

# #1 Using models to control pathogen growth

Foods can be formulated to reduce the growth of potential pathogens. In certain cases, a preferred formulation is one that limits pathogen growth to less than 1 log.

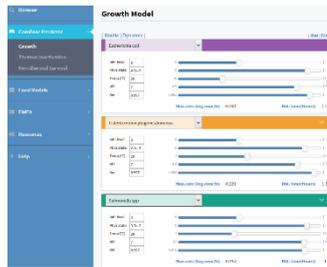
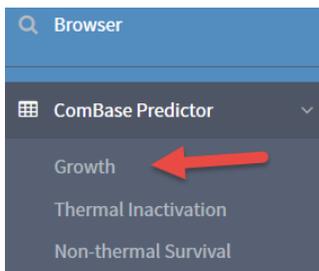
In this case study, you will use ComBase growth models to design a food environment that results in less than 1 log growth of *E. coli*, *Listeria*, and *Salmonella*.

## Intended Learning Objectives

- Understanding features of the ComBase Predictor growth models
- Knowledge about how environment conditions influence pathogen growth
- Awareness of how different pathogens respond to a similar set of conditions

## Instructions

1. Using the ComBase Predictor, select **Growth** models. Next, add *E. coli*, *Listeria*, and *Salmonella* models to the interface.



2. Produce **one** set of environmental conditions that will result in less than 1 log growth of **all three** pathogens at 10°C, over a time period of 72 hours.
3. In the above task, each of the models includes a lag phase. For a fail-safe growth estimate, set the value of the physiological state for each model so that there is **no lag phase**, and then again produce **one** set of environmental conditions that will result in less than 1 log growth of **all three** pathogens at 10°C, over a time period of 72 hours.
4. Questions
  - a. What was the net growth of each pathogen for steps 3 and 4, above?
  - b. Which environmental factor (pH or water activity) had the greatest effect on inhibiting pathogen growth?

## #2 Assessing pathogen growth in different food formulations

*Salmonella* spp. can potentially contaminate and grow in infant formula, especially when the formula is stored at elevated temperature. The ComBase database contains records for the growth of *Salmonella* spp. in infant formula that is hydrated with water, milk, or apple juice.

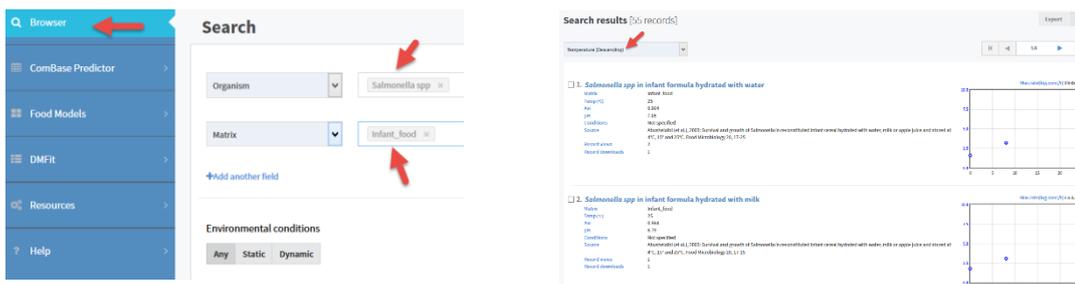
In this case study, you will retrieve data records for *Salmonella* spp. in infant formula, calculate growth rates, and then compare rates among the three different formulations.

### Intended Learning Objectives

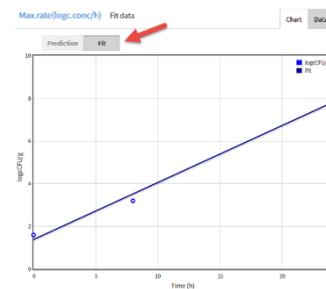
- Understanding features of the ComBase Browser (database)
- Knowledge about how to retrieve data records
- Skills in fitting growth models to data
- Knowing how different formulations affect growth rates

### Instructions

1. Using ComBase Browser, produce search criteria for *Salmonella* spp. and for Infant Food. Next, search for these criteria and then sort the records by Descending Temperature.



2. For each record showing 25°C, record the pH and then fit a model to the curve using the tool located above the individual record. Repeat this for each hydrating liquid (i.e. water, milk, and apple juice).



3. Record each growth rate.

Linear Model [fit]	
R-square:	0.985
SE of Fit:	0.401
Initial value	1.386 ± 0.339
Max Rate	0.267 ± 0.0232

4. Questions

- a. What is the average growth rate for *Salmonella* spp. in each formula?
- b. What environmental factor appears to have the greatest effect on *Salmonella* spp. growth?

### #3 Predictive models to manage pathogen growth in cooled meats

*Clostridium perfringens* is a spore-forming bacterium that can cause foodborne illness, especially in meats that are cooked and then improperly chilled. This typically occurs when whole cuts of meats (primals) are cooked, in which spores survive, and then vegetative cells grow when the cooked meat is improperly cooled.

In this case study you will design temperature cooling profiles that produce less than 1 log *C. perfringens* growth, and also unsafe growth (> 1 log).

#### Intended Learning Objectives

- Understanding features of the ComBase Perfringens Predictor
- Knowledge of how cooling profiles influence outgrowth of *C. perfringens*
- Awareness about how cured versus uncured meat, pH, and water activity affect *C. perfringens* growth

#### Instructions

1. Using the ComBase Food Models, select the **Perfringens Predictor**. Read the Help section by clicking the '?' symbol in the upper right corner.



2. Enter a pH and NaCl/Aw value.

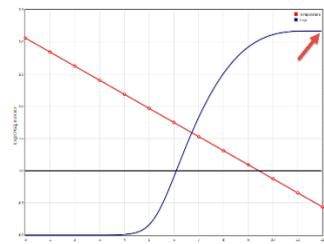
pH [5.2-8.0]

NaCl (%) [0-4]  [Aw | NaCl]

Time(h)	Temp (°C)
0.00	70.00
1.00	65.00
2.00	60.00
3.00	55.00
4.00	50.00
5.00	45.00
6.00	40.00
7.00	35.00
8.00	30.00
9.00	25.00
10.00	20.00
11.00	15.00

3. Enter a time-temperature profile consisting of at least **12 data points** that results in **less than 1 log *C. perfringens* growth**.

4. Click **Predict** and record the net predicted *C. perfringens* growth. Do this for Uncured and Cured meat, as well as different pH and Aw values.



5. Repeat steps 3-5 for a time-temperature profile consisting of at least **12 data points** that results in **more than 1 log *C. perfringens* growth**.

6. Questions

- a. How is *C. perfringens* growth different for cured and uncured meat?
- b. What is the effect of pH?

- c. What is the effect of  $A_w$ /NaCl?

## #4 Using predictive models to estimate food shelf-life

*Listeria monocytogenes* grows at refrigerated temperatures, which may pose a significant risk to immunocompromised individuals who consume certain foods refrigerated for long periods of time. One approach to managing *L. monocytogenes* risk is to formulate food so that *L. monocytogenes* will not grow more than 1 log over the shelf-life of the product.

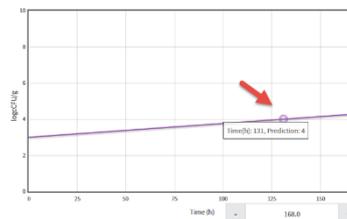
In this case study, you will design food conditions to extend the shelf-life of a food product, while maintaining less than 1 log growth of *L. monocytogenes*.

### Intended Learning Objectives

- Knowledge of features of ComBase Predictor growth models
- Understanding how intrinsic and extrinsic factors of food influence *L. monocytogenes* growth.
- Skill in designing food environments that extend shelf-life, while maintaining low levels of *L. monocytogenes*.

### Instructions

1. Using the ComBase Predictor, select **Growth** models.
2. Next, select the *Listeria monocytogenes/innocua* model interface.
3. Set the conditions to:  
physiological state = 1  
temperature = 5°C  
pH = 5  
 $A_w$  = 0.99  
Time = 168 hours
4. Move the mouse pointer along the line on the graph, and estimate the time where growth increases by 1 log from the initial level.



5. Next, examine the effects of other *Listeria monocytogenes/innocua* models (e.g. lactic, acetic, CO<sub>2</sub>, and nitrite) at extending 5°C shelf-life by **3-times longer**.
6. Questions
  - a. How can the following models be used to design a food environment that will extend shelf-life 3 time longer at 5°C?
    - *Listeria monocytogenes/innocua* CO<sub>2</sub>
    - *Listeria monocytogenes/innocua* acetic
    - *Listeria monocytogenes/innocua* lactic

- *Listeria monocytogenes/innocua* nitrite

## #5 Thermal inactivation of bacterial pathogens

Heat is a common method to inactivate bacterial pathogens in food. However, the amount of heat necessary to produce a specific inactivation rate varies according to bacterial species/strains and the food matrix. In thermal processing, 'D-value' is commonly used to designate the sensitivity of bacteria in a specific food matrix. D-value is the time (normally in minutes) to reduce a bacterial population by 1 log.

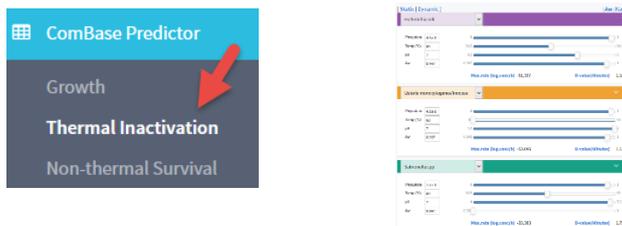
In this case study, you will compare thermal inactivation of three bacterial pathogens using ComBase Predictor models.

### Intended Learning Objectives

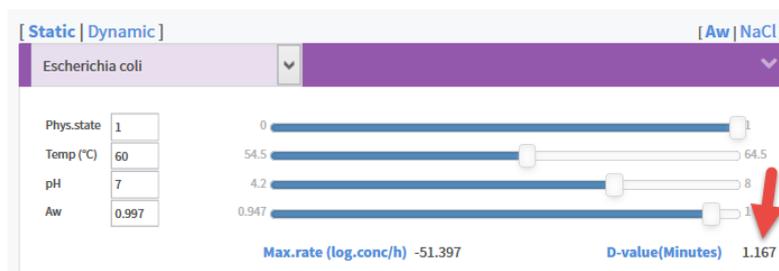
- Understanding features of ComBase Predictor thermal inactivation models
- Knowledge of how temperature, pH and water activity affect D-values
- Awareness of differences in thermal sensitivity among different pathogens

### Instructions

1. Using the ComBase Predictor, select **Thermal Inactivation** models. Next, select the **Escherichia coli**, **Listeria monocytogenes/innocua**, and **Salmonella** model interfaces.



2. Set identical conditions for each bacterial species, and then record and compare D-values. Change the 'time' scale as needed to better visualise the profiles.



3. Next, change the pH and water activity (Aw)/NaCl to different values, keeping them the same for all three species. Record and compare D-values. Change the 'time' scale as needed to better visualise profiles.

### 4. Questions

- a. Which species is more sensitive to heat?
- b. How do pH and water activity influence inactivation?
- c. Which parameter (pH or Aw) has a greater influence on inactivation?
- d. At the same pH and water activity, give an example of temperatures that produce approximately the same D-value.