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CHRONIC METFORMIN, TURMERIC TEA AND LOW-DOSE ACUTE CHLORPROMAZINE ATTENUATE PROGRESSION OF COVID-19: PRELIMINARY STUDY AND REVIEW

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ABSTRACT

The pathobiology of COVID-19 caused by SARS-CoV2 is complex, evolving and intertwined with aging and age-related diseases. The high infectivity of COVID-19 is attested to by the increasing daily infection and mortality figures. The increased neutrophilia and increased neutrophil-to-lymphocyte ratio in COVID-19 may be the key factors responsible for the progressive lung and other organ impairment in this disease. They may also be the central culprits in the early thrombophilia, hypoxia, dysregulation of immune-inflammatory pathways, visceral adiposity, increased plasminogen activator inhibitor-I (PAI-I) and other features of obesity-insulin resistance that may be signatures of the disease. In the present report, the combination of metformin and turmeric tea is shown to statistically significantly ($P < 0.05$) decrease the cardiometabolic risk factors of increased BMI, cholesterol, FBS, MAP, uric acid and neutrophil-to-lymphocyte ratio which may be relevant both for the metabolic syndrome and COVID-19. The combination of chronic metformin, turmeric plus acute low-dose chlorpromazine also prevented the progress of COVID-19 from a mild flu-like illness to severe disease in an adult patient. The combination of metformin and turmeric tea thus may serve as preventive agents of the disease; and with acute chlorpromazine display enhanced curative effects for COVID-19; functions which are urgent and compelling today especially as preliminary reports suggest decreased vaccine efficacy in the elderly.

KEYWORDS: SARS-CoV2, COVID-19, Metformin, Turmeric, Chlorpromazine, Lung injury, Prevent.

INTRODUCTION

COVID-19 caused by SARS-CoV2 virus, a variant of SARS-CoV virus (centreforhealthsecurity.org), has caused a pandemic which has claimed more than 600,000 lives and with more than 16 million people infected. Of the beta-coronaviruses of greatest human clinical importance, HCoV-HKUI, HCoV-OC43, SARS-CoV, MERS, and SARS-CoV2, it is SARS-CoV2 that has an astonishingly efficient human-to-human transmission and infectivity.[1,2] Bats and rats have been the culprits harbouring emerging viruses.

The SARS-CoV2 is a novel coronavirus with a large (approximately 29,903 nucleotides) positive sense, single stranded RNA containing 14 functional open reading frames (0RFs). Its genome shares 79% and 50% sequence identity with SARS-CoV and MERS-CoV genomes respectively. Open reading frames are parts of a reading frame (sequence of nucleotide triplets) that has potential to be transcribed into RNA and translated into protein. 2 large ORFs (ORF Ia and Ib) constitute the replicase gene which encodes proteins required for viral RNA synthesis. ORFIa and Ib encode the polyproteins ppIa and ppIb which are cleaved by viral proteases (papain-like protease (PLpro) and 3Chymotrypsin-like protease or main protease (3CLpro) to yield 16 non-structural proteins (NSPs) which are translated first.[3] The remaining 12 ORFs encode the 4 structural proteins (spike (S), membrane (M), nucleocapsid (N) and envelope (E) proteins) and 8 accessory proteins (ORFS 3a, 3b, 6, 7a, 7b, 8b, 9b and 14). The structural proteins are involved in entry, replication, assembly and propagation into cells containing the angiotensin converting enyme 2 (ACE2) receptor.[4] The nucleocapsid protein is key to SARS-CoV2 infection and pathogenesis; while the accessory proteins regulate infection but do not incorporate into the virion.[5]

Cell Entry Mechanisms Of SARS-CoV2 And Its Implication

SARS-CoV2 and SARS-CoV use human ACE2 as entry receptor and human proteases as entry activator to access endosomes with eventual fusion of viral and lysosomal membranes. Transmembrane protease serine 2 (TMPRSS2) and cathepsins have cumulative effects with the proprotein convertase furin. Furin first cleaves the activation sequence at the S1/S2 boundary before further cleavage by TMPRSS2.[2,6] The entry process results in endocytosis and translocation of both the virus and ACE2 into endosomes located within cells.

ACE2 down-regulation induced by viral invasion may be especially detrimental in males, people with older age, obesity, hypertension, diabetes and in cardiovascular disease where the patients may have relative baseline ACE2 deficiency. ACE2 deficiency after viral invasion might amplify the dysregulation between the adverse ACE-angiotensin II-ATI receptor axis and the protective ACE2-angiotensin (1-7)-mitochondrial assembly (Mas) receptor axis thus favouring thrombus, fibrosis, vasoconstriction, edema, permeability and inflammation in the lungs.[7,8,9,10] Also ACE2 attenuates endothelial dysfunction and raised plasminogen activator inhibitor-I (PAI-I) levels while increasing vasodilator prostacyclin and nitric oxide release.[11,12]

Host immune responses are triggered by danger signals from infected cells or free virions, which are recognised by innate immune cells. There is ThI activation with involvement of interferon-gamma, IL-2 and lymphototoxin mediated by cytotoxic lymphocytes. Antigen presenting cells (APCs) present processed CoV2 antigen to CD4 + T-helper cell by further increasing ThI activation, increasing NF-kappaB and IL-17 signaling pathways. Recruitment of neutrophils and monocytes occur; with enhancement of IL-1, IL-6, IL-8, IL-21, TNF-alpha, MCP-1, GM-CSF and CRP. Elevated proinflammatory cytokines and chemokines can be seen in asymptomatic to mild cases. Exuberant cytokine production resulting in cytokine storm exacerbates the severity of coronavirus disease-19 (COVID-19). This may be due to failure of “front-line” immune anti-viral defence, leading to a macrophage activation syndrome –like state or secondary hemophagocytic lymphohistiocytosis (sHLH) with hypercytokinemia.[13] Lymphopenia (T and B cells) and neutrophil infiltration into infected sites contribute to the pathogenesis of COVID-19.

The increased infectivity and virulence of SARS-CoV2 compared to SARS-CoV may be explainable by the prion-like domains in SARS-CoV2 spike proteins which enable higher affinity to ACE2.[14] Additionally, immune evasion strategies and increased infectivity by SARS-CoV2 include the lying-down position of the receptor binding domain (RBD) hidden in the spike protein compared to that of SARS-CoV,[2] and deployment of NSP 14 and NSP 16 to ensure viral RNA cap modification in order to avoid recognition by pattern recognition receptors (PRRs). NSP 1 and 3 may block host innate immune response. ORF6 could inhibit JAK-STAT signaling, blocking its nuclear translocation and inhibiting anti-viral defence.

Coronavirus Disease-19 (COVID-19)

Risk factors for COVID-19 include age over 60 years, smoking habit, underlying non-communicable diseases such as obesity, type 2 diabetes mellitus, hypertension, cardiac disease, chronic lung disease, cerebrovascular disease, chronic kidney disease, immunosuppression and cancer.

Symptoms of COVID-19 include fever (83-99%), cough (52-82%), fatigue (44-70%), anorexia (40-84%), shortness of breath (11-40%), myalgias (11-35%), anosmia and ageusia. Other non-specific symptoms include sore throat, nasal congestion, headache, diarrhea, nausea and vomiting. Fever may be absent in the elderly. Central nervous system symptoms of anxiety, depression, agitation, insomnia may be present without respiratory symptoms (*WHO Clinical Guidance, 2020*). Some patients have reported persistent insomnia and hiccups which have been attenuated by omega-3 fatty acids (*personal communication*). The incubation period of COVID-19 is 2-14 days.

COVID-19 may be asymptomatic in about 35% of cases when infection can still be transmitted (CDC). Of the symptomatic cases, mild cases constitute 40%, moderate cases 40%, severe cases 15%, critical cases 5%. The shortness of breath increases as the severity increases. Clinical signs of pneumonia are present in moderate cases and this gets severe in severe cases when supplemental oxygen may be needed. ARDS, sepsis and septic shock complicate the critical cases when patients may need extracorporeal membrane oxygenation (ECMO) or even ventilator.

Public health measures of mask usage, frequent hand washings and social distancing bolstered by economy-crippling lock-downs and avoidance of massive crowds are widely advocated to help prevent infection as well as contain the pandemic. Actual containment may wait for the development of a potent vaccine and/or effective universal drug treatment since there seems to be poor development of long-lasting immunity in response to the virus, and this imperils effective development of herd immunity.[15] An important consideration for vaccination for the elderlies is the question of vaccine effectiveness in the elderly age-group due to immunosenescence.[16,17]

Clinical Study

A 55-year old medical staff and housewife presented in our clinic, during the labour dispute, at Oseghale Oriaifo Medical Centre, Ekpoma. She had been on self-quarantine and treatment

for mild/moderate COVID-19, having been tested positive by RDT and PCR. She was reacting to chloroquine while she was placed on self-quarantine by having intolerable pruritus, and was therefore brought to our clinic for advice. She also had persistent headache, body pains, difficulty in breathing, chest tightness and worsening cough. Her insomnia and anxiety worsened, and fasting blood sugar and blood pressure rose unexplainably (from 95. 6 mg/Dl to 125.2 mg/dL; and mean arterial pressure (MAP) from 94.6 mmHg to 118.4 mmHg respectively). Chest X-ray was negative for pneumonia; bur CT-scan was not done. The treatment in our clinic led to her close contact with 5 persons including staff who had been on metformin and turmeric tea as agents for diabetes prevention/treatment (see below). Subsequent tests showed these had no symptoms or very mild illness. 3 other exposed individuals not on metformin and turmeric developed moderate COVID-19 as found out on referral. Her husband of 76 years with BMI of 24.31 Kg/M^2 displayed only malaria-like symptoms which responded to chloroquine tablets and omega-3 fatty acid, 1000 mg daily for seven days.

Patient was placed on chlorpromazine (50 mg nocte) and azithromycin 250 mg twice a day for 10 days. She also had cimetidine 400 mg twice daily for 3 days; and ginger tea. There was marked improvement to her breathing difficulty, insomnia, anxiety, headache, cough and myalgia (body pains) after the first night of medication and this was maintained. Her blood pressure and sugar gradually returned to base-line levels over the next 6 weeks and repeat RDT and PCR turned negative. The contacts who developed mild COVID-19 symptoms were also self-quarantined and had only paracetamol as anti-pyretic.[18]

Effect of chronic metformin and turmeric tea on selected cardiometabolic risk factors for the metabolic syndrome

Preliminary study was done on 8 volunteers with similar controls. Test individuals had 1,500 mg-2,000 mg of metformin for treatment of type 2 diabetes or 750 mg – 1,000 mg for its prophylaxis. They also had turmeric tea (half-teaspoonful) twice a day. Treatment had lasted for 4 months. Laboratory tests and analysis were supervised by our technologist. Results are presented in Table I.

STATISTICAL ANALYSIS One-way Analysis of Variance was applied using SSPS version 17 for analysis. For Post-hoc tests, we used Duncan Multiple Range. Mann-Whitney non-parametric test was used when comparing the means of only two samples. Data are presented

as mean ± standard error of mean. The difference was considered to be significant at P < 0.05. (n = 8)

Results show that effects of turmeric had additive effects with metformin. Metformin and turmeric more significantly lowered most cardiometabolic risk factors which are metabolic syndrome-related traits. Metformin and turmeric additively lowered body mass index (BMI), cholesterol, fasting blood sugar (FBS), mean arterial pressure (MAP), WBC, neutrophil counts but non-significantly increased lymphocyte count. They had no significant effect on platelet count. Metformin and turmeric tea also had additive effect in lowering uric acid level; while prolonging prothrombin time (PT) and bleeding time.

Results may explain beneficial effect observed with metformin and turmeric tea in preventing progression of COVID-19; especially when acute low-dose chlorpromazine is added. The low-dose acute addition of chlorpromazine could explain the rapid improvement also of the CNS manifestations including the insomnia, body pains and fatigue.

Table 1: Effect of chronic metformin + turmeric administration on selected cardio-metabolic parameters which are metabolic syndrome-related traits.

	Control	Metformin	Metformin + Turmeric
BMI (kg/m^2)	29.45±2.76	28.26±3.15*	26.03±2.04**
Cholesterol (mg/dl)	220.50±2.22	185.46±3.92*	165.48±3.45**
FBS (mg/dl)	105.42±6.12	98.23±3.64*	78.42±5.86**
MAP (mmHg)	112.21±6.33	97.46±4.84*	79.25±4.76**
WBC (x10^9/l)	6.65±6.51	6.42±2.09*	6.01±3.45**
Neutrophil (x10^9/l)	3.60±3.42	3.10±4.15*	3.01±4.16**
Lymphocytes (x10^9/l)	2.85±1.74	2.86±0.34	2.86±1.26
Platelets (x10^9/l)	219.48±55.26	219.48±28.35	218.46±35.88
Uric Acid (mg/dl)	6.56±0.68	6.56±0.94*	3.36±1.56**
Prothrombin Time (s)	6.54±1.08	6.54±1.15*	25.44±0.32**
Bleeding Time (s)	68.54±1.18	68.54±1.18	107.26±3.67**

Table I: Metformin and turmeric tea more significantly (**) lowered BMI, cholesterol, FBS, MAP, WBC, neutrophil counts but non-significantly increased lymphocyte count. Metformin and turmeric had no significant effect on platelet count. Metformin and turmeric also additively lowered uric acid levels while prolonging prothrombin time and bleeding time significantly.

DISCUSSION

AMPK Activators, Metformin And Turmeric Tea, Attenuate Obesity And Type 2 Diabetes Which Are Risk Factors For COVID-19

Age above 60 years, type 2 diabetes and high BMI are associated with negative outcome in COVID-19.[19] Obesity, which is attenuated by metformin,[20] increases NLRP3 inflammasome activation, increases low-grade inflammation and chemokines. Obesity also lowers neutralizing antibodies and effector memory T-cells during a viral infection. Type 2 diabetes or insulin resistance may increase TMPRSS2 activity and consequently enhance viral replication in the host.[21] The cardiometabolic risk factors positively impacted by metformin and turmeric may be traits related to both the metabolic syndrome and COVID-19 (see Table I). There may be a bidirectional relationship between COVID-19 and the central features of the metabolic syndrome via SARS-CoV2 deleterious effect on the pancreas to enhance insulin resistance plus the increased visceral adiposity, atherogenic dyslipidemia, and endothelial dysfunction induced by COVID-19.[22,23,24] There may be amplification of PAI-I and tissue factor increase and decreased endothelial nitric oxide (NO) bioavailability.[25,26,27,28,29] Workers have shown that PAI-I which is down-regulated by metformin may be directly involved in thrombosis,[25] white adipose tissue dysfunction, insulin resistance, RAS activation and lung inflammation.[30] Metformin is also known to upregulate the anti-viral, vasodilator and anti-thrombotic endothelial nitric oxide which decline is accelerated by COVID-19.[31] The Diabetes Prevention Program (DPP) study showed that metformin prevents progression of type 2 diabetes. We have previously reported that calorie restriction and metformin reduce BMI and attenuate progression of pre-diabetes,[32,33] which it may effect via growth differentiation factor-15 (GDF-15) increases to suppress appetite and promote weight loss.[34] Metformin and calorie restriction were previously shown to have same signatures (gene profiling),[35] both decreasing inflammation and oxygen consumption. Importantly, metformin and turmeric (which contains curcumin as principal polyphenol), both AMPK activators, are synergistic in lowering BMI and blood sugar levels,[36] reversing immune senescence and inflammaging, inhibition of virus via enhancing insulin sensitivity and preventing NLRP3- and SARS-CoV2-induced cytokine storm.[37,38] Interestingly, omega-3 fatty acids (docosahexaenoic acid and eicosapentaenoic acid) which also activate AMPK display metformin-mimetic effects. Metformin, a geroprotective, which displays anti-inflammatory effect irrespective of diabetes status shows significant benefit in reducing mortality and morbidity in COVID-19 patients.[39,40,41]

Metformin, Turmeric And Calcitriol Interact To Decrease Inflammasome Activation By Uric Acid And Prion-like Protein

Metformin, turmeric and calcitriol, the active form of vitamin D, exerts pronounced impact on ACE2-Ang (1-7)-Mas receptor axis with enhanced expression of ACE2 which may attenuate acute lung injury.[42,43] Metformin, in the presence of vitamin D, reinforces the interaction between vitamin D receptor (VDR) and its co-activation protein to attenuate renin angiotensin system (RAS), uric acid levels and tissue inflammation.[44] Our results show metformin and turmeric additively lower uric acid levels (Table I). A dysregulated prionoid protein-uric acid-viral RNA interaction, which may upend innate and adaptive immune mechanisms, may be implicated in COVID-19 pathobiology,[45] as well as in aging, hypertension and type 2 diabetes mellitus.[46,47,48] The omega-polyunsaturated fatty acids (w-PUFAs) may also decrease uric acid and prion-like proteins. NLRP3 inflammasome activation by prion-like protein and uric acid which may contribute to the acute kidney injury (AKI) and cytokine storm (cytokine release syndrome) in COVID-19 may be attenuated additively by metformin, vitamin D and vitamin C,[49,50,51,52,53] by curcumin,[54] and by oxypurinol. (XORTX, 2020) Metformin is synergistic with vitamin D3 by preventing the degradation of VDR.[55] Colchicine which attenuates inflammasome activation, gouty attacks and prion-like mechanism-induced cell death has been used for the cytokine release syndrome in COVID-19.[56]

Metformin, Chlorpromazine And Turmeric Prevent SARS-CoV2 cell entry

Metformin could lead to phosphorylation of ACE2 via AMPK to decrease viral entry.[27] Metformin and turmeric could synergistically lower cholesterol levels to attenuate ACE2-mediated viral entry which is upregulated by high cholesterol, similarly to the statins and essential fatty acids which may function to inhibit HMG-CoA reductase.[57,58,59] Chlorpromazine inhibits clathrin-mediated endocytosis of SARS-CoV2 and enhances IgM.[60,61] Chlorpromazine decreases salivary and CNS load of SARS-CoV2 and thus may help in preventing co-transmission. Curcumin from turmeric binds directly to RBD of viral S protein,[62,63] destabilizing the structural integrity of the receptor.[64] Calorie restriction, metformin, curcumin, chlorpromazine, the 4-amino- quinoline anti-malarials such as chloroquine may inhibit prion-like proteins,[65,66,67,68,69] and may decease affinity of SARS-CoV2 to ACE2 to reduce infectivity.

Metformin And Turmeric Upregulate ACE2 To Attenuate Acute Lung Injury

Metformin and turmeric enhance ACE2 down-regulated by viral-induced endocytosis;[63] a down-regulation accentuated by RAS over-activation in diabetes and hypertension, for example.[7,70] SARS-COV2 upregulation of ADAM-17 which facilitates viral entry is attenuated by AMPK.[6,71,72] Both metformin and turmeric inhibit RAS over-activation which is upregulated by ADAM-17. Enhanced ACE2 shedding resulting from RAS over-activation and subsequent ADAM-17 upregulation drives pathogenesis. Increased membrane ACE2 shedding occasioned by the catalytic activity of ADAM-17 results in loss of ACE2 at the membrane and impaired conversion of angiotensin II into Ang (1-7) peptides.[73] Diets rich in fats decrease ACE2 activity, which favors obesity-induced hypertension, and increase the deleterious angiotensin II effects. This attenuation of ACE2 may be countered also by w-PUFAs which may function to inhibit angiotensin converting enzyme and endogenous HMG-CoA reductase.

Metformin, Turmeric And Chlorpromazine Inhibit SARS-CoV2 Replication

Metformin and turmeric may inhibit viral replication by upregulating interferon-stimulating genes (ISGs),[71] and inhibiting viral main protease via AMPK.[73,74] Metformin and Curcumin could also inhibit SARS-CoV2 replication and COVID-19 severity via AMPK inhibition of mTOR;[17,75,76,77] an effect which may be potentiated by omega-3 fatty acids which may also inhibit SARS-CoV2 viral replication via mTOR inhibition. Very recent report shows chlorpromazine also inhibits SARS-CoV2 replication.[78]

Metformin, Chlorpromazine, Turmeric Tea Inhibit Inflammation Associated With COVID-19 Progression

Interleukin-17 is a human homologue of viral nucleocapsid which acts as a potent proinflammatory cytokine production by activated memory T cells.[63] IL-17 is known to induce the mobilization, recruitment and activation of neutrophils. It also stimulates the expression of several pro-inflammatory cytokines and chemokines by a broad range of cellular targets, including epithelial cells, endothelial cells and macrophages. AMPK activators such as metformin and curcumin suppress gut commensal-induced IL-I beta production, a critical step for Th17 differentiation.[79,80] Chlorpromazine may suppress TNF production, a pivotal mediator of endotoxic shock, even in those resistant to dexamethasone.[81,82] Chlorpromazine may also effectively synergise with the AMPK activators, metformin, azithromycin and turmeric, to inhibit gut dysbiosis, IL-6, NF-kappa,

IL-17 and neutrophil activation. Both chlorpromazine and metformin are known to increase the protective cytokine, IL-10,[81] and attenuate endotoxic shock. They ameliorate sodium and fluid transport to prevent diffuse alveolar damage and lung function decline.

Metformin And Chlorpromazine Enhance Lung Function And May Prevent Silent Hypoxia In COVID-19

Chlorpromazine is a vasodilator in pulmonary vascular bed preconstricted by hypoxia. It enhances membrane integrity, preventing membrane peroxidation and reverses ischaemic mitochondrial dysfunction.[83,84,85,86] SARS-CoV2 displays hepcidin-mimetic effects. The hepcidin antagonist, metformin stands to down-regulate the hyperferritinemia, low hemoglobin, low serum iron, thrombocytopenia, increased lactate and lactate dehydrogenase which are the laboratory findings in some COVID-19 patients with silent hypoxia. Hepcidin is increased in diabetes and obesity and pairs the higher levels of glycated, dysfunctional hemoglogin contributing to the hypoxemia made worse by mitochondrial dysfunction and oxidative stress. Metformin could synergise with curcumin in turmeric tea and with chlorpromazine to enhance mitochondrial biogenesis and antagonise SARS-CoV2 effects in bone marrow via CD 147. It could also via enhanced nitric oxide production by capillary endothelium regulate mitochondrial function and oxygen consumption and be additive with turmeric which inhibits arginase.[87] Nitric oxide increases oxygen saturation percentage of hemoglobin as oxyhemoglobin and oxygen saturation of arterial blood.[88] This has relevance for acute respiratory distress syndrome. [89] Thus they are positioned to attenuate the breathing difficulties and possibly hypoxemia which may manifest at all stages of COVID-19. They enhance mitochondrial function to enhance cell adaptation to hypoxia similarly to calorie restriction and w-PUFAs.[90,91,92,93] They also stand to prevent the septic shock and ARDS which may be present in critical COVID-19.[92]

Metformin, Turmeric And Chlorpromazine May Attenuate The Coagulation Dysfunction In COVID-19

The increased risk of thrombus formation and disseminated intravascular coagulation in patients with type 2 diabetes is made worse with super-added COVID-19.[94] In this report, metformin and turmeric additively increase prothrombin time which may be shortened in mild COVID-19 patients, thereby preventing increased coagulation. In severe disease, metformin stands to inhibit the disseminated intravascular coagulation induced by neutrophil extracellular traps (see below). W-PUFAs also may inhibit disseminated intravascular

coagulation (Patent W01993016691A1). Studies have shown that coagulation dysfunction is a major cause of death in patients with severe COVID-19,[95,96,97] who may have micro-clots in the tiny capillaries of the small air-sacs that mediate gas exchange.[98] Importantly, there may be thrombus persistence due to down-regulation of genes involved in angiogenesis.[99] Age and platelet peaks have been shown to influence mortality in COVID-19,[100] while metformin via GDF-15 prevents platelet integrin activation and thrombus formation.[101] Metformin, curcumin and omega-3 fatty acids could be the new anti-platelet drugs. Extended thromboprophylaxis with oral anti-coagulants have been advised, since this has benefit compared with low-molecular weight heparin.[102] Metformin increases bleeding time (confirmed by this study), inhibits platelet activation by decreasing the release of mitochondrial DNA (mtDNA) and suppresses proinflammatory and prothrombotic processes.[103,104,105] Notably, metformin enhances the angiogenic potential of endothelial progenitor cells, increases migration of bone marrow-endothelial progenitor cells (BM-EPCs) via activating AMPK/eNOS pathway.[106] Metformin, which enhances the anti-coagulant activated protein C (APC),[107] may have additive effects with the anti-platelet functions of curcumin and chlorpromazine to become relevant in preventing consumptive coagulopathy or disseminated intravascular coagulation.[108,109,110]

Deleterious Role Of Neutrophilia In COVID-19

Neutrophils display a key role in COVID-19 pathology by increasing the inflammatory and hemorrhagic lesions in the lungs of patients. Neutrophil activation and neutrophil degranulation are the most activated biological processes in the SARS-CoV2 infection, as well as the basophilia and lymphopenia with increased neutrophil- to- lymphocyte ratio which predicts severe illness in COVID-19.[111,112] There is an association between circulating white blood cell counts, CT-determined visceral fat burden and obesity-insulin resistance.[113,114] Age, obesity, WBC count and inflammatory cytokines are significant predictors of PAI-I concentration, a key factor linking fibrinolysis and age-related diseases with worse prognosis,[115] Neutrophil elastase (ELANE) which enhance neutrophil maturation and neutrophilia may additively with arginase I and chemokine C-X-C ligand (CXCL1) attract neutrophils to inflammation site. Metformin reverses the lymphopenia in COVID-19 and rebalances the neutrophil- to- lymphocyte ratio.[111,116] Metformin upregulates lymphocyte count in COVID-19 patients compared to control, decreases neutrophil count and promotes T-regulatory and CD8 memory cells.[117] Metformin-induced enhancement of insulin sensitivity inhibits arginase which may induce immunosuppression and may co-

operatively act with turmeric tea in this regard.[87,118] Also, chlorpromazine decreases leukocyte count, attenuate neutrophil activation and inhibits pulmonary arginase.[119,120] Neutrophils induce NETosis, complement activation, thrombus formation to cause alveolar cell damage and enhance cytokine release (necroinflammation).[121] Metformin, chlorpromazine and curcumin or omega-3 fatty acids may additively decrease neutrophil counts and activation to prevent neutrophil-induced lung, cardiac damage and neurological manifestations in COVID-19.[122,123]

Neutrophil Extracellular Traps And COVID-19

Neutrophil extracellular traps or NETs, which promote thrombus generation,[124,125] are released upon exposure to infection agents, sterile inflammation, type 2 diabetes, obesity, arterial hypertension, autoimmune stimuli and cancer. Increased mitochondrial reactive oxygen species can increase NET formation in aged individuals.[126,127,128,129] NETs induce macrophages to secrete IL-I beta which induces IL-6 strongly associated with pro-inflammatory states, and which enhances further NET formation. IL-I beta, IL-6, IL-8 and TNF-alpha seem to be involved in the enhanced release of NETs in systemic inflammatory response syndrome.[130] A signalling loop between macrophages and neutrophils can lead to progressive uncontrollable inflammation-the cytokine storm.[131] Metformin blunts NETosis *in vitro* and reduces circulating NETosis biomarkers *in vivo*,[128] thereby may synergise with chlorpromazine, curcumin or even the specialised proresolvin mediators (SPM) from omega-3 fatty acids in protecting surfactant generation by type II pneumocytes.[132,133,134,135]

Metformin, Curcumin, Chlorpromazine Inhibit Mast cell Degranulation

Mast cells are located at the junction points of the host and external environment, at places of early introduction of antigen.[136] Mast cells located in the submucosa of the respiratory tract are activated by SARS-CoV2 to release histamine, proteases followed by the inflammatory mediators IL-I, IL-6 and TNF-alpha.[137] Mast cells mediate microvascular inflammatory response to systemic hypoxia,[137] and may aid leukocyte recruitment in hypoxia via MCP-I. The chromones cromolyn, nedocromil and ketotifen have been suggested as antidote to this leukocyte recruitment and increased vascular permeability during early COVID-19. Increased endothelial cell permeability which contributes to lung injury in COVID-19 has been attributed to histamine H2-receptor activation,[139] and this may respond to famotidine and cimetidine. Vitamin D deficiency enhances mast cell activation.

Metformin inhibits IgE and aryl hydrocarbon receptor-mediated mast cell activation, suppresses the alarmin, IL-33 signaling and neutrophil recruitment via inhibition of glycolysis.[140,141] These actions of metformin may be additive with that of chlorpromazine, curcumin and docosahexaenoic acid which also inhibit mast cell degranulation and prevent the deleterious effect of the mast cell in COVID-19 subsets.[142,143;144]

CONCLUSION

Important preventive roles of the combination of chronic metformin, turmeric plus acute low-dose chlorpromazine on SARS-CoV2 virus infectivity is highlighted with this report. They may additively inhibit SARS-coV2 viral entry, replication and resultant necroinflammation. The drug combination of metformin and turmeric decrease the cardiometabolic risk factors which may be a commonality between COVID-19 and the metabolic syndrome. Mechanistically, they may inhibit the prion-like domain of the RBD of the viral spike protein, prevent the hypoxia and attenuate the thrombus generation, neutrophilia and hyperinflammation in the disease which may be the hallmarks of disease progression.

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