

1 **Supplementary files**

2 **Figure S1. Expression pattern and immunological correlation of Siglec15 in**
3 **pan-cancers.** (A-C) The expression pattern of Siglec15 of pan-cancers in TCGA,
4 TCGA combined with GTEx, and Oncomine. The asterisks indicated a significant
5 statistical p value calculated with Mann-Whitney U test. (*P < 0.05; **P < 0.01; ***P
6 < 0.001). (D) The expression of Siglec15 in BLCA cohort in Oncomine. (E-F) The
7 expression of Siglec15 in cancer cell lines in BioGPS and CCLE. (G) The expression
8 of Siglec15 in normal tissues. (H) Quantitative RT-PCR (qPCR) estimation on
9 Siglec-15 mRNA levels in 30 paired bladder cancer and normal tissues.

10
11 **Figure S2. Prognostic analysis of Siglec15 for overall survival in pan-cancers.** (A)
12 The prognostic analyses of Siglec15 in pan-cancers using a univariate Cox regression
13 model. Hazard ratio >1 indicated a risk factor and hazard ratio <1 represented a
14 protective factor. (B-G) The prognostic analyses of Siglec15 in pan-cancers using
15 Kaplan-Meier method and log-rank test. Only cancers in which Siglec15 was a
16 significant prognostic biomarker were shown.

17
18 **Figure S3. Prognostic analysis of Siglec15 for disease specific survival in**
19 **pan-cancers.** (A) The prognostic analyses of Siglec15 in pan-cancers using a
20 univariate Cox regression model. Hazard ratio >1 indicated a risk factor and hazard
21 ratio <1 represented a protective factor. (B-G) The prognostic analyses of Siglec15 in
22 pan-cancers using Kaplan-Meier method and log-rank test. Only cancers in which
23 Siglec15 was a significant prognostic biomarker were shown.

24
25 **Figure S4. Prognostic analysis of Siglec15 for progression free survival in**
26 **pan-cancers.** (A) The prognostic analyses of Siglec15 in pan-cancers using a
27 univariate Cox regression model. Hazard ratio >1 indicated a risk factor and hazard
28 ratio <1 represented a protective factor. (B-G) The prognostic analyses of Siglec15 in
29 pan-cancers using Kaplan-Meier method and log-rank test. Only cancers in which
30 Siglec15 was a significant prognostic biomarker were shown.

31
32 **Figure S5. Correlations between Siglec15 and TMB, MSI in pan-cancers.** (A)
33 Correlation between Siglec15 and TMB in pan-cancers. (B) Correlation between
34 Siglec15 and MSI in pan-cancers. The asterisks indicated a significant statistical p
35 value calculated with spearman correlation analysis. (*P < 0.05; **P < 0.01; ***P <
36 0.001).

37
38 **Figure S6. Multi-omics analysis of Siglec15 in BLCA.** (A) CNV pattern of Siglec15
39 in BLCA. (B) Effect of Siglec15 CNV pattern on the expression of Siglec15 mRNA.
40 The asterisks indicated a significant statistical p value calculated with Mann-Whitney
41 U test (*P < 0.05; **P < 0.01; ***P < 0.001). (C) Effect of Siglec15 methylation on
42 the expression of Siglec15 mRNA. (D-E) The top 30 mutational genes in low and
43 high Siglec15 group respectively. The upper barplot showed the TMB, the number on

44 the right showed the mutation frequency. The right barplot represented the proportion
45 of variant types. (F) Overview of mutation profiles in BLCA.

46

47 **Figure S7. Correlations between Siglec15 and the tumor associated immune cells**
48 **calculated with TIMER algorithm.** The p value was calculated with spearman
49 correlation analysis.

50

51 **Figure S8. Correlations between Siglec15 and the tumor associated immune cells**
52 **calculated with CIBERSORT-ABS algorithm.** The p value was calculated with
53 spearman correlation analysis.

54

55 **Figure S9. Correlations between Siglec15 and the tumor associated immune cells**
56 **calculated with quanTIseq algorithm.** The p value was calculated with spearman
57 correlation analysis.

58

59 **Figure S10. Correlations between Siglec15 and the tumor associated immune**
60 **cells calculated with xCell algorithm.** The p value was calculated with spearman
61 correlation analysis.

62

63 **Figure S11. Correlations between Siglec15 and the tumor associated immune**
64 **cells calculated with TISIDB algorithm.** The p value was calculated with spearman
65 correlation analysis.

66

67 **Figure S12. Correlations between Siglec15 and the tumor associated immune**
68 **cells calculated with TIP algorithm.** The p value was calculated with spearman
69 correlation analysis.

70

71 **Figure S13. Correlations between Siglec15 and the tumor associated immune**
72 **cells calculated with MCP-counter algorithm.** The p value was calculated with
73 spearman correlation analysis.

74

75 **Figure S14. Correlations between Siglec15, tumor associated immune cells and**
76 **immune phenotypes.** (A) Correlations between Siglec15 and the effector genes of
77 five tumor associated immune cells . (B-E) Correlations between Siglec15 and four
78 critical marker genes of macrophages. (F) Expression of Siglec15, PD-L1, and CD8 in
79 the bladder cancer microarray (TMA) cohort were detected using
80 immunohistochemistry. Representative images of CD8, PD-L1, and Siglec15 in three
81 immune phenotypes were displayed. The scale bars correspond to 200 μ m. (G) CD8
82 positive rates in the three immune phenotypes in BLCA TMA cohort detected by
83 immunofluorescence. (H) Correlation between Siglec15 positive rates and CD8
84 positive rates detected using immunohistochemistry. (I) Correlation between PD-L1
85 positive rates and CD8 positive rates detected using immunofluorescence. (I)
86 Correlation between PD-L1 positive rates and Siglec15 positive rates detected using
87 immunofluorescence.

88

89 **Figure S15. Correlations between Siglec15 and the immunological status, the**
90 **enrichment scores of therapeutic signatures, and the molecular subtype in**
91 **GSE32894 cohort.** (A-C) Correlation between Siglec15 and immunomodulators,
92 effector genes of tumor associated immune cells, and inhibitory immune checkpoints
93 in BLCA. (D) Correlation between Siglec15 and enrichment scores of immunotherapy
94 predicted signatures. The asterisks indicated a significant statistical p value calculated
95 with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P < 0.001). (E) Correlation
96 between Siglec15 and molecular subtype and bladder cancer signatures. (F)
97 Correlation between Siglec15 and enrichment scores of therapeutic signatures,
98 including radiotherapy, targeted therapy.

99

100 **Figure S16. Correlations between Siglec15 and the immunological status, the**
101 **enrichment scores of therapeutic signatures, and the molecular subtype in**
102 **GSE31684 cohort.** (A-C) Correlation between Siglec15 and immunomodulators,
103 effector genes of tumor associated immune cells, and inhibitory immune checkpoints
104 in BLCA. (D) Correlation between Siglec15 and enrichment scores of immunotherapy
105 predicted signatures. The asterisks indicated a significant statistical p value calculated
106 with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P < 0.001). (E) Correlation
107 between Siglec15 and molecular subtype and bladder cancer signatures. (F)
108 Correlation between Siglec15 and enrichment scores of therapeutic signatures,
109 including radiotherapy, targeted therapy.

110

111 **Figure S17. Correlations between Siglec15 and the immunological status, the**
112 **enrichment scores of therapeutic signatures, and the molecular subtype in**
113 **IMvigor210 cohort.** (A-C) Correlation between Siglec15 and immunomodulators,
114 effector genes of tumor associated immune cells, and inhibitory immune checkpoints
115 in BLCA. (D) Correlation between Siglec15 and enrichment scores of immunotherapy
116 predicted signatures. The asterisks indicated a significant statistical p value calculated
117 with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P < 0.001). (E) Correlation
118 between Siglec15 and molecular subtype and bladder cancer signatures. (F)
119 Correlation between Siglec15 and enrichment scores of therapeutic signatures,
120 including radiotherapy, targeted therapy.

121

122 **Figure S18. Correlations between Siglec15 and the immune signatures in CR**
123 **subgroup of IMvigor210 cohort.** (A-C) Correlation between Siglec15 and
124 immunomodulators, effector genes of tumor associated immune cells, and inhibitory
125 immune checkpoints in BLCA. (D) Correlation between Siglec15 and enrichment
126 scores of immunotherapy predicted signatures. The asterisks indicated a significant
127 statistical p value calculated with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P
128 < 0.001).

129

130 **Figure S19. Correlations between Siglec15 and the immune signatures in PR**
131 **subgroup of IMvigor210 cohort.** (A-C) Correlation between Siglec15 and

132 immunomodulators, effector genes of tumor associated immune cells, and inhibitory
133 immune checkpoints in BLCA. (D) Correlation between Siglec15 and enrichment
134 scores of immunotherapy predicted signatures. The asterisks indicated a significant
135 statistical p value calculated with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P
136 < 0.001).

137

138 **Figure S20. Correlations between Siglec15 and the immune signatures in PD**
139 **subgroup of IMvigor210 cohort.** (A-C) Correlation between Siglec15 and
140 immunomodulators, effector genes of tumor associated immune cells, and inhibitory
141 immune checkpoints in BLCA. (D) Correlation between Siglec15 and enrichment
142 scores of immunotherapy predicted signatures. The asterisks indicated a significant
143 statistical p value calculated with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P
144 < 0.001).

145

146 **Figure S21. Correlations between Siglec15 and the immune signatures in SD**
147 **subgroup of IMvigor210 cohort.** (A-C) Correlation between Siglec15 and
148 immunomodulators, effector genes of tumor associated immune cells, and inhibitory
149 immune checkpoints in BLCA. (D) Correlation between Siglec15 and enrichment
150 scores of immunotherapy predicted signatures. The asterisks indicated a significant
151 statistical p value calculated with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P
152 < 0.001).

153

154 **Figure S22. Correlations between Siglec15 and the hyper-progression associated**
155 **genes, and the predictive accuracy of Siglec15 for molecular subtype in four**
156 **validation sets.** (A) Correlation between Siglec15 and CNV pattern of
157 hyper-progression associated genes in BLCA. The p value was calculated with Fisher
158 t test. (B) Correlation between Siglec15 and mRNA expression of hyper-progression
159 associated genes in BLCA. The asterisks indicated a significant statistical p value
160 calculated with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P < 0.001). (C-F)
161 The predictive value of Siglec15 for molecular subtype in four independent validation
162 sets, including an immunotherapy cohort (IMvigor210), a neoadjuvant-chemotherapy
163 cohort(GSE70691), and two general bladder cancer cohorts(GSE31684, GSE48277).

164

165 **Figure S23. Role of Siglec15 in predicting molecular subtypes and therapeutic**
166 **sensitivities in Xiangya cohort.** (A) Correlations between Siglec15 and molecular
167 subtypes and bladder cancer signatures. (B) ROC curves indicated the predictive
168 accuracy of Siglec15 in predicting molecular subtypes. (C) Correlations between
169 Siglec15 and enrichment scores of therapeutic signatures, including radiotherapy,
170 targeted therapy.

171

172 **Figure S24. Differentially expressed RNAs between the Siglec15 groups, the**
173 **immune score groups and the stromal score groups.** (A-F) Differential analysis of
174 RNAs between Siglec15 groups, immune score groups and stromal score groups. The
175 criterias for determining differentially expressed RNAs were set as adjusted P value <

176 0.01 and $|\log_{2}FC| > 1$. (G) 1500 common immune-related differential RNAs shown in
177 Venn diagram. (H-I) There is no intersection between RNAs up-expressed among
178 high Siglec15 group and RNAs up-expressed among high immune/stromal score
179 groups. Similarly, there is no intersection between RNAs down-expressed among high
180 Siglec15 group and RNAs down-expressed among high immune/stromal score groups.
181 (J) There is 1010 common RNAs between RNAs down-expressed among high
182 Siglec15 group and RNAs up-expressed among high immune/stromal score groups.
183 (K) There is 490 common RNAs between genes up-expressed among high Siglec15
184 group and RNAs down-expressed among high immune/stromal score groups.

185

186 **Figure S25. GO and KEGG analysis of 1500 common differentially expressed**
187 **RNAs.** (A-C) Biological process, cellular component, and molecular function of 1500
188 common differentially expressed RNAs. (D) KEGG analysis of 1500 common
189 differentially expressed RNAs.

190

191 **Figure S26. Performance of the IRS RNA-expression profiles in predicting**
192 **survival in five independent validation cohorts.** (A-E) Survival in high vs. low-IRS
193 patients depicted by KM plots and ROC curves to depict the accuracy of IRS in five
194 bladder cancer cohorts. (E) Survival in high vs. low-IRS patients depicted by KM
195 plots and ROC curves to depict the accuracy of IRS in GSE135222 (lung cancer).

196

197 **Figure S27. Performance of the IRS RNA-expression profiles in predicting**
198 **survival in subgroups of IMvigor210 cohort.** (A-H) Survival in high vs. low-IRS
199 patients depicted by KM plots and ROC curves to depict the accuracy of IRS in eight
200 subgroups of IMvigor210 cohort, including CR/PR subgroup, SD/PD subgroup, IC0
201 subgroup, IC1 subgroup, IC2 subgroup, TC0 subgroup, TC1 subgroup, TC2
202 subgroup.

203

204 **Figure S28. Performance of the IRS RNA-expression profiles in predicting**
205 **survival in subgroups of IMvigor210 cohort.** (A-H) Survival in high vs. low-IRS
206 patients depicted by KM plots and ROC curves to depict the accuracy of IRS in eight
207 subgroups of IMvigor210 cohort, including deserts phenotype subgroup, excluded
208 phenotype subgroup, inflamed phenotype subgroup, ECOG score0 subgroup, ECOG
209 score1 subgroup, ECOG score2 subgroup, pre-platinum therapy subgroup, no
210 pre-platinum therapy subgroup.

211

212 **Figure S29. IRS predicted the clinical response of cancer immunotherapy.**

213 (A) Correlation between Siglec15 and IRS. (B-C) Correlations between IRS and the
214 pan-cancer T cell inflamed score, and the inhibitory immune checkpoints. (D-E)
215 Correlations between the IRS and immunomodulators, and the tumor-associated
216 immune cells. (F) Correlations between the IRS and the activities of cancer immunity
217 cycles. (G) Correlations between the IRS and the enrichment score of immunotherapy
218 predicted pathways. The asterisks indicated a significant statistical p-value calculated

219 with Mann-Whitney U test (*P < 0.05; **P < 0.01; ***P < 0.001).

220

221 **Figure S30. Comparisons of the accuracy in predicting ICB response and**
222 **survival probability between IRS and TIDE algorithms in GSE78220.**

223 (A) The accuracy of two algorithms in predicting ICB response. (B-C) Survival in
224 high vs. low TIDE, and in high vs. low IRS patients depicted by KM plots. (D-E)
225 ROC curves to depict the accuracy of TIDE and IRS in predicting survival
226 probability.

227

228 **Figure S31. Comparisons of the accuracy in predicting ICB response and**
229 **survival probability between IRS and TIDE algorithms in GSE91061.**

230 (A) The accuracy of two algorithms in predicting ICB response. (B-C) Survival in
231 high vs. low TIDE, and in high vs. low IRS patients depicted by KM plots. (D-E)
232 ROC curves to depict the accuracy of TIDE and IRS in predicting survival
233 probability.

234

235 **Figure S32. Comparisons of the accuracy in predicting ICB response and**
236 **survival probability between IRS and TIDE algorithms in PMID29301960.**

237 (A) The accuracy of two algorithms in predicting ICB response. (B-C) Survival in
238 high vs. low TIDE, and in high vs. low IRS patients depicted by KM plots. (D-E)
239 ROC curves to depict the accuracy of TIDE and IRS in predicting survival
240 probability.

241

242 **Figure S33. Comparisons of the accuracy in predicting ICB response and**
243 **survival probability between IRS and TIDE algorithms in IMvigor210.**

244 (A) The accuracy of two algorithms in predicting ICB response. (B-C) Survival in
245 high vs. low TIDE, and in high vs. low IRS patients depicted by KM plots. (D-E)
246 ROC curves to depict the accuracy of TIDE and IRS in predicting survival
247 probability.

248

249

250 **Table S1.** Characters of data sets; Abbreviations of cancer types; Cox analysis in
251 TCGA BLCA cohort.

252

253 **Table S2.** Correlations between Siglec15 and 122 immunomodulators, PD-L1,
254 PD-1,CTLA-3, LAG-3.

255

256 **Table S3.** Comparisons of the cancer-immunity cycle activity between Siglec15
257 groups.

258

259 **Table S4.** Infiltration level of tumor associated immune cells in BLCA estimated by
260 using different algorithms.
261
262 **Table S5.** Comparisons of the effector genes of tumor associated immune cells
263 between Siglec15 groups.
264
265 **Table S6.** Comparisons of the inhibitory immune checkpoints between Siglec15
266 groups.
267
268 **Table S7.** The pan-cancer T cell inflamed score of TCGA BLCA patients.
269
270 **Table S8.** The CNV patterns of hyper-progression associated genes in BLCA.
271
272 **Table S9.** Detailed information of immunotherapy predicted signatures, bladder
273 cancer signatures and other therapeutic signatures.
274
275 **Table S10.** Comparisons of therapeutic sensitivity of drug genes used in BCLA
276 between Siglec15 groups.
277
278 **Table S11.** Molecular subtypes of patients in TCGA BLCA cohort.
279
280 **Table S12.** Expression of Siglec15, PD-L1, and CD8 in the clinical validation set.
281
282 **Table S13.** Comparisons of the enrichment score of therapeutic predicted pathways
283 between Siglec15 groups.
284
285 **Table S14.** Correlations between Siglec15 and immunomodulators in Xiangya cohort.
286
287 **Table S15.** 1500 common differentially expressed RNAs.
288
289 **Table S16.** GO, KEGG analysis of 1500 common differentially expressed RNAs.
290
291 **Table S17.** Protein-protein interaction analysis of 1500 differentially expressed
292 RNAs.
293
294 **Table S18.** Univariate cox analysis of 1500 common differentially expressed RNAs.
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297