

Pseudomonas aeruginosa: A review of their Pathogenesis and Prevalence in Clinical Settings and the Environment

Klriisa Streeter, Mohammad Katouli*

Genecology Research Centre, Faculty of Science, Health, Education and Engineering, University of the Sunshine Coast, Maroochydore DC, Queensland, Australia

*Corresponding Author: Mohammad Katouli, Genecology Research Centre, Faculty of Science, Health, Education and Engineering, University of the Sunshine Coast, Maroochydore DC, Queensland 4558, Australia. Email: mkatouli@usc.edu.au; Tel: (+61) 7 5430 2845

Submitted: November 10, 2014; Revised: January 6, 2015; Accepted: January 7, 2015

The genus *Pseudomonas* consists of more than 120 species that are ubiquitous in moist environments such as water and soil ecosystems and are pathogenic to animals and humans. Within the genus of *Pseudomonas*, *P. aeruginosa* is most frequently associated with human infections. The bacterium is regarded as an opportunistic pathogen, primarily causing nosocomial infections in immunocompromised patients. The existing knowledge regarding the pathogenesis of *P. aeruginosa* has mainly been obtained through studying clinical isolates; particularly those involved in causing chronic lung infection in cystic fibrosis patients. Nosocomial infections commonly associated with *P. aeruginosa* include ventilator-associated pneumonia, catheter-associated urinary tract infections, wound infections in severe burn patients and septicemia with their pathogenesis shown to be multifactorial. The bacterium is also capable of producing a number of toxins via the type III secretion system, as well as secreting enzymes and proteins including elastase, phospholipase C and siderophores. However, *P. aeruginosa* is also a waterborne pathogen, commonly found in environmental waters as well as in other sources such as sewage treatment plants. The public health implication of these bacteria whilst in the environment has not been fully investigated. Here we review our present knowledge about the pathogenesis of *P. aeruginosa* in clinical settings and the environment.

Key words: *Pseudomonas aeruginosa*, Virulence factors, Pathogenesis

1. Background

Included in the category of genus *Pseudomonas* are genus *Pseudomonas* which contains more than 120 species that are all-over in moist environments such as water and soil ecosystems and infective to plants, animals and humans (1, 2). *Pseudomonas* species are easily detectable on agar due to the production of pigments such as pyoverdine which is a yellow-green, fluorescent pigment, and pyocyanin that is a blue-green pigment (3-6). Within the *Pseudomonas* species, *P. aeruginosa* is most frequently associated with causing human infection; however, it naturally exists in the environment (7, 8). The bacterium is regarded as an opportunistic pathogen, primarily causing nosocomial infections in immunocompromised patients (9-12). However, it is capable of causing a wide-spectrum of infections when normal physiological function is disrupted, including damaged epithelial barriers (13), depleted neutrophil production (14), altered mucociliary clearance (15) and the use of medical devices (16, 17). *P. aeruginosa* is rarely associated with causing chronic infections in previously healthy patients, although fatal cases of *P. aeruginosa* infections in previously healthy people have been reported (18, 19).

2. Context

2.1. *P. aeruginosa* causing respiratory tract infections

P. aeruginosa is well known for its ability to establish permanent residency in the airways of cystic fibrosis (CF) patients, resulting in the recurrence of chronic lung infections, progressive decline in lung function and increased morbidity and mortality rates (20, 21). The mechanism by which *P. aeruginosa* colonizes CF patients relies mainly on the pathogenesis of this genetically inherited lung disease. The disease is attributed to a gene mutation in cystic fibrosis transmembrane conductance regulator (CFTR) protein; a chloride channel which is involved in maintaining homeostasis

in epithelial tissues (15, 22, 23). Dysfunction of CFTR channels disrupts the regulation of chloride ion transport across the epithelia, resulting in sodium hyper absorption and impaired mucociliary clearance (15, 22, 23). Thick viscous mucus resides in the airways causing obstruction and blockages, and mucus hypoxia promotes *P. aeruginosa* colonization (24, 25). Non-CF patients are also susceptible to respiratory tract colonization of *P. aeruginosa*, especially patients with chronic obstructive pulmonary disease (COPD) (26). COPD patients display similar symptoms to CF patients such as decreased mucociliary clearance, and under these predisposing conditions *P. aeruginosa* is able to colonize and cause infection (15, 26).

P. aeruginosa is also a common causative agent of hospital-acquired pneumonia (HAP), in immunocompromised individuals (27). Colonization of the respiratory tract is initiated by the contamination of medical equipment and/or cross-colonization from other patients (27, 28). HAP is frequently acquired by patients using mechanical ventilation, termed ventilator-associated pneumonia (VAP) (29). *P. aeruginosa* is frequently isolated from hospital medical equipment, due to the bacterium's ability to survive in biofilms (16, 30). Previous studies have associated VAP with prolonged use of ventilation and prolonged duration in intensive care units (ICU) (29).

2.2. *P. aeruginosa* causing urinary tract infections

Urinary tract infections (UTI) caused by *P. aeruginosa* usually occur secondary to catheterization, instrumentation or surgery. Catheterization of the urinary tract is the major cause of nosocomial acquired-UTI by *P. aeruginosa* (31). Catheters are utilized by pathogens as a source of host entry, attaching to the catheter surface in well-constructed biofilms (16, 17, 30). Furthermore, the insertion of the catheter may also disrupt mucosal epithelial layers, promoting bacterial colonization (31, 32).

2.3. *P. aeruginosa* causing skin and soft tissue infections

P. aeruginosa is the most commonly isolated bacterium colonizing severe burns and wound infections (33-35). Wound infections caused by multidrug resistant (MDR) *P. aeruginosa* have been associated with high morbidity and mortality rates worldwide (33, 35). Estahbanati and colleagues (2002) investigated *P. aeruginosa* isolated from burn wound infections in a burn centre in Tehran and found that the majority of isolates were MDR and less than half of the patients were discharged from the centre (34). Nosocomial outbreaks of *P. aeruginosa* have been reported in surgical wounds, causing post-operative wound infections (36, 37). In addition, *P. aeruginosa* can disseminate from the initial infection site and enter the bloodstream, causing septicaemia (36). High mortality rates of *P. aeruginosa* septicaemia have previously been reported (38, 39).

Mild skin infections can occur in previously healthy people, associated with *P. aeruginosa* contamination in swimming pools, hot tubs and other water sources (40-43). Follicular dermatitis caused by *P. aeruginosa* has previously been described as an itchy rash with a red base and white pustules (44). In addition, nail diseases (e.g. onycholysis) are susceptible to colonization of *P. aeruginosa*, and is commonly referred as "green nail syndrome" (45-47). Paronychia infection has been associated with prolonged exposure to moist environments (e.g. swimming). McNeil and colleagues (2001) investigated an outbreak of *P. aeruginosa* infections in post-surgical patients, reporting severe onycholysis and onychomycosis in a nurses' thumbnail as the primary source (47).

2.4. *P. aeruginosa* causing bacterial keratitis

P. aeruginosa is the leading cause of bacterial keratitis (48), and occurs in patients with pre-existing ocular disease, in post-ocular surgery patients and in individuals who use contact lens. *P. aeruginosa* has been shown to adhere to the disrupted corneal epithelial cells, and internalize rapidly (49, 50). Contact-lens associated keratitis is mediated by the extended use of contact lens that has been shown to disrupt the epithelial surface of the cornea, causing cornea abrasions (49-51). Furthermore, bacterial keratitis initiated by contact lens contamination has been associated with patient noncompliance with appropriate contact lens care (52).

2.5. *P. aeruginosa* causing 'swimmers ear' infections

Otitis externa, commonly known as 'swimmers ear', is an inflammation or infection of the external auditory canal, due to prolonged exposure to moisture and/or the insertion of foreign objects (e.g. cotton tips) (53, 54). It is well known that *P. aeruginosa* is the most common pathogen of otitis externa, strongly associated with swimming in contaminated recreational pools (55).

2.6. Pathogenesis of *P. aeruginosa*

The pathogenesis of *P. aeruginosa* has been extensively studied and proven to be a multifactorial process, mediated by quorum sensing. *P. aeruginosa* possess two quorum sensing systems, *las* and *rhl* that facilitate cell to cell communication through production of signalling molecules termed autoinducers to target specific receptors for activation (56-58). However, a high population density is necessary for the concentration of inducers to go beyond threshold. Maximal receptor activation induces gene expression of several virulence factors and biofilm formation (57-58). There are a number of virulence properties that help *P. aeruginosa* to colonize

and evade the host's immune system to cause infection, including adhesions, the type III secretion system and other secreted proteins.

2.7. Adhesions

Adhesion of bacteria to the host cell surface is the first step toward colonization and initiation of the disease (59). In *P. aeruginosa*, adhesion is mediated by type IV pili and the formation of biofilms (60-77). Type IV pili are the filamentous appendages attached to the cell surface of the bacterium. More than 40 genes involved in the biogenesis and function of type IV pili have been previously identified (60). Hahn and colleagues (61) have also shown that type IV pili accounted for 90% of the adherence, function and virulence in a mouse infection model. Type IV pili also assist in facilitating 'twitching motility'; the retraction and extension of pili, to facilitate bacterial movement along the host cell surface (62-64). As a result, the surface movement assists in the formation of microcolonies, which develop into a mature biofilm (65-66).

P. aeruginosa also secretes extracellular polysaccharides; alginate, polysaccharide-encoding locus (*pel*) and polysaccharide synthesis locus (*psl*) that are also involved in forming the biofilm matrix embedded around microcolonies (66-68). Biofilms protect the bacterium from the host immune system components, as well as resistance to antibiotics (69). Alginate is commonly described as 'slime', that is commonly associated with sputum cultures from CF patients, suggesting that mucoid phenotypes of *P. aeruginosa* are important for persistence and establishing permanent residency in the lungs of CF patients (70). It has been previously shown that mucoid phenotypes interfere with antibiotic effectiveness by decreasing uptake (71). Alginate has also been shown to inhibit phagocytosis and scavenge free radicals (72, 73). However, non-mucoid phenotypes (alginate deficient) have the ability to form biofilms, by utilizing *pel* and *psl* (74, 75). A study by Ma et al. (76) has shown a decrease in binding to the airway epithelial cells in *psl*-mutant strains, suggesting *psl* is necessary for adhesion.

Previous studies have compared virulence and antibiotic-resistance profiles of mucoid (alginate producing) and non-mucoid (alginate deficient) biofilm phenotypes (74, 77). Mittal et al. (77) correlated biofilm-producing *P. aeruginosa* with increased renal tissue damage compared to non-mucoid phenotypes of *P. aeruginosa*, suggesting that the biofilm production contributes to the pathogenicity and host damage in *P. aeruginosa* urinary tract infections. In contrast, Wozniak et al. (74) studied antibiotic resistance profiles in wild-type and alginate-mutant biofilms and reported no differences in the antibiotic resistance.

In addition to type IV pili and biofilm formation, adhesion is also mediated by other various cell surface features, including lipopolysaccharides (LPS). LPS is a large molecule found on the outer surface membrane in most Gram-negative bacteria (78). The LPS structure is heterogeneous in its lipid A and O-antigen structure, presenting in two glycoforms in *P. aeruginosa* (78). LPS serve as recognition molecules to the innate immune system, responsible of causing bacterial infection-induced inflammation (78, 79). The induced inflammatory response is variable, associated with the level of acylation in lipid A, whereby a fully hexa-acylated lipid A is necessary for a vigorous response (80). LPS signal transduction is a complex process mediated by binding LPS to lipopolysaccharide-binding protein (LPB) to form LPS-LBP complexes, that are transferred to CD14 receptors, for secondary activation of toll-like receptor-4 (79, 80). Previous find-

ings have shown that initial isolation of *P. aeruginosa* from CF patients contain O-antigen, however upon establishment of chronic infection, the same strain of *P. Aeruginosa* was found to be LPS O-antigen-deficient (81, 82).

2.8. Type III secretion system

P. aeruginosa interacts with the host via a protein, needle-like appendage known as the type III secretion system (T3SS). The T3SS injects toxic effector proteins into the cytosol of eukaryotic cells to inhibit cellular function for bacterial survival. Four effector toxins have been identified in *P. aeruginosa* and include exoenzymes S, T, U, and Y. ExoS and ExoT are bifunctional, possessing GTPase activating protein (GAP) activity and ADP-ribosyltransferase activity (84, 85), while ExoY is an adenylate cyclase (86). Previous studies have shown that ExoS, ExoT and ExoY function to acquire cytolytic activity, inhibiting epithelial cell invasion and preventing phagocytosis (86-88). Furthermore, these proteins are associated with disruption of endothelial cell junctions, via cell retraction and rounding to alter the cytoskeleton (86-88).

However, ExoU is a potent cytotoxin with phospholipase A₂ (PLA₂) activity; the ability to cleave phospholipid membranes to cause cell lysis (89, 90). ExoU has also been shown to acquire cytolytic activity to various cell types including epithelial cells, macrophages and fibroblasts (83, 89, 90). In addition, the PLA₂ activity initiates inflammation due to the production of arachidonic acid for cyclooxygenase and lipoxygenase pathways, resulting in prostaglandins production (91).

Feltman and colleagues (92) have previously reported that *P. aeruginosa* isolates possess either *exoS* or *exoU* genes, not both. On the other hand, previous studies by Wolfgang *et al* (93) have shown that this feature did not exist among clinical strains, concluding that all clinical isolates carried both *exoS* and *exoU* genes. A high prevalence of ExoS production has been documented in *P. aeruginosa* isolates from urinary tract and wound infections (92, 94). Conversely, ExoU has reported to be highly associated in Pseudomonal lung infections, including hospital-acquired pneumonia (85, 95). In fact, it has been shown that a significant reduction in lung pathology and virulence is associated with ExoU deletion in mutated isolates (96). Consistently, ExoU production in animal models and in patients has been strongly associated with the acceleration of lung injury (89, 97-99). In contrast, Dacheux and colleagues (99) associated CF isolates with a low prevalence of ExoU (i.e. 10%), which is similar to the study by Feltman and colleagues (92), who found that ExoU in only found in 8% of CF isolates. This suggests that virulence properties of *P. aeruginosa* causing lung infections i.e. pneumonia are dissimilar to lung infections associated with CF patients. Previous studies have suggested that ExoT is a non-variable virulence property, which is prevalent in all clinical isolates of *P. aeruginosa* (92, 100). In addition, Feltman and colleagues (92) also reported ExoY to be present in most clinical isolates of *P. aeruginosa*, supporting a similar finding by Dacheux and colleagues (99).

2.9. Other secreted virulence properties of *P. aeruginosa*

P. aeruginosa secretes a haemolytic and a non-haemolytic phospholipase C (PLC). Studies have shown that non-haemolytic PLC has no pathogenic activity, whereas haemolytic PLC degrades phosphatidylcholine and sphingomyelin that are commonly found within eukaryotic membranes and host lung surfactant, principally to cause lung injury (101, 102). Additionally, Meyers *et al* (102) also reported that

inactive forms of PLC were unable to cause an inflammatory effect. In addition, *P. aeruginosa* secretes elastase (LasB); a metalloproteinase involved in the host colonization and tissue damage (103). Previous studies have suggested LasB as an important virulence factor for bacterial survival from the host immune system, through degradation elastin and collagen (103), immune components including cytokines, chemokines, IL-2 and IL-8 and inactivation of immunoglobulin A and G (104, 105). Previous studies have shown a high prevalence of the *lasB* gene in *P. aeruginosa*, irrespective of their isolation origin (106-108).

P. aeruginosa secretes yellow-green fluorescent pigment known as pyoverdinin (4), that functions as a siderophore to acquire and transport iron from the environment through specific protein receptors on the outer membrane (109, 110). Three types of pyoverdins have been identified, although each strain of *P. aeruginosa* produces only one type (109). Animal models have suggested that pyoverdinin is a virulence-associated factor aspyoverdinin-deficient mutants demonstrate no virulence (110). A variety of phenazines are also secreted by *P. aeruginosa*, including pyocyanin. Pyocyanin is a blue redox-active secondary metabolite that is responsible for the production of a blue-green pigment, commonly used in rapid diagnosis of *P. aeruginosa* (111-112). Previous studies have suggested that pyocyanin is a major virulence factor, interfering with numerous cell functions (112).

2.10. Antibiotic resistance in *P. aeruginosa*

Antibiotic resistant (AR)-*P. aeruginosa* are strongly associated with nosocomial infections, that are a worldwide health concern due to the increasing development of MDR strains (i.e. resistance to at least three antibiotics). Various therapeutic challenges exist with MDR *P. aeruginosa* due to the limit of effective treatment strategies. Current literature is strongly associated inadequate empirical treatment with increased rates of mortality and morbidity (113-114). *P. aeruginosa* is intrinsically resistant to various antibiotics due to a low permeability in the outer membrane, which acts as a selective barrier (115, 116). However, this bacterium is a highly diverse pathogen that is capable of adaptation to the surrounding environment. When subjected to antibiotic selective pressure, the induced response facilitates bacterial survival and develops antibiotic resistance (115). The emergence of antibiotic resistance has been reported during host colonization of CF patients, whereby *P. aeruginosa* strains develop and acquire resistance during antimicrobial therapy (117). Studies by Messadi and colleagues (118) have reported a strong correlation between increased use of ciprofloxacin with increased prevalence of ciprofloxacin resistant strains. Therefore, another factor associated with the increase in MDR-*P. aeruginosa* is due to the frequent use of antimicrobial agents.

In addition, these bacteria utilize different mechanisms to resist antibiotics, although the presence of a mechanism does not always justify the source of resistance (113, 117). Therefore, many researchers have suggested that a variety of mechanisms are involved including the production of inactivating enzymes, target site modification, utilisation of efflux pumps and chromosomal mutations (119). *P. aeruginosa* produces β -lactamases; enzymes that hydrolyze the peptide bond of the β -lactam ring to inactivate the antibiotics (120). *P. aeruginosa* is able to produce various β -lactamases, including extended-spectrum β -lactamases (ESBL), metallo- β -lactamases (MBL) and chromosomal cephalosporinase (AmpC).

Various types of MBLs have been described previously and are carried on integrons (121). Previous studies have reported that MBL-producing *P. aeruginosa* in blood stream infections are associated with high morbidity and mortality rates (122). AmpC differs to the other β -lactamases for that the *ampC* gene is present in all strains of *P. aeruginosa*, although it requires a gene mutation, to cause hyper production and resistance (123).

Carbapenems are commonly used to treat *P. aeruginosa* infections due to their effectiveness against β -lactamase producing *P. aeruginosa*. An increased prevalence of carbapenem-resistant *P. aeruginosa* has been reported in hospitals worldwide (124, 125). A study conducted between 1996 and 2003 in a United Kingdom medical centre has reported the prevalence of imipenem resistant-*P. Aeruginosa* increasing from 2% to over 40%, with the majority of the isolates belonging to a single clone (125). Similarly, Vitkauskienė and co-workers (126) studied the prevalence of carbapenem-resistant *P. aeruginosa* over five years at a tertiary university hospital and showed an increase of 53% to 88% of carbapenem-resistant *P. aeruginosa*. In addition, OprD is a specialised porin located in the outer membrane of *P. aeruginosa* that facilitates the influx of amino acids, peptides and imipenem (a carbapenem antibiotic). Conformational changes in the external loops 2 and 3 of OprD have been shown to inhibit imipenem entrance into the bacterium (127).

P. aeruginosa is also capable of acquiring resistance through mutation in specific chromosomal genes (115). Fluoroquinolone resistance is acquired by modifying type II topoisomerases; DNA gyrase (*gyrA* gene) and topoisomerase IV (*parC* gene), to inhibit antibiotic binding (128, 129). Previous studies have suggested that the first target for fluoroquinolones activity is DNA gyrase, followed by topoisomerase IV as the secondary target (129). However, Salma and colleagues (130) have reported quinolone-resistant *P. aeruginosa* isolates without mutations in *gyrA* and *parC* genes, suggesting that another resistance mechanism is also used for the acquisition of fluoroquinolone resistance, such as efflux pumps. Efflux pumps rapidly remove toxic substances (e.g. antibiotics) out of the cytoplasm as a result of hyper expression of the *mexR* efflux gene (131, 132). The efflux pumps are a three component system; the outer membrane protein, the energy-dependant pump and a linker protein (132). In *P. aeruginosa*, four different efflux systems belong to the resistance-nodulation-division (RND) family including MexAB-OprM, MexCD-OprJ, MexEF-OprN, and MexXY-OprM (132). Studies have shown a wide-spectrum of substrates specificity for each efflux pump, including β -lactams and fluoroquinolones for MexAB-OprM and MexCD-OprJ (133). Current literature suggests that the overexpression of one or more Mex pumps is associated with MDR-*P. aeruginosa* in clinical settings (131-133).

2.11. *P. aeruginosa* in the environment

P. aeruginosa as a waterborne pathogen is a growing concern to public health sectors. Many sources of environmental water could potentially be acting as a reservoir for potentially pathogenic strains of *P. aeruginosa* (8, 41). Various studies have shown that water resources (including sewage treatment plants and river water) are highly polluted with pathogenic bacteria including *P. aeruginosa* (134, 135). Public recreational swimming pools have also shown *P. aeruginosa* contamination (136-138). In addition, outbreaks of whirlpool-associated folliculitis and UTI have previously been reported (139, 140). Grobe and colleagues (137) showed

mucoid phenotypes (possessing alginate) were able to survive in chlorinated water better than non-mucoid phenotypes. Despite this, non-mucoid strains of *P. aeruginosa* exist in swimming pool water (106). The presence of *P. aeruginosa* in swimming pools is associated with public health risks. Many studies have associated otitis externa, dermatitis, folliculitis and chloronychia with the use of swimming pools and hot tubs (141).

Lutz and Lee (142) studied the prevalence of antibiotic resistant *P. aeruginosa* in various swimming pools and hot tubs. These researchers found 96% of their isolates to be MDR including resistance to amikacin, aztreonam, gentamicin, ticarcillin/clavulanic acid and trimethoprim/sulfamethoxazole (142). In contrast, Tirodimos and colleagues (42) isolated *P. aeruginosa* from hydrotherapy pools that have only shown 20% resistance to these antibiotics. It is generally accepted that the environmental strains of *Pseudomonas* show very little resistance to antibiotics however this assumption has not been fully verified. One major source of antibiotic resistant *P. aeruginosa* in the environment is the untreated hospital wastewater (UHW). It has been shown that *P. aeruginosa* strains isolated from UHW are commonly resistant to a number of antibiotics (3, 143). For instance, Fentefria and colleagues (143) compared *P. aeruginosa* strains isolated from UHW and surface waters and showed a higher prevalence of MDR *P. aeruginosa* in the UHW than the surface water.

P. aeruginosa is also an inhabitant of soil that can be pathogenic to plants (144). Pitondo-Silva and colleagues (145) isolated *P. aeruginosa* from soil of different crops. These researchers found the majority of isolates to be resistant to aztreonam and Ticarcillin that are commonly used in the treatment of *P. aeruginosa* in CF patients (145). Thus, Wolfgang *et al.* (93) compared the genome content of *P. aeruginosa* isolates from clinical and environment sources' and concluded that gene conservation exists for *P. aeruginosa*.

2.12. Genetic diversity of *P. aeruginosa*

The complete genome of *P. aeruginosa* has been sequenced by Stover *et al* (147), reporting a genome size of 6, 262, 403 base pairs, which suggests that *P. aeruginosa* has the largest genomes amongst bacteria, with an estimated 5570 open reading frames (ORF). Of these ORF, 372 have been defined as functional genes, encoding proteins involved in cell adhesion and motility (e.g. type IV pili and exopolysaccharides), virulence factors (e.g. exoenzymes and the type III secretion system), LPS synthesis enzymes and other secreted proteins involved in the pathogenesis as described above. Other genes expressed in *P. aeruginosa*, are encoded for regulatory networks and outer membrane proteins (e.g. OprD porin family) and efflux systems for antibiotic resistance. Current literature suggests that the large genome size and genome complexity are responsible for the ability of this bacterium to adapt and thrive in a diverse range of environments (93). This high diversity has also resulted in the presence of large clonal groups of these bacteria in the environment.

Various genotypic typing methods are commonly used to identify persistent clones of these bacteria in clinical settings or the environment. Tielens and colleagues (148) have shown that *P. aeruginosa* strains isolated from UTI and catheter-associated UTIs are highly heterogeneous. Interestingly, these researchers found that some of their strains were closely related to *P. aeruginosa* clone C; which is a worldwide clone frequently isolated from the lung of CF patients (148). In a recent study, Naicker (135) examined the prevalence of Gram-

negative bacteria including *P. aeruginosa* in UHWW and their transition to a receiving STP and survival through its treatment processes. The results showed *P. aeruginosa* in UHWW were genetically distinct, although some strains were present at different times in the STP. Despite these findings, some researchers have observed that environmental strains of *P. aeruginosa* are genotypically and functionally equivalent to those isolated from clinical infections (7, 93, 147).

3. Conclusion

Hospitals and healthcare settings are regarded as reservoirs for large numbers of pathogenic *Pseudomonas* strains. Wastewaters from hospitals may contain a large number of these bacteria some of which can also be MDR. Recent studies on the prevalence of *P. Aeruginosa* in UHWW and their dissemination in the environment suggest that certain clonal groups of these bacteria have the ability to survive transmission to the STPs and then through to the finally treated effluent before being released into surface waters. The presence and persistence of these bacteria in environmental waters may pose a great risk to the public health and requires further work to fully characterize and quantify the input of MDR *P. Aeruginosa* strains from the hospitals compared with those originating from the general community or other wastewater- related sources.

Conflict of Interests

The authors declare that there is no conflict of interests with the organization that sponsored this research and publications arising from this research.

Acknowledgments

None to declare.

Authors' Contributions

Klriisa Streeter contributed to 60% and Mohammad Katouli to 40% of study.

Funding/Support

None to declare

References

- Peix A, Ramirez-Bahena MH, Velazquez. Historical evolution and current status of the taxonomy of genus *Pseudomonas*. *Infect Genet Evol.* 2009; 9(6):1132-47.
- Spiers AJ, Buckling A, Rainey PB. The causes of *Pseudomonas* diversity. *Microbiology.* 2000; 146(10): 2345-50.
- Meyer JM. Pyoverdines: pigments, siderophores and potential taxonomic markers of fluorescent *Pseudomonas* species. *Arch Microbiol.* 2000; 174(3):135-42.
- Lamont IL, Martin LW. Identification and characterization of novel pyoverdine synthesis genes in *Pseudomonas aeruginosa*. *Microbiology.* 2003; 149(4): 833-42.
- Frank LH, DeMoss RD. On the biosynthesis of pyocyanine. *J Bacteriol.* 1959; 77(6):776-82.
- Young G. Pigment production and antibiotic activity in cultures of *Pseudomonas aeruginosa*. *J Bacteriol.* 1947; 54(2):109-17.
- Coggan KA, Wolfgang MC. Global regulatory pathways and cross-talk control *Pseudomonas aeruginosa* environmental lifestyle and virulence phenotype. *Curr Issues Mol Biol.* 2012; 14(2):47-70.
- Pirnay JP, Matthijs S, Colak H, Chablain P, Bilocq F, Van Eldere J, et al. Global *Pseudomonas aeruginosa* biodiversity as reflected in a Belgian river. *Environ Microbiol.* 2005; 7(7):969-80.
- Defez C, Fabbro-Peray P, Bouziges N, Gouby A, Mahamat A, Daures JP, et al. Risk factors for multidrug-resistant *Pseudomonas aeruginosa* nosocomial infection. *J Hosp Infect.* 2004; 57(3):209-16.
- Ferroni A, Nguyen L, Pron B, Quesne G, Brusset MC, Berche P. Outbreak of nosocomial urinary tract infections due to *Pseudomonas aeruginosa* in a paediatric surgical unit associated with tap-water contamination. *J Hosp Infect.* 1998; 39(4):301-7.
- Riou M, Carbonnelle S, Avrain L, Mesaros N, Pirnay JP, Bilocq F, et al. In vivo development of antimicrobial resistance in *Pseudomonas aeruginosa* strains isolated from the lower respiratory tract of intensive care unit patients with nosocomial pneumonia and receiving antipseudomonal therapy. *Int J Antimicrob Agents.* 2010; 36(6):513-22.
- Brown SP, Cornfort DM, Mideo N. Evolution of virulence in opportunistic pathogens: generalism, plasticity and control. *Trends Microbiol.* 2012; 20(7):336-42.
- Engel J, Balachandran P. Role of *Pseudomonas aeruginosa* type III effectors in disease. *Curr Opin Microbiol.* 2008; 12(1):61-6.
- Bodey GP. Microbiologic aspects in patients with leukaemia. *Human Pathol.* 1974; 5(6):687-98.
- Koch C, Hoiby N. Pathogenesis of cystic fibrosis. *Lancet.* 1993; 341(8852):1065-9.
- Donlan RM. Biofilms and device-associated infections. *Emerg Infect Dis.* 2001; 7(2):277-81.
- Morris NS, Stickler DJ, McLean RJ. The development of bacterial biofilms on indwelling urethral catheters. *World J Urol.* 1999; 17(6):345-50.
- Huhlescu S, Simon M, Lubnow M, Kaase M, Wewalka G, Pietzka AT, et al. Fatal *Pseudomonas aeruginosa* pneumonia in a previously healthy woman was most likely associated with a contaminated hot tub. *Infect.* 2001; 39(3):265-9.
- Hatchette TF, Gupta R, Marrie TJ. *Pseudomonas aeruginosa* community-acquired pneumonia in previously healthy adult: case report and review of the literature. *Clin Infect Dis.* 2000; 31(6):1349-56.
- Nixon GM, Armstrong DS, Carzino R, Carlin JB, Olinsky A, Robertson CF, et al. Clinical outcome after early *Pseudomonas aeruginosa* infection in cystic fibrosis. *J Paediatr.* 2001; 138(5):699-704.
- Smith JJ, Travis SM, Greenberg EP, Welsh MJ. Cystic fibrosis airway epithelial fail to kill bacteria of abnormal airway surface fluid. *Cell.* 1996; 85(2):229-36.
- Schwiebert EM, Benos DJ, Egan ME, Stutts MJ, Guggion WB. CFTR is a conductance regulator as well as a chloride channel. *Physiol Rev.* 1999; 79(Supp 1):145-66.
- Sheppard DN, Welsh MJ. Structure and function of the CFTR chloride channel. *Physiol Rev.* 1999; 79(1):23-45.
- Terheggen-Lagro SWJ, Rijkers GT, Van der Ent CK. The role of airway epithelium and blood neutrophils in the inflammatory response in cystic fibrosis. *J Cyst Fibros.* 2005; 4(2):15-23.
- Worlitzch D, Tarran R, Ulrich M, Schwab U, Cekici A, Meyer KC, et al. Effects of reduced mucus oxygen concentration in airway *Pseudomonas* infections of cystic fibrosis patients. *J Clin Invest.* 2002; 109(3):317-25.
- Martinez-Solano L, Macia MD, Fajardo A, Oliver A, Martinez JL. Chronic *Pseudomonas aeruginosa* in chronic obstructive pulmonary disease. *Clin Infect Dis.* 2008; 47(12):1526-33.
- Fujitani S, Sun HY, Yu VL, Weingarten JA. Pneumonia due to *Pseudomonas aeruginosa*: part 1: epidemiology, clinical diagnosis and source. *Chest.* 2011; 139(4):909-19.
- Bergmans D, Bonten M, van Tiel F, Gaillard C, van der Geest S, Wilting R, et al. Cross-colonisation with *Pseudomonas aeruginosa* of patients in an intensive care unit. *Thorax.* 1996; 53(12):1053-8.
- Parker CM, Kutsogiannis J, Muscedere J, Cook D, Dodek P, Day AG, et al. Ventilator-associated pneumonia caused by multidrug-resistant organisms or *Pseudomonas aeruginosa*: prevalence, incidence, risk factors and outcomes. *J Crit Care.* 23(1):18-26.
- Inglis TJJ. Evidence for dynamic phenomena in residual tracheal tube biofilm. *Br J Anaesth.* 1993; 70(1):22-4.
- Mittal R, Aggarwal S, Sharma S, Chhibber S, Harjai K. Urinary tract infections caused by *Pseudomonas aeruginosa*: a mini review. *J Infect Public Health.* 2009; 2(3):101-11.
- Maki DG, Tambyah PA. Engineering out the risk for infection with urinary catheters. *Emerg Infect Dis.* 20017(2):342-7.
- Ressner RAMC, Griffith ME, Rasnake MS, Hospental DR, Wolf SE. Outcomes of bacteremia in burn patients involved in combat operations overseas. *J Am Coll Surg.* 206(3):439-44.
- Estahbanati HK, Kashani PP, Ghanaatpisheh F. Frequency of *Pseudomonas aeruginosa* serotypes in burn wound infections and their resistance to antibiotics. *Burns.* 2002; 28(4):637-41.
- Armour AD, Shankowsky HA, Swanson T, Lee J, Tredget EE. The impact of nosocomially-acquired resistant *Pseudomonas aeruginosa* infection in a burn unit. *J Trauma.* 2007; 63(1):164-71.
- Ranjjan KP, Ranjan N, Bansal SK, Arora DR. Prevalence of *Pseudomonas aeruginosa* in post-operative wound infection in a referral hospital in Haryana, India. *J Lab Physicians.* 2010; 2(2):74-7.
- Jombo GT, Akpan S, Epoke J, DenenAkaa P, Odey F. Multi-drug resistant *Pseudomonas aeruginosa* infections complicating surgical wounds and the potential challenges in managing post-operative wound infections: University of Calabar Teaching Hospital experience. *Asian J Trop Med.* 2010; 3(6):479-82.
- Gang RK, Bang RL, Sanyal SC, Mokaddas E, Lari AR. *Pseudomonas aeruginosa* septicemia in burns. *Burns.* 1999; 25(7):611-6.
- Patel BM, Paratz JD, Mallet A, Lipman J, Rudd M, Muller MJ, et al. Characteristics of bloodstream infections in burn patients: an 11-year retrospective study. *Burns.* 2012; 38(5):685-90.

40. Guida M, Galle F, Mattei ML, Anastasi D, Liquori G. Microbiological quality of the water of recreational and rehabilitation pools: a 2-year survey in Naples, Italy. *Public Health*. 2009; 123(6):448-451.
41. Moore JE, Heaney N, Millar BC, Crowe M, Elborn JS. Incidence of *Pseudomonas aeruginosa* in recreational and hydrotherapy pools. *Commun Dis Public Health*. 2002; 5(1): 23-6.
42. Tirodimos I, Arvanitidou M, Daravessis L, Bisiklis A, Alexiou-Daneel S. Prevalence and antibiotic resistance of *Pseudomonas aeruginosa* isolated from swimming pools in northern Greece. *Health J*. 2010; 16(7):783-7.
43. Ratnam S, Hogan K, March SB, Butler RW. Whirl pool-associated folliculitis is caused by *Pseudomonas aeruginosa*: report of an outbreak and review. *J Clin Microbiol*. 1986; 23(3):655-9.
44. Hopkins RS, Abbott DO, Wallace LE. Follicular dermatitis outbreak caused by *Pseudomonas aeruginosa* associated with a motel's indoor swimming pool. *Public Health Rep*. 1981; 96(3):246-9.
45. Daniel CR, Iorizzo M, Piraccini BM, Tosti A. Simple onycholysis. *Cutis*. 2011; 87(5):226-8.
46. Hengge UR, Bardeli V. Images in clinical medicine. Green nails. *N Engl J Med*. 2009; 360(11):1125.
47. McNeil SA, Nordstrom-Lerner L, Malani PN, Zervos M, Kauffman CA. Outbreak of sternal surgical site infections due to *Pseudomonas aeruginosa* traced to a scrub nurse with onychomycosis. *Clin Infect Dis*. 2001; 33(3):317-23.
48. Nagachandrika T, Kumbar U, Dumpatic S, Chary S, Mandatharac PS, Rathni VM. Prevalence of contact lens related complications in a tertiary eye centre in India. *Cont Lens Anterior Eye*. 2011; 34(6):266-8.
49. Ramphal R, McNiece MT, Polack FM. Adherence of *Pseudomonas aeruginosa* to the injured cornea: a step in the pathogenesis of corneal infections. *Ann Ophthalmol*. 1981;13(4): 421-5.
50. Stern GA, Lubniewski A, Allen C. The interaction between *Pseudomonas aeruginosa* and the corneal epithelium. *Arch Ophthalmol*. 1985; 103(8):1221-5.
51. Roberston DM, Petroll WM, Jester JV, Cavanagh HD. Current concepts: contact lens related *Pseudomonas* keratitis. *Cont Lens Anterior Eye*. 2007; 30(2):94-107.
52. Yeung KK, Forister JFY, Forister EF, Chung MY, Han S, Weissman BA. Compliance with soft contact lens replacement schedules and associated contact lens-related ocular complications: the UCLA contact lens study. *Optom*. 2010; 81(11):598-607.
53. Wang MC, Liu CY, Shiao AS, Wang T. Ear problems in swimmers. *J Chin Med Assoc*. 2005; 68(8):347-52.
54. Nussinovitch M, Rimón A, Volovitz B, Raveh E, Prais D, Amir J. Cotton-tip applicators as a leading cause of otitis externa. *Int J Pediatr Otorhinolaryngol*. 2004; 73(4):168-72.
55. Ninkovic G, Dullo V, Saunders NC. Microbiology of otitis externa in the secondary care in United Kingdom and antimicrobial sensitivity. *Auris Nasus Larynx*. 2008; 35(4):480-4.
56. De Kievit TR, Iglewski BH. Bacterial quorum sensing in pathogenic relationships. *Infect Immun*. 2000; 68(9):4839-49.
57. Smith RS, Iglewski BH. *Pseudomonas aeruginosa* quorum sensing as a potential antimicrobial agent. *J Clin Invest*. 2003; 112(10):1460-5.
58. O'Loughlin CT, Miller LC, Drescher K, Semmelhack MF, Bassler BL. A quorum-sensing inhibitor blocks *Pseudomonas aeruginosa* virulence and biofilm formation. *Proc Natl Acad Sci USA*. 2013; 110(44):17981-6.
59. Pizzaro-Cerda J. Bacterial adhesion and entry into host cells. *Cell*. 2006; 124(4):715-27.
60. Bertrand JJ, West JT, Engel JN. Genetic analysis of the regulation of type IV pilus function by the cAMP chemosensory system of *Pseudomonas aeruginosa*. *J Bacteriol*. 2010; 192(4):994-1010.
61. Hahn HP. The type-4 pilus is the major virulence-associated adhesion of *Pseudomonas aeruginosa* – a review. *Gene*. 1997; 192(1):99-108.
62. Burrows LL. *Pseudomonas aeruginosa* twitching motility: type IV pili in action. *Annu Rev Microbiol*. 2012; 66: 493-520.
63. Zolfaghar I, Evans DJ, Fleiszig MJ. Twitching motility contributes to the role of pili in corneal infection caused by *Pseudomonas aeruginosa*. *Infect Immun*. 2003; 71(9):5389-93.
64. Mattick JS. Type IV pili and twitching motility. *Annu Rev Microbiol*. 2002; 56:289-314.
65. Nivens DE, Ohman DE, Williams J, Franklin MJ. Role of alginate and its O-acetylation in formation of *Pseudomonas aeruginosa* microcolonies and biofilms. *J Bacteriol*. 2001; 183(3):1047-57.
66. Lam J, Chan R, Lam K, Costerton JW. Productions of mucoid microcolonies by *Pseudomonas aeruginosa* within infected lungs in cystic fibrosis. *Infect Immun*. 1980; 28(2):546-56.
67. Colvin KM, Irie Y, Tart CS, Urbano R, Whitney JC, Ryder C, et al. The pel and psl polysaccharides provide *Pseudomonas aeruginosa* structural redundancy within the biofilm matrix. *Environ Microbiol*. 2012; 14(8):1913-28.
68. Byrd MS, Pang B, Mishra M, Swords WE, Wozniak DJ. The *Pseudomonas aeruginosa* exopolysaccharide psl facilitates surface adherence and NF-kappaB activation in A459 cells. *MBio*. 2010; 1(3):e00140-10.
69. Noiby N, Ciofalo O, Johansen HK, Song ZJ, Moser C, Jensen PO, et al. The clinical impact of bacterial biofilms. *Int J Oral Sci*. 2011; 3(2):55-65.
70. Pritt B, O'Brien L, Winn W. Mucoid *Pseudomonas* in cystic fibrosis. *Am J Clin Pathol*. 2007; 128(1):32-34.
71. Bayer AS, Speert DP, Park S, Tu J, Witt M, Nast CC, et al. Functional role of mucoid exopolysaccharide (alginate) in antibiotic-induced and polymorphonuclear leukocyte-mediated killing of *Pseudomonas aeruginosa*. *Infect Immun*. 1991; 59(1):302-8.
72. Schwarzmann S, Boring JR. Antiphagocytic effect of slime from a mucoid strain of *Pseudomonas aeruginosa*. *Infect Immun*. 1971; 3(6):762-7.
73. Simpson JA, Smith SE, Dean RT. Alginate inhibition of the uptake of *Pseudomonas aeruginosa* by macrophages. *J Gen Microbiol*. 1988; 134(1):29-36.
74. Wozniak DJ, Wyckoff TJO, Starkey M, Keyser R, Azadi P, O'Toole GA, et al. Alginate is not a significant component of the extracellular polysaccharide matrix of PA14 and PA01 *Pseudomonas aeruginosa* biofilms. *Proc Natl Acad Sci USA*. 2003; 100(13):7907-12.
75. Stapper AP, Narasimhan G, Ohman DE, Barakat J, Hentzer M, Molin S, et al. Alginate production affects *Pseudomonas aeruginosa* biofilm development and architecture, but it not essential for biofilm formation. *J Med Microbiol*. 2004; 53(7):679-90.
76. Ma L, Jackson KD, Landry RM, Parsek MR, Wozniak DJ. Analysis of *Pseudomonas aeruginosa* conditional psl variants reveals roles for the prl polysaccharide in adhesion and maintaining biofilm structure post attachment. *J Bacteriol*. 2006; 188(23):8213-21.
77. Mittal R, Aggarwal S, Sharma S, Chhibber S, Harjai K. Urinary tract infections caused by *Pseudomonas aeruginosa*: a mini review. *J Infect Public Health*. 2009; 2(3):101-11.
78. Pier BP. *Pseudomonas aeruginosa* lipopolysaccharide: a major virulence factor, initiator of inflammation and target for effective immunity. *Int J Med Microbiol*. 2007; 297(5):277-95.
79. Villar J, Maca-Meyer N, Perez-Mendez L, Flores C. Bench-to-bedside review: understanding genetic predisposition to sepsis. *Crit Care*. 2004; 8(3):180-9.
80. Backhed F, Normark S, Schmeda EKH, Oscarson S. Structural requirements for TLR4-mediated LPS signalling: a biological role for LPS modifications. *Microbes Infect*. 2003; 5(12):1057-63.
81. Evans DJ, Pier GB, Coyne MJ, Goldberg JB. The *rfb* locus from *Pseudomonas aeruginosa* strain PA103 promotes the expression of O antigen by both LPS-rough and LPS-smooth isolates from cystic fibrosis patients. *Mol Microbiol*. 2006; 13(3):427-34.
82. Hancock RE, Mutharia LM, Darveau RP, Speert DP, Pier BG. *Pseudomonas aeruginosa* isolates from patients with cystic fibrosis: a class of serum-sensitive, nontypable strains deficient in lipopolysaccharide O side chains. *Infect Immun*. 1983; 42(1):170-7.
83. Hauser AR, Engel JN. *Pseudomonas aeruginosa* induces type-III-secretion-mediated apoptosis of macrophages and epithelial cells. *Infect Immun*. 1999; 67(10):5530-7.
84. Galle M, Jin S, Bogaert P, Haegem M, Vandenaebel P, Beyaert R. The *Pseudomonas aeruginosa* type III secretion system has an exotoxin s/t/y independent during acute lung infection. *PLoS One*. 2012; 7(7):e41547.
85. Atkories K, Barbieri JT. Bacterial cytotoxins: targeting eukaryotic switches. *Nat Rev Microbiol*. 2005; 3(5):397-410.
86. Kaufman MR, Jia J, Zeng L, Ha U, Chow M, Jin S. *Pseudomonas aeruginosa* mediated apoptosis requires the ADP-ribosylating activity of exoS. *Microbiol*. 2000; 146(Pt10):2531-41.
87. Hritonenko V, Mun JJ, Connie T, Simon NC, Barbieri JT, Evans DJ, et al. Adenylate cyclase activity of *Pseudomonas aeruginosa* exoY can mediate bleb-niche formation in epithelial cells and contributes to virulence. *Microb Pathog*. 2011; 51(5):305-12.
88. Prasain N, Alexeyev M, Balczon R, Stevens T. Soluble adenylate cyclase-dependent microtubule disassembly reveals a novel mechanism of endothelial cell retraction. *Am J Physiol Lung Cell Mol Physiol*. 2009; 297(1):73-83.
89. Finck-Barbancon V, Goranson J, Zhu L, Sawa T, Wiener-Kronish JP, Fleiszig SMJ, et al. ExoU expression by *Pseudomonas aeruginosa* correlates with acute cytotoxicity and epithelial injury. *Mol Microbiol*. 1997; 25(3):547-57.
90. Sato H, Frank DW, Hillard CJ, Feix JB, Pankhaniya RR, Moriyama K, et al. The mechanism of action of the *Pseudomonas aeruginosa*-encoded type III cytotoxin, exoU. *EMBO J*. 2003; 22(12):2959-69.
91. Plotkowski MC, Feliciano LFP, Machado GBS, Cunha LG, Freitas C, Saliba AM, et al. ExoU-induced procoagulant activity in *Pseudomonas aeruginosa*-infected airway cells. *Eur Respir J*. 2008; 32(6):1591-8.
92. Feltman H, Schult G, Khan S, Jain M, Peterson L, Hauser AR. Prevalence of type III secretion genes in clinical and environmental isolates of *Pseudomonas aeruginosa*. *Microbiology*. 2001; 147(Pt 10):2659-69.
93. Wolfgang MC, Kulasekara BR, Liang X, Boyd D, Yang Q, Miyada CG, et al. Conservation of genome content and virulence determinants among clinical and environmental isolates of *Pseudomonas aeruginosa*. *Proc Natl Acad Sci USA*. 2003; 100(14):8484-9.
94. Mitov I, Strateva T, Markova B. Prevalence of virulence genes among Bulgarian nosocomial and cystic fibrosis isolates of *Pseudomonas aeruginosa*. *Braz J Microbiol*. 2010; 41(3):588-95.

95. Schuler G, Feltman H, Rabin SDP, Martin CG, Battle SE, Rello J, et al. Secretion of the toxin exoU is a marker for highly virulent *Pseudomonas aeruginosa* isolates obtained from patients with hospital-acquired pneumonia. *J Infect Dis.* 2003; 188(11):1695-706.
96. Kurahashi K, Kajikawa O, Sawa T, Ohara M, Frank DW, Martin TR, et al. Pathogenesis of septic shock in *Pseudomonas aeruginosa* pneumonia. *J Clin Invest.* 1999; 104(6):743-50.
97. Finnan S, Morrissey JP, O'Garra F, Boyd EF. Genome diversity of *Pseudomonas aeruginosa* isolates from cystic fibrosis patients and the hospital environment. *J Clin Microbiol.* 2004; 42(12):5783-92.
98. Shaver CM, Hauser AR. Relative contributions of *Pseudomonas aeruginosa* ExoU, ExoS, and ExoT to virulence in the lung. *Infect Immun.* 2004; 72(12):6969-77.
99. Dacheux D, Toussaint B, Richard M, Brochier G, Croize J, Attree I. *Pseudomonas aeruginosa* cystic fibrosis isolates induce rapid type III secretion-dependent, but ExoU-independent, oncosis of macrophages and polymorphonuclear neutrophils. *Infect Immun.* 2000; 68(5):2916-24.
100. Berthelot P, Attree I, Plesiat P, Chabert J, de Bentzmann S, Pozzetto B, et al. Genotypic and phenotypic analysis of type III secretion system in a cohort of *Pseudomonas aeruginosa* bacteremia isolates: evidence for a possible association between O serotypes and *exo* genes. *J Infect Dis.* 2003; 188(4):512-8.
101. Jackson AA, Gross MJ, Daniels EF, Hampton TH, Hammons JH, Vallet-Gely I, et al. Anr and its activation by PlcH activity in *Pseudomonas aeruginosa* host colonization and virulence. *J Bacteriol.* 2013; 195(13):3093-104.
102. Meyers DJ, Berk RS. Characterization of phospholipase C from *Pseudomonas aeruginosa* as a potent inflammatory agent. *Infect Immun.* 1990; 58(3):659-66.
103. Heck LW, Morihara K, Abrahamson DR. Degradation of soluble laminin and depletion of tissue-associated basement membrane laminin by *Pseudomonas aeruginosa* elastase and alkaline protease. *Infect Immun.* 1986; 54(1):149-53.
104. Leduc D, Beaufort N, de Bentzmann S, Rousselle JC, Namane A, Chingnard M, et al. The *Pseudomonas aeruginosa* lasB metalloproteinase regulates the human urokinase-type plasminogen activator receptor through domain-specific endoproteolysis. *Infect Immun.* 2007; 75(8):3848-58.
105. Theander TG, Kharazmi A, Pedersen BK, Christensen LD, Tvede N, Poulsen LK, et al. Inhibition of human lymphocyte proliferation and cleavage of interleukin-2 by *Pseudomonas aeruginosa* proteases. *Infect Immun.* 1988; 56(7):1673-7.
106. Bradbury RS, Roddam LF, Merritt A, Reid DW, Champion AC. Virulence gene distribution in clinical, nosocomial and environmental isolates of *Pseudomonas aeruginosa*. *J Med Microbiol.* 2010; 59(Pt 8):881-90.
107. Martins VV, Pitondo-Silva A, Manco LDM, Falcao JF, Freitas SDS, Silveira WDD, et al. Pathogenic potential and genetic diversity of environmental and clinical isolates of *Pseudomonas aeruginosa*. *APMIS.* 2014; 122(2):92-100.
108. Lanotte P, Watt S, Mereghetti L, Dartiguelongue N, Rastegar-Lari A, Goundeau A, et al. Genetic features of *Pseudomonas aeruginosa* isolates from cystic fibrosis patients compared with those of isolates from other origins. *J Med Microbiol.* 2004; 53(Pt 1):73-81.
109. Lamont IL, Martin LW. Identification and characterization of novel pyoverdine synthesis genes in *Pseudomonas aeruginosa*. *Microbiology.* 2003; 149(Pt 4):833-42.
110. Meyer JM, Neely A, Stintzi A, Georges C, Holder IA. Pyoverdine is essential for virulence of *Pseudomonas aeruginosa*. *Infect Immun.* 1996; 64(2):518-23.
111. Ho Sui SJ, Lo R, Fernandes AR, Caulfield MDG, Lerman JA, Xie L, et al. Raloxifene attenuates *Pseudomonas aeruginosa* pyocyanin production and virulence. *Int J Antimicrob Agents.* 2012; 40(3):246-51.
112. Jimenez PN, Koch G, Thompson JA, Xavier KB, Cool RH, Quax WJ. The multiple signalling systems regulating virulence in *Pseudomonas aeruginosa*. *Microbiol Mol Biol Rev.* 2012; 76(1):46-65.
113. Aloush V, Navon-Venezia S, Seigmen-Igra Y, Cabili S, Carmeli Y. Multi-drug resistant *Pseudomonas aeruginosa*: risk factors and clinical impact. *Antimicrob Agents Chemother.* 2006; 50(1):43-8.
114. Schechner V, Gottersman T, Schwartz O, Korem M, Maor Y, Rahav G, et al. *Pseudomonas aeruginosa* bacteremia upon admission: risk factors for mortality and influence of inadequate empirical antimicrobial therapy. *Diagn Microbiol Infect Dis.* 2001; 71(1):38-45.
115. Lambert PA. Mechanisms of antibiotic resistance in *Pseudomonas aeruginosa*. *J R Soc Med.* 2002; 95(suppl 41):22-6.
116. Bredenstein EBM, de la Fuente-Nunez C, Hancock REW. *Pseudomonas aeruginosa*: all roads lead to resistance. *Trends Microbiol.* 2011; 19(8):419-25.
117. Carmeli Y, Troillet N, Eliopoulos GM, Samore MH. Emergence of antibiotic-resistant *Pseudomonas aeruginosa*: comparison of risks associated with different antipseudomonal agents. *Antimicrob Agents Chemother.* 1999; 43(6):1379-82.
118. Messadi AA, Lamia T, Kamel B, Salima Q, Monia M, Said BR. Association between antibiotic use and changes in susceptibility patterns of *Pseudomonas aeruginosa* in an intensive care burn unit: a 5-year study, 2000-2004. *Burns.* 2008; 34(8):1098-102.
119. Allen HK, Donato J, Huimi Wang H, Cloud-Hansen KA, Davies J, Handelsman J. Call of the wild: antibiotic genes in natural environments. *Nat Rev Microbiol.* 2010; 8(4):251-59.
120. Majiduddin FK, Materon IC, Palzkill TG. Molecular analysis of beta-lactamase structure and function. *Int J Med Microbiol.* 2002; 292(2):127-37.
121. Walsh TR, Toleman MA, Poirel L, Nordmann P. Metallo-beta-lactamases: the quiet before the storm? *Clin Microbiol Rev.* 2005; 18(2):306-25.
122. Marra AR, Pereira CAP, Gales AG, Menezes LC, Cal RGR, de Souza JMA, et al. Bloodstream infections with metallo-beta-lactamase-producing *Pseudomonas aeruginosa*: epidemiology, microbiology, and clinical outcomes. *Antimicrob. Agents Chemother.* 2006; 50(1):388-90.
123. Juan C, Macia MD, Gutierrez O, Vidal C, Perez JL, Oliver A. Molecular mechanisms of beta-lactam resistance mediated by AmpC hyper production in *Pseudomonas aeruginosa* clinical strains. *Antimicrob Agents Chemother.* 2005; 49(11):4733-8.
124. Gales AC, Menezes LC, Silbert S, Sader HS. Dissemination in distinct Brazilian regions of an epidemic carbapenem-resistant *Pseudomonas aeruginosa* producing SPM metallo-beta-lactamase. *J Antimicrob Chemother.* 2003; 52(4):699-702.
125. Crespo MP, Woodford N, Sinclair A, Kaufmann ME, Turton J, Glover J, et al. Outbreak of carbapenem-resistant *Pseudomonas aeruginosa* producing vim-8, a novel metallo-beta-lactamase, in a tertiary centre in Cali, Colombia. *J Clin Microbiol.* 2004; 42(11):5094-101.
126. Vitkauskienė A, Skrodenienė E, Dambrauskienė A, Bakšyte G, Macas A, Sakalauskas R. Characteristics of carbapenem-resistant *Pseudomonas aeruginosa* strains in patients with ventilator-associated pneumonia in intensive care units. *Medicina (Kaunas).* 2011; 47(12):652-6.
127. Li H, Luo F, Williams BJ, Blackwell TS, Xie CM. Structure and function of oprD protein in *Pseudomonas aeruginosa*: from antibiotic resistance to novel therapies. *Int J Med Microbiol.* 2012; 302(2):63-8.
128. Nakano M, Deguchi T, Kawamura T, Yasuda M, Kimura M, Okano Y, et al. Mutations in the *gyrA* and *parC* genes in fluoroquinolone-resistant clinical isolates of *Pseudomonas aeruginosa*. *Antimicrob Agents Chemother.* 1997; 41(10):2289-91.
129. Yonezawa M, Takahata M, Matsubara N, Watanabe Y, Narita H. DNA gyrase *gyrA* mutations in quinolone-resistant clinical isolates of *Pseudomonas aeruginosa*. *Antimicrob Agents Chemother.* 1995; 39(9):1970-2.
130. Salma RM, Dabboussi FA, Kassaa IM, Khudary RH, Hamze MM. *gyrA* and *parC* mutations in quinolone-resistant clinical isolates of *Pseudomonas aeruginosa* from Nini hospital in North Lebanon. *J Infect Chemother.* 19(1):77-81.
131. Li XZ, Nikaïdo H, Poole K. Role of mexA-mexB-oprM in antibiotic efflux in *Pseudomonas aeruginosa*. *Antimicrob Agents Chemother.* 1995; 39(9):1948-53.
132. Sadeghifard N, Valizadeh A, Zolfaghary MZ, Maleki MH, Maleki A, Mohebi R, et al. Relationship between the presence of the nalC mutation and multidrug resistance in *Pseudomonas aeruginosa*. *Int J Microbiol.* 2012; 2012:575193.
133. Poole K. Multidrug efflux pumps and antimicrobial resistance in *Pseudomonas aeruginosa* and related organisms. *J Mol Microbiol Biotechnol.* 2001; 3(2):255-64.
134. Thompson JM, Gundogdu A, Stratton HM, Katouli M. Antibiotic resistant *Staphylococcus aureus* in hospital wastewaters and sewage treatment plants with special reference to methicillin-resistant *Staphylococcus aureus* (MRSA). *J Appl Microbiol.* 2013; 114(1):44-54.
135. Naicker S. 2012. Extended spectrum beta-lactamase producing Gram-negative strains in untreated hospital wastewater and their survival in sewage treatment plants. Honours thesis. University of the Sunshine Coast, Queensland, Australia.
136. Guida M, Galle F, Mattei ML, Anastasi D, Liquori G. Microbiological quality of the water of recreational and rehabilitation pools: a 2-year survey in Naples, Italy. *Public Health.* 2009; 123(6):448-51.
137. Grobe S, Wingender J, Flemming HC. Capability of mucoid *Pseudomonas aeruginosa* to survive in chlorinated water. *Int J Hyg Environ Health.* 2001; 204(2-3):139-142.
138. Moore JE, Heaney N, Millar BC, Crowe M, Elborn JS. Incidence of *Pseudomonas aeruginosa* in recreational and hydrotherapy pools. *Commun Dis Public Health.* 2002; 5(1):23-6.
139. Ratnam S, Hogan K, March SB, Butler RW. Whirlpool-associated folliculitis caused by *Pseudomonas aeruginosa*: report of an outbreak and review. *J Clin Microbiol.* 1986; 23(3):655-9.
140. Salmen P, Dwyer DM, Vorse H, Kruse W. Whirlpool-associated *Pseudomonas aeruginosa* urinary tract infections. *JAMA.* 1983; 250(15):2025-6.
141. Hajjartabar M. Poor-quality water in swimming pools associated with substantial risk of otitis externa due to *Pseudomonas aeruginosa*. *Water Sci Technol.* 2004; 50(1):63-7.
142. Lutz JK, Lee J. Prevalence of antimicrobial-resistance of *Pseudomonas aeruginosa* in swimming pools and hot tubs. *Int J Environ Res Public Health.* 2011; 8(2):554-64.
143. Fuentefria DB, Ferreira AE, Graf T, Corcao G. *Pseudomonas aeruginosa*: spread of antimicrobial resistance in hospital effluent and surface water. *Rev Soc Bras Med Trop.* 2008; 41(5):470-3.

144. Plotnikova JM, Rahme LG, Ausubel FM. Pathogenesis of the human opportunistic pathogen *Pseudomonas aeruginosa* PA14 in *Arabidopsis*. *Plant Physiol*. 2000; 124(4):1766-74.
145. Pitondo-Silva A, Martins VV, Fernandes AFT, Stehlin EG. High level of resistance to Aztreonam and Ticarcillin in *Pseudomonas aeruginosa* isolated from soil of different crops in Brazil. *Sci Total Environ*. 2014; 473-4:155-158.
146. Stover CK, Pham XQ, Erwin AL, Mizoguchi SD, Warrener P, Hickey MJ, et al. Complete genome sequence of *Pseudomonas aeruginosa* PA01, an opportunistic pathogen. *Nature*. 2000; 406(6799):959-64.
147. Alonso A, Rojo F, Martinez JL. Environmental and clinical isolates of *Pseudomonas aeruginosa* show pathogenic and biodegradative properties irrespective of their origin. 1999; 1(5):421-30.
148. Tielen P, Narten M, Rosin N, Biegler I, Haddad I, Hogardt M, et al. Genotypic and phenotypic characterization of *Pseudomonas aeruginosa* isolates from urinary tract infections. *Int J Med Microbiol*. 2010; 301(4):282-92.

How to cite this article: Streeter K, Katouli M. *Pseudomonas aeruginosa*: A review of their Pathogenesis and Prevalence in Clinical Settings and the Environment. *Infection Epidemiology and Medicine*. 2016; 2(1): 25-32.