DuPont Qualicon RiboPrinter® System

APPLICATION PROFILE

Hospital investigators enlist RiboPrinter® system to discover dangerous bacteria contaminating neonatology unit

For the fragile premature Infants In a neonatology unit, the key to survival often depends on a controlled and closely monitored environment that remains free of bacterial contamination.
When a number of these delicate babies In a French hospital were diagnosed with septicemia, epidemiologists focused on finding the source quickly and accurately.

The Investigation begins

The hospital's investigators took samples from the babies' nursery, including the new incubators that had recently been added to the neonatology unit. Unlike the older models that only allowed 60% humidity in the enclosed environment, these new units had the advantage of permitting an 80% level.

The epidemiologists also collected clinical samples from the babies: Blood, abdominal fluid, samples from the trachea of one patient and the shunt of another were examined by hospital staff microbiologists who used traditional biochemical methods.

For this investigation, however, these methods didn't provide the answer. They created confusion. What investigators needed to do was match the bacteria in the babies' blood to something in the seemingly sterile environment. Several days into the investigation, the most advanced biochemical tests has gotten scientists no closer to solving the problem.

Confusing identifications

That the babies were sick from the same bacterium was certain, but no source of the illness could be found. Bacteria, including *Pseudononas fluorescens,* was found in a number of locations in the nursery, but the biochemical tests indicated that the babies were infected with *Ralstonia picketti.*

In desperation, the hospital called the scientists at the Institut Pasteur de Lille (IPL), whose staff, many of them experts in genomics, have expertise in a variety of DNA fingerprinting techniques. Investigators believed that if they could find identical fingerprints in the environment and in the babies, they could track and eliminate the source of the infections.

The Center for Molecular typing at the institute relied on the RiboPrinter® Microbial Characterization system from Qualicon to quickly find the needed match. The bacterium that

was making the babies sick had a fingerprint identical to one of an organism found in the humidifying reservoir of the babies' incubator. However, the water supply coming into the incubator did not contain that bacterium.

Somehow, bacteria were contaminating the supposedly sterile water that was being used to moisten the air.

Multiple methods

Investigators obtained reference (known) strains of the two suspect bacteria – *Pseudomonas fluorescens* and *Ralstonia picketti* – from a well- respected culture collection. The RiboPrinter® system determined that the bacterium found in the reservoir was neither one of these organisms. The IPL scientists then used DNA sequencing – a method of "decoding" subfragments of DNA- on the offending organism. This told IPL scientists that the bacterium was known as MC5, an organism that Is very closely related to *Ralstonia solanacearum* and was formerly a member of the *Pseudomonas* family.

This close relationship was probably what had caused all the confusion. However MC5 Is so new a strain It didn't even have a name. But one important factor about this bacterium was known; MC5 degrades plastic. With this Information, investigators could conclude that bacteria was most probably leaching from the incubators' plastic reservoir into the water that was being used to humidify the babies' enclosure.

The hospital was quickly notified and the new incubators were replaced with older models that doctors felt they could trust. Then epidemiologists waited to see if they'd gotten rid of the problem by getting rid of the incubators.

Conclusion

They had. The investigators were right and the problem was solved.

Staff at the hospitals returned the incubators to the manufacturer with the recommendation that their design be reviewed and that the formulation of the plastic be investigated. Somehow, MjC5 had gotten into the units' reservoirs. It was now up to the makers of the incubator to find out how.

For doctors, nurses and epidemiologists at the hospital, there was tremendous relief. The source of the problem had been found and eliminated. In addition, the staff had learned how quickly and effectively DNA fingerprinting could help solve a serious problem. They also discovered how important it was to keep careful and accurate records that could connect individual patients with their environment.

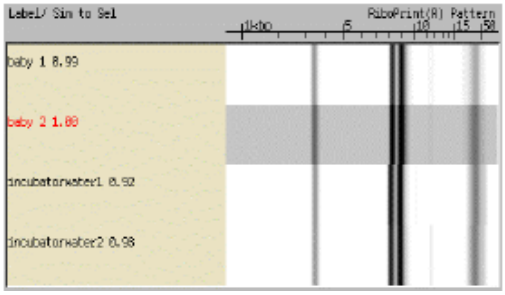

Figure 1. Identical RiboPrint® patterns for the strains isolated from the patients and from the water in the incubator reservoirs showed which bacterium was infecting the babies

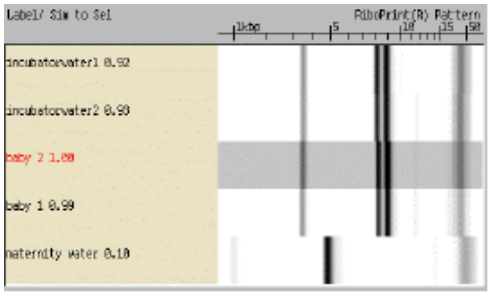

Figure 2. The RiboPrint® pattern for the strain isolated from the incoming water pipes did not match the patterns of the strains isolated from the reservoirs or from the patients

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