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Research Article / Araştırma Makalesi

EXPANDING REMDESIVIR'S THERAPEUTIC SPECTRUM FROM ANTIVIRAL PROPERTIES TO ANTICANCER ACTIVITY THROUGH MTOR INHIBITION

REMDESİVİRİN TERAPÖTİK SPEKTRUMUNUN MTOR İNHİBİSYONU ARACILIĞIYLA ANTİVİRAL ÖZELLİKLERDEN ANTİKANSER AKTİVİTESİNE GENİŞLETİLMESİ

Zeynep Büşra AKSOY[1], Gökçe Yağmur SUMMAK[1,2], Mehmet Altay ÜNAL[1]*, Açelya YILMAZER-AKTUNA[3], Mustafa GÜZEL[4,5,6]*

[1]Ankara University, Stem Cell Institute, Ankara, Türkiye
[2]Ankara University, Graduate School of Health Sciences, Ankara, Türkiye
[3]Ankara University, Faculty of Engineering, Department of Biomedical Engineering, Ankara, Türkiye
[4]Istanbul Medipol University, Research Institute for Health Sciences and Technologies (SABITA), Center of Drug Discovery and Development, İstanbul, Türkiye
[5]Istanbul Medipol University, School of Pharmacy, Department of Basic Pharmaceutical Sciences, İstanbul, Türkiye
[6]Istanbul Medipol University, Health Sciences Institute, Department of Molecular Medicine and Biotechnology, İstanbul, Türkiye

ABSTRACT

Objective: *The dysregulation of the mammalian target of rapamycin (mTOR) pathway is a prominent feature of various cancers, rendering it an attractive target for therapeutic intervention. While some mTOR inhibitors have been employed in cancer treatment, their use is often associated with significant complications. Here, we propose a novel approach to drug repurposing, focusing on Remdesivir, a known antiviral agent, as a potential mTOR inhibitor for cancer therapy.*
Material and Method: *In this study to evaluate the effect of Remdesivir on mTOR patway we performed in silico molecular docking and in vitro experiments including MTT, ELISA, qPCR, and Western Blotting with Caco2 colorectal cancer cells.*
Result and Discussion: *Through computational analyses, we conducted in-silico screenings to identify Remdesivir's interaction with the mTOR protein. Our findings suggest promising binding affinity and molecular interactions, indicating the potential for Remdesivir to inhibit mTOR activity. To validate this hypothesis, we performed molecular experiments using Caco2 colorectal cancer cells to investigate Remdesivir's ability to suppress epithelial-mesenchymal transition (EMT), a critical process in cancer metastasis, following mTOR inhibition. Our study represents a significant advancement in identifying new therapeutic targets using computational methodologies. Repurposing Remdesivir as an mTOR inhibitor offers a cost-effective and expedited approach to drug development, leveraging its known safety profile. Additionally, our findings suggest the potential for Remdesivir to reduce cancer progression by targeting the mTOR pathway and suppressing EMT-associated processes. Overall, this study underscores the promise of*

* **Corresponding authors:** Mehmet Altay Ünal, Mustafa Güzel
E-mail: altay.unal@ankara.edu.tr, mguzel@medipol.edu.tr
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computational drug repurposing in expediting the discovery of targeted therapies. The successful validation of Remdesivir as an mTOR inhibitor paves the way for advanced preclinical and clinical investigations, providing a fresh perspective on cancer treatment and therapeutic innovation.
Keywords: *Anticancer drug development, drug repurposing, mTOR inhibition, remdesivir bioactivity*

ÖZ

Amaç: *Memelilerde rapamisin hedefi (mTOR) yolunun düzensizliği, çeşitli kanser türlerinde belirgin bir özelliktir ve bu da onu terapötik müdahaleler için cazip bir hedef haline getirir. Bazı mTOR inhibitörleri kanser tedavisinde kullanılmış olsa da bu ilaçların kullanımı genellikle önemli komplikasyonlarla ilişkilidir. Bu çalışmada, ilaç yeniden konumlandırmaya yönelik yeni bir yaklaşım öneriyoruz: bilinen bir antiviral ajan olan Remdesivir'in, kanser tedavisinde potansiyel bir mTOR inhibitörü olarak değerlendirilmesi.*
Gereç ve Yöntem: *Bu çalışmada Remdesivir'in mTOR yolağı üzerindeki etkisini değerlendirmek için in silico moleküler kenetlenme (docking) ve in vitro Caco2 kolorektal kanser hücreleri ile MTT, ELISA, qPCR ve Western Blotting yöntemleri ile analiz edilmiştir.*
Sonuç ve Tartışma: *Hesaplamalı analizler yoluyla, Remdesivir'in mTOR proteini ile etkileşimini belirlemek amacıyla in-silico taramalar gerçekleştirdik. Bulgularımız, Remdesivir'in mTOR aktivitesini inhibe etme potansiyelini ortaya koyan umut verici bağlanma afinitesi ve moleküler etkileşimler göstermektedir. Bu hipotezi doğrulamak için, mTOR inhibisyonu sonrasında kanser metastazında kritik bir süreç olan epitel-mezenkimal geçişi (EMT) baskılayıp baskılamadığını incelemek üzere Caco2 kolorektal kanser hücreleri kullanılarak moleküler deneyler gerçekleştirildi. Çalışmamız, hesaplamalı yöntemlerle yeni terapötik hedeflerin belirlenmesinde önemli bir ilerleme sunmaktadır. Remdesivir'in mTOR inhibitörü olarak yeniden konumlandırılması, bilinen güvenlik profili sayesinde maliyet etkin ve hızlandırılmış bir ilaç geliştirme yaklaşımı sunmaktadır. Ayrıca, bulgularımız Remdesivir'in mTOR yolaklarını hedefleyerek ve EMT ile ilişkili süreçleri baskılayarak kanser ilerlemesini azaltma potansiyeline sahip olduğunu göstermektedir. Genel olarak, bu çalışma, hedefe yönelik tedavilerin keşfini hızlandırmada hesaplamalı ilaç yeniden konumlandırmanın vaatlerini vurgulamaktadır. Remdesivir'in mTOR inhibitörü olarak başarılı şekilde doğrulanması, ileri düzey preklinik ve klinik araştırmaların önünü açmakta ve kanser tedavisinde yenilikçi bir bakış açısı sunmaktadır.*
Anahtar Kelimeler: *Antikanser ilaç geliştirme, ilaç yeniden hedefleme, mTOR inhibisyonu, remdesivir biyoaktivitesi*

INTRODUCTION

Cancer is the most common cause of death and is a multifactorial disease [1,2]. Understanding the biological mechanisms of cancer in humans is crucial for the development of effective drugs in studies aimed at cancer treatment. It is essential to comprehend the type of cancer, its fundamental characteristics, and molecular mechanisms [3]. Therapeutic products developed against cancer constitute the highest revenue in the pharmacological market. It is expected that the estimated revenue in 2024 will exceed 142 billion dollars. When comparing the costs required for developing a new drug with anticancer activity and the costs of repositioning an already existing drug, drug repositioning is both faster and more economical when the process is evaluated.

The mammalian target of the Rapamycin (mTOR) pathway plays a crucial role in cancer development and progression. This pathway is frequently dysregulated in various cancers, including breast cancer, prostate cancer, ovarian cancer, and pancreatic cancer [4-7]. The dysregulation of the mTOR pathway is associated with uncontrolled cell cycle progression, cell growth, and survival, which are hallmark features of cancer cells [8,9]. Consequently, targeting the mTOR pathway has emerged as a promising therapeutic strategy for cancer treatment [10,11]. Studies have shown that mTOR inhibitors, such as rapamycin and temsirolimus, have demonstrated antitumor effects in various cancer types, including oral squamous cell carcinoma and prostate cancer [12,13]. Furthermore, the use of mTOR inhibitors in combination with other targeted or traditional chemotherapies is being actively investigated in clinical trials for prostate cancer [11]. Additionally, the mTOR pathway has been implicated in the maintenance of cancer stem-like cells, indicating its significance in tumor progression

and resistance [14]. The dysregulation of the mTOR pathway in cancer is often associated with the activation of downstream signaling molecules, such as Akt, which further promotes tumorigenesis and metastasis [5,15]. While mTOR inhibitors hold promises as cancer therapeutics, it is important to consider potential complications associated with their use. In conclusion, the mTOR pathway is a critical regulator of cancer cell behavior and represents an attractive target for cancer therapy. The dysregulation of this pathway contributes to tumorigenesis, metastasis, and resistance to treatment. Targeting the mTOR pathway, either alone or in combination with other therapeutic agents, holds significant potential for improving cancer treatment outcomes.

Preliminary results from a study indicated that patients who received remdesivir had a median recovery time of 11 days, compared to 15 days in those who received a placebo, suggesting a positive impact on recovery time [16]. Additionally, remdesivir's antiviral activity against COVID-19 has been assessed in humans, and it has been used to treat severe COVID-19 patients as an emergency use, further supporting its potential effectiveness [17]. Furthermore, the first approval of remdesivir as a treatment for COVID-19 reflects its potential as a remedy for severe cases [18]. However, it is important to note that the utility of remdesivir in treating SARS-CoV-2 may vary based on the setting. A study from a developing country reported conflicting results with respect to mortality in patients with severe COVID-19, indicating the need for further investigation into its effectiveness in different healthcare contexts [19]. Additionally, concerns have been raised regarding the adverse effects of remdesivir in patients, highlighting the importance of carefully monitoring its use during the treatment of COVID-19 [20]. The mechanism of action of remdesivir involves inhibiting the SARS-CoV-2 polymerase, which may contribute to its antiviral activity against the virus [21]. This mechanism provides insight into how remdesivir may exert its therapeutic effects in COVID-19 patients. In conclusion, while remdesivir has shown potential as a therapeutic option for COVID-19, further research is needed to fully understand its effectiveness, especially in different healthcare settings, and to carefully assess its adverse effects.

The identification and development of novel therapeutic strategies continue to be a pressing need in the field of drug discovery. In recent years, computational methods, specifically in-silico approaches, have emerged as powerful tools for exploring new avenues in drug repurposing. Repurposing existing drugs for new therapeutic targets accelerates the drug development process, taking advantage of known safety profiles and facilitating a quicker transition to clinical applications. One such promising target is the mechanistic target of rapamycin (mTOR) protein, a central regulator of cellular processes involved in growth, proliferation, and survival. Dysregulation of mTOR signaling has been implicated in various diseases, including cancer, neurodegenerative disorders, and metabolic syndromes. Consequently, targeting mTOR has become a focus of intense research for the development of innovative and effective therapeutic interventions.

In this study, we introduce a novel approach to drug repurposing, focusing on identifying potential candidates for mTOR protein inhibition using in-silico methods. Our research is based on the increasing evidence supporting the reuse of established drugs to address the challenges of drug development by providing alternative, cost-effective solutions. The selected candidate for repurposing, Remdesivir, has shown promising potential in early in-silico screenings, indicating possible interactions with the mTOR protein. Through detailed computational analysis, we investigate the binding affinity, molecular interactions, and structural implications of repurposing Remdesivir as an mTOR inhibitor. In molecular experiments conducted using the colorectal cancer cell line Caco2, we explore whether Remdesivir may exhibit anticancer activity by suppressing epithelial-mesenchymal transition (EMT) via the Hif1a/Twist/ß-catenin axis following mTOR inhibition. This study represents a significant step towards the rational design of therapeutic interventions, leveraging the efficiency and predictive power of computational methodologies. By repurposing Remdesivir for mTOR inhibition, we aim to contribute to the development of targeted therapies with potential applications across a spectrum of diseases. The success of this in-silico approach and experimental validations could pave the way for further *in-vivo* studies and clinical investigations, offering a novel perspective on drug discovery and therapeutic innovation.

MATERIAL AND METHOD

Cell Culture

Caco2 cell line was used and cultured with Dulbecco's Modified Eagle Medium-High Glucose (DMEM-HG) (Cat #11965092) completed with 10% Fetal Bovine Serum (FBS)(Cat #A5256801) and 1% penicillin-streptomycin (10,000 U/ml) (P/S) (Cat # 15140122) in a humidified incubator at 37°C and 5% CO_2. Cell culture maintained with weekly passages with trypsin-EDTA (0.25%), phenol red (Cat #25200056).

Drug Concentrations

To prepare the main stocks of both drugs, Remdesivir and Rapamycin were dissolved in dimethyl sulfoxide (DMSO) (Cat #D2650) with a final concertation of 30 mM and stored at -20°C. Further dilutions are done with complete DMEM-HG media with 10% FBS and 1% P/S.

MTT

Main stock of Remdesivir diluted with complete 500 µM, 200 µM, 100 µM, and 75 µM complete cell culture media. Main stock of Rapamycin diluted at 10 µM, 5 µM, 2 µM, 1 µM, 500 nM, 400 nM, 300 nM, 200 nM, 100 nM, 80 nM, 40 nM, 20 nM, 10 nM, and 5 nM with complete cell culture media. To assess the toxicity of the drugs and cell viability, 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide (MTT) assay was used. 10.000 cells/per well-seeded 96-well plate. After 24 hours, drug treatments were done and incubated for 48 hours. At the end of incubation time, 10 µl of MTT solution was added to wells and incubated for 4 hours. Formed formazan crystals dissolved with 100 µl DMSO. The measurement was taken at the end of the incubation period with absorption at 570 nm using an Epoch Microplate Spectrophotometer (Biotech Instruments USA).

ELISA

Total proteins are isolated with homemade NP-40 lysis buffer composed of 50 mM Tris-HCl (pH:8.5), 150 mM NaCl, 1% NP-40 (Cat #56741) and 1X Protein inhibit cocktail (Cat #635673) at final volume. BSA standard curve and total protein concentration were measured with BCA Protein Assay Reagent (Pierce, Rockford, Illinois, USA) following the manufacturer's protocol. For the ELISA, 100µg of total protein was used for the detection of mTOR with Human mTOR (Mammalian Target of Rapamycin) ELISA Kit (Elabscience, E-EL-H1655). Given manufacturer’s instructions followed, and absorbance was measured at 450nm with an Epoch Microplate Spectrophotometer (Biotech Instruments USA).

qPCR

Following the manufacturer's protocol, total RNA isolation was done with the NucleoSpin RNA kit from MACHEREY-NAGEL (USA) (Cat # 740955). After isolation, concentration of RNAs was measured with an Epoch Microplate Spectrophotometer with Take3 apparatus (Biotech Instruments USA). Due to the manufacturer's protocol, 1000 ng RNA was used for the iScript™ cDNA Synthesis Kit (Biorad, 1708891). SsoAdvanced Universal SYBR Green Supermix (Biorad, 1725272) was used for qPCR reactions.

Western Blot Analysis

Total proteins from control, and Remdesivir-treated cells were isolated. Protein lysates were evaluated by BCA assay (Thermo Fisher, USA). 30 µg protein from each group was loaded to 12% SDS-PAGE gel. Following the transfer and blocking steps with 5% BSA in TBS-0.1% Tween, the membrane was incubated with Hif1a (Cell Signaling Technology, #36169) and ß-catenin (Cell Signaling Technology, #8480) primary antibody and ß-actin (Cell Signaling Technology, #3700) primary antibody which was used as house-keeping control. Anti-rabbit (Cell Signaling Technology, #7074) and anti-mouse (Cell Signaling Technology, #7076) secondary antibodies were used, respectively. Band intensities were calculated using Image J (NIH, USA). Hif1a and ß-catenin band intensities were normalized according to total protein control.

Statistical Analysis

Experiments were performed in triplicate (n=3) for robust statistical validity. Statistical evaluations were performed using GraphPad software. Student's t-test was used to evaluate numerical variables with normal distribution among experimental groups. For multiple comparisons, Tukey's test, one-way ANOVA, and two-way ANOVA tests were employed, with significance set at $p<0.05$.

***In silico* Analysis**

Molecular docking experiments were performed utilizing the Autodock Vina software [22,23]. The receptor protein structure (4DRI) was obtained from the protein database. Prior to the docking calculations, the structure underwent a process to eliminate water molecules and other heteroatoms. Utilization of BIOVIA Discovery Studio 2021 (BIOVIA, Dassault Systèmes, Discovery Studio, 2021, San Diego: Dassault Systèmes, 2021) facilitated the preparation of the docking process and the subsequent analysis of the results.

RESULT AND DISCUSSION

In this study, new candidate molecules for the inhibition of the mTOR protein were screened using *in silico* methods, and it was found that Remdesivir yielded significant preliminary results. *In silico* analyses performed within the scope of the study showed that remdesivir may be a good mTOR blocker and this hypothesis was also demonstrated by *in vitro* studies. Detailed in-silico analysis showed that the binding energy of remdesivir to mTOR protein was -9.8 kcal/mol (Table 1). This value was -19.2 kcal/mol for rapamycin using the same method. Since rapamycin is a natural mTOR inhibitor (the mTOR protein is named after rapamycin – the mechanistic target of rapamycin), this value should be quite low. One of the most interesting points in the study is that the total interaction energy with remdesivir is also quite low and the ratio between the interaction values obtained by *in silico* analysis (Table 1) is confirmed by *in vitro* studies.

Table 1. Binding affinity of Rapamycin and Remdesivir with mTOR protein

Compound	Affinity (kcal/mol)
Rapamycin	-19.2
Remdesivir	-9.8

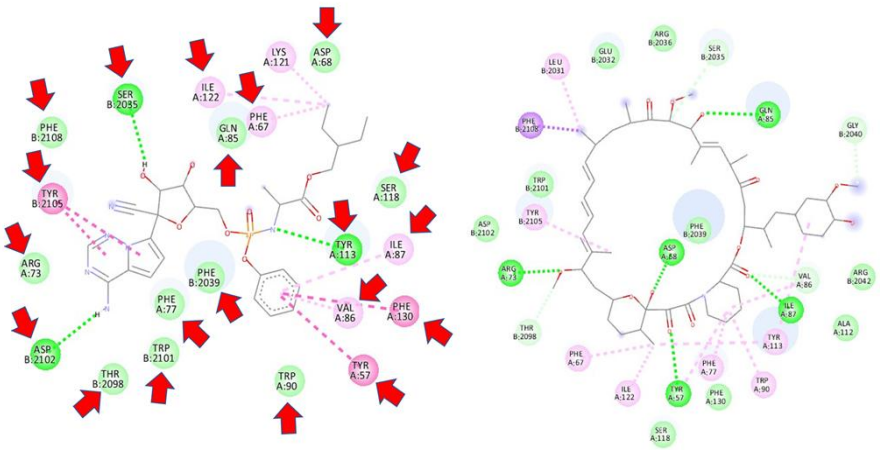

Figure 1. mTOR protein amino acid interactions of Remdesivir and Rapamycin

The comprehensive computational analysis revealed that remdesivir interacts with all amino acid residues that rapamycin interacts with, except for residue LYS121 (Figure 1); conversely, rapamycin does not interact with residue LYS121.

Table 2. Comparison of interaction residues, bond types, and bond lengths (Å) between Rapamycin and Remdesivir with mTOR protein

Residue	**Bond Type**		**Bond Length (A°)**	
	Rapamycin	Remdesivir	Rapamycin	Remdesivir
ASP68	Hydrogen	van der Waals	2.83	
PHE67	Pi-Alkyl	Pi-Alkyl	5.04	4.82
GLN85	Hydrogen	van der Waals	2.89	
ILE122	Pi-Alkyl	Pi-Alkyl	4.73	5.10
SER2035	Carbon Hydrogen	Hydrogen	3.32	2.33
PHE2108	Pi-Sigma	van der Waals	3.61	
TYR2105	Pi-Alkyl	Pi-Alkyl	4.23	3.9
ARG73	Hydrogen	van der Waals	2.39	
ASP2102	van der Waals	Hydrogen		2.65
THR2098	Carbon Hydrogen	van der Waals	3.39	
TRP2101	van der Waals	van der Waals		
PHE77	Pi-Alkyl	van der Waals	5.25	
PHE2039	van der Waals	van der Waals		
TRP90	Pi-Alkyl	van der Waals	4.58	
TYR57	Hydrogen	Pi-Pi	2.86	5.84
VAL86	Pi-Alkyl	Pi-Alkyl	2.32	4.85
PHE130	van der Waals	Pi-Pi		5.78
TYR113	Pi-Alkyl	Hydrogen	5.25	2.40
ILE87	Hydrogen	Pi-Alkyl	2.08	5.32
SER118	van der Waals	van der Waals		

Rapamycin (C51H79NO13 - 914.187 g/mol) is a larger molecule compared to Remdesivir (C27H35N6O8 - 602.585 g/mol) and therefore it can interact more with mTOR. Detailed *in silico* evaluation revealed that remdesivir binds to all residues that rapamycin binds to except residue LYS121; rapamycin does not bind to residue LYS121. As seen in Table 2, except for the hydrogen bond with residue SER2035, there are no hydrogen bonds with unchanged bond characteristics. The length of the hydrogen bond of rapamycin with SER2025 residue is 3.32 A°, while the length of the bond with remdesivir is 2.33 A°. Here we can say that the interaction becomes stronger due to the shortening of the bond. Apart from the hydrogen bond whose profile was preserved, the profiles of Pi bonds with PHE67, ILE122, TYR2105, VAL86 and van der Waals bonds with TRP2101, SER118 were preserved in both drugs. Here, Remdesivir showed a radical increase in the length of the bond with VAL86 However, the bond profiles with ASP68, GLN85, ARG73, THR2098, TYR57, TYR57, ILE87 changed from Hydrogen -> van der Waals, Hydrogen -> van der Waals, Hydrogen -> van der Waals, Hydrogen -> van der Waals, Hydrogen -> van der Waals, Hydrogen -> Pi, Hydrogen -> Pi, respectively. One of the interesting findings is that rapamycin forms van der Waals interaction with ASP2102 residue while hydrogen bonding with Remdesivir. Accordingly, the distance with the ASP2102 residue decreased and the interaction became stronger.

To investigate whether Remdesivir inhibits the mTOR protein, the Caco2 cell line was employed. For this purpose, an MTT cell viability assay was conducted to determine the dose of Remdesivir to be applied *in vitro*. The doses tested in Caco2 cells were 75 µM, 100 µM, 200 µM, and 500 µM, respectively. Experiments were carried out with 75 µM Remdesivir application to examine molecular changes without cytotoxic effects on the cells (Figure 2A).

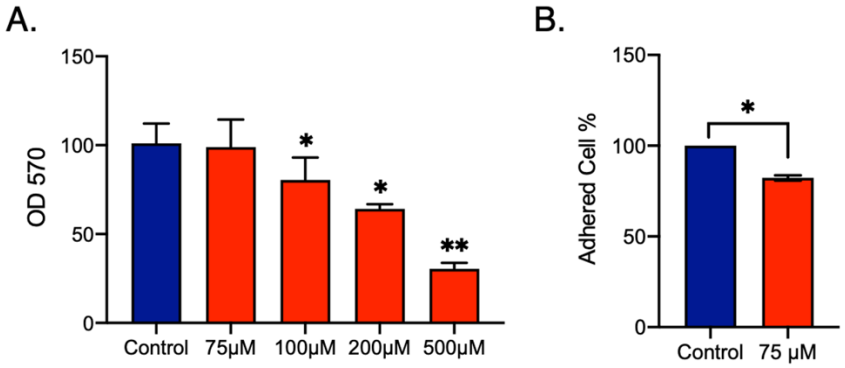

Figure 2. (A) The effect of Remdesivir at 75, 100, 200, and 50 µM concentrations on Caco2 cells in 48h. (B) The percentage of adhered cells after 48h Remdesivir treatment. Blue bars represent the cells treated with the only cell culture media (Control), and red bars represent those treated with Remdesivir. Data were expressed as the mean ± SD (n = 3). *p < 0.05, **p<0.005

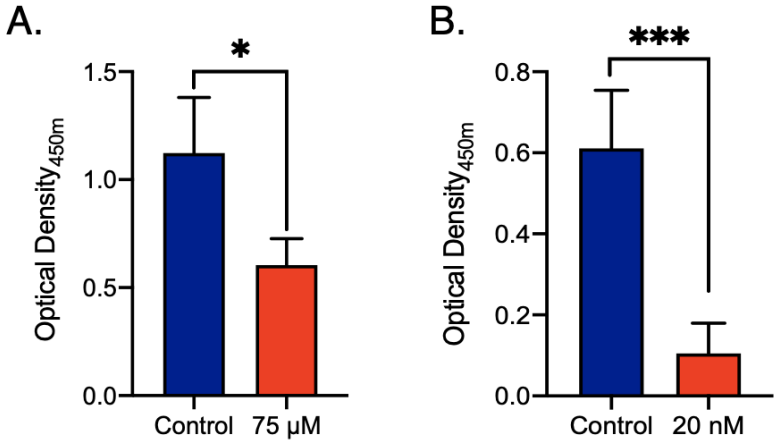

Figure 3. Alteration of mTOR levels after Remdesivir tor Rapamycin treatment. Blue bars represent the cells treated with the only cell culture media (Control), and red bars represent the cells treated either with Remdesivir (A) or Rapamycin (B). Data were expressed as the mean ± SD (n = 3). *p < 0.05, ***p<0.0005

To assess the functional impact of the selected dose on the cells, a cell adhesion assay was performed initially. A decrease in adhesion in Caco2 cells was observed 48 hours after the application of 75 µM Remdesivir (Figure 2B). After confirming the functional effect of the selected dose on the cells, ELISA assay was conducted to determine whether Remdesivir induces mTOR inhibition. Rapamycin (20 nM) was used as a positive control parallel to Remdesivir application (Figure 3). ELISA results indicated that Remdesivir halved the expression of mTOR (Figure 3A and 3B). To better evaluate the anticancer activity of Remdesivir on Caco2 cells, the expressions of Twist (Figure 4A), a transcription factor involved in epithelial-mesenchymal transition (EMT), and Hif1a (Figure 4B), critical in both EMT and metastasis development, were examined. Our results demonstrated a significant decrease in the expression of two genes in Caco2 cells upon the application of 75 µM Remdesivir.

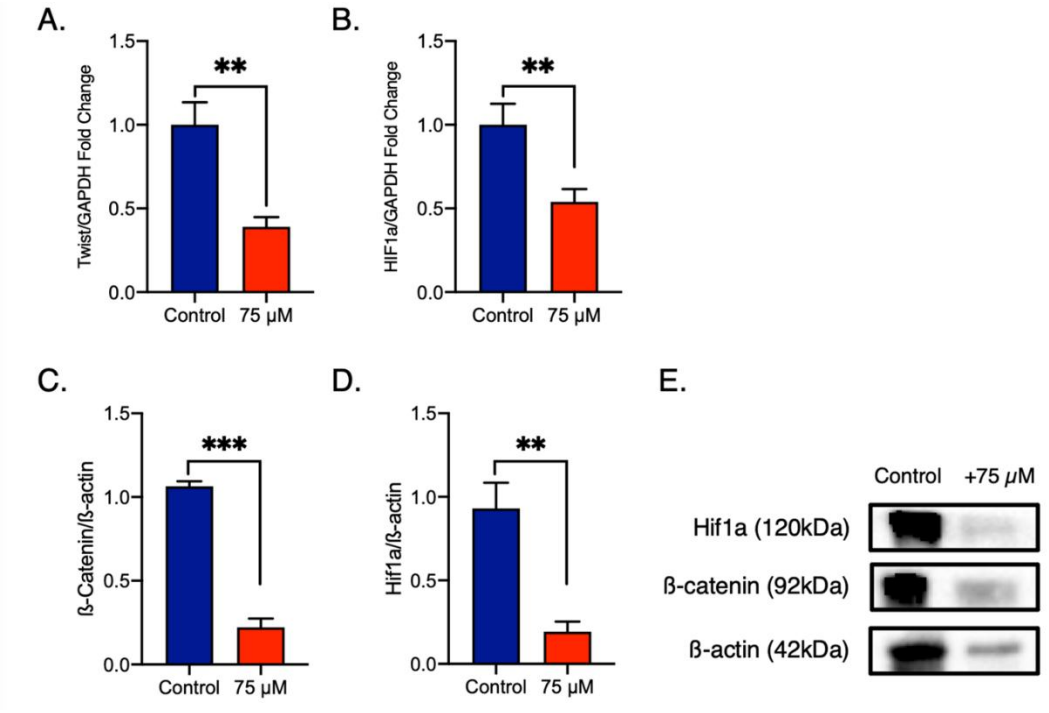

Figure 4. Effects of Remdesivir on Caco2 cells. The mRNA expression levels of (A) TWIST and (B) HIF1a were analyzed by qRT-PCR. The protein expression levels of (C) ß-catenin and (D) Hif1a were analyzed using western blot analysis. Blue bars represent the cells treated with the only cell culture media (Control), and red bars represent those treated with Remdesivir. Protein levels were normalized with their ß-actin level. The representative band images are shown in (E). qRT-PCR data were expressed as the mean ± SD (n = 3), and Western blot data were expressed as the mean ± SD (n = 3). **p < 0.005, ***p<0.0005

In our proposed molecular pathway, following the inhibition of mTOR by Remdesivir, a decrease in Hif1a expression and subsequently in Twist expression was anticipated. The decrease in Hif1a expression was confirmed at both the transcript and protein levels (Figure 4B-4D). Another anticipated decrease following the suppression of Twist and Hif1a expression was in the expression of ß-catenin, critical in EMT. Confirmation was obtained for the decrease in ß-catenin expression at the protein level as well (Figure 4C). This study demonstrates that Remdesivir acts as an mTOR inhibitor and its downstream effects can suppress EMT via the Hif1a/Twist/ß-catenin axis in the colorectal cancer cell line Caco2, thereby eliciting an anticancer response.

The main findings of the study indicate the potential of Remdesivir to inhibit the mTOR signaling pathway and its potential anticancer effects in Caco2 cells. The comparable interaction energy of Remdesivir with the mTOR protein to that of a potent mTOR inhibitor like rapamycin highlights the compound's potential as an mTOR inhibitor. This suggests that Remdesivir could be effective in regulating the mTOR signaling pathway, thereby potentially inhibiting the growth, and spread of cancer cells.

The methodological approach of the study has facilitated a comprehensive evaluation of Remdesivir's efficacy as an mTOR inhibitor. The dose determined via the MTT cell viability assay showed no cytotoxic impact on the cells and was considered approfpriate for investigating cellular functional alterations. Functional analyses such as cell adhesion assays and ELISA were utilized to assess the effects of Remdesivir on cell adhesion capacity and changes in mTOR protein expression, respectively.

In the study aimed at examining the anticancer effects of Remdesivir in Caco2 cells, a significant decrease in the expression of genes associated with EMT was observed. Particularly, the decrease in the expression of Hif1a suggests that Remdesivir may suppress the EMT process, thereby inhibiting the

invasive properties of cancer cells. Hif1a, notably associated with hypoxia and alterations in the tumor microenvironment, plays a crucial role in metastasis, invasion, and cell proliferation in cancer [24]. Therefore, the low expression of Hif1a induced by Remdesivir suggests its potential to limit the aggressive characteristics of cancer cells.

Furthermore, molecular pathway analysis elucidates the changes in the expression of EMT-associated factors following Remdesivir's function as an mTOR inhibitor. The inhibition of the mTOR signaling pathway by Remdesivir is believed to be the cause of the observed decrease in the expression of factors such as Hif1a, Twist, and ß-catenin associated with EMT. Particularly, Twist is known as a significant regulator of the EMT process and is considered an indicator of cancer metastasis [25]. The reduction in Twist expression induced by Remdesivir suggests its potential contribution to inhibiting metastatic properties.

These findings suggest that Remdesivir may contribute to reducing the invasive characteristics of cancer cells. The function of Remdesivir as an mTOR inhibitor and the changes in the expression of EMT-associated factors imply its potential to limit the aggressive behaviors of cancer cells. However, these findings need to be validated in a broader range of cancer cell lines and clinical samples. Further research should explore these mechanisms more thoroughly to enhance our understanding of Remdesivir's potential in cancer treatment and contribute to the advancement of treatment approaches. Additionally, conducting *in vivo* studies to determine the potential of Remdesivir in cancer therapy is crucial. These studies can evaluate the usability of Remdesivir as a new therapeutic strategy in cancer treatment and provide a new direction for research in this field.

Remdesivir is an antiviral drug used in the treatment of viral infections such as COVID-19 [26]. Its side effects are generally mild to moderate, with commonly reported manageable gastrointestinal disturbances. The most frequent adverse effects include nausea, vomiting, headache, dizziness, and skin rash. More severe side effects may involve increased liver enzymes, renal function impairment, and coagulation abnormalities [16]. On the other hand, previously identified adverse effects of mTOR inhibitors tend to be more diverse and severe.

mTOR is a protein that regulates cellular growth, proliferation, and metabolism, and mTOR inhibitors are known for their immunosuppressive and antitumor effects [27]. Commonly encountered adverse effects of clinically used mTOR inhibitors include metabolic disturbances, particularly hyperlipidemia and hypertriglyceridemia. Additionally, they can induce hyperglycemia and increase the risk of infections, particularly fungal and viral infections, especially in immunocompromised patients [28]. In addition to these side effects, gastrointestinal disturbances such as nausea, vomiting, and diarrhea, as well as peripheral edema and pulmonary toxicity, have been associated with the use of mTOR inhibitors [29,30].

Therefore, when evaluating the use of remdesivir as an alternative to currently used mTOR inhibitors, considering their documented side effects, remdesivir may be considered less harmful. Consequently, remdesivir could be regarded as a safer treatment alternative.

This study highlights the potential of Remdesivir as an inhibitor of the mTOR signaling pathway and its potential anticancer effects in Caco2 cells. The comparable interaction energy of Remdesivir with the mTOR protein to that of rapamycin enhances its promise as an mTOR inhibitor. Additionally, it supports its potential to regulate the mTOR pathway and potentially inhibit cancer cell proliferation and dissemination. Methodologically, the approach of the study allowed for a comprehensive evaluation of Remdesivir's efficacy, demonstrating no cytotoxicity and enabling the exploration of cellular functional changes. The investigation of Remdesivir's impact on EMT-associated gene expression, particularly significant reductions observed in Hif1a, suggests its potential to suppress EMT and inhibit cancer cell dissemination. Molecular pathway analysis further supports this, demonstrating alterations in EMT-associated factors following Remdesivir's mTOR inhibition. Besides, this research brings to light the promising capacity of Remdesivir as an inhibitor of mTOR and its potential anti-cancer characteristics in laboratory experiments. More precisely, the assessment of Remdesivir's effectiveness and safety in animal subjects and ultimately in clinical trials will be crucial for determining its therapeutic importance in cancer therapy. Moreover, exploring potential combination treatments involving Remdesivir with other specific agents or traditional chemotherapy medications could amplify its efficacy and expand its relevance across various cancer categories. Furthermore, clarifying the

mechanisms behind Remdesivir's dual functionality as an antiviral substance and an mTOR inhibitor might reveal a new understanding of cancer biology and therapeutic strategies. Sustained interdisciplinary cooperation and rigorous analysis are vital for leveraging the complete therapeutic potential of Remdesivir in the battle against cancer. Further exploring these mechanisms will enhance our understanding of Remdesivir's potential in cancer therapy and advance treatment strategies. Additionally, conducting *in vivo* studies to determine its clinical utility is crucial, indicating the potential for Remdesivir to open up a new therapeutic avenue in cancer treatment.

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AUTHOR CONTRIBUTIONS

Concept: Z.B.A., G.Y.S., M.A.Ü., A.Y.A.; Design: Z.B.A., G.Y.S., A.Y.A., M.G.; Control: A.Y.A., M.G.; Sources: Z.B.A., G.Y.S.; Materials: M.A.Ü., M.G.; Data Collection and Processing: Z.B.A., G.Y.S., M.A.Ü., A.Y.A., M.G.; Analysis and/or Interpretation: M.A.Ü.; Literature Review: Z.B.A., M.A.Ü., M.G.; Manuscript Writing: Z.B.A., G.Y.S., M.A.Ü., A.Y.A.; Critical Review: Z.B.A., M.A.Ü., A.Y.A., M.G.; Other: -

CONFLICT OF INTEREST

The authors declare that there is no real, potential, or perceived conflict of interest for this article.

ETHICS COMMITTEE APPROVAL

The authors declare that the ethics committee approval is not required for this study.

REFERENCES

1. Afrin, H., Salazar, C.J., Kazi, M., et al. (2022). Methods of screening, monitoring and management of cardiac toxicity induced by chemotherapeutics. Chinese Chemical Letters, 33(6), 2773-2782. [CrossRef]
2. Siegel, R.L., Miller, K.D., Fuchs, H.E., et al. (2021). Erratum to “cancer statistics, 2021”. CA:A Cancer Journal for Clinicians, 71(4), 359-359. [CrossRef]
3. Alshehri, S., Imam, S.S., Rizwanullah, M.D., et al. (2021). Progress of cancer nanotechnology as diagnostics, therapeutics, and theranostics nanomedicine: Preclinical promise and translational challenges. Pharmaceutics, 13(1), 24. [CrossRef]
4. Feng, T.T., Zheng, L., Liu, F., et al. (2016). Growth factor progranulin promotes tumorigenesis of cervical cancer via PI3K/Akt/mTOR signaling pathway. Oncotarget, 7(36), 58381-58395. [CrossRef]
5. Li, X.Y., Yang, Q.F., Yu, H.Y., et al. (2014). LIF promotes tumorigenesis and metastasis of breast cancer through the AKT-mTOR pathway. Oncotarget, 5(3), 788-801. [CrossRef]
6. Rousseau, B., Guillemin, A., Duvoux, C., et al. (2019). Optimal oncologic management and mTOR inhibitor introduction are safe and improve survival in kidney and liver allograft recipients with carcinoma. International Journal of Cancer, 144(4), 886-896. [CrossRef]
7. Yu, S.W., Shen, G.X., Khor, T.O., et al. (2008). Curcumin inhibits Akt/mammalian target of rapamycin signaling through protein phosphatase-dependent mechanism. Molecular Cancer Therapeutics, 7(9), 2609-2620. [CrossRef]
8. Papavassiliou, K.A., Zoi, I., Gargalionis, A.N., et al. (2019). Polycystin-1 affects cancer cell behaviour and interacts with mTOR and Jak signalling pathways in cancer cell lines. Journal of Cellular and Molecular Medicine, 23(9), 6215-6227. [CrossRef]
9. Wu, X.J., Li, S.H., Hu, X., et al. (2019). mTOR signaling upregulates CDC6 via suppressing miR-3178 and promotes the loading of DNA replication helicase. Scientific Reports, 9, 9805. [CrossRef]
10. Dong, Y.Y., Gong, Y.X., Kuo, F.S., et al. (2022). Targeting the mTOR pathway in hurthle cell carcinoma results in potent antitumor activity. Molecular Cancer Therapeutics, 21(2), 382-394. [CrossRef]

11. Morgan, T.M., Koreckij, T.D., Corey, E. (2009). Targeted therapy for advanced prostate cancer: Inhibition of the PI3K/Akt/mTOR pathway. Current Cancer Drug Targets, 9(2), 237-249. [CrossRef]
12. German, S., Aslam, H.M., Saleem, S., et al. (2013). Carcinogenesis of PIK3CA. Hereditary Cancer in Clinical Practice, 11, 5. [CrossRef]
13. Okui, T., Shimo, T., Fukazawa, T., et al. (2010). Antitumor effect of temsirolimus against oral squamous cell carcinoma associated with bone destruction. Molecular Cancer Therapeutics, 9(11), 2960-2969. [CrossRef]
14. Matsubara, S., Ding, Q., Miyazaki, Y., et al. (2013). mTOR plays critical roles in pancreatic cancer stem cells through specific and stemness-related functions. Scientific Reports, 3, 3230. [CrossRef]
15. Lee, Y.K., Lee, W.S., Kim, G.S., et al. (2010). Anthocyanins are novel AMPKα1 stimulators that suppress tumor growth by inhibiting mTOR phosphorylation. Oncology Reports, 24(6), 1471-1477. [CrossRef]
16. McMahon, J.H., Udy, A., Peleg, A.Y. (2020). Remdesivir for the treatment of Covid-19-preliminary report. New England Journal of Medicine, 383(10), 992-993. [CrossRef]
17. Frediansyah, A., Nainu, F., Dhama, K., et al. (2021). Remdesivir and its antiviral activity against COVID-19: A systematic review. Clinical Epidemiology and Global Health, 9, 123-127. [CrossRef]
18. Lamb, Y.N. (2020). Remdesivir: First Approval. Drugs, 80(13), 1355-1363. [CrossRef]
19. Malik, M.I., Zafar, S.A., Malik, M., et al. (2022). The utility of remdesivir in SARS-CoV-2: A single tertiary care center experience from a developing country. Exploratory Research in Clinical and Social Pharmacy, 5, 100107. [CrossRef]
20. Doggrell, S.A. (2020). Remdesivir, a remedy or a ripple in severe COVID-19? Expert Opinion on Investigational Drugs, 29(11), 1195-1198. [CrossRef]
21. Wakchaure, P.D., Ghosh, S., Ganguly, B. (2020). Revealing the inhibition mechanism of RNA-Dependent RNA Polymerase (RdRp) of SARS-CoV-2 by remdesivir and nucleotide analogues: A molecular dynamics simulation study. Journal of Physical Chemistry B, 124(47), 10641-10652. [CrossRef]
22. Eberhardt, J., Santos-Martins, D., Tillack, A.F., et al. (2021). AutoDock Vina 1.2.0: New docking methods, expanded force field, and python bindings. Journal of Chemical Information and Modeling, 61(8), 3891-3898. [CrossRef]
23. Trott, O., Olson, A.J. (2010). Software news and update autodock vina: Improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. Journal of Computational Chemistry, 31(2), 455-461. [CrossRef]
24. Bui, B.P., Nguyen, P.L., Lee, K., et al. (2022). Hypoxia-Inducible Factor-1: A novel therapeutic target for the management of cancer, drug resistance, and cancer-related pain. Cancers, 14(24), 6054. [CrossRef]
25. Ribatti, D., Tamma, R., Annese, T. (2020). Epithelial-mesenchymal transition in cancer: A historical overview. Translational Oncology, 13(6), 100773. [CrossRef]
26. Levien, T.L., Baker, D.E. (2023). Remdesivir. Hospital Pharmacy, 58(5), 420-430. [CrossRef]
27. Panwar, V., Singh, A., Bhatt, M., et al. (2023). Multifaceted role of mTOR (mammalian target of rapamycin) signaling pathway in human health and disease. Signal Transduction and Targeted Therapy, 8(1), 375. [CrossRef]
28. Gomez-Pinillos, A., Ferrari, A.C. (2012). mTOR signaling pathway and mTOR inhibitors in cancer therapy. Hematology-Oncology Clinics of North America, 26(3), 483-505. [CrossRef]
29. Pallet, N., Legendre, C. (2013). Adverse events associated with mTOR inhibitors. Expert Opinion on Drug Safety, 12(2), 177-186. [CrossRef]
30. Zou, Z.L., Tao, T., Li, H.M., et al. (2020). mTOR signaling pathway and mTOR inhibitors in cancer: Progress and challenges. Cell and Bioscience, 10(1), 31. [CrossRef]