Note: formulas or values can be propagated through an excel column by selecting a cell(or cells), clicking the square in the bottom right-hand corner of the cell, and dragging down. This will also propagate simple patterns (e.g., a repeating list of dates). If you try to copy a single value this way, sometimes it will try to make up a pattern (e.g., take a single date and follow it with the next date); if this happens, it might just need more data (e.g., select two cells with the same date instead of one).

Protocol for randomizing temperature treatment days:

1. The problem: Clustering similar temperatures/repeatedly having high or low temperatures early or late in the order 🡪 families or ages not evenly distributed across treatments.
2. Trying (for each 5-day set): randomize 1st day. If it’s low (12 or 20), randomly select 2nd day from high (28, 36, 40) and vice versa (28 included in both groups). Sample 3rd day from the remaining numbers. If it’s high, select a low, if it’s low, select a high temp. Whatever is left is day 5.
3. So families are always split between high/low treatments (for Option 1) and high/low temps aren’t as likely to be clustered (for Option 2).

Datasheet protocol for split into two cohorts:

1. Create datasheet with gen1\_ID, MID, wing\_date, and blank columns for test\_date, test\_temp, and test\_age.

\*\*Make sure the date columns are in date Format, and the other columns are in General!

\*\*Excel does not understand dates with a dot format, so dot dates should be converted into / dates in a new column using the command =SUBSTITUTE(AA, “.”,”/”), where AA is the original date column. Use these new dates for wing\_date. To copy/paste without bringing the formula, right click and select “Paste Special”🡪”Values”

2. Add a randomizer column with =rand()

3. Sort the data by wing\_date, then MID, then randomizer\*\*Addendum: add a cohort column and sort by that instead of wing\_date

4. Assign members of each family to the next two test dates in their now randomized order, with the first half going to the earlier of the two dates. If families have an odd number of members, flip a coin for the middle bug (randomizer<0.5=earlier date; randomizer>0.5=later date).

5. Once all test\_dates have been entered, re-sort the data by test date and fill in the test\_temp column

6. Set the test\_age column to subtract the wing\_date column from the test\_date column. Make sure the test\_age column remains in General format (not date)

7. Run through R script to visualize relationships between age, family, and test temperature

Datasheet protocol for grouping cohorts and dividing among treatments:

1. Create a sheet as in steps 1-2 above.

2. Add a “8-day interval” column that covers wing\_dates in each 8-day interval (because this is how long it takes to run 5 trials with an off-day each week)

3. Sort data by 8-day interval, then MID, then randomizer.

4. Individuals are now randomly ordered within family. Assign individuals test dates in order 1st date, 2nd date, 3rd date, 4th date, 5th date, repeating. If the number of individuals in a family is not divisibly by 5, assign the remainder randomly (randomizer<0.2=1st date, 0.2<randomizer<0.4=2nd date, 0.4<randomizer<0.6=3rd date, 0.6<randomizer<0.8=4th date, randomizer>0.8=5th date)

5. Once all test dates have been entered, sort by test date and fill in the test\_temp column.