A quantitative framework for investigating the reliability of empirical network construction: supplemental information

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Appendix S1: Mathematical framework

A naive quantification of uncertainty

We start by considering how to naively quantify an interaction probability and its associated uncertainty *for an interaction that has not yet been observed*. Consider the case where a pair of species have been observed co-occurring n times, of which they have been observed to interact in $k = 0$ cases. To evaluate the uncertainty of this interaction, consider the occurrence of an interaction as a Bernoulli trial. In this framework, the number of successes k over n trials will follow a binomial distribution:

$$L \sim Bin(n, \lambda), \tag{1}$$

$$P(L = k | \lambda, n) = \binom{n}{k} \lambda^k (1 - \lambda)^{n-k}. \tag{2}$$

The parameter λ, the probability of observing an interaction over an infinite time interval and area, is the quantity we want to estimate from empirical data. The maximal likelihood estimate (MLE) of λ is straightforward to find given k and n:

$$\lambda_{MLE} = \frac{k}{n}. \tag{3}$$

Posterior distribution of the interaction probability

Here we adopt a Bayesian approach to estimate the posterior distribution of the parameter λ :

$$\underbrace{P(\lambda | k, n)}_{Posterior} = \frac{\overbrace{P(k | \lambda, n)}^{Likelihood} \overbrace{P(\lambda)}^{Prior}}{\underbrace{P(k | N)}_{Normaliser}}. \tag{4}$$

According to the above description, the likelihood is simply the binomial distribution (Eq. 2). Since λ is a probability, it is bounded between 0 and 1 and the most appropriate prior distribution is the beta:

$$\lambda \sim Beta(\alpha, \beta), \tag{5}$$

which has two shape parameters, α and β.

The beta-binomial distribution is a conjugate distribution of the binomial distribution. This allows us to analytically compute the posterior distribution of a binomial model with a beta prior distribution. We can re-write the posterior distribution of λ as:

$$P(\lambda | k, n) = \frac{\lambda^{\alpha+k-1}(1 - \lambda)^{\beta+n-k-1}}{B(\alpha + k, \beta + n - k)}, \tag{6}$$

where the function B is the beta function. The posterior distribution of λ therefore follows the beta distribution with new parameters $\alpha' = \alpha + k$ and $\beta' = \beta + n - k$.

Appendix S2: Calculating uninformative and informative priors

To create Bayesian distributions for the probability of interactions, we must begin with a prior distribution. This distribution reflects our prior knowledge (or lack thereof) about the likelihood of a given interaction. Priors may be selected to be uninformative, so as not to bias the posterior distribution in any particular direction, or may be informative and based on some prior knowledge about the system.

Uninformative prior

In the absence of any external information, an uninformative prior is the most conservative hypothesis for the distribution of λ. The beta distribution is in this case a uniform distribution, specified with hyper parameters $\alpha = 1$ and $\beta = 1$.

Distribution of connectance

The ecological network literature boasts a collection of networks for which connectance has been calculated and for which we can thus define the connectance distribution. Connectance is measured as $C = L/S^2$, where L is the number of interactions and S is the number of species. It measures the filling of an interaction matrix and thereby expresses the average probability that any two species interact with each other. If we know only the mean $\overline{C}$ and the variance σ_C^2 of the distribution of C, then the beta parameters could be computed as follows using the method of moments:

$$\alpha = \overline{C}\left(\frac{\overline{C}(1 - \overline{C})}{\sigma_C^2} - 1\right), \tag{7}$$

$$\beta = (1 - \overline{C})\left(\frac{\overline{C}(1 - \overline{C})}{\sigma_C^2} - 1\right). \tag{8}$$

Degree distribution or interaction probabilities

The degree of a node in a network is defined as its number of connections to other nodes. The degree distribution of a network is then the probability distribution of these degrees over the whole network and the normalised degree (i.e., degree divided by the number of potential interaction partners) could therefore be interpreted as an interaction probability. It is consequently possible to use the degree distribution to inform the prior distribution. The degree distribution could come from several networks, from a similar network (e.g. a known network at slightly different location) or from the network of interest if interaction probabilities for some species are already documented. The latter approach allows researchers to apply information from known, abundant species to the rarest species for which interactions are less frequently documented. That is, we can use detection probabilities for interactions between abundant species to set bounds on detection probabilities for interactions involving rare species.

If our focal network describes a system similar to that in a known network, we can use the distribution of interaction probabilities in that network to inform our prior. The probability of any interaction L_{ij} depends on the degrees of species i and j. Using normalised degrees Δ_i and Δ_j, we can obtain the probability of interaction $L_{ij}=\Delta_i \times \Delta_j$. Similar to the procedure for degree distribution, the distribution of these interaction probabilities can be used to establish a prior distribution before any data from the focal network are collected. For distributions of either degrees or interaction probabilities, the procedure for the estimation of the hyper parameters follows the same approach as described above for connectance except that each measurement is at the individual interaction level instead of the network level.

Trait-matching function

As a fourth and final approach, it may be possible to obtain the prior distribution of λ using the outcome of a trait-matching model, provided such a model has been parameterised using external data and relevant traits are available. In such a case, the prior distribution would follow the function $P(\lambda|\mathbf{T}) = f(\mathbf{T})$ based on a set of traits for both species $\mathbf{T}$. There are several techniques available to perform this inference of interaction probability, some of which are Bayesian, and we refer interested readers to Bartomeus *et al.* (2016) and Weinstein & Graham (2017) for recent reviews about this topic. Note that in this case the prior might not be beta-distributed and numerical methods might be required to compute the posterior distribution.

Trait-matching functions reduce uncertainty around the feasibility of interactions. Interactions between species with extremely mis-matched traits are very unlikely, and we can therefore be more confident that an unobserved interaction does not occur. This approach could be extended to models including environmental or phenological constraints, allowing researchers to reduce process uncertainty as well. Such models are, however, likely to be large and therefore may pose a risk of over-fitting.

A quantitative example

The Bayesian framework can be illustrated with a simple quantitative example. Suppose we have $n = 10$ observations of co-occurrence between species i and species j in a given time interval and area, and $k = 3$ observations of interactions. The maximum likelihood estimate of the interaction probability is simply $\lambda_{MLE} = 3/10 = 0.3$.

Now consider we know that species i is known to interact with 10 species (other than species j), which have the following degrees:

 degree=c(14, 4, 2, 3, 17, 6, 2, 15, 1, 1) .

If the network has 20 species total, this gives the normalised degrees:

 norm_degree=c(0.65, 0.20, 0.10, 0.15, 0.85, 0.30, 0.10, 0.75, 0.05, 0.05).

Species i has a normalised degree of 0.55 (it interacts with species j and 10 other

species). We can combine the normalised degree of i with the normalised degrees of its interaction partners to obtain the following set of interaction probabilities for species i and each of its interaction partners:

$int\_probs = c(0.358,\ 0.110,\ 0.055,\ 0.082,\ 0.468,\ 0.165,\ 0.055,\ 0.412,\ 0.028,\ 0.028)$.

The mean of these interaction probabilities is 0.176, approximately two-thirds the λ_{MLE} obtained from the observed data. We can use the distribution of these interaction probabilities as our prior distribution and estimate the uncertainty surrounding our λ_{MLE}. With some simple R code (function "calculate_parameters", *Appendix S7*), we obtain prior parameters α=0.998 and β=4.63. Using these priors in equations **??** and **??** above (or in the R function "calculate_distribution" in *Appendix S7*), we find a prior $\bar{\lambda}$=0.177 and var(λ)=0.026. Adding the observed data ($n = 10$, $k = 3$) and using the same code, we obtain posterior parameters α'=4.00 and β'=11.6 and a posterior $\bar{\lambda}$=0.256 and var(λ)=0.012. Comparing the posterior distribution to the prior, we see that the posterior is closer to the observed data and that the additional data about interactions between species i and j has reduced the variance. We may also wish to calculate a credible interval (analogous to the frequentist confidence interval). This is also quite straightforward in R (see function "credible_interval" in *Appendix S7*). In this case, a 95% credible interval for $\bar{\lambda}$ is (0.080, 0.491).

Now, consider the case where the two species have never been observed interacting across n trials, i-e. $k = 0$. The question is then "what is the probability that these two species do not interact"? Since it is not possible to prove that the two species could never interact (strictly speaking, in a Bayesian approach $\lambda = 0$ is impossible), we must fix a threshold below which we consider that there is no interaction ($\lambda \sim 0$). We call this threshold probability $\lambda*$. We then use the cumulative distribution function to estimate $P(\lambda < \lambda * | L = 0, n)$ for different n. The function "samples_for_threshold" in *Appendix S7* calculates distribution function for $\lambda*$ with an increasing number of trials. This yields a surprising result: it requires >24 observations of no interactions to be 95% sure that the interaction probability is smaller than $\lambda*$=0.1 (recall Fig. **??**, Box 2). Note the special case where there is no observation of the two species co-occurring, $n = 0$. In this situation, the posterior distribution converges to the prior distribution since the data include no information on the probability with which species might interact should they co-occur.

Appendix S3: Details of *Salix* data collection

The *Salix*-galler-parasitoid meta-network dataset collected by Kopelke *et al.* (2017) consists of a single community type sampled across Europe: willow (*Salix*) species, willow-galling sawflies (Hymenoptera, Tenthredinidae, Nematinae, Euurina), and their natural enemies (hymenopteran parasitoids and coleopteran, lepidopteran, dipteran, and hymenopteran inquilines). The data were collected over 29 years (1982-2010) at 374 unique locations across Europe ranging from Sicily to the Arctic. Since the dataset includes repeated visits to a number of sites, the data set contains 641 site-visits. Here we take the more conservative approach and pool visits to the same site for a sample size of 374 sub-networks.

The meta-network consists of 1,173 different interactions between 52 *Salix* nodes, 92 herbivore nodes, and 126 parasitoid nodes. Interactions were determined by dissecting and rearing gall inhabitants from 165,424 galls. Some sites were visited repeatedly, for a total of 641 site-visits. We consider these to be repeated samples and take the 374 unique sites as our sample size.

Appendix S4: Results for *Salix*-galler component

Computing the prior and posterior distributions

As with the galler-natural enemy component (presented in the main text), we estimated frequencies of *Salix*-galler interactions based on the normalised degree of each species (*Appendix S6*). We obtained prior parameters of α=8.72, β=305 for the *Salix*-galler component. These prior parameters were then used to estimate the posterior distribution.

For species where $n = 0$ (3,986/4,992 *Salix*-galler pairs in our dataset), estimates for the mean and variance of λ_{ij} can be calculated directly from the prior distribution (*Appendix S6*). For the *Salix*-galler network, these parameters were: $\bar{\lambda}$=0.028, $\text{var}(\lambda)$=8.60×10^{-5}; note that this mean is much lower than that obtained using a prior based on Barbour *et al.* (2016, Data available from the Dryad Digital Repository: https://doi.org/10.5061/dryad.g7805). Note that, despite the intensive sampling in the **??** dataset, most *Salix*-galler pairs were never observed co-occurring (3,986/4,992; Fig. S1).

For species with $n > 0$, the posterior distribution will depend on the values of n and k. Here we are interested only in cases where $k = 0$. This gives α'=α and β'=$\beta + n_{ij}$. For a pair of species which co-occurred at all 374 sites and was never observed to interact, the distribution for the *Salix*-galler network would become $\bar{\lambda}_{ij}$=1.27 × 10^{-2}, $\text{var}(\lambda_{ij})$=1.82 × 10^{-5}. As n decreases, the distribution widens and moves away from the origin (Fig. S4). The 95% credible interval for hypothetical *Salix*-galler pairs widened from (0.006, 0.022) if the pair co-occurred at all sites to (0.013, 0.049) if they co-occurred at none (Table **??**).

How many samples are required to reach a minimal precision

In our dataset, the entire 95% credible interval for the probability of an interaction between a *Salix*-galler pair was (0.013, 0.049). We may therefore be 95% confident that the interaction probability for *Salix* and galler species that have not been observed co-occurring is below 0.05. As the peak of the prior distribution for the probability of interaction between *Salix* and galler probabilities is around 0.02 (Fig. S4), to be 95% confident that the interaction probability for these species is below 0.01 would require 1029 observed co-occurrences with no interaction - far more than the number of sites in the (Kopelke *et al.*, 2017) dataset.

Scaling up to network metrics

As with the galler-natural enemy component, we calculated posterior probability distributions for *Salix*-galler pairs that were not observed interacting. For each posterior web, we calculated the connectance of each web, as well the mean links per *Salix*, mean links per galler, and nestedness (NODF). Likewise, we created a suite of detection-filtered networks by randomly sampling 99%, 95%, 90%, 80%, 70%, 60%, and 50% of the interactions included in each posterior network. We then calculated the same network properties as described above.

We find, perhaps not surprisingly, that the posterior webs for the Salix-galler network had higher connectances than the original, observed web (C=0.028 for the observed web

and $0.082 \leq C \leq 0.096$ for the posterior webs; Fig. S6A). The number of links per *Salix* species in the observed web (L_{Salix}=2.71) was similar to those in the posterior webs ($2.53 \leq L_{Salix} \leq 3.19$; Fig S6B). The number of links per galler, however, was lower in the observed web (L_{galler}=1.47) than in the posterior webs, accounting for the increased connectance ($4.67 \leq L_{galler} \leq 5.88$; Fig. S6D). There was a more substantial difference in the nestedness of the observed and posterior webs: the observed network had NODF=0.560 while the posterior networks were more nested ($1.39 \leq NODF \leq 1.94$). Even the networks sampled with a detection filter of 50% had non-zero nestedness (Fig. S6C). This last result highlights the potential for the possibility for network structure to vary when considering the possibility that unobserved species pairs may interact.

Figure S1: A) Most pairs of *Salix* and gallers were never observed co-occurring despite the high levels of replication in our example dataset. For those pairs that were observed together at least once ($n_{ij} > 0$), the number of observed co-occurrences was generally small (<10). Here we show a histogram of the number of pairs of species observed co-occurring at least once. The 3986 *Salix*-galler pairs never observed co-occurring are omitted from the histogram. **B)** Most pairs of species that were observed at the same site were never observed interacting. Here we show a histogram of the number of observed interactions within pairs of co-occurring species. Species which co-occurred but never interacted are included. **C)** Here we show, for each species pair, the number of observed interactions plotted against the number of observed co-occurrences. *Salix*-galler pairs either are never observed interacting or interact almost every time they co-occur. IThe red, dashed line indicates a 1:1 relationship between interactions and co-occurrences.
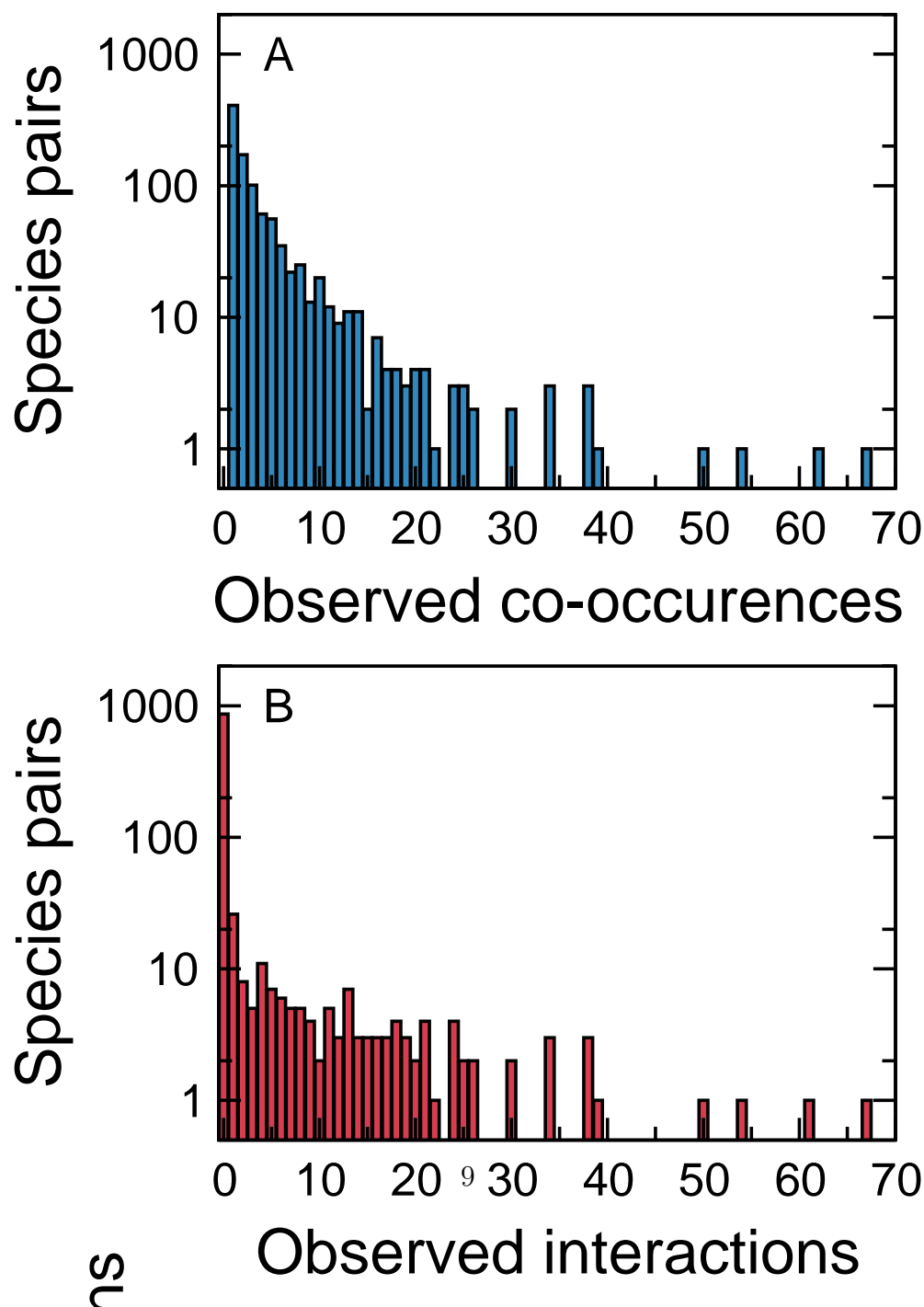
Figure S2: Using prior distributions based on the *Salix*-galler network sampled at a single site in Kopelke *et al.* (2017), we can calculate posterior distributions for the probability of interaction (λ) between two species that have not yet been observed interacting. Here we show posterior distributions for λ ranging from the prior distribution ($n = 0$ observed co-occurrence) to the distribution obtained when the pair of species has been observed co-occurring 100 times. The distribution narrows and approaches zero as the sample size increases. Likewise, the maximum likelihood estimator for the mean probability of interaction (diamonds at top) approaches zero and the 95% credible interval (lines at top) narrows as sample size increases. Dashed lines indicate threshold probabilities of 0.1, 0.05, and 0.01.
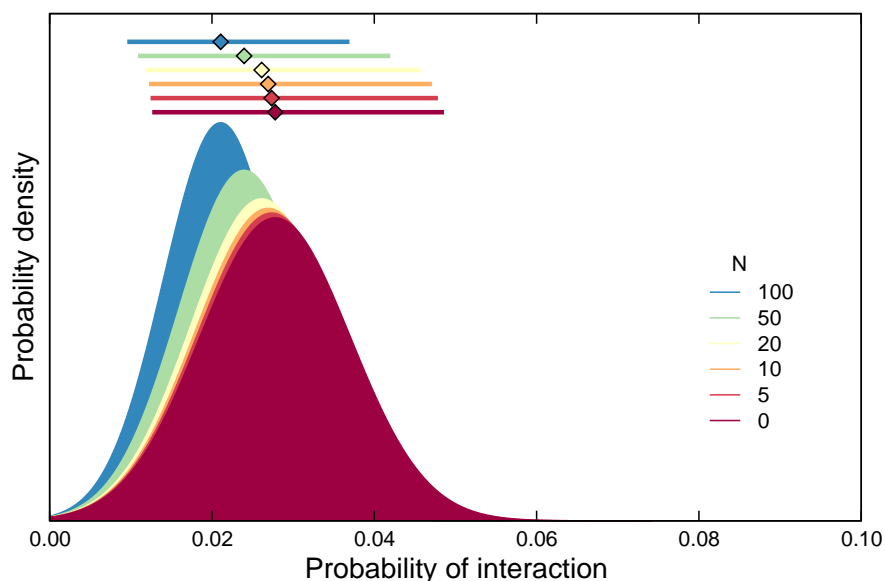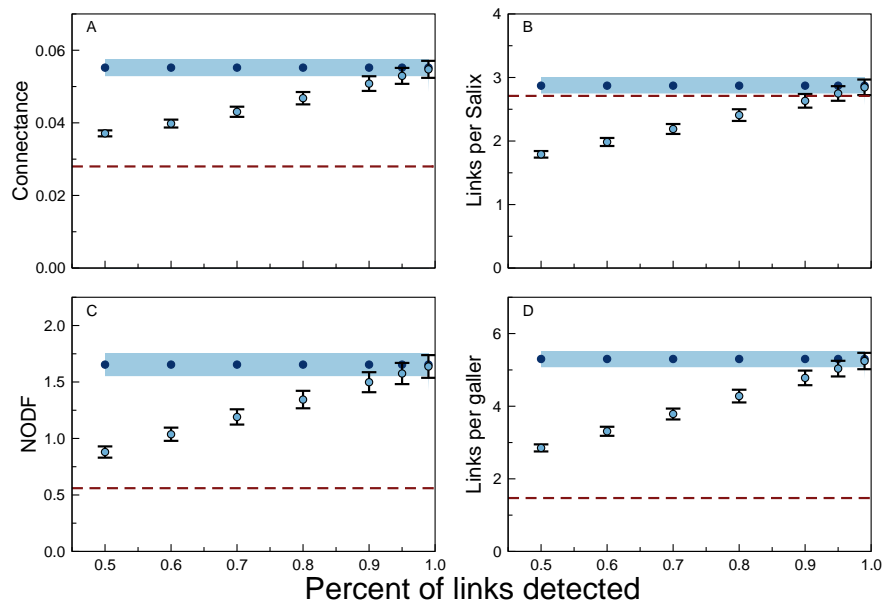
Figure S3: Here we show the mean connectance, links per resource (*Salix* in the *Salix*-galler networks and gallers in the galler-natural enemy networks), links per consumer, and nestedness (NODF) for networks assembled using posterior distributions based on a single sub-network in the Kopelke *et al.* (2017) dataset (Zillis). We created 100 "posterior-sampling" networks and then, for each of these, created 100 "detection-filter" networks by randomly sampling 50%-99% of the interactions included in the posterior-sampling network. This simulates imperfect detection of interactions in the field. Each point represents the mean network property (e.g., connectance) obtained from a set of 100 detection-filter networks, plotted against the value of the network property in the posterior-sampling network used to create the detection-filter networks. For each property and both network types, the posterior-sampling networks cover a relatively small range of network properties than the range covered by networks with varying detection probabilities. The value of each property decreases with the proportion of links included in the detection-filter networks.

Appendix S5: Analyses using alternative prior

We repeated our analyses using an alternative prior derived from a study of a similar *Salix*-galler-natural enemy system in North America (Barbour *et al.*, 2016; Barbour, 2016). We note that this study used several genotypes of *Salix hookeriana* rather than different *Salix* species and so did not produce networks of similar size and connectedness to those in (Kopelke *et al.*, 2017). To obtain the priors based on Barbour *et al.* (2016), we estimated frequencies of *S. hookeriana* genotype-galler interactions based on the normalised degree of each node (species or genotype) in each network component.

Creating a binary interaction network

Barbour *et al.* (2016); Barbour (2016) sampled 145 branches from *Salix hookeriana* of 26 genotypes. They recorded galls made by four species of Cecidomyid midges on each branch. We transformed these data to a binary genotype-galler network matrix, where an entry ij was 1 if galler i was observed on any branch of genotype j and 0 otherwise. In total, this network contained 75 realised galler-genotype interactions out of a potential 104. One *S. hookeriana* genotype did not interact with any gallers and was removed from the network. The R code used to extract this network follows:

prior_web_data=read.csv("tree_level_interaxn_all_plants_traits_size.csv")

 # Remove extra data on parasitoids, etc.
prior_web_data[,31:57]<-NULL
prior_web_data[,4:26]<-NULL
prior_web_data[,2]<-NULL

 # Build a web with interactions=1 for any galler observed on any genotype
prior_web=matrix(nrow=26,ncol=4)
for(r in 1:length(levels(prior_web_data$Genotype))){
    gen=levels(prior_web_data$Genotype)[r]
    subset=prior_web_data[which(prior_web_data$Genotype==gen),]
    for(c in 1:4){
     if(sum(subset[,2+c])>0){
      prior_web[r,c]=1
     } else {
      prior_web[r,c]=0
     }}}
 # Remove non-interactive genotype to obtain the final web
prior_web<-prior_web[which(rowSums(prior_web)>0),]

To extract the galler-parasitoid network from the same study above:

```
prior_web_data=read.csv("tree_level_interaxn_all_plants_traits_size.csv")
prior_web_data <- prior_web_data[,8:26]

# Build a web with interactions=1 for any parasitoid on any galler
prior_web=data.frame()

# each column contains all interactions for a particular parasitoid-galler combination
for(column in 1:ncol(prior_web_data)){
    # the column name gives the species which makes the gall,
    #followed by the species emerging from the gall
    galler_emerged=strsplit(colnames(prior_web_data)[column], "_")
    galler = galler_emerged[[1]][1]
    emerged = galler_emerged[[1]][2]

    # the strength of the interaction is the sum of the column
    (number of galls across all branches sampled)
    linkstrength = sum(prior_web_data[,column])
    prior_web[as.character(galler),as.character(emerged)] <- linkstrength
    }
# Make the NAs into 0
prior_web[is.na(prior_web)] <- 0

# remove emerging gallers with no parasitoids
prior_web_para_only <- prior_web[,c(2:6,8:9,12)]

# convert interaction strengths to 1
prior_web[which(prior_web>0)] <- 1
write.csv(prior_web_para_only, file="prior_web_para_only.csv", quote=F)
```

Prior and posterior distributions

Using the binary networks described above, we obtained prior parameters $\alpha=2.51$ and $\beta=1.89$ for the *Salix*-galler component and $\alpha=1.34$, $\beta=9.49$ for the galler-natural enemy component of the network. We then calculated prior distributions using these parameters. For the *Salix*-galler network, the prior distribution was: $\bar{\lambda}=0.570$, var(λ)=0.056. The prior distribution for the galler-natural enemy network was: $\bar{\lambda}=0.124$, var(λ)=0.010. As with the results in the main text, we can update this prior distribution using observed co-occurrences. In the most extreme case where a pair of species co-occurred at all 374 sites and was never observed interacting, our distributions would become $\bar{\lambda}_{ij}=6.63 \times 10^{-3}$, var($\lambda_{ij}$)=$1.74 \times 10^{-5}$ for the *Salix*-galler network and $\bar{\lambda}_{ij}=3.49 \times 10^{-3}$, var($\lambda_{ij}$)=$9.03 \times 10^{-6}$ for the galler-natural enemy network.

Credible intervals and sampling requirements

The 95% credible intervals calculated using the priors derived from Barbour *et al.* (2016) were much wider than those reported in the main text (Fig. S4). For hypothetical *Salix*-galler and galler-natural enemy pairs these intervals widened from (0.001, 0.017) and (<0.001, 0.11) for pairs observed co-occurring at all 374 sites without any observed interaction to (0.152,0.931) and (0.008, 0.364) for *Salix*-galler and galler-natural enemy pairs that were never observed co-occurring.

If we want to be 95% confident that the interaction probability for two species in the *Salix*-galler network is below 0.1, 0.05, or 0.01, we would need 51, 106, and 550 observed co-occurrences with no observed interaction, respectively (Fig. S5). The number of samples required to be 95% confident that the interaction probability between galler and natural enemy species is below a threshold also increases quickly as the threshold decreases; we would need 25, 62, and 352 observed co-occurrence with no observed interaction for threshold interaction probabilities of 0.1, 0.05, and 0.01, respectively.

Scaling up to network metrics

We find, perhaps not surprisingly, that the posterior webs for the Salix-galler network all had much higher connectance than the original, observed web (C=0.028 for the observed web and $0.528 \leq C \leq 0.568$ for the posterior webs; Fig. S6A). Likewise, the number of links per *Salix* and galler species in the observed web (L_{Salix}=2.71 and L_{galler}=1.47, respectively) were much lower than those predicted in the posterior webs ($27.4 \leq L_{Salix} \leq 29.5$ and $50.6 \leq L_{galler} \leq 54.5$, respectively; Fig. S6C,E), even when filtering with a detection probability of only 50%. Nestedness was also much higher in the posterior networks ($14.2 \leq NODF \leq 15.4$; Fig. S6G) than in the observed network (NODF=0.560). There are two possible explanations for these discrepancies: either the data of Barbour *et al.* (2016) is simply too different from that of Kopelke *et al.* (2017) to offer an appropriate prior for our dataset, or the true detection probability for links between *Salix* and galler species is much less than 50%. As the scale of the two datasets is quite different (genotypes of a single *Salix* species in Barbour *et al.* (2016), various *Salix* species in our dataset), we suspect the former is more likely.

Figure S4: Using prior distribution based on the *Salix*-galler and galler-natural enemy networks in Barbour *et al.* (2016), we can calculate posterior distributions for the probability of interaction (λ) between two species that have not yet been observed interacting. Here we show posterior distributions for each λ in each network ranging from the prior distribution ($n = 0$ observed co-occurrence) to the distribution obtained when the pair of species has been observed co-occurring 100 times. The distribution narrows and approaches zero as the sample size increases. Likewise, the maximum likelihood estimator for the mean probability of interaction (diamonds at top of each panel) approaches zero and the 95% credible interval (lines at top of each panel) narrows as sample size increases. **A)** The posterior distributions for the *Salix*-galler component are always wider and farther from zero than those for **B)** the galler-natural enemy component. This is likely because the prior distribution for the galler-natural enemy component of the network was both narrower and had a mean closer to that in the Kopelke *et al.* (2017) data than did the *Salix*-galler component.
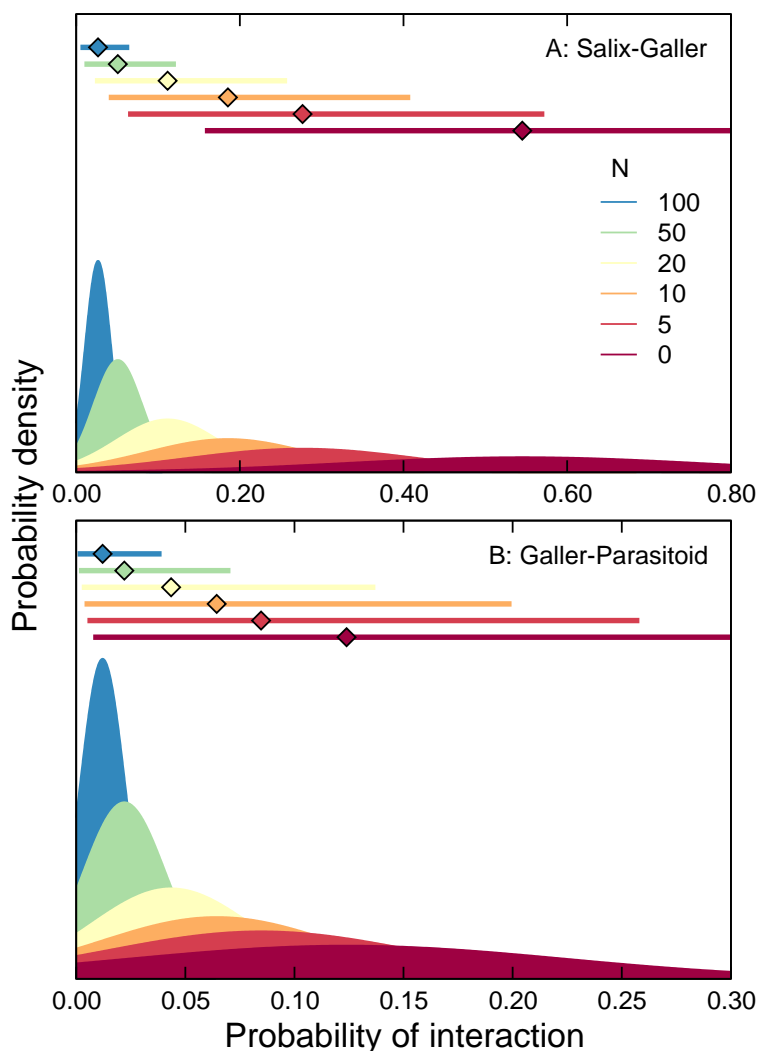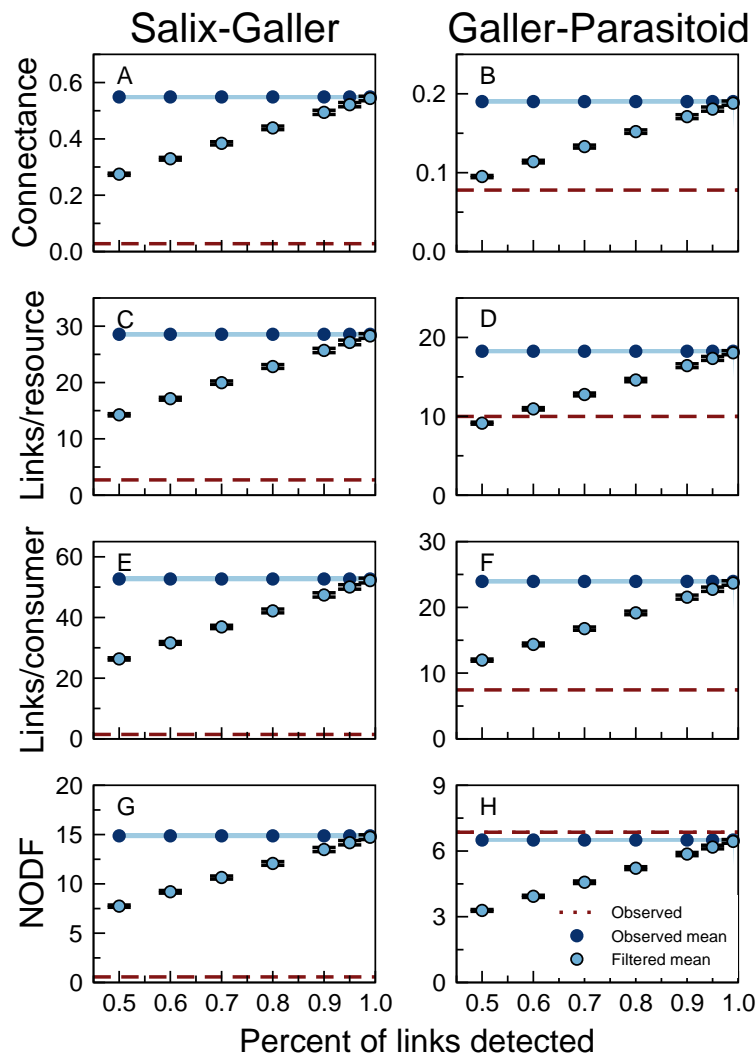
Figure S5: The number of samples required to achieve a given level of confidence that an interaction probability λ_{ij} is below a given threshold varies with both parameters. With a low threshold, our confidence that λ_{ij} is below the threshold increases rapidly with repeated observation of co-occurrence without interaction. Here we show the cumulative density functions for threshold probabilities of 0.5 (solid line), 0.25 (dashed line), 0.1 (dash-dot line), and 0.05 (dotted line) as well as the points at which the cdf reaches 0.90 (orange square), 0.95 (red circle), and 0.975 (blue diamond) for each threshold value. The large ticks along the x-axis indicate the number of samples associated with each of these points. Note that the number of samples required to reach any given threshold is larger for **A)** the *Salix*-galler network than for **B)** the galler-natural enemy network. In the galler-natural enemy network, the credible interval for a pair of species with no observed co-occurrences was 0.364. All pairs of species therefore have $\lambda<0.5$.

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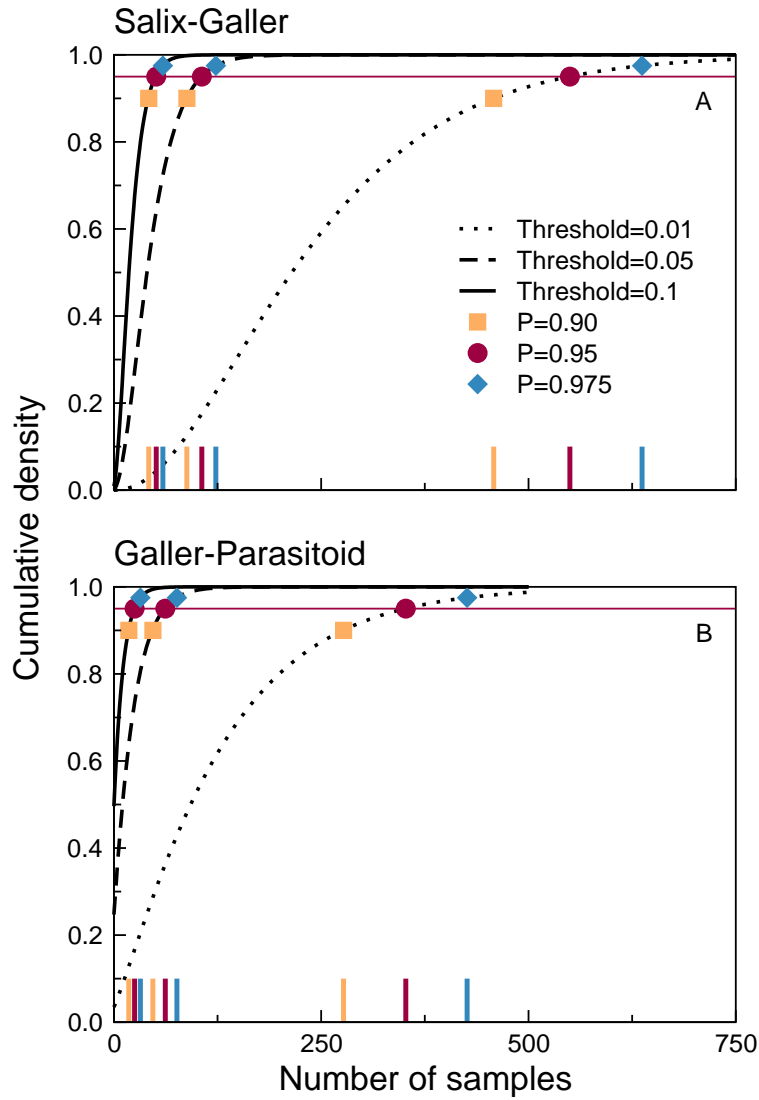
Considering the galler-natural enemy networks, the connectance, mean links per galler, and mean links per natural enemy were also much lower in the observed web (C=0.078, L_{galler}=9.99, and $L_{naturalenemy}$=7.45, respectively) than in the posterior webs ($0.183 \leq C \leq 0.196$, $17.5 \leq L_{galler} \leq 18.9$, and $23.0 \leq L_{naturalenemy} \leq 24.8$). When the detection probability was relatively low (i.e., 50%), however, the properties of randomised networks became similar to those in the observed webs (Fig. S6B,D,F). Nestedness was higher in the observed network (NODF=6.85) than in the posterior networks ($6.30 \leq NODF \leq 6.74$; Fig. S6H), but network structure was much more similar in the observed and posterior galler-natural enemy networks than in the *Salix*-galler networks. The Barbour *et al.* (2016) network likely provides a better prior here than for the *Salix*-galler networks, as both networks included different galler and natural enemy species. Although the two networks differ vastly in scale, and the inclusion of multiple *Salix* species likely causes structural differences from Barbour *et al.* (2016) despite the similar resolution of gallers and natural enemies, this result nevertheless suggests that even such a large and well-replicated network as that in Kopelke *et al.* (2017) is missing many interactions. This should be a strong warning to researchers comparing structural characteristics between networks - small structural differences are likely to be masked by substantial noise resulting from sampling uncertainty.

Figure S6: Here we show the mean connectance, links per resource (*Salix* in the *Salix*-galler networks and gallers in the galler-natural enemy networks), links per consumer, and nestedness (NODF) for networks assembled using posterior distributions based on a smaller, North American *Salix*-galler-natural enemy system (Barbour *et al.*, 2016; Barbour, 2016). We created 100 "posterior-sampling" networks and then, for each of these, created 100 "detection-filter" networks by randomly sampling 50%-99% of the interactions included in the posterior-sampling network. This simulates imperfect detection of interactions in the field. Each point represents the mean network property (e.g., connectance) obtained from a set of 100 detection-filter networks, plotted against the value of the network property in the posterior-sampling network used to create the detection-filter networks. For each property and both network types, the posterior-sampling networks cover a relatively small range of network properties than the range covered by networks with varying detection probabilities. The value of each property decreases with the proportion of links included in the detection-filter networks.

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Appendix S6: Obtaining prior interaction probabilities

Degree distributions and interaction probabilities

Obtaining degree distributions from the binary interaction network is straightforward. To do this, we simply divide the total number of observed interaction partners for each species (row or column sums) with the number of potential interaction partners (number of column or rows). Degree distributions calculated, we can then calculate the probability of each interaction as the product of the normalised degrees of the two species. For example, the probability of an interaction between two species with normalised degrees of 0.5 (each interacts with half of the available partners) is 0.25. R code used to do this follows:

 # Now get the degree distributions from the web
deg_dist_Salix=rowSums(prior_web)/ncol(prior_web)

 # Remove one genotype that never interacted
deg_dist_Salix<-deg_dist_Salix[which(rowSums(prior_web)>0)]
deg_dist_galler=colSums(prior_web)/nrow(prior_web)

 # Interaction probabilities are the product of plant and galler probabilities
int_probs=as.numeric(deg_dist_galler%*%t(deg_dist_Salix))

 # And for the galler-parasitoid web:
deg_dist_parasitoid=rowSums(prior_web_para_only)/ncol(prior_web_para_only)
deg_dist_parasitised=colSums(prior_web_para_only)/nrow(prior_web_para_only)
gp_int_probs=as.numeric(deg_dist_parasitised%*%t(deg_dist_parasitoid))

Appendix S7: R code for calculating posterior distributions and confidence intervals

The following simple functions are implemented in the R language. They are also provided as R code in a separate file. Priordata is the list of interaction frequencies from the prior data, n is the number of sites with observed co-occurrence of species i and j, and k is the number of sites with an observed interaction ij. When calculating the prior distribution, both n and k are 0. In the main text we assume 100% confidence in observed interactions and therefore consider only cases where k=0. Only n varies between species pairs.

To calculate the parameters α and β of a prior or posterior distribution:

```
calculate_parameters<-function(priordata,n,k){

    # Calculate prior parameters
    start=list(shape1=1,shape2=1)
    pars=fitdistr(x=priordata,'beta',start=start,lower=c(0,0))$estimate
    alpha=pars[[1]]
    beta=pars[[2]]

    # Update the parameters with data. If n=0 and k=0, no change.
    alpha_prime=alpha+k
    beta_prime=beta+n-k
    pars2=c(alpha_prime,beta_prime)
    return(pars2) }
```

All of the following functions use the parameters returned by "calculate_parameters". To calculate the maximum likelihood estimates of the mean and variance of the probability of interaction λ_{ij}:

```
calculate_distribution<-function(pars){

    # alpha and beta may be from a prior or posterior distribution
    alpha=pars[[1]]
    beta=pars[[2]]

    # Calculate the MLE of the mean
    mu_numerator=alpha
    mu_denominator=alpha+beta
    mu=mu_numerator/mu_denominator

    # Calculate the MLE of the variance
    sig_numerator=alphabeta
    den1=alpha+beta
    den2=den1**2
    sig_denominator=den1*den2
    sigma2=sig_numerator/sig_denominator
    return(c(mu,sigma2))}
```

To calculate a credible interval based on the prior or posterior distribution, for given lower and upper bounds:

credible_interval<-function(pars,p_lower,p_upper){

    alpha=pars[[1]]
    beta=pars[[2]]
    lowCI=qbeta(p=p_lower,shape1=alpha,shape2=beta)
    highCI=qbeta(p=p_upper,shape1=alpha,shape2=beta)
    return(c(lowCI,highCI))}

To calculate the number of samples required to reach a given level of confidence that the probability of interaction between two species (λ_{ij}) that have not been observed co-occurring is below a threshold value:

samples_for_threshold<-function(threshold,confidence,pars){

    alpha=pars[[1]]
    beta=pars[[2]]
    n=seq(0,100,1)
    k=0
    cdf=pbeta(threshold,shape1=alpha,shape2=beta+n)
    samples=length(which(cdf<confidence))
    return(samples) }

Table S1: Here we give the lower and upper bounds of 95% credible intervals for the probability of interaction λ between *Salix*-galler or galler-natural enemy pairs that have been observed co-occurring n times but have never been observed interacting.

n	*Salix*-galler		galler-natural enemy	
	Lower bound	Upper bound	Lower bound	Upper bound
0	0.013	0.049	5.39×10^{-4}	0.304
1	0.013	0.048	4.82×10^{-4}	0.276
2	0.013	0.048	4.35×10^{-4}	0.253
5	0.012	0.048	3.37×10^{-4}	0.203
10	0.012	0.047	2.45×10^{-4}	0.152
15	0.012	0.046	1.93×10^{-4}	0.121
20	0.012	0.046	1.59×10^{-4}	0.101
25	0.012	0.045	1.35×10^{-4}	0.087
50	0.011	0.042	7.72×10^{-5}	0.050
100	0.010	0.037	4.16×10^{-5}	0.027
150	0.009	0.033	2.84×10^{-5}	0.019
200	0.008	0.030	2.16×10^{-5}	0.014
374	0.006	0.022	1.18×10^{-5}	0.008

Appendix S8: Calculating the credible interval around a probability estimate

Here we describe the derivation of the Clopper-Pearson credible interval for the estimated probability of interaction λ of a pair of species observed co-occurring n times and interacting k times. As we are most interested in the probability of interaction between species pairs that have never been observed co-occurring, we consider only the case where $k = 0$ over a variety of n. This is straightforward to do in R (see the function "credible_interval" in *Appendix S7*).

First, we must obtain the α and β parameters for the prior distribution. In this study we obtained these parameters using the R (R Core Team, 2016) function fitdist from the package fitdistrplus (Delignette-Muller & Dutang, 2015). Once α and β are known, we can update them using our observed data. Specifically, we are interested in $\alpha' = \alpha + k$ and $\beta' = \beta + n - k$. These parameters can then be used to calculate a credible interval using the R (R Core Team, 2016) function qbeta. In the table below, we present the 95% credible intervals for *Salix*-galler and galler-natural enemy pairs with different numbers of observed co-occurrences (n) and no observed interactions ($k = 0$), calculated using prior information derived from the Zillis sub-network (Kopelke *et al.*, 2017).

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