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Article title: Assessing the Effects Imputation on ED Values

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Abstract

Introduction

Evidence from the fossil record and present-day studies argue we are in the midst of, or entering, a sixth mass extinction (Barnosky et al. 2011; Ceballos et al. 2015), such that more species than ever are declining and/or in danger of extinction across a range of environments (Wake & Vredenburg 2008; Thomas et al. 2004). Habitat destruction (Brooks et al. 2002), invasive species (Molnar et al. 2008), climate change (Pounds et al. 2006), and disease (Lips et al. 2006) are some of the leading causes of species declines globally. Conservation biologists seeks to overcome these declines and their detrimental effects on species populations. Even still, researchers and conservationists are confronted with "Noah's Ark problem" (Weitzman 1998), or an unfortunate reality of insufficient, finite resources to confront the increasing amount species requiring conservation effort.

Conservation triage has provided an efficient decision making process for allocating finite resources to obtain the greatest return (Bottrill et al. 2008). One of these triage strategies which have been introduced and used most widely is EDGE (Isaac et al. 2007). This method prioritizes species according to two metrics: evolutionary distinctiveness (ED) and global endangerment (GE). ED measures relative contributions to phylogenetic diversity made by each species within a particular clade (Isaac et al. 2007). Such contributions are assessed by quantifying the amount of branch length which is unique to each species within the overall phylogeny. GE values are assessed by assigning numerical values to each of the World Conservation Union (IUCN) Red List Categories. As species become increasingly threatened and are placed into more concerning categories (e.g. from Vulnerable to Endangered), the GE numerical value increases. Increases in either ED or GE place a particular species at a higher priority for conservation effort.

In the event of missing DNA or trait data, species are often difficult or not able to be placed onto a phylogeny. Even in the face of such uncertainty and missing data, it is understandable

that conservation biologists want to make prioritizations. However, if we are using a quantitative method for prioritizing species, we should remain consistent even when uncertainty arises. To our knowledge, a proper and efficient method for prioritizing species where there is missing data is still untested. This issue pertains mainly to the calculation of ED than GE. The IUCN has collected data on most major clades, and has a strategy for assigning Red Listing values these species which we know little information and are considered Data Deficient (DD). IUCN and other conservation organizations support focus on DD species just the same as Critically Endangered and Endangered species to ensure consistency (Rodrigues et al. 2006). The major area of uncertainty in phylogenetic prioritisation is phylogenetic data. In the past, missing species data and poorly resolved trees have been addressed using imputation (Collen et al. 2011; Isaac et al. 2012; Jetz et al. 2014). However, to our knowledge, there has been no systematic investigation of the efficacy of such imputation, both in terms of the accuracy with which imputed ED values are estimated, and the effect on other known species' scores. Indeed, it is unclear whether any significant information on ED is gained by imputing species which cannot be placed on the phylogeny. It is also not well understood how simply removing missing species, compared to performing imputation, would effect ED values. It may be that simply excluding missing species may be less intrusive than imputation. In searching for a solution for missing species, we may be negatively affecting correct ED values and disrupting EDGE rankings in the process. As the desire to use ED and phylogenies for conservation triage grows, the importance of such tests and a consensus on how to resolve cases of phylogenetic uncertainty becomes more urgent.

Here, we assess and compare the impact that missing species versus phylogenetic imputation has upon ED values. In doing so, we hope to understand the effect that both methods have on ED values and offer a viable solution for dealing with missing data species. Missing species were simulated and removed from trees in two ways: randomly and in a phylogenetically biased manner. Additionally, we tested how ED values were affected by resolving and imputing polytomies of varying sizes on a phylogeny. We found that ED values are by

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₆₄ removing missing species. Here is a reference to figure.

Methods

₆₆ We attempted to demonstrate the effects of removing or imputing species which cannot be
₆₇ placed onto a phylogeny. While assessing the impact of dropping species and phylogenetic
₆₈ imputations, we were primarily focused on ED values. In testing the effects on these values,
₆₉ we remain focused upon ED as a single variable. We exclude GE because it would only
₇₀ add complexity while not providing any additional information to our particular investiga-
₇₁ tion.

₇₂ In each test, we simulated 100 phylogenetic trees and manipulated each tree's tips or clade.
₇₃ All simulations, calculations, and analyses were performed using R (version 3.4.0). Original
₇₄ and manipulated trees were simulated under a pure-birth Yule model using the sim.bdtree
₇₅ function 'geiger' R package (Harmon et al. 2007). This particular model was chosen to
₇₆ maintain simplicity. Results from this simple model should be applicable to other more
₇₇ complex scenarios. Also, to reduce uncertainty, we used the same model throughout each
₇₈ of the simulations. In reality, we would be estimating the parameters of the model which
₇₉ the phylogeny is built upon. The function ed.calc within the R package 'caper' was used to
₈₀ calculate ED values for each tree (Orme 2013).

₈₁ We assessed the impact that removing missing species has upon ED values using the cor-
₈₂ relation of all ED values for the tips remaining within both trees. To evaluate the effect
₈₃ that imputation has upon ED values, we calculated ED for all tips in both the original and
₈₄ manipulated trees while excluding the focal clade where imputation has occurred. These
₈₅ ED values were compared using a correlation. Additionally, we did the same calculations
₈₆ and comparison using only the original focal clade and its' simulated replacement. If miss-

87 ing species have no effect upon ED values, we expect a high, positive correlation coefficient

88 between the original tree and its' manipulated counterpart.

Assessing the impact of missing species on EDGE-listing

90 If missing species has a negative effect on ED values, then the correlation between the ED

91 of the species in the original tree and the same tree with a fraction of tips removed in some

92 manner should be significantly different from 1. To investigate the degree of this effect,

93 we removed tips from simulated trees (Number of taxa = 64, 128, 256, 512, 1024, 2048,

94 4096) at random and by phylogenetic clusters. To assess the effect under varying amounts

95 of uncertainty, fractions of tips dropped ranged from 0 to 0.2 of total tree tips.

96 Missing species at random was simulated by selecting species at random without replacing,

97 and removing those species from the tree. This randomization had no regard for phyloge-

98 netic structure. Missing species related by some character trait was tested by simulating

99 character trait values for each tip. These simulations were all performed under a constant

100 rate Brownian-motion model (par = 0.5, starting root value = 1). Tips were dropped if

101 their character trait values place them into the upper quantile which had been selected to

102 be dropped. More specifically, if the fraction to be dropped was 0.1, species within the 90th

103 quantile of character trait values are dropped. This is equivalent to Felsenstein's threshold

104 model (Felsenstein & Felenstein 2004).

Assessing the impact of phylogenetic imputations

106 We tested the impact of imputing missing species onto a clade of a particular size (sizes 3

107 through 32) which originated from a tree of a particular size (Number of taxa = 64, 128, 256,

108 512, 1024). To simulate the effect that phylogenetic imputation has upon these simulated

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trees, we randomly chose clades within each tree and treated it as a polytomy to be resolved. The clade selected was removed from the original tree and a new separate tree of the same size was simulated under the pure-birth model used before and placed back where the original clade was removed. By doing this , we replicated the process of imputation of a clade which has been resolved. To ensure that this is representative of cases where imputation is used, 100 repetitions of this simulation were performed across different parameter settings.

Results

Assessing the impact of missing species on EDGE-listing

ED values for remaining species were significantly affected by the fraction of species which were removed (Table 1). However, different effects are seen when dropping species at random and in clustered manner (Fig. 1). Dropping species at random has a reduced effect when compared to the effect which dropping species in a clustered manner has on remaining ED values (Fig. 1).

	Estimate	Std. Error	t value	Pr($>$|t|)
(Intercept)	0.9991	0.0003	3752.33	0.0000
Fraction of Species Dropped	-0.3434	0.0021	-164.89	0.0000
Random Treatment	-0.0004	0.0004	-0.96	0.3360
Number of Species Overall	0.0000	0.0000	0.13	0.8932
Fraction of Species Dropped:Random Treatment	0.3129	0.0029	106.25	0.0000
Random Treatment:Number of Species Overall	0.0000	0.0000	9.20	0.0000

Table 1: **ANCOVA model summary describing the effect of dropping species on remaining species ED Values.** The fraction of species dropped significantly affects the the remaining ED values. Dropping the fraction at random had a reduced compared to dropping species in a clustered manner ($F_{29688,5} = 12090$, $R^2 = 0.6706$, p$<$0.0001).
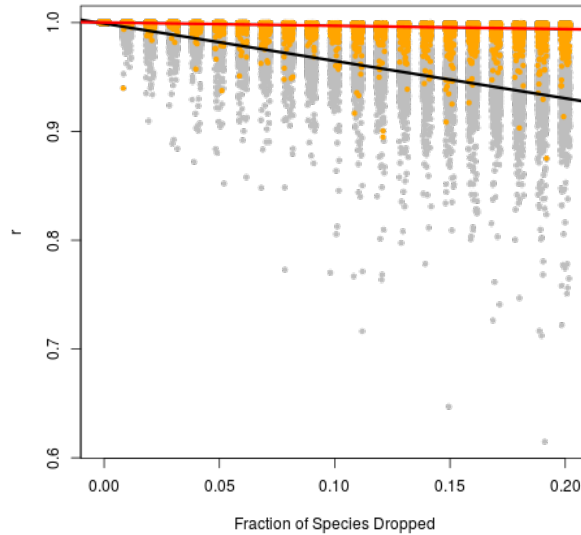
Figure 1: **R-values plotted against the fraction of species dropped at random versus clustered manner.** The color of data points denote whether species were dropped at random (orange; n = 100) or in clustered manner (grey; n = 100). The regression lines are demonstrating the relationship when species are dropped at random (black) and in a clustered manner (red).

Assessing the impact of phylogenetic imputations

ED values for the full tree while excluding the focal clade remain at 1 and unaffected.

However, ED values for the imputed clades are significantly affected by the use of imputation.

As the size of the focal clade increases, the informative value of the ED values within the clade

decreases (Fig. 2). However, even when imputing smaller clades, ED values did not regularly

reflect the true ED values (Table 2). Our analysis demonstrates that measures of the original

phylogeny such as phylogentic diversity (PD), lambda, Colless' Index, skew, and kurtosis do

not provide any indication that imputation would negatively affect ED values (Appendix

A). Additionally, just as imputed ED values did not reflect true ED values, the rankings of

species within the focal clade were altered significantly (Fig. 3). Our model suggests that

with increases in the size of the imputed clade and overall number of species, species within

the clade are ranked farther from their true ranking (Table 3). More specifically, our model

suggests that by imputing a clade of three species within a phylogeny of 128 species, the

species within the clade would be 60 rankings from their true rank on average. ED values

outside of the focal clade were not affected by imputation. While ED values within the focal

clades were affected exclusively by imputation, a notable error rate in ranking crucial species
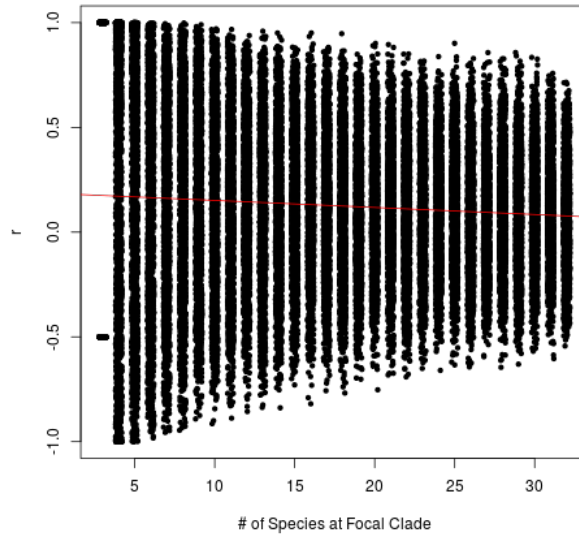
correctly is present (Appendix B).

Figure 2: **R-values plotted against the number of species at focal clade.** Each data point denotes a correlative comparison between ED values within the focal clades where imputation has occurred. The regression line (red) and trend even closer to zero demonstrates the decrease in informative value of the imputed ED values. This is reinforced by the visual narrowing of r-values around zero.

	Estimate	Std. Error	t value	Pr(>|t|)
(Intercept)	0.1852	0.0533	3.47	0.0005
Size of Focal Clade	-0.0034	0.0002	-16.56	0.0000
Size of Phylogeny	-0.0001	0.0001	-0.41	0.6855
PD	0.0001	0.0001	0.37	0.7108
Lambda	-0.0012	0.0524	-0.02	0.9812
Colless' Index	0.0016	0.0022	0.72	0.4687
Skew	0.0043	0.0088	0.48	0.6288
Kurtosis	-0.0005	0.0009	-0.63	0.5269

Table 2: Effect of Clade Size on Imputed ED Values. The intercept describes that the correlation between the true and imputed values begins quite low. As the clade size increases, this correlation only tends toward zero. The total number of species in the full phylogeny along with measures of the true phylogenetic diversity, lambda, Colless' Index, skew, and kurtosis show no significant effect. ($F_{47992,7} = 39.57$, $R^2 = 0.006$, p<0.0001).
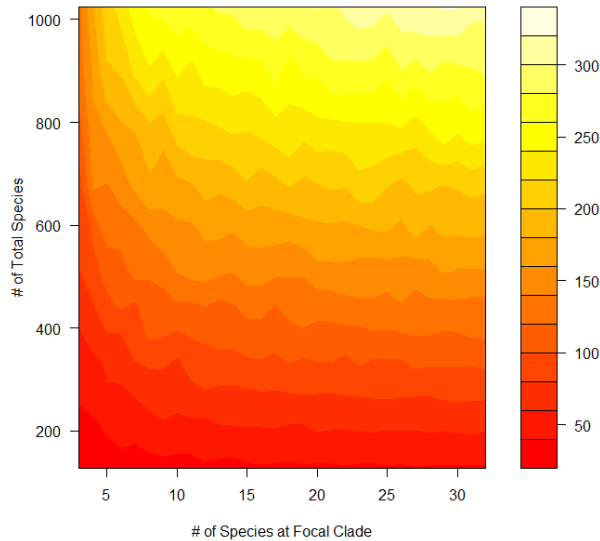
Figure 3: **Mean ranking error of species within the focal clade.** The gradient on the right demonstrates average number of posistions within the full ranking that focal clade species shifted from their true rank. While controlling for the size of the full phylogeny and focal clade, species within the focal clade were, on average, ranked far from the true rank.

	Estimate	Std. Error	t value	Pr($>$|t|)
(Intercept)	-1.6344	0.0332	-49.29	0.0001
Size of Focal Clade	0.0900	0.0010	91.22	0.0001
Size of Phylogeny	0.5179	0.0013	383.99	0.0001

Table 3: Effect of Clade Size and Total Species on Ranking Error. Model demonstrating the relationship between focal clade species ranking error and the size of imputed clade and overall phylogeny. Square-root transformations have been applied to both ranking error and size of phylogeny. Significant increases ranking error are seen when increasing sizes of both the imputed clade and phylogeny ($F_{47997,2} = 77890$, $R^2 = 0.7644$, p$<$0.0001).

Discussion

Acknowledgments

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A. Effect of Measures of the True, Full Phylogenies
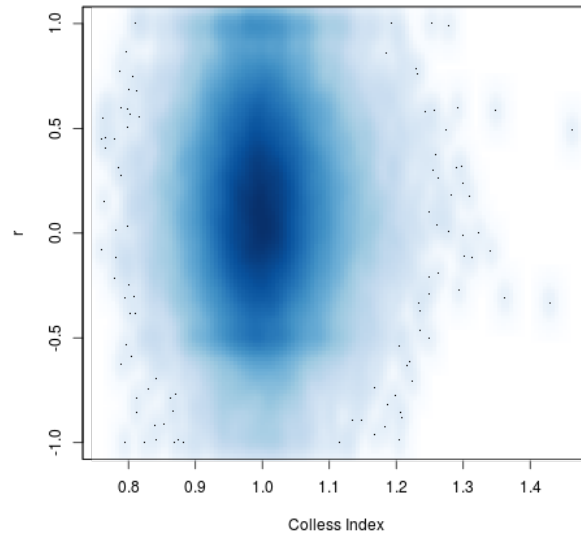

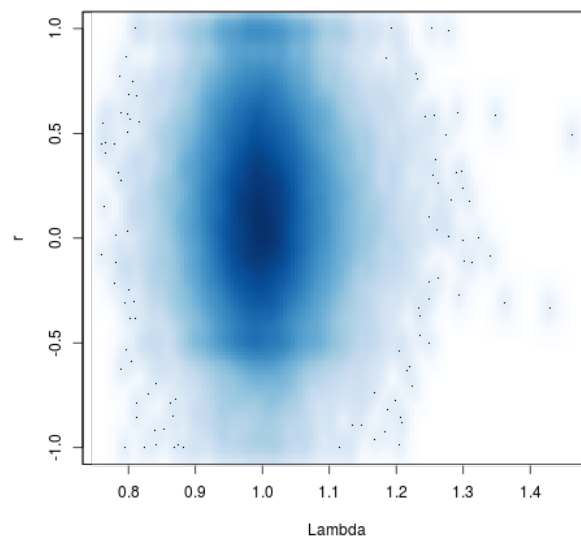Figure 4: **Effect of the True Colless Index of FullPhylogeny.**

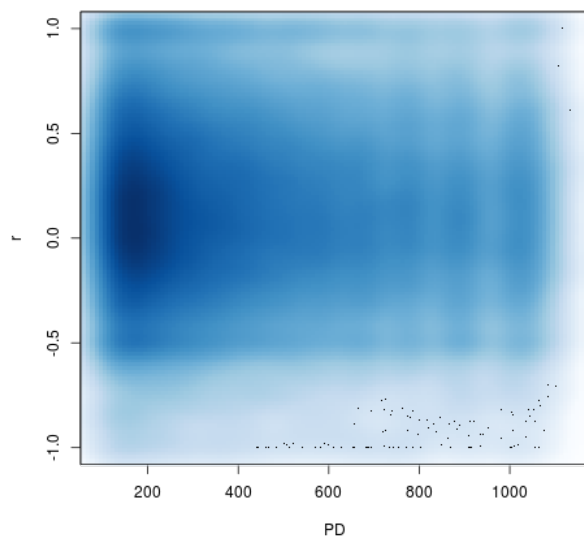Figure 5: **Effect of the True Lambda of Full Phylogeny.**
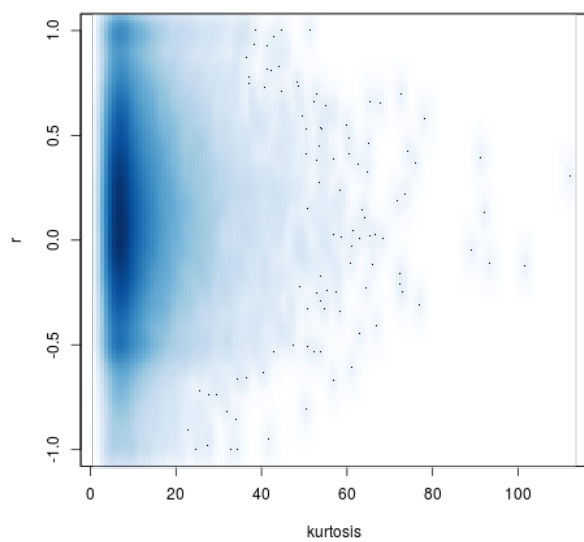
Figure 6: **Effect of True PD of Full Phylogeny.**

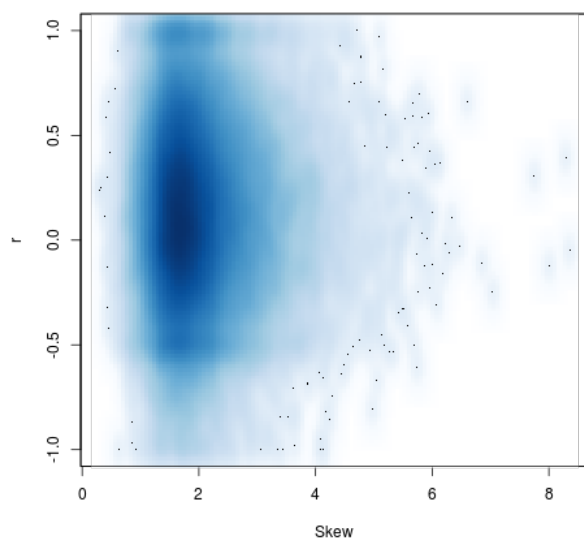Figure 7: **Effect of the True Kurtosis of Full Phylogeny.**

Figure 8: **Effect of the True Skew of Full Phylogeny.**
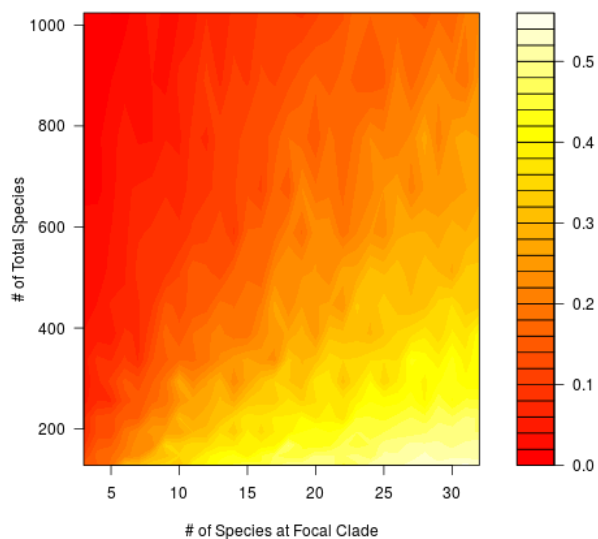
B. Error Rate in Top Rankings

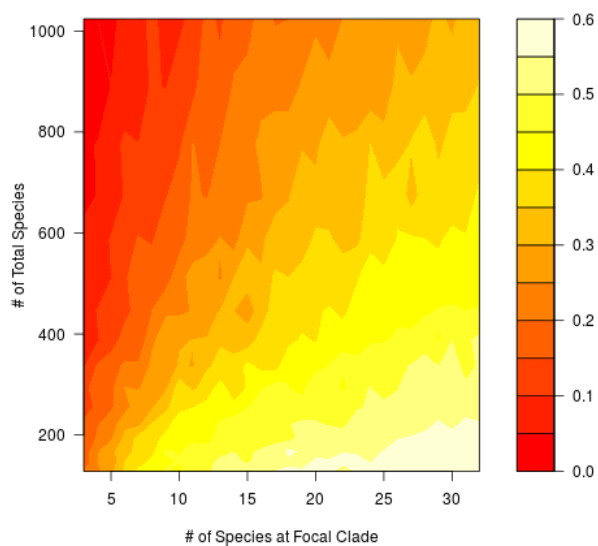Figure 9: **Mean error rate in the ranking of top 50 species.**

Figure 10: **Mean error rate in the ranking of top 100 species.**
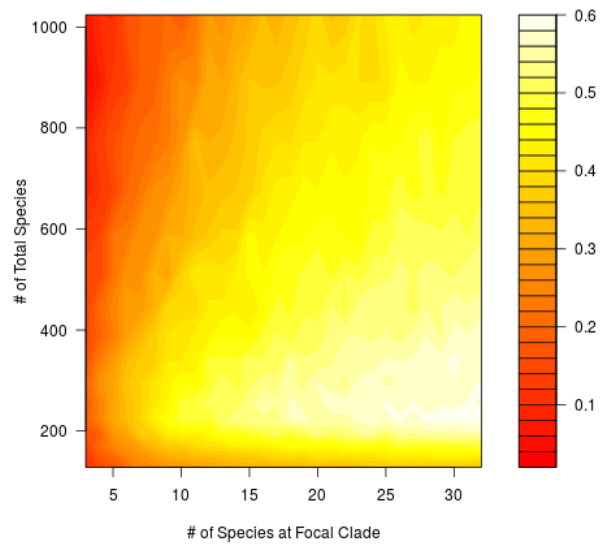
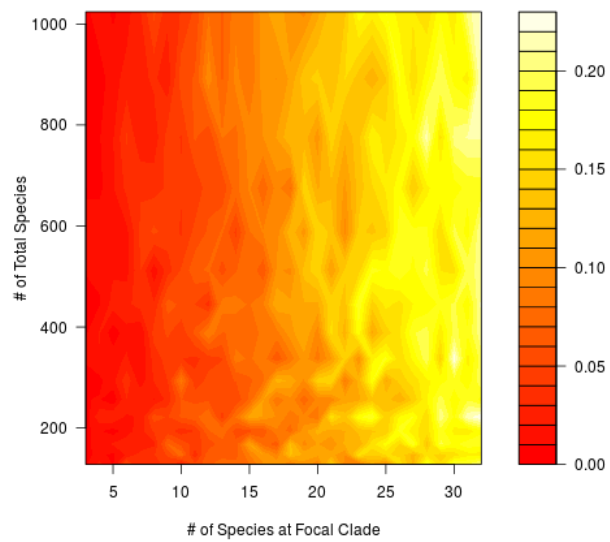Figure 11: **Mean error rate in the ranking of top 200 species.**

Figure 12: **Mean error rate in the ranking of top 5% of species.**

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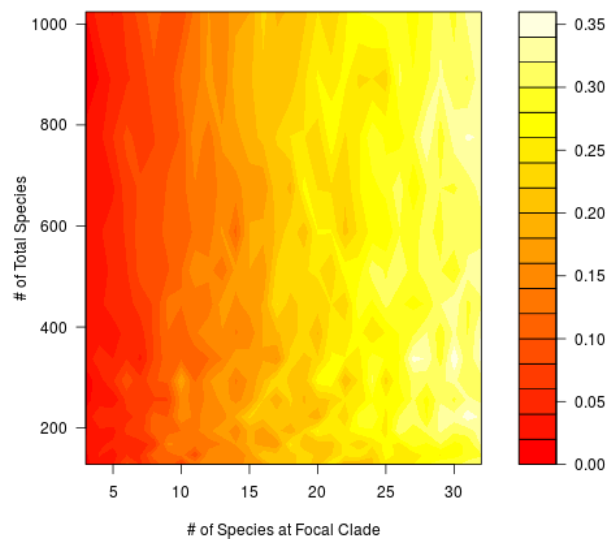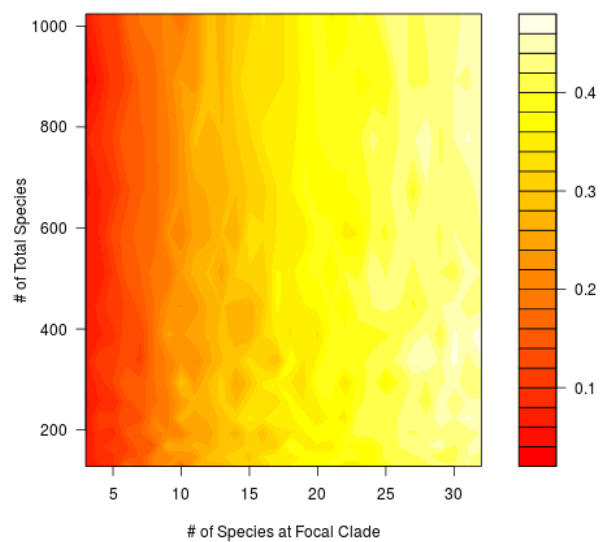Figure 13: **Mean error rate in the ranking of top 10% of species.**

Figure 14: **Mean error rate in the ranking of top 20% of species.**