Forensic investigation into the origin of *Pseudomonas aeruginosa* PA14 — old but not lost

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The description of *Pseudomonas aeruginosa* strain UCBPP-PA14 created a stir in 1995 when Professor Frederick Ausubel’s team (Department of Genetics, Harvard Medical School, Boston MA, USA) first reported its highly virulent properties in both animals and plants due to several common pathogenicity factors [1]. This strain is gradually replacing *P. aeruginosa* PAO1 [2] for pathogenesis research. To date, at least 11 200 publications have reported the use of the PA14 strain (Google Scholar; search terms: *Pseudomonas aeruginosa* PA14; accessed 24 May 2018).

In 2007, I started a search to find the original publication describing *P. aeruginosa* PA14. Professor Ausubel’s group had reported it in 1995 [1]. When I asked Professor Ausubel how he acquired this strain or whether it was previously published, he could not recall. However, buried in the references, citation 7 to be exact is as follows, “The UCBPP *P. aeruginosa* strains, a collection of 30 human, 20 soil, and 25 plant isolates, were obtained from M. Schroth, University of California at Berkeley’ [3]. A call to UC Berkeley revealed that Professor Schroth had retired but that he still came to the University to pick up his mail. Hard to predict his exact comings and goings, they said, but he did appear at some times more than others. I religiously called the department at those times until I was able to speak with him personally in December 2007. The resolution of the PA14 mystery was in front of me. Professor Schroth indicated that the strain came from his collection named UCBPP, the abbreviation for University of California Berkeley Plant Pathology. He had obtained PA14 from Dr Spyros Kominos, a medical microbiologist at Mercy Hospital, Pennsylvania. Professor Schroth sent me a photocopy of his article in which PA14 was first reported [3]. This article showed that the strain PA14 is one of the most virulent clinical isolates to cause extensive plant rot.

During my discussion with Professor Schroth about the *Pseudomonas* genus, I learned that he and his graduate student, David Sands, were the first to report that most plant-pathogenic pseudomonads differed genetically from typical soil-borne pseudomonads [4]. Their finding was met with great scepticism from their highly regarded colleagues from across campus, Roger Stanier and Michael Doudoroff. These were considered among the world’s *Pseudomonas* experts and they could not believe that two young plant pathologists had uncovered a major group of pseudomonads existing right under their noses and studied by plant pathologists for many decades.

Professor Schroth explained that his correspondence with Dr Kominos started when he read one of his publications that dealt with a high number of burn patients infected with *P. aeruginosa* [5]. Dr Kominos was investigating the possible habitats in hospitals where the bacterium may exist, such as on screens in faucets that carry debris. He also thought that raw foods such as vegetables and salads were potential sources [6–8]. Professor Schroth was aware that *P. aeruginosa* had been reported as a plant pathogen [9–13], and thought it would be productive to start a collaborative study with Dr Kominos. Professor Schroth and his students were undertaking a lengthy investigation looking for strains of *P. aeruginosa* in various environments. Dr Kominos sent him isolates from burn patients that included PA14. The result: a characteristic comparison of the various *P. aeruginosa* strains relative to their sources [3, 14–16].

*P. aeruginosa* PA14 became part of a collection of 75 *P. aeruginosa* isolates: 30 human strains, 20 from soil and 25 from plants. The strain PA14 later became part of the international collection of plant-pathogenic bacteria (ICPBP), a collection that dated back to the 1920s. Because of the unfortunate termination of the plant pathology department and other agricultural departments at Berkeley, a contentious event [17], Professor Schroth’s bacterial collections were destroyed, including those collected in the 1920s, despite the fact that there was an endowment to support them. This precious, pre-pesticide collection had been started by plant pathologist Professor Walter Hagemeyer Burkholder (who first described *Pseudomonas cepacia*, which was later named *Burkholderia cepacia*, hence the genus bearing his name) of Cornell, then given to Professor Mortimer Starr, UC Davis, and then to Professor Schroth. The destruction precipitated in Schroth taking an early retirement.
In discussing this subject in 2007, Professor Schroth lamented the fact that so little attention had been given to studying pathogenic microorganisms that occur in nature and how they find their way into human foods. He also believed that earlier publications were pretty much lost [6–8, 14, 15]. I found this to be a fascinating subject as this was around the same time when Salmonella contamination became a concern of the salad industry. The reason why such events are more common today, he said, has to do primarily with post-harvest storage and shipping practices and the environments to which foods are exposed. A breakdown of equipment may lead to a warm and moist climate ideal for bacterial proliferation.

He further explained that food sickness is common in the tropics because the warm and high humidity environments provide an ideal climate for bacterial growth and survival on plants. Pathogenic and non-pathogenic bacteria are part of a natural microflora in and on plants. Bacteria can easily invade plants through natural openings such as stomata. In dry climates, it is rare to find human pathogens on the exterior of plants, and if present at all, they do not survive long. In the tropics, most vegetables are cooked prior to consumption for this reason; eating raw vegetables is a Western practice.

Fast-forward 10 years to November 2017, when I reconnected with Professor Schroth in California. Our thorough and deep discussion included everything from bacteria occurring on plants and which cause food-borne illnesses, to his first publications and present times. I received a new perspective on his published corpus and why his research and cooperative work with Dr Kominos ended rather abruptly. Among the reasons was their unsuccessful solicitations of a grant from the United States National Institutes of Health (NIH) with the comment that Schroth, a plant pathologist, has no business working on a human pathogen. At the same time, he was chided by some plant pathologists that he was not doing research on a real plant pathogen. His research also was not popular with agriculturists and the ornamental industry [14, 15, 18]. With no private sources of support, Professor Schroth discontinued his work with *P. aeruginosa* but not before publishing several seminal papers on the source of *P. aeruginosa*, its ability to donate resistance markers to other pseudomonads and its ability to cause cross-kingdom infections [3, 14, 15, 19, 20].

I ended the conversation by asking why his research was seemingly lost. He said in the sciences, discoveries, no matter their importance, often are not accepted, or are considered of little consequence. Moreover, scientists primarily read only the journals that relate directly to their disciplines. He believes his relationship with Dr Kominos was unique and did not know of any other interdisciplinary teams in the field of plant pathology like his. I informed him that the NIH now promotes extensive multidisciplinary research and that Schroth was just much too ahead of his time.

Professor Schroth expanded, ‘Our research did not require brain power as it was well known by plant pathologists that plants harbor many bacterial species. This is because plants exude all kinds of nutrients from their foliage, and roots, and therefore provide an excellent habitat for both fungi and bacteria, which may include food-poisoning bacteria’. He admits finding it a bit disappointing when he reads reports that food sickness bacteria were found on some vegetables, as if this were a new and inexplicable phenomenon.

Back in 2007, I considered that publishing the origin of PA14 would be a service to the *Pseudomonas* community, and did not know how. At that time, no copies of the book containing this original publication [3] were available as the Raven publishers were no longer in business. In 2016, a search for the original book on Amazon.com led me to acquire a copy priced at $14.84 from a UK-based company (Better World Books). In April 2017, I assumed the position of Co-Editor-in-Chief of the *Journal of Medical Microbiology*. With the help of the Microbiology Society publishing
team, we found that the publishing rights to this book were held by the Elsevier Publishing Company. We have been able to acquire permission to add an abstract and publish verbatim the first publication where PA14 made its debut [3] in the Journal of Medical Microbiology [21].

References

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