

FROM INSIGHT TO ACTION:

Pathogen Mapping to Effectively Address *Cronobacter sakazakii* Contamination in Infant Formula Manufacturing Facility



BACKGROUND

Cronobacter sakazakii is a pathogen of increasing importance to powdered infant formula manufacturers. It is a devastating pathogen that even at low levels can cause death to premature and term infants, the elderly and other immune compromised people. Similar to *Salmonella*, *C. sakazakii* is able to survive in low-moisture foods, like powdered infant formula, for exceptionally long periods of time. *C. sakazakii* contamination can come from, raw materials, the environment, or even found in the process itself.

Recently, an infant formula manufacturer approached bioMérieux for help to understand and mitigate *C. sakazakii* contamination found in the environment, raw material, in-process, and finished product. The diagnostic test confirmed *C. sakazakii* was present, but they had several unanswered questions.

► **An infant formula manufacturer needed to understand and mitigate *Cronobacter* contamination.**

- **Are these the same *C. sakazakii* strains that the normal cleaning methods were not addressing?**
- **Or new *C. sakazakii* issues introduced repeatedly? If so, where from?**
- **How could they better understand the issue so that they could resolve it?**

The issue costs the customer almost a **full week of halted production and over two million dollars** in lost sales, variable and fixed operational expenses, and root cause analysis cost, each time *C. sakazakii* is found. When standard mitigation practices did not resolve the issues, they turned to bioMérieux for help.

With Augmented Diagnostics, bioMérieux offers deep *C. sakazakii* expertise across multiple domains including advanced molecular biology, bioinformatics, and data science to tackle challenges usually not solved by traditional methods. The suite of applications and specialists helped the customer gain never-before-seen insights to effectively mitigate their persistent *C. sakazakii* challenge.

Your Trusted Partner in Augmented Diagnostics

METHODS

► bioMérieux utilized their tools and specialists to gain never-before-seen insights of the contamination.

The manufacturer shared 144 *C. sakazakii* isolates, collected over a twelve-month span, that were found in the factory environment. bioMérieux employed pathogen mapping, which is a combination of sequencing, bioinformatics, data science and process expertise to analyze and provide insights to the customer. The project answered the following questions about the nature of the *C. sakazakii* isolates:



RESULTS



Of the 144 isolates, bioMérieux found three different recurring strains of *C. sakazakii*:

- Strain 1** is recurring with 121 closely related isolates. All isolates of this strain were found in the environment and finished products. **This indicated that this resident strain had found a comfortable niche and was evolving within the facility.** This strain was found across all twelve months of the sampling period – confirming that the mitigations the factory was employing were not resolving the contamination issue.
- Strain 2** is recurring with 5 closely related isolates. They were found in raw material and finished product, but not in the environment.
- Strain 3** is recurring with 2 closely related isolates. They were found first in raw material and finished product and only later in the environment.

Other strains were detected only once and then appear to be resolved.

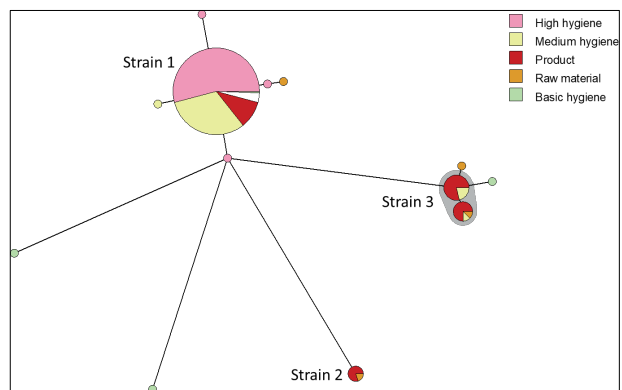


Figure 1: Minimum Spanning Tree demonstrating relationship and evolution of individual strains. A more detailed analysis of strain 1 shows two subclusters with some recent evolution in both clusters, suggesting this strain can spread and find new niches in the processing environment. Strains 2 and 3 were found in raw materials and finished product, indicating that the contamination found in finished product potentially derived from the raw material.

► Analysis confirmed the mitigations the factory was employing were not resolving the contamination issue.



Based on the data available, it is likely that Strain 1 entered the plant prior to the twelve-month period of the project scope. This *C. sakazakii* was not located in a single area of the factory. It originated in a medium hygiene zone, and was spreading across the facility and ultimately reached a high hygiene zone. An approximation of the data is represented below.

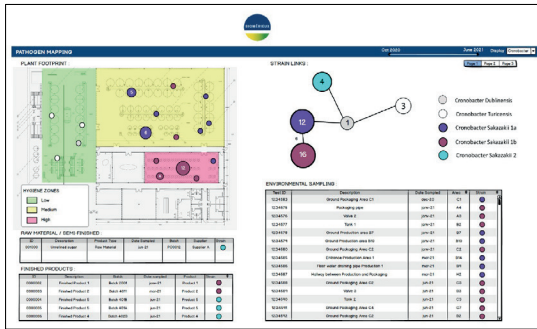


Figure 2: Generic representation of factory layout and hygiene zones, sampling points, isolates found, and isolate relationship. For confidentiality purposes, the diagram does not represent the actual factory layout.



Data from sequencing and environmental sampling can indicate how strains are related and migrating. **To gain true mitigation insights, it is critical to also understand the characteristics and behavior of the strains so they can be eradicated** and addressed appropriately if they reappear in the future.

We applied bioinformatics analysis and expertise and discovered that the particular *C. sakazakii* group in question developed a resistance to silver, copper, and nickel. The manufacturing facility relied on silver anti-microbial coatings in several areas of the different hygiene zones. Knowing these resistance mechanisms exist in the resident *C. sakazakii* strains brought immediate actionable insights to the manufacturer, and they were able to quickly change course in their sanitation practices.



✓ Copper resistance

✓ Nickle resistance

✓ Silver resistance

► **The infant formula company now understood their problem and changed their sanitation practices.**

CONCLUSION

bioMérieux's Augmented Diagnostic approach through robust pathogen mapping diagnostic investigation tools and expertise enabled the company to quickly understand their persistent *C. sakazakii* issues and make data driven decisions to effectively address the problem.

Their focus and diligence, coupled with the collaborative partnership with bioMérieux, not only **saved the company millions of dollars of lost production time, but also ensured they were delivering a safe product to their most precious customers.**

► **bioMérieux's Pathogen Mapping application and expertise enabled the company to save \$2 million dollars and eradicate the contamination.**



