Department of Primary Industries

NSW 2019 Animal Use in Research Statistics

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1. Summary

Information on the use of animals in research in NSW is collected by animal research establishments on a calendar-year basis and submitted to the NSW Department of Primary Industries (DPI) for reporting.

The following information is included in this report:

- General charts to show the trend of animal use since 2010.
- Purpose tables for 2019. There are ten Purpose Categories (see Appendix: *Guide to the categories of reporting*) and these charts show the numbers of animals used, in species groups, for each purpose against the nine categories of procedures (see Appendix: *Guide to the categories of reporting*). The categorisation of procedures aims to give some indication of the 'invasiveness' or 'impact' of the research being undertaken on the animals involved.
- The purpose tables provide a breakdown of the use of all species groups - compared to previous years where breakdown information on species groups was for four categories (laboratory mammals, domestic mammals, birds and primates).
- Fate of animals for 2019: this includes mandatory reporting on the fate of all domestic cats and dogs and voluntary reporting on the fate of other animals (see Appendix: *Guide to the categories of reporting* for the categories of Fate of animals).
- Lethality testing for 2019. The *Animal Research Act 1985* defines a 'lethality test' as *'an animal research procedure in which any material or substance is administered to animals for the purpose of determining whether any animals will die or how many animals will die'*.
- Examples provided by animal research establishments of the implementation of the 3Rs (Replacement, Reduction and Refinement in animal use) in 2019.
- Appendix - Guide to the categories of reporting.

Information provided by research establishments each year also includes the collection of statistics on animals used in the procedure category of "*Observation Involving Minor Interference*". The guidance for inclusion of animals in this procedure category is: *"Animals are not interacted with or, where there is interaction, it would not be expected to compromise the animal's welfare any more than normal handling, feeding, etc. There is no pain or suffering involved."* This procedure category of use potentially includes large numbers of animals. For example, it includes the observation of free-living animals – such as where an aerial survey of birds could record thousands of animals. A chart has therefore been included which excludes this procedure category from the total number of animals used.

Collation and reporting of statistics is reliant upon the information provided by animal research establishments. This means there can be minor differences in the interpretation of

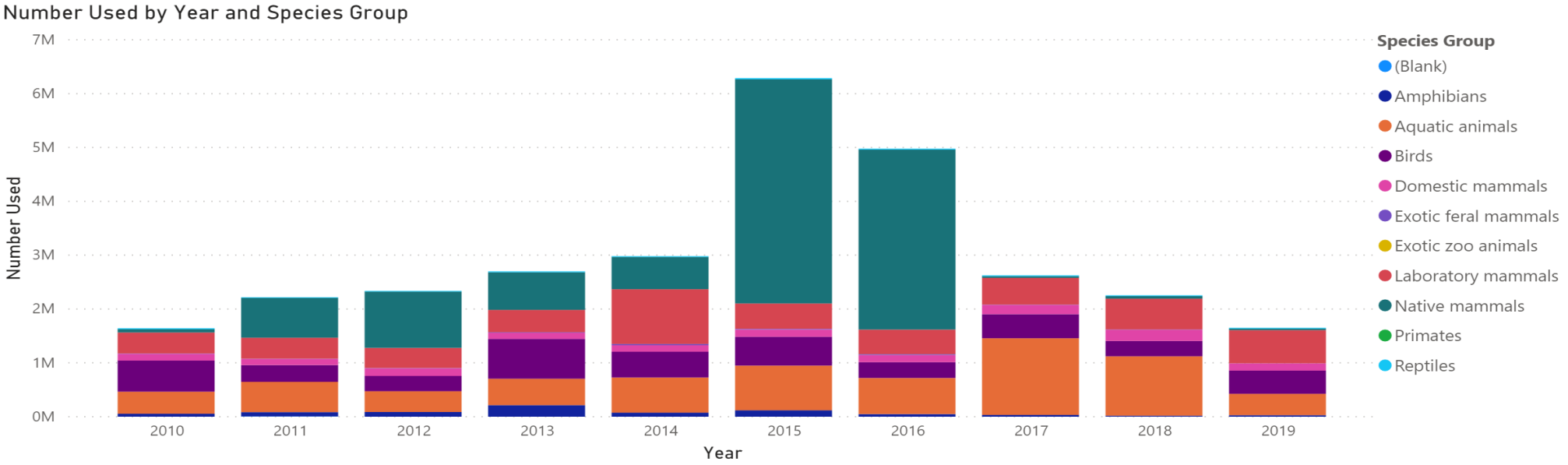
which Purpose and Procedure categories of use are the most appropriate. The magnitude of the information submitted by research establishments means that NSW DPI is not able to verify the data within each individual research project – this is the responsibility of the reporting establishments. However, NSW DPI does, where necessary, undertake verification of information submitted to the level of individual projects at each research establishment. For example NSW DPI seeks to resolve apparent discrepancies reported in category combinations, such as Purpose category *Education* reported with a corresponding Procedure category *Death As An Endpoint*.

Notes:

- For the 2019 reporting year reporting on the Category: *Fate of animals* was mandatory for the use of domestic cats and dogs, and voluntary for other species.
- For the 2019 reporting year, the breakdown of all species groups is included.
- For the 2017 reporting year there was a large increase in the number of aquatic animals used. This was primarily due to two projects which used almost 775,000 fish in the procedure category "Observation Involving Minor Interference". One of these studies involved counting over 500,000 fish by camera recordings.
- For the 2015 and 2016 reporting years there was a large increase in the numbers of animals used. This was primarily due to two projects which involved the aerial counting of bats throughout NSW. There was no interaction with the majority of animals in these projects and these accounted for the reporting of approximately three million animals for each reporting year.
- For the 2016 Animal Use in Statistics Report there were some errors in the item 2 General Charts for the entries for the 2015 year (over-reporting of numbers), and the total for Chart 4 for 2016 (final line not included in the total). These errors have been corrected for subsequent Animal Use in Statistics reports.
- For the 2010 and 2011 reporting years there are species recorded as blank species categories because an incorrect species code was used. The impact of this on the charts is negligible.
- Animals are counted for each year in which they are used. This means, for example, that animals in a project that runs for a period of more than one year will be counted again for each year in which they are used. So the year to year comparisons of animal numbers includes individual animals that are the same across two or more years.

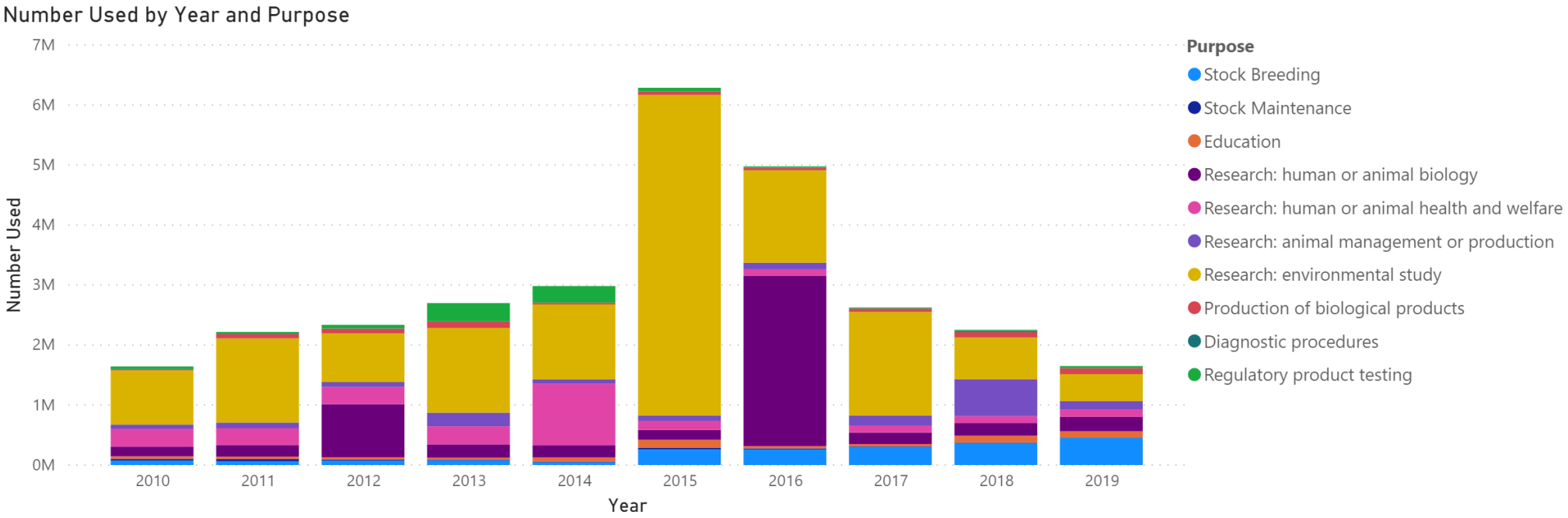
2. General Charts

2.1 Number of animals used over time by species grouping



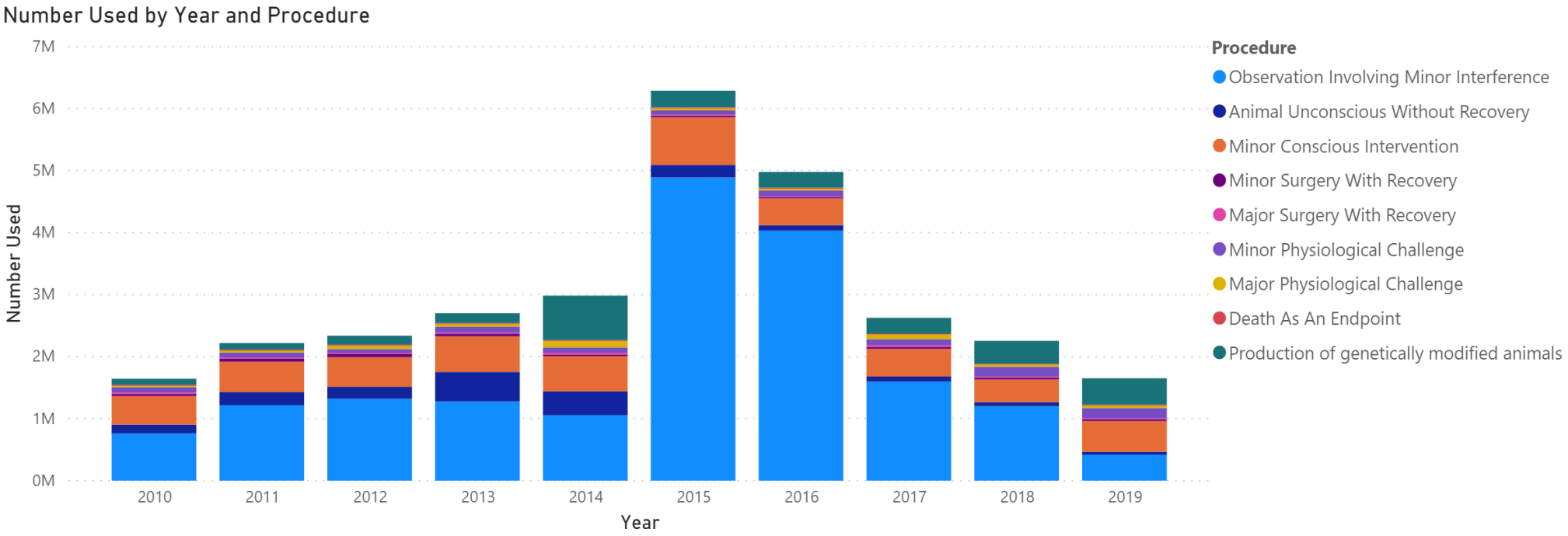
Species group	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	**Total**
	195	5,460									**5,655**
Amphibians	54,992	79,446	87,417	214,616	75,424	118,721	49,008	33,093	18,067	28,413	**759,197**
Aquatic animals	409,917	562,356	386,102	491,114	652,902	830,769	670,514	1,424,101	1,104,172	397,139	**6,929,086**
Birds	576,787	311,690	283,461	739,293	478,754	534,812	292,834	445,877	284,985	430,573	**4,379,066**
Domestic mammals	127,468	114,511	141,288	114,914	120,239	135,679	133,537	172,866	207,125	133,515	**1,401,142**
Exotic feral mammals	5,318	5,195	6,525	9,411	23,200	12,541	15,351	5,338	5,941	1,960	**90,780**
Exotic zoo animals	27	32	71	72	155	83	32	21	37	140	**670**
Laboratory mammals	389,507	388,701	374,037	414,652	1,017,494	470,634	457,431	497,337	572,490	616,812	**5,199,095**
Native mammals	59,870	738,903	1,044,611	697,764	598,737	4,161,992	3,340,256	29,220	45,487	26,954	**10,743,794**
Primates	184	27	18	22	41	179	96	38	44	53	**702**
Reptiles	18,328	12,141	13,398	17,674	15,730	22,067	18,196	17,568	15,595	14,749	**165,446**
Total	**1,642,593**	**2,218,462**	**2,336,928**	**2,699,532**	**2,982,676**	**6,287,477**	**4,977,255**	**2,625,459**	**2,253,943**	**1,650,308**	**29,674,633**

2.2 Number of animals used over time by research purpose



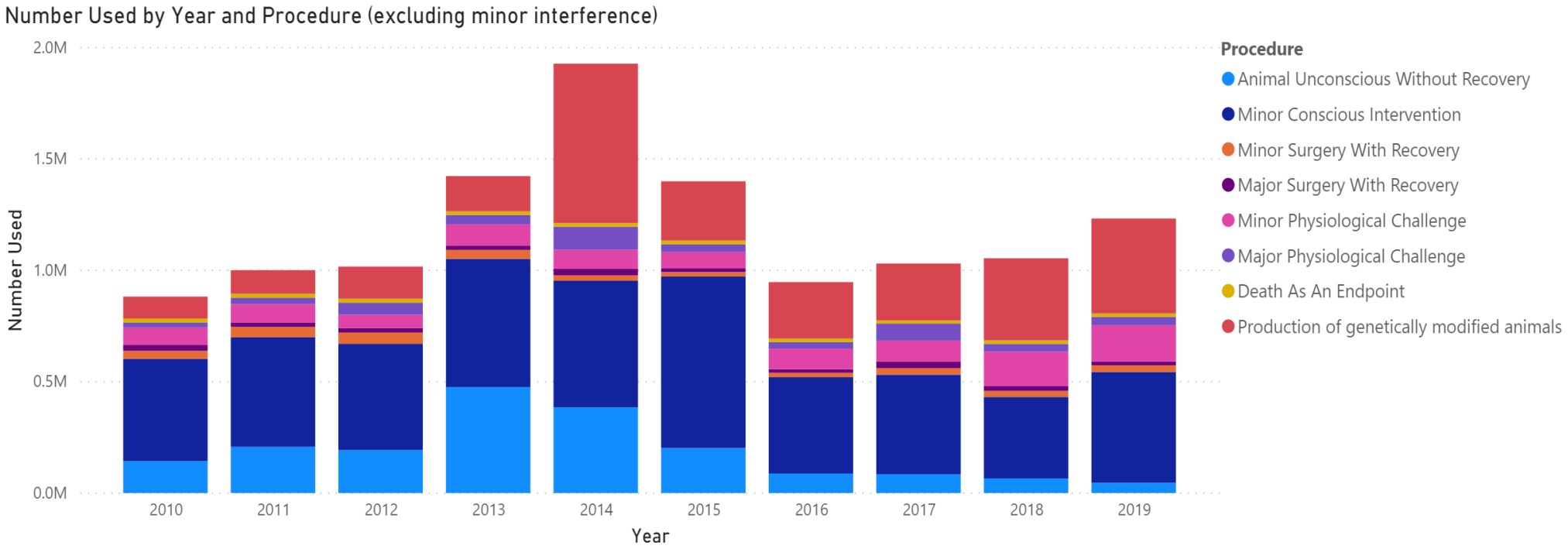
Purpose	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	**Total**
Stock Breeding	75,867	65,936	75,488	80,774	47,116	259,464	263,601	300,720	366,997	452,258	**1,988,221**
Stock Maintenance	27,165	33,850	15,448	7,890	10,500	26,508	13,684	13,204	7,266	5,875	**161,390**
Education	43,344	41,230	40,904	34,960	68,717	135,378	39,301	36,904	114,387	105,110	**660,235**
Research: human or animal biology	158,880	189,450	882,024	218,541	201,636	161,990	2,839,488	190,934	211,778	239,190	**5,293,911**
Research: human or animal health and welfare	298,611	283,546	286,375	303,050	1,024,919	152,375	104,590	116,835	115,119	129,577	**2,814,997**
Research: animal management or production	71,722	94,019	81,831	227,769	76,422	91,603	111,880	167,998	616,015	133,914	**1,673,173**
Research: environmental study	901,504	1,402,726	813,500	1,411,046	1,247,301	5,341,812	1,539,475	1,725,808	694,037	448,737	**15,525,946**
Production of biological products	19,568	74,625	78,419	109,229	28,870	54,811	42,890	55,365	90,866	99,497	**654,140**
Diagnostic procedures	3,630	8,540	1,994	1,031	1,310	766	1,307	1,134	18,186	2,959	**40,857**
Regulatory product testing	42,302	24,540	60,945	305,242	275,885	62,770	21,039	16,557	19,292	33,191	**861,763**
Total	**1,642,593**	**2,218,462**	**2,336,928**	**2,699,532**	**2,982,676**	**6,287,477**	**4,977,255**	**2,625,459**	**2,253,943**	**1,650,308**	**29,674,633**

2.3 Number of animals used over time by research procedure



Procedure	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	**Total**
Observation Involving Minor Interference	760,592	1,217,773	1,320,108	1,276,501	1,054,859	4,887,636	4,030,182	1,595,028	1,199,738	417,598	**17,760,015**
Animal Unconscious Without Recovery	143,155	207,753	192,840	475,557	384,503	201,919	87,443	83,924	65,595	46,055	**1,888,744**
Minor Conscious Intervention	459,712	491,747	477,377	576,018	568,416	769,829	432,697	447,324	365,928	497,414	**5,086,462**
Minor Surgery With Recovery	35,765	46,839	50,552	40,145	24,794	20,530	19,838	29,891	27,274	30,312	**325,940**
Major Surgery With Recovery	25,823	19,643	19,514	18,105	28,592	16,722	16,082	28,436	20,872	17,092	**210,881**
Minor Physiological Challenge	79,070	82,309	60,350	96,384	85,842	73,319	92,516	94,184	155,830	162,130	**981,934**
Major Physiological Challenge	22,625	28,614	54,411	42,647	103,859	34,489	29,148	77,292	34,121	37,880	**465,086**
Death As An Endpoint	17,465	17,767	17,445	15,997	16,351	16,771	15,741	13,982	15,551	15,525	**162,595**
Production of genetically modified animals	98,386	106,017	144,331	158,178	715,460	266,262	253,608	255,398	369,034	426,302	**2,792,976**
Total	**1,642,593**	**2,218,462**	**2,336,928**	**2,699,532**	**2,982,676**	**6,287,477**	**4,977,255**	**2,625,459**	**2,253,943**	**1,650,308**	**29,674,633**

2.4 Number of animals used over time by research procedure excluding "Observation involving minor interference"



Procedure	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	**Total**
Animal Unconscious Without Recovery	143,155	207,753	192,840	475,557	384,503	201,919	87,443	83,924	65,595	46,055	**1,888,744**
Minor Conscious Intervention	459,712	491,747	477,377	576,018	568,416	769,829	432,697	447,324	365,928	497,414	**5,086,462**
Minor Surgery With Recovery	35,765	46,839	50,552	40,145	24,794	20,530	19,838	29,891	27,274	30,312	**325,940**
Major Surgery With Recovery	25,823	19,643	19,514	18,105	28,592	16,722	16,082	28,436	20,872	17,092	**210,881**
Minor Physiological Challenge	79,070	82,309	60,350	96,384	85,842	73,319	92,516	94,184	155,830	162,130	**981,934**
Major Physiological Challenge	22,625	28,614	54,411	42,647	103,859	34,489	29,148	77,292	34,121	37,880	**465,086**
Death As An Endpoint	17,465	17,767	17,445	15,997	16,351	16,771	15,741	13,982	15,551	15,525	**162,595**
Production of genetically modified animals	98,386	106,017	144,331	158,178	715,460	266,262	253,608	255,398	369,034	426,302	**2,792,976**
Total	**882,001**	**1,000,689**	**1,016,820**	**1,423,031**	**1,927,817**	**1,399,841**	**947,073**	**1,030,431**	**1,054,205**	**1,232,710**	**11,914,618**

3. Purpose, Procedure and Species Charts 2019

3.1 Numbers of animals by Purpose and Procedure categories 2019

Numbers of animals by Purpose and Procedure categories 2019

Purpose	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Major Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Death As An Endpoint	Production of genetically modified animals	**Total**
Stock Breeding	31,541	49	27,503	35	8				393,122	**452,258**
Stock Maintenance	1,154	625	2,822	181	1,091	2				**5,875**
Education	82,560	4,481	16,970	897	143	59				**105,110**
Research: human or animal biology	37,818	12,744	99,200	16,800	11,431	30,986	15,568		14,643	**239,190**
Research: human or animal health and welfare	9,503	4,799	69,589	4,725	4,364	9,906	8,154		18,537	**129,577**
Research: animal management or production	40,027	9,206	66,507	2,921		13,345	1,908			**133,914**
Research: environmental study	214,175	13,733	210,164	385	55	135	10,090			**448,737**
Production of biological products	51	6	719	4,221		94,417	83			**99,497**
Diagnostic procedures	129	155	1,111	6		6	1,552			**2,959**
Regulatory product testing	640	257	2,829	141		13,274	525	15,525		**33,191**
Total	**417,598**	**46,055**	**497,414**	**30,312**	**17,092**	**162,130**	**37,880**	**15,525**	**426,302**	**1,650,308**

3.2 Number of animals and species used for Purpose: Stock Breeding 2019

Number of animals used for Purpose: Stock Breeding in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Major Surgery With Recovery	Production of genetically modified animals	Total
Amphibians	**2,201**		**43**				**2,244**
Amphibians	2,201		43				**2,244**
Aquatic animals	**517**		**19**			**10,215**	**10,751**
Fish	517		19			10,215	**10,751**
Birds			**518**				**518**
Native Captive			518				**518**
Domestic mammals	**663**		**1,579**				**2,242**
Cats	67						**67**
Cattle			15				**15**
Dogs	71						**71**
Pigs	25						**25**
Sheep	500		1,564				**2,064**
Laboratory mammals	**27,684**	**49**	**25,303**	**35**	**8**	**382,907**	**435,986**
Guinea Pigs	405						**405**
Mice	24,288	48	25,297	35	8	382,071	**431,747**
Rabbits	59						**59**
Rats	2,932	1	6			836	**3,775**
Native mammals	**25**		**41**				**66**
Native rats and mice	25		41				**66**
Primates	**16**						**16**
Baboons	16						**16**
Reptiles	**435**						**435**
Lizards	330						**330**
Turtles and Tortoises	105						**105**
Total	**31,541**	**49**	**27,503**	**35**	**8**	**393,122**	**452,258**

3.3 Number of animals and species used for Purpose: Stock Maintenance 2019

Number of animals used for Purpose: Stock Maintenance in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Major Surgery With Recovery	Minor Physiological Challenge	Total
Aquatic animals	**151**	**14**					**165**
Fish	151	14					**165**
Birds	**600**						**600**
Poultry	600						**600**
Domestic mammals	**2**					**2**	**4**
Cattle						2	**2**
Dogs	2						**2**
Laboratory mammals	**251**	**611**	**2,822**	**181**	**1,091**		**4,956**
Mice	196	600	2,822		1,091		**4,709**
Rats	55	11		181			**247**
Reptiles	**150**						**150**
Lizards	150						**150**
Total	**1,154**	**625**	**2,822**	**181**	**1,091**	**2**	**5,875**

3.4 Number of animals and species used for Purpose: Education 2019

Number of animals used for Purpose: Education in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Major Surgery With Recovery	Minor Physiological Challenge	Total
Amphibians	**428**	**1,267**					**1,695**
Amphibians	428	1,267					**1,695**
Aquatic animals	**744**	**1,125**	**85**	**151**			**2,105**
Crustaceans (reporting not mandatory)	2						**2**
Fish	742	1,125	85	151			**2,103**
Birds	**76,334**	**723**	**1,003**			**40**	**78,100**
Exotic Captive	5	45	13				**63**
Exotic Wild	4,755						**4,755**
Native Captive	92						**92**
Native Wild	15,102		64				**15,166**
Other birds	165						**165**
Poultry	56,215	678	926			40	**57,859**
Domestic mammals	**3,900**	**251**	**13,932**	**357**	**19**	**10**	**18,469**
Cats	293		23	2			**318**
Cattle	1,728	1	2,857	101	11		**4,698**
Dogs	263		674	18			**955**
Goats	13		1				**14**
Horses	253	1	373	217		4	**848**
Other domestic mammals	9		5				**14**
Pigs	30	148	340				**518**
Sheep	1,311	101	9,659	19	8	6	**11,104**
Exotic feral mammals	**15**		**1**				**16**
Dingo/Wild Dogs	1						**1**
Foxes	5						**5**
Mice	5		1				**6**
Other exotic feral mammals	1						**1**
Rats	3						**3**
Exotic zoo animals	**6**						**6**
Exotic zoo animals	6						**6**
Laboratory mammals	**142**	**1,115**	**1,495**	**389**	**124**	**9**	**3,274**
Guinea Pigs	44	18	9				**71**
Mice	50	402	1,195	382	40		**2,069**
Rabbits	36	2	5				**43**
Rats	12	693	286	7	84	9	**1,091**
Native mammals	**761**		**57**				**818**
Bats	12		2				**14**
Dasyurids	109		48				**157**
Koalas	11						**11**
Macropods	174						**174**
Monotremes	9						**9**
Native rats and mice	82		7				**89**
Possums and gliders	202						**202**
Whales and dolphins	162						**162**
Reptiles	**230**		**397**				**627**
Lizards	125		375				**500**
Other reptiles	30		6				**36**
Snakes	59		16				**75**
Turtles and Tortoises	16						**16**
Total	**82,560**	**4,481**	**16,970**	**897**	**143**	**59**	**105,110**

3.5 Number of animals and species used for Purpose: Research: Human or Animal Biology 2019

Number of animals used for Purpose: Research: human or animal biology in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Major Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Production of genetically modified animals	Total
Amphibians	**3,879**		**2,288**		**50**	**69**			**6,286**
Amphibians	3,879		2,288		50	69			**6,286**
Aquatic animals	**10,261**	**424**	**527**	**2,433**	**30**	**5,328**		**13,828**	**32,831**
Fish	10,261	424	527	2,433	30	5,328		13,828	**32,831**
Birds	**572**	**11**	**75,899**	**1,351**		**186**			**78,019**
Exotic Captive				2					**2**
Exotic Wild		11	6,670						**6,681**
Native Captive			959	7		149			**1,115**
Native Wild	572		66,415	117					**67,104**
Other birds						37			**37**
Poultry			1,855	1,225					**3,080**
Domestic mammals	**105**	**21**	**462**	**18**	**225**	**9**			**840**
Cats	16								**16**
Cattle	30		31						**61**
Dogs	15		301						**316**
Horses	44		30			9			**83**
Other domestic mammals		15							**15**
Pigs					31				**31**
Sheep		6	100	18	194				**318**
Exotic feral mammals	**20**	**17**	**202**						**239**
Cats			1						**1**
Foxes		17	84						**101**
Hares			1						**1**
Mice			2						**2**
Other exotic feral mammals			3						**3**
Rabbits			46						**46**
Rats	20		65						**85**
Exotic zoo animals	**121**		**2**						**123**
Exotic zoo animals	121		2						**123**
Laboratory mammals	**15,466**	**12,206**	**15,705**	**12,800**	**11,125**	**25,117**	**15,518**	**815**	**108,752**
Guinea Pigs		44	91	16					**151**
Mice	10,492	10,845	13,511	9,913	7,432	23,111	15,389	803	**91,496**
Rabbits		27	9	14	234				**284**
Rats	4,974	1,290	2,094	2,857	3,459	2,006	129	12	**16,821**
Native mammals	**6,312**		**962**	**185**		**97**			**7,556**
Bats	2,589			9					**2,598**
Dasyurids			4	7		71			**82**
Koalas			1	92					**93**
Macropods	543		280	57					**880**
Monotremes	4		19	11					**34**
Native rats and mice	95		209						**304**
Other native mammals	1		101						**102**
Possums and gliders	44		334	7		26			**411**
Whales and dolphins	3,035								**3,035**
Wombats	1		14	2					**17**
Primates	**26**			**4**	**1**				**31**
Baboons					1				**1**
Other primates	26			4					**30**
Reptiles	**1,056**	**65**	**3,153**	**9**		**180**	**50**		**4,513**
Lizards	515	65	1,143			180	50		**1,953**
Other reptiles	1		194	1					**196**
Snakes	29		26						**55**
Turtles and Tortoises	511		1,790	8					**2,309**
Total	**37,818**	**12,744**	**99,200**	**16,800**	**11,431**	**30,986**	**15,568**	**14,643**	**239,190**

3.6 Number of animals and species used for Purpose: Research: Human or Animal Health and Welfare 2019

Number of animals used for Purpose: Research: human or animal health and welfare in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Major Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Production of genetically modified animals	Total
Amphibians					**47**				**47**
Amphibians					47				**47**
Aquatic animals		**98**	**389**					**7,412**	**7,899**
Fish		98	389					7,412	**7,899**
Birds	**1,920**	**425**	**52,399**	**595**		**331**	**2,219**		**57,889**
Exotic Wild			97			18			**115**
Native Captive	11								**11**
Native Wild			38	595					**633**
Poultry	1,909	425	52,264			313	2,219		**57,130**
Domestic mammals	**6,842**	**83**	**8,768**	**256**	**149**	**751**	**32**		**16,881**
Cats	194		614	42			32		**882**
Cattle	872	6	2,291		10	113			**3,292**
Dogs	363		1,102	26	32	25			**1,548**
Horses	108		265	6		22			**401**
Other domestic mammals			2	18		80			**100**
Pigs	3,330	10	3,226		91				**6,657**
Sheep	1,975	67	1,268	164	16	511			**4,001**
Exotic feral mammals				**41**					**41**
Rats				41					**41**
Exotic zoo animals	**6**		**5**						**11**
Exotic zoo animals	6		5						**11**
Laboratory mammals	**615**	**4,108**	**7,653**	**3,670**	**4,164**	**8,748**	**5,892**	**11,125**	**45,975**
Guinea Pigs			66	10		77			**153**
Mice	615	4,084	7,475	2,632	3,630	8,372	5,560	11,125	**43,493**
Rabbits				81	13		92		**186**
Rats		24	112	947	521	299	240		**2,143**
Native mammals	**41**	**85**	**331**	**163**					**620**
Bandicoots				45					**45**
Bats			312						**312**
Dasyurids	18			21					**39**
Koalas			19	5					**24**
Macropods		85		2					**87**
Monotremes				1					**1**
Native rats and mice	19			44					**63**
Possums and gliders				43					**43**
Seals	4								**4**
Wombats				2					**2**
Primates					**4**	**2**			**6**
Baboons					4	2			**6**
Reptiles	**79**		**44**			**74**	**11**		**208**
Lizards	62		20			74			**156**
Turtles and Tortoises	17		24				11		**52**
Total	**9,503**	**4,799**	**69,589**	**4,725**	**4,364**	**9,906**	**8,154**	**18,537**	**129,577**

3.7 Number of animals and species used for Purpose: Research: Animal Management or Production

Number of animals used for Purpose: Research: animal management or production in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Total
Aquatic animals	**640**	**5,720**	**720**	**180**			**7,260**
Fish	640	5,720	720	180			**7,260**
Birds	**16,279**	**3,325**	**4,399**	**6**	**8,460**	**1,908**	**34,377**
Exotic Wild	3,640						**3,640**
Native Captive	27		147	6			**180**
Native Wild	4,309						**4,309**
Poultry	8,303	3,325	4,252		8,460	1,908	**26,248**
Domestic mammals	**20,519**	**155**	**61,388**	**2,733**	**4,866**		**89,661**
Cats	68						**68**
Cattle	3,904	5	6,439	3	603		**10,954**
Dogs	144		14		1		**159**
Goats	8,058		36	131			**8,225**
Horses	8		115		29		**152**
Other domestic mammals	44						**44**
Pigs	315		33,036	126			**33,477**
Sheep	7,978	150	21,748	2,473	4,233		**36,582**
Exotic feral mammals	**473**						**473**
Foxes	168						**168**
Hares	19						**19**
Mice	45						**45**
Pigs	3						**3**
Rabbits	238						**238**
Laboratory mammals		**6**					**6**
Guinea Pigs		6					**6**
Native mammals	**1,775**			**2**	**19**		**1,796**
Bandicoots	55						**55**
Dasyurids					19		**19**
Koalas				2			**2**
Macropods	1,500						**1,500**
Possums and gliders	40						**40**
Wombats	180						**180**
Reptiles	**341**						**341**
Lizards	341						**341**
Total	**40,027**	**9,206**	**66,507**	**2,921**	**13,345**	**1,908**	**133,914**

3.8 Numbers of animals and species used for Purpose: Research: Environmental Study 2019

Number of animals used for Purpose: Research: environmental study in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Major Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Total
Amphibians	**12,393**	**30**	**5,698**			**20**		**18,141**
Amphibians	12,393	30	5,698			20		**18,141**
Aquatic animals	**122,916**	**13,682**	**185,716**		**55**	**107**	**10,072**	**332,548**
Cephalopods (reporting not mandatory)	40							**40**
Crustaceans (reporting not mandatory)	2,641		2,579					**5,220**
Fish	120,235	13,682	183,137		55	107	10,072	**327,288**
Birds	**69,542**		**3,312**					**72,854**
Exotic Wild	495		62					**557**
Native Captive			143					**143**
Native Wild	69,047		3,059					**72,106**
Poultry			48					**48**
Domestic mammals	**59**	**21**	**32**					**112**
Cats			8					**8**
Cattle	17							**17**
Deer		21						**21**
Dogs	8		24					**32**
Horses	24							**24**
Other domestic mammals	10							**10**
Exotic feral mammals	**390**		**681**					**1,071**
Cats	34		6					**40**
Dingo/Wild Dogs	48		323					**371**
Foxes	109		43					**152**
Goats	31							**31**
Hares	10							**10**
Horses	3							**3**
Mice	31		153					**184**
Other exotic feral mammals	47		9					**56**
Pigs	30		17					**47**
Rabbits	24		10					**34**
Rats	23		120					**143**
Laboratory mammals	**12**							**12**
Mice	12							**12**
Native mammals	**6,538**		**9,167**	**385**		**8**		**16,098**
Bandicoots	78		97					**175**
Bats	831		3,732	38				**4,601**
Dasyurids	393		1,529	17				**1,939**
Koalas	64		62	53				**179**
Macropods	2,823		1,119	71				**4,013**
Monotremes	39		13	206				**258**
Native rats and mice	847		1,718					**2,565**
Other native mammals	248		141					**389**
Possums and gliders	954		756			8		**1,718**
Whales and dolphins	10							**10**
Wombats	251							**251**
Reptiles	**2,325**		**5,558**				**18**	**7,901**
Lizards	1,987		3,288				18	**5,293**
Snakes	192		133					**325**
Turtles and Tortoises	146		2,137					**2,283**
Total	**214,175**	**13,733**	**210,164**	**385**	**55**	**135**	**10,090**	**448,737**

3.9 Number of animals and species used for Purpose: Production of Biological Products 2019

Number of animals used for Purpose: Production of biological products in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Total
Aquatic animals				**3,580**			**3,580**
Fish				3,580			**3,580**
Birds	**46**		**9**		**94,267**		**94,322**
Native Wild	46						**46**
Poultry			9		94,267		**94,276**
Domestic mammals		**6**	**281**	**445**	**141**		**873**
Cats				7			**7**
Cattle		6		13	6		**25**
Dogs			219		85		**304**
Horses			3	255	11		**269**
Other domestic mammals			2				**2**
Sheep			57	170	39		**266**
Laboratory mammals	**4**			**52**	**9**	**83**	**148**
Rabbits	4				9	83	**96**
Rats				52			**52**
Reptiles	**1**		**429**	**144**			**574**
Lizards	1						**1**
Snakes			429	144			**573**
Total	**51**	**6**	**719**	**4,221**	**94,417**	**83**	**99,497**

3.10 Number of animals and species used for Purpose: Diagnostic Procedures 2019

Number of animals used for Purpose: Diagnostic procedures in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Total
Birds	**10**		**10**			**1,540**	**1,560**
Native Captive			10				**10**
Poultry	10					1,540	**1,550**
Domestic mammals	**32**		**1,101**	**6**			**1,139**
Cats			16				**16**
Cattle			4				**4**
Dogs	16		29	6			**51**
Horses	16		2				**18**
Sheep			1,050				**1,050**
Laboratory mammals	**87**	**155**			**6**	**12**	**260**
Mice	71	130				12	**213**
Rats	16	25			6		**47**
Total	**129**	**155**	**1,111**	**6**	**6**	**1,552**	**2,959**

3.11 Number of animals and species used for Purpose: Regulatory Product Testing 2019

Number of animals used for Purpose: Regulatory product testing in 2019

Species group	Observation Involving Minor Interference	Animal Unconscious Without Recovery	Minor Conscious Intervention	Minor Surgery With Recovery	Minor Physiological Challenge	Major Physiological Challenge	Death As An Endpoint	Total
Birds					**12,334**			**12,334**
Poultry					12,334			**12,334**
Domestic mammals	**640**	**257**	**898**	**141**	**833**	**525**		**3,294**
Cats	20		80		22			**122**
Cattle	387	221	452	129	416			**1,605**
Dogs	29		124		25			**178**
Horses	58		13					**71**
Pigs	22	18	18	12	12			**82**
Sheep	124	18	211		358	525		**1,236**
Exotic feral mammals					**107**		**13**	**120**
Goats							11	**11**
Other exotic feral mammals							2	**2**
Pigs					107			**107**
Laboratory mammals			**1,931**				**15,512**	**17,443**
Guinea Pigs			343				1,068	**1,411**
Mice			631				14,444	**15,075**
Rabbits			685					**685**
Rats			272					**272**
Total	**640**	**257**	**2,829**	**141**	**13,274**	**525**	**15,525**	**33,191**

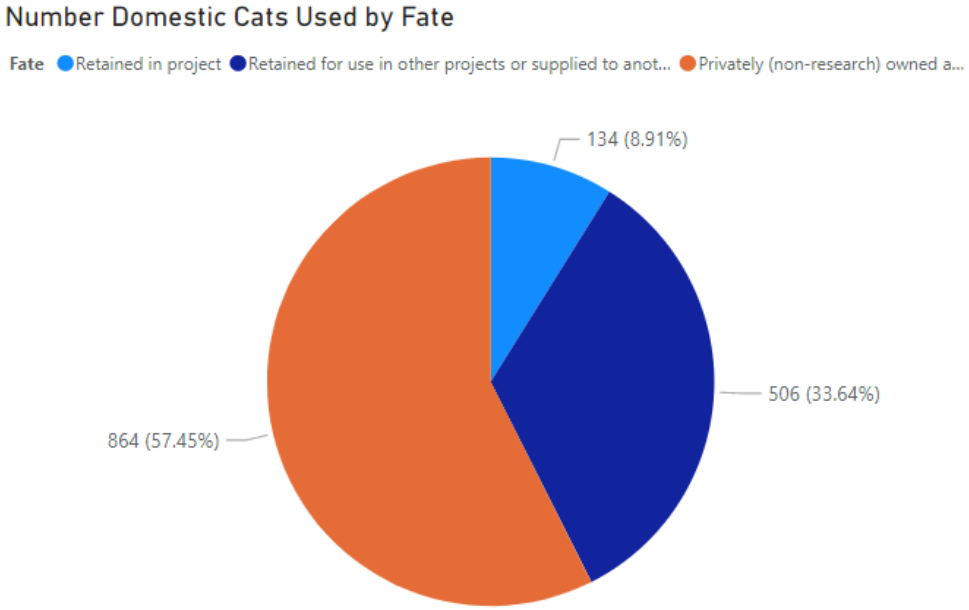
4. Fate of animals

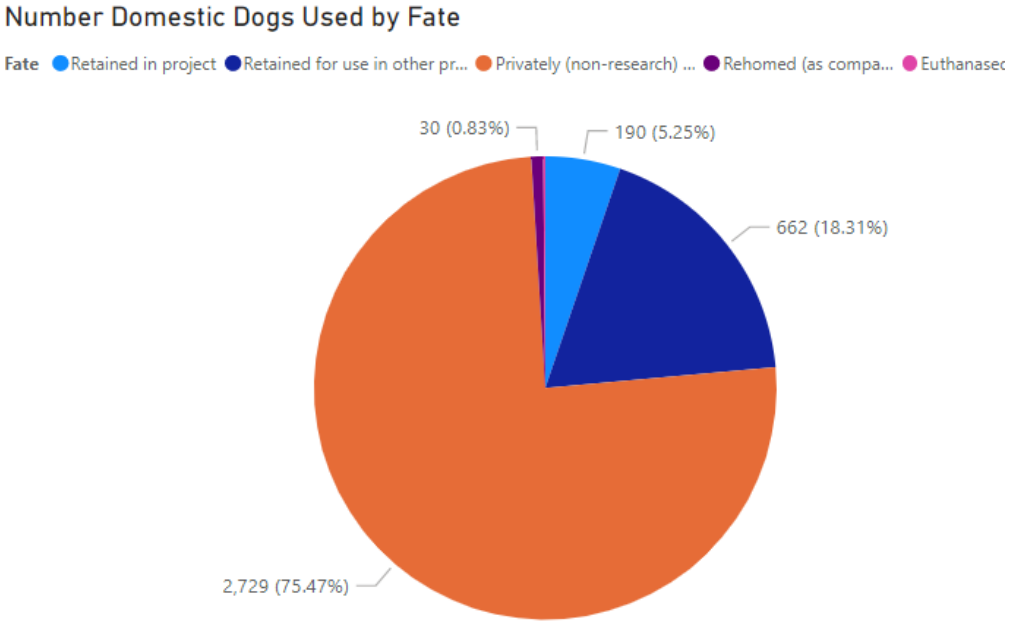
From the 2019 reporting year onwards, reporting on the Fate of animals category is mandatory for the use of domestic cats and dogs, and voluntary for other species.

For domestic cats and dogs the reporting shows:

- The majority of domestic cats and dogs were privately (non-research) owned and remained with the owners (57% (864) domestic cats and 75% (2,729) domestic dogs).
- The remaining 43% (640) of domestic cats were retained in projects or retained for use in other projects or supplied to another establishment/ individual for research.
- Of the remaining 25% (887) of domestic dogs, 24% (852) were retained in projects or retained for use in other projects or supplied to another establishment/ individual for research.
- 30 domestic dogs were rehomed.
- 5 domestic dogs were euthanased or died unrelated to the project.

4.1 Number of domestic cats and domestic dogs used by category: Fate of animals



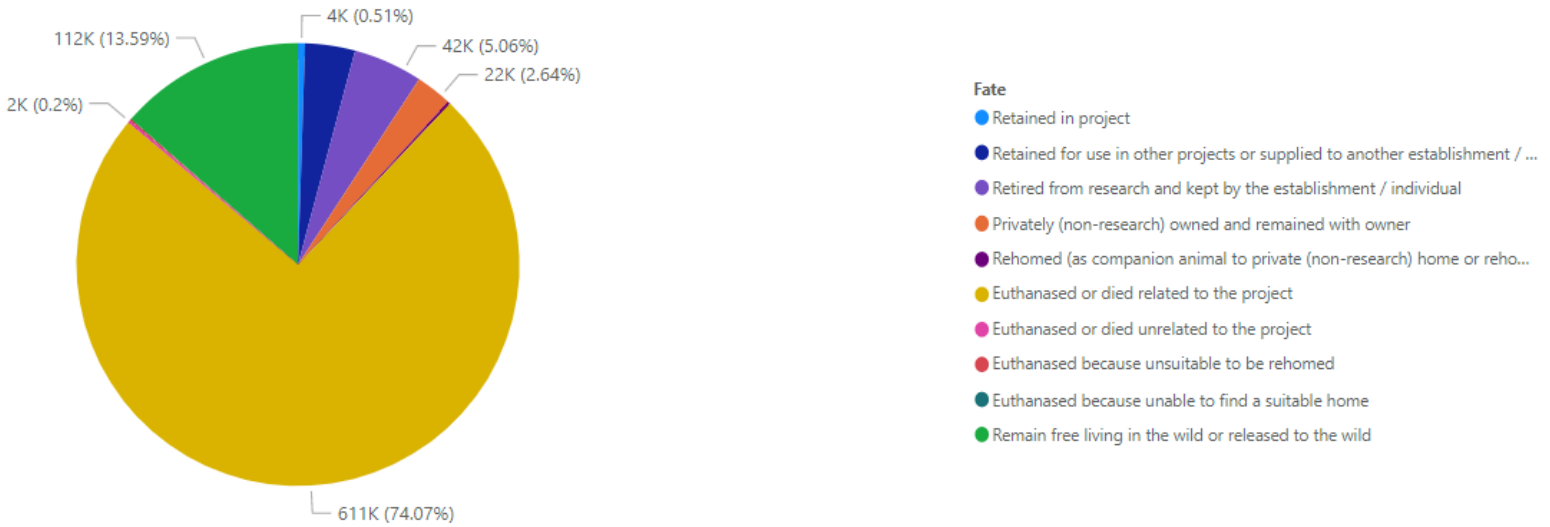


Fate	Cats	Dogs	**Total**
Retained in project	134	190	**324**
Retained for use in other projects or supplied to another establishment / individual for research	506	662	**1,168**
Privately (non-research) owned and remained with owner	864	2,729	**3,593**
Rehomed (as companion animal to private (non-research) home or rehoming organisation)		30	**30**
Euthanased or died unrelated to the project		5	**5**
Total	**1,504**	**3,616**	**5,120**

4.2 Number of animals used by Category: Fate of animals 2019

(Voluntary reporting for species other than domestic cats and dogs)

Number Used by Fate

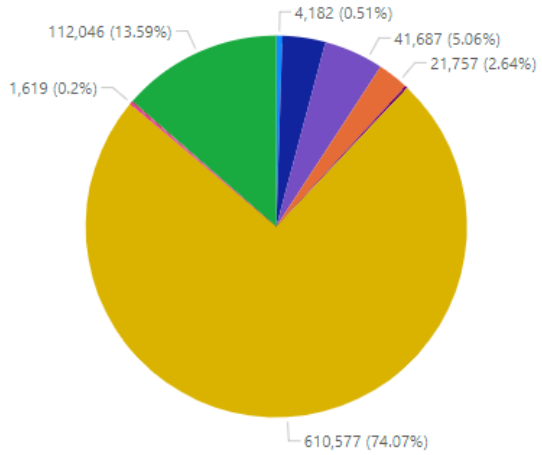


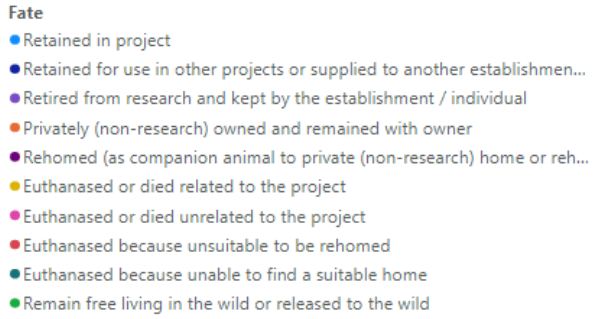
Fate	2019	**Total**
Retained in project	4,182	**4,182**
Retained for use in other projects or supplied to another establishment / individual for research	30,066	**30,066**
Retired from research and kept by the establishment / individual	41,687	**41,687**
Privately (non-research) owned and remained with owner	21,757	**21,757**
Rehomed (as companion animal to private (non-research) home or rehoming organisation)	1,538	**1,538**
Euthanased or died related to the project	610,577	**610,577**
Euthanased or died unrelated to the project	1,619	**1,619**
Euthanased because unsuitable to be rehomed	853	**853**
Euthanased because unable to find a suitable home	17	**17**
Remain free living in the wild or released to the wild	112,046	**112,046**
Total	**824,342**	**824,342**

4.3 Number and species groups used by Category: Fate of animals 2019

(Voluntary reporting for species other than domestic cats and dogs)

Number Used by Fate and Species Group





Fate	Amphibians	Aquatic animals	Birds	Domestic mammals	Exotic feral mammals	Exotic zoo animals	Laboratory mammals	Native mammals	Primates	Reptiles	Total
Retained in project	2,244	289	264	422			127	187	16	633	**4,182**
Retained for use in other projects or supplied to another establishment / individual for research	64	20	144	10,146		16	19,610	20	6	40	**30,066**
Retired from research and kept by the establishment / individual	10			41,608			26	43			**41,687**
Privately (non-research) owned and remained with owner			26	21,711	2	11		7			**21,757**
Rehomed (as companion animal to private (non-research) home or rehoming organisation)		102	1,320	102			14				**1,538**
Euthanased or died related to the project	503	52,173	120,538	4,660	51		432,471	16	1	164	**610,577**
Euthanased or died unrelated to the project		481	693	345			4	88		8	**1,619**
Euthanased because unsuitable to be rehomed			853								**853**
Euthanased because unable to find a suitable home					17						**17**
Remain free living in the wild or released to the wild	15,066	50,914	31,598	832	288			7,754		5,594	**112,046**
Total	**17,887**	**103,979**	**155,436**	**79,826**	**358**	**27**	**452,252**	**8,115**	**23**	**6,439**	**824,342**

5. Lethality testing

The *Animal Research Act 1985* defines a 'lethality test' as *'an animal research procedure in which any material or substance is administered to animals for the purpose of determining whether any animals will die or how many animals will die'*. Lethality tests include, but are not limited to, LD50 tests.

The following are the figures reported on animal use for lethality testing in 2019.

Species	Number used	Number died (not euthanased)	Number euthanased as early endpoint	Procedure	Justification	Alternatives
Guinea Pigs	1046	154	61	Clostridium chauvoei Potency by Challenge in guinea pigs. Vaccinated animals are challenged with test organism in order to demonstrate protection and hence vaccine efficacy.	Regulatory testing required to demonstrate efficacy (potency) of vaccines prior to release. Assessment of in-process or development material to determine suitability for further manufacture.	An in vitro project is currently in place with an ELISA for finished product potency testing of C chauvoei having been developed and validated. This test is available for transfer into the QC Laboratories for routine testing however Regulatory Approval from the APVMA is still pending. It is envisaged that this will be completed by the end of Quarter 4 2020 subject to Regulatory Approval being received.

Species	Number used	Number died (not euthanased)	Number euthanased as early endpoint	Procedure	Justification	Alternatives
Mice	8814	1685	451	Serum neutralisation test in mice: Susceptible animals are challenged with test toxin/antibody dilutions to determine antibody titre.	Regulatory testing required to demonstrate efficacy (potency) of vaccines prior to release. Testing of stability batches and new product formulations.	This test is based upon regulatory guidelines. An in vitro project is currently in place with the objective of replacing animal tests with in vitro tests. Replacement ELISAs for finished product potency testing of C.Septicum, C prefringens Type D and C tetani are being transferred into the QC Laboratories to be used for routine testing by the end of Quarter 3 2020.
Mice	2578	694	139	Total Combining Power test in mice: Susceptible animals are challenged with test antigen/toxin/antibody dilutions to determine potency of antigen preparations.	In-process testing of vaccine constituents to allow evaluation of suitability for further manufacture.	This test is based upon regulatory requirements for the assessment of in-process products. There are no alternatives available at this time however the establishment has embarked on a long-term program to develop in vitro assays which may be used to replace existing in vivo assays

Species	Number used	Number died (not euthanased)	Number euthanased as early endpoint	Procedure	Justification	Alternatives
						subject to requlatory approval of these replacement assavs.
Mice	1312	324	71	L + titration in mice: Susceptible animals are challenged with test toxin in order to determine potency of antigen preparation.	In-process testing of production and development antigen growths to allow stop/go decision during manufacturing process.	This test is based upon regulatory requirements for the assessment of in-process products. There are no alternatives available at this time however the establishment has embarked on a long-term program to develop in vitro assays which may be used to replace existing in vivo assays subject to requlatorv approval of these replacement assays.
Mice	10	4	0	Minimum Lethal Dose in mice. Susceptible animals are challenged with test toxin in order to determine the toxicity of a toxin preparation.	The Minimum Lethal Dose assay is used for the standardisation of test reagents (toxin) and validation of the Serum Neutralisation and Total Combining Power assays.	This test is based upon regulatory requirements for the assessment of in-process products. There are no alternatives currently available.

Species	Number used	Number died (not euthanased)	Number euthanased as early endpoint	Procedure	Justification	Alternatives
Feral Deer	Individual animals cannot be accurately identified at camera trap sites therefore the number of camera events are recorded. For the relevant reporting period camera events recorded per species were: Fallow deer (493) Red deer (3) Sambar deer (103).	2	0	Field trials using a targeted feeding structure and non-lethal or lethal bait types.	Negative impacts associated with overabundant pest herbivore species are well accepted, with feral goats *Capra hircus* listed as a key threatening process under the Commonwealth *Environment Protection and Biodiversity Conservation Act 1999*. Additionally, competition and habitat degradation by feral goats and herbivory and environmental degradation caused by feral deer are listed as Key Threatening Processes in New South Wales under Schedule 4 of the *Biodiversity Conservation Act 2016*. Despite these listings, control techniques for these species appear limited, particularly when compared to the number of techniques available for other pest species. Ongoing field trials continue to identify the potential for further development of a target selective, humane and cost-efficient method as an additional technique for controlling	The purpose of this research is to devise a humane method for killing free-living feral species. There are no alternatives to lethality testing.

Species	Number used	Number died (not euthanased)	Number euthanased as early endpoint	Procedure	Justification	Alternatives
					overabundant herbivore pest species .	
Feral Goats	Individual animals cannot be accurately identified at camera trap sites therefore the number of camera events are recorded. For the relevant reporting period camera events recorded per species were: Feral goat (694)	11	0	Field trials using a targeted feeding structure and non-lethal or lethal bait types.	Negative impacts associated with overabundant pest herbivore species are well accepted, with feral goats *Capra hircus* listed as a key threatening process under the Commonwealth *Environment Protection and Biodiversity Conservation Act 1999*. Additionally, competition and habitat degradation by feral goats and herbivory and environmental degradation caused by feral deer are listed as Key Threatening Processes in New South Wales under Schedule 4 of the *Biodiversity Conservation Act 2016*. Despite these listings, control techniques for these species appear limited, particularly when compared to the number of techniques available for other pest species. Ongoing field trials	The purpose of this research is to devise a humane method for killing free-living feral species. There are no alternatives to lethality testing.

Species	Number used	Number died (not euthanased)	Number euthanased as early endpoint	Procedure	Justification	Alternatives
					continue to identify the potential for further development of a target selective, humane and cost-efficient method as an additional technique for controlling overabundant herbivore pest species.	

6. Examples of methods used to implement the '3Rs'

The following are practical examples of strategies used to implement the '3Rs' (Replacement, Reduction and Refinement in animal use). These examples have all been directly reported by accredited animal research establishments for the 2019 reporting year. Examples relate to 'Replacement' (of animals with other methods), 'Reduction' (in the number of animals used in specific protocols) and 'Refinement' (of techniques used to reduce the impact on animals).

6.1 Replacement

- Projects benefit from donations and exchanges with other institutions, thereby minimising the need to collect live animals in the field. An increasing number of projects are using tissue samples only (often donated from other institutions - e.g. zoos, collected during routine health inspections of animals).
- A researcher has recently been exploring the possibility of using Environmental DNA (eDNA) to minimise the use of live animals in studies. eDNA research consists of sampling surface seawater and filtering free-floating genetic material shed by animals (e.g. fish scales, mucus) to investigate the presence/absence of particular taxa or species within the bodies of water sampled. This is a non-invasive methodology (i.e. no interaction with, disturbance or harm caused to animals) that relies primarily on state-of-the-art DNA metabarcoding of the filtered materials.
- We have developed an ex vivo system to investigate interactions between tumour cells and cells of the bone microenvironment to minimise the impact of intervention studies on live animals.
- We have begun to use ex vivo explants of mouse calvaria which will reduce the impact of tumour burden on these mice as it will replace in vivo studies to some extent.
- Where possible excess tumour tissue will be used for additional analysis such as the tissue engineering platform.
- We have been using in vitro cell cultures in order to study some aspects of cardiac development, which might replace, at least in part, some of our mice usage.
- We have undertaken many in vitro assays to assess the function of MIC-1/GDF15. These have included assessing the effects of MIC-1/GDF15 on cancer cell differentiation, proliferation, apoptosis and tumour cytotoxicity.
- We will only undertake animal experiments to determine if in vitro observations also apply in vivo, where it is not possible to do the work in vitro, or where it seems likely that unexpected or unpredicted results may occur in vivo.

- We will perfuse organs within mouse studies to enable us to come back to different tissue samples and we will make a tissue library/ biobank for other researchers to investigate proteins of interest if required. In vitro cytotoxicity assays and ex vivo antigen presentation/suppression assays will be used for initial testing of therapies not tested before, previous to testing in in vivo models.
- Wherever possible we use patient-derived lymphoblast cell lines, or primary cultures of human monocytes from MKD patients, to perform biochemical and molecular studies on defective protein prenylation and reduce the use of mice.
- We will supplement our data with in vitro techniques in order to reduce our reliance on animal tissue.
- An expert on alternatives remains an advisor to the AEC on alternatives to the use of animals and his services and advice are available to all research staff and students undertaking animal based research.
- The establishment promotes the use of replacements through its Animal Ethics Newsletter 4-5 times per year, its website and through other targeted activities.
- Continued use of technology such as GIS and programs such as the Atlas of living Australia assists in targeting areas or species, reducing the need to undertake some work and in some cases totally replacing it.
- Building strong networks with conservation organisations to replace wild caught animals with animals from captive bred populations for species reintroduction projects.
- Blood samples are used from clinical cases that have been cleared of being infectious.
- Models for anatomical and clinical examinations are being reviewed constantly. Where appropriate these are used to replace the use of animals.
- The use of AD instruments, videos and invertebrates for teaching projects.
- The establishment established a grant scheme with a total funding of $150,000 to support science-based projects with a primary goal of replacing, reducing or refining the use of animals in biomedical research. Several applications were submitted with funding to be awarded in 2020.
- Use of artificial models, e.g. Koken/ Curvet rat, knitted mouse models for training.
- Use of videos and on-line resources for training.
- Mannequins, audio-visual materials, photographs, taxidermied and preserved specimens were used as substitutes for live animals.
- Use of instructional aids for general identification and research of native animals.
- Use of mechanical horse.
- Bandaging and health care procedures are performed on dummies.
- Life size fibreglass replica of horse used for demonstrations.

- Use of photos and wool samples to identify breeds of sheep.
- Use of industry sites where animals are normally kept
- Visit to zoos, aquariums and museums to familiarise students with a range of native animals, eliminating the need for field visits or trapping.
- Use of horse legs from knackery for hoof trimming and basic shoeing.
- Use of cardboards and leather to practise injections and ear tagging.
- Use of condition scoring mannequins to replace the use of live sheep.
- Use of distance education methodology.
- Use of case study data to replace the need for capturing live animals.
- Use of identification tags without live sheep for simulation.
- The use of in vitro kits to study the metabolism of novel radiotracers continued to be implemented. The kits provide an alternative to characterising metabolic fate of drugs in live animals.
- Numerous studies have been carried out using cell-based models of disease.
- The committee challenged researchers to investigate alternative methodologies that do not involve the use of animals for optimising/ascertaining detection limits in radiotracer studies.
- Visual census techniques of fish trophic interactions in the field are a form of replacement for more destructive and intrusive methods such as sacrificing individuals for gut contents analysis.
- Developing and implementing ELISA testing to replace animal testing.
- In the process of removing animal testing completely from our New Zealand product test for Yersinia for deer.
- A number of meta-analyses are conducted which allow research questions to be addressed using existing data replacing the use of live animals and is more powerful than individual studies.
- We worked to find a replacement for the use of guinea pig ileum in Pharmacology practical classes. Through this search, discarded bovine trachea sourced from an abattoir was selected as a suitable tissue for testing. Future work on this project will be to test tissue viability beyond one month and testing using a wider range of compounds. The protocols developed here for dissection, cryopreservation, and smooth muscle experimentation could be applied to the use of other tissues in research and teaching activities.
- Mice have been significantly replaced/reduced in a study due to use of in vitro culture methods to study the Sorcs1 mutations. Only C5781/6J wildtype mice ended up being necessary for experimental use in the project, as our in vitro cell culture in the mouse and rat beta cell lines allowed us to study the BTBR mutation.

- We have successfully created knockout adipocyte cell lines for some of the strains on a protocol, such as ABHD15. This has allowed us to do some of our assays in cells rather than animals. This also enables us to refine which assays are necessary in a particular knockout animal.
- We have replaced the use of live prey animals with commercially bought prey mimics (chicken neck and hen eggs) and/or road killed amphibians for trials that assess feeding behaviour in adult monitors.
- Use of animals that died of natural causes rather than using samples from live animals.
- Use of cell lines, cultivated from tissue obtained from animals that died of natural causes rather than testing on live animals to examine the effect of contaminants.
- Replacement of animals with 3D bio-printed cell culture models of cancer which can mimic the tumour microenvironment.
- Dog Abdominal Surrogate for Instructional Exercise (DASIE) are used for suture training.
- Use of bio-models in surgical training workshops.
- Pilot studies using in vitro techniques.
- The university is moving towards an increased use of computer simulations, and/or the use of deceased animals which have been professionally preserved. Only advanced level teaching activities now require the use of animals.
- Large photo cards have been used in practical classes instead of live animals to teach students how to identify freshwater fish species.
- Mouse cell line culture techniques were establishment for assessing the induction of cell death, replacing the need to utilise cells from animals.
- In vitro cell culture models based on rat brain cells were used in place of animals, in addition to bank tissue samples collected in previous years.
- Immortalised testicular and macrophage cell lines were used to provide proof-of-concept that blood macrophages can transfer infections.
- Immortalised somatic cell lines were used to confirm and validate the inducible viral vector technology.
- Air-liquid Interface (ALI) models of differentiated primary bronchial epithelial cells using human airway cells were used for experiments which did not require the presence of intact immune systems and investigations of the cellular recruitment of the innate or adaptive immune response.
- Conducted in vitro studies using human and mouse melanoma cell lines to test the synthetic version of the peptide sequences discovered during the study.
- Performed immunohistochemistry on existing stored tissue samples, and used a number of online databases to compare peptide characteristics in order to identify which peptides would be most suitable for drug conjugation.

- Developed colonic cell culture techniques to study the effects of cigarette smoke extract and/or other potential therapeutic compounds.
- Cell culture models were conducted in parallel with animal models to translate the findings into human tissue and minimise the number of animals required.
- A MeCap mouse cell line replaced live mice for some experiments.
- Cell lines were used to ensure the specificity of regents prior to use on mouse embryos.
- Developed an in silica model of the hypothalamus-pituitary and adrenal axis for the key adrenal signalling pathways to test hypotheses prior to in vivo work.
- Substituted human sperm donations for spermatozoa work.
- Phantoms (specifically made solutions in plastic tubes containing metabolic concentrations that partially mimic animal tissue) were used during the early phase of a project.
- We have identified the potential development of a world's first organotypic human 3D skin model to study GAS-host interactions. This model involves the use of human cell lines to establish human 3D skin and have immune cells incorporated in order to closely mimic GAS-infected skin in a clinical setting.
- Use of animal mannequins and cadavers for initial training sessions prior to the commencement of training with live animals.
- A fish researcher has started using a photo and video library to start answering questions relating to the behaviour and appearance of different species in intra and interspecific visual communication. This requires no additional need to collect animals at this stage.
- Use of audio-visual material such as videos, slides, interactive computer programs.
- Use of training models to teach techniques (e.g. latex rat for injections).
- Use of abattoir specimens and cadavers.
- Use of plant tissue as a replacement for animal tissue for certain enzymatic assays.
- Use of animals killed in road accidents.
- Researchers moving away from primary cultures and using stem cell differentiation.
- The in-vitro project progressed significantly in 2019. The application for C. chauvoei ELISA as replacement of the guinea pig challenge test was submitted to APVMA on 3 April 2019. This application is still in the assessment phase with the Regulator which is due to be completed in May 2020. It is still expected for the test to be implemented by end of 2020 pending no further requests by the APVMA.
- Staff working for Quality Control have been trained and qualified to perform the C. perfringens D, C. tetani and C. septicum in-vitro tests to replace serum neutralisation (SN) in mice. Due to delays by the supplier in getting critical equipment and software

into the QC department, the tech transfer process was postponed until January 2020. The implementation of all three in-vitro tests is expected to occur in Q1 2020.

- The final SN replacement test is for C. novyi B. There is one final critical reagent in development and is expected to be available by Q2 2020. Assay development shall commence immediately after all critical reagents are available.
- A protocol investigating cytoskeletal dynamics in neurons to drive axonal growth used cell culture for preliminary investigation of drug combination and the mechanisms of action, with only the used, final stage being tested in animal models to replicate human systems.
- A protocol for training research and facility staff in rodent injections used non-animal alternatives to learning in the initial stages of training, such as training videos. A total of 12 videos are used now during this training, prior to use of animal models.

6.2 Reduction

- When the projects involve directly contributing to Collections, efforts are made to only capture minimal number of animals from taxa that are poorly represented in the collections. Similarly, skeletal remains of 10 animals (4 species) were opportunistically collected for the mammal collection.
- Re-use of fistulated cattle across multiple studies.
- The same control dogs for were used for two studies thus reducing the number of dogs required for both studies. These were longer term studies and quite intensive so reducing the number of dogs was good.
- The new application form requires researchers to justify that the number of animals used is appropriate for the level of significance required, the minimum difference expected and the variability for the trait measured. Researchers are also suggested to consult with a bio-statistician.
- All poultry research now required to utilize the 'gene pesti' tool to calculate statistical power required for experimentation.
- We aim to generate a database of the cellular composition of diverse tissues in mouse models of human disease and their response upon therapeutic interventions and therefore will serve as a resource to guide further research. We envision that in the long run this “mouse cell atlas” will save the need to perform many experiments using animals.
- Where possible control groups will be shared.
- The most appropriate statistical analysis methods are employed when planning the experiment.
- We will utilise the information we are generating from the single cell analysis of patient primary and metastatic tumour samples to prioritise genes and pathways we will target in this project.
- The experimental designs used in this project have been calculated to minimise the number of animals used whilst maintaining adequate statistical power. Conditional systems also reduce the number of animals that will be used.

- In complex 2D and 3D tissue culture systems, we currently obtain rat tails and utilise these to create 3D-organotypic assays to partially mimic in vivo cancer progression, invasion and response to treatment whenever possible/appropriate for investigation. Utility of these intermediate 3D assays helps to inform in vivo investigation thereby minimising animal use at early stage of study.
- The scRNAseq data is a hugely efficient way of studying cells in tissues and uses far fewer mice than any other characterization study due to its ability to analyse the whole transcriptome i.e. all 22,000 genes are examined in parallel. Likewise the use of virus studies using AAV8-Sp7-Cre can look at the effects of silencing genes without developing and breeding knockout mouse lines that require very large numbers of mice.
- The use of whole body in vivo imaging will be used at all times, when appropriate for the experimental question/outcome. This significantly reduces the number of mice used because longitudinal studies can be run on a single cohort of mice.
- We will be able to monitor the tumour growth at different time points by measuring bioluminescence. So the same animal will be imaged at different time points without sacrificing the animal and this will reduce the number of animals four times (four time points).
- We will use the same mice for both echocardiography and micromanometry experiments rather than using separate cohorts.
- We will collect several tissues (e.g. fat, liver, muscle) from the same animals to allow a reduction in the total number of animals needed for studies.
- Multicolour flow cytometry will allow concomitant analysis of several parameters in every tissue or cell sample. This will maximise the amount of data obtained from a single experiment and therefore reduce the number of mice required. While not re-using the mice for different experiments, we will take advantage of all possible tissues obtainable in each experiment. For example, sera and peripheral blood mononuclear cells will be obtained from the same mice in one experiment and not from two separate ones; bone marrow from control mice can be extracted and frozen for use in other experiments.
- Use of the same background strain of all knockout mice in our laboratory means that all knockouts can be compared to the same group of wild type animals. This reduces the number of wild type animals we need to study.
- We put our animals through an extensive phenotypic analysis of energy homeostasis, whereby several parameters of energy homeostasis are measured in the same mouse. Procedures are spaced over an interval of 8 weeks, to allow recovery from each measurement. This reduces the number of animals we need to use in our experiments. Tissue is shared with researchers to maximise the use of each animal.
- Animal usage is reduced by using retroviral transduction of spleen cells to inactivate or overexpress a particular molecule in B or T cells instead of producing a corresponding GM line.

- Animal usage is reduced by determining key time points during immune responses to focus studies (eg days 9, 21, 28) rather than analysing at many different time points.
- We will generate transgenic constructs of which expression is monitored by muscle-specific fluorescence protein expression. This technique will enable us to enrich embryos that possibly carry integrated transgenes, contributing to reduce the number of fish that will be raised as founder candidates.
- We have reduced the number of sentinel mice held for health screening by using one mouse twice for sampling of blood by the saphenous vein technique and keeping the mouse for 6 months in place of sacrificing two mice over the same period for blood collection via cardiac bleeding.
- We are using human arterial tissue (endarterectomy specimens) to ascertain a role for myeloperoxidase in destabilization of atherosclerotic plaque. This will allow reducing the overall number of mice used to ascertain a causal relationship between arterial myeloperoxidase activity and formation of unstable atherosclerotic plaque.
- We will snap freeze or fix tissues we would not normally use, in case other colleagues would like to use them in the future.
- The establishment has a statistical consulting unit that is dedicated to providing free advice, support and assistance in experimental design to ensure animal group numbers are correct and will provid meaningful and statistically valid results, minimising animal waste.
- The presence of a statistician at each research ethics drop-in session ensures expert advice is provided on animal numbers and experimental design.
- A reduction on impact on some species in the wild through the continuation of partnerships with other organisations, consolidating monitoring and trapping of species and sharing data.
- The use of museum specimens and tissue sharing wherever possible.
- Increased promotion and requirement of pilot studies, well designed so that pilot results can contribute to a larger study if it goes ahead based on pilot results.
- Reduction in impact on wild animals by running pilots on captive populations before moving on to wild animals (for example transmitter trials).
- Students are placed within small groups where animals are used for surgical or other training. This reduces animal use.
- Breeding programs are designed and maintained to produce stock to order, to reduce numbers and overproduction.
- Genetically modified animals are bred for the desired genotype as far as possible to reduce numbers.
- Animals used for courses are shared between multiple participants to achieve the best learning outcome whilst reducing overall numbers of animals used.
- An ‘Animal Tissue Sharing Program’ is available to all researchers where animals have been humanely killed.

- The use of scavenged tissue is used where possible rather than capturing and sampling live flying foxes.
- The use of retired breeding stock (aged rats) as a terminal blood feed source for mosquitoes and bed bug colonies.
- All researchers are required to review sampling sizes and trapping efforts to minimise handling and capture of native animals. Particular attention is made to where there may be the requirement for relatively large numbers of individuals to be surveyed. Key projects that have carefully considered opportunities for reduction include reducing the number of microbats tagged for radio-tracking for Pilbara mining projects, limiting the number of tagged bats to the minimum necessary to obtain useful data.
- A protocol investigating compressive spinal cord injuring in neonatal and adult rats reduced their sample sizes to the minimum of n=3 for these experiments. Litter sizes were however quite large (n=13 and 14) and the project was able to utilise the extra animals by undertaking MRI (n=3) and transferring excess mums and pups to (n=6) to the training protocols.
- A protocol investigating the effects of cold water pollution on Native Australian Fish ensured that a Power analysis was run prior to conducting experiments, with further power analysis run during analysis after initial results to determine if the original sample size calculated for each group was accurate with the variance in population.
- A protocol developed for the breeding of RAGE, TLR4, and RAGE/TLR4 deficient mice ensures that mouse numbers are kept to the minimum required. The researchers meet with the Facility Manager regularly to consider the number of animals needed for experimental protocols, and the number of animals needed to maintain the colony.
- Statistical evidence that using fewer animals would provide a similar research outcome.
- Animals are not maintained longer than necessary. Researchers request for GM mouse strains to be cryopreserved or culled when no animal experimentation is expected to be done for the immediate period. Regular stocktake of rodent stocks held.
- Less sentinel animals are utilised with animal health monitoring being done on stock animals via dried blood spot sampling as survival procedure. BioResources only makes use of sentinels for blood sampling in immunodeficient mice models.
- AEC recommending use of pilot studies as appropriate.
- Animals used for training are only those designated for culling. Where practicable, dead animals are used to gain experience in technical procedures.
- Avoid performing unnecessary experimental work with animals by conducting a comprehensive review of existing data, available models and alternative experimental approaches before commencing or renewing research projects.
- Frequent communication with researchers so wildtype animals no longer needed are culled soon after genotyping is performed. Prompting researchers of quicker turnover of genotyping results where practicable.
- Tissue sharing for other studies where possible.
- Engage the researchers in consultation on minimizing the number of animals in the proposed project.

- Routine meetings with research groups for colony management to ensure breeding is optimised for experimental or maintenance production only, thereby minimising or eliminating the generation of unrequired animals.
- All animals underwent a health check prior to inclusion in the study. Some animals were excluded from the study based on their mastitis or general health status. This process ensured that only healthy animals were enrolled in the study, as a way of reducing the number of animals used. Up to 1,300 animals were approved to be used in the study; however, only 992 animals were included. From this, statistically significant results have been observed, therefore reducing the need for more animals to be used in future studies.
- The use of CP and UTC groups that were shared between two studies greatly reduced the number of animals that needed to be enrolled in either study.
- Ongoing support of a mailing list to facilitate tissue sharing among researchers, including researchers from other institutions requesting tissue.
- Use of animals that were humanely killed under another approved project for training and assay/in vitro testing.
- Transfer of animals (undesired genotype/sex) from one project to another as approved by the AEC.
- Re-use of animals among multiple projects when ethically justified and as approved by the AEC.
- Pilot studies to ensure the least number of animals are used to obtain statistically valid data.
- Use of in vivo imaging technology to perform repeated measurements on the same animals over the course of an experiment, resulting in a significant reduction of animals used.
- Combination of experiments so that only a single “control” group is used.
- Simulated penning of sheep by demonstration.
- The number of occasions that an animal is used is minimised e.g. lambs are tagged and drenched at the same time as normal management schedule.
- Students working with researchers from other establishments on native animal projects rather than duplicating their own projects.
- Use normal scheduled animal health husbandry routines for teaching activities.
- Weighing and husbandry of cattle are carried out as part of their normal, regular commercial schedule.
- Opportunistic field observations of native fauna.
- Maximum amount of data and student/animal contact is derived from each trapping in order to minimise trapping requirement.
- Only one animal brought in for a demonstration.

- Use of ultrasound machine to replay images from live animal and allows more students to observe procedures used on an animal.
- Use of work placements to reduce the need for students to use animals on campus.
- Re-use of rabbits in studies over 3 studies. Reduction from potential nine animals to three.
- Design of studies to incorporate shared control groups when conducted simultaneously.
- Biostatistician provided valuable advice and challenged investigators to statistically confirm the number of animals requested and to validate statistical data obtained from animal experimentation.
- Where possible, excess animal tissues are frozen and shared by other researchers for protein extraction, eliminating the need to purchase animals for the same task. Excess frozen tissues were also used by researchers to gain expertise and/or optimise in vitro techniques, which will improve the quality of the data received from experimental animals and reduce the need to repeat studies.
- The AEC undertook an evaluation of the mouse breeding colonies held at the establishment. The AEC made recommendations to the institute to close any breeding colonies that are no longer used by researchers. One breeding colony was subsequently closed as it was no longer required. The AEC is requesting regular updates on the status of use of the remaining breeding colonies.
- The AEC with the support of the institute encourages the use of animals for training/competency assessment within the cohort allocated to the project provided the outcomes are not affected in any way.
- There was a request to test new sensor devices that would be fitted to the animals for short periods of time. The use of animals for more than one project was a reduction in the number of animals to be used, and the animals experienced only minimal additional handling as a result.
- Testing the effectiveness of using virtual fencing for encouraging movement and herding of sheep. This project had allowed for a reduction in the number of sheep required by using a previously established learning method developed in a cattle virtual fencing project.
- Euthanasia of a horse for a dissection workshop. This project was considered as a reduction in animal numbers by taking the opportunity to conduct research when euthanasia had been recommended for this horse due to chronic lameness as result of an injury in the past.
- New drugs for blowfly control. The request to change the source of the animals and the location of the experiment was considered a reduction by using animals that were involved in another project.
- Control groups are shared whenever this is feasible for the research being undertaken.

- Surplus SPF males from the breeding farm are preferentially used for Eimeria oocyst production rather than being culled at the time of sex determination.
- Researchers are required to describe what will happen to any excess birds, which may occur when hatched chick numbers are higher than expected. These excess chicks will most often be transferred to another approved project and it is rare that birds are euthanised due wholly to being excess to forecast needs.
- Excess eggs in QC testing may be used for training purposes.
- Egg and bird numbers are continually monitored, with reports versus 'standards' being provided and reviewed at each quarterly AEC meeting. Significant deviations versus expectation may give rise to an Adverse Event report so that immediate action can be taken.
- Collection of pathology samples from animals trapped for fauna surveys to inform disease investigations in collaboration with other institutions – where the additional interventions do not add unduly to animal welfare impacts this reduces the need to trap and handle additional animals.
- All biological information that can be practically attained from each fish will be in order to maximise the value of killing these animals for scientific purposes. This information may be used to create convergent lines of evidence to bolster the results of the present study, used in a subsequent follow-on study, or contributed to colleagues also studying aspects of the biology and ecology of yellowtail kingfish. These include: tissue samples for stable isotope analysis that can help to determine feeding position of this species within the ecosystem, blood samples (10 ml) to measure various aspects of body condition, gonad samples that can indicate the animal's state of sexual maturity, otoliths (ear bones) that can be used to age the fish and fin clips that DNA can be extracted from to determine the genetic connectivity of different population of this species.
- The fishing method (hook-and-line) will catch only a single fish at any one time, and as a result there is no risk of accidentally exceeding the number of fish requested in this application, which could be the case if using trap or gillnet fishing methods that can catch multiple fish simultaneously.
- We will perfuse organs within mouse studies to enable us to come back to different tissue samples and we will make a tissue library/ biobank for other researchers to investigate proteins of interest if required.
- A statistician is appointed to the AEC and attends the meetings to ensure that the appropriate number of animals required for research or teaching are approved.
- Aiming to develop new eDNA assays to detect ornamental fish species not permitted for import to Australia, researchers considered alternatives to collecting tissues samples by euthanasia. As such, they reported that they would include collection of tissue samples from fish collected for museum accession to help reduce the number of fish required from the wild. Total

replacement of tissue samples from euthanasia was not possible as the assay development requires testing and optimization by comparing DNA yield and quality from multiple tissues before selecting the most useful tissue for positive controls.

- The Committee continues to maintain a Biological Non-Human Tissue Database which allows researchers to share excess tissue, thus replacing the use of live animals with the use of stored tissue. In addition, to make these tissues more widely available, the Committee has joined the Ethitex tissue sharing database which facilitates tissue sharing throughout Australia.
- Approval of new techniques for embryo freezing rather than continuous breeding to maintain lines.
- Re-use of animals, where appropriate, after extended recovery interval.
- Consolidating breeding protocols to ensure no overbreeding which in turn reduces the need for culling.
- Rederivation: Animal facilities optimise fostering process and thereby minimise the numbers of female mice used for fostering purposes.
- Training: Animal facilities use mice for training purposes that were identified with an undesired genotype (hence would have been euthanased regardless).
- Sharing: Where possible, mouse lines are shared between different research groups to avoid unnecessary breeding.
- A researcher co-ordinated one of their rumen fluid collections from a project with a researcher from another establishment to allow this researcher to utilise a portion of the rumen fluid collected for in vitro analysis. This replaced the need for this researcher to collect rumen fluid from other cattle.
- Investigated with the sponsor of a project the possibility of collecting additional data from the current project to reduce the requirement for further animal studies in this area.
- We use farm-acquired natural mortalities for junior year necropsy classes for students to focus on learning technical skills. We only use freshly-killed fish for necropsy classes focused on diagnostic techniques (which demand fresh specimens).
- The utilization of the Advanced Microscopy Facility (Axioscan digital imaging) has allowed us to improve data analysis. This is an enormous improvement from previous studies and we have significantly increased the capacity to identify and quantify labelling in different parts of the brain. This refinement means that we gather much more data from each rat, and will reduce the number of rats used for neuroanatomical studies.
- We have reduced the animal numbers required by designing an experimental strategy that involves longitudinal behavioural analyses using a battery of tests. This has dramatically reduced the number of animals used compared to a design that used separate cohorts for each time point and each behavioural test.
- By optimising experimental techniques, such as flow cytometry, we reduce the number of mice required. e.g. we have been using flow cytometry panels that test >25 parameters simultaneously (rather than studying the same parameters in 2 or 3 separate experiments).

- We use frozen tissues from small groups of mice as a pilot to validate the reagents we use for cell labelling first, thus we reduce the number of times the crucial experiment must be undertaken to acquire the minimum statistically defensible number of animals required for the change expected.
- The choice of sheep for this experiment has been with a particular view to reduce the number of animals that would be used for the study. We have used only 2 sheep where we can test 12 sets of control and test scaffolds (6 sets on the back of each sheep) compared to a small animal such as a rabbit, where 12 animals would be required to do an equivalent study.
- Power analysis has been used to detect minimal differences between treatments and the number of animals required to detect a required response.
- Collaborations have allowed multiple sample types to be collected from the same animal for different projects to avoid unnecessary recapture.
- Projects submitted to this AEC almost exclusively use a purpose built poultry trial shed, that by the use of small internal pens allows the use of commercial stocking densities on a miniaturised scale. This in turn reduces the number of birds used from the thousands to the hundreds.
- The university actively encourages researchers working together to develop projects that can be run in parallel, which uses different tissues of the same animals in order to reduce the overall number of animals.
- The faculty has established a tissue bank where unused samples can be stored for future projects by pharmacology students.
- Collected tissues from different disease models are being shared across research disciplines in laboratory based projects.
- The number of different cell types collected from one mouse were maximised, reducing the number of animals and exposures required.
- Xenogen live bioluminescent imaging was used to assess tumour burden in the lung, replacing the need for additional animals to determine tumour burden at each time point.
- Experiments were designed to double up on control groups where possible to reduce the number of animals required.
- Both male and female mice produced were used, to reduce the need to establish larger breeding colonies.
- Multiple procedures on physiological variables were performed within each animal to reduce the required sample size.
- Male neonates produced from breeding through one research group were utilised by another group to reduce the number of animals needed.
- A bank of tissue was collected to assess both reproductive and peripheral tissue from the same cohort, minimising the animal numbers required.
- Tumours were grown on both the left and right flank of each mouse in order to reduce the overall number of mice needed for the project and maximise the tissue samples to be analysed from each mouse.

- The same control mice were used across multiple projects to reduce the number of control animals required.
- Multiple samples from each mouse were taken to reduce the number of mice required, and combined control groups to eliminate the need for more groups for each intervention.
- Compounds were first tested In vitro and only progressed to in vivo if efficacy was shown, thus reducing the animal numbers being used.
- Animal numbers were reduced as 1 or 2 mice per group were enough to provide a sufficient number of cancer cells for ex vivo analysis.
- A cross over study design was used so that control animals were first paired with treated animals, then re-used when controls were paired with controls, thus reducing the need for a new group of control animals.
- A 'within-subject' design was used, which allows one animal to be used for multiple testing parameters, reducing the total number of animals required.
- A colon culture technique was used to collect 6-8 colon fragments from one mouse, rather than requiring 6-8 individual mice.
- Control groups were shared across experiments wherever possible.
- A MeCap mouse cell line was used instead of live mice when an experiment or procedure needed to be optimized.
- Extensive in vitro work was conducted prior to moving to in vivo to limit the number of animals used.
- Advice from an international colleague was implemented to reduce use in later generations, with the numbers for each generation refined on the basis of statistical evaluation.
- Protein channels isolated from cells were used where tissue from one animal produced enough material for 20 experiments.
- Tissues from control group animals were used by multiple researchers.
- Both testis in the animal were used, one to serve as the experimental testis and the other to serve as an internal control. This reduced the animal numbers by 50%.
- The processing of tissues was optimised so that only two sets per time point were required.
- Experiments were performed on human endometrial stromal cells to confirm results and reduce the number of animals required.
- In vitro studies were performed prior to animal use, to reduce animal numbers.
- Transgenic lines were shared with another lab and experimental days were co-ordinated, harvesting multiple tissues from each animal and effectively halving the number of animals used.
- A bank of tissue was collected to assess both endocrine and peripheral tissue from the same cohort.
- One mouse was used for as many tissue/cell collections as possible.
- Both genders and dams were used from each litter.

- Use of transgenic mice allowed targeting recording of orexin neurons, resulting in less animals being needed to record the specific cell type of interest.
- Pathologic activation was used to investigate early embryogenesis rather than using mouse sperm resulting in using less animals.
- Pups of the same sex and litter were randomised into different drug therapy groups to ensure maximal use of littermates.
- Multiple tissues were collected from individual animals, minimising the number of animals required to collect sufficient replicate data for all parameters to be tested.
- In teaching practicums, where two experiments are to be performed by students, half the class will perform one experiment and the other half will perform the other and share the resulting data.
- Experience from previous studies or information from similar studies is utilised to ensure appropriate numbers of animals are used in experiments.
- Where feasible, observational studies are conducted and/or recorded for future use and analysis.
- Where possible, animals are sourced from other approved projects where they would otherwise not be required.
- Researchers are asked to use longstanding and well-established procedures in their research to ensure the minimum number of animals are used.
- Development of an Online Clinical Scoresheet Generator. This allows clinical scoresheets to be generated which are comprehensive and tailored.
- A statistician is sent all protocols to review and is present at meetings. This helps ensure that animal numbers are kept to a minimum while remaining sufficient for scientific validity.
- Mock surgery performed on animal tissue for training purposes or refinement of techniques.
- A researcher working with threatened species of fish has collaborated with other research groups to ensure there is no overlap in species across research groups in the state.
- Several researchers reported stopping their projects once they had adequate results, using much less animals than anticipated.
- Collaborations have provided the supply of fish frames from commercial fisherman. This reduces the number of fish required above the minimum legal size whilst making use of fish that have already been caught and dispatched.
- Samples available that are collected from collection animals and wildlife under our care or that have died. Access to this important material reduces the need for additional interference with animals and has benefited many collaborative researchers through the years.
- Routine husbandry procedures to be performed on animals are coordinated with teaching activities.
- Sharing of unused tissue among researchers.

- Use of pilot projects with reduced animal numbers.
- Obtaining more data from the use of fewer animals by combining objectives.
- Performing statistical power analyses prior to starting experiments to ensure the minimum required animal numbers are used to achieve statistical significance.
- Incorporating animals from one project as breeding stock or experimental animals for a subsequent project, rather than euthanising.
- Minimise use of sentinels which are only used to sample mice in IVC caging.
- Use of a device invented by an establishment researcher to extend the life of neuronal tissue for electrophysiology and imaging which has resulted in less animals being used.
- Use of protocols considered to be current best practice to minimise wastage of tissues collected, thus reducing the overall numbers of animals required in the long term.
- Pilot studies are conducted with a small number of animals to test unknown hypotheses, avoiding the use of large numbers of animals when outcomes are unknown or not guaranteed.
- An improvement in modelling techniques has reduced the need for some types of surveys, and careful survey design optimises the number of trapping events/nights.
- Excess guinea pigs were diverted to breeding.

6.3 Refinement

- In 2019 one project has introduced, and successfully implemented, the use of swabbing amphibians. This is deemed the least harmful and most sensitive method for diagnosing the amphibian chytrid fungus, a disease responsible for amphibian declines worldwide.
- Timetabling of classes is coordinated so that activities are spread over the semester, to avoid over-use of the same animal.
- A project introduced additional methods for humane euthanasia of fish, including Ike jime (a method roughly equivalent to a quick decapitation performed on larger fishes) and spraying clove oil on target fish whilst diving, a method used to anaesthetise and, in combination with already approved Rotenone, euthanise smaller fishes.
- Behavioural enrichment tools are included in studies where deemed appropriate and functional. These may include food scattering, play with handlers, stumps for sheep to climb on, balls hung from the roof for sheep to play with, bones for dogs and cats to chew. Additional play items were introduced this year; these include rope toys and Kongs with treats inside. Dogs

are trained to walk up a set of portable stairs onto a table to assist in blood collection and external parasite assessment. All dogs are routinely leash walked around the facility.

- The new AEC application requires researchers to describe the suitability of the requested species, detail how the design and management of the housing meets the specific needs for the species, what the impacts of transportation will be, and the strategies in place to minimize the stress associated with the procedures. The Committee has also been working with researchers to develop transportation SOP's. The AEC has been working with researchers to develop and trial a number of enrichment objects (e.g. hanging chains and poplar branches for sheep and goats and milk bottles for cattle). All researchers are required to ensure that early humane intervention points are stated and considered throughout the trial. The Committee also requires intensive observation sheets for any projects involving a major physiological challenge.
- This refinement of procedures includes (1) the implementation of an early paralysis detection (EPD) system and (2) monitoring of bone disease via x-ray imaging using the Faxitron system.
- Dual housing exercise cages designed to reduce the single cage stress effects.
- Intraductal injection of cells to form tumours, to minimise the surgery effects compared with orthotopic methods.
- The use of analgesia and anaesthesia.
- Staff specifically trained and experienced in animal technology, therefore any clinical signs of distress are recognised at the earliest onset.
- We are taking the knowledge obtained from our studies of patient samples to prioritise specific mouse experiments and eliminate others that are not clinically relevant.
- All procedures are performed on level 1 so as to minimise stress from transport.
- We refined our tamoxifen delivery techniques to pregnant females so that there is minimal distress to the females and minimal loss of embryos due to technical difficulties.
- Following surgery, animals will be handled for as little time as possible to reduce any unnecessary stress that could lead to sudden death.
- Impact on animals is refined by examining immune responses over the shortest times required (typically <28 days).
- Intrasplenic models are much shorter in length compared to orthotopic models (weeks rather than months) due to the accelerated nature of the model – simulating tumour cells in circulation and the start of metastasis rather than waiting for a primary tumour to metastasise. Orthotopic models are still needed as they determine therapy efficacy in a different way. Using intrasplenic models significantly reduces the overall length of time animals are subject to treatment and therefore reduces the total amount of stress induced.

- We have introduced the use of heating pads under the induction chambers when we ear notch weaning age mice. This helps to maintain their body temperature in the chamber and aids recovery. We see faster recovery times for these mice over the use of a room temperature induction chamber.
- There is a small but consistent occurrence of vaginal malformations (vaginal septas and imperforate vaginas) in the females of the C57Bl6 strain, which is the predominant inbred strain in our facility. Our estimate across our colonies would be 2-7%, depending on the particular mouse line. We have incorporated into the training of new animal care staff, and regularly reinforce, that a check for these conditions should be a standard part of the welfare check of the animals and that no females that have obstructions of the vaginas be used in mating pairs. By this we hope to avoid deaths of the mothers in birth (dystocia) or distress to females on mating. Mice introduced into the facility from other sources are also routinely checked for these conditions if they are to be used for breeding.
- We will use specially adapted cages to allow for social housing, and therefore reduce the stress on the animal and improve its overall welfare.
- The establishment hosted an Animal Welfare Symposium with a range of speakers and attendees from philosophy, NHMRC, Researchers, Veterinarians, Animal Care Staff etc. Biomedical animal facility and wildlife research and conservation site tours were conducted.
- In 2019 the establishment re-started the Animal Ethics Newsletter which goes out to all animal users, animal care staff and facility managers. It contains information on alternatives to animal use, partial replacement options, information from regulators, new technology, veterinary updates etc.
- In 2019 Research Ethics drop-in sessions commenced each month. Research ethics staff, vets and statisticians are present to provide advice and assistance to research staff and ensure they are using best practice methodology, technology and training.
- Research Services in conjunction with the AEC have commenced revising key SOPs/Guidelines for researchers. These help ensure best practice methodology is implemented and the standards and expectations are clear. They are available on the establishment's website.
- Increase in Veterinary support, advice and oversight with a new full time senior veterinarian appointed in addition to the existing veterinary position.
- Continued use and promotion of less invasive sampling for DNA, such as plucking hair or feathers or buccal swabbing over blood sampling, ear biopsy etc.
- Expert veterinary advice is available 24/7 to all staff and students working with animals, including those in the field.
- Continued use of camera traps and ink pad tunnels has decreased the use of live trapping for survey work or determining species presence in an area.

- Continued use of specially designed excluders for wildlife traps has decreased bycatch of non-target species.
- Compulsory training and competency assessment for all animal users in animal ethics and practical techniques.
- Due to teaching requirements a set number of animals are kept on campus and used. These are rotated between classes to minimise individual use.
- Research using organisms such as zebra fish embryos instead of mammals.
- Direct access to experienced staff in Animal Ethics, Welfare and Services and a full-time veterinarian for the entire duration of an AEC project.
- Researchers are encouraged to consult the 'Animal Ethics Review Support Group' for assistance in writing an AEC application.
- Conditions of approval are applied to projects for the Animal Welfare Officer or Animal Facility Supervisor to oversee an initial procedure conducted or competent Animal Services Technicians to conduct the procedure on behalf of the project investigator both within animal facilities and in the field.
- Adoption of an online database for use by both Animal Research and Integrity and researchers to monitor animal usage and avoid potential excess breeding of animals.
- Ongoing training and upskilling of Animal Research and Integrity staff involved in the care and use of animals for scientific purposes.
- Students attend various workplaces to reduce the use of a particular mob of animals.
- Changes to routes of administration for injection and blood collection to meet best practice standards.
- A new application form was developed in 2019 to better obtain the information required by the Code.
- The use of non-capture methods for wildlife research including the use of field surveys, scat surveys, camera traps and drones.
- The Team Leader undertook a review of the rodent enrichment program delivered in facilities and ensured they were standardised across the University and best practice.
- Returning sheep to grassed paddocks as soon as it is safe to do so is preferred over internal pen confinement.
- The use of toys and rewards to encourage and stimulate positive interactions between the animals and their carers provides entertainment and beneficial behavioural activities for non-human primates.
- Alternative methods for survey such as remote cameras, song meters and bat detectors (rather than trapping). We have also introduced a drone standard operating procedure that will enable non-trap techniques such as use of infrared cameras to detect koalas using drones and animal census using drone imagery.
- Refinement of bat survey techniques through adoption of new technologies to refine bat acoustic recording capability and use of infrared cameras at cave entrances to conduct counts, reducing the need for extensive harp trapping.

- Purchase of smaller fyke nets to address minimising entrapment and refinement of fyke net use to separate small and larger fish species reducing predation.
- SOPs have been updated to address extreme temperatures where climate change is increasing the consecutive number of days above 40 Celsius.
- In a protocol investigating forensic tools for illegally trafficked reptiles, the team reduced sampling time per animal to 15 minutes in duplicate as opposed to triplicate. This was determined as many wild animals began escape behaviour during the triplicate run.
- An SOP was developed for non-surgical intratracheal injection into mice, which is a very well-researched procedure in animal research. This was vigorously reviewed by the Chair and the committee.
- A protocol investigating the environment effects of riverine food webs and the growth of Golden Perch used large mesocosms which gave larval fish room to move freely and eat what they would in a natural setting.
- A protocol investigating artificial and natural hollows used by animals ensures that animals are processed inside cloth bags to minimise any stress.
- Camera traps and miniature thermal data loggers were used to provide a non-invasive means of assessing hollow suitability and use by animals.
- A protocol identifying optimal greyhound race track design for canine safety and welfare worked actively with the industry to design an IMU package that is lighter and smaller to reduce any probable discomfort and distress to animals during this project and beyond the research.
- Non-terminal sampling methods are used during routine blood collection for health monitoring.
- Effective use of analgesics and anaesthetics for painful and invasive animal procedures following recommendations by the AEC and veterinary staff.
- Smaller incisions during rodent surgery for faster wound healing.
- Minimum dose of injectable anaesthetics administered for shorter procedures e.g. for imaging purposes enabling faster recovery time.
- Allowing animals to acclimatise to the specific experimental setting and handle them with care to minimise fear and distress.
- Option to use anaesthetised mice when practising subcutaneous injections and tail vein injections during practical orientation sessions with new researchers (minimise harm to animal).
- Continue to make use of Opti-Spot test strips for rodent animal health monitoring, no need to anaesthetise animal, sentinels no longer required as we sample stock animals instead.
- Minimum amount of blood taken from animals during health testing.

- Use of EMLA cream as topical anaesthetic ointment or instillation of local anaesthetic substances in combination with injectable anaesthetics for any painful procedures prior to incision or injection.
- Surgical training – individual training session with researchers who will be performing mice surgical procedures for the first time.
- Ensuring only the right sized surgical instruments are used that are suitable for the species.
- Promote post-procedural monitoring of research animals.
- Increased sharing of information between research groups and institutions.
- Score sheets for all monitoring during approved surgical procedures have been introduced to refine the process and appropriately identify and manage any pain and distress in animals.
- A rich supply of free environmental enrichment has been sourced for our animals allowing us to accommodate budget for further training resources for refinement of techniques in line with best practice in the industry. We have also been able to supply two external facilities with free enrichment.
- Animals were housed on-site in their familiar environment. They were handled by personnel with cattle handling experience, and treated by staff skilled and efficient in the procedures. Prior to treatment animals were kept together in the holding yards, then moved and treated together in groups. Upon release they were free to roam and graze in their regular paddocks. These methods reduced any pain and distress the animals may have experienced.
- The formulations applied in this study all contained active ingredients for local anaesthesia, therefore reducing pain experienced by the animals after mulesing and tail docking. The animals were closely monitored from the time of mulesing and during the first 24 hours after mulesing to observe for signs of pain and distress. An amendment was passed to apply fly-strike preventative to vulnerable control treated lambs on Day 2. This amendment very likely improved animal welfare outcomes as no cases of fly-strike were detected in control treated lambs after this amendment.
- The formulations applied in a study all contained active ingredients for local anaesthesia, therefore reducing pain experienced by the animals after marking (tail amputation and castration of males). An independent veterinarian was present on Day 0 to ensure animal welfare and to provide rescue analgesia post-marking if required.
- Animals were housed on-site in their familiar environment. They were handled by personnel with sheep handling experience and treated by skilled staff and trained farm hands. Once mulesed and treated the lambs were free to graze in the paddock without disturbance. This method was used to reduce distress of animals being separated for long periods of time.
- Lambs underwent observations by personnel prior to study commencement, to acclimate them to human observation. The frequent movement of Cohorts of lambs into and out of the various small pens and yards during the study meant that the lambs were generally very easy to handle and could be weighed and examined with minimal stress. The study was paused

during extreme fly pressure weather (warm, still, humid conditions), so as to reduce the impact on study animals. Animals were pre-treated with Strikeforce-S prior to mulesing, after recognising flystrike problem in earlier cohorts. Additional unhandled lambs were added to the observation yards to act as "friends" to the Cohort being observed once a depressive "group effect" was noted.

- Purchasing of digitally ventilated cages with inbuilt sensors, allowing for 24/7 monitoring of animals and their environment within the cage which greatly improves monitoring of animals undergoing high impact studies over an extended period of time without interference from handling either the cage or the animals themselves.
- Training of researchers in current best practise techniques.
- Improved peri- and post- operative analgesia to reduce pain from surgery.
- Use of ex vivo assays to minimise adverse impact on animals.
- Use of modern trapping techniques and equipment to minimise potential for animal injury. Use of smaller, less invasive tags for identification.
- Increased awareness and use of environmental enrichment.
- Horses are monitored for behavioural changes and replaced regularly. Horse usage is rotated to prevent overuse.
- Using treats and water as training for medication.
- Reduction of lamp size to less intense light; use of red light covers for spotlighting activities.
- For native animals, handling is conducted by the licensed person only, with students observing the techniques.
- Animals are given appropriate rest periods.
- Horse usage recording system to rotate horses and minimise over-use of horses.
- Professional development for teachers to improve skills and knowledge.
- Use of industry sites where animals are housed to minimise stress.
- Uncomfortable procedures e.g. temperature taking only done once.
- Students are referred to Standard Operating Procedures prior to animal use.
- Rotation of locations to minimise repeated exposure to the same native animal colonies.
- Use of non-painful and non-invasive procedures for student activities.
- Animals are monitored closely for signs of stress and distress, and are removed from class if under undue stress.
- Cattle and sheep used on-farm are divided into groups so that they are not re-used for health applications and not more than twice for drafting.
- Animal use monitoring forms identify the number of times an animal has been used.
- Simulations are used to practise and refine techniques before contact with live animals.

- Accommodation of research horses in a large paddock on a professional horse spelling/pre-training farm. Spontaneous collection of naturally voided urine for the purpose of drug analyses.
- New procedures were refined in cadavers before live animals were used. This reduced the potential impact on animal welfare as the procedures involved drilling into the skull of rats.
- New invasive procedures were assessed in the presence of a Veterinarian who was able to provide immediate advice for unexpected impacts, refining the procedures and improving on overall animal wellbeing.
- A request sought permission to apply Leader Products Tail Paint spray to the calves. The purpose was to identify the animals and the order in which serial blood samples were to be collected. The request was considered a refinement as using the numbers to easily identify animals for bleeding in serial bleeds was better for the cattle and easier for the operators.
- The investment in a new Jamesway incubator and hatcher, with validation of amended temperature and humidity profiles in 2018, has resulted in improved hatch rates and healthier day-old chicks through 2019. The use of a dedicated incubator room reduced the need to transport incubated eggs from one incubation area to another, has helped to maintain egg temperature and avoid stressing the embryos at transfer.
- Modifications were made to the Oocysts Production Unit to improve each of bird placement, security from bush fires, general biosecurity from diseases, and potential for cross contamination of antigens. The Company also installed dedicated building and plant facilities to contain security of the new incubator and hatcher.
- Modifications to floor grids in the Grow-Out sheds and to fencing in the Bird Houses was done to improve bird welfare.
- With a focus on environment enrichment, there was the introduction of wood shavings to the floor in the Grow-Out sheds and sustained minimal light intensity through the use of coloured bulbs/tubes.
- Placement of LED lights along the drinking line did help 'start-off' chicks to better identify the drinkers, and then the feeders, during their first week in the GO shed.
- The introduction of music in the GO sheds did help to acclimatise birds to noise from outside the sheds, aiming to ensure the birds were not easily startled.
- Ongoing training and on-going experience in handling poultry which are particularly prone to stress, is standard practice for the Company.
- There was continued 'alarm monitoring', including particular emphasis on temperatures within all Bird Houses, egg incubators, and Grow-Out sheds, power supply to industrial equipment, and building security via external video surveillance of the buildings.
- Rehoming of fish to private tanks when no longer suitable for experimental purposes.
- Remote operated infrared cameras, acoustic recording devices and drones instead of physical trapping.

- Disturbance to the colony: birds will be taken to a 'processing station' adjacent to the colony where burrow density is lower (soil is more stable) and disturbance to the rest of the colony is minimal. Impacts to individual birds: birds will be placed in a breathable cloth bag to prevent injury to the wings when being weighed and measured. The dark colour of the bag offers protection to the bird's eyes which are sensitive to bright light. When working at night, field technicians are required to use red light filters for their head-lamps which are less disruptive to the bird's vision. Handling time will be strictly monitored and kept to a minimum by collecting only a small number of (important) measurements and by sampling feathers rather than blood because it is quicker and less invasive.
- All observations are behavioural, and we only approach the wild horses until the first horse runs. We do not approach bands with very young foals (<1 week old) or heavily pregnant mares.
- Seine nets will be operated by hand-hauling so that the fish are sampled and returned to their environment within 5 minutes of trapping. Seine nets will not be pulled out of water onto shore, a procedure which allows more efficient counting of individuals but can result in higher rates of mortality. Handling of fish will be the minimal required for identification, and in a manner to minimise scale and mucous damage.
- To reduce the effects of the sampling strategy, hand-netting/barrier-netting will be used in favour of other fishing methods, such as pole and line, to minimise any potential bycatch and minimise any suffering in the species being collected.
- The AEC requests that investigators required to handle sheep as part of their research are appropriately skilled in low stress handling techniques to ensure that the impact of any procedures being carried out on them is minimal.
- The Committee continues to encourage researchers to undertake a pilot study if the impact of the proposed study interventions on animal health and well-being is unknown.
- Animal House veterinary managers review protocols with researchers in order to optimise anaesthesia protocols (including monitoring) and analgesia.
- Scoring systems for monitoring of experimental animals have been developed and refined, with the aim of minimising potential pain and distress that animals may experience as part of certain research related procedures.
- The establishment has installed additional silos and feed heads on farms participating in a project to minimise human intervention in administering the treatment product.
- GoPro footage is being utilised in a project to monitor feed residuals and reduce human intervention.
- Trained personnel only administer treatments and collect samples on commercial farms to reduce adverse impacts on animals.
- We have moved away from cognitive tests that cause stress to the animal (e.g. Morris water maze) to the Intellicage system, in which mice are housed in social environments without routine interference from the researcher. This provides both more reliable data and less stress to the animals.

- Seahorse health was also assessed by a licensed vet after experiment completion. These refined procedures allowed us to, with DPI permission, release seahorses back into the collection site instead of being euthanised after experiment completion, as has unfortunately been regular procedure for collected fish in past experiments.
- In terms of GPS collaring, we have purchased some of the latest, high-tech, low-weight GPS collars to minimise the impact of the collars on individual animals. The collars we are using are less than 1/3rd of the weight of most comparable collars on the market (~100g compared with >300g for units from the leading competitor).
- Embryos are now flushed from the uterine horns using a foley catheter, which is much easier to insert than the tubing used previously. Additionally, prior to the embryo recovery practical class taking place, the students practice inserting the foley catheter into ewe reproductive tracts obtained from a local abattoir. These refinements have reduced the amount of time the procedure takes and hence the duration of anaesthesia.
- Learner participants are extensively prepared in the diagnostic, procedural and decision making aspect of trauma surgery prior to the wet lab of the workshop. This not only improves the learning outcomes but also reduces time of anaesthesia and surgical trauma associated with correcting of the simulated lesions.
- From previous protocols, we have optimised the use of absorbable monofilament sutures for muscle layers and Michel clips for the skin layers. So far this has eliminated any suture failure and infection, and drastically reduced the need to re-suture.
- Use of conditioning training to increase trust of animals, increased monitoring and optimised pain management regimes.
- The use of Unmanned Aerial Vehicles (UAVs, Drones) to survey animals and collect biological samples.
- Use of Remotely Operated Vehicles (ROV) and UAV to observe marine animals resulting in less disturbance.
- Limiting the length of time wildlife is held, allowing them to be released after completion of the research.
- Collaboration with other institutions when/where wildlife is studied to reduce impacts on populations and reduce environmental disturbance.
- We have recently appointed an Animal Welfare Officer who has commenced a review of all standard operating protocols in use. Example - Precision livestock management research and general farm monitoring systems (behavioural, activity, weights) both allowing for less invasive and stressful monitoring of livestock herds in research and production settings.
- Work demonstrated that aerial surveys for marine wildlife were less invasive using smaller, quieter drones than using large manned aircraft. Ongoing work therefore used drones.
- Mobile phones are used to record images of fish in photo identification tanks that are then used to identify specimens to reduce time in captivity.
- Methods were employed to minimize interactions between drones and birds, as well as ensure collisions are avoided.
- Selection of sites where students can work on the river bank to minimise time fish are held in captivity.

- Invertebrates were used in student observation studies.
- Surgical techniques were refined prior to the commencement of the project using cadavers, resulting in live animals subsequently recovering very quickly.
- Replaced intra-tracheal inoculation and intravenous anaesthesia with intranasal inoculation with inhaled anaesthesia, which is less invasive, reduces the risk of irritation of the animals, has a reduced procedure time, reduces the risk of anaesthesia related adverse events and removes the pain associated with intravenous anaesthesia.
- Changed the route of administration of pain relief from subcutaneous injection to oral.
- Collected saliva as a non-invasive alternative to blood collection.
- Implemented advances in surgical techniques to provide shorter periods under anaesthesia.
- Reduced the dosages of drugs, infectious inoculations or inflammatory compounds to prevent or reduce potential weight loss.
- Refined surgical techniques to minimise time in surgery and minimise potential adverse events.
- Replaced intravitreal injections with eye drops.
- Used a non-surgical method of embryo transfer to replace the conventional surgical alternative, eliminating the need for anaesthesia and removed the associated surgery related recovery.
- Developed sophisticated in-vitro assays to rapidly detect beneficial biomechanical changes, which reduced the need to maintain the animals for extended periods in-vivo.
- Refined the surgical/recovery location, analgesics being administered, and post-operative recovery procedures, which reduced the impact on animals.
- The AEC issued a Position Statement encouraging researchers to avoid the use of intraperitoneal injections in rodents where possible. This was because of concerns documented in the literature and through our own AEC-approved study that shows this can be an unreliable means of administering substances which also carries a risk of complications.
- An animal temperament assessment matrix developed to assist with the rehoming of rodents.
- A Standard Operating Procedure was developed for the husbandry of decapod crustaceans, which are believed to be sentient species but are not currently covered by NSW animal research legislation.
- Researchers refined a technique after realising they didn't need to use surgery.
- Previously, rats were found to be suffering from seizure-like symptoms in the 0 to 24 hours post microbeam radiation treatment, even with the previous seizure control protocol combining meloxicam, levetiracetam and diazepam. After the implementation of methylprednisolone in the new protocol, it was found that rats experienced a significant reduction in radiation symptoms, likely due to the decrease in inflammation in the brain.

- Ketamine is now applied via subcutaneous rather than intraperitoneal (IP) route. This is reflects the AEC's position statement on IP injection.
- Dorsal von Frey hyperalgesia testing: As a part of a 3Rs grant application and in line with requirements of the AEC for continued approval of the project, the researchers originally planned to assess pain (hyperalgesia) using conventional 'plantar' von Frey testing. In this technique, a single animal is isolated in a Perspex box over a wire mesh floor, allowing access to the bottom of the paws and the assessment of paw withdrawal in response to poking with calibrated fibres (von Frey fibres, a validated measure of increased sensitivity due to pain). This approach can be stressful to animals (isolation, pain of siting on wire mesh etc.) and is of uncertain utility in inflammatory models of pain where spontaneous position of the paw in the correct orientation for testing is unlikely or impossible. With AEC approval and Animal Welfare Officer (AWO) support, the researchers piloted the use of a recent variation of this technique using non-arthritic rats. In this method rats are restrained using a modified towel wrap procedure, securing their front paws, head and torso while their hind paws are positioned on a benchtop in a natural pose. This positioning allows for the application of the von Frey fibre to the top of the paw (dorsal surface) in a manner unlikely to be painful or difficult for an arthritic animal with severe swelling of the hind paws. This work will represent a significant refinement, decreasing the time of restraint, stressful isolation and pain experienced by the animals in this model. We believe this refinement improves the reproducibility of data obtained from this technique and may have applicability in the assessment of pain in other models of chronic inflammation beyond collagen-induced arthritis.
- As part of an amendment approval, the AEC stipulated the following:
  1. That the scoresheet used to monitor the animals be revised to show that any of the following scores would automatically trigger daily scoring:
  An acute weight loss of 7% or above;
  A score of 2 or more for posture;
  A score of 2 or more for activity;
  A score of 2 or more for skin changes suggestive of itchiness (including scaly skin on the face, feet or tail).
  2. That the researchers continue to work with the AWO to develop a suitable means of assessing chronic weight loss.
- Introduction of training in the Scientific Use Code for volunteers on wildlife research studies.
- Introduction of aseptic surgery training for projects using rodents.
- Approval for expansion of the animal yards at the agistment facility.
- The Animal Welfare and Ethics Coordinator (AWEC) served as a fulltime Animal Welfare Officer resulting in:
  Increased informal monitoring of projects
  Direct supervision of hands on rodent training courses

Revision of Animal Ethics Online training course for launch in 2020.

- Use of remote video camera monitoring in all large animal recovery rooms to supplement physical monitoring and increase monitoring out-of-hours and on weekends.
- Extension of Animal House to include an outdoor exercise area for pigs.
- Online animal ethics theory modules and practical training for work with mice, rats and rabbits.
- Continued emphasis on environmental enrichment.
- Fish in one project were rehomed instead of euthanised as originally proposed. Fish recovery after parasite infection was very good and fish health was monitored carefully. At the end of the experiments, fish were quarantined for a week to two weeks before being rehomed.
- Computed tomography (CT) imaging of the wombat gastrointestinal system: the Committee placed a condition on the project to combine the requested imaging procedure with a routine heath check and include the head of the animal when possible to allow for a comprehensive dental assessment.
- Improvements to animal housing and management (e.g. introduction of "buddy cages" to avoid single housing of mice, provision of environmental enrichment).
- Training of researchers (animal monitoring and administration of injections).
- Working with researchers to develop better project specific monitoring checklists to identify, action and report adverse events and the development of an adverse event form.
- The use of less invasive procedures in wildlife studies e.g. sand pads rather than trapping.
- Use of an Observational Only - Field Research Form (No Trapping, Handling or Spotlighting).
- Developing competency assessment procedures.
- Providing alternative procedures to minimise impact.
- Providing mandatory training to use a mobile anaesthetic induction unit (i.e. Stinger anaesthetic machine) and ensuring researchers do not use obsolete anaesthetic techniques.
- Requiring researchers to adopt aseptic technique when conducting surgical procedures in line with current veterinary best practice.
- Requiring new researchers to undertake a mandatory animal ethics course prior to starting work and requesting existing researchers to undertake a follow-up refresher course every 5 years thereafter.
- Decontamination of equipment to be used in direct contact with animals to prevent potential spread of pathogens.

- Thirty-one dogs and three cats were donated to Beagle Freedom, supervised by the AEC during the reporting year, as they were no longer required or suitable for research. There are ongoing efforts to reduce the colony by rehoming surplus animals (dogs and cats), or those that can no longer be used in research due to illness or old age.
- Enrichment and training programmes are in place to condition animals to study procedures.
- The use of cameras and audio equipment (acoustic and ultrasonic) reduces the need to trap animals.
- 43 guinea pigs re-homed through the Research Animal Re-homing Service. By giving them to the re-homing organisation, they were able to be placed as pets in suitable homes rather than being euthanased.
- In 2019, the improved monitoring schedule was embedded for guinea pigs undergoing the C. chauvoei challenge. This has allowed for early intervention/euthanasia of 61 moribund guinea pigs which is a significant improvement on previous years.
- Rabbits on test using new adjuvants had their injection sites shaved to allow for easier monitoring and to reduce the frequency of palpating the injection site.

7. Appendix – Guide to the categories of reporting

The following is the guidance provided in Form L – Animal use statistics on categories for Purpose, Procedure, Species and Fate of Animal.

Column C: PURPOSE

Please note – Purpose Codes now have an A (for Activity) in front of the existing purpose number code in order to help improve accuracy of data entry.

Enter the **most appropriate** numerical code **(A1-A10)** from those listed below to describe the **primary** purpose of the project (one purpose only for each project should be entered).

Purpose Code:	Description:
A1	***Stock breeding*** Breeding projects to produce new teaching or research stock. Include the animals used to produce progeny and any breeders or progeny culled in the process, NOT the final progeny themselves (as these will be counted under the project in which they go on to be used).
A2	***Stock maintenance*** Holding projects for animals maintained for use in other projects. These animals may be maintained under an Animal Research Authority because they require special management. If they are not held under an Authority, (e.g. normal stock animals kept mainly for commercial production, but occasionally used in research) then they are only counted in the project where they are used for teaching/research. *Examples* • *Fistulated ruminants which are maintained under a holding project, for use in other short term feeding trial projects* • *Non-breeding colony of diabetic rats held for research in other projects*
A3	***Education*** Projects carried out for the achievement of educational objectives. The purpose of the project is not to acquire new knowledge, rather to pass on established knowledge to others. This would include interactive or demonstration classes in methods of animal husbandry, management, examination and treatment. *Examples* • *Animals used by veterinary schools to teach examination procedures such as pregnancy diagnosis* • *Sheep used in shearing demonstration classes for students; Dogs used to teach animal care to TAFE students*

A4	***Research: human or animal biology*** Research projects which aim to increase the basic understanding of the structure, function and behaviour of animals, including humans, and processes involved in physiology, biochemistry and pathology.
A5	***Research: human or animal health and welfare*** Research projects which aim to produce improvements in the health and welfare of animals, including humans.
A6	***Research: animal management or production*** Research projects which aim to produce improvements in domestic or captive animal management or production.
A7	***Research: environmental study*** Research projects which aim to increase the understanding of animals' environment or their role in it. These will include studies to determine population levels and diversity and may involve techniques such as observation, radio tracking or capture and release. *Examples* • *Pre-logging or pre-development fauna surveys*
A8	***Production of biological products*** Using animals to produce products other than milk, meat, eggs, leather, fur, etc. *Examples* • *Use of a sheep flock to donate blood to produce microbiological media* • *Production of commercial anti-serum* • *Production of products, such as hormones or drugs, in milk or eggs from genetically modified animals* • *Quality Assurance testing of drugs* **but do not include animals which come under Purpose A10, below.**
A9	***Diagnostic procedures*** Using animals directly as part of a diagnostic process. *Examples* • *Inoculation of day old chicks with ND Virus to determine virulence* • *Water supply testing using fish*
A10	***Regulatory product testing*** Projects for the testing of products required by regulatory authorities, such as the APVMA. **If the product testing is not a regulatory requirement, eg it is part of a quality assurance system only, those animals should be included in the appropriate category selected from above.** (This would be normally be Purpose A8 (Production of biological products) in the case of QA testing.) *Examples* • *Pre-registration efficacy or toxicity testing of drugs and vaccines*

Column D: PROCEDURE

Please note – Procedure codes now have a P (for Procedure) in front of the existing procedure number code in order to help improve accuracy of data entry.

Enter the **highest appropriate** alphanumerical code **(P1-P9)** from those listed below to describe the type of procedures carried out on the animals in the project. The descriptions given are a guide only. **Note:** for each project include additional lines for each procedure category where different animals within the same project are subjected to different procedure categories.

Where 'Death as an endpoint' or 'Production of genetically modified animals ' applies, animals must be placed in these categories (P8 or P9) rather than any others which might also appear appropriate.

Procedure Code:	Description:
P1	***Observation Involving Minor Interference*** Animals are not interacted with or, where there is interaction, it would not be expected to compromise the animal's welfare any more than normal handling, feeding, etc. There is no pain or suffering involved. *Examples* • *Observational study only* • *Breeding animals for supply, where only normal husbandry procedures are used* • *Breeding or reproductive study with no detriment to the animal* • *Feeding trial, such as Digestible Energy determination of feed in a balanced diet* • *Behavioural study with minor environmental manipulation* • *Teaching of normal, non-invasive husbandry such as handling and grooming*
P2	***Animal Unconscious Without Recovery*** Animal is rendered unconscious under controlled circumstances with little or no pain or distress. Capture methods are not required. Any pain is minor and brief and does not require analgesia. Procedures are carried out on the unconscious animal which is then killed without regaining consciousness. *Examples* • *Laboratory animals killed painlessly for dissection, biochemical analysis, etc* • *Teaching surgical techniques on live, anaesthetised patients which are not allowed to recover following the procedure*
P3	***Minor Conscious Intervention*** Animal is subjected to minor procedures which would normally not require anaesthesia or analgesia. Any pain is minor and analgesia is usually unnecessary, although some distress may occur as a result of trapping or handling. *Examples* • *Injections, blood sampling in conscious animal* • *Minor dietary or environmental deprivation or manipulation, such as feeding nutrient-deficient diets for short periods*

	• *Trapping and release as used in species impact studies* • *Trapping and humane euthanasia for collection of specimens* • *Stomach tubing, shearing*
P4	***Minor Surgery With Recovery*** Animal is given appropriate regional or general anaesthesia with as little pain or distress as possible. A minor procedure such as cannulation or skin biopsy is carried out and the animal allowed to recover. Depending on the procedure, pain may be minor or moderate and postoperative analgesia may be appropriate. Field capture using chemical restraint methods is also included here. *Examples* • *Biopsies* • *Cannulations* • *Sedation/anaesthesia for relocation, examination or injections/blood sampling* • *Castration with regional or general anaesthesia and post-operative analgesia*
P5	***Major Surgery With Recovery*** Animal is rendered unconscious with as little pain or distress as possible. A major procedure such as abdominal or orthopaedic surgery is carried out and the animal allowed to recover. Post operative pain is usually considerable and at a level requiring analgesia. *Examples* • *Orthopaedic surgery* • *Abdominal or thoracic surgery* • *Transplant surgery*
P6	***Minor Physiological Challenge*** Animal remains conscious for some or all of the procedure. There is interference with the animal's physiological or psychological processes. The challenge may cause only a small degree of pain/distress or any pain/distress is quickly and effectively alleviated. *Examples* • *Minor infection* • *Minor or moderate phenotypic modification* • *Early oncogenesis* • *Arthritis studies with pain alleviation* • *Induction of metabolic disease* • *Prolonged deficient diets* • *Polyclonal antibody production* • *Antiserum production*
P7	***Major Physiological Challenge*** Animal remains conscious for some or all of the procedure. There is interference with the animal's physiological or psychological processes. The challenge causes a moderate or large degree of pain/distress which is not quickly or effectively alleviated. *Examples* • *Major infection* • *Major phenotypic modification* • *Oncogenesis without pain alleviation*

	• *Arthritis studies with no pain alleviation* • *Uncontrolled metabolic disease* • *Isolation or environmental deprivation for extended periods* • *Monoclonal antibody raising in mice*
P8	***Death As An Endpoint*** This category only applies in those rare cases where the death of the animal is a planned part of the procedures and animals die but are not euthanased. Where predictive signs of death have been determined *and* euthanasia is carried out before significant suffering occurs, they may be placed in category P6 or P7. *Examples* • *Lethality testing (including LD50, LC50)* **It does not include:** death by natural causes; animals which are euthanased as part of the project; animals which are euthanased if something goes wrong; animals euthanased for dissection or for use as museum specimens; or accidental deaths.
P9	***Production of genetically modified animals*** This category is intended to allow for the variety of procedures which occur during the **production** of genetically modified animals. As animals in this category may be subjected to both minor *and* major physiological challenges *and* surgical procedures, this category reflects the varied nature of the procedures carried out. It effectively includes ALL animals used in GM production other than the final progeny which are used in a different category of procedure. *Examples* • *Initial breeding animals for GM production* • *Animals culled as part of the GM production process*

Column E: SPECIES

Please note – the species codes now have an S (for Species) in front of the existing numerical species code in order to help improve accuracy of data entry.

- Enter the alphanumerical code from those listed below to describe the species or species group used in the project.
- The alphanumerical code is not sequential - for each species used select the appropriate numerical code as listed in the table below.
- There are no species codes S15, S19, S22, S25, S26, S44 or S55, and the highest number is S56.
- In filling out the spreadsheet include additional lines for each species where more than one species is used in a project.

Laboratory mammals	S1	Mice
	S2	Rats
	S3	Guinea Pigs
	S4	Rabbits
	S5	Hamsters
	S6	Ferrets
	S7	Other laboratory mammals (not primates)
Domestic mammals	S8	Sheep
	S9	Cattle
	S10	Pigs
	S11	Horses
	S12	Goats
	S14	Deer
	S31	Cats
	S32	Dogs
	S33	Other domestic mammals
Birds	S13	Poultry
	S16	Exotic Captive
	S17	Exotic Wild
	S18	Native Captive
	S20	Native Wild
	S21	Other birds
Aquatic animals	S23	Fish
	S23A	Cephalopods (reporting not mandatory)
	S23B	Crustaceans (reporting not mandatory)
Amphibians	S24	Amphibians
Reptiles	S27	Lizards
	S28	Snakes
	S29	Turtles and Tortoises
	S30	Other reptiles

Primates	S34	Marmosets
	S35	Macaques
	S36	Baboons
	S37	Other primates
Native mammals	S38	Macropods
	S39	Possums and gliders
	S40	Native rats and mice
	S41	Dasyurids
	S42	Wombats
	S43	Koalas
	S44A	Monotremes
	S44B	Bandicoots
	S44C	Bats
	S44D	Other native mammals
	S44E	Seals
	S44F	Whales and dolphins
Exotic feral mammals	S45	Camels
	S46	Cats
	S47	Cattle
	S48	Goats
	S49	Hares
	S50	Horses
	S51	Mice
	S52	Pigs
	S53	Rabbits
	S54	Rats
	S55A	Dingo/Wild Dogs
	S55B	Foxes
	S55C	Other exotic feral mammals
Exotic zoo animals	S56	Exotic zoo animals

Column F: FATE OF ANIMAL

This column **MUST** be completed where species S31 Domestic cats or S32 Domestic dogs have been used.

This column may also be completed where other species are used and it is recommended that this information be provided.

For each project, include additional lines where there are different fates of animals within the same project.

Fate Code	Description
F1	**Retained in project** This is where the project is ongoing and the animal will remain in the project in the next reporting year.
F2	**Retained for use in other projects or supplied to another establishment / individual for research** This is where the animal is kept by the establishment / individual for use in other research projects or supplied to another establishment / individual for use in research.
F3	**Retired from research and kept by the establishment / individual** This is where the animal is kept by the establishment / individual in retirement with no further plans for use in research.
F4	**Privately (non-research) owned and remained with owner** This is where the animal is privately owned and remains with the owner. *Examples:* • *Animal presented to veterinary clinic for treatment and participates in clinical trial* • *Behavioural study with privately owned companion animals*
F5	**Rehomed (as companion animal to private (non-research) home or rehoming organisation)** This is where the animal is rehomed as a companion animal to a private (non-research) home or to a rehoming organisation with the consent of the rehoming organisation.
F6	**Euthanased or died related to the project** This is where the animal is required to be euthanased as an integral part of the research project, or is euthanased or dies during the project as a consequence of the project procedures.

F7	**Euthanased or died unrelated to the project** This is where the animal is euthanased or dies during the project for reasons unrelated to the project. *Example:* • *Animal in long-term food palatability trial euthanased due to unmanageable osteoarthritis*
F8	**Euthanased because unsuitable to be rehomed** This is where the animal is no longer required for research and is euthanased on the basis of an assessment that the animal is unsuitable for rehoming. Reasons the animal is unsuitable for rehoming may include physical, behavioural and biosecurity factors. *Examples:* • *Animals with unmanageable health conditions causing discomfort or distress* • *Animals that have problem behaviours that are unable to be addressed through rehabilitation* • *Animals that could pose a biosecurity risk to other animals, people or the environment* • *Animals that are genetically modified*
F9	**Euthanased because unable to find a suitable home** This is where the animal is no longer required for research and is assessed as suitable for rehoming, but is euthanased because a suitable home is unable to be found.
F10	**Remain free living in the wild or released to the wild** This is where the animal is free living and remains in the wild (including where the animal is captured and released) and where the animal is released to the wild. *Examples:* • *Wildlife fauna surveys* • *Native animal captive breeding and monitored release programs*

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