

REVIEW

Long-term macrolide treatment of chronic inflammatory airway diseases: risks, benefits and future developments

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Summary

Macrolide antibiotics were discovered over 50 years ago and following their use as antimicrobials it became apparent that this group of antibiotics also possessed anti-inflammatory properties. Subsequent clinical trials showed benefits of macrolides as long-term adjuncts in the treatment of a spectrum of chronic inflammatory respiratory diseases, particularly diffuse panbronchiolitis, cystic fibrosis, post-transplant bronchiolitis obliterans and more recently chronic obstructive pulmonary disease (COPD). The evidence for efficacy of macrolides in the long-term treatment of chronic asthma and bronchiectasis is less well established. The mechanism(s) of action of macrolides in the treatment of these diseases remains unexplained, but may be due to their antibacterial and/or anti-inflammatory actions, which include reductions in interleukin-8 production, neutrophil migration and/or function. Macrolides have additional potentially beneficial properties including anti-viral actions and an ability to restore corticosteroid sensitivity. The increased prescribing of macrolides for long-term treatment could result in the development of microbial resistance and adverse drug effects. New macrolides have been developed which do not possess any antimicrobial activity and hence lack the ability to produce microbial resistance, but which still retain immunomodulatory effects. Potentially novel macrolides may overcome a significant barrier to the use of this type of drug for the long-term treatment of chronic inflammatory airway diseases.

Keywords asthma, COPD, macrolides, microbial resistance, non-bacterial macrolides

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Introduction

Long-term macrolide treatment is effective in several chronic respiratory diseases including cystic fibrosis and diffuse panbronchiolitis, and may have a role in others including asthma, chronic obstructive pulmonary disease (COPD), bronchiectasis and post-transplant bronchiolitis obliterans. The purpose of this review is to summarize the strength of evidence for their efficacy in treating chronic respiratory diseases, particularly asthma and COPD, and to consider anti-inflammatory mechanisms accounting for clinical improvement. We also address risks associated with long-term macrolide treatment (Fig. 1) and consider what potential there is for developments in macrolides and future use for long-term treatment of chronic inflammatory airway diseases.

Macrolide antibiotics

Erythromycin was the first macrolide antibiotic and has been used since 1952. This prototypical macrolide consists of a 14-membered macrocyclic lactone ring with two sugar moieties (Fig. 2). Based on this, several other semi-synthetic macrolides have been developed, including clarithromycin; by substituting a methoxy group for the C-6 hydroxyl group of erythromycin [1], and azithromycin; by inserting a methyl-substituted nitrogen in place of a carbonyl-group in the aglycone ring, thus creating a 15-membered ring-structure [1].

These structures confer better acid stability, greater bioavailability and a longer half-life; 3–5 h and 40–68 h respectively [1].

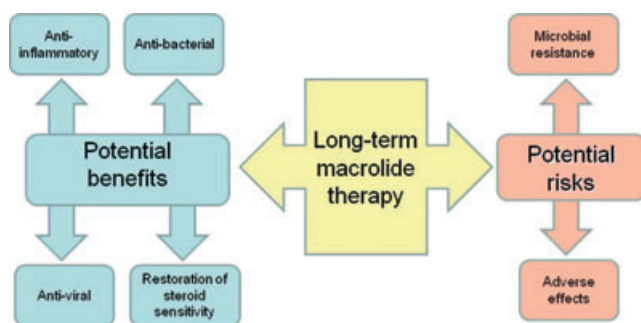


Fig. 1. Summary of potential benefits and risks of long-term macrolide therapy.

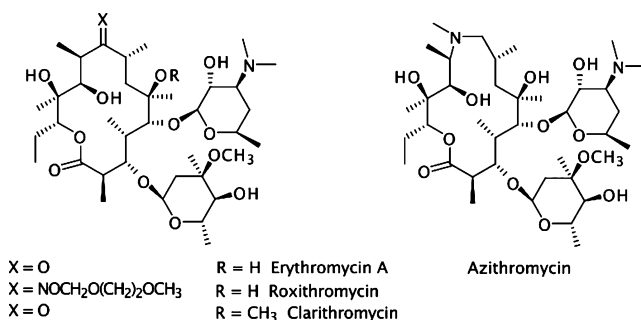


Fig. 2. Macrolide structure Reproduced from [83] with permission.

Efficacy of macrolides in the treatment of chronic inflammatory airway diseases

The efficacy of macrolides, administered as a long-term treatment, has been investigated in several chronic inflammatory respiratory diseases.

Asthma

Anti-inflammatory use of macrolides for asthma was first reported with troleandomycin in the 1960s [2], but hepatic adverse effects restricted further use [3]. Most recent studies have used newer macrolides, such as clarithromycin, roxithromycin and azithromycin for the treatment of chronic asthma and telithromycin for the treatment of acute exacerbations of asthma.

Clarithromycin is the most widely studied macrolide in asthma. A randomized double-blind placebo-controlled study ($n = 21$) with oral corticosteroid-dependent asthma reported that additional clarithromycin, 500 mg twice-daily for 6 weeks, resulted in 30% reduction in mean prednisolone requirement. However, there were no significant improvements in lung function, asthma quality of life or symptom scores [4]. In another randomized placebo-controlled study, ($n = 45$ non-smokers with refractory asthma) reported asthma quality-of-life score and symptoms improved with 2-months clarithromycin 500 mg twice-daily [5]. Studies have reported that short-term treatment with clarithromycin

can improve bronchial hyperresponsiveness (BHR) when added to adult patients with asthma who are already receiving moderate doses of inhaled budesonide [6]. Improvement in spirometry has been more difficult to achieve. A randomized, double-blind placebo-controlled trial of clarithromycin 500 mg twice-daily for 6 weeks ($n = 55$ chronic stable asthma) demonstrated a small improvement in FEV_1 , occurring only in those subjects PCR-positive for *Mycoplasma pneumoniae* or *Chlamydia pneumoniae*. The improvement in lung function was not found in the PCR negative subjects or when both groups were analysed together vs. placebo [7]. Another clarithromycin study ($n = 92$; mild-moderate asthma patients; stratified by PCR-positivity for *M.pneumoniae* or *C.pneumoniae* in bronchial biopsies) [8] showed no improvements in lung-function or asthma control questionnaire score, however, there was an improvement in BHR in the cohort as a whole or those who were PCR-negative; PCR-positive patients did not show improvement. This was a well designed multicentre study with patients undergoing bronchoscopy to obtain the best possible samples for bacteriology and PCR analysis. As such the study findings are likely to be robust and suggest that adding clarithromycin to an inhaled corticosteroid does not improve asthma control in symptomatic patients with mild to moderate asthma.

Roxithromycin, another 14-membered macrolide has also been studied in asthma [9, 10]. A small randomized controlled trial of 14 subjects with aspirin-sensitive asthma, who received 150 mg of roxithromycin twice daily vs. matched placebo, found no changes in FEV_1 or bronchial hyperresponsiveness, but did show improvement in asthmatic symptom scores [9]. In contrast, a large randomized controlled multicentre study in 232 volunteers with asthma and persistent symptoms, receiving 150 mg twice daily of roxithromycin for 6 weeks, found improvements in evening PEF, but no change in morning PEF values or asthma symptom scores [10].

There are few clinical studies of azithromycin in asthma, although it has the advantage of once-daily dosing. In a randomized parallel-group controlled trial ($n = 55$ children; moderate-severe persistent asthma) of azithromycin, montelukast or placebo, plus twice-daily inhaled budesonide and salmeterol, the budesonide maintenance dose was stepped-down until asthma control became inadequate resulting in high treatment failure rate and early trial termination. Data analysis suggested that azithromycin was unlikely to have had an inhaled corticosteroid-sparing effect [11]. In contrast to this conclusion, two other trials using azithromycin showed improvement in BHR. The first ($n = 11$ mild asthma), 250 mg azithromycin, twice-weekly for 8 weeks [12], although the limitations of this study are that it was open label and that it had no control group.

The second was placebo-controlled and demonstrated a reduction in BHR in 16 asthmatic children [13]. A 3-months placebo-controlled study ($n = 45$ adult asthma) reported that azithromycin improved symptom scores and reduced the use of rescue inhalations [14].

Probably the largest macrolide study is the 'Telithromycin in Acute Exacerbations of Asthma' (TELICAST) study; a multicentre, double-blind, randomized, placebo-controlled trial evaluating the efficacy of telithromycin in acute exacerbations. It should be emphasized that the TELICAST study was short-term—10 days of treatment, but the importance of this study rests on both its size and positive outcomes, hence its inclusion in this review. One of the two primary outcomes demonstrated positive improvements in symptom scores. The telithromycin group experienced a 40.4% reduction in symptoms compared with 26.5% in the placebo group (mean difference, -13.9 percentage point; 95% CI -23.4 – -4.3 ; $P = 0.005$). There was no effect on morning PEF, the other primary outcome, but secondary efficacy analysis found there to be significant improvement in FEV₁. Baseline FEV₁ improved until the end of treatment in the telithromycin group by 0.63 L, compared with 0.34 L in the placebo group (mean difference 0.29; 95% CI 0.12–0.46; $P = 0.001$) [15].

Taken together, the results of clinical trials of macrolides in chronic asthma suggest that treatment may improve symptoms and non-specific BHR, which may be independent of *M.pneumoniae* or *C.pneumoniae*, but at present there is little evidence of a sustainable beneficial effect on lung function. All the studies described above have significant limitations, primarily small numbers of participants. None have shown improvement in primary outcome measures, and the only large scale trial was of short treatment duration. There has also been a lack of consistency in controlling for effects in bacterial colonization. A Cochrane review supports the opinion that there is currently insufficient evidence to either support or refute the use of macrolides as anti-inflammatory therapy in patients with chronic asthma [16]. Future studies need to assess macrolide treatment in the management of severe asthma, possibly targeting those patients with associated bronchiectasis or who give a history of chronic mucus hypersecretion [17], as well as in acute asthma, perhaps in conjunction with assessment of changes in the microbial colonization, thus controlling for the potential anti-inflammatory effects of either change in bacterial load or diversity. Currently a number of clinical trials of azithromycin therapy in severe asthma (www.clinicaltrials.gov NCT00266851), smokers with asthma (NCT00852579), exacerbations of asthma (NCT00181272 and NCT01444469), childhood asthma (NCT01233297) and asthma control in adults in the community (NCT00266851), are under way. Hopefully the results of

these clinical trials will help clarify the role of macrolide therapy in the treatment of asthma.

COPD

Few controlled clinical studies have investigated the efficacy of long-term macrolides in COPD. A prospective, double-blind, randomized, placebo-controlled trial examining the effect of 3 months clarithromycin ($n = 67$; moderate-severe COPD) found no significant benefit on health status, exacerbation rate and sputum bacterial load [18]. A larger randomized placebo-controlled study ($n = 115$; stable COPD), erythromycin 250 mg twice-daily, followed for 1 year, demonstrated a reduction in exacerbation frequency [19]. There was no alteration in sputum inflammatory markers interleukin (IL)-6, IL-8 and myeloperoxidase (MPO) or the sputum bacterial flora. A large randomized controlled trial in patients with COPD ($n = 1142$) showed that 250 mg daily azithromycin for 1 year reduced the frequency of exacerbations (Fig. 3) (hazard ratio for time to first exacerbation 0.72 [95% CI 0.63–0.84]), as well as producing a small improvement in quality of life. Azithromycin treatment was associated with slight hearing impairment in a small proportion of patients [20]. This was a well performed study and although the adherence rate with study medication (67%) was lower than expected it provides considerable evidence for the use of azithromycin in patients who give a history of frequent exacerbations of COPD provided local antibiotic resistance is carefully reviewed. Interestingly a subgroup analysis showed that the response to azithromycin was less evident in younger patients (< 65 years), current smokers and those with GOLD stage 4 [20].

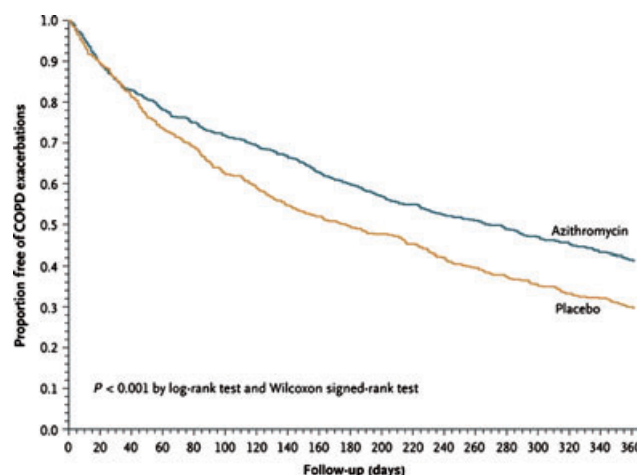


Fig. 3. Proportions of patients free of acute exacerbations of chronic obstructive pulmonary disease in 1 year follow-up. Reproduced from [20] with permission, Copyright © 2011 Massachusetts Medical Society.

Future studies are needed to identify whether certain phenotypes of COPD predict response to long-term azithromycin therapy, such as those with symptoms of chronic bronchitis as well as to assess the long-term safety of macrolide therapy in this patient population. Currently there is only one clinical trial actively recruiting to investigate the use of macrolide (azithromycin) therapy in COPD (www.clinicaltrials.gov NCT00985244). This study is measuring the effect of intermittent dosing (thrice weekly) of azithromycin on exacerbation frequency in COPD.

Other chronic inflammatory airway diseases

Diffuse panbronchiolitis. Diffuse panbronchiolitis (DPB) is characterized by chronic inflammation of the respiratory airways and is largely restricted to the Far East. Macrolides were first used to treat DPB in 1982 in relatively small numbers. In 1998, a large retrospective study of 498 patients demonstrated clear improvement in lung function and survival for patients treated with low dose erythromycin [21]. There appears to be a drug-class effect amongst other 14- and 15-membered macrolide antibiotics, [22] but this does not follow for 16-membered macrolides. Macrolides are now guideline therapy for DPB with recommendation to commence immediately on diagnosis [23]. The evidence for macrolides in DPB is compelling.

Cystic Fibrosis. Cystic fibrosis (CF) has clinical and bacteriological similarities to DPB, and following the establishment of macrolides in DPB treatment, pilot clinical trials were undertaken in CF [24]. A multi-centre randomized controlled trial of azithromycin in patients with chronic pseudomonas infection ($n = 185$) found improvement in mean FEV₁ of 0.097 L and in the frequency of exacerbations [25]. Conversely a more recent RCT with azithromycin in pseudomonas negative CF-adolescents found no improvement in FEV₁ [26]. The reasons for this are not entirely clear but the subjects in the latter study were younger (mean age 10 years cf 20 years in the earlier study) and had normal baseline FEV₁ (%pred 97–99% across the groups cf 68–70% in the former) which could make improvements more difficult to detect. A meta-analysis of prolonged-use azithromycin in CF confirmed the improvement in lung function [27]. This evidence has led to macrolides being a guideline recommendation in CF.

The argument for the use of azithromycin in CF must be balanced by the evidence that other inhaled antibiotics, such as tobramycin, have proven efficacy in reducing exacerbations and preserving lung function [28]. The original articles assessing the efficacy of azithromycin in cystic fibrosis allowed for the admission of patients already on long-term inhaled antibiotics, with

benefit still being conferred in the azithromycin group suggesting a degree of synergism [25].

Non-CF bronchiectasis. Five clinical studies (two randomized) investigated macrolides in non-CF bronchiectasis. The most reproducible finding was decreased sputum volume. This appears to be a class-effect with similar improvements following treatment with azithromycin [29], clarithromycin [30] and erythromycin [31]. Although the evidence for efficacy was only modest and there were relatively few high quality studies, these results were used to support recommendations for long-term macrolide treatment for patients with bronchiectasis and frequent exacerbations [32].

Post-transplant bronchiolitis obliterans. Bronchiolitis obliterans syndrome (BOS) is the leading cause of death following lung transplantation, leading to a progressive decline in FEV₁ with mortality rates from 25% to 50% [33]. Open-label studies involving small patient numbers have found improvement in FEV₁ following treatment with azithromycin; 250 mg for 12–36 weeks [34, 35]. The mechanism suggested was a reduction in airway neutrophilia and IL-8 [35]. A retrospective cohort study ($n = 178$ post-lung transplant BOS), 78 treated with azithromycin 250–500 mg 3-times-per-week showed increased survival, which was improved if commenced early in the development of BOS [36]. Early treatment with azithromycin (at BOS stage 1 and before stage 2) was associated with a decreased risk of death independent of other characteristics [36]. If azithromycin is given at stage 1 BOS, the risk of death has a hazard ratio of 0.29, 95% CI 0.11–0.82, $P = 0.02$ [36]. More recently, a large randomized placebo-controlled trial confirmed the efficacy of azithromycin—this time demonstrating a significant improvement in BOS-free survival at 2 years. This study utilized azithromycin treatment as an adjunct to normal anti-rejection therapy before the onset of any clinical evidence of BOS. Although there was no statistical difference in survival the follow-up period (2 years) was probably too short to identify any differences [37].

Chronic cough. Erythromycin has been used unsuccessfully in a randomized controlled trial in the treatment of chronic cough [38]. The rationale for the trial was the presence of sputum neutrophilia and its association with chronic cough. The premise being that macrolides have been shown to reduce sputum neutrophilia. What still remains undetermined is if neutrophilia is consequent or causative of the inflammation causing the cough. Whilst the sputum neutrophil count reduced significantly in the treatment group, none of the other outcome measures improved.

Mechanism(s) of action

The mechanism(s) of action of long-term macrolide treatment in chronic respiratory diseases remains unresolved. Macrolides have anti-inflammatory properties that are independent from their antibacterial activity and which may be beneficial in reducing airway inflammation. Furthermore, macrolides have potential additional beneficial properties including anti-viral activity and an ability to restore corticosteroid sensitivity.

Anti-inflammatory activity

A part of macrolide anti-inflammatory activity may be a consequence of the reduction in bacterial load afforded by the antibiotic effects [39] in addition to the anti-inflammatory activity that is distinct from its antibacterial activity [1]. The nature and mechanism of the anti-inflammatory activity has been reviewed [40]. The diversity of these activities suggests a variety of mechanisms affecting immune and tissue-derived cell function including cytokine production. There is no single immunomodulatory-axis through which macrolides exert their effect and we will review the most important pathways.

Macrolide effects on inflammatory cells. Neutrophils. Macrolide therapy can reduce neutrophil accumulation in the airway epithelium [5, 13, 35]. This may be associated with macrolides reducing the local production of CXCL-8 (IL-8); a powerful chemotactic factor for neutrophils, as reported in DPB [35, 41, 42]. This mechanism was also suggested in a study using clarithromycin and azithromycin in refractory asthma leading to reduced airway CXCL-8 and neutrophils [5, 13].

Monocyte and macrophages. Macrolides have anti-inflammatory effects on monocytes/macrophages. Classically activated macrophage (M1) phenotypes, induced by stimulation with interferon (IFN) γ and bacterial lipopolysaccharide (LPS) are associated with microbicidal and cytotoxic function and pro-inflammatory cytokine production [43]. Alternatively activated (M2) phenotypes induced by IL-4/13 are associated with Th2-type, immunosuppressive and remodelling responses. Azithromycin reduced the production of pro-inflammatory IL-1 β , and tumour necrosis factor (TNF) α in mouse M1 macrophages [44] and polarized cells towards M2, with reduced pro-inflammatory IL-12 and IL-6, and increased anti-inflammatory IL-10 [45] (Fig. 4). *Ex-vivo* human monocytes treated with clarithromycin could effectively and significantly reduce LPS stimulated IL-8 production in a dose dependant manner [46]. The relevance to lung disease is that alveolar macrophages become polarized during infection towards M1 [47], but

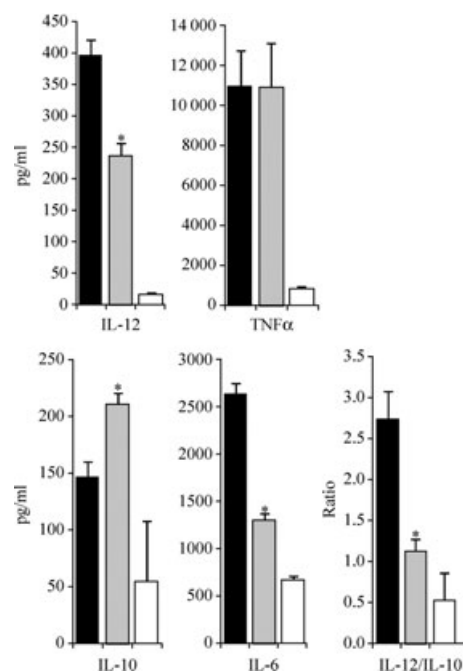


Fig. 4. Azithromycin alteration of cytokine production in the stimulated mouse macrophage cell line J774. J774 cells were cultured with IFN γ overnight in the presence or absence of azithromycin. Cells were stimulated with LPS for 24 h and cytokines were measured from supernatants. Black bars depict IFN γ +LPS; Grey IFN γ +LPS+Azithromycin; White unstimulated. Differences between the two treatment groups were significant $P < 0.05$. Reproduced from [45]. Murphy BS *et al.* Azithromycin alters macrophage phenotype. *Journal of Antimicrobial Chemotherapy* 2008;61[3]:554-560 by permission of Oxford University Press.

their pro-inflammatory activity may be attenuated by macrolides e.g. by reduced production of TNF α , IL-1 β and IL-8 [44]. In addition, polarization towards anti-inflammatory M2 [45] characterized by IL-10 production [48], and scavenger-receptor expression associated with clearing apoptotic cells, and reduced apoptotic bronchial epithelial cells have been observed in COPD patients receiving azithromycin [48].

Macrolide effects on cytokine production. Azithromycin has many anti-inflammatory effects [49] including down-regulation of production of pro-inflammatory mediators e.g. prostaglandin E2, nitric oxide and cytokines TNF α , IL-8, IL-1 α , growth-related oncogene (GRO)- α and soluble vascular cell adhesion molecule (sVCAM)-1. Many of these are chemotactic, activation and survival factors for neutrophils.

IL-8 (CXCL8) is a potent chemoattractant of neutrophils. A randomized double-blind placebo-controlled trial (twice-daily clarithromycin 500 mg) in refractory asthma demonstrated a significant reduction in airway IL-8 levels and neutrophil numbers [5]. Sub-group analysis showed that airway IL-8 protein and

gene expression was predominantly reduced in the non-eosinophilic asthmatic patients. Similarly, azithromycin reduced airway IL-8 mRNA in patients with post-transplant BOS following 3 months treatment [35].

IL-1 β is sufficient to induce neutrophil accumulation in the lung [50], GM-CSF is a neutrophil survival factor [51], and both are derived from macrophage and airway epithelial cells and are central to airway infectious inflammation [47]. In a murine model of LPS-induced pulmonary neutrophilia, azithromycin or clarithromycin were able to reduce airway neutrophilia with striking reductions of IL-1 β and GM-CSF [52].

Molecular mechanisms of macrolide anti-inflammatory activity. Defining molecular mechanisms of anti-microbial macrolide function received the 2009 Nobel prize, yet the mechanisms of anti-inflammatory macrolide function remain unresolved; likely reflecting their complexity. Macrolides accumulate and persist inside leucocytes [1] suggesting effects on cell signalling. Azithromycin can inhibit mitogen-activated protein (MAP) kinases [53]; which regulate cellular processes, e.g. gene expression, cell growth, proliferation, differentiation and survival in response to a variety of extracellular stimuli, especially cytokines including IL-8 and GM-CSF as mentioned above. Putative mechanisms e.g. with clarithromycin suggest altered DNA binding activity of transcription factors NF- κ B and AP-1 [46, 54], and inhibition of synthesis and/or secretion of pro-inflammatory cytokines [40].

In summary, different macrolides can cause reductions in pro-inflammatory cytokines, suggesting that there is a class effect inhibiting airway neutrophilia. Individuals with neutrophilic inflammation may therefore derive greatest benefit. *In vitro* and *in vivo* studies demonstrate efficacy in multiple pathways leading to reductions in the inflammatory response. The evidence for inhibition of inflammatory pathways is widespread, but this does not always or necessarily lead to a clinically relevant or measurable response.

Macrolide effects on mucus production. Macrolides appear not to affect normal physiological secretion of mucus [40], but can reduce hypersecretion [55], possibly by inhibiting production of pro-inflammatory cytokines, e.g. TNF α , that stimulate mucin genes MUC5B and MUC5AC in airway goblet cells [56] and IL-13 that induces goblet cell hyperplasia and mucus hypersecretion [57].

Macrolide microbial activity as a factor in immunomodulation

Evidence against anti-microbial activity of macrolides accounting for their anti-inflammatory activity includes

effective long-term treatment of DPB at sub-antibacterial concentrations, and efficacy when colonized with macrolide-resistant *pseudomonas aeruginosa* [22]. Mechanisms may include macrolide interference with microbial protein synthesis even below minimal antimicrobial concentrations that can inhibit synthesis of virulence factors [58, 59].

Macrolide antiviral effects

Rhinoviruses (RV) cause ~60% of virus-induced asthma exacerbations. Macrolides appear to have inherent antiviral properties, and induce anti-viral responses. Several studies have demonstrated the beneficial effects of macrolides in experimental RV and influenza infections. The addition of a macrolide led to reduced virus titres, most likely as a consequence of inhibition of RV-induced up-regulation of ICAM-1 and also inhibition of pro-inflammatory cytokine production; ICAM-1 is the RV receptor on airway epithelium [60–62].

Corticosteroid-sparing effects

Macrolides may have corticosteroid-sparing effects, first shown with troleandomycin, but the use of this macrolide has been limited due to adverse effects [63]. Corticosteroid-sparing efficacy is limited to case reports [64] and small open-label pilot studies [65], which demonstrate improvement in clinical laboratory endpoints—enhanced sensitivity of lymphocytes to suppression by dexamethasone. Although macrolide antibiotics are inhibitors of CYP3A4 [1] and can increase the plasma concentration of CYP3A4 substrates, which include fluticasone and budesonide, this mechanism is unlikely to contribute to the localized therapeutic effects of inhaled corticosteroids on the airways of patients given macrolides.

Smokers with COPD have decreased histone deacetylase-2 (HDAC2) activity in alveolar macrophages, and this may lead to increased inflammatory gene expression and reduced sensitivity to corticosteroids [66] and a similar mechanism may occur in smokers with asthma [67]. Erythromycin can increase HDAC2 levels *in vitro* [68], suggesting a mechanism by which macrolides therapy may restore corticosteroid sensitivity and improve asthma control in smokers with asthma.

Risks of long-term use of macrolides

Macrolides are licensed for short-term use as antibiotics for acute infections. They are currently being used off-license for long-term treatment and information on the safety of these drugs for this use is limited. Recommendations on the use of macrolides in the treatment of airway diseases should be based on the balance of the

risk of adverse effects vs. clinical effectiveness. Unfortunately for many of the airway diseases in which long-term macrolide therapy is used there is lack of adequate information of both adverse effects and efficacy. With the increasing use of macrolides across the respiratory disease spectrum, large numbers of patients are now taking macrolides on a long-term basis which increases the likelihood of adverse effects and the emergence of drug resistant microbial organisms.

Adverse effects of long-term use

In cystic fibrosis, studies have used macrolide treatment for up to 52 weeks and no serious adverse effects due to the drug were reported. Minor side effects were common, in the form of nausea and diarrhoea. A recent meta-analysis assessed the effect of prolonged use of azithromycin in CF patients [27]. Treatment duration was 13–52 weeks, with a total of 177 subjects in the azithromycin arm, with a mean age 18.1 years. The gastro-intestinal side effects were 72% higher than placebo, with the main complaint being nausea and diarrhoea. Gastrointestinal side-effects may be due to motilin-like activity of macrolides stimulating gastrointestinal motility [1]. The longest treatment phase amongst clinical trials with azithromycin is 2 years [37]. There were no significant differences between the treatment and placebo groups in terms of cardiac, gastrointestinal or emergence of drug-resistant organisms [37].

Oral erythromycin prolongs cardiac repolarisation and is associated with case reports of torsades de pointes. A large retrospective epidemiological study reported that the risk of sudden death from cardiac causes among patients currently using erythromycin was twice as high as those not using any antibiotic and the adjusted rate of sudden death from cardiac causes was five times as high among those who concurrently used CYP3A inhibitors along with erythromycin [69], although this adverse effect is rare [40].

A recent report has suggested that long-term azithromycin may be associated with hearing loss, but it is possible that the investigators were too stringent and that hearing loss was overestimated [20].

Emergence of drug resistant microbial organisms

A worrying potential consequence of the increasing use of long-term macrolides therapy in the management of chronic respiratory diseases is the emergence of drug-resistant microbial organisms. There is great variation in the reported rate of macrolide-resistance to *Streptococcus pneumoniae* (MRSP), ranging from 4 to 70% in worldwide surveillance studies [70]. In a meta-analysis Halpern and colleagues (29 studies; 1998–2003) found the mean resistance of *S. pneumoniae* isolates to

azithromycin was 27.2% (95% CI 24.6–29.7) and to erythromycin was 30.4% (95% CI 28.1–32.7). Too few studies of clarithromycin were included to allow an accurate evaluation of resistance [71].

Macrolides are important for the treatment of mycoplasma infections, and reports of macrolide resistant *M. pneumoniae* are rising. In Japan, the increase was from 0% in 2002 to > 30% in 2006 [72]. With long-term azithromycin use in CF, erythromycin resistant *Staphylococcus aureus* increased from 6.9 to 53.8%, and clarithromycin resistant *Haemophilus spp* from 3.7 to 37.5% [73]. In *Mycobacterium avium* infections, azithromycin treatment is used for long durations in combination with other antibiotics. In one study, no *M. avium* isolate from patients receiving azithromycin in combination with rifabutin and ethambutol developed macrolide resistance [74].

Risk-benefits of long-term macrolide therapy

Benefits of macrolides in selective chronic inflammatory respiratory diseases have been assessed earlier in this review. Safety data collected during long term clinical trials do not suggest that serious side effects (QTc prolongation, liver toxicity) are of significant concern, but it is worth noting that clinical studies exclude patients at risk of developing likely adverse effects with treatment, for example, subjects with background history of severe liver or kidney disease, abnormal electrocardiograms or those with known drug interactions. General safety precautions should be followed in routine clinical practice. A balance needs to be struck between clinical benefit, long-term safety and uniquely to this type of therapy—the incidence and spectrum of microbial resistance. There is a need for large randomized clinical trials for many of the airway conditions in which macrolides are currently used that carefully assess both long-term efficacy and adverse effects.

Future developments

Predicting a therapeutic response to macrolides in airway diseases

The concept of responders and non-responders to macrolide treatment has been proposed [35]. In one study of patients with bronchiolitis obliterans a higher baseline bronchoalveolar neutrophil count predicted a favourable response to macrolide treatment [35]. Induced sputum is a recognized method of categorizing the underlying type of airway inflammation into neutrophilic and eosinophilic phenotypes [75]. Carefully selecting the group of patients most likely to respond to macrolides may be important in decision making when



Fig. 5. Macrolide conjugate.

prescribing, given they appear to exert a greater effect on airway neutrophils and neutrophilic asthma [5].

Non-bacterial macrolides—overcoming the shortcomings of macrolides

Recent development of chemically modified non-antibiotic macrolides that maintain anti-inflammatory properties may overcome the risk of microbial drug resistance. EM703 is a 12-membered-ring derivative of erythromycin that is devoid of anti-bacterial actions, but exhibits anti-inflammatory properties. For example, EM703 inhibits transforming growth factor- β induced proliferation, transcription of type I collagen and collagen production of both human and murine lung fibroblasts [76, 77] and NF κ B activation and IL-8 production in human bronchial epithelial cells [78]. In addition, EM703 can increase HDAC2 levels *in vitro* [68], suggesting a possible mechanism by which this compound may restore corticosteroid sensitivity in COPD and in smokers with asthma [66, 67]. There is evidence in the patent literature of novel compounds that are claimed to demonstrate dissociation of anti-inflammatory effects from antibiotic effects. A derivative of the macrolide azithromycin (CYS0073) has been developed that has anti-inflammatory actions in the absence of any bactericidal properties [79]. This candidate molecule has been found to reduce inflammation in animal models of inflammatory bowel disease and rheumatoid arthritis [79]. If similar anti-inflammatory effects are found in experimental models of airway inflammation then this molecule could be developed as a potential therapy for chronic inflammatory airway diseases in man.

Macrolide antibiotics have the ability to accumulate in inflammatory cells including neutrophils, monocytes and alveolar macrophage as well as epithelial cells [1]. Due to this property, non-antibiotic macrolides have been developed that are conjugated with a second, pharmacologically active drug that has anti-inflammatory properties and as such, these compounds are used to transport and increase anti-inflammatory drug action. Using this property macrolides have been developed that act as a drug carrier for an active compound, such as a corticosteroid [80], cyclo-oxygenase or 5-lipoxygenase inhibitor [81], which is covalently bound to the macrolides (Fig. 5). Macrolide conjugates have been patented that include pyrrolizine and indolizine compounds that act as inhibitors of 5-lipoxygenase and cyclooxygenase [81] or that contain coumarins [82].

Macrolide conjugate compounds that include a corticosteroid can bind to the glucocorticoid receptor, accumulate in cells and can inhibit T cell proliferation, IL-2 production and ovalbumin-induced airway inflammation in mice [80]. In addition, the intracellular localization of the active compound, such as a corticosteroid limits the likelihood of systemic adverse effects and ensures a localized anti-inflammatory action of the active compound. There are no published reports on the clinical efficacy and anti-inflammatory effects of non-bacterial macrolides in the treatment of airway diseases.

Conclusions

Macrolides are now widely used for long-term treatment of chronic respiratory inflammatory disease. Their benefit is uncertain in diseases such as asthma, whereas there is good evidence to support their use in cystic fibrosis, diffuse panbronchiolitis and COPD. The underlying mechanisms are becoming better understood and whilst there is evidence for multiple effects, there is no single mechanism that can account for their anti-inflammatory action. A common theme appears to be the effects on cytokine e.g. IL-8 production and consequently neutrophil recruitment, activation and function. These findings have been consistent across different disease models and individual macrolides. In addition, macrolides show the ability to alter cellular function at the molecular level and through intracellular signalling pathways. There is concern that the widespread use of macrolide antibiotics could result in the development of microbial resistance and adverse drug effects which may ultimately limit the use of this class of drug. Macrolides are not without adverse effects and whilst QT prolongation is the most worrying of these, there is good evidence that these side effects are rarely encountered. Synthetically derived macrolides are under development that may improve anti-inflammatory functions and at the same time abrogate microbial activity. Novel macrolides could overcome a significant barrier to the use of this type of drug for the long-term treatment of chronic inflammatory respiratory diseases. For diseases such as severe asthma, appropriately powered clinical trials are now required to give a definitive answer whether there is any benefit to the chronic use of macrolides or macrolide derivatives. Until these are performed it is likely some patients may be treated inappropriately.

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