**Supplementary materials**

**Cycle threshold values in the context of multiple RT-PCR testing for SARS-CoV-2**

**About time to first test in the present study**

**Supplementary tables**

**Table S1.** Clinical, epidemiological, and laboratory characteristic of 13 patients that presented a cycle threshold (Ct) value ≤33 after day 30 since symptoms onset.

|  |  |  |
| --- | --- | --- |
| **Patients** | **Counts** | **Summary statistics** |
| Age | 13 | 49 (38-84) |
| Men | 5 | 38.5% |
| Women | 8 | 61.5% |
| Death | 0 | 0% |
| Alive | 13 | 100% |
| Beginning of symptoms to hospitalization |  | 6 (1-22) |
| Beginning of symptoms to discharge |  | 25 (15-76) |
| First attention to discharge |  | 20 (11-72) |
| **Comorbidities** | n | % |
| * Hypertension
 | 4 | 30.8 |
| * Cancer
 | 2 | 15.4 |
| * Diabetes mellitus type 2
 | 1 | 7.8 |
| * Chronic kidney disease
 | 1 | 7.8 |
| * Hypothyroidism
 | 0 | 0 |
| * Chronic ischemic cardiomyopathy
 | 0 | 0 |
| * Rheumatoid arthritis
 | 0 | 0 |
| * Lupus
 | 0 | 0 |
| * Hepatic cirrhosis
 | 0 | 0 |
| * HIV
 | 0 | 0 |
| **Symptomatology** | n | % |
| * Fever
 | 8 | 61.5 |
| * Dry cough
 | 7 | 53.9 |
| * Productive cough
 | 3 | 23.1 |
| * Headache
 | 2 | 15.4 |
| * Dyspnea
 | 2 | 15.4 |
| * Odynophagia
 | 2 | 15.4 |
| * Dysphagia
 | 2 | 15.4 |
| * Arthralgia
 | 1 | 7.7 |
| * Asthenia
 | 1 | 7.7 |
| * Myalgia
 | 1 | 7.7 |
| * Runny nose
 | 1 | 7.7 |
| * Abdominal pain
 | 1 | 7.7 |
| * Diarrhea
 | 0 | 0 |
| * Anosmia
 | 0 | 0 |
| * Chest pain
 | 0 | 0 |
| * Nausea
 | 0 | 0 |
| * Vomit
 | 0 | 0 |
| * Dysgeusia
 | 0 | 0 |
| * Conjunctivitis
 | 0 | 0 |
| * Dysuria
 | 0 | 0 |
| * Increased frequency of urination
 | 0 | 0 |
| **Laboratory parameters** | Median | Range |
| White blood cells | 4.9 | (3.1-7.5) |
| Hemoglobin | 15.85 | (13-18.8) |
| Hematocrit | 43.2 | (33-55.9) |
| Platelets | 225 | (121-460) |
| Red cells | 4.99 | (4.22-6.48) |
| Monocytes % | 9.7 | (3.2-14 |
| Eosinophils % | 0.2 | (0-1.9) |
| Lymphocytes % | 26 | (11.9-41) |
| Neutrophils % | 67.5 | (49-87.9) |
| Basophils % | 0.4 | (0.2-1) |
| Glucose | 97 | (73-274) |
| BUN | 13.5 | (11-55) |
| Urea | 27 | (14-117.7) |
| Creatinine | 0.87 | (0.63-14.68) |

**Table S2.** Results of univariate analysis comparing different patient groups and using a dichotomized cycle threshold (Ct) value of ≤33 to discriminate infectious vs. no infectious patients for the first test.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variables** | **Dichotomization** | **Counts (%)** | **Infectious/Non-infectious** | **Cycle values (median, standard deviation)** | **Statistic** | ***p* value** |
| **x2/t/Pearson** |
| Age | >65 | 8 (13.12%) | 1 (5.3%)/7 (16.66%) | 28.87 (4.167) | 0.660/-0.297/ -0.129 | 0.415/0.768/0.321 |
| <65 | 53 (86.89%) | 18 (94.7%)/35 (83.33%) | 29.66 (7.031) |
| Comorbidities | Presence | 14 (23.73%) | 10 (24.4%)/4 (22.2%) | 30.24 (5.988) | 0/0.000/NA | 1/1/NA |
| Absence | 45 (76.3%) | 31 (75.6%)/14 (77.8%) | 28.7 (7.050) |
| Gender | Male | 38 (61.29%) | 27 (62.8%)/11 (57.9%) | 27.83 (7.623) | 0.007/-1.951/NA | 0.781/0.056/NA |
| Female | 24 (38.71%) | 16 (37.2%)/8 (42.1%) | 29.98 (4.473) |
| White blood cells | Normal | 46 (82.14%) | 32 (94.1%)/14 (63.6%) | 29.35 (7.167) | 0.076/0.098/0.42 | 0.707/0.922/0.759 |
| Abnormal | 10 (17.86%) | 2 (5.9%)/8 (36.4%) | 28.08 (4.457) |
| Lymphocytes | Normal | 37 (66.07%) | 27 (67.5%)/10 (62.5%) | 29.66 (5.101) | 0.002/-1.154/-0.060 | 0.761/0.253/0.662 |
| Abnormal | 19 (33.93%) | 13 (32.5%)/6 (37.5%) | 26.74 (8.918) |
| Monocytes | Normal | 52 (92.9%) | 37 (92.5%)/ 15 (93.75%) | 29.35 (6.828) | 0.000/0.509/-0.065 | 1/0.613/0.639 |
| Abnormal | 4 (7.14%) | 3 (7.5%)/1 (6.25%) | 26.11 (5.347) |
| Neutrophils | Normal | 35 (62.5%) | 26 (65%)/9 (56.25%) | 30.29 (5.322) | 0.093/-1.088/-0.003 | 0.557/0.282/0.983 |
| Abnormal | 21 (37.5%) | 14( 35%)/7 (43.75%) | 25.74 (8.428) |
| Eosinophils | Normal | 1(1.79%) | 1 (2.5%)/0 (0%) | 19.34 (NA) | 0/1.371/-0.085 | 1/9.220/0.539 |
| Abnormal | 55(98.2%) | 39 (97.5%)/16 (100%) | 29.04 (6.689) |
| **Platelets** | Normal | 45(80.36%) | 33 (82.5%)/12 (75%) | 29.66 (5.488) | 0.071/-1.190/**0.396** | 0.711/0.239/**0.003\*** |
| Abnormal | 11(19.64%) | 7 (17.5%)/4 (25%) | 26.74 (10.001) |
| Basophils | Normal | 54(96.4%) | 40 (100%)/14 (87.5%) | 28.87 (6.698) | 2.191/1.468/0.094 | 0.078/0.148/0.495 |
| Abnormal | 2(3.6%) | 0 (0%)/2 (12.5%) | 25.88 (10.303) |
| Hemoglobin | Normal | 32 (57.14%) | 23 (57.5%)/9 (56.25%) | 30.26 (7.562) | 0/-0.398/-0.183 | 1/0.692/0.181 |
| Abnormal | 24 (42.86%) | 17 (42.5%)/7 (43.75%) | 28.47 (5.6) |
| Hematocrit | Normal | 24 (42.86%) | 16 (40%)/8 (50%) | 27.9 (8.316) | 0.148/0.099/-0.028 | 0.558/0.197/0.841 |
| Abnormal | 32 (57.14%) | 24 (60%)/8 (50%) | 29.35 (5.268) |
| Erythrocytes | Normal | 35(62.5%) | 24 (60%)/11 (68.75%) | Median 28.27 Std. Deviation 7.263 | 0.093/0.175/-0.190 | 0.761/0.862/0.166 |
| Abnormal | 21(37.5%) | 16 (40%)/5 (31.25%) | 28.70 (5.9) |
| **C Reactive Protein** | Normal | 6(11.11%) | 2 (5%)/4 (28.6%) | 33.11 (4.383) | **3.691**/-1.530/-0.012 | **0.033\***/0.132/0.933 |
| Abnormal | 48(88.88%) | 38 (95%)/10 (71.4%) | 28.47 (6.665) |
| Creatinine | Normal | 38(70.37%) | 26 (66.7%)/12 (80%) | 28.87 (7.404) | 0.395/0.663/-0.187 | 0.508/0.510/0.181 |
| Abnormal | 16(29.63%) | 13 (33.4%)/3 (20%) | 29.98 (4.827) |
| Intensive Care Unit | Admitted | 11(19.3%) | 8 (20%)/3 (17.7%) | 27.88 (6.782) | 0/0.097/NA | 1/0.923/NA |
| Not-admitted | 46(80.7%) | 32 (80%)/14 (82.4%) | 29.35 (6.851) |

**Table S3.** Results of univariate analysis comparing different patient groups and using a dichotomized cycle threshold (Ct) value of ≤ 33 to discriminate infectious vs. no infectious patients for the second test.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variables** | **Dichotomization** | **Counts (%)** | **Infectious/Non-infectious** | **Cycle values (median, standard deviation)** | **Statistics** | ***p* value** |
| **x2/t/Pearson** |
| **Age** | >65 | 14 (13.33%) | 1 (10.5%)/13 (14.9%) | 34.28 (6.365) | 0.470/-1.474/**-0.276** | 0.455/0.146/**0.033**\* |
| <65 | 91 (86.66%) | 17 (89.5%)/74 (85.1%) | 35.33 (3.769) |
| Comorbidities | Presence | 29 (28.16%) | 6 (33.3%)/23 (27.1%) | 33.52 (4.343) | 0.062/-1.975/NA | 0.576/0.053/NA |
| Absence | 74 (71.84%) | 12 (66.7%)/62 (72.9%) | 35.7 (4.163) |
| Gender | Male | 66 (62.86%) | 11 (61.1%)/59 (64.8%) | Median 35.23 Std. Deviation 4.385 | 0/0.276/NA | 1/0.784/NA |
| Female | 39(37.14%) | 7 (38.9%)/32 (35.2%) | 34.55 (4.280) |
| White blood cells (normal 4.5 -10) | Normal | 82(82%) | 17 (94.4%)/65 (78.3%) | 35.15 (4.502) | 1.390/0.843/0.053 | 0.182/0.403/0.690 |
| Abnormal | 18(18%) | 1 (5.6%)/18 (21.7%) | 34.55 (3.352) |
| Lymphocytes(normal 1.1-3.2) | Normal | 57(57%) | 10 (55.6%)/47 (57.3%) | 35.28 (3.741) | 0/0.299/0.186 | 1/0.747/0.157 |
| Abnormal | 43(43%) | 8 (44.4%)/ 35 (42,7%) | 34.40 (4.970) |
| MonocytesNormal (0.3-0.8) | Normal | 89(89%) | 16 (88.9%)/73 (89%) | 35.07 (4.375) | 0/-0.216/0.012 | 1/0.830/0.926 |
| Abnormal | 11(11%) | 2 (11.1%)/9 (11%) | 35.35 (4.051) |
| **Neutrophi****Ls (normal 2.2-4.8)** | Normal | 48(48%) | 11 (61.1%)/37 (45.1%) | 34.16 (3.735) | 0.939/0.578/**-0.259** | 0.299/0.566/**0.047\*** |
| Abnormal | 52(52%) | 7 (38.9%)/45 (54.9%) | 34.41 (4.782) |
| Eosinophils(normal 0.5-2.9) | Normal | 1(1%) | 0 (0%)/1 (1.2%) | 35.33 (NA) | 0/-0.283/0.060 | 1/0.778/0.651 |
| Abnormal | 99 (99%) | 18 (100%)/81 (98.8%) | 34.81 (4.346) |
| Platelets (normal 150-450) | Normal | 85(85%) | 16 (88.9%)/69 (84.2%) | 34.55 (4.333) | 0.021/1.558/0.217 | 1/0.125/0.099 |
| Abnormal | 15(15%) | 2 (11.1%)/13 (15.9%) | 36.91 (3.844) |
| Basophils (normal <0.1) | Normal | 97(97%) | 18 (100%)/79 (96.3%) | Median 35.07 Std. Deviation 4.311 | 0.004/ NA/-0.143 | 1/NA/0.281 |
| Abnormal | 3(3%) | 0 (0%)/3 (3.7%) | 0 |
| Hemoglobin (normal 14-18) | Normal | 64(64%) | 12 (66.7%)/52 (63.4%) | 35.15 (4.882) | 0.00/0.350/0.061 | 1/0.727/0.644 |
| Abnormal | 36(36%) | 6 (33.3%)/30(36.6%) | 34.55 (3.113) |
| Hematocrit (normal 42-52) | Normal | 54(54%) | 12 (66.7%)/42 (51.2%) | 34.17 (4.977) | 0.864/1.454/-0.010 | 0.300/0.151/0.940 |
| Abnormal | 46(46%) | 6 (33.3%)/40 (48.8%) | 35.47 (3.232) |
| Erythrocytes (normal 4.7-6.1) | Normal | 67(67%) | 13 (72.2%)/54 (65.9%) | 34.55 (4.261) | 0.059/0.513/0.016 | 0.783/0.610/0.904 |
| Abnormal | 33(33%) | 5 (27.8%)/28 (34.2%) | 35.41 (4.470) |
| C Reactive Protein (normal<3.1) | Normal | 9(9.38%) | 1 (5.9%)/8 (10.1%) | 33.07 (6.491) | 0.007/0.372/-0.158 | 1/0.711/0.236 |
| Abnormal | 87 (90.63%) | 16 (94.1%)/71 (89.9%) | 35.15 (4.286) |
| Creatinine (normal 0.7-1.3) | Normal | 62 (63.92%) | 12 (66.7%)/50 (63.3%) | 34.16 (4.027) | 0/0.626/0.023 | 1/0.534/0.867 |
| Abnormal | 35 (36.08%) | 6 (33.3%)/29 (36.7%) | 35.93 (4.868) |
| **Intensive Care Unit** | Admitted | 77(75.5%) | 9 (50%)/68 (81%) | 32.88 (3.563) | **6.094**/-0.712/NA | **0.013\***/0.480/NA |
| Not-admitted | 25(24.5%) | 9 (50%)/16 (19%) | 35.34 (4.620) |

**Table S4.** Results of univariate analysis comparing different patient groups and using a dichotomized cycle threshold (Ct) value of ≤ 33 to discriminate infectious vs. no infectious patients for the third test.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variables** | **Dichotomization** | **Counts (%)** | **Infectious/Non-infectious** | **Cycle values (median, standard deviation)** | **Statistic** | ***p* value** |
| **x2/t/Pearson** |
| Age | >65 | 3 (15%) | 1 (33.33%)/2 (66.66%) | 32.81 (4.870) | 1.133/-0.575/-0.403 | 0.142/0.573/0.078 |
| <65 | 17 (85%) | 2 (66.7%)/15 (88.2%) | 35.63 (3.558) |
| Comorbidities | Presence | 6 (30%) | 1 (33.3%)/5 (29.4%) | 33.81 (2.082) | 1.081/-1.793/NA | 0.150/0.090/NA |
| Absence | 14(70%) | 2 (66.7%)/12 (70.6%) | 37.44 (3.979) |
| Gender | Male | 10(50%) | 2 (33.3%)/8 (47.1%) | 34.78 (4.066) | 0.023/-1.121/NA | 0.636/0.277/NA |
| Female | 10(50%) | 1 (66.7%)/9 (52.9%) | 37.44 (2.568) |
| White blood cells (normal 4.5 -10) | Normal | 6 (10%) | 0 (0%)/6 (35.3%) | 35.86 (3.813) | 0/-0.574/0.207 | 1/0.573/0.382 |
| Abnormal | 14(80%) | 3 (100%)/11 (64.7%) | 34.59 (3.262) |
| Lymphocytes(normal 1.1-3.2) | Normal | 8(40%) | 2 (66.7%)/6 (35.3%) | 37.15 (2.256) | 0.506/-1.189/0.222 | 0.320/0.250/0.347 |
| Abnormal | 12(60%) | 1 (33.3%)/11 (64.7%) | 34.13 (4.419) |
| MonocytesNormal (0.3-0.8) | Normal | 17(85%) | 3 (100%)/14 (82.4%) | 35.34 (3.707) | 0.015/0.736/-0.299 | 1/0.471/0.200 |
| Abnormal | 3 (15%) | 0 (0%)/3(17.6%) | 36.73 (3.670) |
| **Neutrophi****Ls (normal 2.2-4.8)** | Normal | 7 (35%) | 1 (33.3%)/6 (35.3%) | 34.70 (2.923) | 0/0.086/0.159 | 1/0.932/0.504 |
| Abnormal | 13 (65%) | 2 (66.7%)/11 (64.7%) | 35.86 (4.441) |
| Eosinophils(normal 0.5-2.9) | Normal | 0 (0%) | 0 (0%)/0 (0%) | 0 | NA/NA/-0.068 | NA/NA/0.776 |
| Abnormal | 20(100%) | 3 (100%)/17 (100%) | 35.34 (3.660) |
| Platelets (normal 150-450) | Normal | 18(90%) | 3 (100%)/15 (88.2%) | 35.63 (3.943) | 0.094/0.229/-0.061 | 1/0.820/0.799 |
| Abnormal | 2(10%) | 0 (0%)/2 (11.8%) | 34.78 (1.590) |
| Basophils (normal <0.1) | Normal | 20(100%) | 3 (100%)/ 17(100%) | 35.34 (3.660) | 0/NA/-0.220 | 1/NA/0.352 |
| Abnormal | 0 (0%) | 0 (0%)/0 (0%) | 0 |
| Hemoglobin (normal 14-18) | Normal | 13(65%) | 3 (100%)/10 (58.8%) | 36.62 (4.054) | 0/-0.488/-0.431 | 1/0.631/0.058 |
| Abnormal | 7(35%) | 0 (0%)/7 (41.2%) | 34.56 (2.741) |
| Hematocrit (normal 42-52) | Normal | 12(60%) | 2 (66.7%)/10 (58.8%) | 37.3 (3.970) | 1.273/-1.480/-0.368 | 0.131/0.156/0.110 |
| Abnormal | 8(40%) | 1 (3.3%)/7 (41.2%) | 34.24 (2.766) |
| Erythrocytes (normal 4.7-6.1) | Normal | 14(70%) | 3 (100%)/11 (64.7%) | 37.15 (3.962) | 0.088/-0.825/0.316 | 0.580/0.420/0.175 |
| Abnormal | 6(30%) | 0 (0%)/6(35.3%) | 34.34 (3.080) |
| C Reactive Protein (normal<3.1) | Normal | 3(15%) | 0 (0%)/3 (17.7%) | 35.89 (2.192) | 0.023/-0.383/0.173 | 1/0.706/0.082 |
| Abnormal | 17(85%) | 3 (100%)/14 (82.3%) | 35.63 (3.933) |
| Creatinine (normal 0.7-1.3) | Normal | 0 (0%) | 0 (0%)/0 (0%) |  | 0/-1.141/-0.298 | 0.627/0.269/0.215 |
| Abnormal | 20 (100%) | 3 (100%)/17 (100%) | 34.34 (4.094) |
| Intensive Care Unit | Admitted | 40(78.4%) | 5 (100%)/35 (76.1%) | 36.09 (2.449) | 0.211/1.208/NA | 0.565/0.244/NA |
| Not admitted | 11(21.6%) | 0 (0%)/11(23.9%) | 34.70 (4.018) |

**Table S5.** Results of univariate analysis comparing different patient groups and using a dichotomized cycle threshold (Ct) value of ≤ 24 to discriminate infectious vs. no infectious patients for the first test. In this table only chi squared is calculated because the group comparisons are the same as in the previous tables.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variables** | **Dichotomization** | **Counts (%)** | **Infectious/Non-infectious** | **Cycle values (median, standard deviation)** | **Statistic** | ***p* value** |
| **x2**  |
| Age | >65 | 8 (13.12%) | 2 (18.2%)/6 (12%) | 28.87 (4.167) | 0.003 | 0.627 |
| <65 | 53 (86.89%) | 9 (81.8%)/44 (88%) | 29.66 (7.031) |
| Comorbidities | Presence | 14 (23.73%) | 4 (30.8%)/10 (21.7%) | 30.24 (5.988) | 0.489 | 0.432 |
| Absence | 45 (76.3%) | 9 (69.2%)/36 (78.3%) | 28.7 (7.050) |
| **Gender** | Male | 38 (61.29%) | 1 (7.7%)/23(46.9%) | 27.83 (7.623) | **6.579** | **0.004** |
| Female | 24 (38.71%) | 12 (92.3%)/26 (53.1%) | 29.98 (4.473) |
| White blood cells (normal 4.5 -10) | Normal | 46 (82.14%) | 11 (84.6%)/35 (72.9%) | 29.35 (7.167) | 0 | 1 |
| Abnormal | 10 (17.86%) | 2 (15.4%)/8 (27.1%) | 28.08 (4.457) |
| Lymphocytes(normal 1.1-3.2) | Normal | 37 (66.07%) | 5 (38.5%)/32 (76.2%) | 29.66 (5.101) | -1.154 | 0.253 |
| Abnormal | 19 (33.93%) | 8 (61.5%)/11 (23.8%) | 26.74 (8.918) |
| MonocytesNormal (0.3-0.8) | Normal | 52 (92.9%) | 11 (84.6%)/41 (95.4%) | 29.35 (6.828) | 0 | 1 |
| Abnormal | 4 (7.14%) | 2 (15.4%)/2 (4.7%) | 26.11 (5.347) |
| **Neutrophi****Ls (normal 2.2-4.8)** | Normal | 35 (62.5%) | 5 (38.5%)/30 (69.8%) | 30.29 (5.322) | 0.913 | 0.298 |
| Abnormal | 21 (37.5%) | 8 (61.5%)/13 (30.2%) | 25.74 (8.428) |
| Eosinophils(normal 0.5-2.9) | Normal | 1(1.79%) | 1 (7.7%)/0 (0%) | 19.34 (NA) | 0.594 | 0.196 |
| Abnormal | 55(98.2%) | 12 (92.3%)/43 (100%) | 29.04 (6.689) |
| Platelets (normal 150-450) | Normal | 45(80.36%) | 9 (69.2%)/36 (83.7%) | 29.66 (5.488) | 0 | 1 |
| Abnormal | 11(19.64%) | 4 (30.8%)/7 (16.3%) | 26.74 (10.001) |
| Basophils (normal <0.1) | Normal | 54(96.4%) | 12 (92.3%)/42 (97.7%) | 28.87 (6.698) | 0 | 1 |
| Abnormal | 2(3.6%) | 1 (7.7%)/1 (2.3%) | 25.88 (10.303) |
| Hemoglobin (normal 14-18) | Normal | 32(57.14%) | 8 (61.5%)/24 (55.8%) | 30.26 (7.562) | 0.021 | 0.741 |
| Abnormal | 24(42.86%) | 5 (38.5%)/19 (44.2%) | 28.47 (5.6) |
| Hematocrit (normal 42-52) | Normal | 24(42.86%) | 7 (53.84%)/17 (39.5%) | 27.9 (8.316) | 0.285 | 0.501 |
| Abnormal | 32(57.14%) | 6 (46.2%)/26 (60.5%) | 29.35 (5.268) |
| Erythrocytes (normal 4.7-6.1) | Normal | 35(62.5%) | 7 (53.9%)/28 (65.1%) | 28.27 (7.263) | 0.068 | 0.73 |
| Abnormal | 21(37.5%) | 6 (46.2%)/15 (34.9%) | 28.70 (5.9) |
| C Reactive Protein (normal<3.1) | Normal | 6(11.11%) | 0 (0%)/6 (14.3%) | 33.11 (4.383) | 0.603 | 0.327 |
| Abnormal | 48(88.88%) | 12 (100%)/36 (85.7%) | 28.47 (6.665) |
| Creatinine (normal 0.7-1.3) | Normal | 38(70.37%) | 9 (75%)/29 (69.1%) | 28.87 (7.404) | 0 | 1 |
| Abnormal | 16(29.63%) | 3 (25%)/13 (30.9%) | 29.98 (4.827) |
| Intensive Care Unit | Admitted | 11(19.3%) | 4 (36.3%)/7 (15.2%) | 27.88 (6.782) | 1.372 | 0.195 |
| Not admitted | 46(80.7%) | 7 (63.6%)/39 (84.8%) | 29.35 (6.851) |