

**Complementary functionalities of extracellular polymeric substances, adhesion ability and hydrophobicity in *Pseudomonas* isolates may help the selection of strategically advantageous microbial inoculants**

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## SUPPLEMENTARY MATERIAL

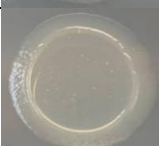
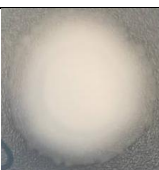
**Table S1** Formulation of the medium ATCC No.14 (a) and PUM buffer (b)

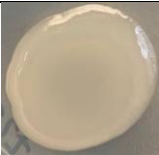
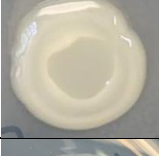
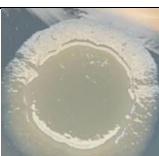
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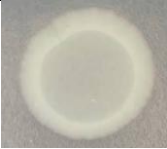
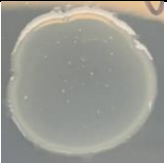
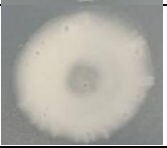
| Component                                            | Concentration (g L <sup>-1</sup> ) |
|------------------------------------------------------|------------------------------------|
| KH <sub>2</sub> PO <sub>4</sub>                      | 0.200                              |
| K <sub>2</sub> HPO <sub>4</sub>                      | 0.800                              |
| MgSO <sub>4</sub> x 7H <sub>2</sub> O                | 0.200                              |
| CaSO <sub>4</sub> x 2H <sub>2</sub> O                | 0.100                              |
| FeCl <sub>3</sub>                                    | 0.001                              |
| Na <sub>2</sub> MoO <sub>4</sub> x 2H <sub>2</sub> O | 0.001                              |
| Yeast extract                                        | 0.500                              |
| Sucrose                                              | 20.0                               |
| (Agar)                                               | 15.0                               |
| pH = 7.2                                             |                                    |

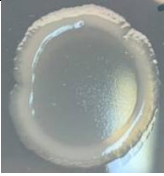
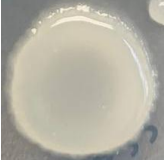
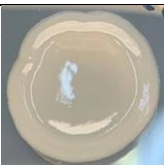
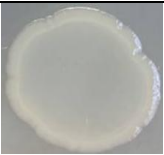
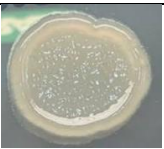
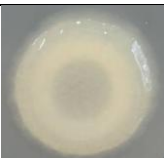
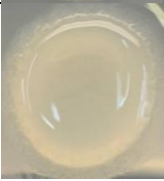
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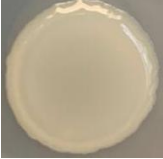
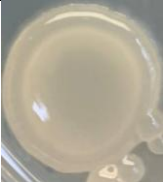
| Component                                          | Concentration (g L <sup>-1</sup> ) |
|----------------------------------------------------|------------------------------------|
| K <sub>2</sub> HPO <sub>4</sub> .4H <sub>2</sub> O | 22.2 g                             |
| KH <sub>2</sub> PO <sub>4</sub> .4H <sub>2</sub> O | 7.26 g                             |
| Urea                                               | 1.80 g                             |
| MgSO <sub>4</sub> .7H <sub>2</sub> O               | 0.20 g                             |
| pH = 7.1                                           |                                    |

| Isolate code | Colony morphology                                                                   | Morphological type and characteristics                                                   | Reaction on CRA (+/-) |
|--------------|-------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------|-----------------------|
| C1           |    | V- Round colonies with limited extracellular material at margins                         | -                     |
| C4           |    | IV- Flat, round colonies that may have curled margins; no visible extracellular material | -                     |
| C9           |    | IV- Flat, round colonies that may have curled margins; no visible extracellular material | -                     |
| C10          |   | I- Circular, raised, mucous; diffuse extracellular material                              | -                     |
| C14          |  | III- Translucent, flat, dry; carpet-like, no matrix                                      | -                     |
| C17          |  | V- Round colonies with limited extracellular material at margins                         | -                     |
| C21          |  | I- Circular, raised, mucous; diffuse extracellular material                              | -                     |
| C22          |  | I- Circular, raised, mucous; diffuse extracellular material                              | -                     |
| C25          |  | V- Round colonies with limited extracellular material at margins                         | -                     |

|     |                                                                                     |                                                                                                            |   |
|-----|-------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------|---|
| C27 |    | II- Crateriform,<br>raised, round;<br>abundant,<br>condensed matrix                                        | + |
| C28 |    | II- Crateriform,<br>raised, round;<br>abundant,<br>condensed matrix                                        | + |
| C32 |    | I- Circular, raised,<br>mucous; diffuse<br>extracellular<br>material                                       | - |
| C33 |    | II- Crateriform,<br>raised, round;<br>abundant,<br>condensed matrix                                        | + |
| C37 |    | II- Crateriform,<br>raised, round;<br>abundant,<br>condensed matrix                                        | + |
| C41 |   | V- Round<br>colonies with<br>limited<br>extracellular<br>material at<br>margins                            | - |
| C42 |  | III-<br>Translucent,<br>flat, dry;<br>carpet-like,<br>no matrix                                            | + |
| C44 |  | IV- Flat, round<br>colonies that may<br>have curled<br>margins; no<br>visible<br>extracellular<br>material | - |
| C47 |  | IV- Flat, round<br>colonies that may<br>have curled<br>margins; no<br>visible<br>extracellular<br>material | - |
| C48 |  | V- Round<br>colonies with<br>limited<br>extracellular<br>material at<br>margins                            | - |

|     |                                                                                     |                                                                                          |   |
|-----|-------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------|---|
| C59 |    | IV- Flat, round colonies that may have curled margins; no visible extracellular material | - |
| C60 |    | V- Round colonies with limited extracellular material at margins                         | - |
| S2  |    | V- Round colonies with limited extracellular material at margins                         | - |
| S3  |    | III- Translucent, flat, dry; carpet-like, no matrix                                      | - |
| S8  |   | II- Crateriform, raised, round; abundant, condensed extracellular matrix                 | + |
| S10 |  | IV- Flat, round colonies that may have curled margins; no visible extracellular material | - |
| S11 |  | I- Circular, raised, mucous; diffuse extracellular material                              | - |
| S12 |  | II- Crateriform, raised, round; abundant, condensed matrix                               | + |
| S18 |  | I- Circular, raised, mucous; diffuse extracellular material                              | - |
| S20 |  | I- Circular, raised, mucous; diffuse extracellular material                              | - |

|     |                                                                                     |                                                                            |   |
|-----|-------------------------------------------------------------------------------------|----------------------------------------------------------------------------|---|
| S21 |    | V- Round colonies with limited extracellular material at margins           | - |
| S22 |    | II- Crateriform, raised, round; abundant, condensed extracellular matrix   | + |
| S23 |    | II- Crateriform, raised, round; abundant, condensed extracellular material | + |
| S25 |    | II- Crateriform, raised, round; abundant, condensed extracellular material | + |
| S28 |   | V- Round colonies with limited extracellular material at margins           | - |
| S29 |  | V- Round colonies with limited extracellular material at margins           | + |
| S31 |  | I- Circular, raised, mucous; diffuse extracellular material                | - |
| S34 |  | II- Crateriform, raised, round; abundant, condensed extracellular material | + |
| S35 |  | II- Crateriform, raised, round; abundant, condensed matrix                 | + |
| S38 |  | II- Crateriform, raised, round; abundant, condensed extracellular material | + |

|     |                                                                                     |                                                                                          |   |
|-----|-------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------|---|
| S40 |    | III- Translucent, flat, dry; carpet-like, no matrix                                      | - |
| S42 |    | II- Crateriform, raised, round; abundant, condensed extracellular material               | + |
| S44 |    | II- Crateriform, raised, round; abundant, condensed extracellular material               | + |
| S48 |    | IV- Flat, round colonies that may have curled margins; no visible extracellular material | - |
| S50 |   | III- Translucent, flat, dry; carpet-like, no matrix                                      | - |
| S54 |  | II- Crateriform, raised, round; abundant, condensed extracellular material               | + |
| S56 |  | II- Crateriform, raised, round; abundant, condensed extracellular material               | + |
| S58 |  | II- Crateriform, raised, round; abundant, condensed extracellular material               | + |
| S60 |  | II- Crateriform, raised, round; abundant, condensed extracellular material               | + |

**Figure S1.** Colony morphology of the 48 isolates grown on ATCC No.14 and positive or negative reaction on CRA after incubation for 48h at 25°C.



**Figure S2** Cell biomass and CR-dyed extracellular matrix after the Congo Red-binding assay

**Table S2** ANOVA Table for b-CR

| Source       | DF         | Sum of squares | Mean square | F-ratio | p-value  |
|--------------|------------|----------------|-------------|---------|----------|
| Isolate      | 47         | 42.1584        | 0.89699     | 82.3260 | < 0.0001 |
| Error        | 96         | 1.0588         | 0.01103     |         |          |
| <b>Total</b> | <b>143</b> | <b>43.2172</b> |             |         |          |

**Table S3** ANOVA Table for r-EPS production by the isolates.

| Source         | DF         | Sum of squares   | Mean square | F-ratio | p-value  |
|----------------|------------|------------------|-------------|---------|----------|
| <b>Isolate</b> | 47         | 13730.785        | 292.144     | 82.3221 | < 0.0001 |
| <b>Error</b>   | 96         | 340.680          | 3.549       |         |          |
| <b>Total</b>   | <b>143</b> | <b>14071.465</b> |             |         |          |

**Table S4** ANOVA Table for hydrophobicity index

| Source       | DF         | Sum of squares   | Mean square | F-ratio | p-value  |
|--------------|------------|------------------|-------------|---------|----------|
| Isolate      | 47         | 13.370.785       | 292.144     | 82.3221 | < 0.0001 |
| Error        | 96         | 340.680          | 3.549       |         |          |
| <b>Total</b> | <b>143</b> | <b>14071.465</b> |             |         |          |

**Table S5** ANOVA Table for cell adhesion

| Source       | DF         | Sum of squares   | Mean square | F-ratio | p-value  |
|--------------|------------|------------------|-------------|---------|----------|
| Isolate      | 47         | 37222.047        | 791.958     | 62.8393 | < 0.0001 |
| Error        | 96         | 1209.880         | 12.603      |         |          |
| <b>Total</b> | <b>143</b> | <b>38431.927</b> |             |         |          |

**Table S6** Regression Analysis of Adhesion with Predictor Variables (b-CR, r-EPS, HI%)

| Term                                                         | Coefficient ( $\beta$ ) | Standard Error | t-ratio         | p-value  | 95% Confidence Interval |
|--------------------------------------------------------------|-------------------------|----------------|-----------------|----------|-------------------------|
| Intercept                                                    | -0.0263                 | 0.0873         | -0.30           | 0.7645   | (-0.2023, 0.1496)       |
| b-CR                                                         | -0.6300                 | 0.1403         | -4.49           | < 0.0001 | (-0.9129, -0.3471)      |
| r-EPS                                                        | 0.14028                 | 0.1116         | 1.29            | 0.2044   | (-0.0812, 0.3688)       |
| HI                                                           | 0.3193                  | 0.1317         | 2.42            | 0.0196   | (0.0536, 0.5849)        |
| <b>Generalized R square = 0.6642, Adj. R square = 0.6408</b> |                         |                |                 |          |                         |
| Parameter                                                    | Estimate                | Standard Error | Wald Chi-Square | p-value  | 95% Confidence Interval |
| Scale                                                        | 0,5980                  | 0,066          | 82              | < 0.0001 | (0.4686, 0,7275)        |

**Table S7** Regression Analysis of hydrophobicity index (HI) with Predictor Variables (b-CR, r-EPS)

| Term                                                         | Coefficient ( $\beta$ ) | Standard Error | t-ratio         | p-value  | 95% Confidence Interval |
|--------------------------------------------------------------|-------------------------|----------------|-----------------|----------|-------------------------|
| Intercept                                                    | -0.006849               | 0.099847       | -0.068          | 0.9456   | (-0.1943, 0.2080)       |
| b-CR                                                         | -0.654623               | 0.125623       | -5.2697         | < 0.0001 | (-0.4045, -0.1019)      |
| r-EPS                                                        | -0.151285               | 0.1266252      | -1.2039         | 0.2350   | (-0.9098, 0.3994)       |
| <b>Generalized R square = 0.5609, Adj. R square = 0.5409</b> |                         |                |                 |          |                         |
| Parameter                                                    | Estimate                | Standard Error | Wald Chi-Square | p-value  | 95% Confidence Interval |
| Scale                                                        | 0.6844                  | 0.0742         | 85              | < 0.0001 | (0.5389, 0.8299)        |