Bond Pet Foods Saves Time and Money in Their Media Optimization Process with the Cell Growth Quantifier

BOND PET FOODS™

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Introduction

Who is Bond Pet Foods?

Bond Pet Foods is a Boulder, Colorado-based company using biotechnology to create meat proteins that are nutritionally comparable to their conventional counterparts but without all the bad stuff- so people, pets, farm animals and the planet all win. Using some of the same processes that are employed in craft brewing, Bond produces high-quality animal proteins through fermentation, harvests them to better meet the nutritional requirements of companion animals, and supplies the ingredients to manufacturers for pet food, treat and supplement applications.

An important part of bioprocess development is selecting the right growth medium that best supports the metabolic needs of your microbial organisms. Not all organisms require the same type or amount of nutrients.

Bond Pet Foods wanted to optimize their media for the best yield while saving resources to lower production costs.

Using SBI's Cell Growth Quantifier (CGQ), Bond Pet Foods ran a media optimization experiment focused on nitrogen sources. They observed significant cost- and time-savings when switching from manual to automated sampling, as well as identifying optimal levels of nitrogen in their media per production run.

The Challenge

When monitoring their microbial shake flask fermentation processes, researchers at Bond Pet Foods relied on manual sampling techniques to asses parameters, including optical density (OD), of their cultures.

The process of manual sampling to measure optical density (OD) involves a multitude of operations and each touchpoint leaves room for contamination and potential human error.

For each sample point, researchers at Bond Pet Foods must interrupt their shake flask experiment. Each flask must be removed, opened under a hood in a sterile environment, and sampled. The data is then manually collected.

Even with qualified technicians performing the right experiments, manual sampling provides low-resolution data. To fully understand the growth behavior of your organism, one would need 24-hour coverage, with sampling intervals of every one to two hours which can still result in rough growth curve estimations. This means that lab technicians would have to sample at times outside of the traditional workday, including nights and weekends, putting a substantial strain on staff.

Challenges of OD Manual Sampling

- **Low-resolution data**
- **Time-consuming**
- **Invasive sampling methods/ disturbed bioprocess**
- **Increased risk of contamination**
- **Increase in human error**

The Solution

There were several key benefits that Bond recognized in SBI's Cell Growth Quantifier over manual sampling when considering solutions to achieve their goal of media optimization while saving both time and money.

Non-invasive Measurements

The CGQ relies on the principle of light backscattering to monitor cell density. Each sensor plate is positioned under the cultivation vessel and measures the biomass non-invasively through the vessel wall. As a non-invasive technology, the CGQ removes the need to interrupt the experiment or open samples, reducing possible points of contamination and helping to preserve the growing environment.

Automated, Continuous, Parallel Measurements in Real-time

Once the plates are installed and the program is established via the software, the CGQ continuously monitors multiple shake flasks in parallel. Data from each connected sensor is brought into the software for real-time data visualization. The complete measurement cycle is now performed with a single push of a button, giving greater productivity elsewhere. Additionally, online sampling eliminates operator-to-operator variability in the measurement assay so that reproducibility increases among experiments.

Compatibility

When it comes to process development, shake flask experiments are an ideal set-up for many purposes and allow the testing of different conditions in lower volumes than bioreactors. Therefore, costs of medium and additives are generally lower, and experiments can be set-up in places with limited space. And unlike microtiter plates, working volumes in flasks are large enough to allow for other analyses. The CGQ was designed to work with shake flasks and can be easily incorporated in existing protocols.

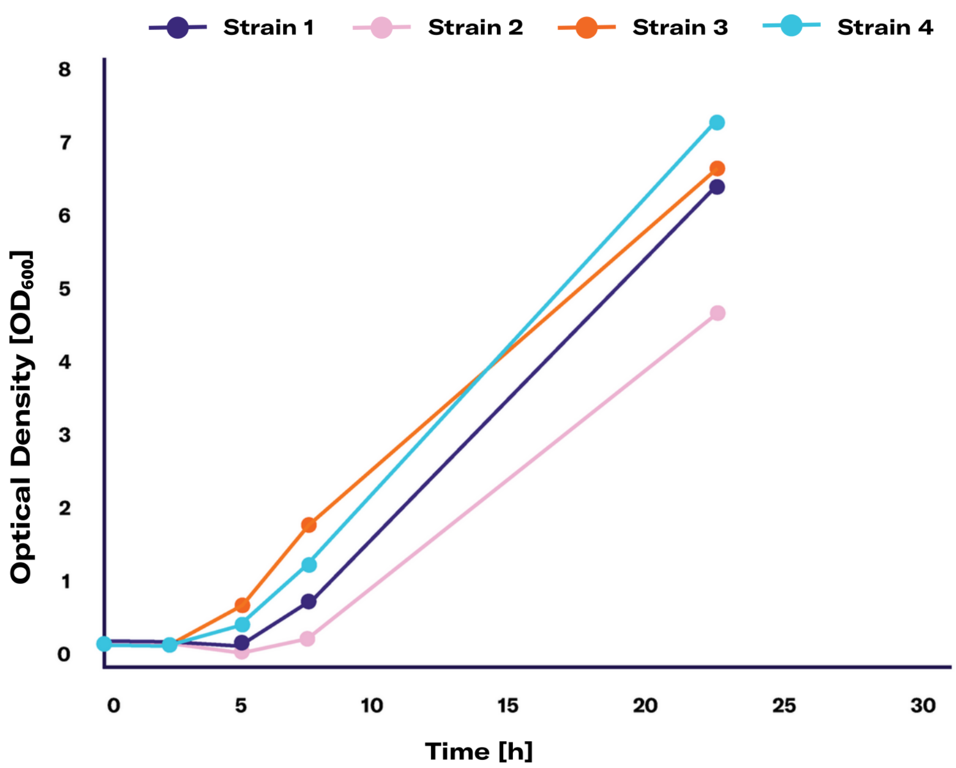
The Results

When asked to explain Bond's experience with SBI's Cell Growth Quantifier, Raul Reveles, Senior Bioprocess Engineer, says "The real power of the CGQ is being able to sample at specific points of interest based on the continuous, high-resolution data. Otherwise with manual sampling, it is just a shot in the dark- you never really know where in their growth curve the organism is."

New Bioprocess Insights

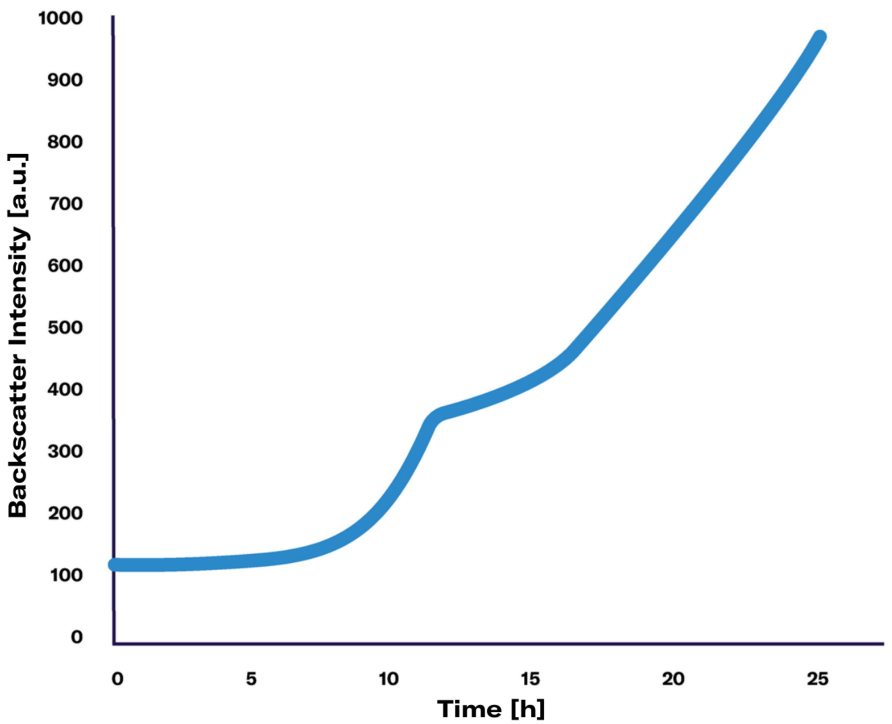
The high-density resolution provided by the CGQ allowed researchers to sample at key points in the organism's metabolism and life cycle, gaining better insight into their bioprocess.

Typical Flask Data



This is an example of a typical growth curve comparison as collected by one of Bond's strain engineers, using traditional flask sampling. Offline sampling is complex and time-consuming, resulting in lower measurement frequency with most pulls being at the start or the end of the experiment.

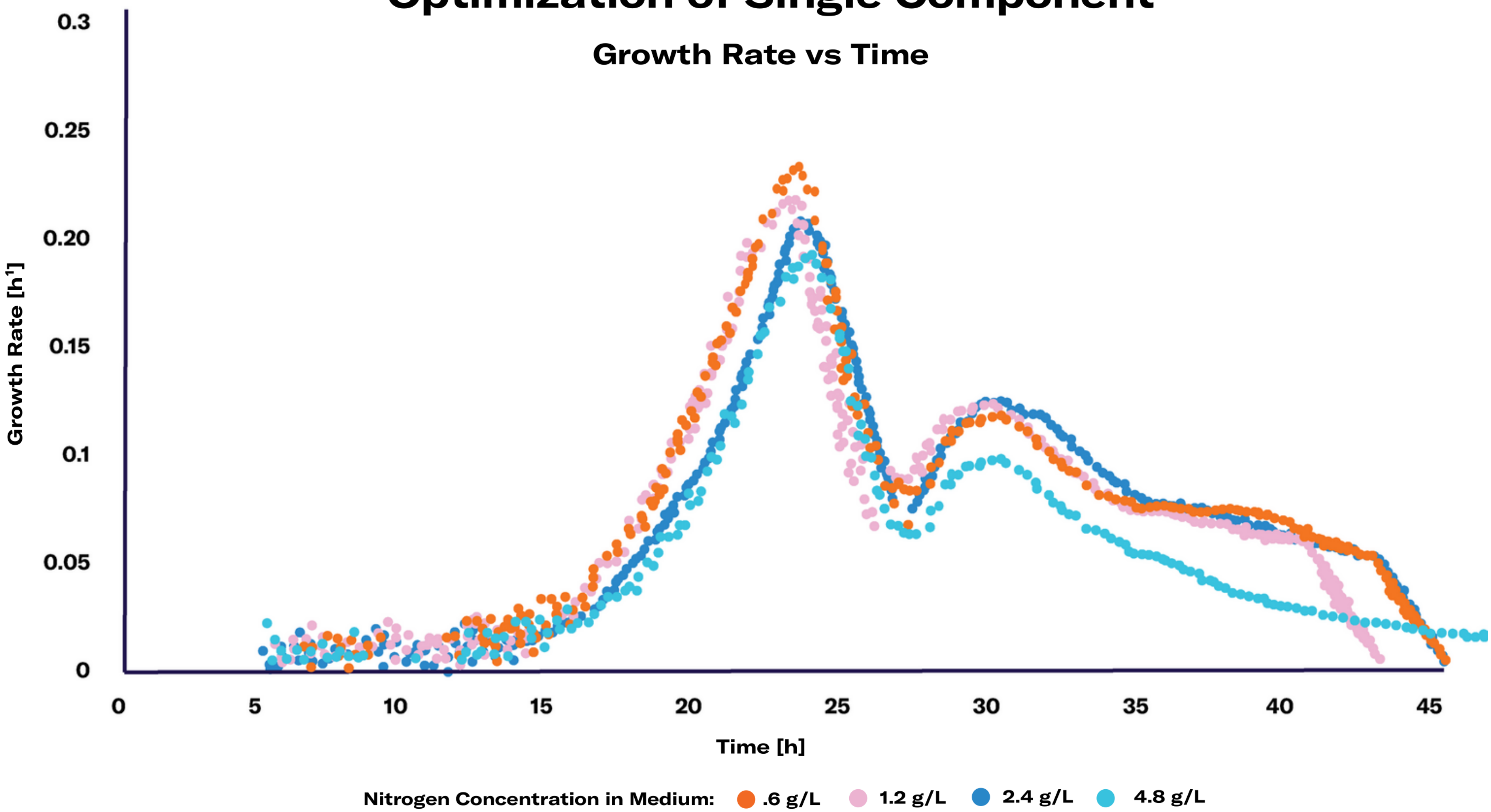
High Resolution Shows Biphasic Growth



This graph illustrates the growth rate resolution available with the CGQ. Due to this, Bond was able to determine that the growth curve of the organism was biphasic and involved consumption of the primary carbon source, followed by a second lag phase, and a switch to consuming the metabolic over flux products.

New Bioprocess Insights

Optimization of Single Component



The graph illustrates the organism's growth rate over time. The standard media recipe (cited from literature) called for 4.8 g/L nitrogen. This concentration was compared to varying levels (4.8, 2.4, 1.2, and 0.6 g/L of nitrogen). The data shows that as the concentration of nitrogen is lowered, the max growth achieved increases. The best performing nitrogen concentration was 0.6 g/L and showed a peak growth rate of 0.185 (a 3.75 hour doubling time).

The Savings

Per Optimization Experiment

It was found that 14 manual sample points were needed in order to obtain a similar resolution to that of the CGQ. Each manual data point took approximately 10 minutes to perform, including all relevant assays and data entry. For the following numbers, 8 flasks were used to used different conditions in replicates.

140 min

Per Flask (Sampling and Flask Manipulations)

- 10 minutes per flask
- 14 sample points to get similar resolution
- 8 flasks
- 10 minutes x 14 sample points = 140 mins per flask
- 8 flasks x 140 mins per flasks = 1120 minutes per experiment

$60

Per Flask in Labor Costs (Standard Labor Costs X 140 Min)

- 1120 minutes per experiment/ 60 minutes per hour = 18.5 hours of labor per experiment
- $30 per hour in labor costs
- 18.5 hours x $30 = $555 saved per experiment in labor costs

The Savings

Per Optimized Production Run

The optimization experiments showed that lower nitrogen concentrations for production are sufficient. Therefore, significant amounts of nitrogen could be saved in the optimized production runs

$840

Saved on Cost of Goods (Per 10K L Volume)

- $840 saved per optimized production run
- 1 run per week
- $840 x 52 runs per year = $43,680 per year

87%

Nitrogen Waste Reduction (From Original Recipe)

Get results like these for your bioprocess!

Let’s work together to find a solution that works best for you.

Speak with an Expert

The Big Picture

After analyzing the savings in both time and resources seen when switching from manual to automatic sampling during the nitrogen-optimization experiment, the following annual savings can be realized for Bond Pet Foods (assuming one production run per week).

$28,860

In Labor Costs per Year

- $555 labor costs x 52 weeks = $28,860 per year in labor costs

962 hours

Saved per Year

- 18.5 labor hours x 52 weeks = 962 hours of labor per year (~40 days of labor per year)

$43,680

Saved on Nitrogen-Based Cost of Goods per Year

- $840 x 52 runs per year = $43,680 per year

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Conclusion

What's Next for Bond Pet Foods?

With the implementation of SBI's CGQ, Bond Pet Foods was able to identify significant savings in both money and time.

Incorporating an automated, non-invasive technology for biomass monitoring gave researchers at Bond unprecedented insights into the growth behavior of their organisms. Access to high resolution, continuous growth curves provided critical information and helped to enhance process development.

As the company continues to grow, Bond is looking to accelerate their process development work by using innovative technologies such as the CGQ to support their mission of creating sustainable, high-quality meat alternatives for pet food applications. They will continue using the CGQ technology to identify additional savings potentials for other processes.

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