Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

International Journal of Antimicrobial Agents 56 (2020) 106053

Contents lists available at ScienceDirect

International Journal of Antimicrobial Agents

journal homepage: www.elsevier.com/locate/ijantimicag

Macrolides and viral infections: focus on azithromycin in COVID-19 pathology

Arianna Pani[a,b], Marinella Lauriola[c], Alessandra Romandini[a,b], Francesco Scaglione[a,b,*]

[a] Department of Oncology and Hemato-oncology, Postgraduate School of Clinical Pharmacology and Toxicology, University of Milan, Milan, Italy
[b] Clinical Pharmacology Unit, ASST Grande Ospedale Metropolitano Niguarda, Milan, Italy
[c] Infectiouos Disease Department, Policlinico di Monza, Monza, Italy

ARTICLE INFO

Article history:
Received 29 April 2020
Accepted 6 June 2020

Keywords:
COVID-19
Macrolides
Azithromycin
Hydroxychloroquine

ABSTRACT

The emergence of the new COVID-19 virus is proving to be a challenge in seeking effective therapies. Since the most severe clinical manifestation of COVID-19 appears to be a severe acute respiratory syndrome, azithromycin has been proposed as a potential treatment. Azithromycin is known to have immunomodulating and antiviral properties. In vitro studies have demonstrated the capacity of azithromycin in reducing production of pro-inflammatory cytokines such as IL-8, IL-6, TNF alpha, reduce oxidative stress, and modulate T-helper functions. At the same time there are multiple clinical evidences of the role of azithromycin in acute respiratory distress syndrome and against Middle East Respiratory syndrome (MERS). Some preliminary evidence has demonstrated controversial results regarding efficacy of azithromycin in combination with hydroxychloroquine in COVID-19. First, a French trial demonstrated 100% virological negativizing of six patients treated with azithromycin plus hydroxychloroquine vs. 57.1% of patients treated with only hydroxychloroquine and 12.5% of the control group ($P < 0.05$). On the other hand, another case series revealed no efficacy at all on 11 patients treated with the same combination and doses. Furthermore, there are some concerns regarding the association of azithromycin and hydroxychloroquine because of potential QT prolongation. In fact, both drugs have this as a potential side effect and evidence regarding the safe use of this combination is controversial. Despite the necessity to quickly find solutions for COVID-19, extreme caution must be used in evaluating the risk-benefit balance. However, based on preclinical and clinical evidence and some preliminary results in COVID-19, azithromycin could have potential in the fight against this new disease.

1. Introduction

Macrolides are bacteriostatic antibiotics that are widely used in clinical practice against many Gram-positive and atypical bacterial species that are commonly associated with respiratory tract infections. In addition to their antibacterial effects, macrolides have been shown to have immunomodulatory and anti-inflammatory effects [1–3]. The severity and mortality of respiratory viral infections, including COVID-19, are associated with the host's excessive inflammatory response characterized by hyper-production of cytokines [4–6]. Preclinical and clinical studies have shown that macrolides regulate the inflammatory response, attenuating the production of anti-inflammatory cytokines and also promoting the production of immunoglobulins [7]. These regulatory effects on the immune response reduce complications of respiratory viral infections [8–10]. Due to these immunomodulating properties, macrolides (e.g. azithromycin, clarithromycin, erythromycin, and fidaxomycin) have been extensively studied for their potential use as adjunctive broad-spectrum therapy for viral respiratory infections including influenza [7,10–13].

This narrative review explored the role of macrolides in COVID-19 pathology. It focused on azithromycin, and considered it as the most suitable macrolide in a possible therapeutic combination. A literature search was performed on MEDLINE with the following search terms: "azithromycin and viral infections", "azithromycin and SARS-CoV2", "azithromycin and COVID-19", "azithromycin and Qt prolongation", and "azithromycin and chloroquine and Qt prolongation". The most up-to-date evidence and all those relevant to the role of macrolides in COVID-19 treatment were selected.

* Corresponding author at: Department of Oncology and Hemato-oncology University of Milan, Italy Via Vanvitelli 32 20129-Milan.
E-mail address: francesco.scaglione@unimi.it (F. Scaglione).

https://doi.org/10.1016/j.ijantimicag.2020.106053
0924-8579/

2. Macrolides in viral infections

Clarithromycin, azithromycin, erythromycin, bafilomycin A1, and telithromycin have shown to have anti-inflammatory and immunomodulatory effects [10]. For this reason, macrolides have been proposed as options for viral respiratory infections presenting an inflammatory basis, including COVID-19. The immunomodulating activities of azithromycin are shown in two different phases of the disease: during the acute phase and at the resolution of the chronic inflammation. In the acute phase, the ability of azithromycin to reduce the production of pro-inflammatory cytokines – such as IL-8, IL-6, TNF alpha, and MMPs – is thoroughly demonstrated [14]. In the resolution phase, this macrolide has been shown to increase neutrophil apoptosis and the oxidative stress related with inflammation. Also, clarithromycin, bafilomycin A1 and erythromycin have been found to inhibit the production of the intercellular adhesion molecule (ICAM)-1 and IL-1β, IL-6, IL-8 and TNF-α in rhinovirus and influenza infection models [11,15–17].

In a study conducted by Murphy et al., azithromycin was associated with a shift of the T-helper phenotype from type I to type II, favoring tissue repair after inflammation. Azithromycin has also been found to attenuate the effects of lipopolysaccharide on lung allograft bronchial epithelial cells [11,18–22]. In addition, this drug is able to significantly reduce the expression of iNOS and the pro-inflammatory macrophage receptor (CCR7) by increasing the activity of arginase and the anti-inflammatory macrophage receptors (MR and CD23) [23–25]. All of these effects are explained by the azithromycin-mediated inhibition of the nuclear factor-kappa B (NF-kB). Azithromycin has also shown in vitro efficacy against Zika virus, reducing viral viability and proliferation of the virus [26]. A paper by Menzel et al. demonstrated that azithromycin can transiently, although strongly, induce interferon expression in the bronchial epithelium of patients with chronic obstructive pulmonary disease (COPD) when infected with rhinovirus [27] and this may explain the ability of azithromycin to reduce exacerbation frequency in COPD patients [28,29].

Despite their well-established anti-inflammatory and immunomodulatory properties, macrolides do not have a direct antiviral effect and have shown controversial results in clinical trials. In one randomized controlled trial, adult patients hospitalized for laboratory-confirmed flu were randomized to receive oseltamivir and azithromycin or oseltamivir alone, both for 5 days. Proinflammatory cytokines decreased more rapidly in the oseltamivir-azithromycin group. However, the decline in viral RNA was not affected by the addition of azithromycin [8]. In another prospective, double-blind, controlled trial in 24 healthy subjects inoculated with rhinovirus (who were seronegative for antibodies prior to the inoculation) and assigned to receive either clarithromycin or trimethoprim-sulfamethoxazole, no effects were noticed in favor of clarithromycin in terms of symptom reduction or white blood cell and neutrophil counts, and in the concentrations of interleukins 6 and 8 in nasal lavage fluid during the cold [30].

In a study by Arabi et al. of 349 patients with Middle East Respiratory Syndrome (MERS) in a critical condition, 136 (39%) received macrolide therapy. Azithromycin was the most commonly used (97 of 136; 71.3%). Macrolide therapy was commonly started before the patient arrived in the intensive care unit (ICU) (51 of 136; 37.5%) or on day 1 in the ICU (53 of 136; 39%). At the time of ICU admission, the baseline characteristics of patients who received and did not receive macrolides were similar, including demographics and the sequential organ failure assessment score. Moreover, no statistically significant between-group differences were found in in-hospital mortality, ICU and 90-day mortality, and hospital length of stay [31].

In children hospitalized for respiratory syncytial virus (RSV) bronchiolitis, clarithromycin was associated with a reduction in hospital length of stay, oxygen need, treatment with β2-agonists, and re-hospitalizations within 6 months [12]. It should be pointed out that this study has been widely criticized because of errors in statistical analysis, methodology and number of enrolled participants [32]. Results on hospital readmissions reported by Tahan et al. have not been confirmed in a larger RCT comparing azithromycin and placebo in children with bronchiolitis [33]. Regarding COVID-19, azithromycin has proven to have an EC50 of 2.12 mM against SARS-CoV-2 in an in vitro screening of Food and Drug Administration (FDA)-approved chemical libraries [34].

In addition to the aforementioned effect on inflammatory response, macrolides could play a prophylactical role in pneumococcal and staphylococcal bacterial complications that occur with a certain frequency as complications of respiratory viral infections.

3. Efficacy of azithromycin in COVID-19

In a French clinical trial of 20 patients treated with hydroxychloroquine compared with 16 controls (patients who were refusing treatment with hydroxychloroquine or had contraindications), six were treated with a combination of hydroxychloroquine 200 mg three times a day for 10 days and azithromycin 500 mg on the first day, followed by 250 mg daily for another 4 days. Comparing the outcomes between patients treated with hydroxychloroquine alone, in combination with azithromycin or controls, the authors found that 100% of patients treated with the combination were virologically healed at day 6 vs. 57.1% of patients treated with only hydroxychloroquine and 12.5% of the control group ($P < 0.05$) [35]. In contrast with this result, Molina et al. reported the outcomes obtained in 11 consecutive patients treated with a combination of hydroxychloroquine plus azithromycin at the same dose scheme reported by Gautret et al.: none of the 11 patients benefited from the treatment [35,36]. Of note, in the case series reported by Molina, eight of 11 patients did have significant comorbidities linked with poor outcomes (obesity, solid and hematological cancer, HIV-infection). One patient was discontinued after 4 days because of QT prolongation.

An update of the study by Gautret et al. reported a favorable outcome (defined as patient discharged not requiring aggressive oxygen therapy) in 65 of 80 patients (81.3%) treated with hydroxychloroquine and azithromycin and a negative viral load test at 6 days in 83% of patients with the combination: 15% required oxygen therapy, three needed ICU admission but then improved and returned to the infectious disease ward, and one died [35].

Two large studies on the efficacy of the combination of azithromycin and hydroxychloroquine were recently published. Rosenberg et al. published a retrospective multicenter cohort study on 1438 hospitalized patients with COVID-19, 735 of whom received hydroxychloroquine plus azithromycin as treatment for COVID-19. Comparing in-hospital mortality of patients who received the combination with that of those who received hydroxychloroquine alone, azithromycin alone or no treatment, no significant differences were observed among the four groups [37]. Also, Mehra et al. reported an outcome against the benefit of using hydroxychloroquine (or chloroquine) with a macrolide (azithromycin or clarithromycin) on a population of 96 032 patients hospitalized for COVID-19. The authors compared in-hospital mortality of patients treated with the combination macrolide/quinoline derivatives with those of patients receiving no treatments for COVID-19; they found that the combinations where associated with an increased risk of mortality [38].

Currently, many ongoing trials are evaluating the efficacy of azithromycin in COVID-19. The schemes predominantly being evaluated are: azithromycin versus placebo, in combination or versus hydroxychloroquine or in triple combination with tocilizumab (NCT04329832, NCT04341870, NCT04334382, NCT04348474,

NCT04332107, NCT04341207, NCT04339426, NCT04329572, NCT04336332, NCT04332094, NCT04335552, NCT04339816, NCT04338698, NCT04328272, NCT04347512, NCT04349592, NCT04345861, NCT04321278, NCT04344444, NCT04322396, NCT04322123, NCT04324463, NCT04334512, NCT04351919, NCT04341727, NCT04345419, NCT04332835, NCT04347031, and NCT04349410). A French trial is also evaluating the efficacy of azithromycin and hydroxychloroquine in the prevention of SARS-CoV-2 infection in health workers exposed to the virus (NCT04344379). Azithromycin is also one of the drugs included in the large adaptive RECOVERY trial, the English national study sponsored by the University of Oxford EudraCT 2020-001113-21.

4. Co-administration of azithromycin and hydroxychloroquine, and QT interval prolongation

Following some reports, the FDA noticed (in 2012) a small increase in cardiovascular deaths and deaths from any cause among patients taking azithromycin for a 5-day cycle course [39]. It was hypothesized that azithromycin could increase the QTc with the risk of arrhythmias. On 12 March 2013, the FDA published a communication on azithromycin safety on heart rhythms, warning on the risk of potentially fatal outcomes [40]. Following the revision of many studies both before and after the public health note, the FDA modified it stating that the potential risk must be assessed when azithromycin is used in the presence of risk factors such as QTc interval prolongation, hypokalemia, hypomagnesaemia, bradycardia, or co-administration with antiarrhythmic drugs such as quinidine, procainamide, dofetilide, amiodarone, and sotalol (drugs associated with prolongation of the QTc interval). In addition, it must be specified that the FDA note was linked to the reporting of torsade de pointes following the use of azithromycin in 12 patients (out of a few million treatments) who had at least two other risk factors each for torsade de pointes. The association of azithromycin with QTc prolongation is controversial and still debated. Preclinical electrophysiological studies have extensively shown that azithromycin does not lengthen the QTc. Azithromycin seems to have a rather low affinity for the hERG channel: at a high concentration of 300 mM, an inhibition of 22.5% of the hERG current was reported, with an IC50 value of 1091 mM, a concentration absolutely unattainable at doses used in humans. In addition, intravenous administration of azithromycin failed to produce a significant prolongation of the QTc interval in dogs with chronic atrioventricular block and there was also no increase in short-term variability (from beat to beat) in the potential repolarization of the monophasic action [41]. Furthermore, azithromycin has been used long-term in patients with COPD or cystic fibrosis without reports of cardiovascular death [28,42]. Numerous other clinical studies have shown that QTc prolongation following azithromycin administration is clinically irrelevant [43–45], but many others have reported higher cardiovascular deaths [45–47] and cardiac arrhythmias, especially in the elderly population [48,49]. A meta-analysis of 33 observational studies on 22 601 032 patients found a statistically significant increase in myocardial infarction risk associated with macrolides use (OR = 1.15, 95% CI 1.01–1.30), but authors noted that erythromycin and clarithromycin were associated with a higher risk, compared with azithromycin (OR = 1.58, 95% CI 1.18–2.11 vs. OR = 1.41, 95% CI 1.11–1.81, respectively) [50].

5. Chloroquine/hydroxychloroquine azithromycin interaction

The proposed mechanism of QT prolongation induced by drugs is virtually the same for all medications. It is caused by a block in the outward IKr current, which is mediated by the potassium channel encoded by the *KCNH2* gene. The reversibility with drug discontinuation can be associated with a modification in extracellular potassium concentrations [51]. For this synergistic mechanism, co-prescription of QT-prolonging medications is associated with a higher mortality rate [52].

Some studies have been carried out to assess the risk of QT prolongation and fatal arrhythmias associated with the concomitant use of azithromycin and hydroxychloroquine, especially in patients with malaria. Since alterations in the duration of cardiac action potential are a measure of cardiac instability associated with a new onset of ventricular fibrillation, some authors have evaluated this parameter in guinea pigs. Pigs were anesthetized after the administration of azithromycin alone, chloroquine alone or in combination, reaching drug plasma concentrations clinically used to manage malaria. Chloroquine alone produced a marked increase in the duration of action potential and azithromycin did not. Azithromycin alone or in combination with chloroquine did not increase the action potential beyond the basic chloroquine responses, with no additional responsibility for arrhythmia [53]. In 2012, in order to use combination therapy to protect pregnant patients against malaria and sexually transmitted infections, Pfizer conducted a randomized, placebo-controlled, parallel study of 116 healthy controls who received 1000 mg of chloroquine alone or in combination with increasing doses of azithromycin (500 mg, 1000 mg and 1500 mg). Concomitant administration of chloroquine with azithromycin increased the QTc interval by 5 ms, 7 ms and 9 ms, respectively [54]. There was a very close correlation between the azithromycin dose and the increase in the QTc interval. Over the years, various studies have tested the combination of azithromycin-chloroquine or hydroxychloroquine in patients with malaria, with no reports of cardiovascular death [55,56]. The azithromycin-chloroquine or hydroxychloroquine combination is currently in use in Africa, India, and Thailand for the treatment of malaria.

More recently, a study on the use of Mobile Cardiac Outpatient Telemetry (MCOT) to monitor QTc prolongation and any occurring arrhythmias was carried out. The authors reported that out of 28 urgent alerts received by the cardiologist (by 18 of 117 patients) five alerts were for QTc prolongation. In one case, hydroxychloroquine needed to be discontinued because of QT prolongation [57]. In addition, 34.2% of patients were on treatment with at least another QT-prolonging medication.

On the other hand, in a recent COVID-19 study comparing safety and efficacy of chloroquine at high-dosage (600 mg twice daily for 10 days) versus chloroquine at low-dosage (450 mg twice daily on day 1 and once daily for 4 days) all patients were receiving azithromycin in association [58]. Eleven of 73 (15.1%) experienced QTc > 500 ms, with eight of 57 (14.0%) COVID-19 confirmed cases. QTc prolongation was more frequently found in the high-dosage group (18.9% vs. 11.1%) than in the low-dosage group. Two patients in the high-dose group experienced ventricular tachycardia before death but torsade de pointes were absent. Of note, 90% of patients were also on treatment with oseltamivir, which is also known to increase QTc. Also, previously mentioned studies by Rosenberg et al. and Mehra et al. respectively found a greater proportion of patients who experienced cardiac arrest (15.5%) and abnormal ECG findings (27.1%) among those receiving hydroxychloroquine plus azithromycin compared with hydroxychloroquine alone (13.7% and 27.3%, respectively) and azithromycin alone (6.2% and 16.1%, respectively) and neither drug (6.8% and 14.0%, respectively) [37]. An increased risk of de-novo ventricular arrhythmia during hospitalization correlated with the use of hydroxychloroquine or chloroquine with a macrolide (8.1%, 5.106 HR, 95% CI 4.106–5.983; 6.5%, 4.011, 3.344–4.812, respectively) [38]. In addition, an evaluation of the FDA's Adverse Event Reporting System (FAERS) from 1969 to Q3/2019, using a disproportionality analysis method, revealed that hydroxychloroquine/chloroquine alone were not associated with an increase in safety signal, while azithromycin alone

or in combination with hydroxychloroquine/chloroquine was associated with an increase in safety signal [59].

6. Precautions for the clinical use of the combination

Data from the literature on the risk of QT increase show that this side effect occurs in particular populations: long QT syndrome, bradycardias [60], arrhythmias, female sex [61], advanced age [60], and people with electrolyte imbalances and/or pre-existing cardiac pathologies. Hypokalemia seems to be one of the major triggers [62].

Since the potential risk for QT prolongation was reported, both the American College of Cardiology [63] and the European Society of Cardiology [64] have defined recommendations for the use of azithromycin and hydroxychloroquine in combination for COVID-19. In patients, especially the elderly, who start combination therapy with azithromycin and hydroxychloroquine the following steps are recommended:

- careful evaluation of the patient's clinical features
- correction of hypokalemia to a level $>$ 4 mEq/L and of hypomagnesemia to a level of $>$ 2 mg/dL
- discontinuation of any therapy with proton pump inhibitors [65] (with the exclusion of patients with a documented history of an ulcer, or Zollinger-Ellison syndrome): they notoriously reduce the absorption of potassium and magnesium. To control a possible rebound in the production of hydrochloric acid that occurs with the suspension of proton pump inhibitors, the use antacid drugs (for example sucralfate) is suggested, being careful to distance intake of at least 3 hours from COVID-19 therapies.

7. Conclusions

There is some promising evidence regarding the use of azithromycin as a potential treatment for COVID-19, but more structured studies should be carried out. However, a benefit-risk assessment must be cautiously performed due to the potential cardiac harm that the association of azithromycin and hydroxychloroquine could cause, especially in more fragile patients such as the elderly, with history of cardiovascular disease or co-medications known to prolong QTc. In particular, some measures must be implemented to provide patients' safety.

Declarations

Funding: No funding.
Competing Interests: None.
Ethical Approval: Not required.

References

[1] Amsden GW. Anti-inflammatory effects of macrolides–an underappreciated benefit in the treatment of community-acquired respiratory tract infections and chronic inflammatory pulmonary conditions. J Antimicrob Chemother 2005;55(1):10–21.
[2] Kanoh S, Rubin BK. Mechanisms of action and clinical application of macrolides as immunomodulatory medications. Clin Microbiol Rev 2010;23(3):590–615.
[3] Zarogoulidis P, Papanas N, Kioumis I, Chatzaki E, Maltezos E, Zarogoulidis K. Macrolides: from in vitro anti-inflammatory and immunomodulatory properties to clinical practice in respiratory diseases. Eur J Clin Pharmacol 2012;68(5):479–503.
[4] Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020;395(10223):497–506.
[5] Liu Q, Zhou YH, Yang ZQ. The cytokine storm of severe influenza and development of immunomodulatory therapy. Cell Mol Immunol 2016;13(1):3–10.
[6] Wang J, Nikrad MP, Travanty EA, Zhou B, Phang T, Gao B, et al. Innate immune response of human alveolar macrophages during influenza A infection. PLoS One 2012;7(3):e29879.
[7] Bermejo-Martin JF, Kelvin DJ, Eiros JM, Castrodeza J, Ortiz de Lejarazu R. Macrolides for the treatment of severe respiratory illness caused by novel H1N1 swine influenza viral strains. J Infect Dev Ctries 2009;3(3):159–61.
[8] Lee N, Wong CK, Chan MCW, Yeung ESL, Tam WWS, Tsang OTY, et al. Anti-inflammatory effects of adjunctive macrolide treatment in adults hospitalized with influenza: A randomized controlled trial. Antiviral Res 2017;144:48–56.
[9] Lendermon EA, Coon TA, Bednash JS, Weathington NM, McDyer JF, Mallampalli RK. Azithromycin decreases NALP3 mRNA stability in monocytes to limit inflammasome-dependent inflammation. Respir Res 2017;18(1):131.
[10] Min JY, Jang YJ. Macrolide therapy in respiratory viral infections. Mediators Inflamm 2012;2012:649570.
[11] Suzuki T, Yamaya M, Sekizawa K, Hosoda M, Yamada N, Ishizuka S, et al. Erythromycin inhibits rhinovirus infection in cultured human tracheal epithelial cells. Am J Respir Crit Care Med 2002;165(8):1113–18.
[12] Tahan F, Ozcan A, Koc N. Clarithromycin in the treatment of RSV bronchiolitis: a double-blind, randomised, placebo-controlled trial. Eur Respir J 2007;29(1):91–7.
[13] Zhang C, Xu Y, Jia L, Yang Y, Wang Y, Sun Y, et al. A new therapeutic strategy for lung tissue injury induced by influenza with CR2 targeting complement inhibitor. Virol J 2010;7:30.
[14] Lin SJ, Kuo ML, Hsiao HS, Lee PT. Azithromycin modulates immune response of human monocyte-derived dendritic cells and CD4(+) T cells. Int Immunopharmacol 2016;40:318–26.
[15] Jang YJ, Kwon HJ, Lee BJ. Effect of clarithromycin on rhinovirus-16 infection in A549 cells. Eur Respir J 2006;27(1):12–19.
[16] Yamaya M, Shinya K, Hatachi Y, Kubo H, Asada M, Yasuda H, et al. Clarithromycin inhibits type a seasonal influenza virus infection in human airway epithelial cells. J Pharmacol Exp Ther 2010;333(1):81–90.
[17] Suzuki T, Yamaya M, Sekizawa K, Hosoda M, Yamada N, Ishizuka S, et al. Bafilomycin A(1) inhibits rhinovirus infection in human airway epithelium: effects on endosome and ICAM-1. Am J Physiol Lung Cell Mol Physiol 2001;280(6):L1115–27.
[18] Culic O, Erakovic V, Cepelak I, Barisic K, Brajsa K, Ferencic Z, et al. Azithromycin modulates neutrophil function and circulating inflammatory mediators in healthy human subjects. Eur J Pharmacol 2002;450(3):277–89.
[19] Parnham MJ, Erakovic Haber V, Giamarellos-Bourboulis EJ, Perletti G, Verleden GM, Vos R. Azithromycin: mechanisms of action and their relevance for clinical applications. Pharmacol Ther 2014;143(2):225–45.
[20] Yamauchi K, Shibata Y, Kimura T, Abe S, Inoue S, Osaka D, et al. Azithromycin suppresses interleukin-12p40 expression in lipopolysaccharide and interferon-gamma stimulated macrophages. Int J Biol Sci 2009;5(7)::667-78..
[21] Poachanukoon O, Koontongkaew S, Monthanapisut P, Pattanacharoenchai N. Macrolides attenuate phorbol ester-induced tumor necrosis factor-alpha and mucin production from human airway epithelial cells. Pharmacology 2014;93(1-2):92–9.
[22] Gielen V, Johnston SL, Edwards MR. Azithromycin induces anti-viral responses in bronchial epithelial cells. Eur Respir J 2010;36(3):646–54.
[23] Cory TJ, Birket SE, Murphy BS, Hayes D Jr, Anstead MI, Kanga JF, et al. Impact of azithromycin treatment on macrophage gene expression in subjects with cystic fibrosis. J Cyst Fibros 2014;13(2):164–71.
[24] Gensel JC, Kopper TJ, Zhang B, Orr MB, Bailey WM. Predictive screening of M1 and M2 macrophages reveals the immunomodulatory effectiveness of post spinal cord injury azithromycin treatment. Sci Rep 2017;7:40144.
[25] Murphy BS, Sundareshan V, Cory TJ, Hayes D Jr, Anstead MI, Feola DJ. Azithromycin alters macrophage phenotype. J Antimicrob Chemother 2008;61(3):554–60.
[26] Retallack H, Di Lullo E, Arias C, Knopp KA, Laurie MT, Sandoval-Espinosa C, et al. Zika virus cell tropism in the developing human brain and inhibition by azithromycin. Proc Natl Acad Sci USA 2016;113(50):14408–13.
[27] Menzel M, Akbarshahi H, Tufvesson E, Persson C, Bjermer L, Uller L. Azithromycin augments rhinovirus-induced IFNbeta via cytosolic MDA5 in experimental models of asthma exacerbation. Oncotarget 2017;8(19):31601–11.
[28] Albert RK, Connett J, Bailey WC, Casaburi R, Cooper JA Jr, Criner GJ, et al. Azithromycin for prevention of exacerbations of COPD. N Engl J Med 2011;365(8):689–98.
[29] Taylor SP, Sellers E, Taylor BT. Azithromycin for the Prevention of COPD Exacerbations: The Good, Bad, and Ugly. Am J Med 2015;128(12) 1362 e1361-1366.
[30] Abisheganaden JA, Avila PC, Kishiyama JL, Liu J, Yagi S, Schnurr D, et al. Effect of clarithromycin on experimental rhinovirus-16 colds: a randomized, double-blind, controlled trial. Am J Med 2000;108(6):453–9.
[31] Arabi YM, Deeb AM, Al-Hameed F, Mandourah Y, Almekhlafi GA, Sindi AA, et al. Macrolides in critically ill patients with Middle East Respiratory Syndrome. Int J Infect Dis 2019;81:184–90.
[32] Kneyber MC, Kimpen JL. Antibiotics in RSV bronchiolitis: still no evidence of effect. Eur Respir J 2007;29(6):1285.
[33] McCallum GB, Morris PS, Chatfield MD, Maclennan C, White AV, Sloots TP, et al. A single dose of azithromycin does not improve clinical outcomes of children hospitalised with bronchiolitis: a randomised, placebo-controlled trial. PLoS One 2013;8(9):e74316.
[34] Franck Touret MG, Barral Karine, Nougairède Antoine, Decroly Etienne, de Lamballerie Xavier, Coutard Bruno. In vitro screening of a FDA approved chemical library reveals potential inhibitors of SARS-CoV-2 replication. bioRxiv 2020.
[35] Gautret P, Lagier JC, Parola P, Hoang VT, Meddeb L, Mailhe M, et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. Int J Antimicrob Agents 2020:105949.

[36] Molina JM, Delaugerre C, Le Goff J, Mela-Lima B, Ponscarme D, De Castro N. No evidence of rapid antiviral clearance or clinical benefit with the combination of hydroxychloroquine and azithromycin in patients with severe COVID-19 infectio. Med Mal Infect 2020;50(4):384.
[37] Rosenberg ES, Dufort EM, Udo T, Wilberschied LA, Kumar J, Tesoriero J, et al. Association of Treatment With Hydroxychloroquine or Azithromycin With In-Hospital Mortality in Patients With COVID-19 in New York State. JAMA 2020.
[38] Mandeep MR, Desai SSD, Ruschitzka F, Patel AN. Hydroxychloroquine or chloroquine with or without a macrolide for treatment of COVID-19: a multinational registry analysis. Lancet 2020.
[39] http://wayback.archive-it.org/7993/20170112032314/http://www.fda.gov/Drugs/DrugSafety/ucm304372.htm (accessed april 2020).
[40] https://www.fda.gov/drugs/drug-safety-and-availability/fda-drug-safety-communication-azithromycin-zithromaxor-zmax-and-risk-potentially-fatal-heart (accessed April 2020).
[41] Thomsen MB, Beekman JD, Attevelt NJ, Takahara A, Sugiyama A, Chiba K, et al. No proarrhythmic properties of the antibiotics Moxifloxacin or Azithromycin in anaesthetized dogs with chronic-AV block. Br J Pharmacol 2006;149(8):1039–48.
[42] Altenburg J, de Graaff CS, Stienstra Y, Sloos JH, van Haren EH, Koppers RJ, et al. Effect of azithromycin maintenance treatment on infectious exacerbations among patients with non-cystic fibrosis bronchiectasis: the BAT randomized controlled trial. JAMA 2013;309(12):1251–9.
[43] Mosholder AD, Mathew J, Alexander JJ, Smith H, Nambiar S. Cardiovascular risks with azithromycin and other antibacterial drugs. N Engl J Med 2013;368(18):1665–8.
[44] Sutton SS. Is cardiovascular risk a concern when prescribing azithromycin. JAAPA 2017;30(1):11–13.
[45] Svanstrom H, Pasternak B, Hviid A. Use of azithromycin and death from cardiovascular causes. N Engl J Med 2013;368(18):1704–12.
[46] Ray WA, Murray KT, Hall K, Arbogast PG, Stein CM. Azithromycin and the risk of cardiovascular death. N Engl J Med 2012;366(20):1881–90.
[47] Rao GA, Mann JR, Shoaibi A, Bennett CL, Nahhas G, Sutton SS, et al. Azithromycin and levofloxacin use and increased risk of cardiac arrhythmia and death. Ann Fam Med 2014;12(2):121–7.
[48] Maisch NM, Kochupurackal JG, Sin J. Azithromycin and the risk of cardiovascular complications. J Pharm Pract 2014;27(5):496–500.
[49] Choi Y, Lim HS, Chung D, Choi JG, Yoon D. Risk Evaluation of Azithromycin-Induced QT Prolongation in Real-World Practice. Biomed Res Int 2018:1574806.
[50] Gorelik E, Masarwa R, Perlman A, Rotshild V, Muszkat M, Matok I. Systematic Review, Meta-analysis, and Network Meta-analysis of the Cardiovascular Safety of Macrolides. Antimicrob Agents Chemother 2018;62(6).
[51] Yang T, Roden DM. Extracellular potassium modulation of drug block of IKr. Implications for torsade de pointes and reverse use-dependence. Circulation 1996;93(3):407–11.
[52] Freeman BD, Dixon DJ, Coopersmith CM, Zehnbauer BA, Buchman TG. Pharmacoepidemiology of QT-interval prolonging drug administration in critically ill patients. Pharmacoepidemiol Drug Saf 2008;17(10):971–81.
[53] Fossa AA, Wisialowski T, Duncan JN, Deng S, Dunne M. Azithromycin/chloroquine combination does not increase cardiac instability despite an increase in monophasic action potential duration in the anesthetized guinea pig. Am J Trop Med Hyg 2007;77(5):929–38.
[54] Pfizer Labs, 2013 https://www.accessdata.fda.gov/drugsatfda_docs/label/2013/050693s023,050730s031lbl.pdf (accessed April 2020).
[55] Kimani J, Phiri K, Kamiza S, Duparc S, Ayoub A, Rojo R, et al. Efficacy and Safety of Azithromycin-Chloroquine versus Sulfadoxine-Pyrimethamine for Intermittent Preventive Treatment of Plasmodium falciparum Malaria Infection in Pregnant Women in Africa: An Open-Label, Randomized Trial. PLoS One 2016;11(6):e0157045.
[56] Sagara I, Oduro AR, Mulenga M, Dieng Y, Ogutu B, Tiono AB, et al. Efficacy and safety of a combination of azithromycin and chloroquine for the treatment of uncomplicated Plasmodium falciparum malaria in two multi-country randomised clinical trials in African adults. Malar J 2014;13:458.
[57] Chang D, Saleh M, Gabriels J, Ismail H, Goldner B, Willner J, et al. Inpatient Use of Ambulatory Telemetry Monitors for COVID-19 Patients Treated with Hydroxychloroquine and/or Azithromycin. J Am Coll Cardiol 2020.
[58] Borba MGS, Val FFA, Sampaio VS, Alexandre MAA, Melo GC, Brito M, et al. Effect of High vs Low Doses of Chloroquine Diphosphate as Adjunctive Therapy for Patients Hospitalized With Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Infection: A Randomized Clinical Trial. JAMA Netw Open 2020;3(4.23):e208857.
[59] Sarayani A, Cicali B, Henriksen CH, Brown JD. Safety signals for QT prolongation or Torsades de Pointes associated with azithromycin with or without chloroquine or hydroxychloroquine. Res Social Adm Pharm 2020.
[60] Drew BJ, Ackerman MJ, Funk M, Gibler WB, Kligfield P, Menon V, et al. Prevention of torsade de pointes in hospital settings: a scientific statement from the American Heart Association and the American College of Cardiology Foundation. Circulation 2010;121(8):1047–60.
[61] Makkar RR, Fromm BS, Steinman RT, Meissner MD, Lehmann MH. Female gender as a risk factor for torsades de pointes associated with cardiovascular drugs. JAMA 1993;270(21):2590–7.
[62] Vandael E, Vandenberk B, Vandenberghe J, Willems R, Foulon V. Risk factors for QTc-prolongation: systematic review of the evidence. Int J Clin Pharm 2017;39(1):16–25.
[63] Roden DM, Harrington RA, Poppas A, Russo AM. Considerations for Drug Interactions on QTc Interval in Exploratory COVID-19 Treatment. J Am Coll Cardiol 2020;75(20):2623–4.
[64] Naksuk N, Lazar S, Peeraphatdit TB. Cardiac safety of off-label COVID-19 drug therapy: a review and proposed monitoring protocol. Eur Heart J Acute Cardiovasc Care 2020 2048872620922784.
[65] Lazzerini PE, Bertolozzi I, Finizola F, Acampa M, Natale M, Vanni F, et al. Proton Pump Inhibitors and Serum Magnesium Levels in Patients With Torsades de Pointes. Front Pharmacol 2018;9:363.