



Centre for
Tropical Livestock
Genetics and Health

Analysing sources of phenotypic variation with linear models

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Sources of phenotypic variation with linear models

Learning objectives:

- Introduction to linear models
- Parts of a model
 - Equation
 - Effects – fixed and random
 - Expectations, assumptions, limitations
- Developing a model
- Matrix format



$$y_i = \mu + F_i + bx_i + e_i$$

Workhorse of statistical analysis of many kinds of data

Central tool in quantitative genetics

Very versatile & powerful

Note: every model is an assumed simplification of reality

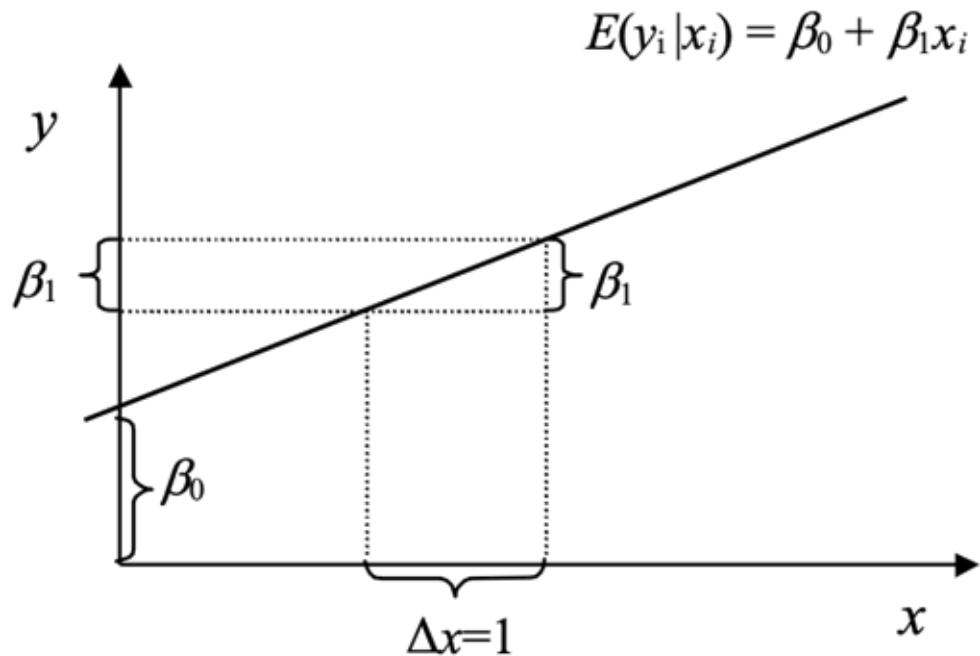


Figure 7.2 Interpretation of parameters for simple linear regression

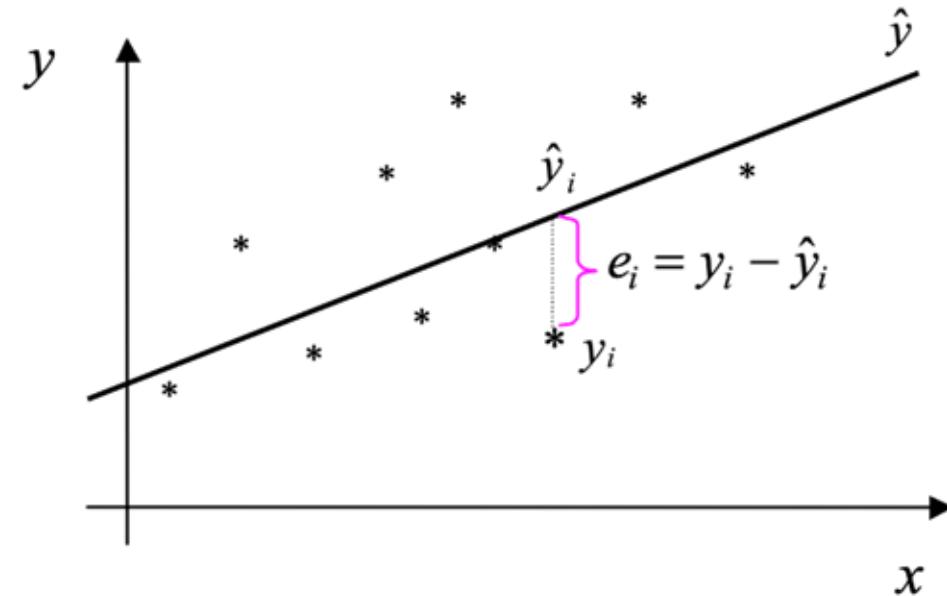


Figure 7.4 Estimated or fitted line of the simple linear regression



Parts of a model

1. The equation
2. Expectations and variance-covariance matrices of random variables
3. Assumptions, restrictions and limitations

A linear model is not complete without all parts

Statistical procedures and strategies are only determined based on a complete model



The equation

$$y_i = \mu + F_i + bx_i + e_i$$

y_i - Dependent variable

μ - Intercept (population reference value)

F_i - Factor with levels

bx_i - Covariate

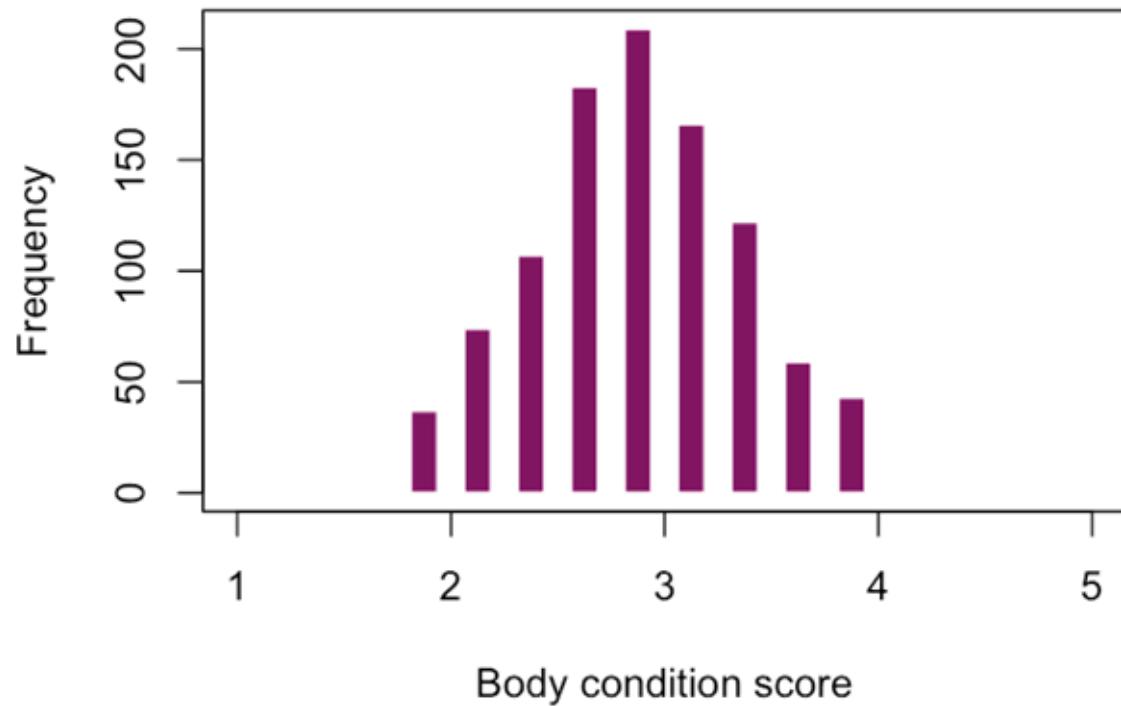
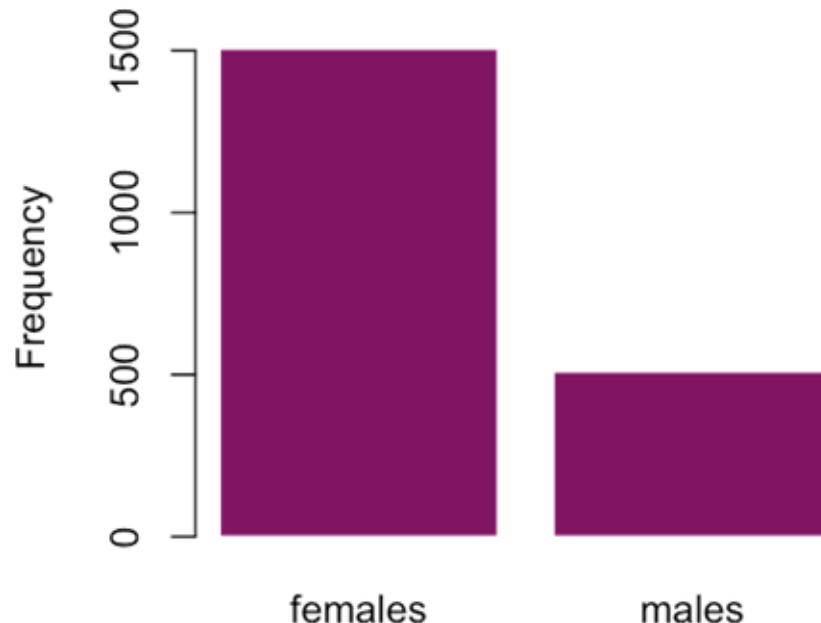
Fixed or random?

e_i - Residual

(environmental effects/noise, other effects/model misspecification, data errors, ...)

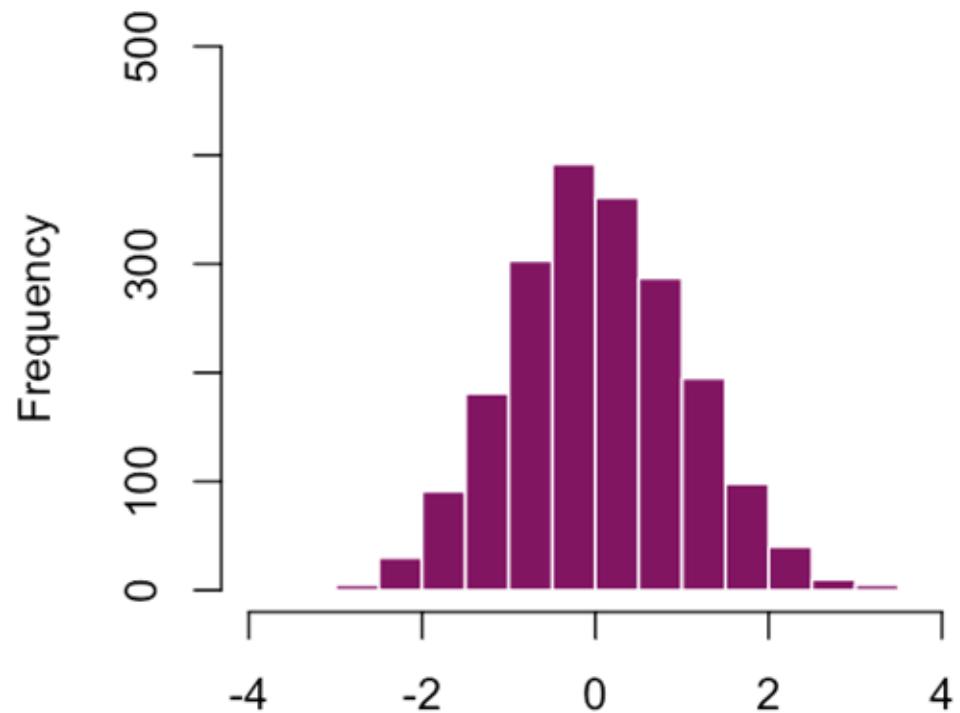


Discrete





Continuous





Fixed vs. random effects

	Fixed	Random
Number of levels	Small or limited to a fixed number	Large
Inferences	Limited to that set of levels	Going to be made to an entire population of conceptual levels
New experiment made	Same factor levels	Completely different levels
Levels of a factor selection	Selected among possible available levels	Sampled from an infinitely large population
Number of records per parameter	Many records	Not many ... with many records, random effect behaves practically as a fixed effect
	Regressions of a continuous factor	



Fixed vs. random effects

Diet (ex. diets A, B and C)	Animal
Breed (ex. Holstein, Angus, Jersey)	Season (ex. Wet and dry seasons)
Contemporary group	Permanent environment



Fixed vs. random effects

Diet (ex. diets A, B and C) - fixed	Animal
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Fixed vs. random effects

Diet (ex. diets A, B and C) - fixed ✓ Limited to a small, fixed number of levels ✓ Inferences limited to the sets of levels	Animal
Breed (ex. Holstein, Angus, Jersey)	Season (ex. Wet and dry seasons)
Contemporary group	Permanent environment



Fixed vs. random effects

Diet (ex. diets A, B and C) - fixed <ul style="list-style-type: none">✓ Limited to a small, fixed number of levels✓ Inferences limited to the sets of levels	Animal - random <ul style="list-style-type: none">✓ Inferences going to be made to an entire population of conceptual levels
Breed (ex. Holstein, Angus, Jersey) – fixed <ul style="list-style-type: none">✓ Limited to a small, fixed number of levels✓ Inferences limited to the sets of levels✓ Levels of the factor selected among possible available levels	Season (ex. Wet and dry seasons) - fixed <ul style="list-style-type: none">✓ Limited to a small, fixed number of levels✓ Inferences limited to the sets of levels✓ New experiment will have same levels
Contemporary group - fixed <ul style="list-style-type: none">✓ Inferences going to be made to an entire population of conceptual levels✓ New experiment made has completely different levels	Permanent environment - random <ul style="list-style-type: none">✓ Number of levels is large



Expectations, assumptions and limitations

Which data was sampled or collected?

Were individuals randomly selected, or met some requirement?

Did data arise from many environments, at random, or chosen?



How to develop a model

Information in literature on what should be included in model

Discuss with others (colleagues, industry, etc. – not everyone will agree what is relevant, which effects are fixed or random)

Test models and change as evidence accumulates

Justify your model

Consider non-linear or other types of models if appropriate



Developing a model – example for beef calf weights

Weights on beef calves taken at 200 days of age are shown in the table below:

Males	Females
198	187
211	194
220	202
	185



Developing a model – example for beef calf weights

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y_{ij}
 y_{ij} 200-day weight



Developing a model – example for beef calf weights

Weights on beef calves taken at 200 days of age are shown in the table below:

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211	194
220	202
	185

$$y_{ij} = s_i$$

y_{ij} 200-day weight

s_i effect of sex of calf (fixed effect)



Developing a model – example for beef calf weights

Weights on beef calves taken at 200 days of age are shown in the table below:

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198	187
211	194
220	202
	185

$$y_{ij} = s_i + c_j$$

y_{ij} 200-day weight

s_i effect of sex of calf (fixed effect)

c_j effect of calf (random effect)



Developing a model – example for beef calf weights

Weights on beef calves taken at 200 days of age are shown in the table below:

Males	Females
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$$y_{ij} = s_i + c_j + e_{ij}$$

y_{ij} 200-day weight

s_i effect of sex of calf (fixed effect)

c_j effect of calf (random effect)

e_{ij} residual, or unexplained, variation (random effect)



Developing a model – example for beef calf weights

Expectations

$$E(y_{ij}) = s_i$$

$$E(c_j) = 0$$

$$E(e_{ij}) = 0$$

$$Var(c_j) = \sigma_c^2$$

$$Var(e_{ij}) = \sigma_e^2$$

$$Cov(c_j, c_{j'}) = 0$$

$$Cov(e_{ij}, e_{ij'}) = 0$$

$$Cov(e_j, e_{i'j'}) = 0$$

$$y_{ij} = s_i + c_j + e_{ij}$$

y_{ij} 200-day weight

s_i effect of sex of calf (fixed effect)

c_j effect of calf (random effect)

e_{ij} residual, or unexplained, variation (random effect)



Developing a model – example for beef calf weights

Assumptions and limitations

- Calves are the same breed
- Reared in the same environment and time period
- Maternal effects ignored
- Calf effects contain all genetic effects (direct and maternal)
- Weights were accurately recorded at 200 days of age

$$y_{ij} = s_i + c_j + e_{ij}$$

y_{ij} 200-day weight

s_i effect of sex of calf (fixed effect)

c_j effect of calf (random effect)

e_{ij} residual, or unexplained, variation (random effect)



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Matrix algebra



$$y_{ij} = \mu_i + a_i + e_{ij}$$



$$\mathbf{y} = \mathbf{X}\mathbf{b} + \mathbf{Z}\mathbf{a} + \mathbf{e}$$

$$y_{11} = \mu_1 + a_1 + e_{11}$$

$$y_{21} = \mu_2 + a_2 + e_{21}$$

$$y_{31} = \mu_3 + a_3 + e_{31}$$



$$\begin{bmatrix} y_{11} \\ y_{21} \\ y_{31} \end{bmatrix} = \begin{bmatrix} x_{11} & x_{12} \\ x_{21} & x_{22} \end{bmatrix} \begin{bmatrix} b_{11} \\ b_{21} \end{bmatrix} + \begin{bmatrix} z_{11} & z_{12} & z_{13} \\ z_{21} & z_{22} & z_{23} \\ z_{31} & z_{32} & z_{33} \end{bmatrix} \begin{bmatrix} a_{11} \\ a_{21} \\ a_{31} \end{bmatrix} + \begin{bmatrix} e_{11} \\ e_{21} \\ e_{31} \end{bmatrix}$$

Efficient

- Data storage, and computations run
- A system of linear models can be written in one matrix equation



A rectangular array of numbers

$$\mathbf{M} = \begin{bmatrix} m_{11} & m_{12} & m_{13} \\ m_{21} & m_{22} & m_{23} \\ m_{31} & m_{32} & m_{33} \end{bmatrix}$$



A rectangular array of numbers

$$M = \begin{bmatrix} m_{11} & m_{12} & m_{13} \\ m_{21} & m_{22} & m_{23} \\ m_{31} & m_{32} & m_{33} \end{bmatrix}$$

↑
Columns

← Rows



A rectangular array of numbers

$$M = \begin{bmatrix} m_{11} & m_{12} & m_{13} \\ m_{21} & m_{22} & m_{23} \\ m_{31} & m_{32} & m_{33} \end{bmatrix}$$

First row

Second column

Rows

Columns

The diagram shows a 3x3 matrix M with elements m_{ij}. A green arrow points down from the top row to the element m₁₂, labeled 'First row'. A green arrow points right from the first column to the element m₁₂, labeled 'Second column'. A blue arrow points left from the second column to the matrix, labeled 'Rows'. A red arrow points up from the bottom row to the matrix, labeled 'Columns'.



A rectangular array of numbers

Column vector

$$\mathbf{c} = \begin{bmatrix} c_{11} \\ c_{21} \\ c_{31} \end{bmatrix}$$

$$\mathbf{M} = \begin{bmatrix} m_{11} & m_{12} & m_{13} \\ m_{21} & m_{22} & m_{23} \\ m_{31} & m_{32} & m_{33} \end{bmatrix}$$

First row

Second column

Rows

Row vector

Columns

$\mathbf{r} = [r_{11} \quad r_{12} \quad r_{13}]$

Scalar $s = [s_{11}]$



A rectangular array of numbers

Column vector

$$M = \begin{bmatrix} -20 & 4 & 6 \\ 9 & 60 & -2 \\ 33 & -15 & 8 \end{bmatrix}$$

$$c = \begin{bmatrix} 2.5 \\ 5.0 \\ -1.2 \end{bmatrix}$$

Row vector

$$r = [3 \quad 9 \quad 11]$$

$$E = \begin{bmatrix} x & y + 1 & \sqrt{x - y} \\ (m + n)/n & a^2 & \log d \end{bmatrix}$$

Scalar $s = [13]$



A rectangular array of numbers

Matrix

Naming convention:

Matrix – boldface & uppercase

Vector – boldface & lowercase

Scalar - lowercase

Column vector

$$\mathbf{c} = \begin{bmatrix} 2.5 \\ 5.0 \\ -1.2 \end{bmatrix} \quad 3 \times 1$$

$$\mathbf{M} = \begin{bmatrix} -20 & 4 & 6 \\ 9 & 60 & -2 \\ 33 & -15 & 8 \end{bmatrix} \quad \begin{matrix} \text{order} \\ 3 \times 3 \end{matrix}$$

Row vector

$$\mathbf{r} = [3 \quad 9 \quad 11] \quad 1 \times 3$$

$$\mathbf{E} = \begin{bmatrix} x & y + 1 & \sqrt{x - y} \\ (m + n)/n & a^2 & \log d \end{bmatrix} \quad 2 \times 3$$

Scalar $s = [13]$ 1×1



Information on matrices that we need this week

- Special matrices
- Addition
- Multiplication
- Transpose
- Inversion



Information on matrices that we need this week

- Special matrices
- Addition
- Multiplication
- Transpose
- Inversion

Bonus!

Other helpful matrix operations to know
for animal breeding

Slides included at the end

Markdown file with how to do matrix
operations in R



Special square matrices

Square – equal
number of rows and
columns

$$s = \begin{bmatrix} 4 & 7 & 0 \\ 0 & -5 & 3 \\ -7 & 3 & 11 \end{bmatrix}$$



Special square matrices

Square – equal
number of rows and
columns

$$s = \begin{bmatrix} 4 & 7 & 0 \\ 0 & -5 & 3 \\ -7 & 3 & 11 \end{bmatrix}$$

Diagonal – all off-
diagonal elements are
zero

$$d = \begin{bmatrix} 9 & 0 & 0 \\ 0 & 3 & 0 \\ 0 & 0 & -1 \end{bmatrix}$$



Square – equal
number of rows and
columns

$$S = \begin{bmatrix} 4 & 7 & 0 \\ 0 & -5 & 3 \\ -7 & 3 & 11 \end{bmatrix}$$

Diagonal – all off-
diagonal elements are
zero

$$D = \begin{bmatrix} 9 & 0 & 0 \\ 0 & 3 & 0 \\ 0 & 0 & -1 \end{bmatrix}$$

Special square matrices

Identity – diagonal
matrix, where all
diagonal elements are
1

$$I = \begin{bmatrix} 1 & 0 & 0 \\ 0 & 1 & 0 \\ 0 & 0 & 1 \end{bmatrix}$$



Two matrices can be added together if they have the same number of rows and columns, ie. They are of the same order and conformable

$$c_{ij} = a_{ij} + b_{ij}$$

$$\mathbf{C} = \mathbf{A} + \mathbf{B} = \begin{bmatrix} 9 & 2 \\ -11 & 5 \end{bmatrix} + \begin{bmatrix} 13 & -7 \\ 8 & 4 \end{bmatrix}$$

$$= \begin{bmatrix} 9 + 13 & 2 + (-7) \\ -11 + 8 & 5 + 4 \end{bmatrix}$$

$$= \begin{bmatrix} 22 & -5 \\ -3 & 9 \end{bmatrix}$$



Multiplication

Two matrices can be multiplied if the number of columns in the first matrix equals the number of rows in the second

The product matrix has an order of the number of rows of the first matrix by the number of columns of the second

$$c_{ij} = \sum_{j=i}^m \sum_{i=1}^n \sum_{k=1}^z a_{ik} b_{kj}$$

$$C = EG = \begin{bmatrix} -1 & 5 & 3 \\ 6 & -2 & 4 \\ 4 & -3 & 6 \end{bmatrix} \begin{bmatrix} 4 & -5 \\ 2 & 3 \\ -6 & 1 \end{bmatrix}$$

$$= \begin{bmatrix} -1(4) + 5(2) + 3(-6) & -1(-5) + 5(3) + 3(1) \\ 6(4) + (-2)(2) + 4(-6) & 6(-5) + (-2)(3) + 4(1) \\ 4(4) + (-3)(2) + 6(-6) & 4(-5) + (-3)(3) + 6(1) \end{bmatrix}$$

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For J , with order $i \times k$, the transpose, J' has order $k \times i$

$$J = \begin{bmatrix} 5 & 3 \\ -1 & 4 \\ 6 & -2 \end{bmatrix}$$

$$J' = J^T = \begin{bmatrix} 5 & -1 & 6 \\ 3 & 4 & -2 \end{bmatrix}$$



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Transpose

For J , with order $i \times k$, the transpose, J' has order $i \times k$

$$J = \begin{bmatrix} 5 & 3 \\ -1 & 4 \\ 6 & -2 \end{bmatrix}$$

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A matrix is not equal to its transpose



For J , with order $i \times k$, the transpose, J' has order $i \times k$

$$J = \begin{bmatrix} 5 & 3 \\ -1 & 4 \\ 6 & -2 \end{bmatrix}$$

$$J' = J^T = \begin{bmatrix} 5 & -1 & 6 \\ 3 & 4 & -2 \end{bmatrix}$$

$$W = \begin{bmatrix} 7 & 2 & -4 \\ 2 & 3 & 0 \\ -4 & 0 & -9 \end{bmatrix}$$

$$W' = \begin{bmatrix} 7 & 2 & -4 \\ 2 & 3 & 0 \\ -4 & 0 & -9 \end{bmatrix}$$

A matrix is not equal to its transpose ... Unless it is a symmetric matrix



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A matrix is not equal to its transpose ... Unless it is a symmetric matrix



When \mathbf{W}^{-1} is multiplied by \mathbf{W} ,
an identity matrix (\mathbf{I}) is the
product

Only square matrices can be
inverted

For diagonal matrix, \mathbf{Y} , the
inverse is the reciprocal of the
diagonal elements

$$\mathbf{W}^{-1}\mathbf{W} = \mathbf{I} = \begin{bmatrix} 1 & 0 & 0 \\ 0 & 1 & 0 \\ 0 & 0 & 1 \end{bmatrix}$$

$$\mathbf{Y} = \begin{bmatrix} 5 & 0 & 0 \\ 0 & -10 & 0 \\ 0 & 0 & 2 \end{bmatrix}$$

$$\mathbf{Y}^{-1} = \begin{bmatrix} \frac{1}{5} & 0 & 0 \\ 0 & -\frac{1}{10} & 0 \\ 0 & 0 & \frac{1}{2} \end{bmatrix}$$



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Inverse

$$\mathbf{Z} = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix}$$



Calculate the determinant (the difference between the product of the two diagonal elements and the two off-diagonal elements)

$$\mathbf{Z} = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix}$$

$$\mathbf{Z}^{-1} = \frac{1}{z_{11}z_{22} - z_{12}z_{21}} \begin{bmatrix} z_{22} & -z_{12} \\ -z_{21} & z_{11} \end{bmatrix}$$



Calculate the determinant (the difference between the product of the two diagonal elements and the two off-diagonal elements)

Reverse diagonal elements,
multiplying off-diagonal
elements by -1

$$Z = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix}$$

$$Z^{-1} = \frac{1}{z_{11}z_{22} - z_{12}z_{21}} \begin{bmatrix} z_{22} & -z_{12} \\ -z_{21} & z_{11} \end{bmatrix}$$



Calculate the determinant (the difference between the product of the two diagonal elements and the two off-diagonal elements)

Reverse diagonal elements,
multiplying off-diagonal
elements by -1

Divide all elements by the
determinant

$$Z = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix}$$

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Divide all elements by the determinant

Inversion – 2x2 matrix example

$$Z = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix} = \begin{bmatrix} 3 & -1 \\ 2 & 5 \end{bmatrix}$$

$$Z^{-1} = \frac{1}{z_{11}z_{22} - z_{12}z_{21}} \begin{bmatrix} z_{22} & -z_{12} \\ -z_{21} & z_{11} \end{bmatrix}$$

$$= \frac{1}{3(5) - (-1)(2)} \begin{bmatrix} 5 & -(-1) \\ -2 & 3 \end{bmatrix}$$

$$= \begin{bmatrix} 0.29 & 0.06 \\ -0.12 & 0.18 \end{bmatrix}$$



$$y_{ij} = \mu_i + a_i + e_{ij}$$

y_{ij} - record j of the i^{th} animal, dependent/response variable

μ_i - fixed environmental effects of i^{th} animal (year of birth, sex, etc.)

a_i - random additive genetic effect of i^{th} animal

e_{ij} - residual (environmental effects, noise, other effects, model misspecification) effect for record j of the i^{th} animal



The equation in matrix format

$$y_{ij} = \mu_i + a_i + e_{ij}$$



$$\mathbf{y} = \mathbf{X}\mathbf{b} + \mathbf{Z}\mathbf{a} + \mathbf{e}$$

y vector of observations

b vector of fixed effects (year of birth, sex, etc.)

a vector of random additive effects

e vector of random residual effects

X and **Z** incidence matrices relating records to fixed and random effects



$$y_{ij} = \mu_i + a_i + e_{ij}$$



$$\mathbf{y} = \mathbf{X}\mathbf{b} + \mathbf{Z}\mathbf{a} + \mathbf{e}$$

y

vector of observations

b

vector of fixed effects (year of birth, sex, etc.)

a

vector of random additive genetic effects

e

vector of random residual effects

X and **Z** incidence matrices relating records to fixed and random effects

The equation in matrix format

$$y_{11} = \mu_1 + a_1 + e_{11}$$

$$y_{21} = \mu_2 + a_2 + e_{21}$$

$$y_{31} = \mu_3 + a_3 + e_{31}$$



$$\begin{bmatrix} y_{11} \\ y_{21} \\ y_{31} \end{bmatrix} = \begin{bmatrix} x_{11} & x_{12} \\ x_{21} & x_{22} \end{bmatrix} \begin{bmatrix} b_{11} \\ b_{21} \end{bmatrix} + \begin{bmatrix} z_{11} & z_{12} & z_{13} \\ z_{21} & z_{22} & z_{23} \\ z_{31} & z_{32} & z_{33} \end{bmatrix} \begin{bmatrix} a_{11} \\ a_{21} \\ a_{31} \end{bmatrix} + \begin{bmatrix} e_{11} \\ e_{21} \\ e_{31} \end{bmatrix}$$



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In matrix format, with fixed effects only

$$\mathbf{y} = \mathbf{X}\mathbf{b} + \mathbf{e}$$

$$[\mathbf{X}'\mathbf{X}][\widehat{\mathbf{b}}] = [\mathbf{X}'\mathbf{y}]$$

$$[\widehat{\mathbf{b}}] = [\mathbf{X}'\mathbf{X}]^{-1}[\mathbf{X}'\mathbf{y}]$$



In matrix format, mixed effects model

$$\mathbf{y} = \mathbf{X}\mathbf{b} + \mathbf{Z}\mathbf{a} + \mathbf{e}$$

$$\begin{bmatrix} \mathbf{X}'\mathbf{X} & \mathbf{X}'\mathbf{Z} \\ \mathbf{Z}'\mathbf{X} & \mathbf{Z}'\mathbf{Z} + \mathbf{I} \frac{\sigma_e^2}{\sigma_a^2} \end{bmatrix} \begin{bmatrix} \hat{\mathbf{b}} \\ \mathbf{a} \end{bmatrix} = \begin{bmatrix} \mathbf{X}'\mathbf{y} \\ \mathbf{Z}'\mathbf{y} \end{bmatrix}$$

$$\begin{bmatrix} \hat{\mathbf{b}} \\ \mathbf{a} \end{bmatrix} = \begin{bmatrix} \mathbf{X}'\mathbf{X} & \mathbf{X}'\mathbf{Z} \\ \mathbf{Z}'\mathbf{X} & \mathbf{Z}'\mathbf{Z} + \mathbf{I} \frac{\sigma_e^2}{\sigma_a^2} \end{bmatrix}^{-1} \begin{bmatrix} \mathbf{X}'\mathbf{y} \\ \mathbf{Z}'\mathbf{y} \end{bmatrix}$$



Mixed effects model example

Ex. Weights on beef calves taken at 200 days of age.

Males	Females
198	187
211	194
220	202
	185

$$y_{ij} = s_i + c_j + e_{ij}$$

y_{ij} 200-day weight

s_i effect of sex of calf (fixed effect)

c_j effect of calf (random effect)

e_{ij} residual, or unexplained, variation
(random effect)



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Mixed effects model example

$$\mathbf{y} = \mathbf{X}\mathbf{s} + \mathbf{Z}\mathbf{c} + \mathbf{e}$$

\mathbf{y} vector of weights

\mathbf{s} vector of effect of sex

\mathbf{c} vector of effect of calf

\mathbf{e} vector of random residual effects

\mathbf{X} and \mathbf{Z} incidence matrices



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\mathbf{X} and \mathbf{Z} incidence matrices

$$\begin{bmatrix} \hat{\mathbf{s}} \\ \mathbf{c} \end{bmatrix} = \begin{bmatrix} \mathbf{X}'\mathbf{X} & \mathbf{X}'\mathbf{Z} \\ \mathbf{Z}'\mathbf{X} & \mathbf{Z}'\mathbf{Z} + I \frac{\sigma_e^2}{\sigma_c^2} \end{bmatrix}^{-1} \begin{bmatrix} \mathbf{X}'\mathbf{y} \\ \mathbf{Z}'\mathbf{y} \end{bmatrix}$$



Males	Females
198	187
211	194
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Constructing matrices

$$\mathbf{y} = \mathbf{X}\mathbf{s} + \mathbf{Z}\mathbf{c} + \mathbf{e}$$

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198	187
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$$\mathbf{y} = \begin{bmatrix} 198 \\ 211 \\ 220 \\ 187 \\ 194 \\ 202 \\ 185 \end{bmatrix}$$

Constructing matrices

$$\mathbf{y} = \mathbf{X}\mathbf{s} + \mathbf{Z}\mathbf{c} + \mathbf{e}$$

\mathbf{y} vector of weights

\mathbf{s} vector of effect of sex

\mathbf{c} vector of effect of calf

\mathbf{e} vector of random residual effects

\mathbf{X} and \mathbf{Z} incidence matrices

$$\begin{bmatrix} \hat{\mathbf{s}} \\ \mathbf{c} \end{bmatrix} = \begin{bmatrix} \mathbf{X}'\mathbf{X} & \mathbf{X}'\mathbf{Z} \\ \mathbf{Z}'\mathbf{X} & \mathbf{Z}'\mathbf{Z} + I \frac{\sigma_e^2}{\sigma_c^2} \end{bmatrix}^{-1} \begin{bmatrix} \mathbf{X}'\mathbf{y} \\ \mathbf{Z}'\mathbf{y} \end{bmatrix}$$



Males	Females
198	187
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$$\mathbf{y} = \begin{bmatrix} 198 \\ 211 \\ 220 \\ 187 \\ 194 \\ 202 \\ 185 \end{bmatrix} \quad \mathbf{X} = \begin{bmatrix} 1 & 1 \\ 1 & 1 \\ 1 & 1 \\ 1 & 0 \\ 1 & 0 \\ 1 & 0 \\ 1 & 0 \end{bmatrix}$$

weights mean sex

Constructing matrices

$$\mathbf{y} = \mathbf{X}\mathbf{s} + \mathbf{Z}\mathbf{c} + \mathbf{e}$$

\mathbf{y} vector of weights

\mathbf{s} vector of effect of sex

\mathbf{c} vector of effect of calf

\mathbf{e} vector of random residual effects

\mathbf{X} and \mathbf{Z} incidence matrices

$$\begin{bmatrix} \hat{\mathbf{s}} \\ \mathbf{c} \end{bmatrix} = \begin{bmatrix} \mathbf{X}'\mathbf{X} & \mathbf{X}'\mathbf{Z} \\ \mathbf{Z}'\mathbf{X} & \mathbf{Z}'\mathbf{Z} + \mathbf{I} \frac{\sigma_e^2}{\sigma_c^2} \end{bmatrix}^{-1} \begin{bmatrix} \mathbf{X}'\mathbf{y} \\ \mathbf{Z}'\mathbf{y} \end{bmatrix}$$



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$$\mathbf{y} = \begin{bmatrix} 198 \\ 211 \\ 220 \\ 187 \\ 194 \\ 202 \\ 185 \end{bmatrix} \quad \mathbf{X} = \begin{bmatrix} 1 & 1 \\ 1 & 1 \\ 1 & 1 \\ 1 & 0 \\ 1 & 0 \\ 1 & 0 \\ 1 & 0 \end{bmatrix} \quad \mathbf{Z} = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 1 \end{bmatrix}$$

weights mean sex calf 1 ... calf 7

Constructing matrices

$$\mathbf{y} = \mathbf{X}\mathbf{s} + \mathbf{Z}\mathbf{c} + \mathbf{e}$$

\mathbf{y} vector of weights

\mathbf{s} vector of effect of sex

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\mathbf{X} and \mathbf{Z} incidence matrices

$$\begin{bmatrix} \hat{\mathbf{s}} \\ \mathbf{c} \end{bmatrix} = \begin{bmatrix} \mathbf{X}'\mathbf{X} & \mathbf{X}'\mathbf{Z} \\ \mathbf{Z}'\mathbf{X} & \mathbf{Z}'\mathbf{Z} + I \frac{\sigma_e^2}{\sigma_c^2} \end{bmatrix}^{-1} \begin{bmatrix} \mathbf{X}'\mathbf{y} \\ \mathbf{Z}'\mathbf{y} \end{bmatrix}$$



Males	Females
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$$y = \begin{bmatrix} 198 \\ 211 \\ 220 \\ 187 \\ 194 \\ 202 \\ 185 \end{bmatrix} \quad X = \begin{bmatrix} 1 & 1 \\ 1 & 1 \\ 1 & 1 \\ 1 & 0 \\ 1 & 0 \\ 1 & 0 \\ 1 & 0 \end{bmatrix} \quad Z = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 1 \end{bmatrix}$$

weights mean sex calf 1 ... calf 7

Constructing matrices

$$y = Xs + Zc + e$$

y vector of weights

s vector of effect of sex

c vector of effect of calf

e vector of random residual effects

X and Z incidence matrices

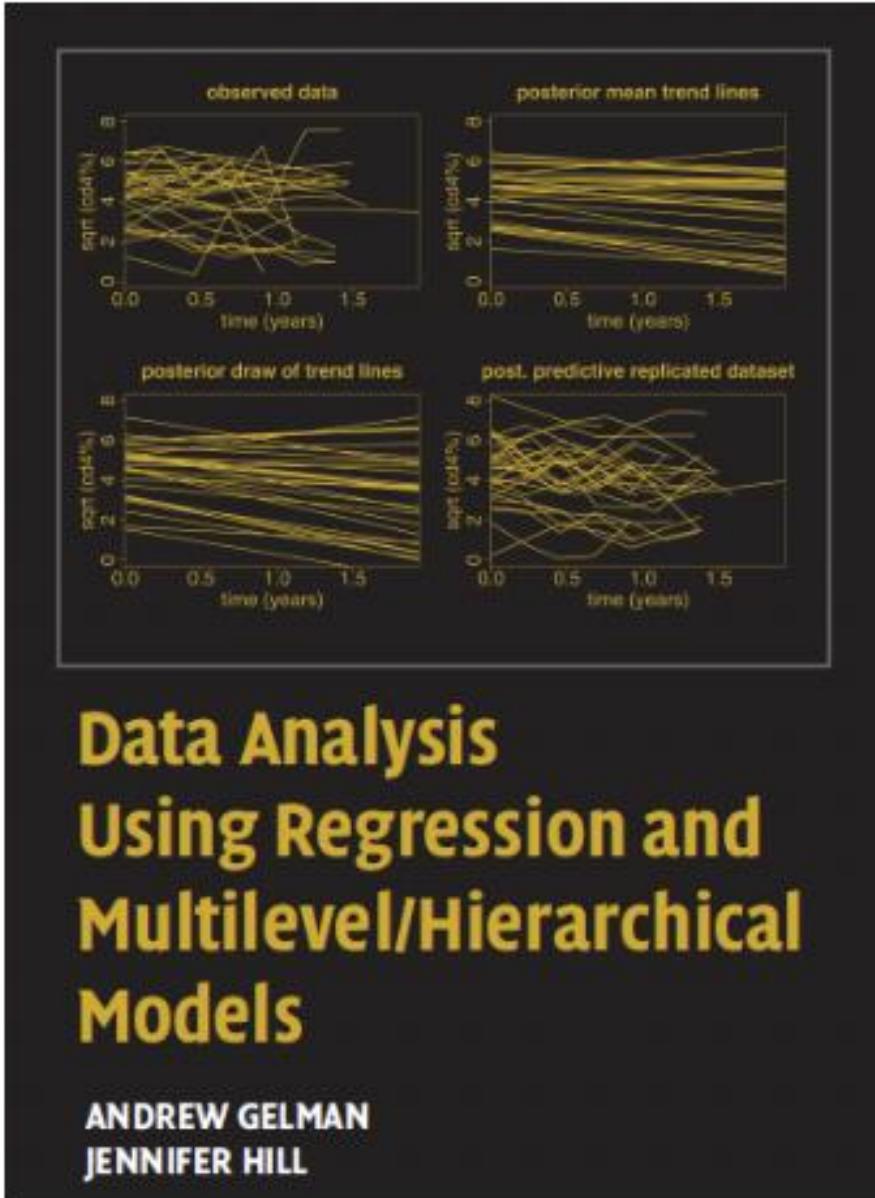
$$\begin{bmatrix} \hat{s} \\ c \end{bmatrix} = \begin{bmatrix} X'X & X'Z \\ Z'X & Z'Z + I \frac{\sigma_e^2}{\sigma_c^2} \end{bmatrix}^{-1} \begin{bmatrix} X'y \\ Z'y \end{bmatrix}$$

I = 7 by 7 incidence matrix (because random effect has seven levels (calves))

For this lecture, we will assume $\frac{\sigma_e^2}{\sigma_c^2}$ is known



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Study material



Sources of phenotypic variation with linear models

Learning objectives:

- Introduction to linear models
- Parts of a model
 - Equation
 - Effects – fixed and random
 - Expectations, assumptions, limitations
- Developing a model
- Matrix format



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Extra slides



Square – equal number of rows and columns

$$S = \begin{bmatrix} 4 & 7 & 0 \\ 0 & -5 & 3 \\ -7 & 3 & 11 \end{bmatrix}$$

Diagonal – all off-diagonal elements are zero

$$D = \begin{bmatrix} 9 & 0 & 0 \\ 0 & 3 & 0 \\ 0 & 0 & -1 \end{bmatrix}$$

Identity – diagonal matrix, where all diagonal elements are 1

$$I = \begin{bmatrix} 1 & 0 & 0 \\ 0 & 1 & 0 \\ 0 & 0 & 1 \end{bmatrix}$$

Special (square) matrices

Triangular

Lower triangular, all elements above the diagonal are zero

$$L = \begin{bmatrix} 7 & 0 & 0 \\ 2 & 3 & 0 \\ -4 & 15 & 9 \end{bmatrix}$$

Upper triangular, all elements above the diagonal are zero

$$U = \begin{bmatrix} 13 & -2 & 7 \\ 0 & 3 & 11 \\ 0 & 0 & -1 \end{bmatrix}$$

Symmetric – elements above the diagonal equal to the corresponding elements below the diagonal

$$B = \begin{bmatrix} 7 & 2 & -4 \\ 2 & 3 & 0 \\ -4 & 0 & -9 \end{bmatrix}$$



Two matrices can be added together if they have the same number of rows and columns, ie. They are of the same order and conformable

$$c_{ij} = a_{ij} + b_{ij}$$

$$\mathbf{C} = \mathbf{A} + \mathbf{B} = \begin{bmatrix} 9 & 2 \\ -11 & 5 \end{bmatrix} + \begin{bmatrix} 13 & -7 \\ 8 & 4 \end{bmatrix}$$

$$= \begin{bmatrix} 9 + 13 & 2 + (-7) \\ -11 + 8 & 5 + 4 \end{bmatrix}$$

$$= \begin{bmatrix} 22 & -5 \\ -3 & 9 \end{bmatrix}$$



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$$= \begin{bmatrix} 9 + 13 & 2 + (-7) \\ -11 + 8 & 5 + 4 \end{bmatrix}$$

$$= \begin{bmatrix} 22 & -5 \\ -3 & 9 \end{bmatrix}$$



A matrix can be subtracted from another if they have the same number of rows and columns, ie. They are of the same order and conformable

$$c_{ij} = a_{ij} - b_{ij}$$

$$\begin{aligned} C &= A - B = \begin{bmatrix} 9 & 2 \\ -11 & 5 \end{bmatrix} + \begin{bmatrix} 13 & -7 \\ 8 & 4 \end{bmatrix} \\ &= \begin{bmatrix} 9 - 13 & 2 - (-7) \\ -11 - 8 & 5 - 4 \end{bmatrix} \\ &= \begin{bmatrix} -4 & 9 \\ -19 & 1 \end{bmatrix} \end{aligned}$$



Two matrices can be multiplied if the number of columns in the first matrix equals the number of rows in the second

The product matrix has an order of the number of rows of the first matrix by the number of columns of the second

Multiplication

$$c_{ij} = \sum_{j=i}^m \sum_{i=1}^n \sum_{k=1}^z e_{ik} g_{kj}$$

$$C = EG = \begin{bmatrix} -1 & 5 & 3 \\ 6 & -2 & 4 \\ 4 & -3 & 6 \end{bmatrix} \begin{bmatrix} 4 & -5 \\ 2 & 3 \\ -6 & 1 \end{bmatrix}$$

$$= \begin{bmatrix} -1(4) + 5(2) + 3(-6) & -1(-5) + 5(3) + 3(1) \\ 6(4) + (-2)(2) + 4(-6) & 6(-5) + (-2)(3) + 4(1) \\ 4(4) + (-3)(2) + 6(-6) & 4(-5) + (-3)(3) + 6(1) \end{bmatrix}$$

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Multiplying a scalar with a matrix

Each element of the matrix is multiplied by the scalar

$$\begin{aligned} \mathbf{C} &= 4 \times \mathbf{E} = \mathbf{E} \times 4 = 4 \times \begin{bmatrix} -1 & 5 & 3 \\ 6 & -2 & 4 \\ 4 & -3 & 6 \end{bmatrix} \\ &= \begin{bmatrix} 4(-1) & 4(5) & 4(3) \\ 4(6) & 4(-2) & 4(4) \\ 4(4) & 4(-3) & 4(6) \end{bmatrix} \\ &= \begin{bmatrix} -4 & 20 & 12 \\ 24 & -8 & 16 \\ 16 & -12 & 24 \end{bmatrix} \end{aligned}$$



Multiplying a scalar with a matrix

Each element of the matrix is multiplied by the scalar

$$\begin{aligned} \mathbf{C} &= 4 \times \mathbf{E} = \mathbf{E} \times 4 = 4 \times \begin{bmatrix} -1 & 5 & 3 \\ 6 & -2 & 4 \\ 4 & -3 & 6 \end{bmatrix} \\ &= \begin{bmatrix} 4(-1) & 4(5) & 4(3) \\ 4(6) & 4(-2) & 4(4) \\ 4(4) & 4(-3) & 4(6) \end{bmatrix} \\ &= \begin{bmatrix} -4 & 20 & 12 \\ 24 & -8 & 16 \\ 16 & -12 & 24 \end{bmatrix} \end{aligned}$$



For **H** (order $r \times s$), and **E** (order $m \times n$), the direct product, **C**, has an order of $rm \times sn$

$$\mathbf{C} = \mathbf{H} \otimes \mathbf{E} = \begin{bmatrix} h_{11}\mathbf{E} & h_{12}\mathbf{E} \\ h_{21}\mathbf{E} & h_{22}\mathbf{E} \end{bmatrix}$$

$$\mathbf{C} = \begin{bmatrix} 4 & -5 \\ 2 & 3 \end{bmatrix} \otimes \begin{bmatrix} -1 & 5 & 3 \\ 6 & -2 & 4 \\ 4 & -3 & 6 \end{bmatrix}$$

$$= \begin{bmatrix} -4 & 20 & 12 & 5 & -25 & -15 \\ 24 & -8 & 16 & -30 & 10 & -20 \\ 16 & -12 & 24 & -20 & 15 & -30 \\ -2 & 10 & 6 & -3 & 15 & 9 \\ 12 & -4 & 8 & 18 & -6 & 12 \\ 8 & -6 & 12 & 12 & -9 & 18 \end{bmatrix}$$



Direct product of matrices – Kronecker product

For \mathbf{H} (order $r \times s$), and \mathbf{E} (order $m \times n$), the direct product, \mathbf{C} , has an order of $rm \times sn$

$$\mathbf{C} = \mathbf{H} \otimes \mathbf{E} = \begin{bmatrix} h_{11}\mathbf{E} & h_{12}\mathbf{E} \\ h_{21}\mathbf{E} & h_{22}\mathbf{E} \end{bmatrix}$$

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$$= \begin{bmatrix} -4 & 20 & 12 & 5 & -25 & -15 \\ 24 & -8 & 16 & -30 & 10 & -20 \\ 16 & -12 & 24 & -20 & 15 & -30 \\ -2 & 10 & 6 & -3 & 15 & 9 \\ 12 & -4 & 8 & 18 & -6 & 12 \\ 8 & -6 & 12 & 12 & -9 & 18 \end{bmatrix}$$



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For two matrices of the same
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Hadamard product

$$\mathbf{H} \odot \mathbf{X}$$
$$(\mathbf{H} \odot \mathbf{X})_{ij} = h_{ij} x_{ij}$$

$$\mathbf{H} \odot \mathbf{X} = \mathbf{X} \odot \mathbf{H}$$



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$$\mathbf{E} \odot \mathbf{X} = \begin{bmatrix} -1 & 5 & 3 \\ 6 & -2 & 4 \\ 4 & -3 & 6 \end{bmatrix} \odot \begin{bmatrix} 7 & 2 & -4 \\ 2 & 3 & 0 \\ -4 & 0 & -9 \end{bmatrix}$$



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Transpose

For J , with order $i \times k$, the transpose, J' has order $i \times k$

$$J = \begin{bmatrix} 5 & 3 \\ -1 & 4 \\ 6 & -2 \end{bmatrix}$$

$$J' = J^T = \begin{bmatrix} 5 & -1 & 6 \\ 3 & 4 & -2 \end{bmatrix}$$



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J is not equal to J'



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A matrix is not equal to its transpose ... Unless it is a symmetric matrix



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Dot product

The sum of the products of two equal length vectors

$$\mathbf{a} \cdot \mathbf{b} = \sum a_i b_i$$



The sum of the products of two equal length vectors

$$\mathbf{a} \cdot \mathbf{b} = \sum a_i b_i$$

$$\begin{aligned}\mathbf{a} &= [7, -2, 1] \\ \mathbf{b} &= [3, 5, -4]\end{aligned}$$

$$\begin{aligned}\mathbf{a} \cdot \mathbf{b} &= 7(3) + (-2)(5) + 1(-4) \\ &= 7\end{aligned}$$



The sum of the products of two equal length vectors

$$\mathbf{a} \cdot \mathbf{b} = \mathbf{ab}'$$

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Trace

The sum of the diagonal elements of a matrix

$$\mathbf{W} = \begin{bmatrix} w_{11} & w_{12} & w_{13} \\ w_{21} & w_{22} & w_{23} \\ w_{31} & w_{32} & w_{33} \end{bmatrix}$$

Scalar

$$tr(\mathbf{W}) = \sum w_{ii}$$



The sum of the diagonal elements of a matrix

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$$\begin{aligned} tr(\mathbf{W}) &= 7 + 3 + (-9) \\ &= 1 \end{aligned}$$



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When \mathbf{W}^{-1} is multiplied by \mathbf{W} ,
an identity matrix (\mathbf{I}) is the
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Only square matrices can be
inverted

For diagonal matrix, \mathbf{Y} , the
inverse is the reciprocal of the
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$$\mathbf{W}^{-1}\mathbf{W} = \mathbf{I} = \begin{bmatrix} 1 & 0 & 0 \\ 0 & 1 & 0 \\ 0 & 0 & 1 \end{bmatrix}$$

$$\mathbf{Y} = \begin{bmatrix} 5 & 0 & 0 \\ 0 & -10 & 0 \\ 0 & 0 & 2 \end{bmatrix}$$

$$\mathbf{Y}^{-1} = \begin{bmatrix} \frac{1}{5} & 0 & 0 \\ 0 & -\frac{1}{10} & 0 \\ 0 & 0 & \frac{1}{2} \end{bmatrix}$$



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Inversion – 2x2 matrix example

$$\mathbf{Z} = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix}$$



Calculate the determinant (the difference between the product of the two diagonal elements and the two off-diagonal elements)

Inversion – 2x2 matrix example

$$\mathbf{Z} = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix}$$

$$\mathbf{Z}^{-1} = \frac{1}{z_{11}z_{22} - z_{12}z_{21}} \begin{bmatrix} z_{22} & -z_{12} \\ -z_{21} & z_{11} \end{bmatrix}$$



Calculate the determinant (the difference between the product of the two diagonal elements and the two off-diagonal elements)

Reverse diagonal elements, multiplying off-diagonal elements by -1

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Inversion – 2x2 matrix example

$$Z = \begin{bmatrix} z_{11} & z_{12} \\ z_{21} & z_{22} \end{bmatrix} = \begin{bmatrix} 3 & -1 \\ 2 & 5 \end{bmatrix}$$

$$Z^{-1} = \frac{1}{z_{11}z_{22} - z_{12}z_{21}} \begin{bmatrix} z_{22} & -z_{12} \\ -z_{21} & z_{11} \end{bmatrix}$$

$$= \frac{1}{3(5) - (-1)(2)} \begin{bmatrix} 5 & -(-1) \\ -2 & 3 \end{bmatrix}$$

$$= \begin{bmatrix} 0.29 & 0.06 \\ -0.12 & 0.18 \end{bmatrix}$$



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Rank

The number of linearly independent rows or columns

A square matrix with rank equal to the number of rows or columns is **full rank**



$$\begin{aligned}2x_1 + 5x_2 + 2x_3 &= y_1 \\5x_1 + 3x_2 + 1x_3 &= y_2 \\-3x_1 + 2x_2 + 1x_3 &= y_3\end{aligned}$$

$$\begin{bmatrix} 2 & 5 & 2 \\ 5 & 3 & 1 \\ -3 & 2 & 1 \end{bmatrix} \begin{bmatrix} x_1 \\ x_2 \\ x_3 \end{bmatrix} = \begin{bmatrix} y_1 \\ y_2 \\ y_3 \end{bmatrix}$$

$$\mathbf{N}\mathbf{x} = \mathbf{y}$$

$$r(\mathbf{N}) = 2$$

The number of linearly independent rows or columns

A square matrix with rank equal to the number of rows or columns is **full rank**

In the example, \mathbf{x}' (vector of solutions) cannot be estimated due to lack of information (because a unique inverse does not exist for \mathbf{N} because it is not full rank)

If a square matrix is not full rank, the determinant is zero, and a unique inverse does not exist



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Generalised inverse

$$NN^{-}N = N$$

A **generalised inverse** can
be calculated for a
singular matrix

Generalised inverses are
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$$NN^{-}N = N$$

$$N = \begin{bmatrix} 2 & 5 & 2 \\ 5 & 3 & 1 \\ -3 & 2 & 1 \end{bmatrix}; A = \begin{bmatrix} 2 & 5 \\ 5 & 3 \end{bmatrix}$$



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Eigenvalues are useful in simplifying multivariate evaluations when transforming data

Sum of eigenvalues of a square matrix is equal to its trace (sum of diagonal elements)

Product of eigenvalues is equal to the determinant

For a symmetric matrix, the rank is equal to the number of non-zero eigenvalues



Eigenvalues and eigenvectors

If N is a square symmetric matrix:

$$N = UDU'$$

D is a diagonal matrix, the canonical form of N , containing eigenvalues of N , and U is an orthogonal matrix of the corresponding eigenvectors

$$\begin{aligned}|N - dI| &= 0 \\ Nu - du &= 0\end{aligned}$$

d is one of the eigenvalues of B , and u is the corresponding eigenvector



Cholesky decomposition

Symmetric positive definite matrices can be decomposed into a product of a lower triangular matrix and its transpose:

$$K = VV'$$

$$\begin{bmatrix} 9 & 3 & -6 \\ 3 & 5 & 0 \\ -6 & 0 & 21 \end{bmatrix} = \begin{bmatrix} v_{11} & 0 & 0 \\ v_{21} & v_{22} & 0 \\ v_{31} & v_{32} & v_{33} \end{bmatrix} \begin{bmatrix} v_{11} & v_{21} & v_{31} \\ 0 & v_{22} & v_{32} \\ 0 & 0 & v_{33} \end{bmatrix}$$



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transpose



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💡 transpose

$$\begin{bmatrix} 9 & 3 & -6 \\ 3 & 5 & 0 \\ -6 & 0 & 21 \end{bmatrix} = \begin{bmatrix} v_{11}(v_{11}) + 0(0) + 0(0) & v_{11}(v_{21}) + 0(v_{22}) + 0(0) & v_{11}(v_{31}) + 0(v_{32}) + 0(v_{33}) \\ v_{21}(v_{11}) + v_{22}(0) + 0(0) & v_{21}(v_{21}) + v_{22}(v_{22}) + 0(0) & v_{21}(v_{31}) + v_{22}(v_{32}) + 0(v_{33}) \\ v_{31}(v_{11}) + v_{32}(0) + v_{33}(0) & v_{31}(v_{21}) + v_{32}(v_{22}) + v_{33}(0) & v_{31}(v_{31}) + v_{32}(v_{32}) + v_{33}(v_{33}) \end{bmatrix}$$

💡 multiplication



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$$\begin{bmatrix} 9 & 3 & -6 \\ 3 & 5 & 0 \\ -6 & 0 & 21 \end{bmatrix} = \begin{bmatrix} v_{11}^2 & v_{11}v_{21} & v_{11}v_{31} \\ v_{11}v_{21} & v_{21}^2 + v_{22}^2 & v_{21}v_{31} + v_{22}v_{32} \\ v_{11}v_{31} & v_{21}v_{31} + v_{22}v_{32} & v_{31}^2 + v_{32}^2 + v_{33}^2 \end{bmatrix}$$



Symmetric positive definite matrices can be decomposed into a product of a lower triangular matrix and its transpose:

$$K = VV'$$

$$\begin{bmatrix} 9 & 3 & -6 \\ 3 & 5 & 0 \\ -6 & 0 & 21 \end{bmatrix} = \begin{bmatrix} v_{11}^2 & v_{11}v_{21} & v_{11}v_{31} \\ v_{11}v_{21} & v_{21}^2 + v_{22}^2 & v_{21}v_{31} + v_{22}v_{32} \\ v_{11}v_{31} & v_{21}v_{31} + v_{22}v_{32} & v_{31}^2 + v_{32}^2 + v_{33}^2 \end{bmatrix}$$

$$\begin{aligned} k_{11} &= 9 = v_{11}^2 \\ v_{11} &= \sqrt{9} = 3 \end{aligned}$$



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$$3 = 3(v_{21})$$



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$$k_{11} = 9 = v_{11}^2$$

$$v_{11} = \sqrt{9} = 3$$

$$k_{31} = v_{11}v_{31}$$

$$-6 = 3(v_{31})$$

$$v_{31} = -2$$

$$k_{32} = v_{21}v_{31} + v_{22}v_{32}$$

$$0 = 1(-2) + 2(v_{32})$$

$$v_{32} = 1$$

$$k_{21} = v_{11}v_{21}$$

$$3 = 3(v_{21})$$

$$v_{21} = 1$$

$$k_{22} = v_{21}^2 + v_{22}^2$$

$$5 = 1 + v_{22}^2$$

$$v_{22} = \sqrt{4} = 2$$

$$k_{33} = v_{31}^2 + v_{32}^2 + v_{33}^2$$

$$21 = (-2)^2 + (1)^2 + v_{33}^2$$

$$v_{33} = \sqrt{16} = 4$$



Cholesky decomposition

Symmetric positive definite matrices can be decomposed into a product of a lower triangular matrix and its transpose:

$$K = VV'$$

$$\begin{bmatrix} 9 & 3 & -6 \\ 3 & 5 & 0 \\ -6 & 0 & 21 \end{bmatrix} = \begin{bmatrix} v_{11}^2 & v_{11}v_{21} & v_{11}v_{31} \\ v_{11}v_{21} & v_{21}^2 + v_{22}^2 & v_{21}v_{31} + v_{22}v_{32} \\ v_{11}v_{31} & v_{21}v_{31} + v_{22}v_{32} & v_{31}^2 + v_{32}^2 + v_{33}^2 \end{bmatrix}; \quad V = \begin{bmatrix} 3 & 0 & 0 \\ 1 & 2 & 0 \\ -2 & 1 & 4 \end{bmatrix}$$

$$\begin{aligned} k_{11} &= 9 = v_{11}^2 \\ v_{11} &= \sqrt{9} = 3 \end{aligned}$$

$$\begin{aligned} k_{31} &= v_{11}v_{31} \\ -6 &= 3(v_{31}) \\ v_{31} &= -2 \end{aligned}$$

$$\begin{aligned} k_{32} &= v_{21}v_{31} + v_{22}v_{32} \\ 0 &= 1(-2) + 2(v_{32}) \\ v_{32} &= 1 \end{aligned}$$

$$\begin{aligned} k_{21} &= v_{11}v_{21} \\ 3 &= 3(v_{21}) \\ v_{21} &= 1 \end{aligned}$$

$$\begin{aligned} k_{22} &= v_{21}^2 + v_{22}^2 \\ 5 &= 1 + v_{22}^2 \\ v_{22} &= \sqrt{4} = 2 \end{aligned}$$

$$\begin{aligned} k_{33} &= v_{31}^2 + v_{32}^2 + v_{33}^2 \\ 21 &= (-2)^2 + (1)^2 + v_{33}^2 \\ v_{33} &= \sqrt{16} = 4 \end{aligned}$$