Acinetobacter baumannii is classified by Infectious Diseases Society of America (IDSA) as one of the six main multidrug resistant (MDR) microbes in hospitals worldwide, so it causes a variety of diseases such as hospital and community acquired infection. Here, we investigate physical and biological methods to treat A. baumannii infections by using Cold Atmospheric Plasma (CAP) and polymyxin B (PMB), also we investigate the role of OmpA gene in adhesion and pathogenicity of A. baumannii. We used two strains of A. baumannii in this study, ATCC 17904 and HHR1; both strains were exposed to Dielectric Barrier Discharge- Cold Atmospheric Plasma (DBD-CAP) at 20 KV, after sterilization test, several methods were done to analyze the effect of DBD-CAP on bacterial morphology, proteins and DNA. After that, A. baumannii (ATCC 17904 & HHR1) were treated with PMB (1, 2.5, 5, 7.5, 10 and 20) µg ml-1 and then study the action of PMB on A. baumannii growth, motility and biofilm formation. As shown in the results that HHR1 is more resistance to DBD-CAP than ATCC 17904. At the first point, the growth of both strains was largely affected by plasma and this influence was increased by increasing the time of exposure, also the plasma affected the DNA especially OmpA gene and standard strain exhibit more sequence variations also this result proved the important of OmpA in pathogenicity of A. baumannii so when this gene affected by plasma the bacterial cell was destroyed. In addition, plasma also has been showed to damage proteins and morphology thus the bacterial cells was elongated and aggregated. The results also demonstrate that A. baumannii HHR1 is more resistance to PMB than standard strains except for biofilm formation where ATCC 17904 was more resistance than HHR1. Firstly, the effect of PMB on A. baumannii was increase by increase the Concentration and the time of incubation, and also PMB was shown to be deleterious on A. baumannii growth spatially at last concentration (20 µg ml-1). Furthermore, PMB affects motility and the expression level of OmpA gene, also this result proved the role of OmpA gene as regulator to other genes (adeB, bap, and blaPER-1) that participate in adhesion and antibiotic resistance.