Managing *Listeria* Risk in Fresh Produce Using Predictive Models
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Rationale and Objectives

Many foods are perishable and require time/temperature control throughout their shelf life. In many cases this control is required to ensure quality, but in an uncertain number of situations, it may also be required for food safety (to control foodborne pathogen growth).

This issue is often discussed by fresh produce producers and buyers and is now exacerbated by several federal regulations and policies including the Preventive Controls Rule and Sanitary Transportation Rule.

There is an urgent need for short-term science-based parameters on this topic. This project focuses on the pathogen most likely to grow at the temperature range of interest (*Listeria monocytogenes*) and will use "off-the-shelf" computer models in the form of ComBase Predictor (https://browser.combase.cc/ComBase_Predictor.aspx?model=1.

This progress report has a variety of predictions, comparing relative risk of *Listeria monocytogenes* growth for different conditions to guide science-based risk management decisions.

This preliminary work will not consider the effect of spoilage organisms or competitive microflora. This work will also not explicitly consider whole versus cut produce, although the availability of moisture and nutrients are known to affect bacterial growth rates.

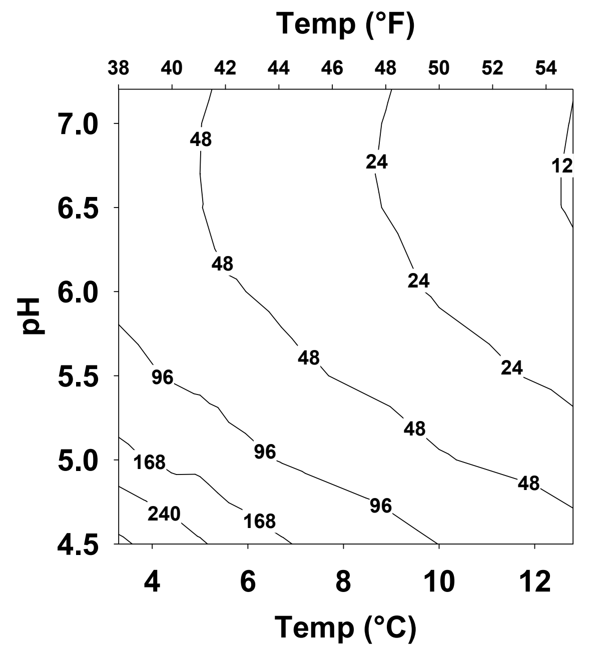
This work assumes that the predictions from ComBase are correct. Since these predictions are based on growth of microorganisms in microbiological growth media without any competitive microflora, such predictions are generally conservative (i.e. organisms grow more slowly in the real world). The model prediction shown below assume that the organism experiences no lag phase, and a very high (0.997) water activity, and that there is no upper population limit, which are all conservative (failsafe) biases.

These assumptions will make the models *highly* conservative, and thus quite robust and able to withstand scrutiny from regulatory agencies or over-zealous inspectors. It is quite likely that actual pathogen growth on specific produce commodities are much less than the model predictions, and in some cases no growth or slow decline would actually be observed in the real world.

Relative impact of parameters of interest

The figure below shows the relative importance of assumed pH and temperature on the time required in hours for a one longer than increase in the concentration of *L. monocytogenes*.

It is clear from this figure that both pH and temperature are important. Although the pH of fresh produce can vary from product to product, from cultivar to cultivar, and even from item to item, the table below may be useful in conceptualizing these differences.



Produce	pH
Apple, low	4.5
Apple, high	5.0
Cabbage	5.5
Romaine	6.0
Honeydew	6.7
Avocado	7.2

Assessing risk by relative equivalence

One way to determine the relative risk of holding food out of temperature control would be to look at currently allowed practices, determine the risk of those practices, and then look for equivalent risk for other time temperature conditions.

The table below assumes a pH of 6, and as noted above a very high water activity (0.997). For purposes of discussion, we can assume that this example relates to romaine lettuce. I know from conversations with the project steering committee that a UC Davis shelf life study concluded that the shelf life of romaine lettuce was approximately 21 days. I also know from those conversations that it is typical to label the product with a 17-day shelf life. We also know that when this product is harvested it is not cooled immediately.

We know the current best practices and food code requirements mean that fresh produce should be stored below 41°F. The example below considers product stored at 38 and 40°F in the first two columns, and a variety of other storage temperatures in the columns to the right. Note that the typical shelf life (17 days), as well as the UC Davis shelf life (21 days) are shown in bold. If we use the model to predict the relative increase in *L. monocytogenes* for the scenarios, the relative increases are shown in the text in green yellow and red boxes. If we make the assumption that 17 days at 40°F represents "acceptable" conditions, and the same temperature at 21 days represents "borderline" conditions, we can generate equivalent relative increases in the concentration of *L. monocytogenes* for other time temperature scenarios. These are shown by the color of the boxes in the table below. Note that this table is available as an Excel spreadsheet, and will automatically change color with the input of different values for "green light", and "yellow light".

	Temp (°F)	38.0	40.0	41.0	42.0	44.0	45.0	50.0	55.0
	Temp (°C)	3.3	4.4	5.0	5.6	6.7	7.2	10.0	12.8
Time (d)	Time (h)								
0	0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
1	24	0.3	0.4	0.4	0.5	0.6	0.7	1.1	1.6
2	48	0.5	0.7	0.8	0.9	1.2	1.4	2.2	3.3
3	72	0.8	1.1	1.2	1.4	1.8	2.0	3.3	4.9
4	96	1.0	1.4	1.7	1.9	2.4	2.7	4.4	6.6
5	120	1.3	1.8	2.1	2.4	3.0	3.4	5.5	8.2
6	144	1.5	2.1	2.5	2.8	3.6	4.1	6.6	
7	168	1.8	2.5	2.9	3.3	4.3	4.8	7.8	
8	192	2.1	2.9	3.3	3.8	4.9	5.4		
9	216	2.3	3.2	3.7	4.3	5.5	6.1		
10	240	2.6	3.6	4.1	4.7	6.1	6.8		
11	264	2.8	3.9	4.6	5.2	6.7	7.5		
12	288	3.1	4.3	5.0	5.7	7.3	8.2		
13	312	3.3	4.7	5.4	6.2	7.9			
14	336	3.6	5.0	5.8	6.6				
15	360	3.9	5.4	6.2	7.1				
16	384	4.1	5.7	6.6	7.6				
17	408	4.4	6.1	7.0					
18	432	4.6	6.4	7.5					
19	456	4.9	6.8	7.9					
20	480	5.1	7.2						
21	504	5.4	7.5						

17 days	Green light	6.1
21 days	Yellow light	7.5

Relative risk for actual data

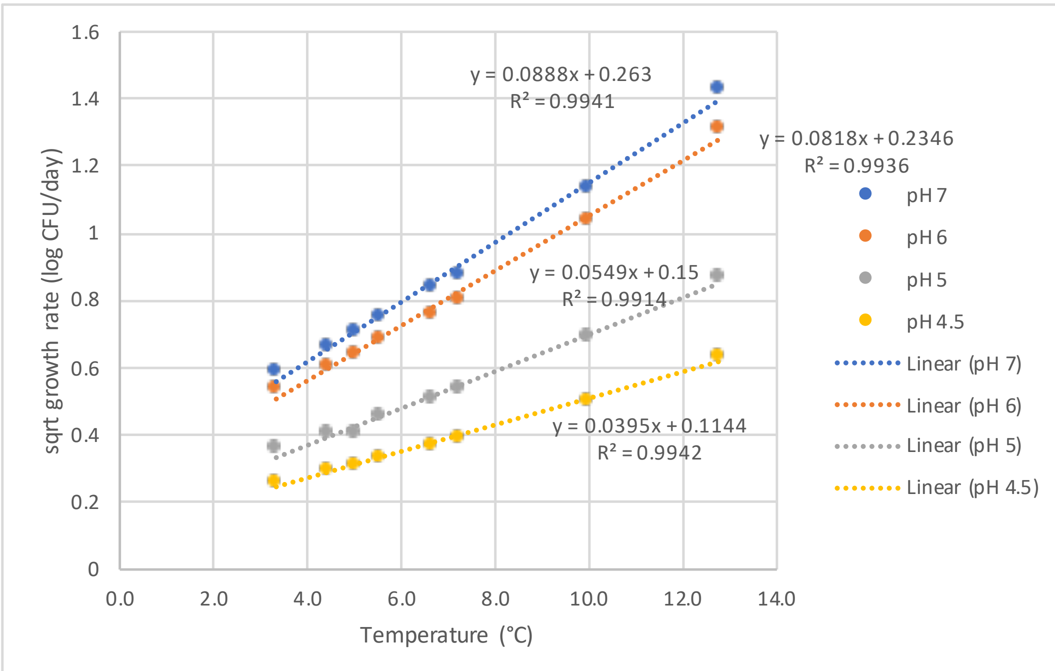
A third analysis was conducted for the March 2019 progress report using actual time and temperature data from 12 different shipments sent in the summer of 2018. As with the analysis above, a pH of 6.0 was assumed, and dynamic temperature modeling predicted the relative increase in *L. monocytogenes* over the course of the shipment. The details of that analysis are not included, but many of the “over 41” runs show less than a 1 log predicted increase in *L. monocytogenes*. As the runs get longer and as temperatures increases the *L. monocytogenes* predictions increase too.

Relative risk prediction tool

Because Listeria can grow at "proper" refrigeration temperatures, this presents the industry with an opportunity to compare the risk of product held out of temperature control with product that is been held in temperature control for different periods of time.

Data from the ComBase modeling program was extracted to create different models for the effective temperature on growth rate at four different pH values: 7, 6, 5, and 4.5.

A plot of these data with regression curves shown below.



Data from the regression analysis was used to create a simple spreadsheet that would allow someone to assume a specific time and temperature that a given product was held out of temperature control. The spreadsheet then calculates the equivalent amount of time that would pose the same increase in risk (due to increase in the concentration of listeria), for the four different pH values and three different "proper" temperature values: 38, 40, and 41 °F.

This spreadsheet could be a useful tool in defending against minor temperature fluctuations on products. For example let's assume that a given product was held for 4 hours at 55°F. If this was questioned by the regulatory authority, the spreadsheet shows that this temperature deviation (assuming a pH of 6) is equivalent to 25.5 hours at 38 °F, 18.3 hours at 40 °F or 15.8 hours at 41 °F all of which would currently be allowed by the regulations. A screenshot showing this particular scenario is shown below, but any possible scenario can be calculated by the spreadsheet.

Inputs			
What was the undesirable temperature the product experienced?	55 °F		(only 33.8 °F to 55 °F)
How many hours was the product at that temperature?	4 hours		
Outputs			
pH 7			
This is eqivalent to a Listeria risk from storing product at:			
	25.0 hours	at	38 °F
	18.1 hours	at	40 °F
	15.6 hours	at	41 °F
pH 6			
This is eqivalent to a Listeria risk from storing product at:			
	25.5 hours	at	38 °F
	18.3 hours	at	40 °F
	15.8 hours	at	41 °F
pH 5			
This is eqivalent to a Listeria risk from storing product at:			
	26.2 hours	at	38 °F
	18.7 hours	at	40 °F
	16.1 hours	at	41 °F
pH 4.5			
This is eqivalent to a Listeria risk from storing product at:			
	25.3 hours	at	38 °F
	18.2 hours	at	40 °F
	15.8 hours	at	41 °F

Further Research

Although this project is completed with the submission of this final report, additional research is ongoing in collaboration with a separately funded CPS project with Laura Strawn at Virginia Tech. We are planning to poster/technical presentations at the IAFP meeting this summer covering both data collected as part of a literature review, as well as data collected in the strong lab. Although not part of this report, those data validate that the models proposed here are failsafe (i.e. conservative or airing in the direction of food safety). Additional research on going in the Schaffner lab (not funded by CPS) will further validate these models.