

A UCI TMF TUTORIAL: FINDING TARGETING VECTORS AND DOWNLOADING SEQUENCE FOR A MUTANT ALLELE OF YOUR GENE OF INTEREST VIA THE INTERNATIONAL KNOCKOUT MOUSE CONSORTIUM (IKMC) MARTSEARCH WEB PAGE

Enter the name of your gene of interest here – e.g. in this case “Fndc3a”

Viewing IKMC Data in Ensembl

Most of the products/data found within this portal can also be viewed as DAS tracks in the [Ensembl genome browser](#). Follow these links to have the tracks automatically activated for you:

[e! IKMC alleles in Ensembl Mouse](#) [e! IKMC alleles on orthologous genes in Ensembl Human](#)

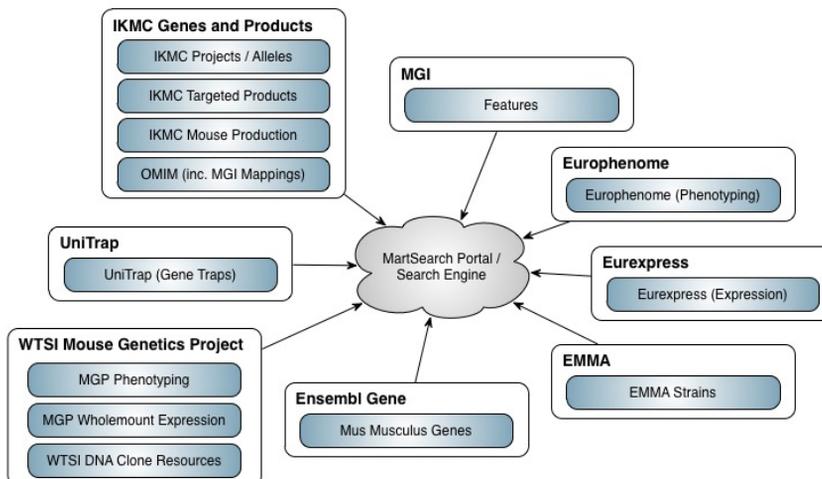
Federated Searches

The links below take you to the [standard MartView interface](#) for several examples of federated queries:

1. Find all IKMC targeted ES cells for genes encoding transcription factors on Chromosome 1. (This query joins IKMC Projects/Alleles to Ensembl).
2. Find all IKMC mice available from the EMMA Repository with information on the vector used to make the mutation. (This query joins IKMC Targeting Repository to Mouse Production data).
3. Find all IKMC targeted ES cells for genes expressed in heart. (This query joins IKMC Projects/Alleles to EurExpress).

The Biomarts

This portal integrates information on IKMC mouse knockout resources with numerous other relevant datasets, including Ensembl, Europhenome, EurExpress and EMMA. For more information about this portal and the way in which it unites and searches the data, please see the [about page](#).

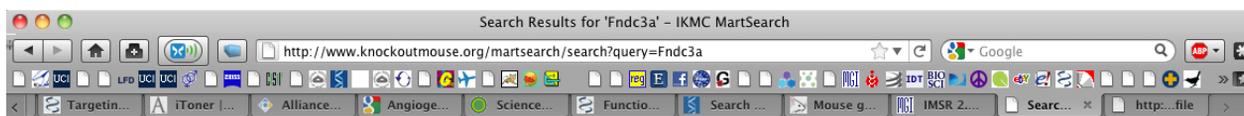


Information for Developers

All the code and data that we produce is open-source and free to use. The following links will guide you to our source-code and documentation on how you can interact with the services we provide.

- [All the code used to create this portal](#)
- [The ruby API used to interact with the Biomarts](#)
- [Using our search engine in your application](#)

The results page



IKMC MartSearch

A- A A+

HOME BROWSE HELP ABOUT

Search Results for 'Fndc3a'

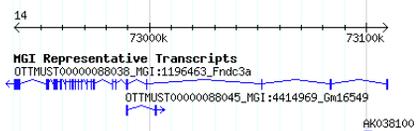
[Explain These Search Results](#)

▼ Fndc3a

Datasets: [Gene Details](#) [Europhenome](#) [Eurexpress](#) [Mice](#) [IKMC Vectors and ES Cells](#) [IKMC Trapped Products](#) [Other Mutants/Resources](#)

▼ Gene Details

Marker Name(s):	fibronectin type III domain containing 3A view this gene in MGI
Marker Type:	protein coding gene
Synonyms:	1700094E19Rik, D14Ert453e, F730017H24Rik, Fndc3, sys
Location:	Chr14:72937753-73109810(-)



Related Human Conditions (from OMIM) - *no related Human Condition*

▶ More Information

Data provided by Mouse Genome Informatics (MGI), Ensembl

[view original data](#)

▼ Europhenome

no data available

Click here to view the details about the mutant alleles generated

In this case, both targeting vectors (DNA) and ES cells with a targeted mutation are available for purchase

[view original data](#)

no data available

▼ IKMC Vectors and ES Cells

	Vectors	ES Cells
EUCOMM (Project: 44191)	order	order

[view details](#)

Data provided by The International Knockout Mouse Consortium (IKMC)

[view original data](#)

▼ IKMC Trapped Products

! Please note that the data supplied within this dataset is currently incomplete. This view is supplied for evaluation purpose only.

	5'		3'			
	UNI000007835#1	UNI000007836#2	UNI000007837#3	UNI000007838#4	UNI000007839#5	
TIGM	1	3	0	7	1	12 traps
EUCOMM	0	0	1	1	0	2 traps
NorCOMM	0	0	0	0	0	0 traps
	1 trap	3 traps	1 trap	8 traps	1 trap	

Data provided by UniTrap

[view original data](#)

▼ Other Mutants/Resources

International Mouse Strain Resource (IMSR): 24 strains/ES cell lines
 International Gene Trap Consortium (IGTC): 28 gene traps
 Targeted mutations in MGI: 2 mutants
 Other mutations in MGI: 27 mutants

Data provided by Mouse Genome Informatics (MGI)

[view original data](#)

The Project Report page – more details of the mutant alleles



IKMC MartSearch

A- A A+

HOME BROWSE HELP ABOUT

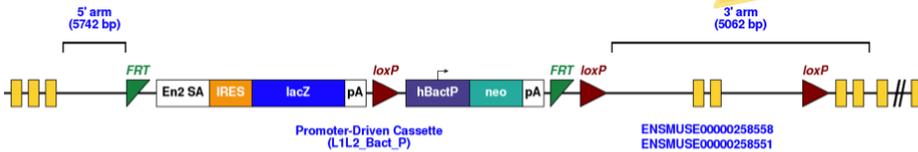
IKMC Project Report – EUCOMM (ID: 44611)

Fndc3a MGI:1196463 ENSMUSG00000033487 OTTMUSG00000034682

Pre-pipeline Designs Vectors ES Cells ES Cells - Targeting Confirmed Mice

Currently, two types of targeted alleles are available. Both alleles are “mutant first”; i.e. the presence of the [splice acceptor – IRES- lacZ – poly A] cassette may result in a truncated, non-functional protein product. The locus can be reverted to a nearly wildtype sequence by expression of Flp recombinase, which will excise sequences between the Frt sites. In the case of the “conditional potential” allele, this will leave one or more exons of the gene flanked by two loxP sites (“floxed”). Expression of cre recombinase should cause deletion of the (in this example) two floxed exons. The location of the loxP sites is designed to generate a frame-shift mutation that should result in degradation of any remaining mRNA via nonsense-mediated decay (NMD) systems.

ES Cell Clones With Conditional Potential



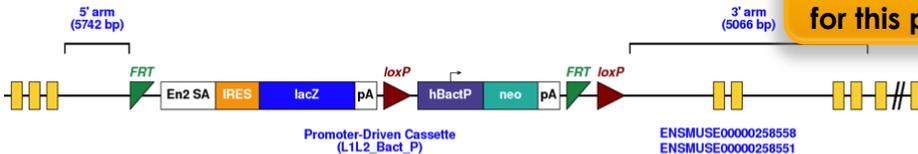
Ensembl (mouse) Ensembl (human) UCSC (mouse) UCSC (human) Southern Blot Tool

ES Cell Clone	Targeting Vector	Allele	Allele Type	Floxed Exons	Parental ES Cell Line	Genbank File	Mouse	QC Data
order HEPD0602_3_D12	PG00187_Z_3_F06	Fndc3a ^{tm1a} (EUCOMM)Hmgu	Knockout-First – Reporter Tagged Insertion (Promotor Driven Cassette)	ENSMUSE00000258551 – ENSMUSE00000258558	JM8A3.N1	view	no	view (about)

[show/hide more ES cells](#)

Click here to view the Genbank formatted entry for this predicted mutation

ES Cell Clones Without Conditional Potential



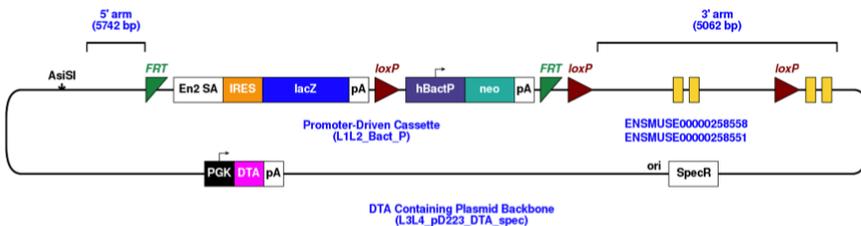
Note: Mutations of type "Without Conditional Potential" are correctly targeted clones that have lost the 3' LoxP site. These mutations cannot be converted into conditional alleles.

Ensembl (mouse) Ensembl (human) UCSC (mouse) UCSC (human) Southern Blot Tool

ES Cell Clone	Targeting Vector	Allele	Allele Type	Floxed Exons	Parental ES Cell Line	Genbank File	Mouse	QC Data
order HEPD0602_3_B11	PG00187_Z_3_F06	Fndc3a ^{tm1e} (EUCOMM)Hmgu	Targeted Non-Conditional (Promotor Driven Cassette)	ENSMUSE00000258551 – ENSMUSE00000258558	JM8A3.N1	view	no	view (about)

[show/hide more ES cells](#)

Targeting Vectors



DTA Containing Plasmid Backbone (L3L4_pD223_DTA_spec)

Details of the predicted mutant allele in GenBank format

Mozilla Firefox

http://www.knockoutmouse.org/targ_rep/alleles/2567/escell-clone-genbank-file

Google

LOCUS allele_193256_ENSMUSE00000258558-ENSMUSE00000258551_L1L2_Bact_P 38157 bp dna linear UNK

ACCESSION unknown

DBSOURCE accession design_id:193256

COMMENT cassette : L1L2_Bact_P

COMMENT design_id : 193256

FEATURES

Feature	Location/Qualifiers
primer_bind	complement(9309..9358) /label=G5 /type="G5" /note="G5"
primer_bind	15002..15051 /label=U5 /type="U5" /note="U5"
exon	221..458 /db_xref="ens:ENSMUSE00000258582" /label=ENSMUSE00000258582 /note="ENSMUSE00000258582"
exon	5368..5637 /db_xref="ens:ENSMUSE00000258572" /label=ENSMUSE00000258572 /note="ENSMUSE00000258572"
exon	7197..7255 /db_xref="ens:ENSMUSE00000258566" /label=ENSMUSE00000258566 /note="ENSMUSE00000258566"
LRPCR_primer	7912..7939 /type="GF3" /label=GF3 /note="GF3"
LRPCR_primer	8795..8821 /type="GF4" /label=GF4 /note="GF4"
misc_feature	9309..15051 /label=5 arm /note="5 arm"
misc_feature	complement(15079..15091) /label=R1 Gateway /note="R1 Gateway"
misc_feature	complement(15079..15091) /label=B1 site /note="B1 site"
misc_feature	15092..15103 /label=L1 Gateway /note="L1 Gateway"
misc_feature	15092..15103 /label=B1 site /note="B1 site"
misc_feature	15116..15163 /label=Frt /note="Frt"
intron	15164..16038 /label=En2 intron /note="En2 intron"
misc_feature	16036..16038 /label=SA /note="SA"
exon	16039..16040 /label=En2 exon /note="En2 exon"
misc_feature	16252..16812 /label=ECMV IRES /note="ECMV IRES"
gene	16823..19869 /gene="lacZ" /label=lacZ /note="lacZ"
misc_feature	19904..20159 /label=SV40 pA /note="SV40 pA"
misc_feature	20163..20196 /label=loxP /note="loxP"
promoter	20222..20742 /label=human beta actin promoter /note="human beta actin promoter"
CDS	20768..21562 /label=NeoR /note="NeoR"
misc_feature	21760..22003 /label=SV40 pA /note="SV40 pA"
misc_feature	22020..22067 /label=Frt /note="Frt"
misc_feature	22074..22107 /label=loxP /note="loxP"
misc_feature	22123..22133 /label=B2 site /note="B2 site"
misc_feature	22123..22133 /label=L2 Gateway /note="L2 Gateway"
misc_feature	22134..22146 /label=R2 Gateway /note="R2 Gateway"
misc_feature	22134..22146 /label=B2 site /note="B2 site"
primer_bind	complement(22174..22223) /label=U3 /type="U3" /note="U3"
primer_bind	22979..23028 /label=D5 /type="D5" /note="D5"

The information is in GenBank format. Before we can view and annotate this data using Lasergene's "SeqBuilder" application, we need to reformat the information

Copy and paste the information from your browser into a suitable text editor application – e.g. MS-Word

LOCUS allele_193256_ENSMUSE00000258558-ENSMUSE00000258551_L1L2_Bact_P 38157 bp dna linear UNK

ACCESSION unknown

DBSOURCE accession design_id=193256

COMMENT cassette : L1L2_Bact_P

COMMENT design_id : 193256

FEATURES

primer_bind Location/Qualifiers
complement(9309..9358)
/label=G5
/type="G5"
/note="G5"

primer_bind
15002..15051
/label=U5
/type="U5"
/note="U5"

exon
221..458
/db_xref="ens:ENSMUSE00000258582"
/label=ENSMUSE00000258582
/note="ENSMUSE00000258582"

exon
5368..5637
/db_xref="ens:ENSMUSE00000258572"
/label=ENSMUSE00000258572
/note="ENSMUSE00000258572"

exon
7197..7255
/db_xref="ens:ENSMUSE00000258566"
/label=ENSMUSE00000258566
/note="ENSMUSE00000258566"

LRPCR_primer
7912..7939
/type="GF3"
/label=GF3
/note="GF3"

LRPCR_primer
8795..8821
/type="GP4"
/label=GP4
/note="GP4"

misc_feature
9309..15051
/label=5 arm
/note="5 arm"

misc_feature
complement(15079..15091)
/label=R1 Gateway
/note="R1 Gateway"

misc_feature
complement(15079..15091)
/label=B1 site
/note="B1 site"

misc_feature
15092..15103
/label=L1 Gateway
/note="L1 Gateway"

misc_feature
15092..15103
/label=B1 site
/note="B1 site"

misc_feature
15116..15163
/label=FrT
/note="FrT"

intron
15164..16038
/label=En2 intron
/note="En2 intron"

misc_feature
16036..16038
/label=SA
/note="SA"

exon
16039..16040
/label=En2 exon
/note="En2 exon"

misc_feature
16252..16812
/label=ECMV IRES
/note="ECMV IRES"

gene
16823..19869
/gene="lacZ"
/label=lacZ
/note="lacZ"

misc_feature
19904..20159
/label=SV40 pA
/note="SV40 pA"

misc_feature
20163..20196
/label=loxP
/note="loxP"

promoter
20222..20742
/label=human beta actin promoter
/note="human beta actin promoter"

CDS
20768..21562
/label=Neor
/note="Neor"

misc_feature
21760..22003
/label=SV40 pA
/note="SV40 pA"

misc_feature
22020..22067
/label=FrT
/note="FrT"

misc_feature
22074..22107
/label=loxP
/note="loxP"

misc_feature
22123..22133
/label=B2 site
/note="B2 site"

misc_feature
22123..22133
/label=L2 Gateway
/note="L2 Gateway"

misc_feature
22134..22146
/label=R2 Gateway
/note="R2 Gateway"

misc_feature
22134..22146
/label=B2 site
/note="B2 site"

primer_bind
complement(22174..22223)
/label=U3
/type="U3"
/note="U3"

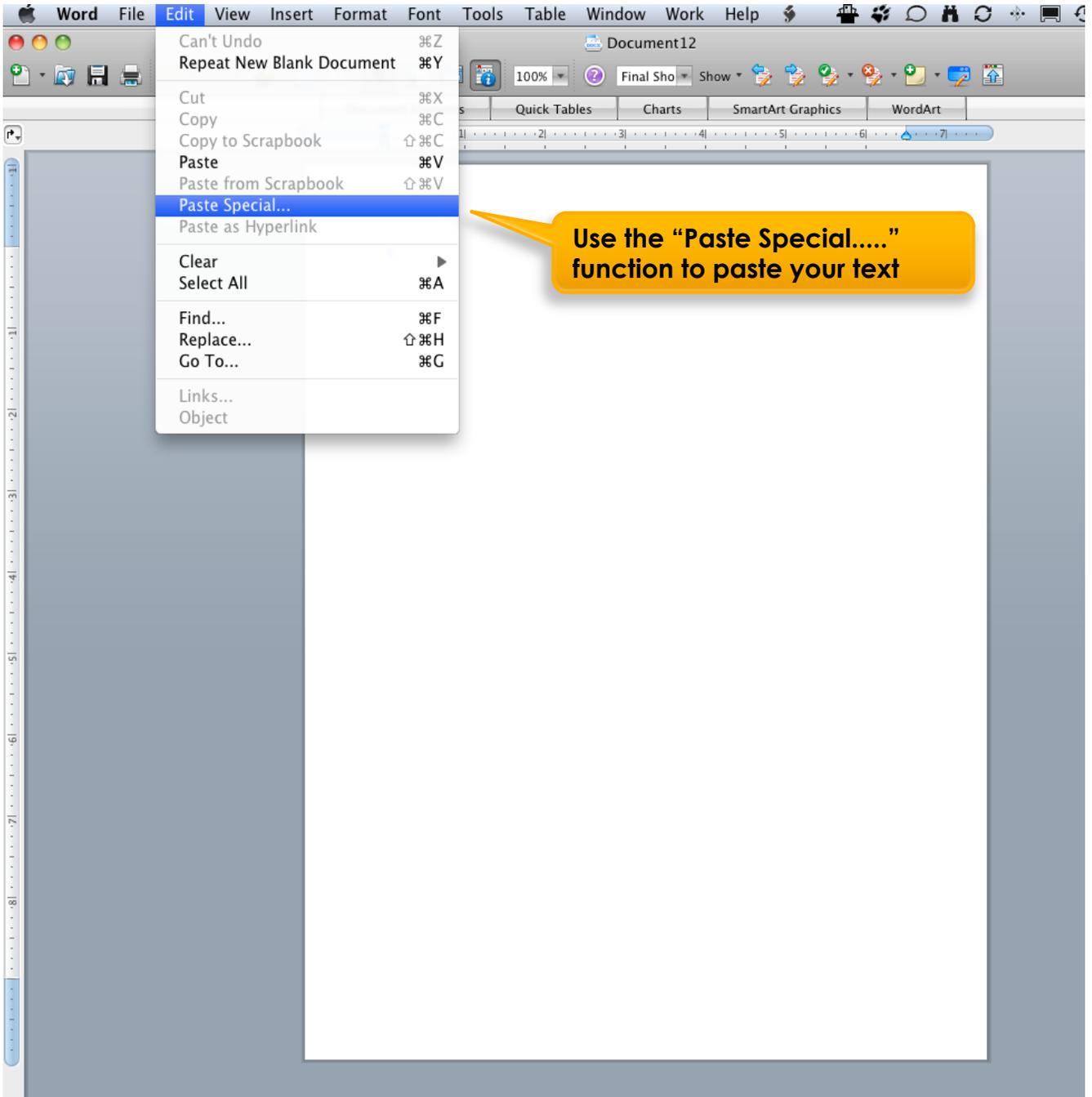
primer_bind
22979..23028
/label=D5
/type="D5"
/note="D5"

exon
22536..22693
/db_xref="ens:ENSMUSE00000258558"
/type="targeted"

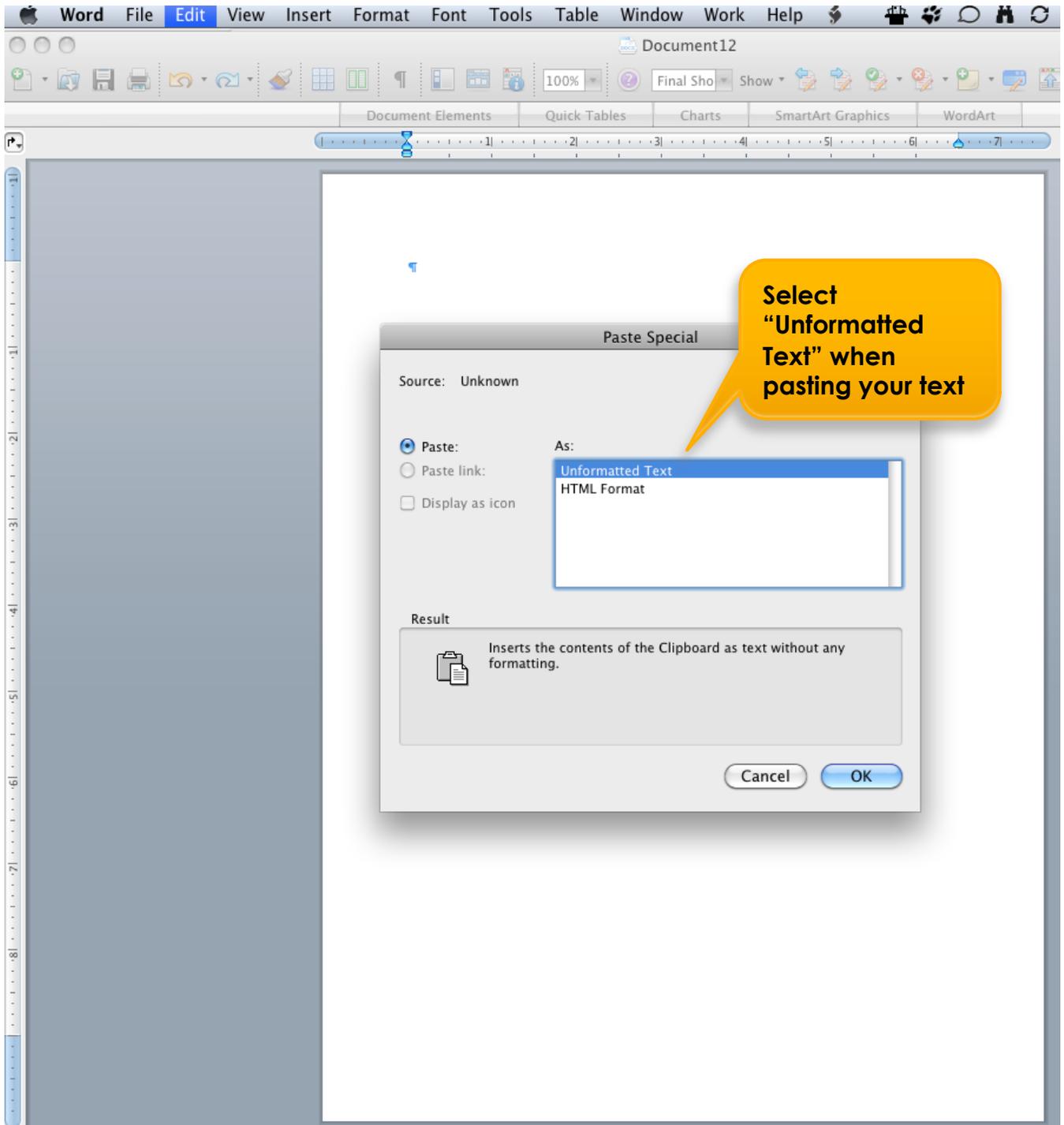
First, "select-all", then "copy"

Copy
Select All
Yahoo!
Search Google for "LOCUS allele_19..."
View Selection Source
Login to LastPass
LastPass
LastPass Fill Forms
Add To Top Sites

Copy and paste the information from your browser into a suitable text editor application – e.g. MS-Word



Copy and paste the information from your browser into a suitable text editor application – e.g. MS-Word



To enable the Lasergene suite's "EditSeq" application to import your data, save the GenBank-formatted information as a text (.txt) file

The image shows a screenshot of the Microsoft Word application interface. The 'File' menu is open, and the 'Save As...' option is highlighted. A yellow callout bubble points to the 'Save As...' option with the following text: "To save your Genbank information in a format that Lasergene's EditSeq application can recognize, select 'Save As...' then select '.txt' (Text) format".

The background of the screenshot shows a document with GenBank-formatted text, including features like 'allele', 'dna', 'ON', 'CE', 'NT', 'ES', 'bind', 'exon', 'LRPCR_primer', and 'misc_feature'. The text is formatted with various annotations and symbols.

To enable the Lasergene suite's "EditSeq" application to import your data, save the GenBank-formatted information as a text (.txt) file

Save As: **Fndc3a_cKO.txt**

Desktop

Word Document (.docx)
Common Formats
Word 97-2004 Document (.doc)
Word Template (.dotx)
Word 97-2004 Template (.dot)
Rich Text Format (.rtf)
✓ Plain Text (.txt)
Web Page (.htm)
PDF
Specialty Formats
Word Macro-Enabled Document (.docm)
Word Macro-Enabled Template (.dotm)
Word XML Document (.xml)
Word 2003 XML Document (.xml)
Single File Web Page (.mht)
Word Document Stationery (.doc)
Speller Custom Dictionary (.dic)
Speller Exclude Dictionary (.dic)
Word 4.0-6.0/95 Compatible (.rtf)

Description
Exports document content
[Learn more about file formats](#)
 Append file extension
Options... Complete
New Folder

Cancel Save

...exon.....16039..16040
...../label=En2 exon
...../note=En2 exon
...misc_feature.....16252..16812
...../label=ECMV IRES
...../note=ECMV IRES

To enable the Lasergene suite's "EditSeq" application to import your data, save the GenBank-formatted information as a text (.txt) file

MS-Word prompts you for a format for text-encoding during the "File Conversion". Use the default setting ("Mac OS")

The screenshot shows a Microsoft Word document with a GenBank file being converted to a text file. The document content is as follows:

```
LOCUS.....allele_193256_ENSMUSE00000258558-ENSMUSE00000258551_L1L2_Bact_P.....
38157 bp.....dna.....linear::UNK
ACCESSION::unknown
DBSOURCE::accession_design_id=193256
COMMENT::cassette::L1L2_Bact_P
COMMENT::design_id::193256
FEATURES.....Location/Qualifiers
.....primer_bind::complement(9309..9358)
...../label=G5
...../type=G5
...../note=G5
.....primer_bind::15002..15051
...../label=US
```

The 'File Conversion - Fndc3a_cKO.txt' dialog box is open, showing the following options:

- Warning: Saving as a text file will cause all formatting, pictures, and objects in your file to be lost.
- Text encoding: Mac OS (Default) MS-DOS Other encoding: [List of encodings]
- Options: Insert line breaks
- End lines with: CR only
- Allow character substitution
- Preview: [Preview of the document content]
- Buttons: Cancel, OK

A yellow callout bubble points to the 'Mac OS (Default)' option in the 'Text encoding' section. Another yellow callout bubble points to the 'OK' button.

Click "OK"

Next, import your saved “.txt” file into Lasergene suite’s “EditSeq” application

You've launched and are in “EditSeq”

Use “Import” in the File menu

Select your “.txt” file

The screenshot displays the EditSeq application window. The top menu bar includes 'File', 'Edit', 'Search', 'Speech', 'Features', 'Goodies', 'Net Search', 'Window', and 'Help'. The main window shows a DNA sequence editor with a ruler at the top and a sequence of nucleotides below. A yellow callout bubble points to the 'Import' option in the File menu.

An 'Import' dialog box is open, showing a file browser view of the Desktop. The file 'Fndc3a_cKO.txt' is selected. A yellow callout bubble points to this file. The dialog also shows a preview of the selected file, which is a plain text file of 57 KB, created and modified on 04/13/11.

At the bottom of the dialog, there are options to 'Enable' the import of 'All Readable Files', with radio buttons for 'DNA' and 'Protein'. The 'Open' button is highlighted.

The GenBank formatted information viewed following import into Lasergene suite's "EditSeq" application

The screenshot displays the EditSeq application interface. At the top, a menu bar includes File, Edit, Search, Speech, Features, Goodies, Net Search, Window, and Help. The main window shows a DNA sequence for 'Fndc3a_cKO-1.seq : SEQUENCE' with a position indicator from 1 to 130. Below the sequence, the GenBank header information is visible, including accession number, source, and features. A yellow callout box on the left states: 'Your data has been imported into EditSeq'. Another yellow callout box on the right points to the GenBank header information, stating: 'The GenBank format header information is located in this field'. A third yellow callout box at the bottom points to the 'exon' features, stating: 'Each of the "features" in the predicted targeted allele are located in this field'. The GenBank header information includes:

```
LOCUS       allele_193256_ENSMUSE00000258558-ENSMUSE00000258551_L1L2_Bact_P          38157 bp    dna    linear    UNK
ACCESSION   unknown
DBSOURCE    accession design_id=193256
COMMENT     cassette : L1L2_Bact_P
            design_id : 193256

BASE COUNT  10623 a   8006 c   8058 g   11470 t
ORIGIN
```

The features section includes:

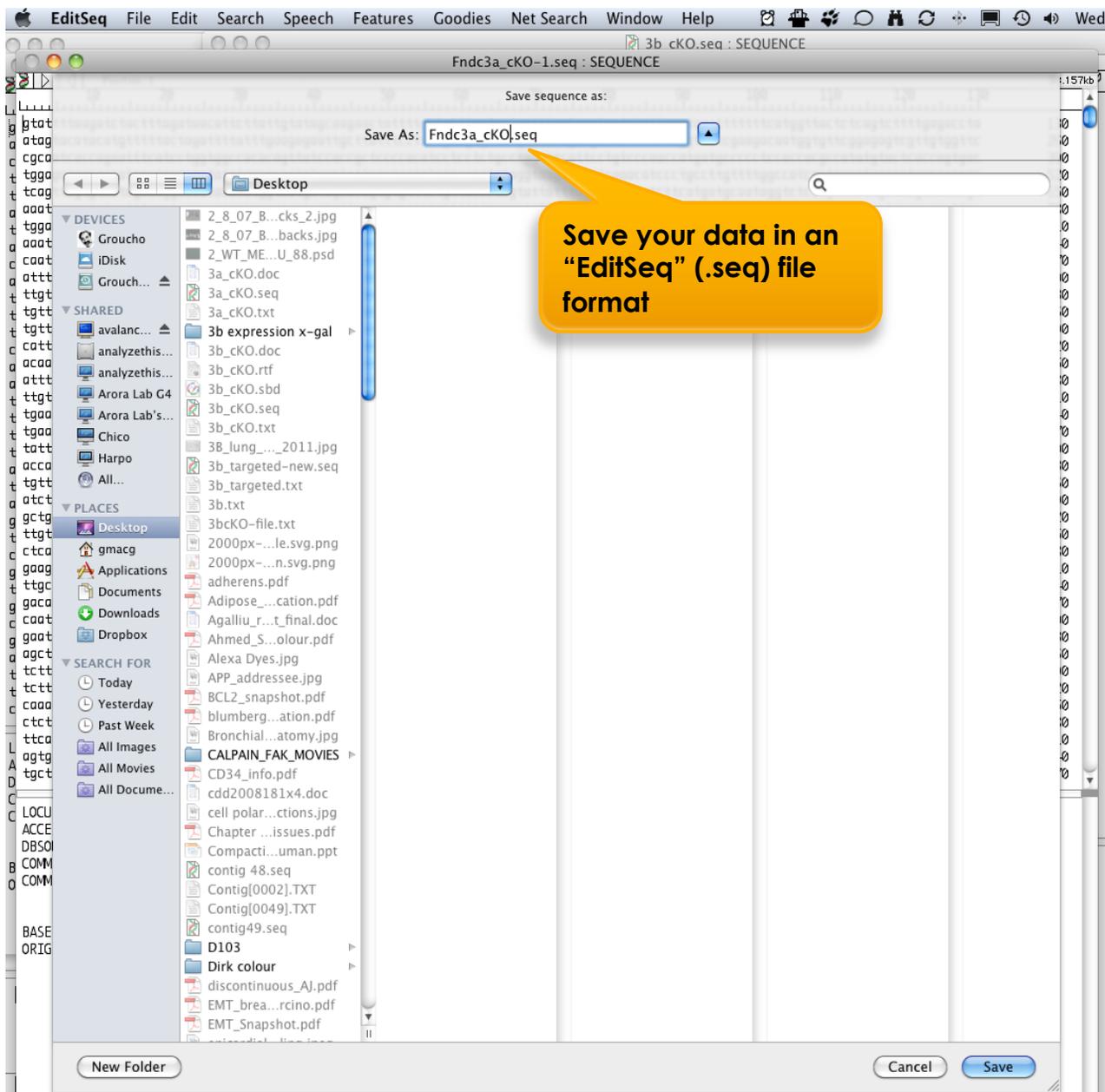
```
primer_bind complement(9309..9358)
/label=G5
/type="G5"
/note="G5"

primer_bind 15002..15051
/label=U5
/type="U5"
/note="U5"

exon 221..458
/db_xref="ens:ENSMUSE00000258582"
/label=ENSMUSE00000258582
/note="ENSMUSE00000258582"

exon 5368..5637
/db_xref="ens:ENSMUSE00000258572"
```

To view the information in Lasergene's "SeqBuilder" application, first save the information in an EditSeq (".seq") file format



Save your data in an "EditSeq" (.seq) file format

primer_bind	complement(9309..9358)
/label=G5	
/type="G5"	
/note="G5"	
primer_bind	15002..15051
/label=U5	
/type="U5"	
/note="U5"	
exon	221..458

To view the information in Lasergene's "SeqBuilder" application, tell SeqBuilder to open the EditSeq (".seq") file

You are now using "SeqBuilder"

Using "File" -> "Open", select your EditSeq (".seq") formatted file

Name Fndc3a_cKO.seq
Kind Lasergene DNA File
Size 53 KB on disk
Created 04/13/11
Modified 04/13/11
Last opened 04/13/11

Enable: All Known Formats

Cancel Open

primer_bind complement(9309..9358)
/label=G5

Your sequence information viewed in Lasergene's "SeqBuilder" application. Remember to save the information in a SeqBuilder (".sbd") file format

You are now using "SeqBuilder"

The screenshot displays the SeqBuilder application window. The top menu bar includes: SeqBuilder, File, Edit, Features, Enzymes, Sequence, Cloning, Priming, Format, View, Net Search, Window, Help. The main window shows a DNA sequence for file 'Fndc3a_cKO.seq' with a position scale from 1 to 38,157 kb. The sequence is annotated with several features represented by colored bars: SV40 pA (orange), Frt (orange), loxP (orange), B2 site (orange), L2 Gateway (orange), R2 G...way (orange), R2 ...way (orange), U3 (pink), target region (orange), and ens:ENSMUSE0000258558 (green). A left sidebar lists various views: Sequence, Feature List, Comment, Linear Map, Circular Map, Primer Design, Primer List, Minimap, Site Summary, Residues, Enzymes Displayed, Features Displayed, ORFs, Full Translations, Partial Translations, Rulers, and Blank Rows. At the bottom, a 'misc_feature' list is visible, showing entries with coordinates and labels like '3 arm', '5 arm', 'B1 site', and 'B2 site'. A yellow callout bubble on the left contains the text 'Annotated sequence information of predicted targeted allele'. Another yellow callout bubble on the right contains the text 'Features in your file'.

Annotated sequence information of predicted targeted allele

Features in your file