

# Eagle: multi-locus association mapping on a genome-wide scale made routine

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Supplementary Table 1: Implementation and methodology attributes of eight computer programs/packages for genome-wide association mapping.

Attributes	Eagle	bigRR	glmnet	LMM-Lasso	MLMM	r2VIM	FaST-LMM	GEMMA
<i>Implementation</i>								
Purpose built <sup>a</sup>	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes
Language	R/C++	R	Python	R	R	C++ and Python <sup>b</sup>	C++	C++
GUI	Yes	No	No	No	No	No	No	No
Documentation	Videos, user-manuals, website, R help	R help	Vignettes, R help	Readme.txt, test script	Vignette, R help	R help	Videos, website user-manuals,	User-manual, website
Additional fixed effects <sup>c</sup>	Yes	Yes	Yes	No	Yes	No	Yes	Yes
Types of trait data	Cont.	Cont., binary, count	Cont., binary, count	Cont.	Cont.	Cont., binary	Cont.	Cont., binary
Data larger than memory	Yes	No	No	No	No	No	Yes	No
<i>Methodology</i>								
Model <sup>d</sup>	LMM	HEM	GLMM	LMM	RF	LMM	LMM, mvLMM	Bayesian Sparse LMM
SNPs fitted <sup>e</sup>	All/multiple	All	All	Multiple	Multiple	Single	Single	Single
Selection type	Model	Variable	Variable	Model	Variable	Variable	Variable	Variable
Threshold free	Yes	No	No	No	Yes	No	No	No

<sup>a</sup> Specifically created for the analysis of GWAS data.

<sup>b</sup> Separate programs, one written in Python, the other C++

<sup>c</sup> Capacity for additional fixed effects (such as age, sex, and/or population structure effects) to be included directly in the model.

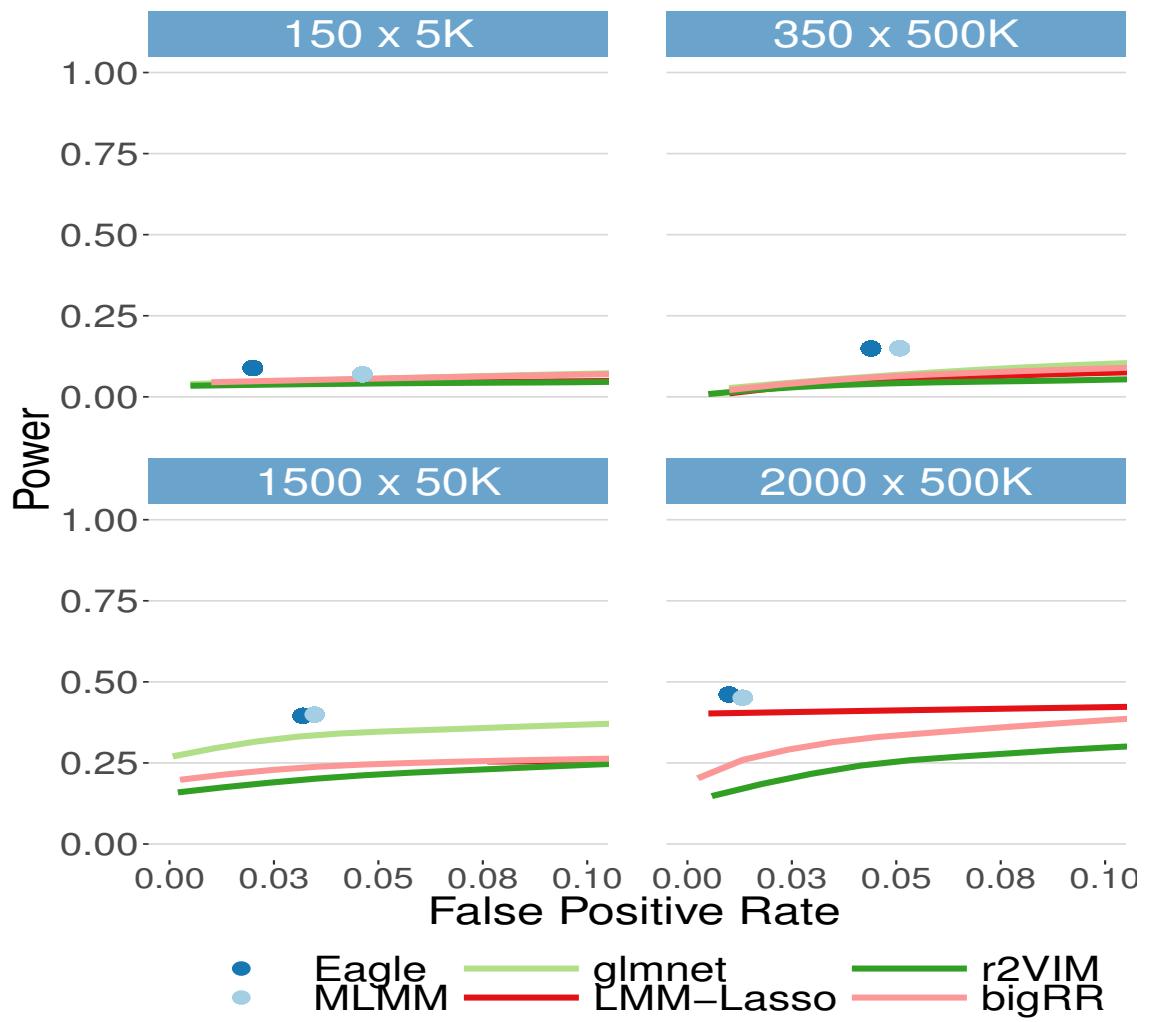
<sup>d</sup> For the different types of model, LMM is linear mixed model, GLMM is generalised linear mixed model, and RF is random forests.

<sup>e</sup> Association is assessed a SNP at a time (single), for multiple SNPs (multiple), or for all SNPs (all). Eagle fits all SNPs but also identifies multiple SNPs (All/multiple) in association with the trait.

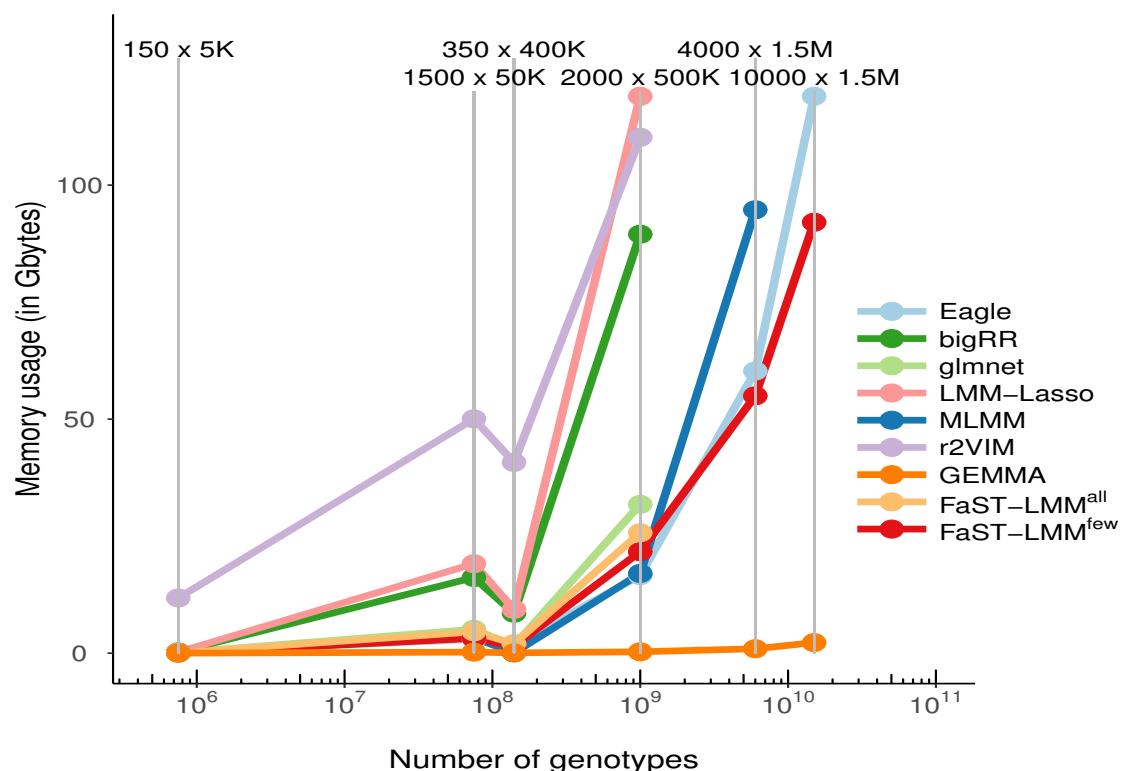
Supplementary Table 2: New SNP-trait associations found from the analysis of the mouse data with Eagle. Due to many of the traits in the study being correlated, genetically, these new findings could be confirmed through their prior discovery with other traits in the original mouse study (Prior Associations).

Method	New SNP-trait Association				Prior Associations		
	Trait	Chrm	Position (Mb)				
Eagle <sup>alt</sup>	Bioch.HDL	6	17.53		Muscles.Sol.g	Muscles.TA.g	Muscles.Plant.g
					Muscles.EDL.g	Muscles.Gast.g	SPPI.ln.pa
Eagle <sup>optimal</sup>	Bioch.LDL	6	17.53		Bioch.Calcium	Bioch.Tot.Cholesterol	
					Bioch.Tot.Protein	Bioch.Albumin	
Eagle <sup>optimal</sup>	Bioch.Tot.Cholesterol	4	134.58		Muscles.Sol.g	Muscles.TA.g	Muscles.Plant.g
	Bioch.HDL	4	134.58		Muscles.EDL.g	Muscles.Gast.g	SPPI.ln.pa
Eagle <sup>optimal</sup>		6	17.53		Bioch.Calcium	Bioch.Tot.Cholesterol	
					Bioch.Tot.Protein	Bioch.Albumin	
BMC.Mean	BMC.Mean	15	86.57		FACS.CD3posCD44posCD4CD8Ratio		
	BMC.Mean.N	5	24.64		FACS.CD3posCD44posCD4CD8Ratio		
BMC.Max.N	BMC.Mean				Muscles.Sol.g	Muscles.TA.g	Muscles.Plant.g
	BMC.Mean.N	15	86.57		Muscles.EDL.g	Muscles.Gast.g	SPPI.ln.pa
BMC.Max.N	Bioch.LDL	6	17.53		Bioch.Calcium	Bioch.Tot.Cholesterol	
					Bioch.Tot.Protein	Bioch.Albumin	

Supplementary Figure 1: Power verse false discovery rates for Eagle and the multi-locus methods. Plots for only those simulation scenarios where all multi-locus methods could be implemented are shown. Eagle has the highest power across the four scenarios but MLMM also performs well. A false discovery rate of greater than 10% is typically not employed in GWASs so the upper limit of the x-axis is 0.1.



Supplementary Figure 2: Memory usage (in gigabytes) of Eagle and the other association mapping programs/packages across the six simulation scenarios. The maximum amount of memory on the computer is 128 gigabytes. The x-axis is on the log scale. GEMMA, a single-locus implementation, had the lowest memory usage. Of the multi-locus implementations, Eagle had the lowest memory usage. Also, it was the only multi-locus implementation able to produce results for data under scenario 10000 x 1.5M. This is due to its ability to handle data larger than the available memory of a computer. FaST-LMM was run where all the SNP data are used to estimate the relationship matrix (FaST-LMM<sup>all</sup>) and where genotype data from every five-hundredth SNP are used to estimate the relationship matrix (FaST-LMM<sup>few</sup>)



Supplementary Figure 3: Manhattan plots of the single-locus analysis of the mouse data with FaST-LMM. Genome-wide significance thresholds for FaST-LMM were calculated, via the Bonferroni correction (pink horizontal line) and permutation (blue horizontal line), at a 5% significance level.

