia.org

## PDF for normal distribution



#### Cumulative distribution function

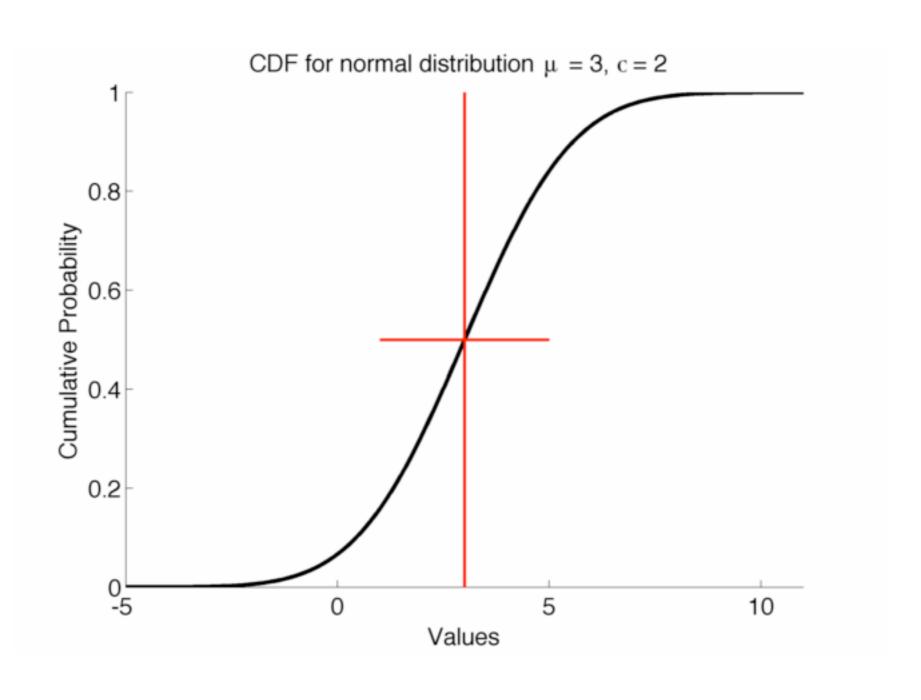
Cumulative distribution function (CDF): how likely is X less than or equal to a particular value.

$$Pr[X \le x] = F(x)$$

The CDF is the integral of the PDF.

The PDF is the derivative of the CDF. Therefore, the parts of the CDF with the steepest slope are the highest points of the PDF, i.e. where most of the values lie.

## CDF for normal distribution



## **Expected Value**

The expected value of a random variable is it's mean. You can calculate the expected value of a random variable X by taking the weighted average of all its possible values. The weights are the probability of X taking each value.

Discrete RV: 
$$E[X] = x_1p_1 + x_2p_2 + \cdots + x_kp_k$$

Continuous RV: 
$$E[X] = \int_{-\infty}^{\infty} x f(x) dx$$

## Sample mean

**Sampling:** When we measure some quantity in an experiment, we think of it as taking samples from a distribution.

**Sample mean:** By taking the average, we are estimating the mean or expected value of the underlying distribution which generated these quantities.

A central problem in statistics: How close is this estimate of the mean (the average of our samples) to the true, underlying mean?

#### Standard Error of the Mean

Suppose we make N measurements of X, sampling from a normal distribution with mean  $\mu$  and standard deviation  $\sigma$ .

If we take the average of these N samples, our estimate of the mean is a normal distribution.

The mean of this sampling distribution is µ

The standard error is  $\sigma$  / sqrt(N).

This means that on average, our estimate will be correct. The spread around the true mean shrinks as 1/sqrt(N).

#### Standard Error of the Mean

Suppose we make N measurements of X which may or not be normally distributed.

If we take the average of these N samples, our estimate of the mean **approaches** a normal distribution as N gets larger (central limit theorem).

The mean of this sampling distribution is µ

The standard error is  $\sigma$  / sqrt(N).

#### Confidence intervals

Based on the data you've collected, you can estimate the true value of some quantity, e.g. the true mean.

This estimate of the quantity isn't perfect. Confidence intervals tell you a range of values where the true value lies with some probability

95% confidence intervals are the range where the true value of the quantity will lie with 95% probability.

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Analysis of Variance

Dimensionality Reduction

- PCA

Final Project

## Statistical hypothesis testing

The point of statistical tests is to cast doubt on the veracity of a **null hypothesis**.

If null hypothesis true, it would be very unlikely to observe the given data.

Statistical tests reject null hypothesis if the un-likelihood of the data crosses a threshold.

This threshold or significance level is typically expressed as a p-value: the likelihood of false-rejections

- i.e. the likelihood that the null hypothesis would be rejected if it were true.

## Statistical hypothesis testing

- 1. State the null (H<sub>3</sub>), and alternative (H<sub>1</sub>) hypotheses
- 2. State the assumptions

```
Independence of samples?
```

Normality?

- 3. Determine an appropriate test statistic
- 4. Derive the distribution of the test statistic under the null hypothesis
- 5. Determine the critical region for the test statistic
- 6. Compute the observed value of the test statistic
- 7. **Reject** or **fail to reject** the null hypothesis (H<sub>0</sub>)
- 8. i.e.: Compute the strongest significance level at which the null hypothesis would be rejected (p-value)

Suppose we monitor scores on some behavioral assay before and after treatment. We take the difference of the scores for each subject.

Each subject's change in scores is xi

1) State the null (and alternative) hypotheses:

**Null hypothesis:** x<sub>i</sub> are drawn from a normal distribution with zero mean.

**Alternative hypothesis:** x<sub>i</sub> are drawn from a normal distribution with non-zero mean.

#### State the assumptions:

All samples are independent.

Changes in scores have a normal distribution. This follows from scores on the test before and after having normal distributions.

#### **Test Statistics**

#### Determine an appropriate test statistic.

A test statistic is a numerical summary of data that reduces the information needed to perform a hypothesis to a single value (or a small number of values).

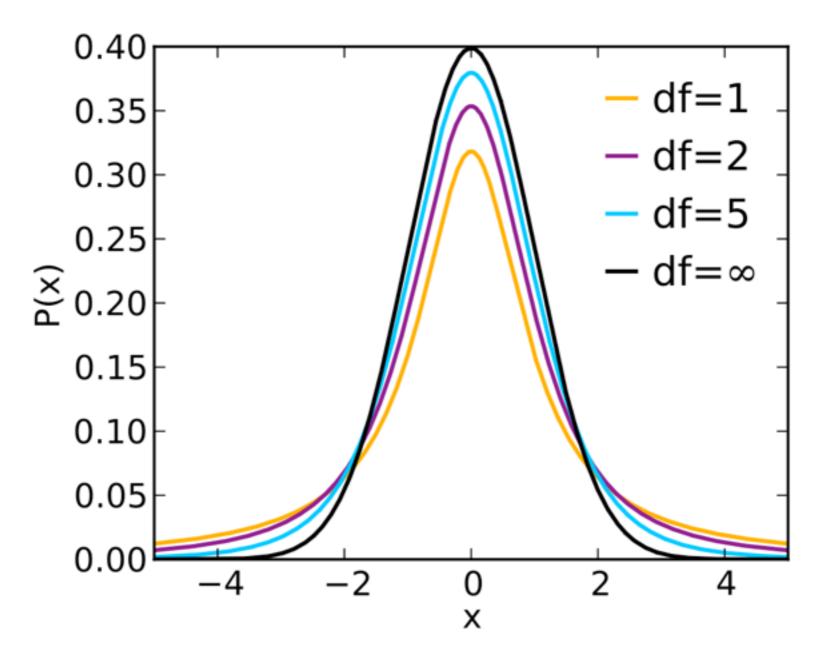
The important point is that we know what this quantity's distribution would look like under the null hypothesis. If the test statistic computed from the data is very unlikely to be drawn from that distribution, we can reject the null hypothesis.

Since the set of values are normally distributed, the Student's t-test is appropriate. Therefore, the test statistic is:

$$T=\frac{X_n-\mu}{\frac{S_n}{\sqrt{n}}} \qquad \text{$\mu$ is zero, the mean for the null hypothesis}$$
 where  $\bar{X}_n=\frac{1}{n}(X_1+\cdots+X_n)$  (sample mean) 
$$S_n^2=\frac{1}{n-1}\sum_{i=1}^n(X_i-\bar{X}_n)^2 \qquad \text{(sample variance)}$$

This T value has a Student's t distribution with N-1 degrees of freedom.

Derive the distribution of the test statistic under the null hypothesis: Student's t-distribution, n-1 df



Source: wikipedia.org

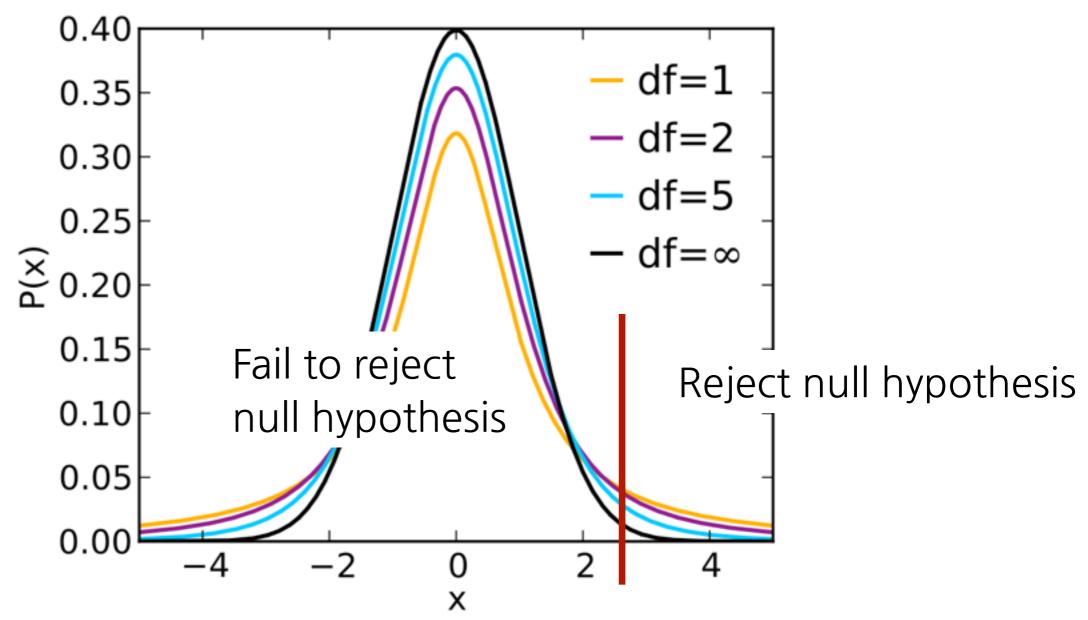
#### Determine the critical region for the test statistic.

Suppose N=6. Then we have 5 degrees of freedom. At the 95% significance level, the critical value for T is **2.447**.

Thus, if  $|T| \ge 2.447$ , we reject the null hypothesis at the 95% significance level.

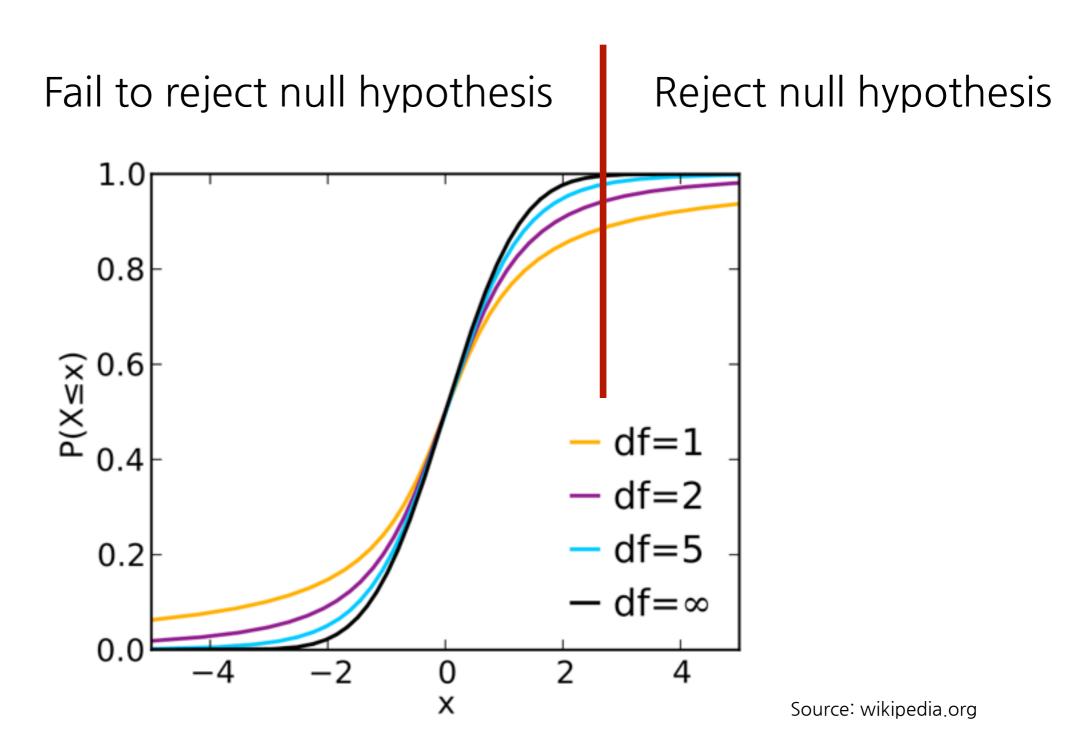
In other words, if the mean were really zero, | T | would be larger than 2.447 only 5% of the time.

Determine the critical region for the test statistic.



Source: wikipedia.org

#### Determine the critical region for the test statistic.



## Student's t-test example

Compute the observed value of the test statistic.

Suppose we calculated T=3.1

Decide to fail to reject or to reject the null hypothesis.

98.66% of the t-distribution with 5 df lies to the left of 3.1.

Therefore we can reject the null hypothesis at the 95% level.

Our p-value is 0.0134

### ttest() function

[h,p,ci,stats] = ttest(X)

Tests against the null hypothesis that the values in vector X are drawn from a normal distribution with zero mean.

h: true if the null hypothesis is rejected

p: p-value associated with the result

ci: 95% confidence intervals for the true value of the mean

stats: a structure that MATLAB can use for doing follow up tests, such as using multcompare.

### ttest() function

[h,p,ci,stats] = ttest(X, nullMean)

Tests against the null hypothesis that the values in vector X are drawn from a normal distribution with mean nullMean.

h: true if the null hypothesis is rejected

p: p-value associated with the result

ci: 95% confidence intervals for the true value of the mean

stats: a structure that MATLAB can use for doing follow up tests, such as using multcompare.

### ttest() function

[h,p,ci,stats] = ttest(X, nullMean, thresh)

Tests against the null hypothesis that the values in vector X are drawn from a normal distribution with mean nullMean.

h: true if the null hypothesis is rejected at threshold thresh (default is 0.05)

p: p-value associated with the result

ci: 95% confidence intervals for the true value of the mean

stats: a structure that MATLAB can use for doing follow up tests, such as using multcompare.

### Paired t-test

In a paired, t-test you make two measurements on the same subject, usually before and after. The null hypothesis is that the two measurements are drawn from distributions with equal means.

Internally, all this does is take the after-before difference for each subject and test against the null hypothesis that these differences have zero mean.

[h,p,ci,stats] = ttest(X, Y)

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### **Analysis of Variance**

Homework

## Analysis of Variance

Analysis of variance (ANOVA) is a set of statistical models and methods for partitioning variance in some quantity into components attributable to different sources.

ANOVA extends the t-test to multiple groups and allows you to test against the null hypothesis that some quantity measured from all of these groups have the same mean.

## Analysis of Variance

[p table stats] = anova1(X, groupNames)

Performs a one-way balanced ANOVA comparing the means of the columns of X.

Each column is a group, each row in that column is a data point. Thus the number of subjects in each group must be equal (i.e. balanced design).

p-value is the significance threshold associating with rejecting the **null hypothesis that all means are the same**.

## Analysis of Variance

The ANOVA test makes the following assumptions about the data:

All sample populations are normally distributed.

All sample populations have equal variance.

All observations are mutually independent.

The ANOVA test is known to be robust with respect to modest violations of the first two assumptions.

### Multiple comparisons

```
[c,m] = multcompare(stats);
```

When you have many groups, allows testing for differences between pairs of groups, without the rate of false positives increasing with each comparison.

In an N-way analysis of variance, you have a single output measurement for each subject, and each subject is described by N factors.

The aim is to determine which of these N factors (or interactions among these factors) affect the output quantity.

Works fine with **unbalanced designs** as well, meaning some groups can have more data points than others.

Each subject/trial/data point etc. is described by a single output measurement Y. It also belongs to one of several groups within each factor.

**Example:** Each data point represents one mouse.

- Output quantity: score on some behavioral assay
- Factor 1: Age group (Young, Old)
- Factor 2: Drug treatment (Control, DrugA, DrugB)

#### Main effects:

- Does the age group affect the score?
- Does the drug treatment group affect the score?

#### Interaction effects:

 Does the age group affect the score differently depending on what drug group a mouse is in? (Equivalently, vice versa?)

```
assayScore = [24 \ 101 \ 56 \ ...]
ageGroup = {'young', 'old', 'young', ... }
treatmentGroup = {'control', 'drugA', 'drugA' ...}
[p t stats terms] = anovan(assayScore, ...
   {ageGroup treatmentGroup}, ...
   'varnames', {'Age Group','Treatment Group'}, ...
   'model', 'interaction');
```

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Analysis of Variance

#### Homework

# Homework

# Statistical Test Reference

## Two sample t-tests: ttest2()

You measure some quantity for two separate groups of subjects, and you want to know whether the means are different between the two groups.

Need to estimate whether the two groups of measurements have equal variances. Is one group more variable than the other?

```
[h,p,ci,stats] = ttest2(X, Y);
```

- Assumes equal variances

```
[h, p, ci, stats] = ttest2(X,Y,thresh,tail,'unequal');
```

- Assumes unequal variances

### Two-tailed versus one-tailed

**Two tailed t-test:** tests the alternative hypothesis that the mean is different from zero, in either direction. You almost always want this one.

One tailed t-test: tests the alternative hypothesis that the mean is different from zero in a pariticular direction (i.e. greater than OR less than zero). This effectively halves your p-value, so people are often skeptical when you use this.

# Tests for normality: Chi-square

h = chi2gof(x)

Performs a chi-square goodness-of-fit test of the default null hypothesis that the data in vector x are a random sample from a normal distribution with mean and variance estimated from x, against the alternative that the data are not normally distributed with the estimated mean and variance.

## Lilliefors test for normality

h = lillietest(x)

Performs a Lilliefors test of the default null hypothesis that the sample in vector x comes from a distribution in the normal family, against the alternative that it does not come from a normal distribution.

### Rank-sum tests

Known as Mann-Whitney U or Wilcoxon rank sum.

Assesses whether quantities in one group tend to be higher than the other.

Doesn't require data to be normal, unlike the t-test.

```
p = ranksum(x, y)
```

## Sign-rank test

Operates analogously to the t-test or paired t-test, assessing whether there a set of numbers has a median different from zero or whether there is a difference in medians between paired measurements.

Doesn't require data to be normal.

```
p = signrank(x)
p = signrank(x,y)
```

## Chi-square variance test

[h, p] = vartest(X, v)

Performs a chi-square test of the null hypothesis that the samples in vector x comes from a normal distribution with variance v, against the alternative that X comes from a normal distribution with a different variance.

Data must be normal.

## Compare variances of two groups

[h, p, ci] = vartest2(X,Y)

Performs an F test of the hypothesis that two independent samples, in the vectors X and Y, come from normal distributions with the same variance, against the alternative that they come from normal distributions with different variances.

ci is a 95% confidence interval for the true variance ratio var(X) / var(Y)

Data must be normal