

# **Use of Immune Profiling Panel to assess the immune response of septic patients for prediction of worsening as a composite endpoint**

Estelle Peronnet, Gabriel Terraz, Elisabeth Cerrato, Katia Imhoff, Sophie Blein, Karen Brengel-Pesce, Maxime Bodinier, Aurore Fleurie, Thomas Rimmelé, Anne-Claire Lukaszewicz, Guillaume Monneret, Jean-François Llitjos

## **Supplementary data**

**Supplementary Table S1** Prototype Immune Profiling Panel (IPP) gene set.

**Supplementary Table S2** Patients baseline characteristics and management in the train and test sets.

**Supplementary Table S3** Patients outcomes in the train and test sets

**Supplementary Table S4** IPP model performances for the prediction of worsening at day 15 and day 28.

**Supplementary Table S5** Enrichment in clinical outcomes at day 15 and day 28 in the two groups identified by the IPP model, on patients alive at D28.

**Supplementary Table S1** Prototype Immune Profiling Panel (IPP) gene set.

| Gene          | Name                                                     |
|---------------|----------------------------------------------------------|
| <b>C3AR1</b>  | Complement C3a receptor 1                                |
| <b>CD177</b>  | CD177 molecule                                           |
| <b>CD3D</b>   | CD3d molecule                                            |
| <b>CD74</b>   | CD74 molecule                                            |
| <b>CIITA</b>  | Class II major histocompatibility complex transactivator |
| <b>CTLA4</b>  | Cytotoxic T-lymphocyte associated protein 4              |
| <b>CX3CR1</b> | C-X3-C motif chemokine receptor 1                        |
| <b>IFNG</b>   | Interferon gamma                                         |
| <b>IL1R2</b>  | Interleukin 1 receptor 2                                 |
| <b>S100A9</b> | S100 calcium binding protein A9                          |
| <b>TAP2</b>   | Transporter 2, ATP binding cassette subfamily B member   |

**Supplementary Table S2** Patients baseline characteristics and management in the train and test sets.

| Variable                        |                    | All<br>(n=332) | Train set<br>(n=217) | Test set<br>(n=115) | p value |
|---------------------------------|--------------------|----------------|----------------------|---------------------|---------|
| <b>Cohort</b>                   |                    |                |                      |                     | 0.868   |
|                                 | MIP Réa            | 245 (74%)      | 159 (73%)            | 86 (75%)            |         |
|                                 | REALISM            | 87 (26%)       | 58 (27%)             | 29 (25%)            |         |
| <b>Baseline characteristics</b> |                    |                |                      |                     |         |
| Gender, male                    |                    | 212 (64%)      | 138 (64%)            | 74 (64%)            | 0.987   |
| Age, years                      |                    | 67 [57-77]     | 68 [57-78]           | 64 [55-75]          | 0.064   |
| Septic shock at inclusion       |                    | 189 (57%)      | 119 (55%)            | 70 (61%)            | 0.348   |
| Initial infection site          |                    |                |                      |                     | 0.689   |
|                                 | Respiratory        | 167 (50%)      | 109 (50%)            | 58 (50%)            |         |
|                                 | Abdominal          | 80 (24%)       | 55 (25%)             | 25 (22%)            |         |
|                                 | Others             | 85 (26%)       | 53 (24%)             | 32 (28%)            |         |
| Initial infection acquisition   |                    |                | 217                  |                     | 0.922   |
|                                 | Community acquired | 222 (67%)      | 146 (67%)            | 76 (66%)            |         |
|                                 | Hospital acquired  | 110 (33%)      | 71 (33%)             | 39 (34%)            |         |
| Charlson score                  |                    | 4 [2-6]        | 4 [2-6]              | 4 [3-6]             | 0.487   |
| SAPS II                         |                    | 55 [42-67]     | 54 [42-68]           | 55 [41-67]          | 0.614   |
| SOFA at admission               |                    | 9 [7-12]       | 9 [7-11]             | 9 [7-12]            | 0.173   |
| <b>Patient management</b>       |                    |                |                      |                     |         |
| Mechanical ventilation          |                    | 223 (67%)      | 147 (68%)            | 76 (66%)            | 0.855   |
| RRT                             |                    | 46 (14%)       | 32 (15%)             | 14 (12%)            | 0.632   |
| Catecholamines                  |                    | 220 (66%)      | 144 (66%)            | 76 (66%)            | 1.000   |
| Corticoids                      |                    | 70 (21%)       | 46 (21%)             | 24 (21%)            |         |

p values correspond to comparisons between train and test sets, and are bold when <0.05.

**Supplementary Table S3** Patients outcomes in the train and test sets

| Variable                   | All<br>(n=332)   | Train set<br>(n=217) | Test set<br>(n=115) | p value |
|----------------------------|------------------|----------------------|---------------------|---------|
| <b>D15 outcomes</b>        |                  |                      |                     |         |
| Observed worsening D15     | 64 (19%)         | 41 (19%)             | 23 (20%)            | 0.923   |
| Death without HAI          | 34 (10%)         | 23 (11%)             | 11 (10%)            |         |
| HAI                        | 30 (9.0%)        | 18 (8.3%)            | 12 (10%)            |         |
| HAI D15 site               |                  |                      |                     | 0.574   |
| Pulmonary                  | 15 (50%)         | 9 (50%)              | 6 (50%)             |         |
| Urinary                    | 3 (10%)          | 2 (11%)              | 1 (8.3%)            |         |
| Bacteraemia                | 4 (13%)          | 2 (11%)              | 2 (17%)             |         |
| Abdominal                  | 2 (6.7%)         | 2 (11%)              | 0 (0.0%)            |         |
| Catheter-related infection | 2 (6.7%)         | 0 (0.0%)             | 2 (17%)             |         |
| Others                     | 4 (13%)          | 3 (17%)              | 1 (8.3%)            |         |
| ICU Free days              | 2.50 [0-8]       | 2 [0-8]              | 3 [0-8]             | 0.519   |
| RRT free days              | 15.0 [13.0-15.0] | 15.0 [12.0-15.0]     | 15.0 [13.5-15.0]    | 0.317   |
| Intubation free days       | 15.0 [12.0-23.0] | 15.0 [12.0-23.0]     | 16.0 [12.0-23.0]    | 0.699   |
| Catecholamines free days   | 12.0 [10.0-14.0] | 12.00 [11.0-14.0]    | 12.00 [9.5-14.0]    | 0.209   |
| <b>D28 outcomes</b>        |                  |                      |                     |         |
| Observed worsening D28     | 118 (35%)        | 79 (36%)             | 39 (34%)            | 0.741   |
| Death without HAI          | 65 (19%)         | 41 (19%)             | 24 (21%)            |         |
| HAI                        | 53 (16%)         | 38 (17%)             | 15 (13%)            |         |
| HAI D28 site               | 65               | 24                   | 41                  | 0.548   |
| Pulmonary                  | 34 (52%)         | 13 (54%)             | 21 (51%)            |         |
| Urinary                    | 11 (17%)         | 3 (13%)              | 8 (20%)             |         |
| Bacteraemia                | 6 (9.2%)         | 3 (13%)              | 3 (7.3%)            |         |
| Abdominal                  | 5 (7.7%)         | 1 (4.2%)             | 4 (9.8%)            |         |
| Catheter-related infection | 4 (6.2%)         | 3 (13%)              | 1 (2.4%)            |         |
| Others                     | 5 (7.7%)         | 1 (4.2%)             | 4 (9.8%)            |         |
| ICU Free days              | 15 [0-21]        | 14 [0-21]            | 16 [0-21]           | 0.366   |
| RRT free days              | 28.0 [13.0-28.0] | 28.0 [3.0-28.0]      | 28.0 [22.5-28.0]    | 0.205   |
| Intubation free days       | 21.5 [0.0-27.0]  | 21.0 [0.0-27.0]      | 22.0 [8.5-26.0]     | 0.631   |
| Catecholamines free days   | 25.0 [19.8-27.0] | 25.0 [18.0-27.0]     | 24.0 [20.0-27.0]    | 0.744   |

p values correspond to comparisons between train and test sets, and are bold when <0.05.

**Supplementary Table S4** IPP model performances for the prediction of worsening at day 15 and day 28.

|                              | <b>Train set (n=217)</b> | <b>Test set (n=115)</b> |
|------------------------------|--------------------------|-------------------------|
| <b>Performance at day 15</b> |                          |                         |
| AUC                          | 0.64 [0.55-0.74]         | 0.69 [0.58-0.80]        |
| Accuracy                     | 0.70 [0.63-0.75]         | 0.70 [0.62-0.78]        |
| Sensitivity                  | 0.46 [0.32-0.61]         | 0.52 [0.33-0.71]        |
| Specificity                  | 0.75 [0.68-0.81]         | 0.75 [0.65-0.83]        |
| PPV                          | 0.30 [0.20-0.42]         | 0.34 [0.21-0.51]        |
| NPV                          | 0.86 [0.79-0.90]         | 0.86 [0.77-0.92]        |
| <b>Performance at day 28</b> |                          |                         |
| AUC                          | 0.67 [0.60-0.75]         | 0.63 [0.52-0.73]        |
| Accuracy                     | 0.69 [0.62-0.74]         | 0.65 [0.56-0.73]        |
| Sensitivity                  | 0.47 [0.36-0.58]         | 0.44 [0.29-0.59]        |
| Specificity                  | 0.81 [0.74-0.87]         | 0.76 [0.66-0.84]        |
| PPV                          | 0.59 [0.46-0.70]         | 0.49 [0.33-0.64]        |
| NPV                          | 0.73 [0.65-0.79]         | 0.72 [0.62-0.81]        |

Performances are reported with [95% confidence interval].

AUC: area under the receiver operating characteristic curve; PPV: positive predictive value; NPV: negative predictive value

**Table S5** Enrichment in clinical outcomes at day 15 and day 28 in the two groups identified by the IPP model, on patients alive at D28.

| Patients alive at day 28 only |                  |                         |                        |              |
|-------------------------------|------------------|-------------------------|------------------------|--------------|
| Variable                      | Test set<br>n=95 | High risk<br>n=23 (24%) | Low risk<br>n=72 (76%) | p value      |
| <b>D15 outcomes</b>           |                  |                         |                        |              |
| ICU Free days                 | 5 [0-8]          | 0 [0-5.50]              | 6 [0.75-8]             | <b>0.004</b> |
| RRT free days                 | 15 [15-15]       | 15 [10-15]              | 15 [15-15]             | <b>0.002</b> |
| Intubation free days          | 17 [13-23]       | 15 [10-21.5]            | 18 [13-24]             | 0.145        |
| Catecholamines free days      | 12 [10-14]       | 11 [9.5-13]             | 12 [11-15]             | <b>0.049</b> |
| <b>D28 outcomes</b>           |                  |                         |                        |              |
| ICU free days                 | 18 [10-21]       | 13 [5-18.50]            | 19 [14.5-21]           | <b>0.006</b> |
| RRT free days                 | 28 [28-28]       | 28 [23-28]              | 28 [28-28]             | <b>0.002</b> |
| Intubation free days          | 23 [17.5-27]     | 18 [13.5-25.5]          | 24 [21-27]             | 0.061        |
| Catecholamines free days      | 25 [23-27]       | 24 [22.5-26]            | 25 [24-28]             | <b>0.049</b> |

p values correspond to comparisons between high-risk and low-risk groups. p values <0.05 are bold