

Service	deliverables	IRC prices
Sperm cryopreservation	At least 5 straws frozen	400 €
Quality control of frozen sperm	QC: IVF > 20% fertilization	400 € ⁽²⁾
Embryo cryopreservation ⁽³⁾	At least 200 homozygous or 300 heterozygous embryos frozen	1.650-4.950 € (case by case)
Rederivation/revival from frozen sperm	At least 4 weaned rederived animals ⁽⁴⁾ or 1 mutant, free of pathogens on Felasa list	2.500 € ⁽¹⁾⁽⁸⁾
	If IVF or embryo transfer is not successful due to - not enough embryos to perform embryo transfer (< 10) - fosters did not get pregnant - no or less than 4 offspring and no mutants	500 € ⁽⁵⁾
Rederivation/revival from frozen embryos	At least 4 weaned rederived animals ⁽³⁾ or 1 mutant, free of pathogens on Felasa list	1.800 € ⁽¹⁾⁽⁸⁾
	If transfer is not successful due to - not enough embryos to do a transfer (< 10) - fosters did not get pregnant - no or less than 4 offspring and no mutants	500 € ⁽⁶⁾
Germfree rederivation	- Sperm freezing of mutant line - IVF with frozen sperm + 2-cell freeze - germfree embryo transfers with fresh or frozen 2-cell embryos - sterility testing of offspring At least 2 offspring, irrespective of genotype	7.000 € ⁽⁷⁾
generation of knockout mice using CRISPR	- in silico design of project - identifying and ordering crRNAs - in silico design PCR screening strategy - optimization of PCR screening - electroporation of 400 C57BL/6J zygotes or 2 founders, whatever comes first - screening offspring for mutant founders - characterization of the different mutant alleles in the most interesting founders by subcloning and sequencing - breeding founders 1 generation and analyzing the mutations in the F1 offspring	6.600 € ⁽⁸⁾

generation of knockin mice using CRISPR technology	<ul style="list-style-type: none"> - in silico design of project - identifying and ordering crRNAs - in silico design PCR screening strategy to test efficiency of crRNAs - optimization of PCR screening to test efficiency of crRNAs - test-electroporation with different crRNA's - screen blasocysts from test-electroporation to identify most efficient crRNA's - in silico design PCR screening strategy to check correct integration of repair template - optimization of PCR screening to check correct integration of repair template - designing and ordering repair template - electroporation/injection of 600 C57BL/6J zygotes or 2 founders, whatever comes first - screening offspring for mutant founders - characterization of the different mutant alleles in the most interesting founders by subcloning and sequencing - breeding founders 1 generation and analyzing the mutations in the F1 offspring 	8.250 € ^(8,9,10)
generation of transgenic mice by zygote injection of cDNA or BAC	<ul style="list-style-type: none"> - injection of 400 C57BL/6J zygotes or 2 founders, whatever comes first 	5.000 € ⁽⁸⁾
ES-cell targeting I (vector integrates by HR)	<ul style="list-style-type: none"> - prepare 24 well DNA from 200-400 picked ES-cell clones - triplicate frozen -80 stock of each clone on 96 well plates - thawing up to 8 positive clones - expanding and freezing stock of 2-3 vials/clone in LN2 - DNA preparation of each positive clone for reconfirmation - karyotyping of each positive clone by chromosome count - 1 new electroporation in case 1st electroporation didn't result in positive clones 	4.500 €
ES-cell targeting II (vector integrates by RMCE)	<ul style="list-style-type: none"> - preparing 24 well DNA from 10 picked ES-cell clones - triplicate frozen -80 stock of each clone on 96 well plates - thawing up to 8 positive clones - expanding and freezing stock of 2-3 vials/clone in LN2 - DNA preparation of each positive clone for reconfirmation - karyotyping of positive clones by chromosome count - 1 new electroporation in case 1st electroporation didn't result in positive clones 	3.000 €

ES-cell targeting III (vector integrates randomly)	<ul style="list-style-type: none"> - preparing 24 well DNA from 100 picked ES-cell clones - triplicate frozen -80 stock of each clone on 96 well plates - thawing up to 8 positive clones - expanding and freezing stock of 2-3 vials/clone in LN2 - DNA preparation of each positive clone for reconfirmation - karyotyping of each positive clone by chromosome count - 1 new electroporation in case 1st electroporation didn't result in positive clones 	3.300 €
ES-cell targeting IV (Flpe or Cre electroporation to remove selection marker)	<ul style="list-style-type: none"> - 24 well DNA from 5-10 picked ES-cell clones sensitive to selection marker that is removed - triplicate frozen -80 stock of each clone on 96 well plates - thawing up to 8 positive clones - expanding and freezing stock of 2-3 vials/clone in LN2 - DNA preparation of each positive clone for reconfirmation - karyotyping of each positive clone by chromosome count - 1 new electroporation in case 1st electroporation didn't result in positive clones 	1.650 €
generation of chimeras by aggregation or blastocyst injection	<ul style="list-style-type: none"> - karyotyping clones by chromosome counting (if not done yet for external clones) - aggregation of 160 morulas or injection of 80 blastocysts or 2 high (>50%) chimeras, whichever comes first - testbreeding of chimeras to check for germline 	4.500 € ⁽⁸⁾
General ES-cell related services	<ul style="list-style-type: none"> - cloning, linearisation and purification of a ROSA26 targeting vector - cloning and testing southern probes (5' and 3') on wt DNA on southern blot - screening targeted ES-cell DNA by southern blot (per 100 clones) 	1.650 € 3.000 € 1.500 €
Derivation of ES-cell lines	<ul style="list-style-type: none"> - flushing blastocysts from uterus - keeping blastocysts in ES-derivation culture medium - picking and expanding outgrowths from blastocysts - karyotyping of each line (up to 5) by chromosome counting - 24 well DNA preparation from each line - freezing 2-3 vials from each line and storing in LN2 	2.000 €

⁽¹⁾ cost of health screening not included

⁽²⁾ 2 attempts are performed

⁽³⁾ only on special request

⁽⁴⁾ irrespective of genotype

⁽⁵⁾ if no QC was done on the sperm batch or if sperm comes from an external stock

⁽⁶⁾ if embryos are from external stock

⁽⁷⁾ TCF: 4.500 €; Germfree facility: 2.500 €

⁽⁸⁾ cage prices for fosters, offspring from fosters, testbreedings, offspring from testbreedings not included

⁽⁹⁾ cost of donor DNA > 500 bases is not included in the end-price and should be payed by the researcher

⁽¹⁰⁾ knockin-founders cannot be guaranteed

⁽¹¹⁾ only for low complexity vectors; complex vectors need to be made synthetically by a company