Barnacles, Darwin and Marine Paint Research

Having been a fish developmental biologist in my younger days and a biomedical scientist in my middle years, my own passion for barnacle research did not come until later, after meeting Prof. Dan Rittschof at Duke University Marine Laboratory and Sister Avelin Mary at Sacred Heart Marine Research Centre (SHMRC) in the early 1990s. Barnacles are not exactly the cute furry creatures one can get passionate about, so I have to admit that the interest was partially clouded by my capitalistic pursuits. That was the time when the ban on tributyl tin (TBT) was just looming on the horizon and there was a mad pursuit to discover the ultimate nontoxic barnacle settlement inhibitor. The search for TBT-free antifouling was the 'holy grail' of the marine paint business.

Before discussing my version of business evolution at Poseidon Ocean Sciences, I wish to digress a little to talk about Darwin and the barnacle. Like many of us in this business, we write about the barnacle, *Balanus amphitrite amphitrite Darwin*, and yet do not give any thought to why Darwin's name came to be part of this nomenclature. So, let me tell you why.

Charles Darwin

The Charles Darwin we are all familiar with is the English naturalist who wrote *On the Origin of Species by Means of Natural Selection* in 1859, which has since become the foundation for our understanding of evolution and the unifying explanation for the diversity of life on earth. He wrote about his theory in 1844, then quickly shelved it inside his desk drawer, specifically instructing his wife to release it for publication only if he died unexpectedly. Darwin was a modest man who shied away from controversies, and he knew his theory would be controversial; it remains so even on this year's 150th anniversary of writing *On the Origin of Species*.

For 20 years, this document remained hidden until he received a letter from a young English naturalist, Alfred Russell Wallace, then living in an island of what is now Indonesia. In a malarial fit, Wallace remembered reading Thomas Malthus' 1798 *Essay on the Principle of Population* (which coincidentally also inspired Darwin) and reached his own *Eureka* moment totally independently. He quickly dispatched a letter to Darwin describing an almost identical theory of evolution. In the typical Darwin sense of fair play, he presented Wallace's ideas and his own at the same time during the meeting of the prestigious Linnean Society, giving equal credit to the idea of Wallace and the share of the controversy as well. Yet, Darwin is credited with the theory of natural selection because his ideas were written while Wallace was yet in his teens, over 20 years before.

Then, you may ask, what did he do for those 20 years? Besides dealing with his failing health and the tragedies in his life, he was consumed by the passion of cataloguing barnacles. His interest in these tiny, ugly creatures began during his famous around-the-world voyage in HMS Beagle. Then, at the age of 26, young Darwin was exploring the Chilean coastline looking for biological specimens when he came upon a conch shell with its thick shell riddled with tiny boreholes. Inside the hole was a microscopic creature, attached by its head to the shell and waving six tiny legs. Knowing that it was a

By **Jonathan R. Matias**, Executive Director | Poseidon Sciences Group, New York, NY

barnacle without a shell, Darwin became even more fascinated since it had never before been described by any naturalist. He was a disciplined taxonomist and organized the chaotic nomenclature of this organism that number over 1000 species. Most of the species were often misnamed during his lifetime. Upon his return to England and immediately after writing his ideas on natural selection, at great expense to his health, he began his day and night obsession with barnacles that lasted for eight years (1846-1854), cataloguing the collection from his voyage and from the hundreds more sent to him by mail from around the world.

What drove this passion about such a mundane organism? Perhaps a clue comes from an earlier anonymous publication in 1844 of a controversial, incendiary, speculative book, *Vestiges of the Natural History of Creation* (later confirmed to be the work of Robert Chambers, a Scottish medical journalist). Widely panned and mocked for its evolutionary ideas even by Darwin's friends, the failure of the book was a great personal disappointment because Darwin expected a similar response to his own ideas in *On the Origin of Species*. Even his best friend, the noted botanist Joseph Hooker wrote, "no one has the right to examine the origin of species who has not minutely described many." Perhaps, one reason for this obsession was indeed to minutely observe a distinct part of the natural world and in so doing earn his right to question their origins. Whatever the reason might be, Darwin started us all on a path of research towards understanding barnacle biology and the commercial opportunities that follow in its wake.

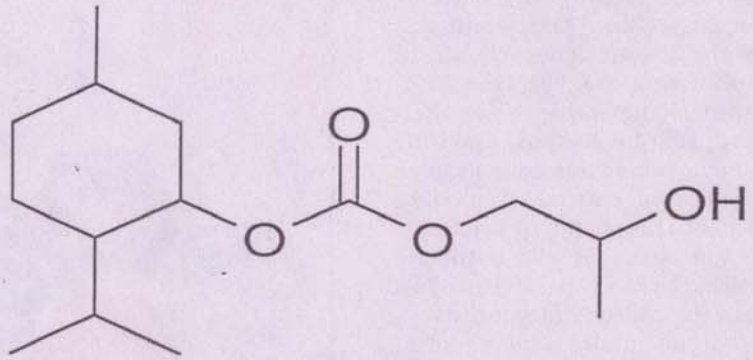
Development of Poseidon Ocean Coatings Research

Though not as dramatic as Darwin's, my adventure towards discovering the ultimate barnacle settlement inhibitors followed similar paths as many of us. The 1990s was a time when marine biotechnology was in its heyday and a lot a promise for a 'cure all' was just beyond the horizon. Scientists were extracting every marine organism they could get a hold of and looking for bioactive chemicals. For us in the marine paint industry, the goals were purely mercenary – finding one chemical, that 'magic bullet' that would bring great income opportunity especially for fledgling companies like mine.

At the time, the main workhorse in barnacle research utilized the method

developed by Dan Rittschof called the barnacle cyprid assay, which still remains the best method to date. Here, the larvae are artificially cultured until they reach the stage called cyprid when they are competent to attach to surfaces. The cyprid assay still remains the best screening method to date. But, it was labor intensive and needed a green thumb to culture the barnacle larvae (and all the microalgae needed as food to sustain them artificially). Considering the sheer number of chemicals I wanted to test and the limited time available, Dan Rittschof suggested I collaborate with Sister Avelin Mary, who was his post-doctoral student and had since returned to India to establish her own laboratory. Sister Avelin got her Ph.D. in biology to become one of India's most celebrated marine scientists. Because the internet had not yet reached her place in the port city of Tuticorin in the mid-1990s, collaboration depended solely on snail mail and fax (both unreliable even at the best of times) between New York and Tuticorin. I often wonder at how patient we were in those days. Yet, we were able to identify an active fragment from the purified extract called juncelin, named after Sister Avelin and the soft coral, Juncela. Through computer simulations of the structure of the fragment, we were able to identify the molecular structures that likely would repel barnacles, not kill them.

FIGURE 1 | Menthol propyleneglycol carbonate



Barnacle Inhibition Development

Finding a barnacle inhibitor that will have any remote chance of being commercial must meet a major hurdle—the cost. Marine paint biocides are an industrial commodity, huge volumes at low cost; a commodity that should cost no more than US $100 per kilogram. Knowing what the structure should look like, we began cross-matching known compounds that are relatively cheap and used either as food ingredients or in pharmaceuticals for human and veterinary use. This was Poseidon's Natural Bioproducts Screening Programme. Our presumption was that we would likely find from such a known inventory a series of compounds, we referred to as the NB series, with similar structures that were reasonably safe and could be mass produced cheaply. Years later, we narrowed our search of NB compounds to small molecules with menthol-like configurations, finally settling on a menthol derivative called menthol propyleneglycol carbonate, a GRAS (generally recognized as safe) food ingredient used as cooling agent in chewing gum and cosmetics (Figure 1). This chemical, based on tests, had repellent characteristics not only against barnacles, but also flies, mosquitoes, termites and even head lice. I suppose barnacles are nothing more than simply bugs in the sea! No wonder agro-chemical companies are taking their crop protection chemicals and testing them out against barnacles too. We just did it the hard way, starting from the ocean. Millions of dollars of R&D money and a decade later, I was hoping for a more exotic looking chemical besides a menthol derivative.

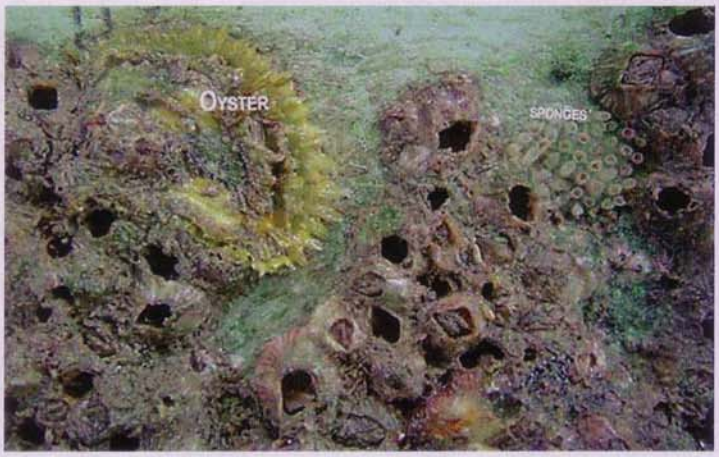
Field Testing

Of course, laboratory testing is never sufficient. We needed field testing to validate the antifouling performance of our test

chemicals on coated panels immersed in the sea. To make this possible, we built our own facilities in several countries. Looking back, that business decision was more likely driven by a Darwinian-like passion rather than business sense. Validating an additive in a marine paint is in itself a test of patience and endurance for both the company and the people involved. It takes years to complete such testing; years of watching barnacles grow.

One of the biggest lessons learned was that there was no short cut, no magic method that can predict the behavior of coatings. Like Sister Avelin said in one magazine interview, "There is no magic in the world [of science] except the magic of hard work." She did not specifically mean marine paint R&D, but it definitely applied to us nonetheless. It is hard work, and only the passionate ones need apply! Marine paint development remains to this day a never-ending series of trials and errors. One increases the odds of success by simply doing as many variations as possible. It is a test of will power for both the scientists and the stockholders.

Along the way our scientists slowly built a research family from the many years of collaborations. One of those enduring relationships, spanning almost a decade and a half, was with Sister Avelin at SHMRC. It is a unique organization in many ways. It is an organization run by nun scientists, yet independent from the daily obligations of the Congregation. Their task is to use marine science to contribute to marine conservation and livelihood development for coastal communities.



Branching out from conducting laboratory assays, Sister Avelin's team built a test platform for shallow submergence of our test panels in one of the world's most aggressive tropical fouling environment. Later on she expanded the shore laboratory we had built to enable simulation of test panels in a unique open sea dynamic test system found nowhere else in the world. A month after completing the shore facilities and the dynamic platform, the 2004 tsunami hit South India and Sri Lanka. I was speaking with Sister Avelin by cell phone as she described the sea rising unexpectedly as she gazed out in the horizon. Then, the connection was cut off for five days. Everything around them was devastated, yet the shore lab and the Sisters survived the rising sea. The laboratory, built on top of stones and gravel, missed the tsunami by a mere inch! The force of the currents snapped off half of the cables holding the platforms stationary, but the workers rode the successive waves unharmed. Miraculously, we did not lose a single life and even a single test panel.

This dynamic test system, now in its 6th year, allowed multiple flat panels to experience the fluid shear forces. It duplicates the erosion of a submerged surface traveling 250,000 miles in a single year. Since the bulk of our research was already completed, we decided to open our facilities for use by other companies to maximize its value to the marine industry. The same program was instituted at our Athena Biosystems testing facility in the Philippine Islands, where subsea test platforms for deepwater submergence at 100 ft below the ocean surface provides testing capabilities for coatings designed for oil rigs and pipelines. Thus, this discovery program has evolved into a company that supports the marine paint industry through contract research – an activity that has now reached the same time scale as Darwin's eight-year barnacle obsession.

This barnacle obsession is shared by many biologists and paint chemists around the world. And there is a never-ending list of questions yet to be answered about barnacle biology and how chemists can employ such knowledge in keeping them off ship bottoms. Now that TBT is legislated out of marine paints by a worldwide ban, and copper seems to be the new target for environmentalists, the chase is on once again for the next best thing. The passion for barnacles continues on for Poseidon Sciences and we can always blame Charles Darwin for getting it all started. ■

For more information see www.poseidonsciences.com, or contact jrmatias@poseidonsciences.com.