

Article

Anti-infective **screening** of selected nine cannabinoids against *Clostridium perfringens* **and** influenza A (H5N1) **neuraminidases**, and SARS-CoV-2 **main protease and spike protein interactions**

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Supplementary file

Figure S1. Protein pairwise alignment of neuraminidase between influenza A and *C. perfringens*²

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Figure S1. Protein pairwise alignment of neuraminidase between influenza A and *C. perfringens*

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Alignment:   Global Protein alignment.
Parameters:  Scoring matrix:  BLOSUM 62

Mol 1 ABU94738.1 (1 to 449)      Mol 2 WP_224383915.1 (1 to 382)
Number of sequences to align:  2
Total length of aligned sequences with gaps:  480 aas
Settings:    Similarity significance value cutoff:  >= 90%

Summary of alignment results:
Total length of aligned sequences with gaps:  480 aas
Matching bases:  81 / 480    (16%)
Gaps:           129 / 480    (26%)

ABU94738.1      1  MNENQKINTIGSICMVIGIVSLMLQIGNMISIWSSHSIQTGNGHQAEFIINANFLTENA
WP_224383915.1  1  MYNNNN---TFEKNLDISHKPEPLILFNKDNNIWNSSKYFR-----IENIQILNDGI

ABU94738.1      60  VASVILAGNSSSLCPVRGWAVHSDNSIRIGSKGDVVFVIREPFIICSHLECRTEFFITCG--
WP_224383915.1  49  ILTF-----SDIRVNGEDD-----HAYIDIASARSTDFGHTWSYD

ABU94738.1     118  ALINDKHNS--GTVKIRSPHRTLMSCPVGEAPSPYNRFFESVAWSASACHDGTSLWTIG
WP_224383915.1  84  IAKNNNRIDSTYSFVMISTIVIT-NTCRILLIAGSNNTINGN---WAMITSARRSTWVQM

ABU94738.1     175  IEGFDNGAVAVLKYNGIITDIKSWNNILRTQESACACVNGSCFTVMTDGFPSNQGASYR
WP_224383915.1  140  IYSDDNGI-----TWSNNIDLTKESEKVKNOESNTIGWLCGVGSG-----

ABU94738.1     235  IFRMERGKIVKVEL---NAPNHYEECSQYEDAGEITCVCRDNWHGSNRPVVSFNQNL
WP_224383915.1  180  -IVMDGIIIVMPAQISLRENNENNYSLIYSHDNGE-----TWIMGNKV-----

ABU94738.1     291  EYQIGYICSGVFGDNPEPNDGTGSCGFPVSPNGAYGIRGFSFRYENGWVIGRIKSTNSRS
WP_224383915.1  224  -----PNSNTSENMVIELGALIM---STFYDYSGYRAAYISHDLGST

ABU94738.1     351  EEWIWF-NGWTEIDSNFSVKQDIVAITDWSGSGSFVQHPETIGLDGIRPCFWVLI--
WP_224383915.1  264  WE-IYEPLNGKVIITCKGSGCQGSFIKATISTGHRIGLISAPKNTNGEYIRDNIAYVMIDE

ABU94738.1     408  ----RG-----RPKESTIWTSGSSSFCGUNSDTW-----SWSWEDGALPFTIDK
WP_224383915.1  323  DDLKGVQEICIPYFKDGNKLGGSSCSFRNGHLSIVYEANGNIEYQDLTPYYSLINKE

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ABU94738.1 is influenza A viral neuraminidase, while WP_224383915.1 is *C. perfringens* neuraminidase.

Table S1. Potency index value and calculation from antibacterial and antiviral neuraminidase experiments

Compound	Influenza A neuraminidase				bacterial neuraminidase			Potency Index*
	Screen	IC50 (μM)	Potency Index	modified Potency Index	Screen	IC50 (μM)	Potency Index	
THC	100.00	38.34	38.34	38.34	99.53	20.3	1.85	20.30
CBD	100.00	20.98	20.98	20.98	97.24	4.53	0.41	4.53
CBG	100.00	8.92	8.92	8.92	100.00	6.92	0.63	6.92
CBN	100.00	340.68	340.68	100	34.87	ND	100	100.00
CBC	0.00	ND	100	100	95.05	56.89	5.18	56.89
CBL	100.00	175.34	175.34	100	94.25	96.12	8.75	96.12
CBT	27.10	ND	100	100	100	150.97	13.74	100.00
CBF	13.86	ND	100	100	-	-	-	
CBE	100.00	0.87	0.87	0.87	-	-	-	
Std	100.00	1	1	1	96.07	10.99	1	10.99
								Compared to std influenza A

Std = standard reference (Positive control). For antiviral neuraminidase, zanamivir (a clinical drug) is used as a reference, while for antibacterial neuraminidase, quercetin is used as a standard reference for antibacterial neuraminidase.

Table S2. Potency index value and calculation from anti-SARS-CoV2 experiments

Compound	SARS-CoV2 main protease			SARS-CoV2 spike protein		
	Screen	IC50 (μM)	Potency Index	Screen	Ratio	Potency Index
THC	-	16.23	38.64	57.82	1.73	72.94
CBD	-	1.86	4.43	56.30	1.78	77.62
CBG	86.99	5.68	13.52	97.60	1.02	2.46
CBN	-	ND	100	71.85	1.39	39.18
CBC	74.96	ND	100	95.25	1.05	4.99
CBL	100.00	5.67	13.50	64.71	1.55	54.53
CBT	62.00	ND	100	53.37	1.87	87.36
CBF	65.29	ND	100	99.22	1.01	0.79
CBE	80.26	ND	100	73.66	1.36	35.75
Std	86.60	0.42	1	100	1	1

Std = standard reference (Positive control). For antiviral main protease, GC376 (comes with an assay kit) is used as a reference, while for anti-SARS-CoV2 spike protein-human ACE2 interaction, pinostorbin is used as a standard reference.

Table S3. Heatmap raw data

	bacterial neuraminidase	Influenza A neuraminidase	SARS-CoV2 main protease	SARS-CoV2 spike protein
Compounds	Potency Index	Potency Index	Potency Index*	Potency Index
THC	1.85	38.34	38.64	72.94
CBD	0.41	20.98	4.43	77.62
CBG	0.63	8.92	13.52	2.46
CBN	100	100	100.00	39.18
CBC	5.18	100	100.00	4.99
CBL	8.75	100	13.50	54.53
CBT	13.74	100	100.00	87.36
CBF	-	100	100.00	0.79
CBE	-	0.87	100.00	35.75

	0
	100