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Rapamycin and Metformin in Treating COVID-19

Avrohom Karp

Avrohom Karp will graduate with a Bachelor of Science degree in May 2021 and will be attending medical school

Abstract

The SARS-CoV-2 virus has spread rapidly, resulting in a global pandemic. There is a great need for an effective drug cocktail therapy to combat Acute Respiratory Distress Syndrome (ARDS), a major cause of death due to COVID-19. The two drugs examined are metformin, an antidiabetic medication, and rapamycin. Rapamycin is often prescribed for transplant patients as it has an immunosuppressive effect. The aim of the investigation was to determine the efficacy of metformin and rapamycin in treating COVID-19, and to examine what an effective protocol would look like. These two drugs both inhibit mTOR and can reduce the body's auto-immune response, which destroys bronchial cells via cytokine storms. Both drugs have a long history of clinical use and have sufficient evidence of efficacy. They possess antiviral properties and downregulate inflammatory markers, making them excellent candidates for further study, both individually and in combination. Rapamycin has been shown to reverse markers of aging and can help repair organ damage. Importantly, metformin can help negate the toxic side effects of the potent rapamycin, while still preserving the positive effects of the compound. Metformin also has been shown to aid those who are at risk of developing ARDS due to comorbidities such as diabetes or hypertension. As such, using metformin as a preventative therapy, either alone or with small doses of rapamycin, may be warranted in patients at risk (Hussain et al, 2020) (Malhotra et al, 2020).

Introduction

The SARS-CoV-2 virus's lethality can be largely owed to Acute Respiratory Distress Syndrome (ARDS), a major cause of death due to COVID-19. The lungs become inflamed and are heavily damaged as fluid from the blood vessels leaks into the lungs, obstructing breathing and depriving the organs of adequate amounts of oxygen. This results in high morbidity and mortality rates. The overall percentage of mortality in patients with ARDS was 40 percent, making the disorder of primary importance in the search to reduce deaths in the patient population (Tzotzos et al., 2020).

The ability to repurpose drugs is vitally important to combat novel pathogens and can shorten the amount of time and money required to develop an effective protocol. Novel viruses often use elements of central pathways that are commonly used by other viruses. Therefore, drugs that have been previously developed can prove efficacious, as they target the same pathway. Additionally, the drugs that are currently in use have already been proven to be effective and safe, which allows their immediate use until another, more pathogen-specific, drug can be developed. Furthermore, modeling the drug after an existing therapeutic agent can shorten the amount of time and energy required to develop a new agent, as portions of the previous drug can be used as a starting point. This can all add to an advantage that may save countless patients' lives as they wait for a new drug to be developed (Husain et al, 2020).

One of the more interesting drugs proposed as a therapeutic involves the use of metformin, an anti-diabetic medication. This drug has been touted for its anti-inflammatory effects and seems to have a positive effect on the mortality rate of patients suffering from COVID-19, even though the exact mechanism of action remains contested. Possible theories range from its ability to act as a strong base, disrupting the viral envelope, to its ability to up-regulate the ACE-2 enzyme, the critical binding site for the spike protein that serves as the virus' entry point (Scheen, 2020).

Another potential drug being offered as therapy is rapamycin. Rapamycin first gained recognition as an anti-rejection medication for transplanted organs. It suppresses the immune system, resulting in less inflammation, and has a protective effect, shielding the patient from ARDS. Rapamycin can ensure that even if a cure for the virus remains elusive, we can mitigate the negative physiological effects of the illness on the patients. It would therefore be advantageous to examine the differing protocols involving metformin and rapamycin for their efficacy in treating COVID-19 generally, and ARDS specifically, in vulnerable patient populations (Husain et al, 2020).

Methods

Several databases (PubMed, Medscape, clinicaltrials.gov) were systematically reviewed for the relevant literature. Papers concerning the effects of metformin and rapamycin were studied extensively, both in relation to COVID-19 as well as other illnesses.

Discussion

Metformin is a drug that is generally used in the treatment of type 2 diabetes as well as other metabolic conditions. Its widespread use and its minor side effects have led to it being christened the "aspirin of the 21st century." It was originally introduced as an anti-influenza medication before being used for its effects on glucose reduction. It works primarily by decreasing the production of hepatic glucose while simultaneously increasing the action of insulin on tissues, combatting a major factor of type 2 diabetes. Metformin did not achieve widespread use in the United States until 1995, yet by the year 2017, it was the fourth most prescribed medication in the United States. Even though the exact mechanisms of metformin are unknown, there are multiple proposed mechanisms of action, including inhibition of the respiratory chain in complex I of the mitochondria and lowered production of cyclic AMP. Cyclic AMP is the major activating factor for

protein kinase A, an essential part of the second messenger pathway involved in hormonal regulation. This greatly assists the uptake of glucose, preventing insulin resistance, the primary cause of type 2 diabetes. Insulin resistance is caused by an overproduction of insulin, which causes the bodily tissues to require more of the molecule to uptake glucose, resulting in the increasing inability of glucose to migrate into the resistive tissue (Sharma et al, 2020).

The SARS-CoV-2 virus initially binds to the angiotensin-converting enzyme 2 (ACE2). The virus uses its spike protein (S) region to bind to the enzyme, forming a complex that allows the virus entry into the cell. After gaining entry to the host, the virus will downregulate the ACE2 receptor, leading to excessive inflammation, including cardiovascular damage and ARDS. The inflammation is due to ACE2/Ang (1-7)/Mas acting within the Renin Angiotensin Aldosterone System (RAAS) as the anti-inflammatory element to counteract the ACE1–Ang II pro-inflammatory arm. The inflammatory arm causes elevated systemic blood hypertension and inflammation. Metformin is hypothesized to be able to counteract the downregulation of the ACE2 receptor through stimulation of the AMPK/mTOR pathway. The AMPK/mTOR pathway is thought to be involved in increasing the upregulation and stability of ACE2, by phosphorylating ACE2 Ser680. (Malhotra et al, 2020). Although clinical evidence *in vivo* is required, there is strong evidence that Metformin upregulates ACE2, from studies with human umbilical vein endothelial cells (HUVECs) and human embryonic kidney 293 (HEK293T) cells (Zhang et. al. 2018b). Additionally, phosphorylation decreases ubiquitination, extending its half-life. Furthermore, the phosphorylation of the ACE2 enzyme by a large negative phosphate group would change its 3D conformation and sterically hinder the binding complex that the virus uses for entry. In this way, metformin can decrease the ability of the virus to enter the host and reduce mortality through its upregulation of the ACE-2 receptor (Malhotra et al, 2020).

There is evidence that by upregulating ACE-2, Metformin can exhibit a protective effect on the cardiopulmonary system through the AMPK pathway. AMPK activation often leads to ACE2 conformational changes leading to SARS-CoV-2 having a greater difficulty binding to the receptor (Malhotra et. al. 2020). A study found that in animal models “Transgenic mice overexpressing the phosphomimetic ACE2 S680D exhibit less damage in pulmonary vasculature under injurious conditions” (Zhang et al., 2018). Metformin has also been shown to have a healing effect on the pulmonary system in lipopolysaccharides in animal models (Jian et al., 2013).

The glucose-lowering effects of metformin seem to

have a significant effect on survivability. Hyperglycemia has been labeled a major risk factor in various Chinese studies studying pulmonary illnesses. (Chen et al, 2015). A study of patients with diabetes and COVID-19 found that patients with well-controlled blood glucose levels had a much better prognosis and overall reductions in mortality (Crouse et al, 2020). As such, metformin’s proven ability to aid in the treatment of type 2 diabetes is important for diabetic patients that contract COVID-19. Another study has demonstrated that there is a direct correlation between the SARS-CoV-2 virus and diabetes, leading to speculation that the virus can enter the cells of the pancreatic islets where there is an expression of the ACE2 enzyme. This leads to damage to the pancreatic beta cells, which produce insulin, leading to transient type 2 diabetes mellitus (Yang, 2010). This would indicate that the regulation of diabetes is of primary importance for the treatment of COVID-19. Metformin’s previously stated effects on insulin sensitivity would make it an ideal candidate.

Two retrospective studies of COVID-19 patients with diabetes from China in early 2020 found that Metformin could provide benefit. In a study of diabetic patients, the mortality rate for patients on metformin was lower than the group that did not receive the drug. This was despite the metformin patients having higher levels of fasting glucose on admission. The length of the hospital stay was the same between the two groups. (Luo et al. 2020). The SARS-CoV-2 and Diabetes Outcome study in France initially showed that patients who were given metformin before admissions had a lower mortality rate after seven days (Cariou et al, 2020). However, multiple confounding factors needed to be accounted for, which once considered, rendered the findings no longer significant.

In a retrospective analysis studying whether Metformin had any favorable effects 1,213 type 2 diabetic patients with COVID-19 in 16 hospitals in Hubei, China were examined. They excluded confounding variables such as eliminating patients with a glomerular filtration rate of less than 30ml/min or who had cirrhosis. Patients exclusively using insulin were also eliminated. They looked at patients treated with metformin and other drugs compared to just other diabetic medications. They used propensity score matching to adjust for confounding variables and used a Cox regression model to account for the changes in clinical conditions throughout the patients’ stay. They discovered that the length of stay and the 28 days all-cause mortality rate were both unchanged, though metformin was associated with higher levels of acidosis (Lui et al, 2020). Metformin had a protective effect, guarding against heart failure and the body’s various inflammatory responses. Markers of heart failure,

inflammation, and cardiac injury were examined, and all were found to be lower. With these data taken together, according to the results of the study, Metformin exhibits a positive effect on cardiac failure and inflammation (Cheng et al., 2020). This was particularly true in severe cases but did not translate into an overall reduction in mortality. The studies' discovery that Metformin does not influence mortality was contrasted by alternate findings in which researchers had found an improvement in patients using Metformin (Luo et al. 2020). It is possible that since in the Luo study, patients on insulin were included in the non-Metformin group, there was a confounding variable that biased the results.

The poor outcomes of diabetic patients can be explained by several factors. Patients with diabetes generally live with greater levels of overall inflammation daily, putting their already inflamed tissues at an increased risk of destruction of the bronchial epithelium by the body's immune system, resulting in ARDS (Azar et al, 2020). Elevated levels of glucose in patients with diabetes is an additional risk factor as high blood glucose levels have been shown to depress the immune system (Ceriello et al, 2020). These reasons make metformin a viable candidate for the treatment of diabetes in COVID-19 patients, particularly as its glucose-lowering effects can help mitigate the negative role of diabetes (Kow et al, 2021). Additionally, there seem to be positive effects independent of the reduction of blood glucose. Various biochemical mechanisms are involved in Metformin's suppression of inflammatory cytokines, which would aid in the prevention of an overwhelming immune response on the part of the host (Lui et al, 2020).

Metformin's can play a role in the reduction of inflammation in patients either with or at a high risk of contracting ARDS (Acute Respiratory Distress Syndrome). Metformin has proven to be effective at diminishing cytokine storms, an immune response that damages the body's cells. It does this by inhibiting interleukin 1 α and 1 β , important kinases for pro-inflammatory action. The standard measure of systemic inflammation is the neutrophil to lymphocyte ratio, which is used as a marker. Tracking these markers showed an overall reduction of inflammatory cytokines. In a follow-up study of nondiabetic patients with heart failure, it was demonstrated that metformin suppressed plasma cytokines (Cameron et al, 2016). This makes metformin a viable choice in the prevention of inflammation and the formation of ARDS.

The pathway that metformin uses in its anti-inflammatory effects is by the inhibition of tumor necrosis factor- α -dependent I κ B (I κ B kinase) degradation. In an experiment, researchers treated mouse hepatocytes

with metformin. They found that "Metformin treatment for 3 hours suppressed TNF α -induced degradation of the NF- κ B negative regulator I κ B while modulating AMPK and mammalian target of Rapamycin signaling in a dose-dependent manner" (Cameron et al, 2016). Metformin also inhibited signaling downstream by inhibiting the cytokines that are normally produced and activated, 5' AMP-activated protein kinase (AMPK). AMPK is an enzyme that increases glucose uptake and is often thought of as a negative regulator of inflammation. In the same vein, IL-1 β (interleukin-1- β), and IL-6 (interleukin-6) which are both markers of TNF- α -dependent inhibition, were greatly reduced (Cameron et al, 2016).

A 30-day study examining the mortality of nursing home patients found that metformin was associated with significantly less mortality (Lally et al, 2020). Additionally, a retrospective analysis was made of claims made by the United Health group's Clinical Discovery database. This study analyzed the records of enrollment of COVID-19 patients across all 50 states, as well as the various pharmacy claims and laboratory reports. They found that when metformin was given to adults with type 2 diabetes mellitus or obesity there was a marked decrease in mortality exclusively for women, with men receiving no benefit. This finding partially confirms the proposed theory that metformin would be beneficial based on its known effects of decreasing levels of tumor-necrosis-factor α (TNF α). TNF α is of particular importance as patients with COVID-19 have shown to have remarkably high levels of it in their lung tissue. Patients with diabetes also have high levels of TNF α , as diabetes has been shown to further elevate levels. Metformin also promotes the upregulation of anti-inflammatory cytokine, IL-10. Additionally, The study found that TNF α inhibitors were associated with reduced mortality (Bramante et al, 2020).

This study, while interesting, has several noticeable flaws, including the obese and overweight patient samples and the lack of strength associated with retrospective analysis. Additionally, patients may have been prescribed Metformin previously, due to its prevalence, which may have gone unreported. It is important to stress that although a helpful effect was observed, these were individuals with other comorbidities such as T2DM and obesity. The protective effect may be much less pronounced with a patient group that has a lower risk factor and absence of these comorbidities.

Another factor in analyzing the results of the study is the difference in the sensitivity required to activate mast cells between males and females. Mast cells are an early indicator for the SARS-CoV2 immune response. Females exhibit a far greater increase in TNF α than their male

counterparts. Metformin would therefore exhibit a greater positive effect in females, as they are naturally predisposed to a more severe reaction that can be inhibited by the drug. Women and men differed in their cytokine responses even though the levels of the ACE2 receptor were equal (Bramante et al, 2020).

Rapamycin or Sirolimus is a potent anti-transplant rejection drug that can suppress and inhibit mTOR. mTOR is a serine/threonine-protein kinase that is composed of a two-part protein complex named mTORC1 and mTORC2. mTORC1 is the complex that is sensitive to rapamycin and other more common factors such as oxygen, glucose, and various amino acids. mTORC2 is insensitive to rapamycin and acts as an effector of insulin/IGF-1 (Insulin-like growth factor-1). The proteins S6, p70S6K and 4E-BP1 are the point of control for many cellular functions when phosphorylated by mTORC1. This controls protein synthesis and the cell's self-destruction mechanism known as autophagy. mTORC2 is mostly required for other kinases such as Protein Kinase B. One of the reasons that the mTOR pathway is so vital is that it regulates pivotal moments in the life cycle of a cell including metabolism, transcription, proliferation, and eventually, cell death. This has led mTOR, and by extension, rapamycin to be often studied in the examination of aging, also known as cellular senescence (Husain et al, 2020).

Rapamycin first rose to prominence when it was discovered to block the immune system generally, and T cell proliferation specifically. Additionally, mTOR is involved in the cellular division cycle, playing a role in the transition of the G1 to the S phase. Rapamycin as an inhibitor blocks the cell cycle. This would suggest that mTOR can serve as an important chokepoint in mitigating the spread of the virus as it could block the proliferation of infected cells (Husain et al, 2020).

In numerous studies examining the mortality of patients with COVID-19, it was found that a primary factor in the occurrence of infectious diseases was blood Vitamin D concentration. In a study of elderly patients, Vitamin D helped reduce the inflammatory response in the upper respiratory epithelium and lowered the risk of developing intense symptoms (Grant et al, 2020). Vitamin D has also been shown to interact with the ACE2 enzyme, limiting the virus's entry into the cell, as this serves as the entry point for the SARS-COV-2 virus. Evidence would suggest that the mechanism of action for the positive effects of Vitamin D is the suppression of the mTOR pathway through multiple mechanisms. One of these mechanisms involves a regulator known as "regulated in development and DNA damage response 1 (REDD1), a suppressor of mTOR activity", which is stimulated by the 1,25(OH)2D

form of Vitamin D (Husain et al, 2020).

In all, it would be reasonable to suggest that the numerous benefits of Vitamin D are achieved through the inhibition of mTOR and can therefore be stimulated directly by an mTOR inhibitory agent such as rapamycin.

The anti-aging effects of Rapamycin can be of great use as well, with the drug being shown to extend the life of mice significantly (Harrison et al, 2009). But, importantly for the treatment of COVID-19, Rapamycin has been shown to rejuvenate damaged tissues such as damaged cardiac cells and increase the vitality of hematopoietic stem cells (Guarda et al, 2004). It may be that as proteins are continuously synthesized, they acquire damage and various misfolding that inhibit their function and are the prime indicators of age-related diseases. The domain that is affected by rapamycin is mTORC1. Inhibiting mTORC1 reduces protein synthesis and causes the cell to induce autophagy, thereby recycling the damaged components. COVID-19 has been proven to affect the elderly population at an increased rate, with higher mortality being attributed, making rapamycin a potential anti-aging drug that can mitigate the negative effects of age-related vulnerabilities (López-Otín et al, 2013). Additionally, it might go some way in aiding with the reversal of organ damage that is a hallmark of critical cases of COVID-19.

mTOR regulates metabolic processes that help to serve as signaling for anabolic (building) and catabolic (dismantling) processes in the cell. mTOR inhibition has been shown to protect against high fat-induced obesity in mice by regulating the breakdown of glycogen and other processes involving glucose. This could make rapamycin useful in mitigating the negative effects associated with insulin resistance and obesity in COVID-19 patients, which has been proven to result in negative health outcomes (Saxton et al. 2017).

Acute respiratory distress syndrome (ARDS) is caused by the breakdown of upper respiratory epithelia that results from a cytokine storm that destroys the body's tissues. This can lead to multi-organ failure that can often be fatal. The various cytokines that are released are "IL-2, IL-7, IL-10, MCP-1 (monocyte chemoattractant protein), MIPIA (Macrophage Inflammatory Proteins) and TNF- α (Tumor Necrosis Factor- α)". Rapamycin's primary effect as an immunosuppressive drug can decrease the levels of the cytokines in the body, making it a useful tool for dealing with cytokine storms (Costela-Ruiz et al., 2020). Rapamycin's targeting and inhibition of a wide variety of cytokines make it an ideal drug for the suppression of harmful immune responses, making it potentially more useful than the drug Tocilizumab and other monoclonal antibodies that merely target individual cytokines.

Rapamycin's inhibition of the mTOR pathway remains

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very promising not only for its pleiotropic effects on the cell's regulatory mechanisms but also for its anti-immune properties. It is important to mention, however, that rapamycin's side effects can be rather unpredictable. This makes it necessary to monitor the effects of rapamycin in each patient and to educate them about the potential negative side effects. There has been a proposal to examine upstream mutations in mTORC1 signaling to determine ideal candidates for therapeutic interventions using rapamycin. This would help mitigate the negative side effects experienced by members of the patient population.

T-cell senescence is prevalent in long term infections and cancer and is a state of T-cell dysfunction. Cytokine storms can play a role in inducing T-cell apoptosis and necrosis, leading to overall lower T-cell counts. Patients with COVID-19 have been observed to have lower CD4+, CD8+, and total T-cell numbers which are all implicated in lowering the survival rate of patients with the illness. Even when the CD4+ and CD8+ cells are present in severely ill COVID-19 patients, they exhibit less function overall and are unable to secrete perforin, granzyme, and IFN- γ , all of which are cytotoxic molecules. The senescent markers PD-1 and Tim-3 are also present at higher levels. Cells that are senescent release certain cytokines and molecules that are indicative of the cell's status and are known as the "senescence-associated secretory phenotype (SASP)". Rapamycin, being an mTOR inhibitor, can suppress SASP and by extension the cytokine storm that results from T-cell senescence. Therefore, administration of Rapamycin in the early phase of the cytokine storm might prevent the emergence of a severe form of COVID-19 through the downregulation of SASP (Omarajee et al, 2020).

Rapamycin has a history of being beneficial regarding respiratory infections, and has been shown to reduce the recovery time in H1N1 and SARS patients. A study that examined 38 patients with H1N1-induced pneumonia, reported that Rapamycin was associated with positive outcomes in the overall prognosis of patients and shortened their time on a ventilator. It also was associated with significantly increased viral clearance, lower rates of hypoxemia, and reduced multiple organ dysfunction. Both H1N1 and SARS-CoV-2 activate mTOR, leading to lung inflammation, fever, and other intense immune reactions. Rapamycin may provide a significant benefit by inactivating mTOR and therefore IL-1 β secretion, the mediator of inflammation (Wang et al., 2014).

Rapamycin is currently undergoing phase two clinical trials to determine its efficacy in a 30-patient sample that seeks to improve clinical outcomes in COVID-19 patients (NCT04341675). The study will last for 14 days with 2mg being given orally daily. The primary outcome is

to determine the proportion of patients that do not need advanced respiratory support by 28 days. The secondary outcomes involve tracking changes in lymphocyte concentrations as biomarkers as well as the proportion of patients requiring general respiratory support.

There have already been numerous studies done on the use of rapamycin and metformin in the treatment of a plethora of cancers and tumors, including pancreatic and breast cancers (Faria et al, 2019; Amin et al, 2019). These studies have determined the combination to be safe and effective, with the combination able to target slightly different pathways than any of the two drugs alone. Rapamycin can cause glucose intolerance and insulin resistance if taken long term. These effects may be mitigated by metformin, due to metformin causing increased insulin sensitivity in tissues. The ITP (Intervention Testing Program) reported that the effect of both rapamycin and metformin on longevity when taken together was far superior to the effect of each drug alone. It should be noted that in mice the combination of the two drugs did reduce the effective concentration of rapamycin in females and metformin in both sexes. The final concentrations were still within the range to be clinically useful (Strong et al, 2016).

In a study conducted on mice, examining the effects of metformin on glucose, the animals were given both rapamycin and metformin. Interestingly, in contrast to studies that had been conducted previously, the researchers found that metformin did not inhibit mTOR and stopped rapamycin from inhibiting mTOR in the liver. The researchers theorized that metformin could alleviate the disfunction in gluconeogenesis that was found in other mTOR mutant mice (Kim et al, 2020).

Metformin and rapamycin would seem uniquely suited for COVID-19 patients with obesity and diabetes. These patients can be aided by glucose mitigation of metformin but can also cycle lower doses of rapamycin for the potent mTOR effects. In a study of rats being given rapamycin, the researchers attempted to see if they could mitigate the increased hepatic gluconeogenesis caused by rapamycin administration by combining it with metformin. The female mice in the study exhibited significantly lower gluconeogenesis, thereby implying that the metformin served to remove the harmful side effects of rapamycin (Weiss et al, 2018). Here, as in our previous discussion, the effect was sex-specific for many of the reasons outlined previously.

In a study examining metformin and rapamycin on the proliferation of pancreatic cancer cell growth, the optimal therapeutic dosage was determined to be (20 mmol/l) Metformin + Rapamycin (200 ng/ml) *in vivo*. The study discovered that the combination was vastly more effective than monotherapy at inhibiting mTOR (Zhang et al, 2018).

Conclusion

In all, given the evidence that metformin reduces the mortality and morbidity in diabetic patients, it should remain a drug of primary importance in the treatment of COVID-19 in diabetic patients. The effects of the drug on insulin resistance can play an important part in reducing the immunosuppression found in diabetic patients due to the abundance of glucose in the bloodstream. It should be noted that the guidelines urging concern about metformin inducing acidosis in patients was specifically applicable to patients already in multi-organ failure. Additionally, even though the prevalence of acidosis was higher in patients that were given metformin, it did not affect the mortality rate (Cheng et al, 2020). As such, the positive effects of metformin would make it an excellent candidate for further study, not only in patients with pre-existing comorbidities but as a possible therapeutic given its anti-inflammatory properties. Metformin's significant effects on the RAAS system can make it an important agent in the reduction of inflammatory cytokines that are the hallmarks of COVID-19.

Regarding metformin in the treatment of patients with COVID-19 and diabetes, it should be noted that many of the studies are retrospective and examine patient outcomes using statistical analyses. They seek to determine the efficacy of metformin in mitigating the detrimental effects of diabetes on COVID-19 patients. The positive effects of metformin previously noted, there is a paucity of data of metformin being given to COVID-19 patients without diabetes. Many of the studies specifically look at the outcomes of patients who were previously given metformin to control diabetes before the virus and were then examined to determine the beneficial effects specifically on patients with diabetes and COVID-19. But for such a widely prescribed drug such as metformin, it is shocking that there exists so few clinical trials for individuals without diabetes as a comorbidity. Many papers propose that metformin's anti-inflammatory effects could be of use to the wider population to reduce the negative immune response, but as of the date of this paper, there have not been any clinical trials examining the exact outcomes of the drug on a non-diabetic patient population.

There may be several reasons for the lack of clinical trials, among them, that even if metformin does show some benefit in its ability to prevent an overzealous immune response, other drugs currently being used or examined in the treatment of COVID-19 are even more effective through a similar mechanism. Tocilizumab, for example, has already been shown to help in CAR T-cell-induced cytokine release syndrome, which bears many similarities to the cytokine storm that can occur in COVID-19.

A common side effect of Tocilizumab, however, is an elevation in blood pressure, which has proven damaging in COVID-19 patients (Jones et al, 2010). Therefore, it would be efficacious to conduct a thorough study of metformin, given its ability to be tolerated by a wide subset of the population and its proposed benefits in its ability to downregulate the immune system.

Additionally, many of the monoclonal antibody therapies that are being examined specifically exert their effects on one target, while metformin has been shown to operate across many different pathways. Metformin lowers inflammation and oxidative stress, all the while enhancing the immune system of patients.

Rapamycin can be a dangerous compound that must be handled with caution due to its ability to act as a potent inhibitor of mTOR. While mTOR inhibition can be beneficial in the reduction of inflammation, it is important not to inhibit the pathway completely. A study examining the effects of rapamycin on elderly patients discovered that total inhibition of mTOR stops the function of T cells and leads to complete immunosuppression. While some suppression of the immune system is favorable due to its effect on cytokine storms, the body's natural defenses must remain viable. As such, the dose of rapamycin must be carefully monitored to be safe and effective. In that same study, they determined that the most effective dose at promoting a healthy immune response in elderly patients was the lowest dose of rapamycin followed by a flu shot (Mannik et al., 2014). This has a great deal of significance as of early 2021, due to the availability of the mRNA COVID-19 vaccine currently seeing widespread use. The use of rapamycin followed by a vaccine should possibly be explored as a method of enhancing immunity in vulnerable patient populations. This could maximize the effectiveness of a protocol involving not only the currently available vaccine, but also carries implications for other protocols involving vaccinations.

The combination of rapamycin and metformin used in pancreatic cancer has been proven to be safe and effective. The dose used was the optimal dose to lower the expression of mTOR in pancreatic cells, and it stands to reason that this dosage would work in other cases where mTOR should be partially inhibited, such as in COVID-19. Rapamycin inhibits mTOR directly, while metformin does it through the AMPK pathway (Zheng et al, 2020).

The dose of rapamycin administered must be carefully controlled to avoid negative side effects. Metformin can temper some of these effects, which can include insulin resistance and glucose intolerance, through its ability to promote insulin sensitivity. Rather than administering the two drugs as a cocktail, risk can be assessed by the

administration of each drug separately. This would allow a thorough examination of the differing effects while still allowing metformin to counter the deleterious side effects of rapamycin.

An important point in discussions concerning the treatment of viruses is the health of the host. It has been demonstrated that immunocompromised patients and those with significant comorbidities fare far worse than patients without these complications. Metformin has the dual role of not only directly combatting the virus, but also reinforcing the immune system of the host. The ability to reduce comorbidities (e.g.: diabetes) should not be treated flippantly. A major factor in the morbidity rate in the United States has been a large proportion of the population with high blood pressure and obesity (Azar et al, 2020). These conditions have been proven to have significant negative effects. If metformin can provide a mechanism to reduce these additional comorbidities, the patient's health outcomes could be vastly improved.

Taking metformin to prevent an extremely negative reaction to COVID-19 should be considered. Being that metformin is a mild mitochondrial toxin that blocks mTOR, it would be a useful preventative drug. mTOR inhibitors have been shown to block viral action and express innate antiviral gene expression. In previous studies, mTOR inhibitors were effective against other coronaviruses. Prescribing metformin to vulnerable patient populations as a preventative drug may grant the benefits of mTOR inhibition without rapamycin's toxic side effects (Benedetti et al, 2020).

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