Assessment Questions

As just discussed, Harmful Algal Blooms (HABs) are a concern in various ocean regions. This activity is based on results from research conducted in the Northeast Atlantic during 2014 and 2017 on a HAB group called *Pseudo-nitzschia*. Some species in this group produce a potent neurotoxin called Domoic Acid. You will be figuring out what species were present by using the DNA extracted from *Pseudo-nitzschia* diatoms present in water samples. You will then be calculating the percent abundance of six species in each sample, representing those data as pie graphs, and then comparing those results between years (2014 vs. 2017) and from month to month. The thought questions are designed to reinforce your ability to see patterns over time and to speculate about which species are toxic (based on comparisons with Domoic Acid levels measured at the time of sampling in 2017). This toxin is of concern for human health as it causes an illness known as amnesic shellfish poisoning, and also sickens wildlife.

The teacher will break you up into groups. There are 8 dates for which you must calculate the percent (%) contribution of each species. Perhaps each student could complete two dates.

1. Use the information in the following table to identify the species found in the "trace" files for your assigned date/s on page 3. There can be multiple species for any given date. Once you identify the species present in each sample, then you will calculate the proportional abundance (percent contribution) of each species and add it to the Exercise 1 sheet on page 4 and color in the pies.

   Example calculation: The DNA fragment size (base pairs) is oriented on the x axis and the proportional abundance on the y axis. If there is only one peak on the trace file, then there was only one species detected. If, for example, the peak was at 200 base pairs on the x axis, then using the table below, you can determine that this peak represents the species *Pseudo-nitzschia seriata*. You would then use the peak height to determine the proportional abundance of this species (ranging between 0 and 1). In this example, the peak height is 1. If you multiply by 100 to obtain proportional abundance expressed as percent total, then there is a 100% occurrence of that species. Fill in your assigned dates from 2017 and then color in the corresponding pie using the assigned color system.

Species	DNA Fragment Size (base pair)
Pseudo-nitzschia pungens	142
Pseudo-nitzschia australis	150
Pseudo-nitzschia seriata	200
Pseudo-nitzschia delicatissima	168
Pseudo-nitzschia seriata	200
Pseudo-nitzschia plurisecta	226

2. After you finish your pies, cut them out, put the date on the back, and tape them onto the graph below (Exercise 2; page 5). When was the toxin (Domoic Acid) level the highest?
   ___September 2017___

3. **The teacher will project the completed graph with all the pies provided (Answer Key page 9).** Which species may be responsible for the high levels of Domoic Acid, which can be harmful to humans as well as wildlife? You can abbreviate the first word (the genus) by writing "P." Note that you should underline a genus and species to denote its Latin origin when writing by hand:

   P. australis

4. Are there any other species that might contain Domoic Acid? Which ones? What evidence do you have for this?
   Yes, *P. seri* because the Domoic Acid levels were slightly elevated when they were present.

Now let's take a closer look at any similarities over time. We refer to these as temporal changes. Using Exercise 3, complete the pies for 2014 on page 7 using the data in the provided table on page 6. Once those are completed, you will be comparing them to the pies you created for 2017 and answering the following questions:

1. What similarities or differences do you see within the same month (early vs. late) but within the same year?
   These responses vary. Sometimes the exact same species are present, but in different proportions. Sometimes there are more species for the 2014 date than for 2017 (or vice versa).

2. What similarities or differences do you see across seasons (summer vs. fall) both within and between years?
   These responses vary. There were species that tended to be found early in the year (summer), like *P. deli*. (in both years). *P. pungens* tended to be found later in the year (late summer and fall) in both years. It was often the dominant species when present in 2014 (late summer through fall). *P. aust* was only found in late summer and fall of 2017. *P. pluri* was found only once (fall) in 2014 but then mostly in spring in 2017, with the exception of early October. In both years, there were usually only two species present. These were usually *P. pungens* and *P. seri* in 2014, but *P. pungens* and *P. aust* in 2017.

3. What similarities or differences do you see between years?
   These responses vary. There were species that tended to be found early in the year, like *P. deli*. (in both years). *P. pungens* tended to be found later in the year (in both years). It was often the dominant species when present in 2014. It was never the dominant species in 2017, although it was tied with *P. aust* at 50% in early September 2017. *P. aust* was only found in 2017. *P. pluri* was found once once (late October) in 2014 and mostly in early 2017, with the exception of early October. In both years, there were usually only two species present. These were usually *P. pungens* and *P. seri* in 2014, but *P. pungens* and *P. aust* in 2017. There were always at least two species in 2017. The only times there was only one species present was in 2014 (summer).

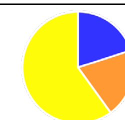
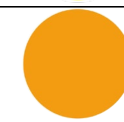
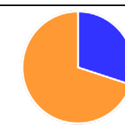
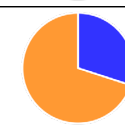
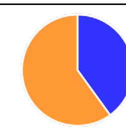
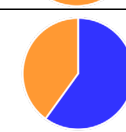
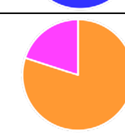
4. What changes in the natural environment could be responsible for these differences (seasonal and annual)?
   There could be changes in the physical environment such as with temperature. There could also be biological factors that differed like the numbers and types of predators or competitors. Diatoms need light to photosynthesize and that could vary seasonally and annually, as could temperature.

5. Why do you think *P. australis* is present in the region in 2017 but not 2014?

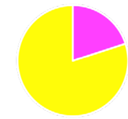
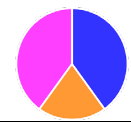
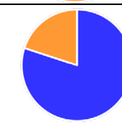
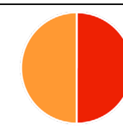
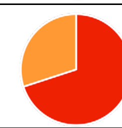
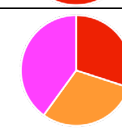
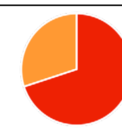
The ultimate source of *P. australis* is not known; however, scientists hypothesize that this species originated from the Scotian Shelf, and was transported to the region by ocean currents.

Exercise 1 – Answer Key

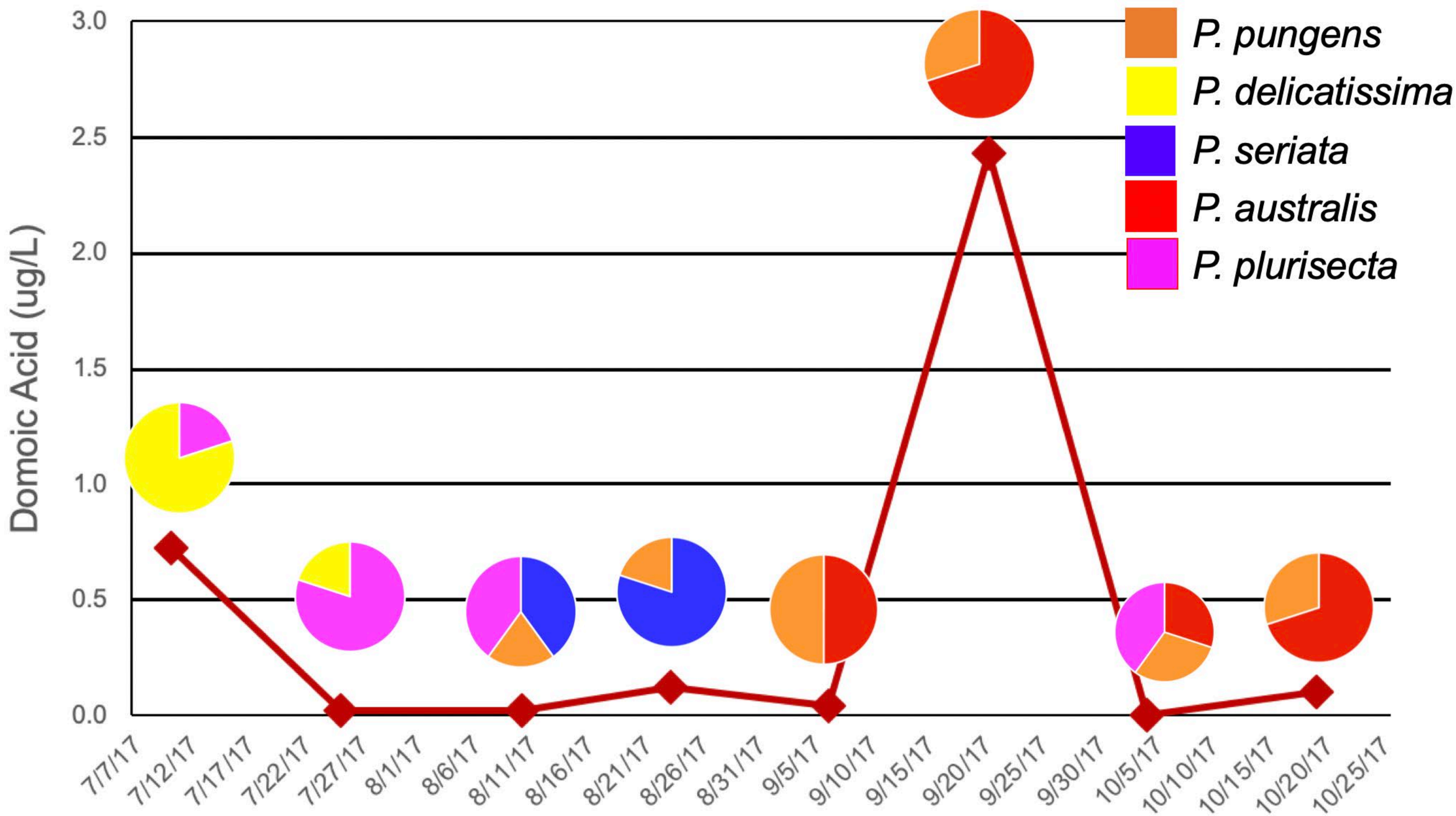
Community composition in 2014

Date	*P. pungens*	*P. delicatissima*	*P. seriata*	*P. australis*	*P. plurisecta*	
	Orange	**Yellow**	**Blue**	**Red**	**Pink**	
7/7/14	0	1	0	0	0	
7/18/14	0.2	0.6	0.2	0	0	
8/12/14	1	0	0	0	0	
8/27/14	0.7	0	0.3	0	0	
9/9/14	0.7	0	0.3	0	0	
9/22/14	0.6	0	0.4	0	0	
10/3/14	0.4	0	0.6	0	0	
10/30/14	0.8	0	0	0	0.2	

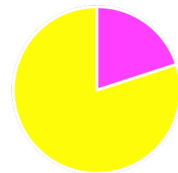
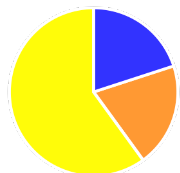
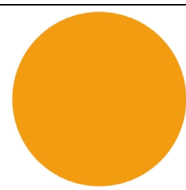
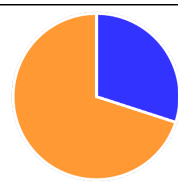
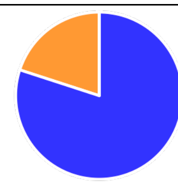
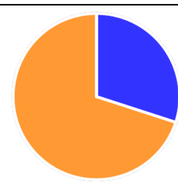
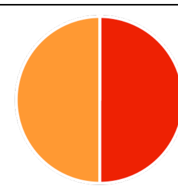
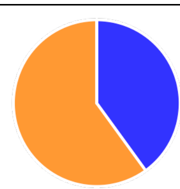
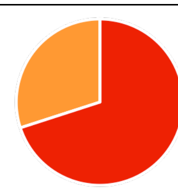
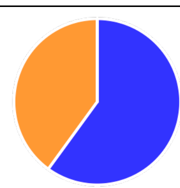
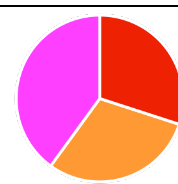
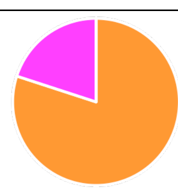
Community composition in 2017

Date	*P. pungens*	*P. delicatissima*	*P. seriata*	*P. australis*	*P. plurisecta*	
	Orange	**Yellow**	**Blue**	**Red**	**Pink**	
7/10/17	0	0.8	0	0	0.2	
7/25/17	0	0.2	0	0	0.8	
8/10/17	0.2	0	0.4	0	0.4	
8/23/17	0.2	0	0.8	0	0	
9/6/17	0.5	0	0	0.5	0	
9/20/17	0.3	0	0	0.7	0	
10/4/17	0.3	0	0	0.3	4	
10/19/17	0.3	0	0	0.7	0	

Exercise 2 – Answer Key



Exercise 3 - Answer key

Time period	2014		2017	
Early July	**7/7/14**		**7/10/17**	
Late July	**7/18/14**		**7/25/17**	
Early Aug	**8/12/14**		**8/10/17**	
Late Aug	**8/27/14**		**8/23/17**	
Early Sept	**9/9/14**		**9/6/17**	
Late Sept	**9/22/14**		**9/20/17**	
Early Oct	**10/3/14**		**10/4/17**	
Late Oct	**10/30/14**		**10/19/17**