



chaVoc	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
egrGar	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
pygAde	F	K	G	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
aptFor	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
nipNip	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
anaPla	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
melGal	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
galGal	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	V	E
strCam	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aptRow	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aptHaa	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aptOwe	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
rhePen	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
rheAme	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
droNov	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
casCas	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
eudEle	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
notPer	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
tinGut	F	K	G	W	S	G	G	L	-	-	-	-	-	-	-	-	-	-
cryCin	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aquChr	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
halLeu	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
lepDis	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
colliv	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	T	E
cucCan	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
picPub	F	K	G	-	P	G	H	L	E	L	-	#	A	A	E	G	A	E
calAnn	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
chaPel	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
faiPer	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
melUnd	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
corBra	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
pseHum	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
taeGut	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	V	E
geoFor	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	V	E
ficAlb	F	K	-	-	-	-	-	-	-	-	-	-	G	Q	-	-	-	-
serCan	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	V	E

MALMA\_AF344483  
MALMA\_AF344519  
MALMA\_AF344467  
MALMA\_AF344470  
MALMA\_AF344507  
BBOBB\_AJ235773  
HANCO\_AF323190  
HANCO\_AF323188  
CRNCO\_AY530929  
CRNCO\_L14395  
CRNCO\_AY530927  
CRNCO\_AF190433  
CRNCO\_L11217  
BERRA\_AF203487  
COMMY\_AF281478  
PCIER\_AJ428893  
MGVER\_Z80200  
SLAER\_AF421109  
TSTER\_Z83149  
SYMER\_Z80192  
SLAER\_AF421108  
PPLER\_AF419239  
SPTER\_Z83135  
LE CER\_AF077656  
LE CER\_Z80183  
LE CER\_Z80202  
LE CER\_Z80180  
LE CER\_AF077652  
LE CER\_AF077653  
LE CER\_Z80177  
LE CER\_Z80181  
LE CER\_AF022134  
PMOER\_Z83144  
THPER\_AF213818  
THPER\_AF213819  
THPER\_AF213817  
THPER\_AF213815  
PRMER\_AF395005  
PRMER\_AF394965  
PRMER\_AF395006  
PRMER\_AF213794

A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	N	W	S	P	E	L	A	A	A	C	A	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	#	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	#	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	#
A	S	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	I	W	K	E	I	K	F
A	A	E	G	N	T	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	R	F
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	A	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	I	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	A	V	W	K	E	I	K	F
A	R	#	G	N	E	I	I	R	E	A	S	K	W	S	P	-	-	-	-	-	-	-	-	-	-	-	-	-	-
A	R	E	G	N	E	I	I	P	A	A	S	K	W	S	P	E	L	A	A	A	C	E	I	W	K	E	I	K	F
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	Q	F
A	R	E	G	N	E	I	I	R	E	A	T	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	-	-	-	-	-	-	-	-	-	-	-	-	-	-	R	E	#	S	K	W	S	
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	#	V	W	K	E	I	K	F
A	R	E	G	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
A	R	Q	G	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	#	A	S	K	W	S	P	E	L	A	A	V	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	A	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	A	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	N	L
A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	I	I	R	A	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	#	E	I	K	F
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
A	R	E	G	N	E	V	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	V	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
A	R	E	G	N	E	V	I	R	E	A	C	K	W	S	P	G	L	A	A	A	C	E	V	W	K	E	S	K	F

2019

2021

Sample alignments from recently published papers

These 45,367 hierarchical orthologous groups, or HOGs, were filtered to retain 16,151 HOGs with sequences for at least four species. Protein sequences were aligned with MAFFT v. 7.245 ([Kato and Standley, 2013](#)), and filtered in three steps. First, entire columns were excluded if missing in more than 30% of species, had sequence in fewer than 10 taxa, or was missing in two of the three of the main taxonomic groups (paleognaths, neognaths, or non-avian outgroups). Second, poorly aligned regions were masked according to [Jarvis et al. \(2014\)](#) using a sliding-window similarity approach. Third, columns were removed using the same criteria as the first round. Next, entire sequences were removed from each alignment if they were over 50% shorter than their pre-filtered length or contained excess gaps. Finally, entire HOGs were removed if they contained more than three sequences for any species, did not have more than 1.5x sequences for the given number of species present in the alignment, or were less than 100 base pairs long. Nucleotide sequences for all remaining HOGs were aligned with the codon model in Prank v. 150803 ([Löytynoja and Goldman, 2008](#)). In total, 11,247 HOGs remained after all alignment and filtering steps.

**After the application of “due diligence” alignment masking and filtering techniques**



# Sample alignments from recently published papers

chaVoc	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
egrGar	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
pygAde	F	K	G	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
aptFor	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
nipNip	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
anaPla	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
melGal	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
galGal	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	V	E
strCam	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aptRow	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aptHaa	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aptOwe	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
rhePen	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
rheAme	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
droNov	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
casCas	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
eudEle	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
notPer	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
tinGut	F	K	G	W	S	G	G	L	-	-	-	-	-	-	-	-	-	-
cryCin	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
aquChr	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
halLeu	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E
lepDis	F	K	G	F	R	G	G	L	D	V	S	H	G	Q	T	G	A	E

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MALMA_AF344483	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
MALMA_AF344519	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
MALMA_AF344467	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
MALMA_AF344470	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
MALMA_AF344507	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
BBOBB_AJ235773	A	R	E	G	N	E	I	I	R	E	A	S	N	W	S	P	E	L	A	A	A	C	A	V	W	K	E	I	K	F
HANCO_AF323190	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
HANCO_AF323188	A	R	E	G	N	E	I	#	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
CRNCO_AY530929	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
CRNCO_L14395	A	#	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
CRNCO_AY530927	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	#
CRNCO_AF190433	A	S	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
CRNCO_L11217	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	I	W	K	E	I	K	F
BERRA_AF203487	A	A	E	G	N	T	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	R	F
COMMY_AF281478	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
PCIER_AJ428893	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	A	I	K	F
MGVER_Z80200	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	I	W	K	E	I	K	F
SLAER_AF421109	A	R	E	G	N	E	I	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	A	V	W	K	E	I	K	F
TSTER_Z83149	A	R	#	G	N	E	I	I	R	E	A	S	K	W	S	P	-	-	-	-	-	-	-	-	-	-	-	-	-	-
SYMER_Z80192	A	R	E	G	N	E	I	I	P	A	A	S	K	W	S	P	E	L	A	A	A	C	E	I	W	K	E	I	K	F
SLAER_AF421108	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
PPLER_AF419239	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	Q	F
SPTER_Z83135	A	R	E	G	N	E	I	I	R	E	A	T	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
LECER_AF077656	A	R	E	G	N	E	I	I	-	-	-	-	-	-	-	-	-	-	-	-	-	-	R	E	#	S	K	W	S	
LECER_Z80183	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	#	V	W	K	E	I	K	F
LECER_Z80202	A	R	E	G	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
LECER_Z80180	A	R	Q	G	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
LECER_AF077652	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
LECER_AF077653	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
LECER_Z80177	A	R	E	G	N	E	I	I	R	#	A	S	K	W	S	P	E	L	A	A	V	C	E	V	W	K	E	I	K	F
LECER_Z80181	A	R	E	G	N	E	I	I	R	A	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
LECER_AF022134	A	R	E	G	N	E	I	I	R	A	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
PMOER_Z83144	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
THPER_AF213818	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
THPER_AF213819	A	R	E	G	N	E	I	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	N	L
THPER_AF213817	A	R	E	G	N	E	I	I	R	E	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
THPER_AF213815	A	R	E	G	N	E	I	I	R	A	A	S	K	W	S	P	E	L	A	A	A	C	E	V	W	#	E	I	K	F
PRMER_AF395005	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
PRMER_AF394965	A	R	E	G	N	E	V	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
PRMER_AF395006	A	R	E	G	N	E	V	I	R	E	A	C	K	W	S	P	E	L	A	A	A	C	E	V	W	K	E	I	K	F
PRMER_AF213794	A	R	E	G	N	E	V	I	R	E	A	C	K	W	S	P	G	L	A	A	A	C	E	V	W	K	E	S	K	F

2021

After the application of “due diligence” alignment masking and filtering techniques



Immune genes are hotspots of shared positive selection across birds and mammals

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