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Supporting information for article:

Molecular determinants of vascular transport of dexamethasone in COVID-19 therapy

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A

1	10	20	30	40	50	60		
ESA	DTHKSEIAKRFNDLGEKEFEGLVIVAFS	SYLOQCPFFEDHV	KLVNEVTEFAK	KCAADESAE				
HSA	DTHKSEIAKRFNDLGEKEFKA	LVIAF	SYLOQCPFFEDHV	KLVNEVTEFAK	TCVADESAE			
70	80	90	100	110				
ESA	NCDKSLEHTLFGDKLCTVATLR	ATYGEIADCC	EKQEPERNECFLIMKDDNPNLPKL	KPEP				
HSA	NCDKSLEHTLFGDKLCTVATLR	ATYGEIADCC	EKQEPERNECFLIMKDDNPNLPRLV	RPEK				
120	130	140	150	160	170			
ESA	DAQQCARFQEDPDKFLGKYLYE	VARRHPYFYGP	PELLFHARESYKA	FTTECCPADDKLA	CIP			
HSA	DAQQCARFHDNEETFLRKYLYE	IARHRPYFYGP	PELLFHAKRYKA	FTTECCQADKA	ACCLP			
180	190	200	210	220	230			
ESA	KLDALKERILLSSAKER	RLKCSE	QFGERAVKAWA	VARLSQKFPKA	FAEVSKI	VTDLTK		
HSA	KLDALKERDEGKA	SSAK	RLKCSE	QFGERAVKAWA	VARLSQKFPKA	FAEVSKI	VTDLTK	
240	250	260	270	280	290			
ESA	VHRECCCHGDLLE	CADDRADLAKYICE	HQDSISGKLKACCD	KPLQKSHCIAEV	KEDDILPS			
HSA	VHRECCCHGDLLE	CADDRADLAKYICE	HQDSISSKLKE	CCEKPLQKSHCIAEV	ENDEMPA			
300	310	320	330	340	350			
ESA	DLPALAADF	AEDKEICKYKD	AKDVFLGTFLYEYS	RRHPDYSVS	LLPLIAKTYEA	TLEKC		
HSA	DLPALAADF	VESKDKVCKNYAB	AKDVFLGTFLYEY	ARRHPDYSVV	LLPLIAKTYEA	TLEKC		
360	370	380	390	400	410			
ESA	CAPADPACYRTV	VFDCPTPLVEEP	KSLVRKNC	DLFEEVGEYD	FQNAIIVRYT	KKAPQVST		
HSA	CAPADPACYAKV	VFDFEPPLVEEQNL	IKONC	ELFEOLGEYK	FQNAIIVRYT	KKVPQVST		
420	430	440	450	460	470			
ESA	PTLVEIGRTLGKVGS	RCCR	LPESEPCSPNH	ALALINR	LCVLHEKTPVSEKI	TKCCTS		
HSA	PTLVEVSNMLGKVGS	RCCR	PEAKMPC	ARDYL	SVVLNQLCVLHEKTPVSDRV	TKCCTS		
480	490	500	510	520	530			
ESA	LAARRPCFSALE	LDEGYVPKE	RATFTFHADIC	TLPEDE	ERQIKKQ	QAI	ELVKHKPKAT	
HSA	LVNRRPCFSALE	VDET	YVPKE	NAETFTFHADIC	TLSEKER	QIKKQ	QAI	ELVKHKPKAT
*								
540	550	560	570	580				
ESA	KEQLKT	VUGNES	AFVAKCC	GREDK	CF			
HSA	KEQLKAV	MDDFAAFV	KEKCC	KADDK	CF			

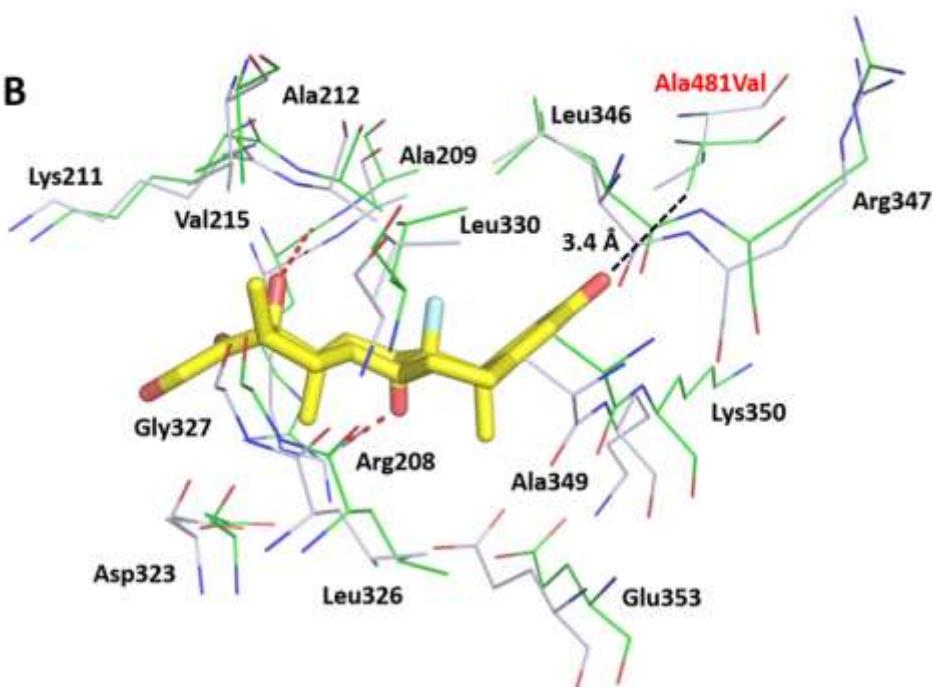
B

Figure S1 Comparison of ESA and HSA. **(A)** Alignment of ESA and HSA sequences. Identical residues are highlighted in red; residues involved in dexamethasone binding to DS7 in ESA and analogous residues in HSA are marked with stars. **(B)** Superposition of DS7 binding dexamethasone in ESA (PDB ID: 6XK0) and analogous site in HSA (PDB ID: 4K2C). Dexamethasone is shown in stick representation with carbon atoms in yellow, oxygen atoms in red and fluoride atom in cyan. Residues within 5 Å are shown in line representation, residues from ESA are shown with carbon atoms in green while residues from HSA are shown with carbon atoms in gray, oxygen and nitrogen atoms are red and blue, respectively. Residue numbers correspond to positions in ESA. Ala481 is the only residue at this site that differ in HSA (Val in HSA). Hydrogen bonds are shown as red dashes.

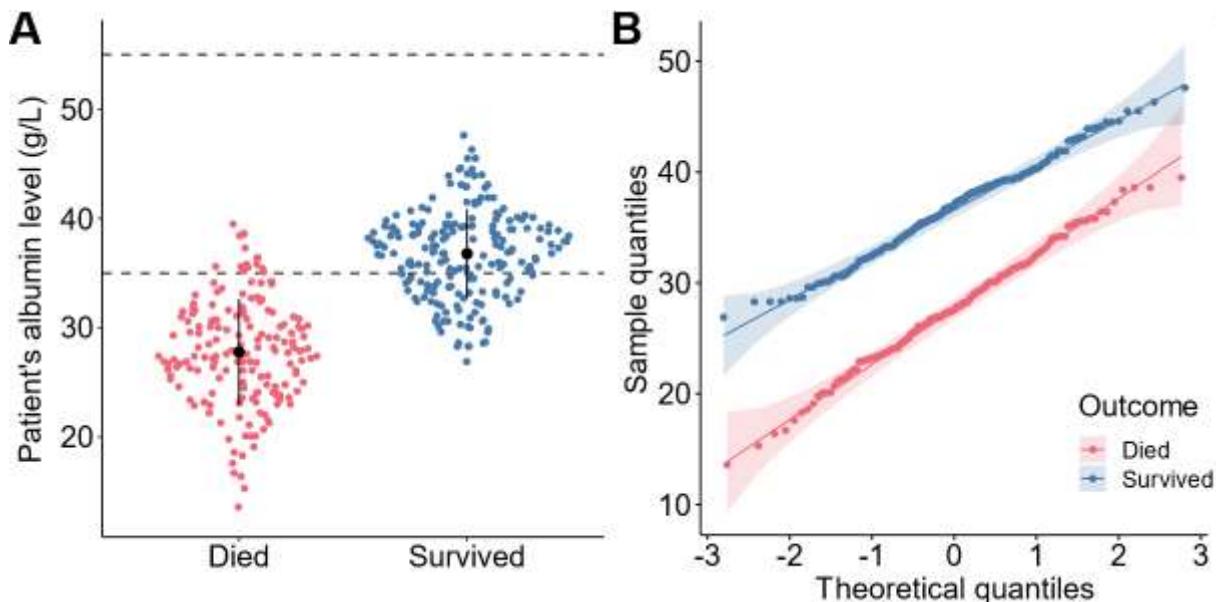


Figure S2 Distribution of COVID-19 patients grouped by outcome. **(A)** Violin strip charts (sina plots) of patients that died (red) and (survived), after being admitted to Tongji Hospital. **(B)** Q-Q plot for the albumin levels of patients that died and survived COVID-19. The shaded areas represent 95% confidence intervals.

Table S1 Associations between gender, age, glucose level, albumin level and survival or death in univariate and multivariate logistic regression analyses.

Analysis	Odds ratio of survival (95% CI)	p-values
Univariate analysis (unadjusted models)		
Albumin level	1.56 (1.44–1.71)	<i>p</i> < 0.001
Female	2.82 (1.83–4.37)	<i>p</i> < 0.001
Age	0.90 (0.88–0.92)	<i>p</i> < 0.001
Glucose level	0.76 (0.70–0.81)	<i>p</i> < 0.001
Multivariate analysis (adjusted model)		
Albumin level	1.51 (1.37–1.69)	<i>p</i> < 0.001
Female	2.19 (1.04–4.72)	<i>p</i> < 0.05
Age	0.92 (0.89–0.94)	<i>p</i> < 0.001
Glucose level	0.89 (0.81–0.96)	<i>p</i> < 0.01

Table S2 RMSD values [Å] for aligned C α atoms of ESA-steroid complexes and ligand-free ESA and HSA structures.

	ESA-dexamethasone (PDB ID: 6XK0)	ESA-testosterone (PDB ID: 6MDQ)	ESA-ligand free (PDB ID: 3V08)	HSA-ligand free (PDB ID: 4K2C)
ESA-dexamethasone (PDB ID: 6XK0)	-	1.1	0.4	1.6
ESA-testosterone (PDB ID: 6MDQ)	1.1	-	1.1	1.6
ESA-ligand free (PDB ID: 3V08)	0.4	1.1	-	1.7
HSA-ligand free (PDB ID: 4K2C)	1.6	1.6	1.7	-

Table S3 Comparison of mean (s.d) of albumin levels and patient outcomes in different studies. N denotes the sample size of the study. ICU: Intensive Care Unit, ARDS: Acute Respiratory Distress Syndrome.

Country	N	Group 1 (g/L)	Group 2 (g/L)	Study
Spain	48	ICU: 29.0 (5.2)	Non-ICU: 39.2 (4.2)	(Rica <i>et al.</i> , 2020)
China*	21	Severe: 29.6 (28.6–33.0)	Moderate: 37.2 (35.8–38.8)	(Chen <i>et al.</i> , 2020)
China*	201	ARDS: 30.4 (27.15–33.35)	Non-ARDS: 33.7 (30.95–36.30)	(Wu <i>et al.</i> , 2020)
China*†	2623	Died: 31.1 (27.9–34.2)	Non-critical: 36.6 (33.2–40.4) Critical: 32.2 (29.6–35.7)	(Huang <i>et al.</i> , 2020)
China‡	910	Severe: 35.0 (2.4)	Non-severe: 40.5 (2.2)	(Aziz <i>et al.</i> , 2020)
China	373	Died: 27.8 (4.9)	Survived: 36.8 (4.1)	current

*Albumin level characterized by the median and quartiles (Q1–Q3); †Albumin levels measured at admission; ‡Meta-analysis of data from different hospitals

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