## **Bio-based disinfectants:** a development journey

A proudly South African company called Biodx has developed a cost-effective disinfectant technology that comes from nature and works with nature. MechChem Africa visits the company's Modderfontein premises and talks to CEO, Burt Rodrigues.

iodx is on a journey to reduce society's dependence on synthetic chemicals. "With financial support from TIA and the IDC, along with technical support from CSIR, we've spent the last 12 years relentlessly researching and developing antimicrobial technologies. As a result, we can now manufacture an organic, 100% biodegradable disinfectant compound that contains no chlorine, ethanol or aldehydes," says Rodrigues.

Called DECONT-X<sup>™</sup>. Biodx's formulation is 99.9% effective against bacterial species tested. "It is better than non-corrosive, it actively inhibits corrosion," Rodrigues continues, adding that Biodx is now manufacturing four variants: Microdx<sup>™</sup>, Vitrodx<sup>®</sup>, Indusdx<sup>®</sup> and Agridx<sup>®</sup>, all of which offer groundbreaking solutions for food & beverage; medical/ pharmaceutical; industrial/manufacturing; and agricultural industries, respectively.

#### In the beginning

Born in Mozambique of Portuguese parents, Burt Rodrigues came to South Africa as a teenager and attended a technical high school in Kroonstad. After finishing matric, his parents returned to Portugal, but Rodrigues remained in the country and enrolled at Wits Technikon to study electronics.

After a year, he won an AECI Bursary to do chemical engineering at Wits. During his third year, however, he decided to get married, so joined AECI and began 'working the benches' at the company's Modderfontein Laboratories. Initially, he performed analytical work on the ammonia and methanol bench, then he moved onto polymers and resins, and finally onto nitro-glycerine mixed acid (NGMA) and nitro-cotton mixed acid (NCMA) analysis.

In 1991/1992, Rodrigues joined Monroe Hickson's Organics, which later became Akulu Marchon, as works chemist, which began his journey into detergents and first generation quaternary ammonium compounds (QACs) such as Benzalkonium chloride or alkyldimethylbenzylammonium chloride (BACs), which were widely used as disinfectants in those days. "Although now considered harmful, these are still manufactured today," he savs

"The early BACS could neither be neutralised nor biodegraded, so they were very environmentally unfriendly. In the 50s, 60s

and 70s, however, this had not yet emerged as a problem as the accumulation of pollution was not yet evident.

"Today we have to manufacture compounds that can be broken down so that they decompose/biodegrade without any long-term effects on the environment. Unfortunately, however, the older compounds are still available and they are cheap, so some people still persist in using them," Rodrigues tells MechChem Africa.

### A problem is identified

Rodrigues went on to form a small company for the supply of appropriate detergents and disinfectants for commercial and industrial applications. "We were initially supplying a relatively standard range of chlorine-based and BAC 50 products. But then enzymes started to become popular," he relates.

Enzymes are complex molecules that act as catalysts to speed up the process of breaking down biochemical compounds such as carbohydrates, proteins and fats. They have an active site, which latches onto to a piece of the molecule being broken down. The enzyme lowers the activation energy needed to break



the molecule's bonds and, as soon as the bonds have broken, the molecule is released leaving the enzyme unchanged.

"At Baragwanath Hospital, we were supplying a BAC 50 disinfectant for general use. Then another problem was encountered: grease, fat and other debris was continually clogging up the drains and they needed a long-term solution to keep the drains clear and clean.

"We had just started to import digestive enzymes from Novozymes, the biggest manufacturer in the world, which were very effective at breaking up fats and protein cells. As well as blocking drains, waste fat is an ideal food sources for microbes, which then produce biofilm, which starts to constrict the drain further.

"So we tested and started to supply enzymes to Baragwanath to digest the fat in these drains and unblock them. This worked very well, for about a month.

"The hospital was understandably unhappy and a further investigation began, in which we discovered an incompatibility between enzymes and chlorine- or BAC-based disinfectants. So by 2001/2002, we already knew there was a problem. You could not use environmentally friendly enzymes alongside traditional chemical disinfectants," says Rodrigues.

"This was the point at which we realised that something new was necessary. And today, our DECONT-X<sup>™</sup>-based solutions can work alongside enzymes without destroying their digestive power," he notes.

Further elaborating on the problems associated with BACs and chlorine-based disinfectants, Rodrigues says that although modern compounds with longer chain lengths have now been formulated that are biodegradable, the big problem with traditional disinfectant chemicals is that 'the microbes' develop resistance.

"Microbes and bacteria grow in colonies, feeding on the proteins and carbohydrates around them. Chemical disinfectants such as

Three views of the Biodx 20 t processing plant at Modderfontein Laboratory Services, which is being used at a capacity of 20 t per month to develop and test the product range. The company is now building and commissioning a new 100 t plant on its own premises



by rupturing the bacterial cell walls because of a micro voltage charge difference. This equilibrium variation in the charge is enough to rupture the cell wall, allowing the fluid protecting the nucleus of the cell to leak out," Rodrigues explains.

"The disinfecting compound generally remains unaffected, leaving it available to continue to attack more of the targeted bacteria. But to survive, the cells develop resistance alarmingly quickly. The microbe world is tiny, but the standard operating procedure for life applies: they eat, reproduce and die – and this cycle happens in minutes. This means that evolution happens at a vastly accelerated rate. We have measured a sample of 300 000 colony-forming units (CFUs) in the morning and by the afternoon, the sample was up to 800 000 CFUs. Some microbes will always survive a treatment, and the generations of surviving offspring therefore become increasingly resistant.

"Chlorine is widely believed to be the best water disinfectant. The maximum allowable dose in potable water, however, is 5 ppm. But the microbial load has adapted to cope with 5 ppm, and resistance goes up in orders of magnitude. So a microbe resistant to a 5 ppm dose will guickly require 50 ppm to neutralise it, a dose that is too high for human health," Rodrigues notes. When it comes to cleaning surfaces, the

chlorine solutions and BACs kill 'microbes'

same applies. If cleaning a stainless steel surface with a 5 ppm solution when a 2 ppm solution is enough, it will create microbes that can only be killed using a 50 ppm solution, and this level of concentration is now going to corrode the working surface," he explains, adding: "microbial survival has built-in intelligence. One or two will always survive and it is only a question of time before dilution levels have to go up."

Another survival defence mechanism is the formation of biofilm, which provides a protective layer for a colony, inside which the organisms can survive the harsh surroundings. "Biofilm forms from polysaccharid, which is a by-product of microbial reproduction. This in itself contaminates the water and affects viscosity and conductivity. It dries like fibreglass, though, which prevents the disinfectant from accessing bacteria inside.

Microbes recognise the presence of a







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lethal environment and they will only emerge when it is safe to do so. All disinfectants only work when the target organism is in solution, so the microbes need to leave their 'nest' and come into the solution before treatment can be affected

"And here is where our innovation comes in. We asked ourselves the question, how can we entice microbes and bacteria to come into a disinfecting solution so that they can be killed?" Rodrigues tells MechChem Africa.

"At the same time, we wanted the solution to biodegrade in the process, so that by the time disinfection is complete, there are no harmful bacteria left and no disinfectant solution either. The clean-up job needs to be done without having any resulting negative environmental impact or residual harmful substances," he adds.

Chemistry dictates that the more stable the compound, the less biodegradable it will be, while less stable substances, which biodegrade faster, tend to have a weak disinfecting performance. "Overcoming this dilemma was another key challenge. We had to develop a stable molecule with a high disinfecting performance that readily biodegrades," Rodrigues continues, "from a solution that can 'trick' the survival instincts of the bacteria and bring them out from behind their microfilm shields."

#### **Biodx's solution from nature**

On realising that enzymes and traditional disinfectants were incompatible, Rodrigues began to seek a solution from the market. Having failed, however, he decided to explore the possibility of developing something new.

"I was attracted to the biotechnological revolution, which arrived here in the mid-1990s. There was white (industrial), green (agriculture), blue (water) and red (medical) biotechnology emerging at that time and, following a world tour, Thabo Mbeki launched a South African development drive that resulted in incubators being set up to give Biotech some local direction.

"We formed a company called CADKEM and submitted an application in 2004 to an incubator called Egoli Bio, which was quickly approved. We simply proposed to develop a bio-based disinfectant using citrus extracts. CADKEM was closed a few years later and we opened Biodx to better reflect the direction we had adopted."

Why citrus, we ask? "We needed a sustainable crop, because the feedstock has to be grown, so manufacturing is dependent on the growing seasons and seasonal volumes. We also knew that our grandmothers used lemon juice to clean and disinfect, so we decided that citrus, which was abundant in the Eastern Cape at the time, was an ideal place to start," he responds.

From there on, it became a scientific research exercise. "We knew that citrus iuices contain antibacterial compounds, but these are not present all the time. Antibacterial generation is triggered by survival instincts when under attack.

"If you throw a naked person He says that citrus juice reacts similarly.

into the snows of Siberia, three possibilities emerge. The person will die immediately; they will die later trying to escape; or they will find resources in that environment to enable them to survive. Natural instincts drive the survival responses and the body automatically and instantly generates the adrenalin needed to escape and survive," argues Rodrigues. If the juice is put under pressure and sees an imminent threat to life, the juice naturally produces antibacterial substances to defend itself.

"So the trick is to use a trigger to make the juice think that it is under attack. It then produces antibodies - antifungal, antibacterial and antimicrobial compounds. After stabilising these compounds, we react them with a fourth generation QAC, a quaternary ammonium carbonate, to end up with a stable and powerful disinfectant.

"Also, because the juice consists largely of carbons and proteins, the microbes don't see a toxic environment, so they will come out from behind their microfilm shield, enabling our antimicrobial disinfectants to attack them."

As part of the proof of concept, Modderfontein Laboratory Services agreed to assist Biodx by putting together a small plant on its Modderfontein site, which is being used at a capacity of 20 t per month to develop and test the product range. This plant, the process and the resulting products, are now certified by SABS and substantiated by NRCS.

"And we are now building a new 100 t plant of our own that we are busy commissioning. We hope to get this certified before the end of 2018 and we are currently very close to being operational," Rodrigues reveals.

As with the pilot plant, the base product, DECONT-X<sup>™</sup>, will be manufactured and used to make the four variants: Indusdx, Microdx, Vitrodx and Agridx.

nificant amounts of work to get the approvals required for widespread use, a process that can take years. Two trials of its Agridx, for an apple producer, have been successfully completed and a full-scale pilot is currently being implemented to treat a warehouse of crated apples waiting to be processed.

The challenge is to optimise the dosage so that all the apples are preserved for the time required without any residual unreacted

On the agricultural side, Biodx is doing sig-

Biodx is involved in two trials of Agridx® for a local apple producer. Initial trails have been successful and a full-scale pilot is currently being implemented to treat a warehouse of crated apples

waiting to be processed.

Agridx remaining when the apple is sold or processed."We strive to match the treatment time with the time it takes for the disinfectant to biodegrade, so that no microbes, disinfectant or residual chemical compounds remain at the start of processing.

"We are already achieving this balance very successfully with our Indusdx disinfectant for water treatment, which is an ideal substitute for chlorine-based treatments." he continues. "And we have now created a cheaper alternative for industrial water treatment. Called DECONT-A, this product is based on using the ascorbate extracts from our juice. It is not fully natural, but it is fully biodegradable and very affordable for the treatment of industrial wastewater." he adds.

DECONT-A has a significantly positive effect on bio-fouling, particularly for processes that are using reverse osmosis (RO) membranes. The advantage is that it disinfects the water and biodegrades completely by the time it leaves the clarifier. The water can then go through RO plants free of any active ingredient that might block the membranes used," he tells MechChem Africa.

"Vitrodx is also being well received for clean-in-place (CIP) use. This product offers solutions against *listeriosis*-type outbreaks and many producers who do not have problems right now are taking pre-emptive action to make sure their foods remain safe," he notes.

"At Biodx we believe that working with nature can benefit both the Earth and the bottom line of industrial, food, health and agricultural producers. We will never stop working to enable better solutions that enable a better world," Rodrigues concludes.