Peptide Therapeutics for COVID-19

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As the world struggles to cope with the current COVID-19 pandemic, in this edition of "Peptide Highlights" we present a survey of peptide drugs which are being investigated as potential therapies for the disease.

INTRODUCTION

COVID-19 is caused by the SARS-CoV-2 coronavirus, which enters the body through the oronasal route and rapidly spreads to the lungs, causing a hypoxic pneumonia. In severe cases, a hyperinflammatory syndrome and development of acute respiratory distress syndrome (ARDS) follows, which necessitates mechanical ventilation of the lungs and is accompanied by a high mortality rate. Furthermore, both clinical experience and results of autopsy studies indicate that coagulopathy is a prominent feature in the pathogenesis of severe COVID-19 disease.

Drugs being developed or investigated to treat COVID-19 may be broadly categorized into antivirals, which act at the early stages of the disease process by inhibiting viral entry into the host cells and subsequent viral replication, and immunomodulatory, anti-inflammatory drugs acting at a later stage to modulate the inflammatory response of the host immune system or to counteract coagulopathy (Lipworth, 2020). Drugs aimed at treating the underlying pathophysiology of ARDS may be considered as another category (Horie, 2020).

The urgent need for COVID-19 therapies has resulted in the testing in clinical trials of repurposed drugs, either already approved for clinical use in indications other than COVID-19 or currently in clinical trials for other indications. We have witnessed a remarkable acceleration of drug regulatory procedures, which would otherwise be too slow to deal with the need for immediate therapeutic solutions. Likewise, rapid funding initiatives have enabled research groups to adapt their efforts to produce and test potential therapies.

Global efforts to identify effective interventions for the prevention and treatment of COVID-19 have resulted in more than 2,200 trials completed or underway (Global Coronavirus COVID-19 Clinical Trial Tracker). One recent systematic review of randomized trials investigating COVID-19 prevention and treatment, identified 381 single interventions (Karlsen, 2020). Despite these remarkable efforts, an efficacious treatment has as yet proven elusive (Siemieniuk, 2020).

PEPTIDE-BASED THERAPIES FOR COVID-19 IN THE RESEARCH PHASE

With respect to peptide-based therapies for COVID-19, a wealth of research data from *in vitro* and pre-clinical studies reveal some promising candidates for future clinical development. To name a few examples:

a) Defensin-like peptide, P9R

In experiments in mice, the defensin-like peptide P9R has shown broad spectrum antiviral activity against respiratory viruses including influenza virus and SARS-CoV-2 (Zhao, 2020). The antiviral effect of P9R has been shown to be due to direct binding to virus particles as well as inhibition of the virus-host acidification of endosomes required for viral entry into the host cells.

b) Integrin binding peptide, ATN 161N

The integrin binding peptide, ATN161N, (Ac-PHSCN-NH_2), has been shown in a pre-clinical, ELISA-based method to interfere with binding of the SARS-CoV-2 spike protein (S) to the ACE2 receptor in host cells (Beddingfield, 2020).

c) Snake-venom derived bradykinin-potentiating peptide (BPP-10c)

Venom from the Brazilian arrowhead viper, *Bothrops jararaca*, contains peptides, so-called bradykinin-potentiating peptides (BPPs), which inhibit the angiotensin-converting enzyme (ACE) and thereby potentiate the effects of bradykinin. The angiotensin-converting enzyme (ACE) cleaves angiotensin I to generate angiotensin II and causes increased blood pressure and vasoconstriction. BPPs were the first bradykinin agonists and ACE inhibitors used to reduce blood pressure in patients with hypertension. BPPs augment bradykinin-related effects by interacting directly on bradykinin receptors. The interaction of BPPs with both the renin-angiotensin (RAS) as well as kinin-kallikrein (KKS) systems indicated their potential use to treat COVID-19. The peptide BPP-10c (ENWPHPQIPP) strongly decreases angiotensin II by inhibiting ACE, increasing bradykinin-related effects on the bradykinin 2 receptor (B2R), increasing NO-attributed antioxidant, antiinflammatory and neuroprotective effects and exhibiting direct neural antihypertensive effects. For these reasons it was hypothesized that BPP-10c may be an excellent anti-COVID-19 treatment due to its ability to counteract most of the deleterious effects of SARS-COV-2 on both RAS and KKS (Gouda, 2020).

Many other research groups have produced promising results with peptides which interfere with viral entry to host cells, giving an optimistic picture for availability of candidates for future clinical trials (Baig, 2020; Elnagdy, 2020; Huang, 2020; Maiti, 2020). For a comprehensive review, the reader is referred to Van Patten, 2020.

PEPTIDE THERAPIES FOR COVID-19 IN CLINICAL TRIALS

Amongst the numerous clinical trials and treatments under investigation, there are nine peptides which are currently being tested as potential therapies for COVID-19. These are listed in Table 1. As can be seen in Table 1, all clinical trials testing peptides as therapies for COVID-19 are at least in Phase II, indicating that the peptides have been repurposed, and have either received regulatory approval or are being tested in ongoing clinical trials for an indication or disease condition other than COVID-19. Table 2 gives more information about each peptide, describing its structure and origin, as well as function and likely mode of action.

Of the nine peptides being tested in clinical trials as COVID-19 therapies, one, namely degarelix is aimed at inhibition of viral binding to the ACE2 receptor in host cells. Degarelix suppresses production and reduces availability of the host transmembrane protease serine protease 2 (TMPRSS2) required for cleavage of SARS-CoV-2 spike protein (S), followed by membrane fusion and delivery of viral genetic material to the host cell. Viral binding to the host cell receptor ACE2, implicates the RAS system in COVID-19 pathophysiology and RAS disequilibrium might also influence modulation of the inflammatory response in the lungs. Angiotensin-(1-7) is downregulated in COVID-19 patients; its use as a therapy is aimed at improvement of respiratory function, and prevention of the coagulopathy observed in COVID-19, owing to the antithrombotic effect of angiotensin-(1-7). Cyclosporine, metenkefalin, oxytocin and tridecactide, through their immunomodulatory function, are aimed at reducing inflammation during the later stages of severe COVID-19. The LSALT peptide is a selective dipeptidase-1 antagonist that prevents leukocyte adhesion to endothelial cells reducing inflammation and subsequent organ damage and is thus aimed at prevention of acute lung and kidney injury in COVID-19 patients. Last, but not least, solnatide, which has successfully undergone a Phase II trial in patients with moderate-to-severe ARDS, is aimed at treating the underlying lung pathophysiology in mechanically-ventilated COVID-19 patients. Solnatide activates the amiloride-sensitive sodium ion channel, ENaC, thereby improving alveolar fluid clearance, reducing lung oedema and enabling improved oxygenation of the lungs (Table 1).

Table 1. Clinical trials involving peptide therapies under investigation as treatments for COVID-19 (last updated 11th September, 2020).

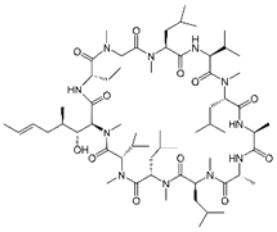
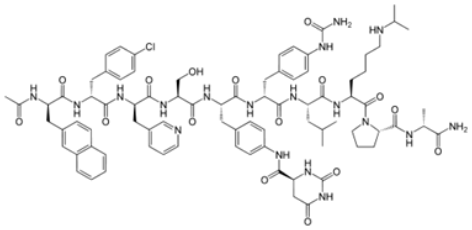
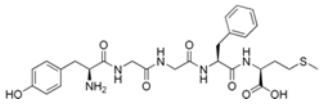
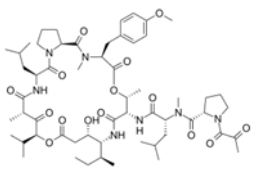
Peptide	Trial Registry Entry Number	Title of Trial	Phase
Angiotensin 1-7	NCT04332666	Angiotensin-(1,7) Treatment in COVID-19: the ATCO Trial	II
Cyclosporine	EUCTR2020-001262-11	Randomized, controlled, blinded clinical trial to evaluate the efficacy and safety of cyclosporin A treatment associated with standard treatment versus standard treatment only in hospitalized patients with confirmed coronavirus infection	
	NCT04392531	Clinical Trial to Assess Efficacy of cYclosporine Plus Standard of Care in Hospitalized Patients With COVID19	IV
Degarelix	NCT04397718	Hormonal Intervention for the Treatment in Veterans With COVID-19 Requiring Hospitalization (HITCH): A Multicenter, Phase 2 Randomized Controlled Trial of Best Supportive Care (BSC) vs BSC Plus Degarelix	II
LSALT peptide	NCT04402957	LSALT Peptide vs. Placebo to Prevent ARDS and Acute Kidney Injury in Patients Infected With SARS-CoV-2 (COVID-19)	II
Metenkefalin[a]	NCT04374032	Clinical Trial to Evaluate the Efficacy and Safety of an Immunomodulatory Therapy for the Treatment of Patients With Moderate to Severe COVID-19 Infection	II
Oxytocin	NCT04386447	Phase II RCT to Assess Efficacy of Intravenous Administration of Oxytocin in Patients Affected by COVID-19	II
Plitidepsin	NCT04382066	Proof of Concept Study to Evaluate the Safety Profile of Plitidepsin in Patients With COVID-19	
Solnatide	EUCTR2020-001244-26	COVID-19: Efficacy of solnatide to treat pulmonary permeability oedema in SARS-Cov-2 positive patients with moderate-to-severe ARDS - a pilot-trial.	II
Tridecactide[a]	NCT04374032	Clinical Trial to Evaluate the Efficacy and Safety of an Immunomodulatory Therapy for the Treatment of Patients With Moderate to Severe COVID-19 Infection	II

[a] Metenkefalin and tridecactide are formulated in the drug Enkorten (Tradename).

CONCLUSION

Peptide therapies for COVID-19 under investigation in clinical trials mainly address the later stages of severe manifestation of the disease characterized by hyperinflammation, coagulopathy and respiratory failure. Current research efforts promise to bear fruit in producing antiviral peptide drugs which will address earlier stages of the disease and focus on preventing SARS-CoV-2 virus cell entry. There is room for cautious optimism concerning peptide therapeutics against COVID-19.

Table 2. Properties of peptide drugs being investigated in clinical trials as treatments for COVID-19.

Peptide	Sequence/structure	Origin/function/action
Angiotensin 1-7	DRVYIHP	Natural peptide ligand for G-protein coupled receptor MAS1. Has vasodilator and antidiuretic effects. Has an antithrombotic effect that involves MAS1-mediated release of nitric oxide from platelets.
Cyclosporine		Isolated in 1971 from the fungus *Tolypocladium inflatum* and came into medical use in 1983. Immunosuppressant.
Degarelix		Synthetic peptide derivative of natural gonadotropin releasing hormone (GnRH) decapeptide. Degarelix binds to GnRH receptors in the pituitary gland, blocking their interaction with GnRH, inducing a fast reduction in luteinizing hormone (LH), follicle-stimulating hormone (FSH) and in turn, testosterone suppression. Approved for treatment of advanced prostate cancer by FDA in 2008.
LSALT peptide	LSALTPSPSWLKYKAL	Binds to dipeptidase-1 (DPEP-1) but does not inhibit its biologic enzymatic activity, potentially minimizing off-target or other adverse effects. LSALT peptide inhibits leukocyte recruitment in multiple experimental disease models through the direct inhibition of leukocyte adhesion to DPEP-1 present in lungs, kidney, and liver.
Metenkefalin[a]		Endogenous opioid and beta-endorphin. Targets the delta-opioid receptor. Immunomodulatory.
Oxytocin	[CYIQNCPLG] (cyclo Cbeta1-Cbeta6)	Peptide hormone produced in hypothalamus and involved in social bonding, sexual reproduction and childbirth. Synthetically produced oxytocin first introduced into clinical use in 1962 for induction of labour, postpartum haemorrhage and lactation.
Plitidepsin		Cyclic depsipeptide isolated from sea squirt, *Aplidium albicans*. Exhibits antitumour, antiviral and immunosuppressive activities. Shows promise in shrinking tumours in pancreatic, stomach, bladder, and prostate cancers.
Solnatide	[CGQRETPEGAEAKPWYC] (cyclo Cbeta1-Cbeta17)	Synthetic peptide which mimics the lectin-binding domain (TIP) of tumour necrosis factor (TNF). Activates epithelial Na+ channel, ENaC, promoting alveolar fluid clearance and reducing pulmonary oedema.
Tridecactide[a] (alpha-corticotropin 1-13)	SYSMEHFRWGKPV	Synthetic deacetylated, deaminated peptide analogue of the naturally occurring alpha-melanocyte stimulating hormone (alpha-MSH). Targets the melanocortin receptor. May have antipyretic, anti-inflammatory, and antimicrobial effects. Immunomodulatory.

[a] Metenkefalin and tridecactide are formulated in the drug Enkorten (Tradename).

REFERENCES

Baig MS, Alagumuthu M, Rajpoot S, Saqib U (2020) Identification of a Potential Peptide Inhibitor of SARS-CoV-2 Targeting its Entry into the Host Cells. *Drugs R D.* Sep;20(3):161-169. doi: 10.1007/s40268-020-00312-5. PMID: 32592145.

Beddingfield B, Iwanaga N, Chapagain P, Zheng W, Roy C, Hu T et al (2020) The Integrin Binding Peptide, ATN-161, as a Novel Therapy for SARS-CoV-2 Infection. bioRxiv 2020.06.15.153387; doi: https://doi.org/10.1101/2020.06.15.15338

Cytel. Global Coronavirus COVID-19 Clinical Trial Tracker (2020) https://www.covid19-trials.org/.

Elnagdy S, AlKhazindar M (2020) The Potential of Antimicrobial Peptides as an Antiviral Therapy against COVID-19. *ACS Pharmacol Transl Sci.* Jun 16;3(4):780-782. doi: 10.1021/acsptsci.0c00059. PMID: 32821884.

Gouda AS, Mégarbane B. (2020) Snake venom-derived bradykinin-potentiating peptides: A promising therapy for COVID-19? *Drug Dev Res.* Aug 5:10.1002/ddr.21732. doi: 10.1002/ddr.21732. Epub ahead of print. PMID: 32761647.

Horie S, McNicholas B, Rezoagli E, Pham T, Curley G, McAuley D, O'Kane C, Nichol A, Dos Santos C, Rocco PRM, Bellani G, Laffey JG. (2020) Emerging pharmacological therapies for ARDS: COVID-19 and beyond. *Intensive Care Med*. Jul 11:1–19. doi: 10.1007/s00134-020-06141-z. Epub ahead of print. PMID: 32654006.

Huang X, Pearce R, Zhang Y (2020) *De novo* design of protein peptides to block association of the SARS-CoV-2 spike protein with human ACE2. *Aging (Albany NY).* Jun 16;12(12):11263-11276. doi: 10.18632/aging.103416. Epub 2020 Jun 16. PMID: 32544884.

Karlsen APH, Wiberg S, Laigaard J, Pedersen C, Rokamp KZ, Mathiesen O. (2020) A systematic review of trial registry entries for randomized clinical trials investigating COVID-19 medical prevention and treatment. *PLoS One.* Aug 20;15(8):e0237903. doi: 10.1371/journal.pone.0237903. PMID: 32817689.

Lipworth B, Kuo CR, Chan R (2020) Emerging pharmacotherapy for COVID-19. *J R Coll Physicians Edinb*. Jun;50(2):133-137. doi: 10.4997/JRCPE.2020.210. PMID: 32568282.

Maiti BK (2020) Potential Role of Peptide-Based Antiviral Therapy Against SARS- CoV-2 Infection. *ACS Pharmacol Transl Sci.* Jul 24;3(4):783-785. doi: 10.1021/acsptsci.0c00081. PMID: 32821885.

Siemieniuk RA, Bartoszko JJ, Ge L, Zeraatkar D, Izcovich A, Pardo-Hernandez H et al. (2020) Drug treatments for covid-19: living systematic review and network meta-analysis. *BMJ*. Jul 30;370:m2980. doi: 10.1136/bmj.m2980. PMID: 32732190.

VanPatten S, He M, Altiti A, F Cheng K, Ghanem MH, Al-Abed Y (2020) Evidence supporting the use of peptides and peptidomimetics as potential SARS-CoV-2 (COVID-19) therapeutics. *Future Med Chem.* Jul 16:10.4155/fmc-2020-0180. doi: 10.4155/fmc-2020-0180. Epub ahead of print. PMID: 32672061.

Zhao H, To KKW, Sze KH, Yung TT, Bian M, Lam H et al. (2020) A broad-spectrum virus- and host-targeting peptide against respiratory viruses including influenza virus and SARS-CoV-2. *Nat Commun.* 2020 Aug 25;11(1):4252. doi: 10.1038/s41467-020-17986-9. PMID: 32843628.