Evaluation of Biofilm Formation Capacity of Acinetobacter baumannii Isolated from Clinical Samples in Baghdad Hospitals using Phenotypic Methods

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Abstract: During the period from November/2016 to March/2017, a total of 475 specimens from various clinical sources (wounds, burns, urine, sputum, blood and throat) were collected from patients suffering different infections from a number of hospitals in Baghdad city (Karkh and Resafa), as follows: (Al-Yarmuk, Al-Karama, Al-Karkh, Al- Kadimia, Martyr Gazi Al-Hariry, the Medical city, Al-Kindi Teaching, Al-Imam Ali, Ibn-Al Balady and Baghdad Teaching Hospitals). Acinetobacter baumannii isolates in selective (CHROMagar) and specific differential medium, microscopic features, biochemical tests and API20 NE and VITEK-2 system at probability 98%. Furthermore the identification of isolates depending of molecular method by using polymerase chain reaction (PCR) technique which is used to amplify specific gene by using special primers, to genus level was confirmed finally by detection of 16SrRNA gene while identification of A.baumannii isolates to species level was done by blaOXA-51-like gene, giving ratio 100%. The results showed that the 83 A.baumannii isolates were obtained from clinical specimens (17.47%), distributed according to the sources from highest to lowest percentage as follow in 31(25.20%) wound, 21(19.81%) burn, while 10(12.82%) urin, 8(11.94%) sputum, 7(12.72%) blood and low percentage 6(13.04%) from throat. This study focused on the phenotypic to determine methods the ability of biofilm production using three methods includes: Congo red agar (CRA), Tube method (TM) and microtiter plates (MTP). The results showed a significant differences between all 83 A.baumannii isolates. A seventy four isolates (89.15%) have the ability to adherence and produce slim layer with significant differences in thickness degrees (strong, moderate and weak). While 9 isolates which represented (10.84%) from isolates have no ability to adherence and produce slim layer. When comparing between these methods to detect strong biofilm production isolates, the results showed that MTP assay was excelled on CRA and TM at 58(69.87%), 48(57.83%) and 45(54.21%), respectively measured by optical densities values at 630 nm. Conclusion: The study concludes that there a positive correlation between biofilm formation and multi drug resistance A. baumannii. Each of the three phenotypic methods used for detection of biofilm formation has its advantages and disadvantages. But MTP method is most widely used and was considered as standard test for detection of biofilm formation.

Keywords: Acinetobacter baumannii, CHROMOagar acinetobacter, Biofilm Formation, phenotypic methods.

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Introduction: Acinetobacter baumannii considered as a typical opportunistic pathogen that almost exclusively affects predisposed individuals, and classified as a nosocomial pathogen involved in infections that mainly affect critically ill patients. On the other hand, in immune compromised patients several species,
in particular *A. baumannii* can cause severe, life threatening infections (1).

Another major risk factor for *A. baumannii* infections is the widespread use of broad-spectrum antibiotics (2).

One of the distinctiveness of *A. baumannii* which contributes to its extensive survival in the environment is its ability to biofilm formation, or communities of intercommunicate bacterial cells associated with a surface and encased in an extracellular matrix of carbohydrates, nucleic acids, proteins, and other macromolecules (3).

*A. baumannii* is can adhere and colonizer to different abiotic surfaces in the hospital environment, such as glass and plastic, keyboards, pillows, gloves, beds and medical equipment. Prolonged periods survival on dry surfaces and high levels of resistance contribute to persistence of *A. baumannii* and complicates eradication during outbreaks (4).

Hypothesized that biofilm development mechanisms in *A. baumannii* from three important factors which contribute to the perseverance of *A. baumannii* in the hospital environment specifically, resistance to major antimicrobial drugs, resistance to desiccation, resistance to disinfectants and disease generation is closely associated with its ability to form biofilm and colonize on biotic and abiotic surfaces (1).

Biofilm formation has also been correlated with MDR in clinical *A. baumannii* isolates it’s to increased antimicrobial resistance. Formation of a biofilm is commonly a characteristic of *A. baumannii* clinical isolates and the ability to form a biofilm has been linked to pathogenesis in this organism (5).

The present study aims at studying the capacity of *Acinetobacter baumannii* isolates for Biofilm Formation.

**Materials and methods:**

**Clinical specimens collection:**

During the period between from November 2016 to March 2017, a total of 475 clinical specimens were collected from patients hospitalized in Baghdad city (Karkh and Resafa).

Different sources collected from (Burns swab, Wounds swab, Sputum swab, Urine, Blood and Throat swab). All specimens were collected by using clean sterile containers and transport swabs damped with normal saline, and transported to the laboratory immediately. The time between samples collection and bacteriological exam never exceed 1-2 hrs.

**Isolation and Identification of *A. baumannii* isolates:**

Each specimen was streaked using direct method of inoculation on a selective media to investigated for the presence of *A. baumannii*. Typical *Acinetobacter CHROMagarTM Acinetobacter/ MDR* medium, MacConkey agar and blood agar and then incubated under aerobic conditions for 24 hrs at 37°C (6). Then confirmatory identification was reconfirmed of *A. baumannii* according to Macroscopic and Microscopic examination, biochemical tests, API20
NE kit, Vitek-2 system and genotyping by PCR were studies.

**Genotypic Techniques detection:**

**Extraction of Genomic DNA:**

Genomic DNA of *A.baumannii* clinical isolates was extracted by using Genomic DNA extraction kit a commercial purification system (Genomic DNA Minni Kit Geneaid, Thailand), and quantification of purity and concentration of genomic DNA using a spectrophotometrically (Nano-drop) instrument to measure the optical density (O.D), and stored at -20°C.

**Primers selection and Identification used PCR technique:**

In this study, monoplex PCR assay was performed as a confirmative test is used to amplify specific gene by using special primers, to genus level was confirmed finally by detection of *16SrRNA* gene while identification of *Acinetobacter baumannii* isolates to species level was done by *blaOXA-51*-like gene. This assay was done according to method described by (7), as in Table (1) and the programs for each gene that mention in Table (2). All PCR assays were accomplished in institute of genetic engineering and biotechnology.

### Table (1): Primers used in this study.

<table>
<thead>
<tr>
<th>Primer Name</th>
<th>Primer Sequence Oligo sequence F (5’….3’) Reverse</th>
<th>Product size (bp)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>16SrRNA</em></td>
<td>5’- CAG CTC GTG TCG TGA GAT GT -3'</td>
<td>150</td>
<td>(Higgins et al., 2004)</td>
</tr>
<tr>
<td><em>blaOXA-51</em></td>
<td>5’- TAA TGC TTT GAT CGG CCT TG -3'</td>
<td>353</td>
<td>(Woodford et al., 2006)</td>
</tr>
</tbody>
</table>

R: Reverse primer and F: Forward primer

### Table (2): Programs of PCR Thermocycling Conditions.

<table>
<thead>
<tr>
<th>Gene</th>
<th>Initial denaturation</th>
<th>Temperature (°C) / Time</th>
<th>Cycling condition</th>
<th>Final extension</th>
<th>Cycle Number</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>16S rRNA</em></td>
<td>94 °C for 4 min.</td>
<td>94 °C for 35 sec.</td>
<td>55 °C for 35 sec.</td>
<td>72 °C for 40 sec.</td>
<td>30</td>
</tr>
<tr>
<td><em>bla OXA-51</em></td>
<td>95 °C for 5 min.</td>
<td>94 °C for 45 sec.</td>
<td>52 °C for 40 sec.</td>
<td>72 °C for 45 sec.</td>
<td>30</td>
</tr>
</tbody>
</table>

**Agarose gel electrophoresis:**

To provides a simple and efficient method for purification of genomic DNA and the amplified PCR products. The DNA fragments and PCR products were separated and visualized by horizontal agarose-gel electrophoresis (8).

**Biofilm Formation measurement:**

A variety of different assays were used to evaluate production of biofilm
formation to all *A. baumannii* isolates was studied in different medium, on various materials with some modifications:

**Culturing on Congo red agar plates (CRA) (qualitative assay):**

The morphology of the colonies and their phenotypic changes were studied using CRA medium as previously described by (9) with some modifications. The CRA medium is composed of Brain heart infusion broth (BHIB) (37 g/L), sucrose (50 g/L), agar agar (15 g/L) dissolve in 900 ml of deionized water Congo red stain was prepared (0.8 g/L) was mixed with 100 ml of deionized water vigorously, thus the concentrated aqueous solution was sterilized by autoclaving separately from other medium constituents, after cooling the dye solution and medium to 55°C, it mixed and pour in Petri-dishes, left to solidify and use to detect biofilm formation.

The colonies were streaked on Congo red agar plates and incubated aerobically for 24 hours at 37°C to obtain single bacterial colonies.

**Tube Method (TM) (quantitative assay):**

*A. baumannii* isolates were tested for biofilm formation was investigated by the glass test tubes adherence test proposed by (10, 1) with some modifications. 10 ml Trypticase soya broth (TSB) with 1% glucose was inoculated with single colonies of the test bacterial strains on nutrient agar individually and the cultures were incubated statically for 48 hrs at 37°C, The experiment was performed in duplicates. After incubation, the contents were decanted and washed with phosphate buffer saline PBS (pH 7.3) and dried. The tubes were stained with crystal violet or safranin (0.1%) for 7 min, each tube was then gently rotated to ensure uniform staining of any adherent material on the inner surface and the contents gently decanted. Excess stain was removed and washed with D.W. Tubes were dried in inverted position and observed for biofilm formation.

The scoring for tube method was done according to the results of the control strains.

**Microtitre culture plate method (MTP) (quantitative assay):**

This analyze determined by microtitre plate method and considered by (10) is performed by a spectrophotometric method, which measures the total biofilm biomass (bacterial cells and extracellular matrix). Biofilm formation assays and the standard test for detection of biofilm formation as follow (11) with some modifications. Briefly, *A. baumannii* isolates were grown overnight in in 10 ml of trypticase soy broth (TSB) with 1% glucose for 24 hrs at 37°C. The culture was diluted fresh medium in a ratio of 1:90 in TSB-1 % glucose, then 200 μl of diluted cell suspension was filled in each well of a sterile 96 well polystyrene flat-bottom microtitre plates, and 200 μl aliquots of only TSB + 1% glucose were dispensed into microtiter plate to serve up as a control, followed by the plates were covered and incubated for 24 hrs at 37°C.

After three carefully washes with phosphate-buffered saline (PBS) (pH 7.2), then the attached bacteria were
fixed with 200 mL of 99% methanol per well for 15 minutes. Any remaining biofilm was stained with crystal violet (1% v/v) for 30 min and wells were washed again with PBS. The dye bound to the adherent cells was resolubilized with 200 μl of 95% ethanol for 20 min, and were fixed the attached bacteria, and the optical density (OD) was quantified at 630 nm using an ELISA reader. Each assay was performed in triplicate and the average optical density was considered.

According to their optical densities, the adherence capability of each bacterial cell was classified into the following four categories as follows: if OD ≤ ODc, the bacteria were non-biofilm (−); if ODc < OD ≤ 2 × ODc, the bacteria were weakly biofilm producer (+); if 2 × ODc < OD ≤ 4 × ODc, the bacteria were moderately biofilm producer (++) if 4 × ODc < OD, the bacteria were strongly biofilm producer (+++) (12).

**Results and Discussion:**

**Distribution of A. baumannii according to infection:**

The results exposed that a total number of 83 (17.47%) isolates of *A. baumannii* were obtained from 475 clinical specimens which initially were diagnosed in hospitals from different source. The distributions of the isolates according to specimens types are shown in Figure (1).

The results of current study is correlated with the local studies in Iraq conducted by (14) they diagnosed and identified that isolation in different percentage rates of nosocomial infections caused by *A. baumannii* from same sources and that agreement with present study. Furthermore these results were paralleled with the other comparable studies in world such: (15, 16) in Iran they isolated *A. baumannii* bacteria from different sources of clinical isolates.

The reasons for the variation in rate and percentage of isolation of *A. baumannii* between studies may be due to several factors such as place of collection (unit of hospital), geographic site and the difference in time of collection as well as the variation in the number of the studied samples (17).

![Figure (1): Percentages of *A. baumannii* isolates among different clinical samples.](image-url)
**Bacterial identification:**

The phenotypic identification of the *A. baumannii* isolates using bacteriological methods, Grams staining, colonial morphology and microscopically characteristics Figure (2). Then characteristics of isolates were subjected identified by biochemical test Table (3), and confirmatory identification by Api 20 NE and Vitek-2 system. Accurate identification and typing of bacterial isolates are essential, particularly when determining strains involved in hospital outbreaks (18). Results of current study shown to be agreement with all different other studies in this aspect like studies of (19).

![Figure (2): Colonies of *A. baumannii* isolates on selective medium CHROMagar Acinetobacter](image)

**Table (3): Morphological and Biochemical identification results of *A. baumannii*.**

<table>
<thead>
<tr>
<th>No.</th>
<th>Biochemical Test</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Gram stain</td>
<td>(-)</td>
</tr>
<tr>
<td>2.</td>
<td>Microscopic shape</td>
<td>Coccobacilli</td>
</tr>
<tr>
<td>3.</td>
<td>Growth at 44°C</td>
<td>(+)</td>
</tr>
<tr>
<td>4.</td>
<td>Lactose fermentation</td>
<td>(-)</td>
</tr>
<tr>
<td>5.</td>
<td>Hemolysin production</td>
<td>(-)γ hemolysis</td>
</tr>
<tr>
<td>6.</td>
<td>Oxidase test</td>
<td>(-)</td>
</tr>
<tr>
<td>7.</td>
<td>Catalase production test</td>
<td>(+)</td>
</tr>
<tr>
<td>8.</td>
<td>Methyle red</td>
<td>(+)</td>
</tr>
<tr>
<td>9.</td>
<td>Citrate utilization</td>
<td>(+)</td>
</tr>
<tr>
<td>10.</td>
<td>Kligrar iron agar</td>
<td>Alkaline slant /No change bottom, No gas, No H₂S</td>
</tr>
<tr>
<td>11.</td>
<td>Urease production</td>
<td>variable</td>
</tr>
<tr>
<td>12.</td>
<td>Voges- Proskauer</td>
<td>(-)</td>
</tr>
<tr>
<td>13.</td>
<td>Motility test</td>
<td>(-)</td>
</tr>
<tr>
<td>14.</td>
<td>Indoe production</td>
<td>(-)</td>
</tr>
</tbody>
</table>

(+) : positive, (- -): negative

**Genotyping Identification:**

**Genomic DNA Extraction:**

Genomic DNA was successfully extracted from all *A. baumannii* isolates using a commercial Genomic DNA purification Kit (Geneaid) according to instructions of the company. Extraction results were good and the DNA quantification (concentration and
purity) were directly performed by Nanodrop spectrophotometer at 260/280 nm. Extracted DNA concentration from all the isolates ranged between (58 to 142) ng/μl and purity was evaluated ranged between (1.8 to 2). Extracted DNA were confirmed and analyzed by gel electrophoresis in 1% agarose for 30 min at 75 volts and exposed to U.V light in which the DNA appears as compact bands shown in figure (3).

![Figure (3): Genomic DNA bands extracted from A.baumannii isolates on (1% agarose, 75V, for 30 min stained with ethidium bromide). Lane 1-15: DNA of Lane N: Negative control (water).](image)

**Molecular detection of Acinetobacter species by detection 16SrRNA gene:**

In this study identification of the 83 isolates was done used molecular identification A.baumannii. A specific primer for gene responsible for determined the Acinetobacter genous (16SrRNA) would identify all the isolates correctly and can be used to perform a single step Conventional PCR technique. Results exhibited that 83(100%) isolates were A.baumannii using gene 16SrRNA with amplified size of 150bp that present in Table (4) and according to the statistical analysis that show high significant at P<0.01. All isolates gives positive results that amplified fragments (150 bp) bands were separated by electrophoresis on a agarose gel, stained with ethidium bromide, photographed under UV light that showed in Figure (4).

The results of current study were agreement with studies in local area in Iraq by (14) they using the gene 16SrRNA for molecular identification of Acinetobacter clinical isolate with high rate of genomic identification. Moreover agreement with other studies which mentioned that detection and sequencing of this gene is an effective means for the identification of clinical isolates of Acinetobacter by (3).

16SrRNA gene is one of the structural RNA components of the ribosome. Prokaryotes have 5S and 23S species in large subunit and a 16SrRNA species in the small subunit (20). Postulated the importance of 16S-23S internal transcribed spacer to detect some bacterial identification at species level. As reported from sequence analysis studies, 16SrRNA could be used to identify Acinetobacter to the genus level (21).
Molecular identification of *A. baumannii* by detection *blaOXA-51*-like gene:

The results of the PCR products have been confirmed by comparing their molecular weight with 100 bp DNA Ladder by analysis of the bands on gel electrophoresis, to *blaOXA-51*-like gene is present in all 83(100%) of DNA *A. baumannii* isolates with a PCR product size single amplicon 353bp shown in Figure (5) and that present in Table (4) which indicated that all clinical isolates identified as *A. baumannii*.

The current results is parallel with most of the previous studies which evidence the occurrence of *blaOXA-51*-like gene in all clinical isolates of *A. baumannii* and not detected in the other species of *Acinetobacter* spp. (22). This result was in agreement with other researchers in Iran (15, 16) identified of the isolates *A. baumannii* phenotypically, all confirmed as by using *blaOXA-51*-like primers and observing ~ 353 bp band in the agarose gel.

The purpose of *blaOXA-51*-like gene identification of *Acinetobacter* species has several benefits over phenotypic identification. Even though the detection of the *blaOXA-51*-like gene has been considered a simple, rapid and reliable method for *A. baumannii* genomic species identification as these are naturally occurring carbapenemases genes to *A. baumannii*(23).
Table (4): Prevalence of PCR amplified detection genes in *A. baumannii* clinical isolates.

<table>
<thead>
<tr>
<th>No.</th>
<th>Gene name</th>
<th>Number and the percentage of positive isolates n= 83</th>
<th>Number and the percentage of negative isolates n= 83</th>
<th>Chi-Square (χ²) (P-value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>16SrRNA</td>
<td>83 (100%)</td>
<td>0 (0%)</td>
<td>15.00 ** (0.0001)</td>
</tr>
<tr>
<td>2.</td>
<td><em>blaOXA</em>-51</td>
<td>83 (100 %)</td>
<td>0 (0%)</td>
<td>15.00 ** (0.0001)</td>
</tr>
</tbody>
</table>

** (P<0.01).

Detection of the Bacterial Ability for Adherence and Biofilm Formation:

In this study we determine their ability of 83 *A.baumannii* isolates for adherence and to produce slime layer (Biofilm formation) were experienced by using three conventional phenotypic methods, including Congo-red agar (CRA), Tube method (TM) and Microtiter plate (MTP) by ELISA reader Figures (6,7,8).

The results shown that the difference was statistically significant at P<0.01 between the 83 *A.baumannii* isolates to produce biofilm by using these methods, the highest value of biofilm formation 74(89.15%) from isolates have the ability to adherence and produce slim layer while 9(10.84%) from isolates which have no the ability to adherence and produce slim layer Table (5). The positive results for each method divided in to three types according to amount of slime layer that present in Table (6) as the fallowing: (Strong, Moderate and week) producer biofilm, and shown difference significant between these methods.

Figure (6): Detection of slime layer producing *A.baumannii* isolates colonies on Congo red agar method. A- Strong producer biofilm, B- Weak biofilm producer, C- Moderate biofilm producer and D- Non biofilm producer.
Figure (7): Detection of biofilm producers in *A. baumannii* isolates by Tube method. A- Strong biofilm, B- Moderate biofilm producer, C- Weak biofilm producer and D- Non biofilm producer.

Figure (8): Screening of adhesion and motility producers by MTP method, OD (measured at wavelength of 630 nm). A: strong producer biofilm, B: moderate producer biofilm, C: week producer biofilm, D: non biofilm producer and N: negative control.

Table (5): Total *A. baumannii* isolates which have the ability and non ability to adherence and produce slim layer.

<table>
<thead>
<tr>
<th>isolates</th>
<th>Number of isolates</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Isolates which have the ability to adherence and produce slim layer in three methods (CRD, TM and MTP)</td>
<td>74</td>
<td>89.15%</td>
</tr>
<tr>
<td>Isolates which Non-have the ability to adherence and produce slim layer in three methods (CRD, TM and MTP)</td>
<td>9</td>
<td>10.84%</td>
</tr>
<tr>
<td>Total number</td>
<td>83</td>
<td>100%</td>
</tr>
<tr>
<td>Chi-Square (χ²) (P-value)</td>
<td>---</td>
<td>14.06 ** (0.0001)</td>
</tr>
</tbody>
</table>

** (P<0.01).

Table (6): Number and Percentage of 83 *A. baumannii* isolates which have the ability to produce slime layer by using three phenotyping methods

<table>
<thead>
<tr>
<th>Biofilm produce</th>
<th>Congo red agar (CRA) n= 83</th>
<th>Tube method (TM) n= 83</th>
<th>Microtiter plate (MTP) n= 83</th>
</tr>
</thead>
<tbody>
<tr>
<td>Strong ( ++++</td>
<td>48 (57.83%)</td>
<td>45 (54.21%)</td>
<td>58 (69.87%)</td>
</tr>
<tr>
<td>Moderate ( ++</td>
<td>16 (19.27%)</td>
<td>15 (18.07%)</td>
<td>10 (12.04%)</td>
</tr>
<tr>
<td>Weak ( +</td>
<td>10 (12.04%)</td>
<td>14 (16.86%)</td>
<td>6 (7.22%)</td>
</tr>
<tr>
<td>Non ( -</td>
<td>9 (10.84%)</td>
<td>9 (10.84%)</td>
<td>9 (10.84%)</td>
</tr>
<tr>
<td>Total</td>
<td>83 (100%)</td>
<td>83 (100%)</td>
<td>83 (100%)</td>
</tr>
<tr>
<td>Chi-Square (χ²)</td>
<td>--- 10.72 ** (0.0001)</td>
<td>--- 10.35 ** (0.0001)</td>
<td>--- 12.25 ** (0.0001)</td>
</tr>
</tbody>
</table>

** (P<0.01)
These results nearly compatible with the study by (16) which found more among A. baumannii isolates showed strong slime layer on the Congo red agar. Furthermore also agreement with the study by (24) shown the strains are found positive as well as slime-forming onto CRA.

The CRA is selected in an attempt to improved its capability to identify adhesion and biofilm formation by A. baumannii isolates using a specially prepared solid medium by production changes in the procedure and adjusting different physical parameters. Thus the CRA method is simple, sensitive and fast to carry out and the results are usually based on the colony color produced, which ranges from red for non-biofilm producing strains to black for biofilm producing strains, and reproducible and has the advantage that colonies remain viable on the medium (25).

The results of Tube method (TM) show that there was complete agreement with the results of (16) who reported from the detection of adherent by this method was positivity results among A. baumannii isolates. It’s noted for this procedure the presence of high variability among repetitions of this method. Tube method is also called traditional Christensen method this method is as simple and widely use to determine the ability of bacteria to adherence to the smooth surface such as glass tubes (11). The adhesion growth on smooth surface of class tubes results from the presence of polysaccharide on the smooth surface of class tubes because A. baumannii uses glucose which is found in culture media to produce aggressively sticky glucan polymers that facilitate the attachment of cells and adherent to smooth surface lead to promoting biofilm formation (4).

But we in agreement with the preceding studies, Tube method (TM) metod cannot be suggested as general screening test to identify biofilm producing isolates. Similarly, it was difficult to differentiate between strains weak biofilm producers and non-producers (24).

In this and data obtained from biofilm formation were study biofilm was formed on polystyrene microplates parallel with (26) whom found that all A. baumannii have the ability to formation of biofilm in this method. And another by (27) they showed that 12 isolates from 15 total isolates have ability to produce biofilm formation.

Furthermore the results was parallel with the results of other study by (16) and (15) whom evaluation of biofilm formation of A. baumannii isolates by microtiter plate method and gave higher rate of biofilm formation in this method. Microtiter plate is an important instrument for the study of the early stages in biofilm adhesive and allows for the formation of a biofilm on the wall and bottom of a microtiter plate (28). It has been is most widely used and was considered as standard test for detection of biofilm formation because it allows a simple, more accurate, and rapid to quantify contact cell attachment and biofilm formation of different bacterial strains (29).

The variation in biofilm thickness may be ascribed to the differences in isolates ability to produce biofilm, Formation of biofilm plays an important role in the pathogenesis of pathogens
and development of these biofilms is support on the signal mediated QS system, the QS systems play an important role in the pathogenesis and regarding the role in biofilm formation of A. baumannii infections (30).

Even though the principle of adherence detection by MTP depends on a variety of conditions, they remain better and more accurate than CRA and TM for biofilm detection (31).

In conclusion, this study showed the CRA and TM tests could be successfully used to detect A. baumannii isolate adhesion and biofilm producing, observed in this study when the CRA and TM were compared with the MTP test, indicated the MTP test should be the first choice because this test was more sensitive than the CRA and TM tests, MTP allowed an easy and quantitative classification of the A. baumannii isolates. This method MTP gave the best discrimination between strong, moderate producers and non-producers of adherence.

References:


