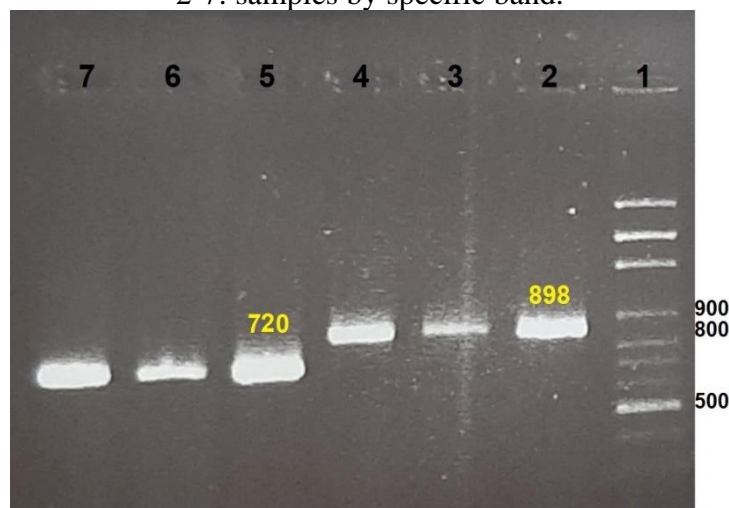


Appendix 1. Some PCR products sent for nucleotide sequencing. 1. Ladder marker 100-1000 bp, 2-7: samples by specific band.



Appendix 2. Mutation analysis of 83 isolates investigated in the study.

	Code	Collection date	Location	Variant	Detected Mutations	Sequencing area of S Protein
1.	2-2511	Feb 2021	Tehran		D138Y, M177I, S477N	42-257 317-575
2.	14-2511	Feb 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144	40-260 306-493
3.	5-2311	Feb 2021	Tehran		---	40-257 308-575
4.	6-2311	Feb 2021	Tehran		D138Y, S477N	42-156 317-575
5.	26-2311	Feb 2021	Tehran	B.1.1.529 (Omicron)	del209/210	40-257 306-433
6.	37-3011	Feb 2021	Tehran	B.1.1.529 (Omicron)	del209/210	43-260 312-545
7.	8-512	Feb 2021	Tehran		D138Y, S477N	46-260 326-578
8.	31-612	Feb 2021	Tehran		D138Y	54-260 527-578
9.	24-612	Feb 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D	52-257 317-578
10	17-612	Feb 2021	Tehran		D138Y, S477N	53-260 317-578
11	5-312	Feb 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D	43-260 310-575
12	5-812	Feb 2021	Tehran		---	52-257 311-578
13	15-912	Feb 2021	Tehran		D138Y, S477N	42-261 316-579
14	25-1012	Feb 2021	Tehran		N501Y, A570D, D614G	53-257 448-704
15	32-1012	Feb 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D,	59-260 472-606
16	17-1112	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, N501Y, A570D	48-260 316-576

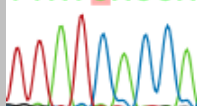
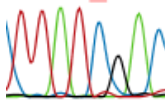
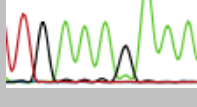

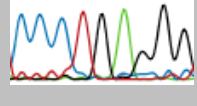
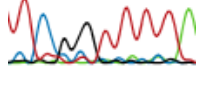
17	36-1112	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100T	55-260 391-443
18	39-1212	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, N501Y,A570D, D614G	42-202 425-702
19	53-1212	Mar 2021	Tehran	B.1.1.7 (Alpha)	del143/144, I100T, Q173H, N501Y,A570D, D614G	72-260 431-683
20	27-1312	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y,A570D, D614G	48-242 425-699
21	28-1312	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, N501Y,A570D, D614G	50-260 425-703
22	54-1412	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, N501Y,A570D, D614G, P681H	43-205 425-702
23	19-1512	Mar 2021	Tehran		D614G	55-231 429-663
24	31-1612	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, N501Y, D614G	43-250 425-703
25	19-1312	Mar 2021	Tehran		Q218E, A222V	49-260 448-565
26	22-1312	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D, D614G	42-261 428-702
27	40-1412	Mar 2021	Tehran		D138Y, S477N, D614G	42-260 429-702
28	43-1412	Mar 2021	Tehran	B.1.1.529 (Omicron)	del209/210, Y145H, G181R, D614G	42-261 437-702
29	1-1712	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D, D614G	50-261 429-703
30	40-1712	Mar 2021	Tehran	B.1.1.529 (Omicron)	del209/210, D614G	42-258 429-704
31	17-1812	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100C, N501Y, D614G	42-260 437-705
32	22-1812	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D, D614G	42-258 435-702
33	36-1812	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D, D614G	42-260 429-703
34	8-1112	Mar 2021	Tehran	B.1.1.529 (Omicron)	del209/210,	42-257 325-576
35	16-2511	Feb 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144, I100C	40-229
36	27-1412	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144	40-260
37	31-2511	Feb 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144	49-255
38	4-1212	Mar 2021	Tehran		Y145H, G181R, 209/210,	42-257
39	12-1712	Mar 2021	Tehran	B.1.1.7 (Alpha)	del69/70, del143/144	49-255
40	10-1012	Feb 2021	Tehran		D138Y	70-259
41	32-1312	Mar 2021	Tehran		S477N, D614G	425-700
42	9-1312	Mar 2021	Tehran		N501Y,A570D, D614G	429-702
43	36-2911	Feb 2021	Tehran		N501Y,A570D	311-576
44	20-112	Feb 2021	Tehran		---	305-475
45	19-212	Feb 2021	Tehran		---	306-416
46	11-2611	Feb 2021	Tehran		S477N	348-575

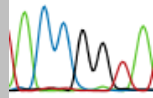
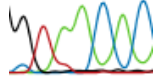
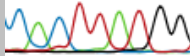
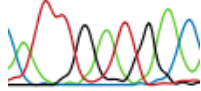
47	69-2811	Feb 2021	Tehran	B.1.617 (Delta)	L452R	348-575
48	63-2811	Feb 2021	Tehran		S477N,	311-576
49	36-3011	Feb 2021	Tehran		--	332-575
50	41-1212	Mar 2021	Tehran		D614G, N679K	429-705
51	17-1212	Mar 2021	Tehran		D614G	428-702
52	38-1312	Mar 2021	Tehran		D614G	423-702
53	26-1612	Mar 2021	Tehran		N501Y, A570D, D614G	423-694
54	3-1112	Mar 2021	Tehran		N501Y	325-578
55	9 (29878)	Dec 2020	Bushehr		D614G	65-240 442-641
56	1 (29092)	Dec 2020	Bushehr	B.1.1.529 (Omicron)	H49Y, del209/210, D614G	48-261 434-703
57	6 (29439)	Dec 2020	Bushehr		D138Y, S477N, D614G	42-260 429-702
58	8 (29864)	Dec 2020	Bushehr		D138Y, S477N, D614G	43-251 434-699
59	10 (29849)	Dec 2020	Bushehr		---	42-257 432-453
60	23 (9)	Apr 2021	Isfahan	B.1.1.7 (Alpha)	del69/70, del143/144	42-260
61	22 (7)	Apr 2021	Isfahan	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, N501Y, A570D, D614G	42-261 442-660
62	25 (13)	Apr 2021	Isfahan	B.1.1.7 (Alpha)	del143/144, N501Y, D614G	71-184 448-681
63	28 (18)	Apr 2021	Isfahan	B.1.1.7 (Alpha)	del143/144, I100T	75-198
64	1-0502	Apr 2021	Arak	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, A570D, D614G	51-258 432-674
65	2-1702	May 2021	Arak	B.1.1.7 (Alpha)	del69/70, del143/144	42-260 434-493
66	5-0602	Apr 2021	Arak	B.1.1.7 (Alpha)	del143/144, I100T	71-261 445-576
67	12-0502	Apr 2021	Arak	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, N501Y, A570D, D614G, P681H	54-170 427-703
68	14-0502	Apr 2021	Arak	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, A570D, D614G, P681H	48-260 429-704
69	19-0202	Apr 2021	Arak	B.1.1.7 (Alpha)	del69/70, del143/144, I100T, D614G	47-260 429-703
70	20-0502	Apr 2021	Arak	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, D614G	38-260 428-703
71	14-1102	May 2021	Arak	B.1.1.7 (Alpha)	del69/70, del143/144, N501Y, A570D, D614G, P681H	51-260 434-704
72	11(1-2641)	Dec 2020	Qom	B.1.1.529 (Omicron)	del210/211, D614G	54-237 445-676
73	12(2)	Dec 2020	Qom		---	53-260
74	18 (8-2635)	Dec 2020	Qom		---	89-209
75	20(10-2672)	Dec 2020	Qom		---	56-209 460-525
76	1(417)	Dec 2020	Shushtar		D614G	46-261 428-704
77	2(401)	Dec 2020	Shushtar	B.1.1.529 (Omicron)	del209/210, D614G,	43-260 437-703

78	3(251)	Dec 2020	Shushtar		A222V, D614G	42-260 428-699
79	5(1454)	Dec 2020	Shushtar		A575S	45-258 436-702
80	6(1046)	Dec 2020	Shushtar	B.1.1.529 (Omicron)	del209/210, D614G	42-258 426-705
81	7(253)	Dec 2020	Shushtar		D614G,	53-260 428-704
82	8(1410)	Dec 2020	Shushtar	B.1.1.529 (Omicron)	del209/210, D614G	49-256 431-705
83	9(693)	Dec 2020	Shushtar	B.1.1.529 (Omicron)	del209/210, D614G	53-260 428-704

Appendix 3. The chromatogram and nucleotide to amino acid translation for each mutation

	Amino acid change		
	Variation	Sequence graph	Chromatogram
1.	H49Y	<p>48 49 50</p> <p>L H S</p> <p>T A C A T T C</p> <p>21710</p> <p>L Y S</p> <p>T A T A T T C</p>	
2.	Δ69/70	<p>67 68 69 70 71</p> <p>A I H V S</p> <p>G C T A T A C A T G T C T C T</p> <p>21770</p> <p>A S</p> <p>G C T A T C T C T</p>	
3.	I100C	<p>99 100 101</p> <p>N I I</p> <p>A C A T A A T A</p> <p>21860</p> <p>N T I</p> <p>A C A C A A T A</p> <p>180</p>	
4.	D138Y	<p>137 138 139</p> <p>N D P</p> <p>A T G A T C C</p> <p>290</p> <p>N Y P</p> <p>A T T A T C C</p>	
5.	Δ143/144	<p>142 143 144 145</p> <p>G V Y Y</p> <p>G G T G T T T A T T A C</p> <p>21990</p> <p>G Y</p> <p>G G T G T T T A C</p>	

6.	Y145H	<p>144 145 146</p> <p>Y Y H</p> <p>A T T A C C A</p> <p>Y H H</p> <p>A T C A C C A</p>	<p>TTATCACCA</p> 
7.	M177I	<p>176 177 178</p> <p>L M D</p> <p>T T A T G G A</p> <p>22090</p> <p>L I D</p> <p>T T A T C G A</p> <p>410</p>	<p>TTATCGAC</p> 
8.	G181R	<p>180 181 182</p> <p>E G K</p> <p>A A G G A A A</p> <p>E R K</p> <p>A A A G A A A</p> <p>420</p>	<p>TGAAAGAAA</p> 
9.	Δ209/210	<p>208 209 210 211</p> <p>T P I N</p> <p>C G C C T A T T A A</p> <p>22190</p> <p>T N</p> <p>C G C C T A A A</p> <p>500 501</p>	<p>CCTA--ATT</p> 
10.	Q218E	<p>217 218 219</p> <p>P Q G</p> <p>C T C A G G G</p> <p>P E G</p> <p>C T G A G G G</p> <p>510</p>	<p>CCCTGAGGG</p> 
11.	A222V	<p>221 222 223</p> <p>S A L</p> <p>C G G C T T T</p> <p>22230</p> <p>S V L</p> <p>C G G T T T T</p>	<p>TCGGTTT</p> 

12.	L452R	<div>451452453</div> <div>YLY</div> <div>TACCTGTA</div> <div>22920</div> <div>YRY</div> <div>TACC GGTA</div> <div>310</div>	<div>ACCGGTA</div> 
13.	S477N	<div>476477478</div> <div>GS T</div> <div>G TAGCAC</div> <div>22990</div> <div>GN T</div> <div>GTAACAC</div> <div>480</div>	<div>GTAACAC</div> 
14.	N501Y	<div>500501502</div> <div>T N G</div> <div>CTAATGG</div> <div>T Y G</div> <div>CTTATGG</div>	<div>CACTTATGG</div> 
15.	A570D	<div>569570571</div> <div>I A D</div> <div>TTGCTGA</div> <div>23270</div> <div>ID D</div> <div>TTGATGA</div>	<div>ATTGATGAC.</div> 
16.	A575S	<div>574575576</div> <div>D A V</div> <div>ATGCTGTC</div> <div>23290</div> <div>D S V</div> <div>ATTCTGTC</div> <div>420</div>	<div>CTGATTCTGT</div> 