## APPENDIX: STATISTICAL TOOLS

## I. Notes on random sampling

## Why do you need to sample randomly?

In order to measure some value on a population of organisms, you usually cannot measure all organisms, so you sample a subset of the population. In order for this subset to be representative of the population as a whole, it cannot be selected subjectively from the population. That is, there should not be any bias about which particular organisms are selected to measure. The way in which we avoid bias is by taking random samples. This means that each individual to be sampled is chosen randomly (or by chance) from the population. Most statistical tests that you use assume that you have sampled randomly.

Consider an example of how nonrandom sampling could affect your results. Suppose you are testing the hypothesis that plants growing in acidic soils have lower growth rates than those growing in neutral soils. You grow plants from seed, then transfer the seedlings to the two soil types. You subconsciously choose more vigorous seedlings to transfer to the neutral soil, and less vigorous seedlings (perhaps those with some herbivore damage) to transfer to the acidic soils. If you observe a difference in growth rates between these two groups, you cannot be sure that it was due to soil differences. If you had randomly chosen which seedling would be transplanted to the two soil types, you could be more certain that any difference in growth was due to the soil.

## Ways to sample randomly

Random sampling can be difficult in field biology. In many other situations, you can assign the individuals numbers, then use a random numbers table to decide which treatment each individual would receive. You sometimes must be creative in devising ways to sample randomly, or at least avoid bias.

Example:
If you need to randomly sample a leaf or branch from a tree, stand with your back to the tree and reach back, sampling the first leaf or branch you happen to touch. Another option is to pick a branch, then choose which leaf you will sample by looking up a number in a random numbers table (close your eyes and put your finger someplace on the page). If the random number is 4 , choose the fourth leaf from the branch tip. You can experiment with other methods. The important thing is to not look at the leaves, and don't choose one because it has few or many galls.

## II. Random Numbers Table

Enter the table at random (e.g., point to it with a pencil). Proceed horizontally or vertically from that point to obtain as many digits or numbers as needed.
$\begin{array}{llllllllll}16363 & 27995 & 41125 & 66592 & 12860 & 51577 & 71692 & 16628 & 67324 & 83419 \\ 39240 & 07412\end{array}$ 661213086334081046077550204707686534333887116464803604174787 965755382641704151945627949971426497017595444767613581655837 521669636747481188624562393595106501205338724789572988686154 910286511212180467553295596324795166343210341942623917969020 $2770928594258398763639442802487499619481 \quad 66351063779782437041$ 911368588224857512991083873647616682777837789531665845441833 711886682904281272067033117899518757725386078122349239102238 867627113159628803005761493257559468978918238252527065337393 833397743638773500812922923649154263150425341746101902291666 249474462342554587053582202055947239609070086312842436119557 281864164292387393819973720272473055876872531467041436165260 328844928367787941165216138414843195097801803199695656488407 794662466741425916166537286449246685445995823126101326622570 117755277819652069920151388362987852711661859539333186123445 662487999813670642901523154748293549488873377746696974684505 800393936959470267164646252922789111995393147137921276135869 993664321390418613371426056876749926772291075911371135464755 339139611224338952940121602949665867466194809147627889475333 $4412959181432333312437576471102072273824798774651675488 \quad 82273$ 044788492968978226939288576772944072272981553461291180620192 388216481447522869154919528851713068420794648170143883492108 974430197361484626154174199773121101593109120338748473808786 857049825129175767849432118842228173549666678758917608697310 424800765540183252560952284493636827272487341577731578268323 648586901031483377404614129821533136264133150504174368687116 961769974695971496228259035237217896776612829145554126215937 284383728768455184339441979448717422503109557796277877651468 286082911612344460253645175226924975871221333692433015738766 889594817224219219865974996376543143451111377159361401579795 716084558548448861564458634124681229551651596613928638513587 543997142835942935945256575225802339217635457717376223951060 597557246586327397164520836755757727479921169311432275478596 179611669869213933888981358122188033903997264246199737538838 162989667157299160616974056814633243536354903909796079283056 449461726917264628222635024863536627236772757162944421073288

## III. Tests of Independence using the G-statistic:

A two-way test of independence is commonly used in ecology for situations in which we want to test whether two different characteristics or conditions occur independently of one another. (An alternative, the Chi Square test, is more commonly used, but G is quite simple to calculate and more closely follows an actual chi-square distribution. An advantage of the chi square test is that students must calculate expected frequencies, so they see for each cell in the table how closely the observed value matches the expected.)

The calculations for a 2-way test of independence follow:

1) $a=\Sigma(f \ln f)$ for cell frequencies
2) $b=\Sigma(f \ln f)$ for row and column totals
3) $c=n \ln n$
4) $G=2(a-b+c)$
5) Compare $G$ with the critical value of $X^{2}$. In a $2 \times 2$ table, there is $(2-1)(2-1)=1$ degree of freedom. Use $\alpha=0.05$. If $\mathrm{G}<$ critical value, accept $\mathrm{H}_{0}$; if $\mathrm{G} \geq$ critical value, reject $\mathrm{H}_{0}$.

## Example:

An ecologist wishes to know whether leaves that have sawfly galls are more susceptible to herbivory by leaf-chewing insects than are leaves without galls. The ecologist recorded presence or absence of leaf chewing damage on 50 leaves with galls and 50 leaves without galls. Here are the results (hypothetical data):

| Galls on leaf | chewing damage on leaf |  |  |
| :--- | :---: | :---: | :---: |
|  | yes | no | Total |
| yes | 31 | 19 | 50 |
| no | 22 | 28 | 50 |
| Total | 53 | 47 | $\mathrm{n}=100$ |

To calculate G :

1) $\mathrm{a}=31 \ln 31+22 \ln 22+19 \ln 19+28 \ln 28=323.7$
2) $\mathrm{b}=50 \ln 50+50 \ln 50+53 \ln 53+47 \ln 47=782.6$
3) $\mathrm{c}=100 \ln 100=460.5$
4) $\mathrm{G}=2(323.7-782.6+460.5)=3.24$
5) In the table of critical values of the chi square distribution, the critical value of G for 1 d.f. and $\alpha=0.05$ is 3.841 .

The ecologist accepts the null hypothesis that chewing damage occurs independently of gall presence on willow leaves at this site and concludes that the observed differences were small enough to have resulted from chance.

## IV. Testing a Poisson Distribution

For ecological events that occur relatively rarely and independently of other events of the same type, the frequency of events should follow a Poisson distribution. For example, adult female sawflies may oviposit on only a small fraction of the leaves of a willow tree. We could test whether sawfly galls occur independently of whether there are other galls on the same leaf by comparing the frequency distribution of galls to a Poisson distribution.
$\mathrm{H}_{0}$ : The number of galls per leaf on willows follows a Poisson distribution.
$H_{a}$ : the number of galls per leaf on willows does not follow a Poisson distribution.
General test procedure: Compare observed frequency data with expected frequencies (Sokal, R. R., and F. J. Rohlf. 1981. Biometry. W.H. Freeman, San Francisco, section 5.3)

|  | Frequency (\# trials) |  |  |
| :---: | :---: | :---: | :---: |
| \#events/trial | observed (f) | expected ( $\hat{\mathrm{f}}$ ) | deviation from expected ( $\mathrm{f}-\hat{\mathrm{f}}$ ) |
| 0 | $\mathrm{f}_{0}$ | $\frac{n}{e^{\bar{X}}}$ |  |
| 1 | $\mathrm{f}_{1}$ | $\frac{n}{e^{\bar{X}}}(\bar{X})$ |  |
| 2 | $\mathrm{f}_{2}$ | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{2}}{2}$ |  |
| 3 | $\mathrm{f}_{3}$ | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{3}}{6}$ |  |
| 4 | $\mathrm{f}_{4}$ | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{4}}{24}$ |  |
| 5 | $\mathrm{f}_{5}$ | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{\text {s }}}{120}$ |  |
| 6 | $\mathrm{f}_{6}$ | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{6}}{720}$ |  |
| 7+ | $\mathrm{f}_{7}$ | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{7}}{5040}$ |  |
| total | $\mathrm{N}=\Sigma \mathrm{f}=$ |  |  |

N is the sample size (number of leaves sampled), $\bar{X}$ (average number of galls per leaf) = total number of galls/total number of leaves. For calculations of expected frequencies, see example data, below.

Example (from real class data, 1998): From a sample of 201 leaves, we found 140 galls. So $\bar{X}=0.70$.

|  | Frequency (\# trials) |  |  |
| :---: | :---: | :---: | :---: |
| \# galls/leaf | observed (f) <br> (= \# leaves) | expected ( $\hat{\mathrm{f}}$ ) | deviation from expected ( $\mathrm{f}-\hat{\mathrm{f}}$ ) |
| 0 | 145 | $\frac{n}{e^{\bar{X}}}=\frac{201}{e^{0.7}}=99.8$ | + |
| 1 | 22 | $\frac{n}{e^{\bar{X}}}(\bar{X})=\frac{201}{e^{0.7}}(0.7)=69.9$ | - |
| 2 | 12 | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{2}}{2}=24.5$ | - |
| 3 | 8 | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{3}}{6}=5.7$ | + |
| 4 | 7 | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{4}}{24}=1.0$ | + |
| 5 | 4 | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{5}}{120}=0.1$ | + |
| 6 | 0 | $\frac{n}{e^{\bar{X}}} \cdot \frac{\bar{X}^{6}}{720}=0$ |  |
| 7+ | 3 | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{7}}{5040}=0$ | + |
| Total | $\mathrm{n}=\Sigma \mathrm{f}=201$ |  |  |

Note that it is best to lump classes with expected frequencies less than 5 . In this example, we would combine leaves with 3 or more galls, giving the table on the following page:

|  | Frequency (\# trials) |  |  |
| :---: | :---: | :---: | :---: |
| \# galls/leaf | observed (f) <br> (= \# leaves) | expected (f) | deviation from <br> expected (f- $\hat{\mathrm{f}})$ |
| 0 | 145 | $\frac{n}{e^{\bar{X}}}=\frac{201}{e^{0.7}}=99.8$ | + |
| 1 | 22 | $\frac{n}{e^{\bar{X}}}(\bar{X})=\frac{201}{e^{0.7}}(0.7)=69.9$ | - |
| 2 | 12 | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{2}}{2}=24.5$ | - |
| $3+$ | 22 | $\frac{n}{e^{\bar{X}}} \bullet \frac{\bar{X}^{3}}{6}=5.7$ | + |
| Total | $\mathrm{n}=\Sigma \mathrm{f}=201$ |  |  |

The data do not fit the Poisson distribution very well. Since the number of leaves with just one gall is much smaller than expected (and the number with 3 or more galls greater), we can conclude that female sawflies do not avoid ovipositing on leaves that already have 1 gall.

